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> Published by The Arab Society for Plant Protection

# SUNN PEST MANAGEMENT A Decade of Progress 1994-2004

Edited by

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International Center for Agricultural Research in the Dry Areas (ICARDA) Aleppo, Syria



Published by the Arab Society for Plant Protection

#### ISBN 978-9953-0-1063-2

Published by the Arab Society for Plant Protection P.O. Box 113-6057, Beirut, Lebanon E-mail: aspp@terra.net.org Website: http://www.asplantprotection.org/

## Citation

Sunn Pest Management: A Decade of Progress 1994-2004. 2007. Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini and Safaa G. Kumari (Eds.). Published by the Arab Society for Plant Protection, Beirut, Lebanon. 432 pp.

#### **Availability of Publication**

This publication is available on request to: ICARDA, P.O. Box 5466, Aleppo, Syria

Back cover photo by Jafar Mohaghegh, *Plant Pests and Diseases Research Institute, Tehran, Iran.* A typical overwintering site for Sunn Pest in the high mountains of Iran

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Printed in Beirut, Lebanon.

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# Foreword

Wheat is the staple food in the Central and West Asia and North Africa (CWANA) region. Annual per capita consumption of wheat in CWANA is about 185 kg, the highest in the world. The region accounts for 23% of global wheat area but only 14% of global wheat production. Wheat productivity is only 1.5 t/ha, compared with the world average of 5 t/ha. The main limiting factors include drought—70% of wheat acreage in CWANA is affected by drought—heat and cold stress, diseases and pests. To meet domestic demand, many countries in the region have to import large quantities of bread wheat, which places a heavy burden on national economies.

Of the various wheat pests, the Sunn Pest is the most devastating. It can cause up to 100% crop loss. It affects about 15 million hectares of wheat annually, and over US\$150 million is spent each year on pesticides in the Sunn Pest-prone areas. In addition to the high cost, insecticides pose risks to human health and the environment. In collaboration with a range of partners, ICARDA has been developing integrated pest management (IPM) options for Sunn Pest. The partners include NARS in CWANA; the University of Vermont, USA; CABI Bioscience; and the Natural Resources Institute, University of Greenwich, UK.

Scientists from 23 countries in CWANA and Eastern Europe (where wheat and barley crops suffer Sunn Pest infestation) met at ICARDA headquarters on 19-22 July 2004 for the Second International Conference on Sunn Pest on the theme 'Enhancing International Cereal Production Capacity for Food Security'. The conference was coorganized by ICARDA, the University of Vermont and the Arab Society for Plant Protection.

Research findings presented and discussed during the conference highlighted various technical possibilities of IPM: safeguarding the complex of natural enemies, use of insect-killing fungi, host plant resistance, cultural practices, and selective use of pesticides. IPM will reduce production costs, benefit national economies and significantly reduce environmental pollution and health hazards.

While very encouraging progress has been made in research, several priority issues were identified to further enhance the field implementation of effective, sustainable and environmentally sound IPM strategies. Significant investment in both research and on-farm field activities must be made through regional approaches and international collaboration. Because insects do not recognize country borders, a regional strategy is critical, if we are to reduce the negative impact of Sunn Pest on food security. The ultimate goal is to maintain Sunn Pest populations at low levels using an integrated approach in a well coordinated regional program.

Mahmoud Solh Director General ICARDA

# Preface

Sunn Pest is a broad term covering a group of insect species that cause serious damage to wheat and barley in the developing world. One species, *Eurygaster integriceps* Puton, stands out as the most prevalent, and causes the most severe damage. Yield loss in wheat can reach 90-100% when insect populations are high. Also important is the fact that this species injects a chemical into the grain while feeding. This chemical destroys glutin, imparts an off-flavor to the flour subsequently made, and prevents the final product, bread, from rising properly. In certain parts of the world even cattle refuse to feed on the hay made from Sunn Pest infested fields.

Sunn Pest is found from North Africa through West Asia and countries of the former Soviet Union, and east to Pakistan. It causes problems throughout its range and management is difficult. Currently the primary defense is to spray crops with insecticides.

In 1993 FAO and ICARDA convened the *First International Conference on Sunn Pest.* About 25 scientists attended. The consensus of the group was that research should be revived, with emphasis on integrated pest management (IPM) strategies. In 1996 ICARDA, NARS and the University of Vermont initiated a concentrated effort to develop new approaches. These initial efforts were supported by funds from the United States Agency for International Development (USAID) and the Conservation, Food and Health Foundation. Several years later the Department for International Development (DFID) in the United Kingdom funded a major proposal on research and development for Sunn Pest IPM. A cohesive group of NARS staff in West Asian countries, international institutions and universities started work on many aspects of IPM. This group was coordinated by ICARDA.

Excellent progress was made towards effective management of Sunn Pest. In 2004, ten years after the first Sunn Pest conference, a second international conference was convened, and attended by 150 participants from 23 countries. This publication, the proceedings of the *Second International Conference on Sunn Pest*, contains 54 papers on a range of subjects, all of which directly contribute to a clearer understanding of Sunn Pest IPM.

We trust that the contents of these proceedings, representing many different facets of Sunn Pest biology and management, justify our efforts and the sponsors' support.

Editors

Bruce L. Parker Margaret Skinner Mustapha El Bouhssini Safaa G. Kumari

# Acknowledgments

The Second International Conference on Sunn Pest would not have been possible without the efficient help provided by the local organizing committee with assistance from the staff of ICARDA's entomology group. The internal support from ICARDA's visitors service was exemplary. Ms. Rita Nalbandian and Mrs. Tamar Varvarian kept track of the many details involved and always addressed each problem with urgency and efficiency.

Special thanks to Ms. Teri Hata who spent many hours double checking manuscripts and organizing the editors.

The conference was financially supported by the Department for International Development (DFID), the United States Agency for International Development (USAID), the Food and Agriculture Organization of the United Nations (FAO), the Islamic Development Bank (IDB) and the United States Department of Agriculture (USDA).

This proceedings has been made possible with funds from the Food and Agriculture Organization of the United Nations (FAO).

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Participants in the First International Conference on Sunn Pest, ICARDA, 1993



Participants in the Second International Conference on Sunn Pest, ICARDA, 2004



# The CGIAR System-wide Program on Integrated Pest Management: Initiation, Mission, Objectives, Governance and Activities

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#### Abstract

Makkouk, K.M. 2007. The CGIAR System-wide Program on Integrated Pest Management: Initiation, Mission, Objectives, Governance and Activities. Pages 3-7.

A System-wide Program on integrated pest management (SP-IPM) was established by the CGIAR in 1995 as an inter-institutional partnership program that responds to CG governance, administration, funding and reporting, with a mission to increase the quality and usefulness of IPM research for higher productivity, sustainable profitability and healthier harvest of cropping systems and enhance the well-being of people, particularly in resource limited agricultural areas in the developing world. In this paper, I will briefly describe the specific objectives, partnership and governance of the SP-IPM and summarize its achievements during the past eight years.

Keywords: CGIAR, SP-IPM.

#### Introduction

In response to Agenda 21, the action plan developed by the United Nations Conference on Environment and Development, convened in Rio de Janeiro in 1992, the CGIAR established a special Task Force to develop recommendations on how to incorporate such an agenda into the CG system strategy and plan of work. One of the Task Force recommendations was to develop an intercenters IPM initiative. Accordingly, IARCs were invited to a meeting in Oslo, Norway in 1994 to discuss the establishment of such an initiative. CG representatives in the Oslo meeting recommended the establishment of an IARCs-IPM network and articulated a coherent CG policy on IPM. They also nominated IITA to be the convening center for the network. The Centers Directors Committee (CDC) accepted the Oslo meeting recommendations and endorsed the inclusion of two non-CG centers, AVRDC and ICIPE, in the network.

During the first meeting of the IARC-IPM network in the Hague (1995), IARC representatives recommended to CDC and to

the CG Technical Advisory Committee (TAC) the formation of a system-wide program on IPM (SP-IPM) to support and coordinate activities in this field, and to establish an Inter-Center Working Group on IPM (ICWG-IPM) to serve as its steering committee and to provide a forum for discussion of relevant CG policy, strategy and activities. In this meeting a number of project areas were also selected. Thus, the SP-IPM emerged as an interinstitutional partnership program of the CGIAR that responds to CG governance, administration, funding and reporting, with a mission to increase the quality and usefulness of IPM research for higher productivity, sustainable profitability and healthier harvest of cropping systems and enhance the wellbeing of people, particularly in resourcelimited agricultural areas in the developing world.

## Objectives

The main objectives of the SP-IPM are to: (i) promote inter-institutional partnerships and break barriers to IPM development, (ii) promote holistic and ecological approaches/methodologies for IPM technology development, (iii) promote effective communication between stakeholders for informed decision-making, (iv) promote farmer adoption of proven IPM technologies for sustainable and healthier harvests, and (v) promote public awareness and impact of IPM strategies.

# Partners

Partnership in the SP-IPM includes a balanced group of research organizations (eight CGIAR Centers, three IARCs and NARSs through projects and IPM pilot sites), IPM promoting agencies (GIPMF, IPM Forum, IAPPS, BioNET and World Bank), advocacy groups (CGIAR NGO Committee and Pesticide Action Network) and the private sector Crop Life International). Table 1 lists the complete membership in the IIWG.

#### **Governance and Coordination**

The SP-IPM implementation structure includes a Steering Committee (SC), an Inter-Institutional Working Group (IIWG) (Table 1), Thematic Working Groups (TWG) (Table 2) and a coordinating Secretariat. The SC serve as the decision body of the program with membership restricted to the Program Leader and IIWG officially appointed institutional representatives. The Program Coordinator is an ex-officio member. The SC conducts business meetings annually, on the last day of each IIWG meeting.

The IIWG is the discussion forum to make recommendations to the SC, agree on policy and vision, priority problems for which an inter-institutional effort could make a difference, on contractual obligations to strengthen collaboration, promote networking, and review progress. Membership includes SC members, program coordinator, special project coordinators, and leaders of the thematic working groups. Occasionally, IIWG is enlarged to include NARSs staff on SP-IPM implementation activities and representatives of special interest groups to update IIWG on topical IPM issues.

 Table 1.
 Members of the Inter-Institutional

 Working Group (IIWG).
 Image: Comparison of the Inter-Institutional

### CGIAR Centers

- International Institute of Tropical Agriculture (IITA)
- Centro Internacional de lap Papa (CIP)
- Centro Internacional del Mejoramiento de Maiz y Trigo (CIMMYT)
- International Center for Agricultural Research in the Dry Areas (ICARDA)
- International Center for Research on Agroforestry (ICRAF)
- International Institute of Tropical Agriculture (IITA)
- International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)
- International Rice Research Institute (IRRI)
- International Service for National Agricultural Research (ISNAR)
- West Africa Rice Development Association (WARDA)

#### **Other Partners**

- Asian Vegetable Research and Development Center (AVRDC)
- CABI Bioscience
- International Center of Insect Physiology and Ecology (ICIPE)
- FAO/World Bank Global IPM Facility
- IPM Forum
- Pesticide Action Network (PAN)
- representing NGOs
- CropLife International

The TWG examines priority problems and develops a coherent response, which may or may not include development and implementation of special projects. Peer review by thematic groups will insure that the special projects and activities are well designed to ensure inter-institutional research, compliment existing efforts, and that proposed activities are properly prioritized to respond to the most pressing needs. At present, there are nine approved thematic working groups (Table 2).

The Coordinating Secretariat is comprised of the Program Leader and the Coordinator who will catalyze and facilitate approved activities, mobilizes and disseminates technical and material resources, develop information and publicity materials on IPM learning and advocacy, serve as a contact point, seek funding, and organize external evaluation of the program.

**Table 2.** Coordinators of the different SP-IPMThematic Groups.

Thematic Working			
Group		Coordinator/Institute	
1.	Tropical whitefly group	F. Morales/CIAT	
2.	FPR & L methods for IPM	E. Van de Fliert/CIP	
3.	Soil Biota*	A. Bellotti/CIAT A. Yahyaoui/ICARDA	
4.	Functional agro- biodiversity	I. Gordan/ICIPE	
5.	Crop loss and impact assessment*	O. Ortiz/CIP H. de Groottee/CIMMYT	
6.	Biotechnology for IPM	D. Bergvinson. CIMMYT	
7.	Leaf miner fly IPM*	A. Lagnaoui/CIP	
8.	Alien invasive species	M. Tamo/IITA	
9.	IPM policy research	K. Gallagher/FAO- GIPMF	

\* Organized international workshop on the respective theme.

# **Summary of Achievements**

The major activities and achievements of the SP-IPM over the last eight years can be summarized as follows:

Project "Sustainable on integrated management of whiteflies as pests and vectors of plant viruses in the tropics" - The whitefly project started in 1996, covering 12 countries in Latin America and the Caribbean. and 10 in Africa. and involved five IARCs. 26 NARSs and six advanced research institutes in the UK, Germany and the USA. Phase I of the project completed diagnostic surveys to assess/characterize agronomic, socioeconomic, and epidemiological features of whiteflies and whitefly transmitted viruses in cassava, legumes and sweet potato in six countries in Latin America, nine in Africa, three in the Caribbean and in Mexico using the standardized methodologies developed by the partners. Phase II of the project will extend project activities geographically, scientifically and organizationally. This is in line with earlier project recognition that improved understanding of population dynamics, epidemiology of viral diseases, biology, genetic variability, natural enemies and host plant selection will be essential to the development and implementation of integrated management methods.

Global project on "Farmer participatory research and learning for IPM" - This project coordinated by the SP-IPM Task Force on FPR-IPM, in collaboration with the System-wide Program on Participatory Research and Gender Analysis (SP-PRGA), FAO Global IPM Facility, and CABI Biosciences provided research planners, managers, and policy makers with guidelines on the principles and practices of farmer participation that underpin successful IPM. and developed a common vision of what the various stakeholders would need to do differently in order to increase the effectiveness of participatory approaches in IPM. This was accomplished through a series of mentored study-exchange visits between contrasting pairs of successful projects on participatory research and learning. To capitalize on FPR experiences world-wide, mentor exchange visits were followed by a learning workshop at which project participants and representatives from other projects/programs conducted a cross-cutting analysis of the case studies generated by the study tour participants in order to provide practical guidelines on the choice and use of participatory approaches.

Pilot sites for IPM learning and adoption -Although a number of promising IPM options are available, adoption of IPM at the farm level is disappointingly slow in the developing countries. Poor communication between researchers and farmers is believed, by many stakeholders in the agricultural development process, to be a major constraint limiting IPM adoption. Effective partnerships to involve farmers in the research process to develop, test/adapt and disseminate IPM options are also uncommon. Additionally, organizational and policy obstacles and extension bottlenecks discourage the dissemination of proven options. In response to these constraints, SP-IPM introduced "pilot sites" in 2000 as part of its implementation strategy to introduce "best bet" IPM options to farming communities, assist participating organizations to gain experience in developing effective farmerscientist-extension (FSE) partnerships, and increase understanding, dissemination and adoption of IPM options by farmers.

Five pilot sites were selected in major cropping systems or agro-ecologies in Africa (Kenya, Nigeria, Burkina Faso, Morocco, Egypt) where farmers had already identified pests as a principal concern. The selection criteria included prior characterization of other bio-physical and socio-economic features of the candidate; availability of promising IPM options that have the potential to achieve a decisive improvement in the locality: existence of research and development activities to provide a platform for pilot site development; opportunities for achieving new synergies by closer collaboration between IARCs and partners; the presence of strong partners to take primary responsibility for site activities; and identification of a wide extrapolation domain for the results of pilot activities. At each pilot site, FSE teams analyzed production problems, identified farmers' coping strategies, and agreed on "best bet" options to evaluate. There is no room here to discuss in details the activities and results obtained in each pilot site, but in general, results obtained were very encouraging and the successes achieved over the period 2000-2002 encouraged up-scaling of theses activities by some of the NARSs in the respective countries.

International Workshops - Three thematic working groups organized international workshops and facilitated global information exchange to harmonize approaches on: (i) managing soil-borne pests, declining soil fertility, soil borne beneficial organisms and plant health in tropical and sub-tropical agriculture; (ii) developing a common framework and methods for impact assessment, taking into account the economic, environmental, human- and social-capital aspects of IPM; and (iii) addressing problems associated with leaf mining flies in a wide range of cropping systems.

Public awareness of IPM benefits - A key activity of the SP-IPM is information dissemination to increase public and donor awareness of the benefits of IPM and to raise the profile of IPM within donor communities. The program has produced and disseminated various informational resources such as research highlights, CGIAR scientific publications and book chapter contributions in three IPM books, newsletters of partners organizations such as IAPPS and CABI, public awareness brochures (1), publicity videos, and information resources on CD for farmer participatory research and participatory learning, technical report to donors, a video CD "Breaking the Cycle" on participatory IPM of Striga/cereal stem borers (3), adaptation of cassava IPM information into a user-friendly format to facilitate farm-level understanding and use, IPM information exchange workshops (2) in collaboration with the Consortium for International Crop Protection (CICP) and the CGIAR NGO Committee, an updated website (www.spipm.cgiar.org), projects websites (e.g., www.tropicalwhiteflyproject.cgiar.org), listserves (e.g., fpr-ipm@cgiar.org and NGO-IPMnet@cgiar.org), and interactive web-based electronic newsletter in collaboration with the EU-supported RUNnetwork for information brokerage.

# الملخص

مكوك، خالد. 2007. برنامج النظام الواسع على الإدارة المتكاملة للآفة: البدء، المهمة، الأهداف، التوجيه، والنشاطات. الصفحات 3-7.

أسس برنامج النظام الواسع على الإدارة المتكاملة للآفة (SP-IPM) من قبل CGIAR عام 1995 كبرنامج شراكة مؤسساتية متبادلة يستجيب لتوجيه، إدارة، تمويل وتدوين CG، مع مهمة زيادة النوعية والفائدة من دراسة المكافحة المتكاملة للآفات من أجل إنتاجية أعلى، ربحية مستدامة وحصاد أكثر صحة للنظام المحصولي وتحسين رفاهية الناس، وبشكل خاص في المناطق الزراعية محدودة المصدر في العالم النامي. ستصف هذه المقالة باختصار الأهداف الخاصة، الشراكة، وتوجيه SP-IPM وستلخص منجزاته خلال السنوات الثمانية الماضية.

كلمات مفتاحية: CGIAR، SP-IPM.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Making an Economic Miracle: Agricultural Science, Technology, and Economic Growth

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#### Abstract

Gollin, D. 2007. Making an Economic Miracle: Agricultural Science, Technology, and Economic Growth. Pages 9-15.

Economic tools and concepts can be applied usefully to the allocation of resources for agricultural research. Previous research suggests that investments in agricultural research tend to have high rates of return. Economic arguments also explain why the public sector role in agricultural research can be critical: while private actors may have incentives to develop or market chemicals, they have few incentives to develop technologies that do not require the use of purchased inputs. As a result, public investments in agricultural research, such as those on Sunn Pest, can have high payoffs.

Keywords: Priority setting returns to research, Sunn Pest.

### Introduction

The discipline of economics may have little to offer in the efforts to control Sunn Pest directly. Compared to spraying or the development of techniques for rearing entomopathogenic fungi, economics can only offer indirect support for the struggle to help farmers deal with pest problems. But financial constraints and economic language are often invoked to justify the limited funding for research and implementation of Sunn Pest control measures-as indeed, this language is generally used to justify the limited expenditure on many worthy projects. This paper will briefly review the reasons why economic concepts may be appropriate for thinking about control of Sunn Pest. The paper will then offer some reasons for believing that agricultural research, in general, can play an important role in improving productivity and contributing to economic growth. The paper makes no attempt to provide an economic assessment of the importance of Sunn Pest, nor to suggest what budgetary resources should be devoted to Sunn Pest controls: these are questions that would require different methodology and considerable data collection. Instead, this paper draws on theoretical arguments and on the experience of the Green Revolution to argue that agricultural research can contribute to economic growth miracles.

#### Scarce Resources and the Application of Economic Concepts

Economics is often defined as the discipline that studies the allocation of scarce resources among competing uses. This definition seems particularly apt for the discussion of Sunn Pest and the funding of research related to its control. At present, Sunn Pest affects broad areas of West Asia and North Africa (WANA), where it causes direct and indirect crop losses to wheat and barley. By some estimates, government expenditures on Sunn Pest spraying and control amount to \$40 million annually and extend over 15 million ha. On affected areas unprotected by spraying, crop losses of 50-90 percent are possible in wheat, taking into account the fact that grain produced on affected plants may be unusable for bread baking. Many of the farmers affected most directly by Sunn Pest are those growing crops in or near mountainous terrain in dry areas. Many poor families are affected, and for them the crop losses caused by the pest may represent significant income losses.

Until recently, spraying has typically been viewed as a government activity, rather than a farmer responsibility. As such, it represents a substantial drain on public budgets and also imposes considerable external damage to health and the environment. With per capita incomes in the WANA region relatively stagnant since 1980 (Figure 1), and with per capita public expenditures actually declining (Figure 2), there are simply few resources to put in to Sunn Pest control.



Figure 1. GDP per capita, West Asia and North Africa, 1970-2001.



**Figure 2.** Government Final Consumption Expenditure per capita, West Asia and North Africa, 1970-2001.

Sunn Pest control is then competing for scarce public money. But public funding is not the only mechanism by which pest problems can be addressed. Farmers themselves might reasonably be expected to invest in control, either through chemical use or through changes in practices. Private firms might in principle be expected to offer control services or even to provide research. Are these alternative models reasonable? Is public financing essential?

Although alternatives are possible, Sunn Pest in many ways offers a classic example of a "public goods" problem, as economics understands the term. No individual farmer has an incentive to spray at an efficient level. If an individual farmer sprays, other farmers may benefit, without having to share the cost. If many farmers are spraying, each individual farmer has an incentive to "free ride" and to avoid spraying, hoping that he or she can enjoy the benefits of neighbors' actions without bearing the costs. Moreover. individual farmers may use chemicals inefficiently or in ways that damage their health or the environment. As a result, it may be more sensible for governments to provide the spraying services.

Leaving aside spraying, IPM research typically focuses on reducing input use, rather than increasing it. For private firms, there are few reasons to invest in this kind of research, unless it uses some other input intensively (e.g., substitutes a machine for chemicals). Thus, most IPM research has come out of governmental institutions and nongovernmental organizations (NGOs).

As for varietal improvement research, private firms have historically invested little in research on wheat or barley, because those crops are self-pollinated and hence farmers can save seeds from one crop to the next. This limits the attractiveness of the market for improved varieties; a company that invests in research will not be able to recoup much of its research expenditure through seed sales. As a result, in most countries, wheat and barley research have been largely undertaken by the public sector. In developing countries, national agricultural research systems (NARS) have been assisted in this work by international agricultural research centers (IARCs), which have collaborated to breed improved crop varieties for a large number of crops and regions.

The public contribution to Sunn Pest research, management, and control is thus essential, and the resources available for the purpose are limited. Economics offers us some insights into how resources can be allocated and also into the question of whether overall levels of funding are adequate.

This paper has little concrete advice to offer about how Sunn Pest resources are allocated across competing research areas or between research and control. Without detailed data, there is little basis for such statements. Moreover, additional information would be needed to discuss the priority assigned to Sunn Pest within NARS or IARC research programs or within agriculture ministries. There are strong reasons, however, to believe that public spending on agriculture is generally too low in many WANA countries, and there are also reasons to argue that international spending on agricultural research and development is inefficiently low. Put in different terms, this is to suggest that the payoffs to spending on agriculture and agricultural research may be very high.

#### **Agriculture in Public Priorities**

Agriculture and agricultural research often receive fairly low levels of public support in developing countries. In many countries, agriculture has been viewed as a backward sector, and the development theories of the 1950s and 1960s tended to focus on modernization and industrialization. Public spending and capital investments thus tended to be skewed away from agriculture.

For many countries, macroeconomic policies further worked against agriculture. As has been widely documented (e.g., 16, 19), many countries supported their development efforts by promoting domestic industrial interests through various kinds of protection, including tariffs, credit subsidies, and tax breaks. A key implicit form of support was overvaluation of exchange rates. In many countries, governments pursued exchange rate policies (deliberately or not) that had the effect of overvaluing the local currency. This tends to make tradable goods relatively cheap -- a boon for importers of machinery and capital goods, but a penalty for agricultural producers, who often found the domestic currency price of their output driven down to extremely low levels.

The "bias against agriculture" was fairly strong in most WANA countries through the 1990s, with relatively high rates of tariff protection for manufactures and very high levels of non-tariff barriers. A 1997 study noted that, with the policy regimes then in place, "a significant bias against agriculture will remain and continue to inhibit the potential of agriculture to contribute more significantly to domestic output and exports" (5).

In terms of public investment. international investments in agricultural development-and especially in science and technology-have declined considerably in the past 15 years. As noted by Byerlee et al. (2), World Bank lending for agricultural research was at a lower level in nominal terms (unadjusted for inflation) in 2000-02 than in any three-year period since at least 1981. Taking inflation into account, World Bank lending for agricultural research has declined dramatically - a situation exacerbated by the decline in support from many bilateral donors, including USAID.

# Lessons from Theory: Agriculture and Economic Growth

The decline in funding has occurred despite growing evidence that investments in agriculture - and particularly in agricultural science and technology - have high payoffs. A long literature in development economics has recognized that agricultural growth plays an important role in stimulating economy-wide growth in incomes and reductions in poverty. Recent reviews of this literature include Mellor (18) and Timmer (21). These papers offer well-reasoned "stories" for the connection between agricultural growth and economic growth, but only in the past several years have macroeconomic theorists begun to accept that the link might be important. Indeed, a prevalent view has been that agricultural growth might in fact slow or impede the process of economic development. (12, 17).

Recent papers have challenged this notion, including Caselli and Coleman (3), Kongsamut, Rebelo and Xie (15), Irz and Roe (13), Gollin, Parente, and Rogerson (9, 10, 11), and Kogel and Prskawetz (14). Gollin *et al.* (10) show that differences in agricultural productivity levels can account for a substantial fraction of the differences in incomes across countries. Subsequent work, reported in Gollin *et al.* (11) argues that agricultural technology changes can lead to dramatic growth in the short- and medium-run, although long-run income levels are likely to depend on policies affecting other sectors of the economy.

In this research, the authors suggest that subsistence food needs play an important role in constraining development in poor countries. Because productivity is generally quite low, countries must devote large amounts of labor to producing food, and incomes in the rural sector will be correspondingly low. Without improvements in agricultural technology, these economies cannot begin to release labor to other sectors or to produce non-agricultural goods. An implication is that countries might be able to escape this "food problem" if they could import sufficient quantities of food, but with a few exceptions (some of them in the WANA region), most economies are not in a position to do so. Certainly, where populations are large and are dispersed in rural areas (i.e., in countries that have significant agricultural populations) it will not be feasible to import food and distribute it on the scale required.

Gollin *et al.* (11) suggest that low levels of agricultural productivity can slow economies' development by 100 years or more; dramatic improvements in agricultural technology ("Green Revolutions") have the potential to unleash rapid economic growth, although this growth cannot be sustained unless policies for the non-agricultural sector are good.

# Lessons from Policy: Agricultural Research and the Green Revolution

This theoretical literature meshes with a growing empirical literature supporting the links between agricultural productivity change and growth. Some of this literature in turn emphasizes the importance of agricultural growth and agricultural science for poverty reduction; see, for example, Datt and Ravallion (4) or Thirtle *et al.* (20).

One strand of this literature examines the returns to public investments in agricultural research. Numerous studies have found that the returns to agricultural research are high – often extraordinarily high. Even a skeptical review of the literature by Alston *et al.* (1) suggests that rates of return of 30-40 percent are common, echoing previous findings by Evenson and various co-authors.

These estimates are typically based on the costs and benefits of individual research programs, which of course are different from the growth impacts of research. To examine this issue, we need a different methodology. One approach comes from a comprehensive review of crop breeding impacts across 11 crops and all regions, reported in Evenson and Gollin (6) and summarized in Evenson and Gollin (7). This work suggests that investments in IARC research did in fact lead to substantial productivity improvements across crops and regions, though with widely varying degrees of impact. In some regions, such as sub-Saharan Africa, research impacts were quite modest, partly because research on relevant crops (cassava, tropical maize) lagged behind research on rice and wheat, and partly because of ill-conceived research strategies that sought simply to transfer technologies developed elsewhere, rather than to invest in intensive programs of breeding within Africa itself.

Evenson and Gollin (6) also summarize work by Evenson and Rosegrant (8) examining the overall impact of the investments in research through the IARC system. Without attaching a precise value to the rate of return, Evenson and Rosegrant consider the likely outcomes of two counterfactual scenarios - one in which no "Green Revolution" had occurred and productivity grew quite slowly from 1960-2000, and one in which a "lite" Green Revolution had taken place, with no input from IARCs. Depending on the scenario under consideration, Evenson and Rosegrant (8) concluded that these counterfactuals would have left the world substantially worse off, with yields of food crops in developing countries lower by 8-20 percent. Crop prices would have been higher, by 6-20 percent, leading to reduced food consumption; total calorie intake per person would have been 4-14 percent lower in developing countries, with substantial variation across countries. As a consequence, the number of malnourished children would have been higher, by 2-8 percent.

These are large impacts. Although a Green Revolution cannot by itself eliminate hunger or poverty, this research suggests that improvements in agricultural productivity can have a major impact on human welfare.

#### Implications for IPM and Sunn Pest

What does this tell us, if anything, about the value of investments in integrated pest management (IPM) programs generally, or Sunn Pest controls in particular? Perhaps not much; after all, the returns to any individual research program depend on the science that is available, the resources that are brought to bear, and indeed on a certain amount of luck. But the theoretical and empirical evidence lead support to the notion that investments in research can have large payoffs. Not all research investments will prove successful, of

course; if there are no failures, then arguably there is evidence of underinvestment.

But among the questions to ask of a research program, we might consider the following:

- Does the science justify the investments? Is the probability of success sufficient, given the (uncertain) research payoffs and the (more certain) costs?
- Can the research be deferred? Given progress in other fields, is it wise to postpone the research and to take advantage of spillovers from research done on other subjects?
- Will other actors take the lead in carrying out the research? Is it possible to take advantage of spillovers across institutions?
- Of the various research projects that satisfy these criteria, how does it compare in terms of likely payoffs? How does it compare in terms of impact on poverty or equity?

There are many more such questions we could ask, and indeed an entire literature focuses on the economics of research priority setting.

But many IPM programs and Sunn Pest management programs in particular, seem likely to meet these criteria. The science involved is generally well understood. There is no reason to wait for advances in other fields or upstream disciplines (as there might be, for example, in developing vaccines for animal diseases). Other actors are unlikely to do the work that public institutions are currently doing and contemplating. For private sector firms, there is little financial incentive to invest in Sunn Pest management strategies, because there is unlikely to be a saleable product that could allow them to recoup any investments in research. Seeds of resistant crop varieties might well be taken up by farmers, but for wheat and barley, the two most susceptible crops, there is little private sector seed research. This reflects the fact that farmers can generally save the seeds of selfpollinated crops, so private firms cannot recoup much return from investments in research.

Under current circumstances, Sunn Pest research is unlikely to be funded on a large scale by NARS in rich countries, except as a donative collaboration with poor countries. Sunn Pest is not yet a major problem in the rich countries, and consequently it ranks low on the priority list of researchers. Like many agricultural problems of developing countries, then, it is unlikely to be solved without substantial commitments by research institutions in the affected region - ideally working with additional financial and human resources from the international community.

#### Conclusions

Can investments in Sunn Pest IPM programs produce economic growth miracles? Perhaps not on their own. It is surely unrealistic to expect Sunn Pest programs to deliver dramatic improvements in income levels or growth rates. It may not be unrealistic, however, to assume that research on Sunn Pest management and control may have high rates of return or that, like many previous research programs, it may play a role in the long, incremental, and unglamorous - but ultimately vital - process of improving agricultural productivity.

#### الملخص

غولين، دوغلاس. 2007. عمل معجزة اقتصادية: العلوم الزراعية، التقانة، والنمو الاقتصادي. الصفحات 9–15.

يمكن أن تطبق الأدوات والمفاهيم الاقتصادية بشكل مفيد لتوزيع المصادر للبحث الزراعي. أوضحت الدراسات السابقة أن الاستثمارات في البحوث الزراعية تتجه لامتلاك نسب عالية من المردود. تشرح المنازعات الاقتصادية أيضاً لماذا يمكن أن يكون دور القطاع العام حاسماً في البحث الزراعي: بينما لدى القطاعات الخاصة حوافز لتطوير أو تسويق الكيماويات، حيث يملكون حوافز قليلة لتطوير التقانات التي لا تتطلب استخدام مدخلات الانتاج المشتراة. وكنتيجة، الحوافز الحكومية في البحوث الزراعية، حيث الزراعية، كتلك التي على حشرة السونة، يمكن أن تعطي ربحاً عالياً.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# How Can Scientists and Scientific Programmes Connect with Public and Private Enterprise to Further the Cause of IPM

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## Abstract

Brader, L. 2007. How Can Scientists and Scientific Programmes Connect with Public and Private Enterprise to Further the Cause of IPM. Pages 17-24.

The IPM research and development activities over the last 20 years have clearly shown the heterogeneity of the agricultural production systems. Thus, no standard solutions can be offered to address the plant health needs of the individual farmers. Extension staff can offer a set of tools, but the farmers themselves must be able to decide on how these tools can be used most effectively. The traditional extension methods of offering standard solutions are not suitable anymore. Plant health research has increasingly moved in this direction, but the extension services have not. An exception is the Farmer Field School approach that has proven its effectiveness. The lack of appropriate extension approaches is further exacerbated by the registration requirements for plant protection products. IPM researchers have not facilitated the necessary transformation towards more effective farmer support systems. For example, the IPM definition keeps changing because of the evolving understanding of the key elements needed for producing a healthy crop. It is high time to define in broadly understandable terms what IPM is. The goal is to produce a healthy crop with minimum use of synthetic pesticides. On this basis closer collaboration has to be established with extension and regulatory agencies to finally arrive at a joint and appropriate approach. Changes have to take place in a system that is still dominated by strong interest groups with outdated ideas. A real lobbying exercise will have to be undertaken. The IPM research community has little capacity to do this well. It will need effective partners that require clear facts showing what can be gained by growing healthier crops.

Keywords: Integrated Pest Management (IPM), plant health, extension, farmer field school.

#### Introduction

For over 60 years it is known that chemical control has undesirable side effects on the agro-ecosystem leading to a deterioration of the health status of crops. During the 1930s and the early 1940s such undesirable changes were first observed and studied in particular in citrus production in Florida and California, and in apple orchards in Nova Scotia, Canada. Researchers determined that the negative effect on the natural enemy complex led to rapid population growth of new pest species. In citrus growing a sharp increase occurred in various scale insects after spraying with DDT to control among others citrus thrips. The population density of other pest species also increased (4, 13). In apple orchards in Nova Scotia there was a strong increase of various pest species, in particular scale insects and mites, after spraying sulphur preparations to control apple scab (9). Pickett in Canada and DeBach and Ray F. Smith in California were among the pioneers that laid the basis for what we now call Integrated Pest Management (IPM). Their work was supported by many others.

In the developing world the beginning of IPM may be traced back to research carried out in the Ca ete Valley in Peru. The tobacco budworm, *Heliothis virescens* Fabricius, appeared for the first time in the valley in 1939, and the use of insecticides resulted in a rapid build up of the cotton aphid, *Aphis* gossypii Glover. The *Heliothis* problem worsened every year notwithstanding an increased number of insecticide applications. Again the elimination of the natural enemy complex by the insecticides used proved to be the major reason for the serious pest outbreaks. This was exacerbated by the emergence of pesticide resistance. Excellent work to develop improved control systems was undertaken by Barducci, Beingolea and others (2, 3).

#### **IPM a Slow Mover**

Since those early days a tremendous amount of research and development efforts have been carried out with the purpose to ensure the development and introduction of effective IPM practices. This work is not limited anymore to entomology, but is now covering all plant health disciplines. However, notwithstanding this extensive range of research and development activities one notes that IPM, while routinely referred to in agricultural development efforts, is not being applied on a very wide scale. In almost all cases pesticides are still considered the first and easiest approach to address plant health problems. Pesticide statistics over the last decades do show a reduction in the overall quantity of active ingredients of insecticides used, but this is essentially due to the development of new products of which smaller quantities of active ingredient are applied per unit area.

Some of the best IPM research and development work has been undertaken in California, USA. An analysis of pesticide use in this State led to the conclusion, that data from the California Pesticide Use Reports support the hypothesis that relatively little reduction occurred during the decade of the 1990s, even though there is "nationwide infrastructure in the land grant universities, Cooperative Extension Service, and USDA that presumably have a goal of reduction of pesticide use or risk" (5). In the USA the trends in the use of the total amount of active ingredient of the various pesticides since 1964 are as follows (7, 8).

- Insecticides: no reduction from 1964 till 1976, from 1976 till 1982 a reduction of about 30 percent, from 1982 till 1992 a further reduction of 25 percent, during the 1990s the total amount of active ingredient used stayed about the same, some 60 million pounds per year. Thus, in 1997 the amount of insecticide active ingredient used was about 50 percent of the amount used in 1964.

- Fungicides: a gradual increase from 1964 till 1997 of about 200 percent.
- Herbicides: the amount of active ingredient used has increased seven-fold between 1964 and 1997.

Osteen (7, 8) considers that the reduction in insecticide use is primarily due to the replacement of organochlorines by other insecticides that can be applied at much lower rates. In addition, during the 1980s, reduction in commodity prices and land that was idled under Federal Programmes, led to an overall decline in pesticide use. With respect to IPM practices Osteen notes that since 1991 there has been an increase in the use of microbial pesticides, in particular Bacillus thuringiensis Berliner (Bt.) strains, but its use is mainly limited to fruit trees and vegetables. Between 10 and 43 percent of the apple, blueberry, grape, nectarine, plum, prune, sweet cherry, blackberry and raspberry acreage received Bt. applications in 1997. Bt. was used on about half or more of the cabbage, celery, eggplant, tomato, and pepper acreage in 1996. Bt-treated cotton acreage increased form 5 percent in 1992 to 9 percent in 1995, but fell to 2 percent in 1997. This decrease may be associated with the increased planting of Bt. gene containing cotton seed.

During the 1990s there has been an increase in the use of pheromones. For example, pheromones were used for monitoring on 33 percent and for pest control on 7 percent of cotton grown; they were used on a much smaller portion of other field crops. Scouting for insects in field crops ranged from 59 percent for soybeans to 98 percent for fall potatoes, with 66 and 88 percent of the corn and cotton acreage also being scouted. On average, scouting for insects reached 67 percent among all field crops in 1996. But, as noted above, these developments do not seem

to have led to a noticeable reduction in the amount of insecticides used.

IPM became a component of federal agricultural policy in the USA in 1972 with the aim to reduce the risks from pesticide use and to develop more sustainable agricultural production strategies. IPM has indeed been introduced widely in the USA. However, the pesticide statistics indicate that this has had a limited effect on the amount of pesticides used. Epstein and Bassein (5), discuss a number of reasons for this.

- i. IPM adoption against insect pests may entail a change in the specific chemicals used rather than a change in the volume of material applied.
- ii. Fungicide use is generally not driven by the presence of the disease, but rather by the perceived risk of disease or the consequences of disease that occurred in earlier years.
- iii. IPM programmes are usually dominated by chemical control and are better defined as programmes of pesticide management, instead of ecologically based pest management programmes.
- iv. Reduced pesticide use is an economic benefit to growers, but, in many cases, this reduction must be accompanied by an increase in cultural control practices to maintain adequate pest management, entailing additional costs

Osteen (8) refers to a national study of biologically based IPM in the early 1990s, sponsored by USDA and EPA, which concluded that dozens of technical, institutional, regulatory, economic, and other constraints need addressing in order to achieve broader adoption of IPM. Three constraints were identified for all commodity groups:

- (i) lack of funding and personnel to conduct site-specific research and demonstrations;
- (ii) producer perception that IPM is riskier than conventional methods, more expensive, and not a short-run solution; and

(iii) educational degree programmes that are structured towards narrow expertise rather than broad knowledge of cropping systems.

Notwithstanding the wide range of possible examples demonstrating the beneficial impact of the adoption of IPM, the data for the USA seem to confirm the hypothesis that, so far, the overall beneficial effect of IPM in agricultural practice has been rather limited. James et al. (6), while discussing the CGIAR Systemwide Programme on Integrated Pest Management, summarize the situation as follows.

- In the search for sustainable options to increase food security, IPM plays a key role, having the potential to increase the productivity of agricultural systems while minimizing threats to human health and the environment.
- IPM has evolved from pesticide abatement strategies aimed at avoiding the pesticide treadmill into analytical approaches to understand pest status within production ecologies in order to make informed decisions on appropriate actions.
- Although a number of promising options are available, adoption of IPM at farm level is disappointingly slow in the developing countries.
- Thus, the full potential of IPM to reduce shortfalls in food production is poorly realized.
- Many research initiatives focus largely on developing component technologies with minimal understanding of clientoriented approaches in the innovation process, and neglect the key role of policy environment in IPM promotion.

# IPM in Rice Growing, a Success Story

Very impressive results have, however, been achieved over the last 15 years with the introduction of IPM in rice growing in Southeast Asia. An analysis of this successful undertaking may help in defining the conditions needed to increase the success rate of IPM introduction. It should be kept in mind that attempts to introduce IPM in rice growing started only to show lasting beneficial results from the early 1990s onwards when Farmer Field Schools (FFS) were conducted for the first time in Indonesia. An overview of the development and implementation of FFS has been published by Pontius, Dilts and Bartlett in 2002 under the title "Ten years of IPM training in Asia. From farmer field school to community IPM" (10).

The authors note that "the centralized systems were unable to take into account the reality of pronounced agro-ecological diversity within countries, regions and even villages. The inclusion of routine pesticide applications within the input packages (of the rice Green Revolution) often caused severe ecological disruptions, most notably the rise of pest resurgence and (pesticide) resistance".

The FFS allow the farmers to become the decision makers with respect to the action to be taken to grow a healthy crop. The approach is known in particular from rice growing in Southeast Asia, but is now being applied on a wide range of crops in other developing regions. It consists of participatory, nonformal adult education, and so far over 75,000 FFS have been conducted. The authors describe the FFS as an interactive way of deploying science. And, they note that scientific excellence and adherence to ecological principles provide a strong entry point for IPM development. The idea that farmers can and should empower themselves became the acknowledged motivating force among programme developers and field staff. Nowadays, across Asia, farmer empowerment continues to be the foundation of IPM activities. A key element is that the agroecological diversity of the different farmer's fields is taken into account for implementing effective plant health practices. The FFS ensure that the farmers obtain the necessary knowledge and experience for this.

The success of the IPM activities in Indonesia was helped, in particular, by effective policy support. Pontius et al. (10) describe this as follows. "Scientists were able persuade several ministers of the to ineffectiveness of intensive insecticide use (notably, the Department of Agriculture remained unconvinced). The scientists proposed an IPM programme based on (i) a farm-level IPM strategy, (ii) IPM training for technical personnel who would train farmers, and (iii) limiting the availability of broad spectrum insecticides. An inter-ministerial coalition supported the proposal and took it to the president. The result was Presidential Decree No. 3, 1986. The degree called for farmer and field worker IPM training, the banning of 57 broad-spectrum insecticides from use in rice production and the eventual elimination of subsidies for insecticides. The decree created a policy environment at all levels of government that ensured support for rice IPM implementation".

The FFS approach is based on four IPM principles (10) that provide a guide to what farmers should be able to do when they participate in an FFS. They are:

- grow a healthy crop;
- conserve natural enemies;
- conduct regular field observations; and
- become IPM experts.

Farmers have displayed an intellectual curiosity to understand rice agro-ecosystem ecological processes and an eagerness to formulate community-wide approaches to increase the impact of IPM. They are not only taking part in IPM activities, they are taking over IPM activities.

Pontius *et al.* stress that "IPM is a set of practical guidelines for how to best manage a specific crop. The rice IPM FFS learning approach, however, is based on well established theories, supported by good field data". This leads to the following description of IPM. "IPM is a set of practical guidelines, derived from a sound scientific understanding, used by farmers to grow a healthy crop".

It is noted that one of the biggest problems with many of the developments of IPM over the years has been the tendency to generalize and make recommendations for farmers across large and highly heterogeneous areas. The shift from calendar based applications to farmers as IPM managers places an increasingly large burden on the user in terms of ecological knowledge, observation and analysis. FFSs place the control of smallscale agro-ecosystems in the hands of people who manage them.

Getting IPM institutionalized at the field level required working through complex government bureaucratic systems. Centralized bureaucracies are not conducive to participatory IPM. Attention needs to be given to the highest and lowest levels of the system that provide the context for IPM field activities. Policy guidance from top levels can combine effectively with bottom-up pressure generated by farmers.

Schmidt et al. (11) have used the FFS approach for a review of success factors in integrated pest management. It is worthwhile to highlight a number of issues raised by these authors. They note that the feature common to natural resource management and IPM is not only that they represent examples of endeavours to implement sustainable agricultural practices, but also that they are all applied in very complex, and highly diverse, agro-ecological situations where any major change deserves a preceding, careful, holistic analysis. Consequently, the role of the farmer and his or her family in deciding to make changes needs to be included systematically into the entire process of searching and applying new solutions. Under these conditions traditional ways of extension hit the limits. The traditional extension approaches used during the Green Revolution were characterized by

- technologies developed by researchers on research stations;
- top-down transfer of technology by researchers to extensionists, and from these to farmers; and

blanket recommendations for large areas.

Schmidt *et al.* (11) note that, for a variety of reasons, nowadays the concept of economic damage thresholds is a point of disagreement among many IPM experts. They stress that the real breakthrough in IPM development and application in rice growing in Indonesia occurred when the ecological principles of IPM were merged with lessons learned from non-formal education and the FFS extension model was developed. The following important results are listed:

- in 1997 in Indonesia some 600,000 rice farmers had already attended FFSs. These farmers have become the owners of the programme;
- after having attended a FFS farmers apply 80 to 100 percent less insecticide;
- yields remain stable or increase even slightly, which is attributed to increased crop observations;
- reduced costs for inputs by stable yields imply improving benefits for IPM farmers;
- health hazards caused by pesticide application are reduced;
- environmental pollution is reduced;
- the national economy saves finances because pesticide subsidies are abandoned and less pesticides are imported;
- stable interest groups emerge from FFSs;
- FFS graduates spread the principles of IPM to their fellow farmers; and
- local government bodies contribute to the funding of FFSs.

The following success factors in extension for IPM are enumerated.

- (i) Extension and farmers: building confidence between farmers and extensionists, and raising farmers' selfconfidence and decision-making abilities.
- (ii) Extension and research: facilitating action research and furthering participatory technology development, and helping to perceive problems in a common manner.
- (iii) Extension methods and contents: integrating local knowledge and arriving at understandable explanations of complex problems.
- (iv) Broad impact of extension: farmer interactive extension, group approach and IPM as a movement leading to farmers becoming a critical mass.
- (v) Motivation of farmers: attractiveness to farmers and ownership.
- (vi) Extension staff: practical and continuous education and staff motivation.
- (vii) Political environment: favourable policy framework, awareness building with mass media and local situation analysis.
- (viii) Institutional set-up: selection of partners which are favourable to the work to be undertaken.

FFS have definitely proven to be effective tools for the introduction of IPM. Currently FFS are conducted in over 30 countries worldwide. A review of 25 impact studies on FFS leads to the following conclusion (14). "Generally, the case studies reported reductions, sometimes drastic reductions, in pesticide use attributable to the effect of training. There was also a general increase in yield due to the effect of training". In this review it is also noted that the exercise of agro-ecosystem analysis stimulated skills of thinking and communicating which could subsequently be applied to broader areas of people's lives.

## How to Further the Cause of IPM

The IPM research and development activities over the last 20 years have clearly shown the heterogeneity of the agricultural production systems. Thus, standard solutions can not be the answer to address the plant health needs of the individual farmers. A set of tools can be offered by extension staff, but the farmers themselves must be able to decide on how these tools can be used most effectively. The traditional extension methods of offering standard solutions are not suitable anymore. Thus, research and extension staff must in the first place understand the practical problems at the farm level to be able to jointly develop with farmers sustainable solutions. If the cause of IPM is to be furthered in an effective manner, then all persons and institutions concerned should adopt these principles.

IPM researchers have not facilitated the necessary transformation towards more effective farmer support systems. For example, the IPM definition keeps changing because of the evolving understanding of the key elements needed for producing a healthy crop. For an effective dialogue with all stakeholders it is important to define in clearly understandable terms what IPM wants to achieve. The goal should be to produce a healthy crop with minimum use of synthetic pesticides. Researchers need to better understand what it means to produce a healthy crop and how to do it.

A clear definition is also essential to better demonstrate the impact of IPM. For example van den Berg (14) in his review of FFS raises the question what do IPM initiatives attempt to achieve? Is the purpose to reduce insecticide use, to enhance sustainable pest management, or to enhance adaptive crop management? Is it to increase yields, to increase profits, or to improve livelihoods? In an information booklet of the Systemwide IPM Programme (12) it is noted that an impediment to the wider adoption of IPM is the lack of an adequate framework for evaluating the true costs and benefits of crop protection measures. In recent years little progress has been made in developing a new conceptual framework focusing on quantifying the opportunities for IPM interventions within defined biophysical the and policy environments or in gathering relevant data. Improved knowledge on these cost and benefit factors remain a challenge for the research community. This is a major task that still requires extensive research and development efforts.

The Systemwide IPM Programme considers that "an inappropriate regulatory

framework for registration of biopesticides in many countries has provided a special impediment to the development and use of these products in developing countries (12). The Environmental Protection Agency in the USA has established a tier approval system for biological pesticides. These have helped to lower the development costs of biopesticides, which are now estimated around \$5 million per product, compared to \$50-70 million for conventional pesticides. In 1997 the average time to register a biological pesticide was 11 months, compared to 38 months for pesticides (8). conventional However, registration requirements in most countries do not seem to offer effective conditions for new products developed within the IPM context. This merits to be analyzed in much more detail in order to convince authorities to adopt systems favouring IPM implementation.

Closer collaboration has to be established by researchers with extension and regulatory agencies to finally arrive at joint and appropriate approaches that address the above issues in an effective manner. Changes have to take place in a system that is still dominated by strong interest groups with outdated ideas. A real lobbying exercise will have to be undertaken. The IPM research community has little capacity to do this well. It will need effective partners that require clear facts showing what can be gained by growing healthier crops. This 2<sup>nd</sup> International Conference on Sunn Pest can be considered a real success if the promising research results obtained over the last years can be amalgamated into an effective IPM programme that is built on the above principles.

## الملخص

برادر، لوكاس. 2007. كيف يستطيع العلماء والبرامج العلمية الاتصال بالمشروع الحكومي والخاص لتعزيز فكرة المكافحة المتكاملة للآفات. الصفحات 17–24.

أظهرت بحوث المكافحة المتكاملة للآفات ونشاطات التطوير على مدى العشرين سنة الماضية بوضوح عدم تجانس أنظمة الإنتاج الزراعي. وبالتالي، فإن هكذا حلول قياسية يمكن تقديمها لتلبي حاجات صحة النبات للزراع الأفراد. يمكن أن يقدم موظفو الإرشاد مجموعة من الأدوات، لكن يجب أن يكون الزراع أنفسهم قادرين على تقرير كيفية استخدام هذه الأدوات بشكل أكثر فعالية. لم تعد الطرق الإرشادية التقليدية التي تقدم حلولاً قياسية مناسبة بعد الآن. نقدمت أبحاث صحة النبات بهذا الاتجاه بشكل متزايد، لكن يجب أن يكون الزراع أنفسهم قادرين على تقرير كيفية استخدام هذه الأدوات بشكل أكثر فعالية. لم تعد الطرق الإرشاد لم تقم بذلك. هناك استثناء يتمتل في طرائق مدارس الزراع الحقلية التي أثبتت فعاليتها. إن بشكل متزايد، لكن خدمة الإرشاد لم تقم بذلك. هناك استثناء يتمتل في طرائق مدارس الزراع الحقلية التي أثبتت فعاليتها. إن النقص في طرائق الإرشاد الم نقم بذلك. هناك استثناء يتمتل في طرائق مدارس الزراع الحقلية التي أثبتت فعاليتها. إن النقص في طرائق الإرشاد المناسبة هي معاناة إضافية لمتطلبات التسجيل لمنتجات وقاية النبات. لم يسهل باحثوا المكافحة المتكاملة للأفات الانتقال الضروري باتجاه أنظمة أكثر فعالية لدعم الزراع. فعلى سبيل المثال، يتغير تعريف المكافحة المتكاملة للأفات الانتقال الضروري باتجاه أنظمة أكثر فعالية لدعم الزراع. فعلى سبيل المثال، يتغير تعريف المكافحة المتكاملة للأفات بتعابير يمكن فهما بشكل واسع. الهدف هو إنتاج محصول سليم. إنه الوقت المناسب لتحديد ما هي المكافحة المتكاملة للأفات بتعابير يمكن فهمها بشكل واسع. الهدف هو إنتاج محصول سليم باستخدام الحد الأدنى من مبيدات الأفات المصنعة. على هذا المبدأ يجب تأسيس تعاون وثيق مع مؤسسات التشريع والإرشاد للوصول أخيراً إلى طرائق تشاركية مناسبة. المصنعة. على هذا المبدأ يجب تأسيس تعاون وثيق مع مؤسسات التشريع والإرشاد للوصول أخيراً إلى طرائق تشاركية مناسبة. يجب إحداث أدي لا يزل ما يدي لايواس التشريع والإرشاد للوصول أخيراً إلى طرائق تشاركية مناسبة. يجب إحداث أدييرات في الذات ورائق من موسات التشريع والإرشاد للوصول أخيراً إلى طرائق تشاركية مناسبة. المصنعة. على هذا المبدأ يجب تأسيس تعاون وثيق مع مؤسات التشريع والإرشاد للوصول أخيراً إلى طرائق منامية. يجب إحسان مالمسنة يوال في ونائل المنوعة بيفكا وديرا إلى ما ميكان وزان من ونتاج محا

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Green Muscle, from Isolate to Commercial Product

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#### Abstract

#### Kooyman, C. 2007. Green Muscle, from Isolate to Commercial Product. Pages 25-36.

Locusts and grasshoppers are still mainly controlled by means of chemical insecticides. However, progress has been made over the last 15 years to develop alternative means of control. A biological product based on the entomopathogenic fungus Metarhizium anisopliae var. acridum Driver & Milner has been developed over a period of 12 years by the LUBILOSA program, a French acronym meaning biological control of locusts and grasshoppers. The program was executed by CABI, IITA, AGRHYMET and GTZ with funding from the international development agencies of Canada, the Netherlands, Switzerland, Germany, the United Kingdom and the United States of America. The program brought together a group of researchers with a wide diversity of expertise to be able to go through all the necessary steps for the development of an effective and easy-to-use product. These steps included the search for isolates, bioassays to select the best isolates, field testing, (eco) toxicological testing, mass production and storage studies, formulation, application and commercialisation. One isolate was selected on the basis of its efficacy and ease of production. A licensing agreement was signed with the South African company Biological Control Products, which registered the product under the name Green Muscle®. Though the product is now on the market, full acceptance is still some way off. One important hurdle is registration, which has not yet been achieved in the majority of affected countries. Acceptance by the locust control establishment is also still problematic, which primarily has to do with the product's slow speed of kill. Lessons learned in the LUBILOSA program and possible ways forward will be discussed.

Keywords: Biopesticide, entomopathogenic fungus, locust control, Metarhizium.

## Introduction

Locusts and grasshoppers regularly cause extensive and serious damage to crops in many parts of Africa, Asia and the Americas. Locusts are well known for their potential of invading cropping areas in swarms of millions of individuals leaving behind devastated fields and plantations. Fortunately, these invasions are infrequent and may be followed by long periods of recession (21). However, grasshoppers form a more chronic problem in some areas causing serious yield losses in most years.

Locust and grasshopper control is currently carried out with chemical pesticides. For many years, the product of choice was Dieldrin, a persistent pesticide well suited for barrier treatment. However, concern about its negative impact on the environment caused it to be banned in most countries. Most modern pesticides replacing it, like Fenitrothion and Malathion, are much less persistent and have therefore to be applied more frequently in blanket treatments and in larger volumes. So, even though these products are less toxic than Dieldrin, their environmental impact may well be worse.

During the last major desert locust outbreak (1986-1989), donors spent more than US\$200 million and almost 15 million litres of pesticides were applied (1). Environmental pressure groups alerted donors to the side effects of this massive use of chemical pesticides (6). Donors then appealed to the scientific community to find more acceptable locust control methods. One of the submitted proposals led to funding of the LUBILOSA program.

## **Materials and Methods**

#### LUBILOSA

The LUBILOSA (LUtte BIologique contre les LOcustes et les SAuteriaux) program started in late 1989 as a collaboration between the then International Institute of Biological Control (IIBC, now CAB International Bioscience), Ascot, UK, the Biological Control Centre for Africa of the International Institute of Tropical Agriculture (IITA), Cotonou, Benin, and the then Département de Formation en Protection des Végétaux (DFPV, now Programme Majeur Formation) of AGRHYMET, Niamey, Niger. During its 12 year life, the program received funding from the Canadian International Development Agency (CIDA), the Swiss Agency for Development and Cooperation (SDC), the Dutch Directorate General of International Cooperation (DGIS), the UK Department for International Development (DFID), the German Society for Technical Cooperation (GTZ) and the US Agency of International Development (USAID). LUBILOSA's multi-disciplinary team developed a microbial pesticide based on the entomopathogenic fungus Metarhizium anisopliae var. acridum Driver & Milner, a naturally occurring grasshopper pathogen, which has been commercialised under the name Green Muscle®.

The development process went through a number of steps: pathogen surveys, isolate selection, field testing, formulation and application, development of mass production, storage studies, toxicological testing, ecotoxicological studies, socio-economics, commercial licensing and registration. However, the very first step was to decide on the kind of alternative to chemical control.

In the late 1980s, there were a number of possible options for locust and grasshopper control, though only those using chemical insecticides were in general use (Table 1). Since the objective was to find alternatives to chemical insecticides, only botanical insecticides, like neem oil, and natural enemies were considered. The former did have little environmental impact and were already being produced. However, it was difficult to imagine how sufficient numbers of trees could be planted to produce the huge quantities of product needed in times of large locust outbreaks. On the other hand, some pathogens could be mass produced.

Table 1. Grasshopper control options (1989).

Control option Examples	Characteristics		
Chemical insecticides			
Organophosphates (e.g. Fenitrothion)	moderately toxic to mammals, toxic to birds and fish, broad spectrum		
Pyrethroids (e.g. Deltamethrin)	low mammalian and avian toxicity, broad spectrum, insects may recover		
Insect growth regulators (e.g. Diflubenzuron)	non-toxic to vertebrates, affect aquatic arthropods, only effective against nymphs		
Botanical insecticides			
Neem oil (azadirachtin)	little environmental impact, slow action, low mortality, problems with quality (esp. a.i. content)		
Natural enemies			
Vertebrates	difficult to manipulate as control agents		
Insects	mass production too expensive		
Nematodes	species parasitising grasshoppers difficult to mass-produce		
Pathogens	some cause epizootics, some can be mass-produced on artificial media		

Grasshopper pathogens occur in the following groups:

- 1. Viruses: those isolated from grasshoppers not very virulent, production *in vivo*.
- 2. Bacteria: species infecting grasshoppers not effective under field conditions, no *Bt* varieties known that infect grasshoppers, production *in vitro*.
- 3. Protozoa: species infecting grasshoppers not effective under field conditions, production *in vivo*.
- 4. Fungi: virulent isolates available, new discovery that certain fungi (e.g. *Metarhizium, Beauveria*) can be formulated in oil, production *in vitro*.

An analysis of the known grasshopper pathogens showed clearly that fungi were the most attractive group to investigate. An African isolate of *Metarhizium* was already available at IIBC and one of its scientists had discovered that certain hyphomycete fungi are more effective when formulated in oil as opposed to water (19), which opened the possibility of using them in areas where water is scarce. It was therefore decided to concentrate efforts on this group of fungi.

Since locusts and grasshoppers are never exotic pests, classical biological control was not an option. Searching for an exotic fungus that might be very virulent in a new association with local grasshoppers, was considered, but such a fungus may not be as specific to the target insects as a co-evolved fungus. For that reason, the latter kind was deemed preferable for use as a biological control agent (18).

#### **Pathogen Surveys**

At the start of the LUBILOSA program, only 28 isolates of hyphomycete fungi from Orthoptera were available in international culture collections (ARSEF and CABI/IMI), and out of those only 3 were from Africa and none from the Near East. Surveys were conducted in West Africa, Madagascar, Oman and Pakistan, and over a period of 3 years, yielded 179 isolates from locusts and grasshoppers and 2 from crickets (20). The

great majority were isolates of *Metarhizium* anisopliae var. acridum, (originally identified as *M. flavoviride* (Gams & Rozsypal) originating from 13 countries. All isolates collected were stored in the culture collection of CAB International. Subsequently, more isolates have been discovered in other continents indicating a pan-tropical distribution extending into subtropical zones.

#### **Isolate Selection**

The pathogenicity of 159 isolates of Metarhizium and Beauveria was tested in bioassays using adult desert locusts, Schistocerca gregaria Forskål (2). A dose of  $7.5*10^4$  spores in 2 µl was applied under the pronotum and the locusts were then kept individually in containers until death. Pathogenicity was measured as median lethal time (MLT). Out of the 21 most virulent isolates, 17 belonged to M. a. var. acridum. The original isolate present at IIBC, IMI 330189, was among those. It had been discovered by a DFPV team in 1989 near Niamey, Niger, on the grasshopper Ornithacris cavroisi (Finot). Since it was not significantly less pathogenic than any other isolate and had shown good growth on artificial media, a single spore of this isolate was chosen for further development.

#### **Field Testing**

As soon as laboratory work had confirmed the potential of IMI 330189, field testing started. Trials progressed from semi-field (arena) trials (3) through small-scale to large-scale trials (9, 12, 14, 17) and finally operational trials (11, 10, 13). At the time of writing, trials had been carried out in 18 countries in Africa and one in Europe (Spain). Target species included the desert locust and other African locust species and several grasshopper species including the notorious Senegalese grasshopper, *Oedaleus senegalensis* Krauss.

At moderately high afternoon temperatures (28-35°C) and occasional overcast skies, 80-90% mortality was generally achieved in 2-3 weeks (Figure 1). Higher and lower temperatures caused mortality to be slower. Spores of *Metarhizium* persisted for weeks in Sahelian grassland with a half-life of around 7 days during the rainy season. Final mortality of grasshoppers caged on treated vegetation was >90% after 3 weeks and  $\pm 25\%$  after 7 weeks. On sparse vegetation and under very hot conditions, the half-life was shorter.



**Figure 1.** Mean grasshopper population density after application of *Metarhizium* at 100 g/ha and Fenitrothion at 250 g/ha on 50 ha plots (A) and 800 ha plots (B) (13).

The slow mode of action of *Metarhizium* combined with the high mobility of many of the target species caused some experimental problems. E.g., to be able to show the effect of the fungus on *O. senegalensis* in the field, 25 ha proved to be the smallest possible plot size. In the case of locust nymphs, hopper bands instead of fixed plots had to be the experimental units. The effect on adult locusts was very difficult to assess, but a team of three experienced locust specialists was able to make reliable population estimates.

#### **Formulation and Application**

Traditionally, fungal spores used to be suspended in water with or without detergent, but efficacy was often disappointing, to the point where people tended to give up on entomopathogenic fungi. However, in the 1980s, it was discovered that certain hyphomycete fungi can be formulated in oil, which greatly improves their efficacy (19). This enables the use of fungi in relatively hot and dry environments, where water is scarce and water-based formulations quickly evaporate. Once the spores have landed on the insect, ambient relative humidity is not very important. Oil formulations can be used in ultra-low volume (ULV) spraying equipment, which is commonly used in locust control and is more efficient than high volume equipment, especially for spraying large areas. Green Muscle can therefore be used in a variety of environments from desert to humid forest.

Initially, technical concentrate (TC, i.e. dry spores) was taken to the trial site and then suspended in suitable oil (e.g. 30% groundnut oil and 70% kerosene). However, spores did not remain long in suspension and settled on the bottom of containers, including the insecticide tanks of vehicles and aircraft. This caused variable active ingredient content of the spray and the concentrated suspension near the bottom tended to clog the filters. The presence of substrate residue exacerbated the clogging problem.

To solve these problems, a special spore extractor was developed that better separated spores from substrate particles and excluded chains and aggregates of spores. Dry Green Muscle powder now contains 99% single spores and very little substrate residue. In addition, an oil miscible flowable concentrate (OF) was developed, which keeps spores in suspension for a long time (patented). Clogging of filters does not often happen any more. The OF can be used in all available ULV equipment (hand-held, vehicle- or aircraft-mounted) after dilution with diesel or an oil/kerosene mixture. TC suspended in the oil/kerosene mixture can be used in hand-held sprayers, but the reservoir needs to be regularly shaken to prevent settling of spores.

Green Muscle is applied at 0.5-2 l/ha depending on the type of application, the lowest for aerial application and the highest for hand-held sprayers. The active ingredient dose varies with target species and density of vegetation:

- Zonocerus variegates L.: 12.5-25 g/ha
- Oedaleus senegalensis Krauss: 25-50 g/ha
- Hieroglyphus daganensis Krauss: 25-75 g/ha
- Schistocerca gregaria Forskål: 25-50 g/ha
- Locustana pardalina Walker: 25-75 g/ha

#### **Development of Mass Production**

For use in a biopesticide, a pathogen has to be mass-produced. For fungi that can grow on artificial media, there are two possible production systems. Liquid fermentation is relatively easy and cheap, and is attractive for very large-scale (industrial) production. However, the spores produced in such a system are blastospores, which have relatively thin outer walls and are not very resistant to adverse conditions. This restricts their use to humid conditions with little insolation.

LUBILOSA opted for a two phase system that produces robust aerial conidia (5, 7). The first phase is liquid and during this phase, the fungus grows in a nutrient broth containing a carbon and a nitrogen source (often sugar and yeast). After bulking up the biomass in this way, a solid substrate is inoculated with the broth containing the fungus. This substrate can consist of some form of cereal, clay granules or even cloth. The fungus then rapidly exhausts its nutrient supply and starts to sporulate. After drying the substrate for a number of days, the spores are extracted. As mentioned before, a special spore extractor has been developed to separate spores from the substrate and to produce a powder of single spores. At every step of the production process, it is essential to carry out quality control, especially to check for contamination with other, potentially toxic, fungi or human pathogens.

When developing mass production for *Metarhizium*, the question came up whether the program should concentrate on a low-tech system that could be applied by farmers in a kind of cottage industry. However, in the course of our work, it became clear that *Metarhizium* cannot compete well with saprophytic fungi, like *Aspergillus* and *Penicillium*. Since some of these fungi are greenish like *Metarhizium*, one needs a microscope to distinguish them. Poor farmers, who are the ones most often affected by grasshoppers, are not likely to invest in strict quality control measures to avoid producing unwanted fungi.

For that reason, LUBILOSA developed an intermediate technology, which was implemented in IITA's pilot fungus production plant. The technology is described in Jenkins *et al.* (7). High-tech fungus production technology has been developed by producers in Europe, North America and South Africa. Table 2 summarises the different levels of technology.

 Table 2. Fungus production at different levels of technology.

Tech	Investment	
level	level	Features
Low-	Low	Suitable for village
tech	investment	level cottage industry;
		High risk of
		contamination with
		torio funci
		toxic fuligi
Medium-	Moderate	Small batches of
tech	investment	substrate (plastic bags
		etc.);
		Little wastage in case
		of contamination
High-	High	Large batches of
tech	investment	substrate (industrial
		vessels);
		Prevention of
		contamination
		difficult and risk of
		large wastage

The cost of production at the IITA plant is roughly US\$200/kg (5). The product is made available at cost price for experimental purposes. The price charged at the factory gate by one commercial producer (BCP, see below) is around US\$350/kg. At the currently recommended dose rates, this works out at \$4.50 to \$26 per ha, which is high compared to conventional locust control products (\$5-15/ha). Efforts are underway to reduce this price through higher efficiency and largerscale production. If further reductions in dose rate are possible, this will also bring down the cost of application.

#### **Storage Studies**

The ready biopesticide product needs to be stored, first at the manufacturer, then at the local supplier and often by the end user as well. Locust outbreaks cannot be predicted long in advance and when they happen, large quantities of insecticide are needed. Such demand can usually not be met within the necessary time, so the only option is to keep a strategic stock. Even if the stock is rotated whenever orders come in, some product may have to be stored for several years.

The program investigated the best way to store *Metarhizium* for long periods. It turned out that dry spores kept best, provided that they contained less than 6% water (15). Spores are packed in hermetically closed sachets made of aluminium covered with plastic. The shelf life at various temperatures is as follows:

- $4^{\circ}C$ : >6 years
- <20°C: >3 years
- 20-30°C: ±1 year
- 30-40°C: ±6 months

The OF formulation can be stored for at least 6 months, if the temperature does not exceed  $30^{\circ}$ C.

#### **Toxicological Testing**

Any pesticide has to be assessed as to its effect on man and other non-target organisms. For this purpose, the US Environmental Protection Agency (EPA) has developed tiered testing, which is a cost-effective step by step experimentation that passes from laboratory tests through semi-field to field experiments. The first tier involves maximum exposure of experimental animals to the active ingredient. Some of the results of isolate IMI 330189 were as follows:

- Acute toxicity (rat): none
- Inhalation toxicity (rat): none
- LD50 (rat): >2000 mg/kg body weight
  - Infection: none
- Eyes, mucosa (rabbit): slight irritation often observed
- Allergy: rare
- Avian toxicity: no effects observed

If a test has a positive result, the product will undergo a similar test at a realistic dose on an organism kept at artificial conditions that resemble natural ones as much as possible (second tier). In case of another positive result, the product has to move to third tier testing of realistic doses under natural conditions, which usually takes the form of ecotoxicological studies.

#### **Ecotoxicological Studies**

Ecotoxicology examines the impact of introduced substances on the environment, e.g. whether the product gets into ground and surface water and affects aquatic organisms. The effect on soil micro-organisms and mesofauna is also studied as well as the effect on terrestrial vertebrates, bees, silkworms and other beneficial organisms. A number of studies have been conducted by the LUBILOSA program, including studies on non-target arthropods during large-scale applications in eastern Niger and in central and south-western Tanzania (16).

These studies concluded that Green Muscle has very low risk to parasitoid wasps and practically no risk to all other non-target taxa, (silkworms were shown to be susceptible in a tier 2 study). In contrast, Fenitrothion has a broad-spectrum effect on arthropods, especially ants, and is moderately hazardous to mammals. A modern insecticide with low toxicity to mammals, birds and fish, Fipronil, has a very broad-spectrum effect on arthropods, even at low doses, especially on termites.

#### **Host-Pathogen Ecology**

In the course of LUBILOSA's efficacy studies, it was observed that grasshoppers incubated in the sun took longer to die than those incubated in the shade. It was already known that some insects infected by pathogens raise their body temperature higher than normal by basking longer in the sun (or near a light bulb). Our investigations found that locusts and grasshoppers do the same (4) (Figure 2). Normally, they raise their body temperature to around 38°C, if weather conditions allow. A high body temperature is even more important for infected locusts, because Metarhizium stops growing above 35°C. Such individuals spend therefore more time basking than their healthy counterparts, often at the expense of feeding. This is the main reason why infected locusts and grasshoppers consume less.

A consequence of this behavioural fever response is that in the tropics, the fungus does not grow during a significant part of the day, unless the sky is overcast. Typically, the fungus grows for a few hours in the morning and the evening and shuts down during the middle of the day. At night, growth slows down considerably if the temperature drops below 20°C. For this reason, Green Muscle takes weeks to bring about a significant population reduction.

#### Socio-economics

Socio-economic studies were carried out to find out whether farmers would accept a product like Green Muscle (8, 22). The slow mode of action of the product was initially seen as a serious obstacle. However, grasshoppers are often treated before they move into the crops, and even if they are in the crop, consumption of infected hoppers gradually decreases.



Figure 2. Observed body temperatures of brown locust nymphs, *Locustana pardalina* Walker, against ambient temperatures in the Karoo, South Africa (4).

Farmers greatly appreciated the fact that Green Muscle is safe for them and their livestock. However, though they acknowledged that they would save on health care compared to chemical products, the price of Green Muscle was considered too high. Fortunately, recent trials have demonstrated that the originally recommended dose rate can be halved, so that the product has now become much more affordable. Currently, we are testing mixtures of Green Muscle with small doses of pyrethroids to provide some knockdown effect, which will allay concerns about slow mortality.

#### **Commercial Licensing**

At some point, it was felt that the product would only succeed if it was licenced to a commercial company. Large companies had by that time lost interest in biopesticides, so the program searched for small- to mediumsized companies, preferably in Africa. Only one suitable company existed in Africa, Biological Control Products SA (Pty) Ltd. (BCP) of Ashwood near Durban, South Africa. Because of its location, it was offered a licence for southern, eastern and north-eastern Africa and the Middle East. Another company, Natural Plant Protection (NPP) of Noguères, France, was offered a licence for West Africa and Europe.

BCP started commercial production in 1998 after registering Green Muscle in South Africa. NPP, through its mother company Calliope, obtained a temporary sales permit for Green Muscle in the CILSS countries, but did not manage to produce it with their production process. A new production plant has been constructed in Dakar, Senegal, by Fondation Agir pour l'Education et la Santé, which obtained a licence in early 2007.

#### Registration

In most African countries, there are no guidelines for the registration of biopesticides, but the existing procedures for the registration of conventional synthetic pesticides have limited value. There are, however, guidelines in the OECD, US, EU, FAO, South Africa and CILSS. In Kenya and Madagascar, they are at an advanced stage.

The current situation is that BCP has registered Green Muscle in South Africa, Namibia, Sudan, Tanzania and Zambia, while registration is in progress in Algeria and Yemen. Calliope tried to register the product in the CILSS countries, but was only granted a temporary sales permit. The FAO has placed Green Muscle on the list of recommended locust control products, but this does not seem to facilitate the registration process at all.

#### Discussion

The LUBILOSA program succeeded because it integrated the input of collaborators from many disciplines. Because of its professional approach, it managed to attract funding for 12 years. It was the first program of its kind and had to learn certain things the hard way. Similar projects can learn from this. One of the things we learned was the importance of the search for suitable isolates. Though the first isolate in our possession was chosen in the end, this was sheer luck. Only about 30% of the 159 isolates screened were worth a second look. Even some of the isolates from the target insect were not among the best. Moreover, pathogenicity should not be the only selection criterion. For a pathogen to be used in a biopesticide, it has to be easy to grow on artificial media.

Any pesticide will fail if it cannot reach its target. The right formulation and application technique are therefore essential ingredients in the development of a biopesticide. Biologists often tend to neglect these issues. LUBILOSA demonstrated the advantage of formulating spores in oil. Though this may not be appropriate in all situations, it is well worth considering it in the beginning. Apart from enhancing infectivity, oil formulations are better suited to ULV application, which minimises the volume to be sprayed making control operations more efficient and cost-effective. The drawback of ULV application is that it depends on a certain wind speed (ideally between 2 and 5 m/s). This can be overcome to some extent by using mistblowers with ULV attachment. In some cases, oil/water emulsions can be a solution.

A pathogen that cannot easily be produced will not make an affordable biopesticide and will at best be used in small niche markets. For that reason, those that can grow on artificial media are the most attractive. Many people like the idea of farmers producing their own pathogens. However, this is only advisable if there is little danger of contamination with toxic microorganisms or human pathogens. In most cases, it will turn out to be better to leave production to the specialists, i.e. commercial producers. Nowadays, there are a fair number to choose from. Unfortunately, production of most pathogens is not cheap. In the case of fungi, it will be difficult to produce them for less than \$300/kg. That often makes it difficult to compete with chemical products, if those are available for the target insect. It is therefore important to reduce the application dose as much as possible.

The shelf life of the final product should be long enough at ambient temperatures. For commercial success, it should be possible to keep it for at least two growing seasons, preferably without the need for a cold room. In dried form, that is possible for many fungi and other micro-organisms. And at 4°C, many can be kept for years. There may, however, often be a need for cold-chain distribution.

Some *Metarhizium* isolates produce toxins and have undesirable side-effects. It is therefore advisable to test promising isolates fairly early in the development process in order not to waste resources on an unusable isolate. Infectivity studies on beneficial nontarget arthropods should also be carried out early enough. If the chosen isolate does infect some beneficials, this problem can usually be managed rather than that the isolate is abandoned.

Host-pathogen ecology is an important issue. As the LUBILOSA experience shows,

the behaviour of infected insects is likely to be different from healthy ones. The changed behaviour may work in favour of the control effort (like reduced consumption) or against it (like behavioural fever).

Though the LUBILOSA program achieved its goal of developing a safe and easy-to-use product, it did not manage to create a market for it. In my opinion, the biggest obstacle at the moment is registration. The one company that is producing Green Muscle does not have the resources to push registration in many countries at once. Fortunately, a producer has now been found in West Africa, a part of Africa where there is some demand from the market.

A related problem is the current sales price of the product. Even if the recommended dose is reduced, the price will still be difficult to afford for many end users. It is hoped that larger orders will lead to increased production capacity and subsequent reduction in price. However, that may take some years, because any company has to recover the costs of registration, which are substantial.

In the meantime, most sympathetic observers believe that subsidised prices are the only sure way of getting Green Muscle accepted by the market in the short term. Unfortunately, this is a difficult proposition in today's economic policy climate. Still, I hope that African governments and international donors will consider it for the sake of the people's health and that of the environment. It is ironic that development agencies that spent \$17 million on the development of a technically successful product, are now reluctant to help making it a commercial success. With this in mind, programs developing similar products should probably go into partnership with a commercial producer quite early on.

International environmental pressure groups should also take notice, if they are serious about minimising the use of chemical pesticides. They were at the origin of the ban on Dieldrin and the drive for alternatives to chemical insecticides but seem to be absent from the debate now, at least as far as Africa is concerned.

A practically identical product, Green Guard<sup>®</sup>, is on its way to becoming a success in Australia, where it has been adopted by the Australian Plague Locust Commission for wide-spread use. What worked in its favour

was that there was a strong demand for a biological product by organic sheep farmers, who basically refused the use of chemical insecticides on their huge farms. This made the government take notice and facilitated registration in a continent-sized country where a lot of locust control is taking place.

## الملخص

## كويمان، كريستيان. 2007. مركب Green Muscle من العزلة إلى الإنتاج التجاري. الصفحات 25-36.

لا يزلل الجراد والجنادب يكافحان بواسطة مبيدات الحشرات الكيميائية. ولكن، خلال 15 سنة الأخبرة تم إحراز تقدم في تطوير وسائل بديلة للمكافحة، حيث تم على مدى 12 عام تطوير منتج حيوي يعتمد على الفطر الممرض للحشرات Metarhizium anisopliae var. acridum Driver & Milner (وهي كلمة فرنسية مركبة تعني المكافحة الحيوية للجراد والجنادب). نفذ البرنامج من قبل Metarhizium anisopliae var. acridum Driver & Milner و وسائل بديلة للمكافحة، حيث تم على مدى 12 عام تطوير منتج حيوي يعتمد على الفطر الممرض للحشرات Metarhizium anisopliae var. acridum Driver & Milner (وهي كلمة فرنسية مركبة تعني المكافحة الحيوية للجراد والجنادب). نفذ البرنامج من قبل AGRHYMET ، IITA، CABI و GTZ بتمويل من مؤسسات التتمية الدولية في كندا، هولندا، سويسرا، ألمانيا، المملكة المتحدة والولايات المتحدة الأمريكية. ضم البرنامج مجموعة من الباحثين ذوي خبرات متنوعة ليكونوا قادرين على تجاوز كل المراحل الضرورية لتطوير منتج فعال وسهل الاستخدام. شملت هذه المراحل البحث والتولية في كندا، هولندا، سويسرا، ألمانيا، المملكة المتحدة والولايات المتحدة الأمريكية. ضم البرنامج مجموعة من الباحثين ذوي غرات متنوعة ليكونوا قادرين على تجاوز كل المراحل الضرورية لتطوير منتج فعال وسهل الاستخدام. شملت هذه المراحل البحث والتولية في العزلات، الاختبارات السمية، دراسات الإنتاج الكمي عن العزلات، الاختبار الحقلي، (coc) اختبارات السمية، دراسات الإنتاج الكمي والتخزين، التشكيل، التشكيق والاستغلال التجاري. تم اختبار عزلة واحدة على أساس فعاليتها وسهولة إنتاجها. ومن ثم تم توقيع أن المنزين، التشكيل، التطبيق والاستغلال التجاري. تم اختبار عزلة واحدة على أساس فعاليتها وسهولة إنتاجها. ومن ثم تم توقيع أن المنزين، التشكيل، التشيق والاستغلال التجاري. تما عزبي غليونية، التي محبولة في جنوب أفريقيا، التي سجلت المنتج تحت اسم مع شرك تم توقيع أن المنتج في الأمريون، التشكيل، الذي في قبوله بشكل كامل لا يزال صعباً. إحدى العقبات المهمة هي التسجيل، الذي لم يماع ورغم أن الفنتج في الأسان فعاليدان المصابة بهذه الحشرات. كما أن يلقبول من قبل مؤسسة مكافحة الجراد هو أبيقا. والذي والذي في معظم البلدان المصابة بهذه الحشرات. كما أن يلقبل من قبل مؤسسة مكافحة الجراد هو أبيلا منطو على مشاكل، والذي ويم معظم البلدان المرابي.

كلمات مفتاحية: مبيد آفات حيوي، فطر ممرض للحشرات، مكافحة جراد، Metarhizium.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.



## Sunn Pest Status in Iraq

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## Abstract

Hama, N.N., Z.A. Stephan, M.A. Ali and M.L. Aboud. 2007. Sunn Pest Status in Iraq. Pages 39-43.

Since 1989 Sunn Pest, Eurygaster integriceps Puton has been considered an economic pest on wheat and barley grown under rain-fed regions in the northern governorates. From seventies to nineties of the past century a number of new wheat and barley varieties (Mexipac, Abu-Ghraib, IPA 95, 99 & Al-Iraq), were introduced to a large area in central and southern governorates as a part of a national plan to reach food self sufficiency. Consequently, the pest infestation map had changed dramatically because of Sunn Pest invading most wheat and barley grown on irrigated areas in central, middle Euphrates and southern governorates. Thousands of hectares of wheat were sprayed 2-3 times /season in Diwaniya and Al-Najef governorates from 2002-2004 due to the severity of the infestation. Field observations and research data, confirmed the hibernation of Sunn Pest under date palm leaf base, blade weeds and rice hay during winter and early spring. This paper will discuss relevant postulates as an attempt to explain this new situation. Keywords: Central, middle Euphrates & southern governorates, Sunn Pest, wheat.

## Introduction

Iraq has been considered by many authors as among the middle eastern countries which suffered through time from serious damage to its cereals annual production by Sunn Pest, Eurygaster integriceps Puton (7). This is true for wheat and barley grown on rain-fed areas. But the infestation map has changed dramatically from1980-2000 and beyond. During which Sunn Pest colonized wheat and barley fields grown in irrigated regions at most central and middle Euphrates governorates such as Salahaldin, Baghdad, Diyala, Al-Anbar and Babil.

In fact, research data have documented its hibernation and infestation at the above regions (1, 2). From 2001-2004 a number of Sunn Pest outbreaks occurred in Al-Najef, Diwaniya (previously known Qadisya) and Karbala (Personal communication and Internal reports).

Several constraints were facing our national program against Sunn Pest such as total reliance on aerial spraying of broadspectrum insecticides. From 1983-2003 there was a period of recession and scientific blockade, and there was a serious lack of knowledge regarding ecology, biology behavior, distribution and quantitative data relevant to the economic threshold of the Sunn Pest; and country strived to reach national food sufficiency. This led to the introduction of new wheat and barley varieties whose crop characteristics were apparently in favor of the pest growth and development. Several research data have indicated the positive correlation between cultivation of some early maturing wheat and barley varieties and occurrence of Sunn Pest outbreaks (4, 5, 6).

This paper is an attempt to answer the question raised by the Ministry of Agriculture by extrapolating a logical explanation for the dispersal and colonization of cereals grown in most central, middle Euphrates and southern governorates by Sunn Pest. Our emphasis will primarily focus on the introduction and cultivation of new cereal varieties and the role of some cultural practices by farmers and other relevant factors.

## **Material and Methods**

Due to the great difficulties we are facing in conducting any research, this paper was designed to gather data concerning the subject matter depending on the followings: a). by reviewing all available local sources of information such as annual national reports, Ministry of Agriculture & Atomic Energy internal reports and personal communications. As well as we were able to collect specific data regarding the newly introduced wheat and barley varieties such as total cultivated area, maturity classification etc.; b). reviewing abstracts, reports and research paper through the internet; and c). conducting field surveys to evaluate the status of the problem in some target governorates.

On April 2004, a field survey was conducted on barley and wheat fields in four governorates: Diwaniya, Al-Najef, Karbala, and Babil during harvest. Plant protection specialists in each agriculture office who are familiar with the history of pest in their areas were consulted. Several unsprayed wheat fields were scouted with the assistance of a square meter wooden frame. Sampled areas were randomly selected (5-10 locations/field) and insects visually counted/observed. In the laboratory the different stages of the pest were identified and recorded.

## **Results and Discussion**

Our infestation map showed Sunn Pest chronological changes starting in 1989 which was the official declaration date that *E. integriceps* was considered as an economic pest on cereals grown in the northern governorates: Mosul, Arbil, Dehok and Sulaimania. From 1990-1998 it has expanded its original ecological zone into other governorates namely, Salahaldin, Diyala and Baghdad, while from 1999- 2004 it moved further toward central and mid Euphrates governorates such as Babil, Kerbela, Najef and Diwania.

During 1970-1990 the only wheat cultivar grown on irrigated region was Mexipac since the country demand for bread wheat was depending on wheat varieties grown under rain-fed conditions. These were located in the northern part of the country. The first change in the infestation map has been documented in Table 1, which shows the infestation and hibernation of Sunn Pest over most central and western governorates, Salahaldin had the highest insect field and in hibernation densities; 1.93 and 11 insects/m<sup>2</sup> respectively followed by Diyala, Baghdad, Al-Anbar and Babil governorates which had less Sunn Pest in fields and hibernation densities. This was the case until 2001when a dramatic change took place regarding Sunn Pest infestation zones. In that year E. integriceps became an economic pest in Diwaniyia and Al-Najef governorates with an average field density of 10 insects/ m<sup>2</sup>. On year 2002 the infestation spread into the neighboring governorates of Babil and Karbala with an average density of 0.2 and 2.5 insects/m<sup>2</sup> respectively.

**Table 1.** Invasion of wheat fields by Sunn Pest inselected central governorates (1991-1996).

	Field Density (Ins	l sect/ m²)	Hibernation density (insect/ m <sup>2</sup> )
Governorate	Range	Х	Range
Salahaldin	0.70-1.93	1.12	3-11
Diyala	0.60-1.20	0.69	3-5
Baghdad	0.02-0.42	0.28	3-6
Al-Anbar	0.08-0.33	0.24	4-5
Babil	NA	0.01	0.0
Data extrapolated	from Al-Khafa	ıji (1).	

Despite the chemical spraying program the problem continued during the following years (2003-2004) on both Diwaniya and Al-Najef with an average densities from 2.5 to 7.6 and 2 to 3 insects/m<sup>2</sup> for year 2003 and 2.5 and 0.5 to 1.0 insects/ m<sup>2</sup> for year 2004, respectively. Consequently aerial spraying of broad spectrum insecticides was conducted over many of the infested areas (Ministry of Agriculture internal reports and pers.commun.).

To investigate the situation further data presented in Table 2. showed more insight to the problem. Sunn Pest density for unharvested wheat field was 5.5, 4.4, 0.4 and 0.2 insect/  $m^2$  for fields in Al-Najef, Diwaniya, Karbala and Babil respectively. Moreover, insect population structure was 15 to 17.25 and 58-60 % for 3<sup>rd</sup> instar nymphs, adults and 4-5<sup>th</sup> instars nymphs respectively.

In fact data from Table 3 have indicated that these wheat and barley varieties such as IPA 99, Abu-Ghraib, Al-Iraq, Mexipac, were widely cultivated and since they are as medium to late maturing cultivars as well as their agronomic characteristics all together favor, completion of Sunn Pest life cycle. The major new wheat and barley varieties cultivated over most central, middle Euphrates and southern governorates for last 10 years such as IPA 95, IPA99, Abu-Ghraib, Tamooz 2, Al-Iz, Triticalea and Sameer were highly susceptible. Their agronomical and morphological characteristics such as spikelets density, glumes thickness and leaf pubescence are in complete harmonization with the insect life span.

As pointed out earlier the main goal of this paper was to identify the most logical reasons behind the unexpected invasion and out break over most cereals field grown over central, middle Euphrates and some southern governorates by Sunn Pest.

After reviewing available international scientific literature and local relevant research data, reports as well as extrapolation of our data we come to believe that (probably) a set of interacting environmental factors was responsible for the new situation. Our data suggest the following:

A. Country efforts during 1990s to achieve bread wheat sufficiency led to development and cultivation of high yield cereal cultivars. These varieties agronomic characteristics; medium to late maturity

such as IPA 95,99 (wheat and barley), Abu-Ghraib, Tamooz 2, Al-Iz, Al-Iraq and Sameer (Table 3). Other crop properties for these cultivars such as spikelets density, glum thickness and other morphological characteristics could make these varieties more susceptible to pest attacks. Relevant Research data acknowledged the positive correlation between several morphological and agronomic characteristics of the cultivars such as earliness of crop, spikelets density, richness and leaf pubescence and susceptibility of the cultivar to Sunn Pest attack (7). Furthermore, in a two years study by Rezabeigi et al (8) (25 wheat varieties) reported that whole wheat varieties Baiat, Azati, Gholestan, Karadj and Navid were the most resistance and Rashid, Altar, Sadari, Inia, Zardak, Tabusi and Omid seemed more susceptible to Sunn Pest infestation. Moreover, there are significant differences among Varamin and Kermashah populations of Eurygaster both in size and weight and their degree of damage to wheat varieties.

B. Our farmers cultural practices such as cultivation of the same variety on the same area repeatedly for long periods without crop rotation and restricting their cultivation to only land near orchards (hibernation sites), delaying crop harvest and growing both barley and wheat on the same area. All these factors support Sunn Pest survival and completion of their life span. Several researchers studied the relationship between these factors and Sunn Pest biological features. For instance in barley, (5) reported that Sunn Pest completed its life cycle at harvest time. Radjabi (4) in his analysis of Sunn Pest periodic outbreaks pointed out that late and slow harvest led to rapid increase in pest population and consequently a new outbreak. Half of these main wheat and barley cultivars were developed through stimulation of mutation by radiation and the remaining were developed through

classical breeding program. Most of these cultivars were selected primarily for their high yield and drought logging resistance, thus farmers only motivated to grow varieties with additional price incentives. Thus out of 32 registered wheat and barley varieties only several cultivars such as IPA 95,99, Abu-Ghraib, Tamooz 2, Al-Iz, Al-Iraq, Sameer and Triticale are the most widely distributed and cultivated by farmers over thousands of hectares for the last 10 years (Table 3).

**Table 2.** *Eurygaster integriceps* density on unsprayed wheat fields in several middle Euphrates and southern governorates at harvest time, 2004 season.

				Pop. Structure (%)				
	Density (ir	usects/ m <sup>2</sup> )				Ν	lymph	
Governorate	Range	Х	Cultivars	Adult	1	2	3	4-5
Diwaniya	0.0 18.0	4.42	IPA-99 Al-Iraq	25	-	-	17	58
Al-Najef	3.0-9.0	5.5	Mexipac	25	-	-	15	60
Karbala	0.0-1.0	0.4	IPA-99 Mexipac	-	-	-	-	-
Babil	0.0-1.0	0.2	IPA-99 Mexipac Abu-Ghraib	-	-	-	-	-

Table 3. Wheat and barley varieties cultivated in different central and middle Euphrates (1997-2001).

Wheat & Barley		Maturity			
Varieties	1997/1998	1998/1999	1999/2000	2000/2001	Class
IPA-99	42060	35640	379110	203850	Late
Abu-Ghraib	12570	15750	175680	223380	Medium
IPA-95	17910	15150	134610	150510	Medium
Tamooz-2	7620	34380	104370	59010	Late
Intesar	95190	4290	570	750	-
Tamooz-3	2520	8010	7750	720	-
Hashimiya	1050	1110	9900	2610	-
Rabeeae	5790	6570	4920	9750	-
Al-Iz	1050	2940	10470	56070	Late
Lm-Rabeeae	2550	1470	7020	33120	-
Wahat Al-Iraq	4140	1710	3360	28650	-
Adnaniya	1020	1470	6600	20940	-
Aratoom	1800	5340	1500	5730	-
Mexipac	2640	720	5100	2850	-
Noor	1350	3060	3330	2250	-
Latefiya	360	960	2580	2250	-
Tahady	570	2070	600	-	-
Al-Gaid (Al-Iraq)	90	570	1680	13350	-
Saly	2190	570	-	-	-
Triticaly (barley)	-	-	-	-	Late
IPA-99 (barley)	-	-	-	-	Late
Sameer (barley)	-	-	-	-	Medium

Source; State Board for Seed Certification 2002 Annual report.

Data regarding cultivated area is in donum=2500 m<sup>2</sup>.

C. Sunn Pest biological versatility enables the insect to adopt and colonize and hibernate in a new environment (1, 2, 3). Other environmental changes may also play an

additional role such as temperature and relative humidity and destruction of the surrounding range and bush land but these factors are beyond the scope of this paper.

#### الملخص

حما، ن. نزار، زهير أ. اسطيفان، محمد أ. علي ومزهر ل. عبود. 2007. وضع حشرة السونة في العراق. الصفحات 39–43.

منذ 1989، اعتبرت حشرة السونة أفة اقتصادية على القمح والشعير المزروع تحت الظروف المطرية في المحافظات الشمالية. من السبعينيات إلى التسعينيات من القرن الماضي أدخل العديد من أصناف القمح والشعير الجديدة (مكسيباك، أبو غريب، إباء 95، إباء 99 والعراق) إلى منطقة كبيرة في المحافظات الجنوبية والوسطى كجزء من خطة وطنية للوصول إلى الاكتفاء الغذائي الذاتي. بعد ذلك، تغيرت خريطة الإصابة بالآفة بشكل ملحوظ بسبب غزو حشرة السونة لمعظم حقول القمح والشعير المزروعة في المحافظات الجنوبية والوسطى كجزء من خطة وطنية للوصول إلى الاكتفاء الغذائي الذاتي. بعد ذلك، تغيرت خريطة الإصابة بالآفة بشكل ملحوظ بسبب غزو حشرة السونة لمعظم حقول القمح والشعير المزروعة في المناطق المروية في المحافظات الجنوبية والفرات الأوسطى كجزء من خطة وطنية للوصول إلى والشعير المزروعة في المناطق المروية في المحافظات الوسطى، الجنوبية والفرات الأوسط. تم رش آلاف الهكتارات من القمح والشعير المزروعة في المناطق المروية في المحافظات الوسطى، الجنوبية والفرات الأوسط. تم رش آلاف الهكتارات من القمح المعير المزروعة في المناطق المروية في المحافظات الوسطى، الجنوبية والفرات الأوسط. تم رش آلاف الهكتارات من القمح الشعير المزروعة في المناطق المروية في المحافظات الوسطى، الجنوبية والفرات الأوسط. تم رش آلاف الهكتارات من القمح المعير المزروعة في المناطق المروية في المحافظات الوسطى، الجنوبية والفرات الأوسط. تم رش آلاف الهكتارات من القمح والشعير المزروعة في المناطق المروية والنجف خلال الفترة ما بين 2002–2004 بسبب الإصابة الشريدة. أثبتت الملحظات الحقلية وبيانات البحوث، أن البيات الشتوي لحشرة السونة هو تحت قاعدة ورقة نخيل التمر، الأعشاب العريضة الأوراق وقش الحقلية وبيانات المحوث، أن البيات الشتوي لحشرة السونة هو تحت قاعدة ورقة نخيل التمرء الوضع الحديد. الأوراق وقش الحقلية والمرات الأوسل هذه المقالة الفرضيات المتعلقة بذلك كمحاولة لشرح هذا الوضع الحديد. ألوراق وقش الرز خلال الشتاء والربيع المبكر . ستناقش هذه المقالة الفرضيات المتعلقة بذلك كمحاولة لشرح هذا الوضع الحديد.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Sunn Pest Management in Romania

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#### Abstract

Popov, C., A. Brabulescu, F. Muresanu, S. Vasilescu, F. Gogu and I. Dobrin. 2007. Sunn Pest Management in Romania. Pages 45-60.

*Eurygaster integriceps* Puton is one of the most important pests of wheat in Romania. Investigations were done on the key elements of the integrated control of Sunn Pest. The elements were: the ratio between species and their distribution areas in Romania, the size and space distribution of Sunn Pest populations, the study of the annual peculiarities of their life cycle, the relationships between pest and oophagous parasites, and the establishment of the economic damage thresholds and means for their control. *Eurygaster integriceps* infests about 1,000,000 ha of wheat in 24 districts. The contribution of the oophagous parasitoids *Telenomus chloropus* Thomson and *Trissolcus grandis* Thomson, and the economic damage thresholds were calculated differentially in accordance with the crop stage and harvest destination. Forecasting is a major concern in Romania and procedures are based on a methodology developed by ARDI Fundulea, and applied across the country by the District Inspectors for Plant Protection and Phytosanitary Quarantine. Chemical control of Sunn Pest in Romania is supported by the Government.

Keywords: Eurygaster integriceps, Sunn Pest, wheat IPM.

## Introduction

*Eurygaster integriceps* Puton is one of the most important pests of wheat in Romania (2, 5, 6, 8, 9, 24, 27, 29, 30, 33, 39). In damaging areas of *E. integriceps*, security of wheat harvest quality is inconceivable without some energetic protection measures (3, 11, 31, 32, 38).

Investigations covering a great number of years tried to underline the contribution of some elements in outlying the integrated control concept for Sunn Pest. Thus, the following elements have been examined: ratio between species and their distribution areas in Romania, size and space distribution of Sunn Pest populations, study of annual peculiarities of their life cycle, relationships between pest and oophagous parasites, establishing the economic damage thresholds and the means for their control. Thus, the share of various elements has been established, and namely prevalence of *E. integriceps* has been proved, this covering a damaging area of about 1.000.000 ha of wheat in 24 districts: parasites contribution of oophagous (Telenomus chloropus Thomson and Thomson), and the Trissolcus grandis economic damage thresholds, as calculated differentially in accordance with crop vegetation stage and harvest destination. Chemical control of Sunn Pest in Romania is supported by Government, conform Main Undertakings Regarding Organization and Development of Nationwide Campaign of Sunn Pest Control in Romania (17, 18, 25, 39).

#### **Material and Methods**

Investigations extended to the whole country for a great number of years and were aimed at various aspects playing a role in the dynamics of Sunn Pest populations. Ratio between *Eurygaster* species as established in the recent decades on about 15,000 specimens annually has been compared to data published by Montandon (4) and Radulescu & Gruita (40).

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Table 1. Main undertakings regarding organization and development of the national campaign for Sunn Pest control in Romania.

No	Performed action	Execution term	Coordination
1	Establishing the Program for Sunn Pest controlling in Romania, based on wheat cropped surfaces in the damaging areas and on diapausing Sunn Pest population levels	January	MAFRD - CLPQ; ARDI
2	Development of Methods to pursuit the Sunn Pest life-cycle in representative areas within the districts included in the damaging area	January	MAFRD - CLPQ; ARDI
3	Estimation of pesticide amount and means of application needed, depending on the size of forecasted action to be performed	February	MAFRD - CLPQ; ARDI
4	Centralized training (of the all personnel which will participate in the control action of Sunn Pest in the damaging area)	March	MAFRD - CLPQ; DIPPPQ; ARDI
5	Providing necessary insecticides (according to estimated areas) and funds assigned by subsidies	March- April	MAFRD - CLPQ; DIPPPQ
6	Recording biological reserve of Sunn Pest at the end of overwintering by spring samplings in diapausing sites in forests	April 5th-20th	DIPPPQ
7	Designing the surfaces having to be treated against the overwintering adults, according to percentage of mortality during diapause of Sunn Pest populations	April 20th-25th	MAFRD - CLPQ; ARDI
8	Providing terrestrial and aerial means for applying treatments against the Sunn Pest	April	DIPPPQ; Service aircraft; Agricultural producers
9	Checking, identification and homologation of airfields for airplanes and helicopters participating in the control activities, having in view the most efficient use of these	April	MAFRD - DIPPPQ; Specialized units
10	Recording migration of overwintering adults from forests to crops (in representative areas of every district) according to the method established, in view of releasing warnings	April-May	DIPPPQ
11	Recording densities of overwintering adults in wheat crops; designation of wheat plots where chemical treatment is imposed	May, 1st-5th	DIPPPQ; Agricultural producers
12	Release of warning bulletins for applying treatments against the overwintering adults	Dependent on migratory evolution	CLPQ; DIPPPQ
13	Performance of treatments against the overwintering adults	Warned period	DIPPPQ; Agricultural producers
14	Assessment of the treatment efficacy and operative report	After two days	CLPQ; DIPPPQ
15	Observation of pest life-cycle in representative zones of every district, in order to warn treatments against nymphs according to the recommended method	May-June	MAFRD - CLPQ; DIPPPQ; ARDI
16	Estimation of parasitization level of Sunn Pest eggs by oophagous parasitoids, collecting and preservation of biological material in order to identity the species	June	DIPPPQ; ARDI
17	Estimation of surfaces to be treated against the nymphs, depending on adults fecundity, parasitization and climate factor evolution	June	DIPPPQ

#### Table 1 (Cont.)

18	Release of warning bulletins for applying treatments against nymphs	Dependent on pest evolution	MAFRD - CLPQ; DIPPPQ
19	Finishing-off the nymph control program by establishing wheat plots to be treated, according to crop destination (food or seed) and pest density (economic damage threshold)	June	DIPPPQ
20	Performing treatments against nymphs	Warned period	DIPPPQ; Agricultural producers
21	Assessment of treatment efficacy and operative report	After two days	MAFRD - CLPQ; DIPPPQ; ARDI
22	Recording the new Sunn Pest adult populations level in crops before harvest: action represents a first step of forecast for the next year	Prior to harvest	DIPPPQ; ARDI
23	Harvesting wheat in the damaging area as rapidly as possible, before the end of complete feeding of adults, as a technological measure to diminish Sunn Pest populations	July	Agricultural producers
24	Evaluation of pricked grain percentage in wheat harvest in the damaging area	July- August	MAFRD - CLPQ; DIPPPQ
25	Performing autumn samplings to establish the biological reserve of Sunn Pest in diapausing sites	October	DIPPPQ
26	Presentation of the Report regarding the way of achievement of Sunn Pest control program by districts	December	DIPPPQ
27	Evaluation of Sunn Pest population affecting wheat crops in the next year, based on overwintering adults density/m <sup>2</sup>	December	MAFRD - CLPQ; ARDI
28	Establishing the Program for Sunn Pest controlling in Romania, based on wheat cropped surfaces in the damaging areas and on diapausing Sunn Pest population levels	January	MAFRD - CLPQ; ARDI

NOTE : MAFRD - Ministry of Agriculture, Forests and Rural Development

CLPQ - Central Laboratory for Phytosanitary Quarantine

DIPPPQ - District Inspectorate of Plant Protection and Phytosanitary Quarantine

ARDI - Agricultural Research and Development Institute - Fundulea

Based on data of distribution and population level it is possible to limit damaging areas and zones with favorableness degrees for an explosive reproduction of this pest, at the levels of the 41 districts of this country (Figure 1). In the damaging areas of Sunn Pest in Romania, the following surfaces are included 230.200 ha of oak forests (575 forests), favorable to adult diapause, and 30% of whole wheat area from the total of 737.000-1.184.000 ha annually cropped with wheat (Table 2). In autumn, in October, all 575 forests are inspected, and density of overwintering adult diapausing insects is The average number established. of insects/sq.m. of forest, district, zone and whole area, and the total number of insects were

calculated. Inspections are repeated in spring (March), to recalculate data, according to winter mortality. In fields, repeated inspections are performed during April-July, and density of overwintering adults, dynamics of oviposition and its level, parasitization level, density and evolution of the new generation (nymphs and new adults) is established. The insect density is estimated at the end of the control campaign (before adults leave crops for forests), and also the attack level is analyzed (by establishing the percentage of stung kernels) these representing an early step in evaluation of insect population levels which will infest wheat crops the next year (2, 5, 36, 8, 10, 12, 15, 19, 26, 17, 18, 35).

Zone/District	Oak	Cropped wheat (ha)		
Code number	forests (ha)	Total	Affected	
AREAL	230,200	1,363,000-1,945,000	737,000-1,184,000	
Favorable zone I	189,600	1,134,000-1,577,000	685-1,084,000	
Botosani, 07	53,000	45,000-85,000	35,000-60,000	
Braila, 09	3,600	65,000-90,000	50,000-70,000	
Calarasi, 12	15,200	125,000-180,000	75,000-125,000	
Constanta, 14	8,000	100,000-130,000	75,000-100,000	
Dolj, 17	12,000	145,000-190,000	75,000-120,000	
Galati, 18	6,200	65,000-90,000	35,000-65,000	
Giurgiu, 19	7,100	54,000-80,000	45,000-65,000	
Ialomita, 23	14,900	75,000-100,000	60,000-80,000	
Iasi, 24	18,700	40,000-75,000	10,000-25,000	
Ilfov, 25	4,000	20,000-32,000	15,000-24,000	
Mehedinti, 27	10,500	55,000-65,000	20,000-35,000	
Olt, 30	10,100	115,000-145,000	60,000-95,000	
Teleorman, 36	16,000	130,000-160,000	70,000-120,000	
Tulcea, 38	3,300	50,000-80,000	40,000-65,000	
Vaslui, 39	7,000	50,000-75,000	20,000-35,000	
Favorable zone II	40,600	229,000-368,000	52,000-100,000	
Arges, 03	1,000	38,000-54,000	10,000-15,000	
Bacau, 04	16,400	15,000-45,000	5,000-15,000	
Buzau, 10	3,900	40,000-70,000	8,000-13,000	
Dambovita, 16	2,000	30,000-42,000	5,000-11,000	
Neamt, 29	1,400	25,000-35,000	4,000-7,000	
Prahova, 31	4,100	30,000-40,000	7,000-13,000	
Suceava, 35	5,200	17,000-25,000	3,000-6,000	
Valcea, 40	1,000	10,000-22,000	5,000-8,000	
Vrancea, 41	6,000	24,000-35,000	5,000-12,000	

Table 2. Distribution of cropped wheat areas and oak forests, in damaging areas, in Romania.

Table 3. Ratio between Eurygaster spp. and Aelia spp. in Romania 1907-2004.

7		Perio	d:	
Zone	1907*	1939**	1970	2004
AREAL		41 dist	ricts	
Favorable zone I		15 dist	ricts	
DOBROGEA	Y/Y	86/14	97/3	98/2
MUNTENIA	Y/Y	80/20	93/7	98/2
OLTENIA	Y/Y	80/20	93/7	98/2
MOLDOVA	Y/Y	75/25	80/20	87/2
Favorable zone II		9 distr	icts	
MUNTENIA	Y/Y	75/25	85/15	97/3
MOLDOVA	Y/Y	70/30	75/25	95/5
Unfavorable zone		17 dist	ricts	
WEST PLAIN	Y/Y	75/25	80/20	80/20
TRANSILVANIA	Y/Y	65/35	70/30	77/23

## Results and Discussion

## Relationship Between Species, Damaging Areas

Eurygaster and Aelia exist throughout the country in various ratios (Table 3). Eurygaster maura L. and E. austriaca Schrank have been known since the beginning of the past century, as shown in fauna lists by Montandon (4). Early records for these species as pests belong to Radulescu, starting from 1929 (Radulescu, 1937, in Radulescu & Gruita (40)). From 1938-1939 three harmful species were reported: E. maura, E. austriaca and E. integriceps - the last one being recorded for the first time (40). E. testudinaria Geoffroy, mentioned by Popov (8, 11) is devoid of economical significance. Now, the genus Eurygaster exists throughout the country, practically in all surfaces cropped with cereals and primarily wheat (10, 30, 33). However, the damaged area is confined to South and East of the country, in the extra-Carpathian zones, i.e. in Dobrogea, Muntenia, Oltenia and Moldova (3, 6, 9, 10, 17, 18, 24, 35). As it is shown in Table 4, at present, in this area E. integriceps is prevailing over E. maura and E. austriaca to an overwhelming extent, accounting for more than 95%. Moreover, evolution in time of the species ratio showed that this dominance was stabilized during the last three decades. Range expansion of E. *integriceps* started in the 2<sup>nd</sup> and 3<sup>rd</sup> decades of the last century, by successive migrations from the Russian steppe, through the North of the Black Sea, going on towards West and South in Bulgaria, Serbia and the European part of Turkey (10, 27, 25, 29, 30). An illustration of this phenomenon is well expressed by its area in Oltenia, where E. integriceps did not exist during 1938-1939, yet reached 16-50% in 1970 and finally, became strongly prevalent (96-98%) at present (19, 20, 27). In Table 5 is presented the situation in unfavorable zone for E. integriceps.

This damaged area appeared since 1965 and progressively expanded, covering at present 24 districts. Climatic conditions and surfaces cropped with wheat (for multiplication and development) and oak forests (for diapause), differentiate damaged areas under a very favorable zone (15 districts), where the ensemble of factors are steadily optimal, and a less suitable area (9 districts), where the optimal conditions intermittently occur. In the favorable zone I the highest surfaces are cropped with wheat, while oak tree forests represent more than 80% of the area.



**Figure 1.** Area of Sunn Pest in Romania (code number of district). Favorable zone 1 (Muntenia: 9, 12, 19, 23, 25, 36; Oltenia: 17, 27, 30; Dobrogea: 14, 38; Moldova: 7, 18, 24, 39); Favorable zone 2 (Muntenia: 3, 10, 16, 31, 40; Moldova: 4, 29, 35, 41); Unfavorable zone (West Plain: 2, 5, 11, 26, 32, 37; Oltenia: 20; Transilvania: 1, 6, 8, 13, 15, 21, 22, 28, 33, 34)

Inspections performed at diapause start in all oak forests with variable average densities from one zone to another and from one year to the next (Table 6, 7). Multiannual analyses showed the highest density of Sunn Pest was reached only in those districts included in favorable zone I. In these biotopes, pest densities varied from year to year, and their numbers ranged from a few specimens per sq.m. to more than 50 (5, 25, 29). Based on records in autumn samplings (October 15-30) and in spring samplings (April 1-15), the total number for every forest, district and total area can be estimated, this estimation was the basis for predicting Sunn Pest populations every year (Table 8) (11, 17, 18, 27, 31).

Zone/District,		Per	iod:	
Code number	1907*	1939**	1970	2004
AREAL		41 dis	stricts	
Favorable zone I		15 dis	stricts	
Botosani, 07	-	0/66/34	47/33/20	96/2/2
Braila, 09	-	-	88/7/5	98/1/1
Calarasi, 12	0/Y/Y	-	94/3/3	98/1/1
Constanta, 14	0/Y/Y	75/11/14	94/4/2	98/1/1
Dolj, 17	0/Y/Y	0/86/14	36/25/39	97/2/1
Galati, 18	0/Y/Y	45/33/22	85/9/6	98/1/1
Giurgiu, 19	-	-	73/18/9	98/1/1
Ialomita, 23	-	35/20/45	95/3/2	98/1/1
Iasi, 24	-	0/67/33	41/40/19	97/2/1
Ilfov, 25	-	31/28/41	94/3/3	98/1/1
Mehedinti, 27	-	-	16/36/58	96/2/2
Olt, 30	-	0/65/35	50/26/24	98/1/1
Teleorman, 36	0/Y/Y	-	66/21/13	98/1/1
Tulcea, 38	-	87/5/8	94/4/2	98/1/1
Vaslui, 39	-	21/52/27	57/28/15	97/2/1
Favorable zone II		9 dis	tricts	
Arges, 03	0/Y/Y	-	26/54/20	96/2/2
Bacau, 04	-	0/54/46	45/9/46	97/2/1
Buzau, 10	-	-	32/47/21	95/3/2
Dambovita, 16		-	27/52/21	94/3/3
Neamt, 29	-	-	42/20/38	90/4/6
Prahova, 31	-	15/42/43	28/52/20	96/2/2
Suceava, 35	-	0/94/6	0/54/46	90/6/4
Valcea, 40	-	-	25/53/22	93/4/3
Vrancea, 41	-	-	27/40/33	91/5/4

Table 4. Ratio between Eurygaster spp. in favorable zone (1907-2004).

NOTE: 0 - absent; Y - mention; No.- percent (Eurygaster integriceps/Eurygaster maura /Eurygaster austriaca)

Table 5. Ratio between Eurygaster spp. in unfavorable zone (1907-2004).

Zone/District		Period		
Code number	1907*	1939**	1970	2004
AREAL		41 distric	ts	
Unfavorable zone		17 distric	ts	
Alba, 01	-	0/83/17	0/85/15	0/80/20
Arad, 02	-	-	0/43/57	1/44/55
Bihor, 05	0/Y/Y	0/77/23	1/41/58	1/43/56
Bistrita-Nasaud, 06	-	-	0/73/27	0/60/40
Brasov, 08		-	0/65/35	0/63/37
Caras-Severin, 11	-	-	0/46/54	0/50/50
Cluj, 13	0/Y/Y	0/89/11	0/80/20	0/75/25
Covasna, 15	-	-	0/50/50	0/53/47
Gorj, 20	-	-	7/41/52	33/35/32
Harghita, 21	-	-	0/50/50	0/57/43
Hunedoara, 22	-	0/66/34	0/87/13	0/81/19
Maramures, 26	-	-	-	0/50/50
Mures, 28	-	0/40/60	0/62/38	0/63/37
Satu Mare, 32	-	0/79/21	0/58/42	0/64/36
Salaj, 33	-	-	0/61/39	0/59/41
Sibiu, 34	-	-	0/58/42	0/56/44
Timis, 37	-	0/72/28	3/36/58	1/49/50

NOTE: 0 - absent; Y - mention; No. - percent (Eurygaster integriceps/Eurygaster maura /Eurygaster austriaca)

Zone/District Code number	1987	2000	2001	2002	2003
AREAL	14.4	2.9	3.4	6.0	4.4
Favorable zone I	20.8	4.4	5.3	10.1	7.0
Botosani, 07	5.3	0.3	0.3	0.4	0.6
Braila, 09	43.8	8.9	12.9	24.0	9.1
Calarasi, 12	61.8	5.6	12.3	24.4	7.7
Constanta, 14	29.4	8.7	6.4	16.4	5.1
Dolj, 17	19.3	3.4	4.2	5.8	3.4
Galati, 18	24.3	3.5	3.4	7.9	5.4
Giurgiu, 19	23.4	4.5	4.9	4.7	8.2
Ialomita, 23	40.5	3.1	3.1	14.6	7.5
Iasi, 24	2.8	0.8	0.8	1.2	1.1
Ilfov, 25	24.1	0.5	0.5	2.2	2.1
Mehedinti, 27	1.6	0.8	1.2	1.4	2.6
Olt, 30	25.1	1.8	3	4.6	4.4
Teleorman, 36	35.7	4.1	8.4	14.4	12.7
Tulcea, 38	6.4	10.4	13.6	22.5	24.2
Vaslui, 39	14.7	1.5	1.6	1.8	1.3
Favorable zone II	8.1	1.4	1.4	1.9	1.9
Arges, 03	12.4	4.7	4.8	5.2	5.2
Bacau, 04	7.2	2.3	2.8	3.0	2.8
Buzau, 10	11.7	1.4	1.2	1.1	0.8
Dambovita, 16	29.3	1.4	1.3	2.8	2.9
Neamt, 29	0.4	1.7	0.5	2.3	2.1
Prahova, 31	10.5	0.3	0.8	2.0	1.8
Suceava, 35	0.1	0.1	0.1	0.1	0.1
Valcea, 40	1.5	1.1	1.1	0.8	1.2
Vrancea, 41	5.2	0.3	0.4	0.3	0.3

Table 6. Sunn Pest diapausing populations in oak forests, in damaging areas (density./sq.m.), 1987-2003.

These areas were considered stable and the maximum population areas for zones in Dobrogea, Muntenia, Oltenia and Moldova, where *E. integriceps* finds favorable conditions to develop high populations, obviously varying from year to year (30, 31, 32). Monitoring Sunn Pest populations by evaluating the total number of insects in a generation, in diapausing sites differentiated favorable from unfavorable zones.

Analyses of mortality of diapausing Sunn Pest are another significant element for monitoring; yearly data revealed 15-30% mortality, with broad variations between zones and years (Table 9). Overwintering adults were found in the spring in the crops at densities of a few individual/sq.m., and more rarely at densities between 15 and 20 individual/sq.m. The new generation (nymphs and young adults) had higher densities, usually 15-20 specimens/sq.m., or during the years with particularly favorable chemical conditions, 70-94/sq.m. as happened in 1996. In 2003 there was 145/sq.m (Table 10).

#### Life Cycle - Annual Peculiarities

Pursuit of life-cycle evolution in direct relationship with annual ecological peculiarities is of major interest, due to the ability of insects to develop more or less numerous populations, according to the climatic conditions and phenology of the hostplant. This may be passed on or not to the whole life-cycle under optimal conditions, (1, 6, 26, 25, 30, 31). There were favorable climatic conditions for Sunn Pest in 2003 which induced hastening the life-cycle by 2-3 weeks, and increased its reproductive potential by about 25-35%. In 1997 and 2003, field evolution of Sunn Pest was normal (Tables 11 and 12).

Zone/District		Year	r:		Max. density in 1969-
Code number	1969	1978	1987	2003	2003
AREAL	142.8	107.9	185.2	66.2	185.2/1987
Favorable zone I	142.8	107.9	185.2	66.2	185.2/1987
Botosani, 07	-	-	41.4	8.8	49.3/1988
Braila, 09	7.4	36.0	71.5	22.0	85.5/1977
Calarasi, 12	27.0	80.5	185.2	28.0	185.2/1987
Constanta, 14	50.9	85.0	74.0	66.2	118.7/1982
Dolj, 17	3.7	107.9	114.6	12.4	114.6/1987
Galati, 18	7.7	24.6	44.9	3.7	69.4/1982
Giurgiu, 19	11.0	14.6	67.9	11.5	67.9/1987
Ialomita, 23	27.5	74.8	65.7	22.9	80.5/1979
Iasi, 24	-	-	10.0	2.7	10.0/1987
Ilfov, 25	-	12.9	76.5	11.0	76.5/1987
Mehedinti, 27	0.4	74.5	67.3	2.6	74.5/1978
Olt, 30	1.8	92.2	85.0	12.7	132.1/1985
Teleorman, 36	13.3	102.2	139.9	29.8	139.9/1987
Tulcea, 38	142.8	31.9	26.0	21.3	142.8/1969
Vaslui, 39	-	-	44.5	2.5	97.0/1982
Favorable zone II	-	65.6	73.8	23.5	73.8/1987
Arges, 03	-	65.6	24.8	4.8	65.6/1978
Bacau, 04	-	-	23.9	23.5	23.9/1987
Buzau, 10	-	-	19.8	1.3	19.8/1987
Dambovita, 16	-	40.5	64.8	5.5	70.0/1988
Neamt, 29	-	-	-	2.7	2.7/1998
Prahova, 31	-	-	73.8	3.6	73.8/1987
Suceava, 35	-	-	-	4.1	4.1/1998
Valcea, 40	-	-	5.7	2.0	6.9/1993
Vrancea, 41	-	-	19.7	1.1	41.0/1982

Table 7. Maximum density (individuals/sq.m.), recorded at the level of a representative forest in each district.

Besides climatic conditions the physiological preparation factor expressed by the fat-body, is particularly important for population fluctuations (8, 11, 12, 14, 15, 38, 30, 39).

#### **Pest-parasite Relationships**

Parasitization is important and of major concern for control technology. The average parasitisation fluctuates 30-50%, with certain higher values in a few districts (Table 13). At present, protection of natural parasitic fauna is achieved mainly by framing treatments within the optimal time (13, 16, 27, 34, 7, 21, 22). Composition of oophagous parasitic species occurring in Romania revealed a high share for *Telenomus chloropus* and *Trissolcus grandis* (70-80%), among the total of 8 recorded species (21, 22, 39).

# Economic Damage Threshold (EDT) and Control Measures

Overwintering adult control is done only in wheat plots, when densities exceed the EDT, as established as follows:

- 7 adults/sq.m. in plots with normal plant density and vegetation, when spring weather is humid and cool;
- 5 /sq.m. in plots with normal plant density and vegetation, in spring with high temperatures;
- 3 /sq.m. in plots with reduced plant densities, with poor overwintering survival, humid and warm spring;
- 1 2 /sq.m. under unfavorable complex conditions to crops; late emergence of plants in autumn - winter; prolonged winter with persistent snow layer; late spring with excessively dry weather.

Zone/District	1005 1007	1007 1007	2001 2002	2002 2002
Code number	1995-1990	1990-1997	2001-2002	2002-2003
AREAL	29844	5120	6372	17409
Favorable zone I	26567	4506	6101	17080
Botosani, 07	2852	96	121	1899
Braila, 09	303	96	471	3728
Calarasi, 12	2629	442	877	1726
Constanta, 14	3616	1124	834	2138
Dolj, 17	2154	368	429	695
Galati, 18	1328	184	183	422
Giurgiu, 19	1803	317	347	335
Ialomita, 23	913	463	464	2184
Iasi, 24	37	158	157	221
Ilfov, 25	729	25	23	217
Mehedinti, 27	117	88	108	149
Olt, 30	1347	187	305	462
Teleorman, 36	7205	610	1336	2187
Tulcea, 38	215	323	421	697
Vaslui, 39	1318	25	22	20
Favorable zone II	896	948	608	741
Arges, 03	87	40	41	44
Bacau, 04	364	431	93	102
Buzau, 10	173	61	42	49
Dambovita, 16	1434	27	26	12
Neamt, 29	1	16	4	19
Prahova, 31	855	12	32	84
Suceava, 35	1	1	1	1
Valcea, 40	87	40	41	44
Vrancea, 41	354	18	28	13

**Table 8.** Sunn pest diapausing populations (,000,000 overwintering adults), in oak forest, in damaging area, 1995-2003. At each zone is the number of overwintering adults, from table + 000,000

Establishing plots to be treated for larval control: Chemical application is performed only in wheat plots where pest density exceeds the EDT, thus:

- 5 nymphs/sq.m. for plots designed for bread-making;
- 3 nymphs/sq.m. for plots designed as seed multiplication.

Chemical control is the unique practical method of intervention in order to diminish harvest damage (1, 11, 18, 23, 27, 29, 30, 31, 32, 39). Registered insecticides include both organophosphorus and synthetic pyrethroid compounds. The main share in Sunn Pest control has been done with trichlorfon and dimethoate (100% in 1970-1975; 96% in 1985; 89% in 1993 compared with 2% in 1997). After 1985 other insecticides, mainly pyrethroids have been registered. In 1993

pyrethroids were used on 10% of the treated areas (against the Sunn Pest). However after 1998 they represented over 95% of the total treated surface in Romania, especially cypermethrin (30, 31, 32, 33, 37, 39).

Our annual complex monitoring data facilitated Sunn Pest attack forecasts and correct treatment applications.

Chemical control costs were subsidized by the Ministry of Agriculture, Forest, Water and Environment. Treated wheat surfaces varied from one year to another (Table 14). Harvest quality analyses, as established by the percentage of damaged grains (Table 15) (practically performed to all grain production in the damaging area) showed the excellent results of a complex activity for protection of wheat crops in Romania against the particularly harmful outbreaks of Sunn Pest.

Zone/District	1997/	1998/	1999/	2000/	2001/	2002/
Code number	1998	1999	2000	2001	2002	2003
AREAL	28.5	17.0	29.8	12.6	10.7	15.6
Favorable zone I	21.9	15.8	18.7	8.8	9.7	9.3
Botosani, 07	28.6	17	24.2	17.7	8.8	18.2
Braila, 09	55	23.1	20	4.4	11.5	7.5
Calarasi, 12	62	58	31.6	5.3	32.3	16.2
Constanta, 14	9.7	11.8	12.6	4.6	21.8	12.6
Dolj, 17	14.3	12.4	10.4	7.7	5.1	10.7
Galati, 18	38	52	27	5.5	2.2	3.4
Giurgiu, 19	10	14.8	17.8	8.9	5.6	6.6
Ialomita, 23	39	53	30.5	7.6	10.4	7.3
Iasi, 24	28.6	10	32.6	3.7	4.8	12.4
Ilfov, 25	7.3	11	12.6	6.7	2.1	2.3
Mehedinti, 27	8.7	8.9	12	13.1	6.3	5.7
Olt, 30	53	12	28.8	5.7	5.2	4.2
Teleorman, 36	14.1	12	15	20	6.1	11.7
Tulcea, 38	3.8	3	4.8	7.4	8.2	5.2
Vaslui, 39	10.6	20	30.8	7.5	5.8	3.3
Favorable zone II	35.2	18.3	40.8	16.3	11.7	21.8
Arges, 03	4.8	8	15.5	8.4	3.3	3.3
Bacau, 04	2.8	12	25	7.9	7.9	13.1
Buzau, 10	19.4	21	20	24.2	25.2	30.3
Dambovita, 16	13.4	16.7	24	7.5	3.7	4.6
Neamt, 29	6.8	7.3	46.9	9.6	11.8	46.5
Prahova, 31	78	56	58.9	35.7	14.1	14.7
Suceava, 35	10.7	4.5	46.5	12.3	3.3	6.3
Valcea, 40	93.4	33	97.6	27	10.7	56.7
Vrancea, 41	77.3	9	24.7	15.3	23.5	10.6

Table 9. Average adult mortality (%) recorded in diapause 1997-2003.

Table 10. Sunn Pest population densities in wheat fields (no./sq.m.), in damaging areas, in Romania.

Zone/District	Favorable yea	ar	Normal year		
Code number	<b>Overwintering adults</b>	Nymphs	Overwintering adults	Nymphs	
AREAL	1 - 22	10 - 145	1 - 12	2 - 51	
Favorable zone I	2 - 22	28 - 145	1-12	7 - 51	
Botosani, 07	2	28	2	12	
Braila, 09	13	62	1	37	
Calarasi, 12	4	125	3	20	
Constanta, 14	10	140	7	46	
Dolj, 17	22	120	2	25	
Galati, 18	14	95	3	20	
Giurgiu, 19	12	85	1	34	
Ialomita, 23	5	90	7	51	
Iasi, 24	7	30	1	8	
Ilfov, 25	14	63	1	15	
Mehedinti, 27	6	40	3	7	
Olt, 30	21	76	3	18	
Teleorman, 36	7	145	2	35	
Tulcea, 38	12	118	12	38	
Vaslui, 39	5	52	1	14	
Favorable zone II	1 - 9	10 - 45	1 - 7	2 - 20	
Arges, 03	4	38	3	12	
Bacau, 04	2	25	2	10	
Buzau, 10	9	45	6	20	

Table 10 (Cont.)

Dambovita, 16	2	25	5	10
Neamt, 29	1	20	1	5
Prahova, 31	4	27	7	12
Suceava, 35	1	10	1	2
Valcea, 40	9	12	1	3
Vrancea, 41	1	18	1	4

Table 11. Date when first overwintering Eurygaster integriceps adults appeared in wheat fields.

Zone/District	1997		2003		
Code number	Date	t C°	Date	t C°	
AREAL	30.03 - 02.05	25.7	10 - 30.04	18.0	
Favorable zone I	30.03 - 28.04	23.6	10 - 28.04	19.9	
Botosani, 07	25.04	21.7	26.04	21.2	
Braila, 09	9.04	33.9	16.04	11.7	
Calarasi, 12	8.04	15.2	24.04	16.4	
Constanta, 14	12.04	28.2	20.04	11.9	
Dolj, 17	30.03	11.7	17.04	15.3	
Galati, 18	06.04	17.2	18.04	10.9	
Giurgiu, 19	15.04	19.4	24.04	12.8	
Ialomita, 23	17.04	32.1	22.04	16.8	
Iasi, 24	28.04	31.2	28.04	31.2	
Ilfov, 25	10.04	27.7	22.04	2,5	
Mehedinti, 27	15.04	24.2	10.04	20.5	
Olt, 30	18.04	16.8	13.04	16.6	
Teleorman, 36	08.04	24.9	15.04	10.5	
Tulcea, 38	17.04	11.3	23.04	11.9	
Vaslui, 39	14.04	35.1	28.04	37.5	
Favorable zone II	09.04 - 02.05	27.9	23 - 30.04	16.1	
Arges, 03	10.04	32.7	25.04	15.9	
Bacau, 04	27.04	18.4	30.04	15.1	
Buzau, 10	17.04	28.8	30.04	22.1	
Dambovita, 16	21.04	36.4	24.04	6.2	
Neamt, 29	30.04	33.9	23.04	17.2	
Prahova, 31	09.4	22.8	23.04	16.1	
Suceava, 35	02.05	28.3	28.04	16.7	
Valcea, 40	09.04	20.8	27.04	18.8	
Vrancea, 41	14.04	30.2	29.04	16.8	

## Conclusions

Monitoring the Sunn Pest population in Romania is annually carried out in all districts, based on elements as follow:

**Density in diapausing sites by:** average insect number/sq.m in each forest, at diapausing start and end; adults mortality during diapause; and total insect population at forest, district, zone, area level.

**Density under field conditions by:** infestation level by overwintering adults; establishing features of life-cycle (migration, oviposition, nymphal development, adults formation); share of natural parasitism; and infestation level by nymphs for each plot in the damaging area.

Centralization and interpretation of these data lead to setting up average and short-term forecasts for the whole area, these being used to release application warnings for chemical control undertakings, by zones and districts.

Zone/District	Nymphs (stage III)		New a	adults
Code number	1997	2003	1997	2003
AREAL	02 - 30.06	22.05 - 9.06	20.06 - 2.08	11.06 - 1.07
Favorable zone I	02 - 16.06	22.05 - 9.06	20.06 - 9.07	11 - 27.06
Botosani, 07	06.06	06.06	03.07	27-06
Braila, 09	02.06	26.05	26.06	17.06
Calarasi, 12	09.06	29.05	27.06	21.06
Constanta, 14	12.06	26.05	30.06	11.06
Dolj, 17	06.06	23.05	05.07	16.06
Galati, 18	03.06	27.05	30.06	14.06
Giurgiu, 19	03.06	27.05	25.06	22.06
Ialomita, 23	02.06	22.05	26.06	19.06
Iasi, 24	16.06	28.05	09.07	20.06
Ilfov, 25	05.06	29.05	21.06	14.06
Mehedinti, 27	10.06	09.06	20.06	27.06
Olt, 30	08.06	28.05	25.06	20.06
Teleorman, 36	02.06	07.06	24.06	26.06
Tulcea, 38	05.06	31.05	27.06	20.06
Vaslui, 39	08.06	29.05	30.06	27.06
Favorable zone II	02 - 30.06	26.05 - 1.06	25.06 - 2.08	20.06 - 1.07
Arges, 03	06.06	02.06	28.06	23.06
Bacau, 04	28.06	11.06	06.07	01.07
Buzau, 10	02.06	06.06	25.06	28.06
Dambovita, 16	10.06	26.05	05.07	20.06
Neamt, 29	27.06	06.06	25.07	02.07
Prahova, 31	03.06	11.06	08.07	13.07
Suceava, 35	30.06	10.06	02.08	21.07
Valcea, 40	12.06	03.06	29.06	25.06
Vrancea, 41	05.06	27.05	09.07	25.06

**Table 12.** First appearance of nymphs (stage III) and new adults in wheat crops, in 1997 and 2003.

Table 13. Parasitization level (%) of Sunn Pest eggs by oophagous parasites, 2000-2003	Table 13. Parasitization	level (%) of Sun	n Pest eggs by	oophagous	parasites, 2000-2	2003.
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Zone/District	2000	2001	2002	2003
Code number	2000	2001	2002	2003
AREAL	31.3	35.2	34.1	38.2
Favorable zone I	39.9	37.0	39.2	37.5
Botosani, 07	40	35	30	45
Braila, 09	17	18	21	32
Calarasi, 12	15	41	30	44
Constanta, 14	30	20	31	35
Dolj, 17	57	68	66	60
Galati, 18	20	10	6	37
Giurgiu, 19	22	38	26	31
Ialomita, 23	60	38	12	15
Iasi, 24	40	30	35	40
Ilfov, 25	12	12	6	6
Mehedinti, 27	48	49	48	42
Olt, 30	35	25	35	34
Teleorman, 36	60	31	28	48
Tulcea, 38	36	32	57	21
Vaslui, 39	47	33	46	30
Favorable zone II	22.8	33.5	29.0	38.9
Arges, 03	22	40	30	28
Bacau, 04	49	31	33	40
Buzau, 10	25	14	26	46

## Table 13 (Cont.)

Dambovita, 16	35	7	25	12	
Neamt, 29	48	46	45	45	
Prahova, 31	50	66	53	49	
Suceava, 35	38	40	25	46	
Valcea, 40	76	50	60	69	
Vrancea, 41	28	25	22	15	

Table 14. Area (ha) in Romania treated with chemical pesticides for Sunn Pest management. 1996-2003.

Zone/District	1004	2001	2002	2002
Code number	1990	2001	2002	2005
AREAL	749.811	513.362	449.607	398.433
Favorable zone I	663.195	470.703	404.423	336.017
Botosani, 07	8.734	2.860	1.860	0
Braila, 09	43.080	36.779	41.320	33.536
Calarasi, 12	62.106	59.320	64.800	59.547
Constanta, 14	87.697	71.963	42.464	14.830
Dolj, 17	113.662	31.281	25.950	36.382
Galati, 18	37.180	36.947	29.670	14.214
Giurgiu, 19	39.362	12.599	18.150	16.705
Ialomita, 23	55.767	50.290	36.880	21.350
Iasi, 24	11.280	5.230	4.030	3.800
Ilfov, 25	12.801	7.520	5.329	10.037
Mehedinti, 27	10.670	2.200	2.640	3.850
Olt, 30	66.157	47.573	34.630	29.586
Teleorman, 36	54.668	40.518	43.445	49.143
Tulcea, 38	41.700	41.000	23.000	26.230
Vaslui, 39	18.331	24.623	30.255	16.807
Favorable zone II	86.616	42.659	45.184	62.416
Arges, 03	15.077	14.333	17.310	26.209
Bacau, 04	9.742	5.600	6.220	7.690
Buzau, 10	8.626	3.766	5.302	6.309
Dambovita, 16	14.973	7.800	3.250	4.850
Neamt, 29	20.571	0	4.486	6.892
Prahova, 31	500	400	0	0
Suceava, 35	7.385	960	890	1.265
Valcea, 40	4.000	5.200	2.860	3.312

Table 15. Percentage of damaged grains, 1994-2003.

Year	Total grain tons analyzed	Wheat yield%			
		Without attack	Attack level%		
			0.1-2.0	2.1-5.0	over 5.1
1994	2,146.3	22.5	55.2	16.7	5.6
1995	2,229.6	14.5	51.7	25.1	8.7
1996	800.5	12.6	73.7	13.0	0.7
1997	2,308.7	28.4	68.2	3.3	0.1
1998	1,663.9	29.0	68.0	3.0	-
1999	2,310.0	18.6	77.2	4.1	0.1
2000	1,484.3	23.6	70.2	5.6	0.6
2001	2,429.3	20.9	72.9	6.2	-
2002	1,142.9	6.7	70.7	18.2	4.4
2003	302.7	6.4	51.3	29.7	12.6
## الملخص

بويوف، س.، أ. برابوليسك، ف. موريسان، إس. فاسيليسك، ف. غوغو، و ي. دويرين. 2007. إدارة حشرة السونة في رومانيا. الصفحات 45–60.

تعد حشرة السونة (Eurygaster integriceps Puton) من إحدى أهم آفات القمح في رومانيا. تم التحري عن العناصر الأساسية للإدارة المتكاملة لحشرة السونة. كانت العناصر هي: النسبة بين الأنواع ومناطق توزعها في رومانيا، الحجم ومكان التوزع لمجتمعات حشرة السونة، دراسة الخصائص السنوية لدورة حياتها، العلاقة ما بين الآفة وطفيليات البيض، وتأسيس عتبات الضرر الاقتصادي ووسائل مكافحتها. تصيب حشرة السونة حوالي 1,000,000 هكتار من القمح في 24 مقاطعة. قدرت مساهمة طفيليات البيض مالقصادي المكافحتها. تصيب حشرة السونة حوالي 1,000,000 هكتار من القمح في 24 مقاطعة. قدرت مساهمة مختلف حسب مرحلة نمو المحصول والهدف من الحصاد. يحظى التنبؤ بالاهتمام الرئيس في رومانيا وتعتمد الإجراءات على طرائق مطورة بواسطة آردي فوندوليا (ARDI Fundulea)، ومطبقة عبر البلد بواسطة مفتشي المقاطعات لوقاية النبات والحجر الصحي النباتي. وإن المكافحة الكيميائية لحشرة السونة في رومانيا مدعومة من الحومة.

كلمات مفتاحية: Eurygaster integriceps، حشرة السونة، الإدارة المتكاملة لآفات القمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon. Galia Zharmukhamedova and Amangeldy Sarbaev Institute for Plant Protection, Almaty, Kazakhstan.

### Abstract

Zharmukhamedova, G. and A. Sarbaev. 2007. Sunn Pest in Central Asia: An Historical Perspective. Pages 61-66.

The historical and modern status of Sunn Pest is discussed: area, biology, ecology, harmfulness, economic threshold, and control measures. The review of development strategy and tactics of Sunn Pest IPM (past, present, future) is given.

Keywords: Bioecology, control, distribution, harmfulness, Sunn Pest, wheat.

Sunn Pest (*Euryg ster integriceps* Puton.) is one of the major pests of summer and winter wheat in all basic wheat areas in the European part of Russia, a steppe zone of Ukraine, the western and southern regions of Kazakhstan and the Central Asian Republics.

The first data about outbreaks of Sunn Pest in the Ukraine and southern Russia were reported from 1865-1867, in the Caucasus 1896-1900 and in Central Asia 1891-1892. The next outbreaks in various regions of the USSR occurred periodically every 5-7 yrs and sometimes resulted in total destruction of the crop. From 1938 to 1944 Sunn Pest populations universally reached extremely high densities. In wintering places 2,000 to 40,000 bugs/m<sup>2</sup> were found (19). These high populations occurred in the extensive grain zones of the Ukraine, the south of Russia, and the Northern Caucasus, from 1950-1955. A direct loss of wheat during the period from 1936 to 1955 was estimated at 1.3 million tons of grain a year (14).

Extending wheat production to northern and eastern regions of Russia, and development of virgin farm lands in Western Kazakhstan from 1950-1960 led to the expansion of Sunn Pest infestation areas and an increase in their economic importance. Until 1960 Sunn Pest in Western Kazakhstan was an identified species but of no economic importance. But in 1962 the damage of wheat grain by this insect reached 21 %. By 1966 the areas where Sunn Pest overwintered increased

10 times, larval populations increased 20 times and the area sprayed with insecticides for their control 47 times. In addition to providing more wheat for Sunn Pest to feed on development of field-protective forests promoted good wintering sites for the pests, a factor that was not observed earlier. For example, in the Ural region before the development of virgin lands field-protective forests occupied about 4.5 thousand ha, and in 1960 these forests were on  $\sim$ 12 thousand ha (5). All these conditions caused the Sunn Pest outbreaks of 1967-1968 in the Volga region and Western Kazakhstan. In the European part of Russia, outbreaks were local.

During the last 35 yrs the economic value of the pest changed rather considerably. Until 1981 Sunn Pest populations were kept at the lowest level in most areas (4). The highest level of the pest population was observed in 1985-1986, when (only) in Russia Sunn Pest was sprayed with insecticides on 3.5 to 4.5 million ha (36).

The reforming of agriculture in the post-Soviet republics, a lack of material and financial maintenance caused an economic collapse that led to significantly complicating the problems of plant protection, including cereals. Since 1994 Sunn Pest populations increased in the traditional wheat production areas-in the Ukraine, the Stavropol, Krasnodar territories, the Rostov area, Northern Caucasus, the Volga region, the Ural and in Western Kazakhstan. In Russia in 1994 Sunn Pest was treated on 560,000 ha, in 1995 on 869,600 ha, in 1996 on 2314,000 ha in 1997 on 4000,000 ha and in 2000 on 3125,000 ha (1, 8). In these years the damage by Sunn Pest was one of the principal causes of a reduction of quality grain (15).

The wide infestation area and extremely high degree of damage by Sunn Pest were the main reasons for intensive research and development of effective control methods since the end of the XIX century.

The literature of Sunn Pest in Russian is extensive so we concentrated on the more general studies.

The earliest full report on Sunn Pest problems was done by Peredelsky in "Results of Biological Bases of the Theory and Practice of Sunn Pest Control" (20). His bibliography includes 574 sources, 403 of them are by Russian and Soviet scientists. He includes archival materials from various states establishments. Peredelsky gives a historical, literary and critical analysis of systematization, distribution, range, biology and ecology of Sunn Pest and close species of pentatomides that are damaging cereals not only in the USSR, but also in all Palearctik regions. Problems of Sunn Pest population dynamics, damage and control measures are discussed in detail.

It is necessary to note that this work was a component of research of the Central Asian Expedition on Sunn Pest of the Institute of Evolutionary Morphology named after A.N Severtsev, organized by the Academy of Science of the USSR in 1941-1945. Scientists under the direction of Professor D.M Fedotov made a huge theoretical and practical contribution to the study of Sunn Pest and to development of control measures. Results of these scientific expeditions were united in the Proceedings of Academy of Science of the USSR (24, 25, 26, 27).

Results of long-term research (1955-1979) of entomologists of the Ukrainian Plant Protection Institute under Areshnikov's leadership are generalized in the monograph "Sunn Pest and Control Measures" (4). Information on distribution, harmfulness, biology, ecology and population dynamics of Sunn Pest in steppe zone of Ukraine and the south of Russia are presented. A detailed critical analysis of reasons of Sunn Pest outbreaks is included. The achievements of the development and application of a wheat protection system, including preventive, cultural, biological and chemical actions are described. The leading role of chemical control, as one of the major elements of Sunn Pest management, has been scientifically proven.

The bibliography of this monograph includes 211 sources, 165 are Russian and Soviet authors. Dates of plant protection services of the Ministers of Agriculture also have been used.

Profound research on Sunn Pest has been done at the All-Union Plant Protection Institute (now the Russian Institute for Plant Protection- VIZR, St. Petersburg). Scientists of this Institution were the organizers and coordinators of research on Sunn Pest in the USSR and abroad for many years. Data of ecology and physiology of Sunn Pest, population dynamics in various zones of the country and development of new more effective methods of management have been published in 3 proceedings (21, 22, 23). A number of articles were devoted to methodical questions on the development of regional Sunn Pest forecasting and methods of estimating populations.

Research results on hormonal regulations of Sunn Pest and methods of disrupting it (10, 28, 29), on development of Sunn Pest sterilization methods have been published. Attention has been given to studying Sunn Pest parasites and predators and their usefulness in regulation of populations, including methodical questions of cultivation, introduction and application of egg parasites in IPM (6, 12, 13). Under the guidance of the Institute research and production meetings were held annually at the Ministry of Agriculture of the USSR. Two all-Union theoretical meetings were organized, in Kharkov (1969) and Voronezh (1971) where improvements of chemical methods of control, and economic thresholds to improve expediency of applications and prospects of using biological methods for Sunn Pest control were discussed.

In 1961, 1964, and 1967 at the Institute international seminars on Sunn Pest, organized by FAO of the United Nations were held. One of the primary goals of these seminars was to acquaint participants with the general state of Sunn Pest problem in the countries where it causes serious yield losses, and with the achievements developed in the USSR.

In the Central black gumus earth region of Russia and the Volga region Sunn Pest research was conducted by Pavlov (18), Volodichev (36), Grivanov and Antonenko (9), and Emeljanov (7). In Kazakhstan significant research was done by Tilmenbaev (32), Tilmenbaev and Sarbaev (33), and Sarbaev (30). Thus, the bioecology of the pest has been thoroughly investigated in all areas of the former USSR.

Strategies and tactics of protective measures have been constantly improved. From 1936-1953 emphasis was given to crop preservation to prevent quantitative losses and decrease grain damage. Control measures were addressed against the wintered bugs (20). Larval control served as an additional action and was done in fields where their numbers exceeded 10/ m<sup>2</sup>. Basic control measures included manually gathering insects, the use of a various traps, poisoned baits, raking up Sunn Pest and burning the vegetation in overwintering sites. Attempts to use biological measures such as herding hens to feed on overwintering Sunn Pest and releasing egg parasites (34) were unsuccessful.

The second qualitatively new stage of Sunn Pest control development was the period from 1953 to 1968, when instead of ineffective mechanical and physical methods of management the wider practice of using chemical insecticides became popular. During this period emphasis was placed on increasing and improving efficiency and quality of insecticides, such as DDT, metaphos and chlorophos. There was a replacement of unproductive and expensive methods of dusting with spraying, and then with the use of ULV applications.

The economic threshold levels of Sunn Pest were specified and considerably reduced. This stage characterized by completely preventing quantitative losses of crop, but also sharply reducing the negative influence of the pest on grain quality (4).

The third stage of Sunn Pest control history was the period from 1968 to the present, when there was not a maximal suppression on the pest population, but rather a stabilization of population density at a low level without damage to the crop. This required a change in protective measures.

Experience has shown that adult Sunn Pest control, even in the years of pest outbreaks, does not allow protection of the crop from larvae. Because of this major emphasis has been placed on larval control from the beginning of the milky stage-to full grain ripeness. Sunn Pest feeding during this period decreases gluten quality and results in reduced baking standards. In Russia during Sunn Pest outbreaks, at least 3-7 million tons of marketable wheat was nonstandard (38).

For the prevention of quantitative losses of wheat the following thresholds of Sunn Pest harmfulness have been established: 1-2 wintered bugs (tillering-stalkshooting) and 10-15 larvae (flowering-the beginning of grain formation) on 1 m<sup>2</sup>. During the last decades such levels were observed locally in some years. Hence, the necessity for using chemical insecticides for control in wheat fields was rare.

For preservation of high quality gluten according to state standards no more than 2 % damage grains in the crop are permitted. Normally such a level of damage grain does not result in economic losses and the quality of grain is reduced only when 4-9 % damage occurs. Therefore, different threshold levels of larvae were developed. For preventing damage to valuable wheat the threshold is 2 larvae, and for ordinary crops-5-6 larvae/m<sup>2</sup>. The later number of larvae is observed practically everywhere, and thus annual protective measures are required. These threshold levels of Sunn Pest are found in all post Soviet fields (1, 3, 33, 36).

The important part of Sunn Pest management is the complex of preventive and cultural methods, which increase plant resistance to pest damage such as: crop rotations, optimum timing and rates of seeding, correct rates of fertilizes etc. The organization of harvesting has great value. First, wheat with high quality grain should be harvested. The preference should be given to direct harvesting, because larval mortality from mechanical influence is higher than at two-phase harvesting. Mowing of fields in the perimeter (20-25 m), which is always intensively occupied and damaged by Sunn Pest, is necessary. The crop from these areas is stored separately. Skillful use of these measures allows for lowering Sunn Pest damage and receiving high-quality grain.

Modern insecticides, mainly pyrethroides, have a biological efficiency 82-96 % and effective duration of action. Sunn Pest resistance to these pesticides has been reported in the North Caucasian region, in the South of Russia, where it has been used for many years on large areas (1, 37). Intensive application of insecticides does not remove an annual problem of Sunn Pest and complicates a problem in the environment, and is particularly damaging to the natural enemy complex. For example, spring treatments against the overwintered bugs reduced populations of Sunn Pest egg parasites 1.5-3.3 times. Especially, destructive were insecticide and herbicide mixes, resulting in 100 % mortality of useful insects (2).

Studying natural enemies and their efficiency in decreasing Sunn Pest populations and attempts at mass rearing of and introduction of egg parasites has a long history (13, 34). On winter wheat there is no need of applying insecticides when their density is 2 adults/m<sup>2</sup> and infection of the Sunn Pest eggs

by parasites is 40-50 %. Such a situation has been observed when there was 0,6 egg parasites/m<sup>2</sup> and at ratio of predator : Sunn Pest 1 : 2 (2). Mass production of Sunn Pest egg parasites is planned for the future. Technological processes and equipment needs for this have been developed by VIZR (11).

At present Sunn Pest biological control is limited by measures of storage and accumulation of useful insects. For example, introduction of natural enemies in a crop rotation of sunflower, tobacco, Lucerne, millet, corn and rape increased the number of useful insects, and subsequently an increase in control of Sunn Pest's eggs (70-85 % in comparison with 20-40 %) in neighboring wheat fields. Often chemical treatments are cancelled (16).

Research on causes of wheat resistance to Sunn Pest is of great theoretical and practical interest. This work was mainly done at the All-Union Plant Protection Institute (VIZR), beginning in 1958 (17) under the leadership of Professor Shapiro I.D. who showed that wheat resistance to the Sunn Pest depended on their biopolimer complex structure. It was established that Sunn Pest resistant cultivars were characterized by a high content of large starch grains in wheat endosperm. Susceptible varieties were characterized by a large number of small starch grains. Thus, wheat breeding on structural features of biopolymers was the basis for development of breeding wheat varieties for resistance to Sunn pest (31, 35).

opportunity of The using а microbiological method, in particular, application of nematodes against the wintering bugs was studied. Field research was done in Rostov (Russia), with the Yakut strain of Steinernema feltiae Filipjev. This species caused about 48 % mortality of Sunn Pest in wintering places. It was not enough to curtail outbreaks of populations. But in phases of the pest depressions, use of nematodes would considerably reduce volumes of chemical treatments needed (37).

Modern IPM for Sunn Pest must be based on a general system of purposeful construction of complex agroecosystems and agrolandscapes. It provides for maximum heterogeneity of sowings (strengthening the works on immunity), floristically diversity of agrocenosis (with the purposes of strengthening the role of natural enemies), phytosanitary monitoring, using selective pesticides and development of biopesticides on the basis of entomopathogenic microorganisms (16).

## الملخص

زارموخاميدوفا، غ. و أ. ساربايف. 2007. حشرة السونة في وسط آسيا: نظرة تاريخية. الصفحات 61–66. يناقش هذا البحث الوضع التاريخي والحديث لحشرة السونة: المساحة، الحياتية، البيئية، الضرر، العتبة الاقتصادية، وإجراءات المكافحة. سوف تعطى نظرة عامة لتطوير استراتيجيه ووسائل الإدارة المتكاملة لحشرة السونة (في الماضي، الحاضر، والمستقبل).

كلمات مفتاحية: علم البيئة الحيوي، مكافحة، توزع، ضرر، حشرة السونة، قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Biology of Aelia germari Kuster in the Setif Area in Algeria

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### Abstract

Bounechada, M. and M. Fenni. 2007. Biology of *Aelia germari* Kuster in the Setif Area in Algeria. Pages 67-70.

The study was conducted in Setif high plains in the north east of Algeria. The Setif region is one of the important areas for the production of cereal crops. Several insect pests attack cereals, and *Aelia germari* Kuster is one of the most important ones. The objective of this work was to study the life cycle and ecology of *A. germari* under field condition during growing season 1999-2000. The insect had two generation per year and five instars. *A. germari* emerge from winter diapause during early spring (mid-March) when the average temperature is around 18°C. The insect started its summer-winter diapause at the end of August. The duration of the first and second generation was 40 and 35 days, respectively. This study determined the age distribution of *A. germari* populations in relation to the different phenological stages of wheat (*Triticum durum* Desf. var. Mohammed Ben Bachir) and the quality and quantity of damage caused. Information on the different pesticides used against *A. germari* is presented.

Keywords: Aelia germari, chemical control, durum wheat, IPM, Sunn Pest.

## Introduction

Wheat is a major food crop around the world. Various insects and other predators, one of which is Aelia germari Kuster (Hemiptera: Pentatomidae), attack it. This wheat bug breaks down wheat gluten proteins, which are important components in wheat flour. The pest causes losses of 20 to 70% and damage can reach 100% if control measures are not applied. Sunn Pest currently infest an area of around 10-15 million ha worldwide (1). Control is based on the use of pesticides. Data presented in this paper are from a study conducted in the setifian area to compare the phenological development of durum wheat (Triticum durum Desf. var. Mohamed Ben Bachir) and the life cycle and ecology of A. germari. This information can be used for management of the insect in this location and as a survey tool to predict potential cereal damage. Literature on Sunn Pest and cereal bugs in Algeria is limited, and therefore this study provides useful practical information specific to this region.

## Material and Methods

#### **Study Site**

Work was done during the March to August 1999-2000 cropping season in the Setif area (elevation 1100 m.). *Aelia germari* is a common pest in this area. Research was done on 2.5 ha of durum wheat (*Triticum durum* var. Mohamed Ben Bachir). The wheat was planted on 23 November 1999 and harvested on 26 June 2000.

#### Climate

The climatic constraints are rainfall and temperature. This growing season is characterized by an irregular distribution of rain and a high temperature during estival season (May to August). The high plains are marked with rigorous winters. The climate is semi-arid, with cool winters and hot, dry summers (3). The dry season lasts from June to September. The rainfall is irregular. The mean annual rainfall during the growing season is 350 mm. The hottest months are July and August, their average of the maximum temperature are respectively 36°C and 34°C. The insects (second generation) are aestivating

and diapausing form September until mid March.

### **Phenology of Wheat**

For determining the age, distribution of *A. germari* populations in relation to the phenological stage of the crop the Zadok scale is used to measure growth and development of wheat. This measering system ranges from 10 (to designate emergence of the first seedling leaf) to 100 (to designate mature plants with hard grain) (9).

## **Results and Discussion**

#### **Biology of** Aelia germari

To develop appropriate management programs for this cereal pest, further information is needed to better understand its life cycle in the Setif area. Aelia germari normally has two generations per year, although under some unusual conditions, there may be a third. Eggs are laid in groups of twelve divided into two lines of six eggs. The female deposits her eggs on the surface of the stems or leaves of wheat. First generation females can lay 1000 eggs; second generation - 4000 eggs. Aelia germari have five instars. They are completed in 6 wks for the first generation and 5 weeks for the second generation. The egg stage lasts 6-13 days for the first generation and 4-8 days for the second generation. Immatures feed on the leaves and stem for 16-23 days and within 14-18 days newly emerged adults reach sexual maturity. Mating begins 1-2 weeks after the adult stage is reached and females start laying eggs about 1 week after mating. The average lifetime of adults of the first generation is 25 days and 8 to 9 months for the adults of the second generation. Like other Sunn Pest species, only part of A. germari lifecycle is spent in cereal fields (6). The adults of the second generation spend autumn and winter in aestivation and diapause, respectively in areas (on lower elevation mountains) where they are protected from intense heat and severe cold temperatures.

## Distribution of *Aelia germari* in Relation to Different Phenological Stages of Wheat

*Tillering* - The first presence of adults in fields is noted from 28/3/2000. This date corresponded to the end of tillering (scale 26) (Table 1).

*Stem extension* - It began on 9/4/2000 and is finished on 22/5/2000. At this time, we observed the first egg laid by overwintering adults. The first instar was observed on 30/4/2000 corresponding to scale 29 of wheat. Damage caused by adults and first instar was observed on the stems. At 22/5/2000 (scale 31-32), we observed second and third instars and clear reduction of overwintering adults.

*Heading* - It started on 22/5/2000 and finished on 12/6/2000. The dominate instars were 2 and 3 and we noted the appearance of instars 4 and 5 on 9/6/2000 corresponding to the Zadok's scale 55 to 87.

**Ripenning** - Adults of the new generation appeared on 12/6/2000. There was no 3 and 4 instars but a prevalence of instar 5 corresponding to scale 90 of the phenology of We noted on 24/6/2000 wheat. а predominance of new adults and the beginning of mating. This period corresponded to scale 92 of wheat. There were no overwintering adults. Harvest began on 26/6/2000. A great number of new adults died after mating and some produced a new generation that had to develop on wheat straw. New generation adults migrated towards the places of hibernation starting from the end August. These individuals returned to cereal fields in late March the following year.

**Damage** - Like Sunn Pest, nymphs and adults caused damage by feeding on leaves, stems and grains (2). During feeding, they inject chemicals that reduce the baking quality of flour made from damaged grains (5, 8). Wheat bugs like *A. germari*, were found to break down all wheat gluten proteins (1). When the proteins are broken down by this bug, the flour can no longer be used to produce bread etc. Therefore, it is important to the entire wheat food industry to be able to detect the presence of wheat bug damage.



**Table 1**. Relationship between the life cycle of *Aelia germari* and the phenological stage of *Triticum durum* var.Mohamed Ben Bachir in the Setif area during the growing season 1999-2000.

### **Chemical control**

Insecticides are commonly used because they produce results rapidly and effects of materials can be seen. The importance of insecticides for wheat production has not been reduced and there is a challange to use available materials more judiciously. To control *A. germari* several insecticides are used in the Setifian region (Table 2). Fenthion was effective against *A. germari* (4, 7). During the growing season 1999-2000, damage caused by *A. germari* on wheat was low. Climatic conditions (freezing and high temperatures)

and natural enemies contributed to reducing *A*. *germari* populations.

 Table 2. Insecticide rates and concentrations for controlling *Aelia germari* Kuster in Setif area.

Insecticide	Concentration	Rate (Lit/Ha)
Bromophos	350,0 g/l	1.0
Chlorpyriphos-ethyl	38,9 g/l	3.0
Fenthion	51%	1.5
Formothion	330,0 g/l	2.0
Vormidothion	400,0 g/l	2.0

## Conclusions

A. germari is one of the most important pests of cereals in Algeria. When populations are high it is especially serious in the Setifan high plains. Environmental conditions during 1999-2000 contributed greatly in reducing numbers of *A. germari* populations. Differences between the biology of the first and the second generation populations were noted. To make effective chemical applications against this pest, it is important to know these differences and understand the life cycle of the insect and its relationship to the phenology of wheat.

## الملخص

بونشادة، مصطفى ومحمد فني. 2007. حياتية Aelia germari في منطقة سطيف، الجزائر. الصفحات 67–70.

أجريت الدراسة في سهول سطيف المرتفعة في شمال شرق الجزائر، التي تعد واحدة من المناطق الهامة لإنتاج محاصيل الحبوب. تهاجم محاصيل الحبوب عدة آفات حشرية، وتعد حشرة Aelia germari Kuster واحدة من أهم الآفات. إن الهدف من هذا البحث هو دراسة حياتية ودورة حياة حشرة A. germari م تحت الظروف الحقلية خلال موسم النمو 2000/1999. أظهرت النتائج أن للحشرة جيلان في العام وخمسة أطوار. نتبثق A. germari من البيات الشتوي خلال الربيع المبكر (منتصف آذار/مارس) عندما يكون متوسط درجة الحرارة حوالي 18°س. تبدأ الحشرات بياتها الصيفي الشتوي في نهاية آب/أغسطس. بلغت فترة الجيل الأول والثاني 40 و 35 يوماً، على التوالي. حددت هذه الدراسة التوزع العمري لمجتمعات A. germari م بالمراحل الفينولوجية المختلفة للقمح ونوعية وكمية الضرر الذي تسببه. سوف يعرض هذا البحث معلومات عن مبيدات الحشرات المختلفة المستخدمة ضد حشرة A. germari وكمية الضرر الذي تسببه. سوف يعرض هذا البحث معلومات عن مبيدات الحشرات

كلمات مفتاحية: Aelia germari، مكافحة كيميائية، قمح قاسي، IPM، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Biological Parameters of Sunn Pest in Wheat and Barley Fields in Northern Iraq

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## Abstract

Amin, A.M., H.S. Al-Assadi and M.A.J. Al-Izzi. 2007. Biological Parameters of Sunn Pest in Wheat and Barley Fields in Northern Iraq. Pages 71-77.

The appearance of Sunn Pest, *Eurygaster integriceps* Puton, was studied in fields of wheat & barley in the north of Iraq in Erbile district. Twenty cages (100x100x100 cm and 30x30x60 cm) were used either to cover the wheat plants, or to contain pots planted with wheat. One pair of Sunn Pest adults were placed in each cage for oviposition, and newly emerged nymphs were allowed to complete their growth and development within. Incubation period, nymphal development and adult life span were measured. Pre-oviposition, oviposition and post-oviposition periods ranged from 9-11, 19-38 and 1-5 days, respectively. Longevity of hibernated adults ranged from 32-53 and 25-48 days for females and males, respectively. The females mate more than one time in their lifespan. The hibernated females deposited their eggs in April and May. Females lay their eggs in batches of 5-16, each batch contains 8-14 eggs and the total number of eggs for each female ranging between 68-183 eggs. The incubation period ranged from 6 to 14 days with an average 84% egg hatching. The longevity of first, second, third, fourth and fifth instars was 3.4, 5.3, 4.4, 5.5 and 8.4 days, respectively.

Keywords: Biology, Eurygaster integriceps, Sunn Pest, wheat and barley pest.

## Introduction

The Sunn Pest, *Eurygaster integriceps* Puton, is one of the most severe insects invading wheat and barley fields in Iraq (3, 6) and wild sorghum, oats and wild plants grown in wheat fields in the north of Iraq (1, 17). The yield reduction ranged between 20-80% in barley and 20-90% in wheat There are 15 species of Sunn Pest recorded in the Middle East (10), and in particular in Syria, Palestine, Lebanon, Turkey, Iran, Russia, Romania, Bulgaria, Afghanistan, New Zealand and Japan. There are more than 10 million ha (hectare = 4 Donum) of wheat yield invaded by this species in the world and about half of this area is located in the Middle East (21, 22).

The feeding behavior of nymphs and adults, oviposition and damage caused by Sunn Pest were studied on wheat and barley. The objective of this research was to study Sunn Pest biology in wheat and barley fields by using either field cages or field observations. These studies were important for IPM programs for decreasing the insect injury level.

### Materials and Methods

## Biological Studies in Wheat and Barley Fields

Different sizes of metal and wood cages were used in the field:

- 1. Twelve metal cages (100x100x100 cm) covered by window mesh were distributed in 5 ha of wheat in Erbile.
- 2. Six wood cages (30x30x60 cm), the front door made from glass and the sides covered by window mesh were distributed in 5 ha of wheat in Erbile.
- 3. Two wooden cages (30x30x54.5 cm), the front door made from glass and the sides covered by window mesh, plant pots (20x17x15.5 cm and 13.5x11x12.5 cm) containing wheat plants where placed inside each cage.

4. Two wooden cages (25x25x42 cm), the front door made from glass and the sides covered by organza fabric tissue were used for egg hatching and rearing of first and second instars. The hibernated Sunn Pest adults were collected from Seffin Mountain on March 27, 1999, and transferred to insect cages for biological studies. A single male and female were placed in each cage for oviposition, egg incubation period, nymph development and adult longevity. Biological parameter of eggs, nymphs and adults were studied on wheat and barley fields naturally infested by Sunn Pest. The results of field cages were compared with the findings from wheat and barley fields naturally infested by Sunn Pest (5). Sex ratio was determined monthly.

## **Results and Discussion**

### **Biology of Sunn Pest in Wheat and Barley** Fields

Adult mating was observed 4-5 days after males and females were placed in field cages. The pre-oviposition period ranged from 9-11 days from March 27 to April 6 (Table 1). The females started laying eggs on April 5 and continued until May 13, the oviposition period ranged from 19-38 days. The females stayed alive until May 18, the post-oviposition period ranged 1-5 days. However, the hibernated females survived inside the cages for 32 to 53 days.

The hibernated Sunn Pest females deposited their eggs in April and May (Table 2). Some of the females appeared in late March and laid eggs early in April in Koysinjak and Tak-Tak (1998). Egg masses were recorded within 5-19 days after adults appeared in the field. The females resumed egg laying from the end of March to April (6), other females continued laying eggs until mid-May in Iraq (17) and Romania (15).

#### Adult Lifespan

The life span of adults in field cages takes 32-53 days and 25-48 days for female and male, respectively (Table 3). The males die after mating, dead males and females existed on leaves and debris of cereals and grasses in the fields (3). Low temperature (14.5-25.6°C) under laboratory conditions extended the adult lifespan (20).

#### Number of Mating

Female Sunn Pests mate more than one time in their lifespan (Table 4). The first mating occurred during the first 6-10 days, the second mating at 22-36 days of female age, third, fourth and fifth mating at 33-37, 39 and 41 days of female age, respectively.

Mating occurred from April to the beginning of May, the female age ranged 32-53 days and extend to 60 days (12) More than one mating was observed (2, 15).

**Pre-Oviposition** Oviposition **Post-Oviposition** Cage Total No. Dates Dates days Days Dates Days Days 1 27/3 - 5/410 6/4 - 13/538 14/5 - 16/53 51 2 27/3 - 6/411 7/4 - 13/514/5 - 18/55 37 53 3 27/3 - 5/410 6/4 - 9/534 10/5 - 10/51 45 27/3 - 5/44 10 4 6/4 - 12/537 13/5 - 16/551 5 2 27/3 - 6/411 7/4 - 25/419 26/4 - 27/432 3 6 27/3 - 4/49 48 5/4 - 10/5 36 11/5 - 13/510.17 33.5 3 46.67 Avg.

Table 1. Pre-, post and oviposition periods of Sunn Pest in field cages in Northern Iraq.

		Hibernated Sunn Pest records				
Area Name	Year	1 <sup>st</sup> adult appearance	1 <sup>st</sup> records of eggs	Days taken for oviposition		
AinKawa	1998	1/4	12/4	11		
	1999	13/4	22/4	9		
	2000	15/4	22/4	7		
Bansallawa	1998	2/4	13/4	11		
	1999	12/4	17/4	5		
	2000	13/4	23/4	10		
Koshtapa	1998	3⁄4	26/4	13		
	1999	6/4	17/4	11		
	2000	8/4	19/4	11		
Aski-Kalak	1998	1/4	12/4	11		
	1999	15/4	24/4	9		
	2000	17/4	25/4	8		
Shorsh	1998	26/3 *	14/4	19		
	1999	10/4	25/4	15		
	2000	15/5	24/4	9		
Koysinjak	1998	29/3 *	9/4*	11		
	1999	17/4	25/4	8		
	2000	24/4	29/4	5		
Tak-Tak	1998	27/3 *	9/4*	13		
	1999	13/4	21/4	8		
	2000	24/4	4/5 **	10		
Sallah-Idin	1998	28/3 *	14/4	17		
	1999	12/4	23/4	11		
	2000	10/4	18/4	8		
Shaklawa	1998	16/4	30/4	14		
	1999	28/4	5/5 **	7		
	2000	23/4	5/5 **	12		
Harir	1998	10/4	26/4	16		
	1999	20/4	25/4	5		
	2000	17/4	28/4	11		
Khlaifann	1998	17/4	30/4	13		
	1999	25/4	2/5 **	7		
	2000	19/4	1/5 **	12		
Siddik	1998	28/4	10/5 **	12		
	1999	22/4	2 /5 **	10		
	2000	26/4	9/5 **	13		
Rawandozz	1998	27/4	9/5 **	12		
	1999	24/4	5/5 * *	11		
	2000	27/4	8/5 **	11		
Mirkassour	1998	30/4	16/5 **	16		
	1999	28/4	4/5 **	6		
	2000	21/4	30/4	9		
Sidda-kan	1998	5/5 **	20/5**	15		
	1999	3/5 **	15/5**	12		
	2000	30/4	14/5**	14		
Joman	1998	29/4	11/5**	12		
	1999	26/4	6/5**	10		
	2000	29/4	10/5**	11		
Barazan	1998	1/5 **	9/5**	8		
	1999	2/5 **	7/5**	5		
	2000	13/4	22/4	9		
	2000	13/4	22/4	7		

Table 2. Oviposition of Sunn Pest in different areas in the north of Iraq in the years 1998, 1999 and 2000.

\* Early date for oviposition \*\* Late oviposition

**Table 3.** Adult lifespan of Sunn Pest, *Eurygaster integriceps*Puton, inside field cages in NorthernIraq, 1999.

Cage	Female lifespan		Male lifespan		
No.	Period	Days	Period	Days	
1	27/3-16/5	51	27/3-10/5	45	
2	27/3-18/5	53	27/3-13/5	48	
3	27/3-10/5	45	27/3-20/4	25	
4	27/3-10/5	45	27/3-8/5	43	
5	27/3-16/5	51	27/3-12/5	47	
6	27/3-27/4	32	27/3-7/5	42	
7	27/3-13/5	48	27/3-22/4	27	
8	27/3-13/5	48	27/3-5/5	40	
Avg.		46.67		39.63	

**Table 4.** Number of matings of Sunn Pest,*Eurygaster integriceps* Puton, inside field cages inNorthern Iraq, 1999.

	Age	Female					
Female No.	1 <sup>st</sup> Mt	1st2nd3rd4th5thMtMtMtMtMt					
1	6	-	-	-	-	32	
2	9	36	-	-	-	45	
3	7	31	-	-	-	53	
4	7	36	37	-	-	51	
5	6	22	33	-	-	51	
6	10	31	36	39	41	48	
Avg.	7.5	26	35.3	39	41	46.7	

## Sex Ratio

The sex ratio was close to 50-50 during June. In general, a higher number of females was recorded in April, May, and August 1999 to March 2000 due to the death of males after mating (Table 5).

The differences in ratio of males to females was due to the differences of ecological conditions which have adverse effects on insect life cycles in the field and in the overwintering sites (17, 13, 16, 4, 18, 11).

#### **Female Oviposition Behavior**

Sunn Pest deposit their eggs in groups, their numbers ranged from 5 to 16 egg mass per female, 8-14 eggs per mass (Figure 1). The female oviposited 68-83 eggs during its life (Table 6). There are differences in number of masses and number of eggs per mass; it was 12-14 eggs per mass in 2-4 parallel lines (23) and 14-15 egg mass containing 42-210 eggs for the single female (17).

It was recorded that female deposited from 150-200 eggs (8, 14). The migrated females deposited twice number of eggs compared with non-immigrant females (18).

**Table 5.** The ratio of males to females of Sunn Pest, *Eurygaster integriceps* Puton inside field cages in Northern Iraq, 1999 and 2000.

	No. of males and females in the field			
Month	Male Female			
April 1999	9	11		
May	7	13		
June	1	1		
July	5	3		
August	3	7		
September	1	3		
October	1	5		
November	1	9		
December	1	5		
January 2000	6	14		
February	1	2		
March	1	9		

**Table 6.** Oviposition of Sunn Pest, *Eurygasterintegriceps* Puton, inside field cages in NorthernIraq.

Cage No.	Egg masses	Avg. No. eggs per mass	Total eggs
1	16	11.25±0.497	180
2	15	11.13±0.496	167
3	9	13.22±0.324	119
4	13	11.69±0.683	152
5	5	13.60±0.245	68
6	16	11.43±0.524	183
Avg.	12.33	12.05	144.83

## Eggs Hatching (Embryo Development Inside the Eggs)

The newly deposited eggs are brilliant green then they change to faded green with two red spots which are the eyes of the nymph growing inside the egg, the egg color turned to yellow-green before hatching and the embryo appeared as a red stripe inside the egg. The embryo development takes 6 to 14 days (Table 7).

The percentage of hatching ranged between 66.67 to 100% with the average 84.19%. The eggs deposited by females at the end of the oviposition period take a shorter interval for hatching compared with those laid at the beginning of the oviposition period. The incubation period and nymphal egg development depends on host availability and environmental conditions. Different incubation periods were recorded previously; from 7 to 20 days (19), 6 to 16 days (3) and 10 to 20 days under field conditions (11).



**Figure 1.** The correlation between accumulated number of Sunn Pest eggs and the female age on wheat plants inside field cages in 1999 and 2000 in Northern Iraq.

#### Nymphal Development

Sunn Pest nymphs pass through five instars to reach the adult stage. The average duration of each instar was 3.42, 5.32, 4.4, 5.5 and 8.41 days for the  $1^{\text{st}}$ ,  $2^{\text{nd}}$ ,  $3^{\text{rd}}$ ,  $4^{\text{th}}$  and  $5^{\text{th}}$  instars, respectively (Table 8).

The period of nymphal instars was recorded by other authors (6, 11). The total nymphal development takes 30 days (19), ranged from 25-28 days and 27-31 days under field and laboratory conditions, respectively (7) and 45-60 days in Iran (9, 12).

**Table 7.** Number of eggs per mass, incubation period and hatching percent of Sunn Pest eggs in wheat and barley fields in Northern Iraq.

	No. of		
Egg	Eggs	Incubation	
Mass	per	Period	
No.	Mass	(days)	Hatching%
1	14	14	78.57
2	13	14	92.31
3	13	13	76.92
4	14	13	100.00
5	14	13	71.42
6	12	10	91.67
7	12	9	75.00
8	12	11	100.00
9	12	9	66.67
10	8	10	100.00
11	12	8	83.33
12	11	7	72.73
13	10	6	70.00
14	10	6	100.00
Avg.	11.93	10.21	84.19

**Table 8.** The duration of nymphal instars of Sunn

 Pest, *Eurygaster integriceps* Puton, on wheat plants

 inside field cages in Northern Iraq.

Instars No. of Nymph			nph per	h period (days)		
mstars	nymphs	Min.	Max.	Average		
1 <sup>st</sup>	26	3	4	$3.42\pm0.99$		
2 <sup>nd</sup>	25	5	6	$5.32 \pm 0.095$		
3 <sup>rd</sup>	20	4	5	4.40±0.112		
4 <sup>th</sup>	18	5	6	5.50±0.121		
5 <sup>th</sup>	17	6	11	8.41±0.352		
Total	-	23	32	27.05		

## الملخص

أمين، عبد الباسط م.، حسن س. الأسدي، و محمد ع. ج. العزي. 2007. المؤشرات الحيوية لحشرة السونة في حقول القمح والشعير في شمال العراق. الصفحات 71–77.

تم دراسة ظهور حشرة السونة (Eurygaster integriceps)، في حقول القمح والشعير في شمال العراق في مقاطعة إربيل. استخدم عشرون قفصاً (100×100×100 سم و 30×30×00 سم) إما لتغطية نباتات القمح، أو لتحتوي أصص مزروعة بالقمح. وضع في كل قفص زوج من حشرات السونة البالغة لأجل وضع البيض، وسمح للحوريات المنبثقة حديثاً بإكمال نموها وتطورها فيه. تم قياس فترة الحضانة، تطور الحورية، وطول فترة حياة الحشرة البالغة. تراوحت فترات ما قبل وضع البيض، وضع البيض وما بعد وضع البيض من 9–11، 19–38 و 1–5 أيام، على التوالي. تراوح طول فترة حياة البالغات المشتية من 22-53 و 25–48 يوماً للإناث والذكور، على التوالي. نتزاوج الإناث أكثر من مرة خلال فترة حياتها. تضع الإناث المشتية بيضها في نيسان/أبريل وأيار/مايو، في مجموعات من 5–10 مجموعة، كل مجموعة تحتوي 8–14 بيضة والعدد الكلي من البيض لكل أنثى يتراوح بين 68–183 بيضة. تراوحت فترة الحضانة من 6 إلى 14 يوماً متوسط نسبة فقس للبيض للبيض البيض حياة طور الحورية الأول، الثاني، الثالث، الرابع والخامس 3.4، 5.3، 4.4 ولي م عنوي ما بيخة وقل البيض والعدد الكلي من البيض كلمات مقتولية الأول، الثاني، الثالث، الرابع والخامس 3.4، 5.3، 4.4 وم عمول نسبة فقس للبيض والماد الم البيض موات معروبية الأول، الثاني، الثالث، الرابع والخامس 3.4، 5.3، 4.4 و قر م م عليه والديم واليولي م علي الم م علي فترة محياة طور الحورية الأول، الثاني، الثالث، الرابع والخامس 3.4، 5.3، 4.4 و م 2.5 و 4.5 يوماً، على التوالي.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Population Density of Sunn Pest during Hibernation and Aestivation in Northern Iraq

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#### Abstract

## Amin, A.M., M.A.J. Al-Izzi and H.S. Al-Assadi. 2007. Population Density of Sunn Pest during Hibernation and Aestivation in Northern Iraq. Pages 79-88.

The seasonal appearance of Sunn Pest was studied in Northern Iraq. Hibernated insects resumed their activity in the second half of March in 1998, 1999 and 2000. Newly emerged adults appeared in the second half of May and the first week of June. The population density of hibernated adults ranged between 0.58-11.83 adults/10 m<sup>2</sup> in the three years. Females lay eggs during April and the number of egg masses in the fields ranged from 1-39, 1-10 and 1-11 masses/10m<sup>2</sup> during 1998, 1999 and 2000, respectively. Density of nymphs and new adults during the three years ranged from 0.08 to 119.26 and 1.11 to 99.41 insect/10 m<sup>2</sup>, respectively. The movement of the insects within the different regions of the mountain was observed from November, 1999 to March, 2000. The highest population density in the mountain summit and sides decreased gradually from 97.75 and 20.75 in November to 15.33 and 13.33 adults/10 m<sup>2</sup> in March, respectively. The population density increased in the mountain base from 11.75 in November to 25.3 adults/10 m<sup>2</sup> in March. The population density of adults in the mountains; Hassan Beg, Hindrin, Kalander, Shereen, Baradost, Zozic, Korec, Kearan and Safin were 916, 100, 1458, 720, 943, 25, 1095, 766, and 295 adults/10 m<sup>2</sup>, respectively. These numbers were different from those recorded in 2000, in which Hassan Beg, Shereen, Bradost, Korec, Pearan and Safin were: 76, 146, 308, 373, 692 and 117 adults/10 m<sup>2</sup>. During hibernation and aestivation, Sunn Pest migrated to the following plants: Prunus argentea (Lam.) Rehd., Astragalus russelii Banks & Sol., Thymbra vulgaris L. (Thyme), Euphorbia peplus L., Quercus infectoria Olivier, Quercus aegilops L., Pistacia atlantica Desf., Crxpis parviflora, Platanus orientalis L., Teucrium chamaevrys L. subsp. sinuatum (Celak.) Rech., Telephium oligospermum Boiss., Nepeta sp., Silene ampullata Boiss., S. stenobotrys Boiss & Hausskn and Onosma albo-roseum Fisch. et Mey.

Keywords: Eurygaster integriceps Puton, hibernation, population density, Sunn Pest.

## Introduction

The Sunn Pest, *Eurygaster integriceps* Puton, was first recorded in Iraq in 1920 (15) and spread to most areas in the north of the country (Erbile, Dohok, Nainawa and Sulaimanya) (5). It was recorded around Baghdad and Diala (1). Sunn Pest can completely destroy the grain and cause a high economic yield loss, it is the most dangerous pest of wheat, wild sorghum, barley, oats and wild plants grown in wheat fields in Northern Iraq (1, 15). The yield reduction ranged between 20-80% of barley and 20-90% of wheat. Loss of cereal yield is a result of Sunn Pest feeding by piercing and sucking nutrients

from seeds, causing weight reduction of the infested seeds. High populations of Sunn Pest can completely destroy wheat or barley (17). There are two migrations of Sunn Pest, the spring migration of over wintered adults from the mountains to cereal fields (wheat and barley) in the flat areas for feeding and egg laying and the summer migration to mountains where the new adults aestivate and later hibernate. They hibernate in shelters of dry leaves, grass clumps, plants and rock. The object of this research was to study the annual existence of Sunn Pest in the mountains and cereal fields, determination the population density and insect development in the field, the relation between insect migration, summer aestivation and winter hibernation as related to ecological conditions.

## Materials and Methods

## Study the Annual Existence of Sunn Pest and its Developmental Stages in the Fields

The developmental stages of Sunn Pest in wheat and barley fields were studied during the second week of March and the observations continued to the second week of June in the plains of the villages of Ain-Kawa, Binsilawa, Aski-Kalak, Koshtoba, Shorsh, Koisinjack, Tak-Tak, Salah-Eldin, Shaklawa, Harir. Khlaifan. Siddiq, Rawandozz. Mirgasour, Sida-kan, Joman and Barazan (Figure 1). The sampling unit was divided to 10 subunits, each is 1 m<sup>2</sup>. A wooden square frame (100x100 cm), two wooden sides connected by strong cord (3) was used in the field, the frame was randomly thrown in each subunit to find the number of nymphs and adults of Sunn Pest per square meter. The frame was thrown ten times randomly in (X) shape in order to count the number of insect stages per 10 square meters. The egg masses were detected on the broad leaves grasses and on wheat leaves.



**Figure 1.** The Villages of Erbile in which the Sunn Pest Existence where studied

Samples of Sunn Pest stages were transferred to the laboratory for biological studies under field cages. Plant samples were dried for identification by personnel at the Iraqi National Herbarium.

### Study the Annual Existence of Sunn Pest and its Developmental Stages in the Mountains

Most of the studies were initiated in Sefin Mountain in Erbile Governorate.

- Weekly visits to study the summer aestivation of Sunn Pest started from July to November of 1999. The wooden square frame (one square meter) was used to determine the population density of Sunn Pest/10 m<sup>2</sup>. The plants resort to by Sunn Pest for sheltering in the mountain area were identified.
- Bi-weekly visits to study the winter hibernation of Sunn Pest started from December 1999 to March 2000 in Seffin Mountain. The number of hibernated insects underneath the plants was determined. Samples of insects and plants were transferred to the laboratory for identification.
- Sunn Pest in the mountains were counted at three elevations: base (1-650 m), slopes (651-1290 m) and the
- summit (1291-1938 m). A special instrument was used to measure the elevation. The number of dead and alive insects was determined. Temperature and relative humidity were obtained from a Climatologically Station in Erbile City or by direct measurements using thermo or hydro-meters in the hibernation areas.
- Data on Sunn Pest presence and population density were obtained in July in the following mountains; Seffin, Bawakoup, Wazardikan, Bany-harir, Kork, Hindrin, Zozak, Hassan-beck, Bradost, Kalindro-Biran and Sherin.

## **Results and Discussions**

### Annual Existence of Sunn Pest and its Developmental Stages in Wheat and Barley Fields

Weekly surveys of fields in Ain-Kawa, Binsilawa, Aski-Kalak, Koshtoba, Shorsh, Koisinjack, Tak-Tak, Salah-Eldin, Shaklawa, Khlaifan, Siddiq, Rawandozz, Harir, Mirgasour, Sida-kan, Joman and Barazan showed the existence of Sunn Pest adults and nymphs from March to July of 1998, 1999 and 2000. The hibernated adults resumed their activities from the second half of March of 1998, 1999 and 2000. They moved from the mountains to the cereal fields at the end of March. At first they moved down to the base of the mountain where they fed on a number of grasses and gramineous plants grown on mountainside and then moved into wheat and barley fields.

Sunn Pest adults were recorded at the end of March 1998 in Shorsh, Koisinjack, Tak-Tak and Salah-Eldin where the temperature ranged between 13.4 and 18.3°C. Adults appeared in April in Ain-Kawa, Binsilawa, Koshtoba and Aski-Kalak where the temperature ranged between 13-15.1°C. The remaining adults appeared at the end of April and continued to May until the temperature rose to a higher level (Table 1). Sunn Pest resumed their activities at the second week of April and continued through the following weeks in 1999 and 2000, except in Koshtoba in 1999. The 13°C temperature motivated the migration of Sunn Pest from the hibernated areas to wheat and barley fields in Ain-Kawa, Aski-Kalak and Shorsh in 1998 and in Sallah-Eldin in 2000. Migration depends on ecological conditions as well as temperature, relative humidity, rains and winds (4, 7). In other areas, Sunn Pest started their migration at temperature range 16.9-18°C in Russia (10), at 18°C in Turkey (8) and at 22.5°C in Romania (14). The adults fly during the day and night at the temperature range 16-18°C and the photoperiod has a direct effect on Sunn migration in Russia (9).

Table 1. Existence of Sunn Pest, *Eurygaster integriceps* Puton in plain fields in the north of Iraq during the years 1998, 1999 and 2000.

	Date of first record of hibernated Sunn Pest						
Area name	1998	Temp. °C	1999	Temp. °C	2000	Temp. °C	
Ain-Kawa	1/4	13.0	13/4	19.2	15/4	22.0	
Binsilawa	2/4	14.3	12/4	20.8	13/4	25.0	
Koshtoba	3/4	15.1	6/4	20.0	8/4	21.2	
Aski-Kalak	1/4	13.0	15/4	18.9	17/4	25.5	
Shorsh	26/3	13.4	10/4	16.4	15/5	22.0	
Koisinjack	29/3	14.6	17/4	19.9	24/4	21.3	
Tak-Tak	27/3	17.0	13/4	19.2	24/4	21.3	
SalahEldin	28/3	18.3	12/4	20.8	10/4	13.0	
Shaklawa	16/4	25.8	28/4	25.6	23/4	21.9	
Harir	10/4	23.4	22/4	21.8	17/4	25.5	
Khlaifan	17/4	21.6	25/4	26.6	19/4	26.7	
Siddiq	28/4	23.6	22/4	21.8	26/4	23.0	
Rawandozz	27/4	23.6	24/4	23.0	27/4	25.5	
Mirgasour	30/4	19.2	28/4	25.6	22/4	21.5	
Sida-kan	5/5	24.3	3/5	26.0	30/4	19,3	
Joman	29/4	18.4	26/4	26.7	29/4	21.4	
Barazan	1/5	20.6	2/5	26.6	18/4	27.6	

## Population Density of Hibernated Sunn Pest Adults Migrated to Wheat and Barley Fields

The density of hibernated adults in wheat and barley fields ranged from 1-10 adults /10 m<sup>2</sup> in 1998, higher densities were recorded in Joman (10.0), Tak-Tak (7.6), Khlaifan (7.4) and decreased to one adult/10 m<sup>2</sup> in Sida-kan. Higher adult densities were recorded in 1999 in SalahEldin (36.0), Shorsh (23.0) and Joman (8.1) and in 2000 in Shorsh (11.8), Koisinjack (11.2) and Tak-Tak (10.5) (Table 2).

**Table 2.** The highest density of hibernated adults/10  $m^2$  of Sunn Pest, *Eurygaster integriceps* Puton, in the north of Iraq in 1998, 1999 and 2000.

	Highest density of hibernated adults per 10 m <sup>2</sup>					
Area Name	1998	1999	2000			
Ain-Kawa	3.58	6.00	0.78			
Binsilawa	6.00	0.58	1.67			
Koshtoba	2.92	1.75	0.33			
Aski-Kalak	5.58	1.66	0.33			
Shorsh	4.92	23.00	11.83			
Koisinjack	4.58	1.66	11.25			
Tak-Tak	7.60	1.50	10.50			
SalahEldin	6.91	36.00	1.56			
Shaklawa	2.33	4.17	0.64			
Harir	6.64	1.66	3.78			
Khlaifan	7.40	2.25	1.22			
Siddiq	2.92	3.33	7.33			
Rawandozz	6.00	3.83	6.00			
Mirgasour	1.00	6.66	6.22			
Sida-kan	3.00	4.50	2.56			
Joman	10.00	8.16	6.00			
Barazan	3.83	3.75	3.10			

Egg masses were found in wheat and barley in 1998, 1999 and 2000. Females deposited their eggs in April and May. The number of egg masses ranged from 1 to 39, 10 and 11 egg mass for the years 1998, 1999 and 2000, respectively (Table 3).

The first instars were present at the second half of April and May, they developed to fifth instars in May and June (Table 4). Nymphs were in the fields for 12 to 42 days with an average of 29.31 days. Nymphal density ranged from 0.08 to 119.26 nymph/10  $m^2$  for the three years.

**Table 3.** Number of egg masses of Sunn Pest in wheat and barley fields in the north of Iraq in 1998, 1999 and 2000.

	Egg masses of Sunn Pest in the field					
Area	1998		19	99	2000	
name	Apr	May	Apr	May	Apr	May
Ain-Kawa	23	16	5	-	-	-
Bansalawa	14	4	2	-	1	-
Koshtoba	8	2	5	-	2	-
Aski-Kalak	37	2	-	-	-	-
Shorsh	20	6	10	4	-	-
Koysinjack	27	3	3	3	-	-
Tak-Tak	19	4	2	-	-	-
SalahEldin	39	18	4	1	5	1
Shaklawa	-	3	-	1	-	-
Harir	-	30	4	2	2	-
Khlaifan	-	8	-	5	-	2
Siddiq	-	1	-	5	-	2
Rawandozz	-	2	-	3	-	4
Mirgasour	-	1	-	-	3	5
Sida-kan	-	-	-	1	-	-
Joman	-	2	-	6	-	1
Barazan	-	3	-	1	11	1

## Occurrence of Newly Developed Adults of Sunn Pest

The new adults of Sunn Pest coexisted with other instars in wheat and barley. Their appearance was generally in May but some adults were found late in June 2000 in the Siddiq, Mirgasour and Barazan areas (Table 5).

The adults fed for 10-20 days on wheat spikes, storing nutrients and then made their way upward to the mountains. At that time the air temperature in the fields ranged between 21-34.8°C. The higher temperatures caused the insects to migrate more quickly from the fields to the mountain areas.

### Density of Newly Developed Sunn Pest Adults

Nymphal development was studied on wheat and barley plants under field conditions. The density of newly developed adults was higher than the hibernated adults for the years 1998, 1999 and 2000 (Table 6). It ranged from 1.11 to 99.41 adults/10 m<sup>2</sup> for the three years. Higher densities were recorded in the

			Nymph density and existence in the field			
		First record	Density ran	nge per 10m <sup>2</sup>	Last record	Nymph
Area	Year	of 1 <sup>st</sup> Instars	Low	High	of 5 <sup>th</sup> instars	existence (days)
Ain-Kawa	1998	25/4	1.58	38.16	26/5	31.0
	1999	14/4	0.66	46.91	24/5	40.0
	2000	26/4	0.66	10.86	23/5	27.0
Bansalawa	1998	26/4	0.83	29.14	3/6	38.0
	1999	4/5	4.66	46.33	18/5	14.0
	2000	25/4	3.44	10.22	20/5	25.0
Koshtoba	1998	2/5	0.58	25.58	4/6	33.0
	1999	22/4	0.16	39.00	24/5	32.0
	2000	2/5	0.77	19.45	28/5	26.0
AskiKalak	1998	2/5	1.00	115.00	3/6	32.0
	1999	27/4	8.33	38.75	20/5	23.0
	2000	29/4	7.59	14.22	23/5	24.0
Shoursh	1998	29/4	1.58	51.00	8/6	40.0
	1999	27/4	0.08	48.44	31/5	34.0
	2000	16/5	15.67	35.50	6/6	21.0
Koiysinjack	1998	21/4	0.42	30.50	2/6	42.0
5 5	1999	21/4	1.33	33.62	28/5	37.0
	2000	10/4	0.92	28.42	24/5	14.0
Tak-Tak	1998	23/4	0.17	37.50	3/6	41.0
	1999	25/4	0.41	26.00	25/5	30.0
	2000	29/4	0.25	18.14	18/5	19.0
SalahAldin	1998	28/4	0.16	34.26	4/6	37.0
	1999	16/4	0.58	48.92	26/5	40.0
	2000	3/5	4.56	42.89	28/5	25.0
Shaklawa	1998	6/5	0.16	26.50	15/6	40.0
	1999	10/5	0.82	49.25	2/6	23.0
	2000	6/5	5.11	20.63	5/6	30.0
Harir	1998	4/5	0.50	44.91	10/6	37.0
	1999	29/5	0.08	119.26	1/6	33.0
	2000	20/4	0.67	23.78	1/6	42.0
Khlaifan	1998	7/5	0.75	31.91	17/6	41.0
	1999	1/5	2.83	20.83	31/5	30.0
	2000	8/5	4.00	12.89	31/5	23.0
Siddia	1998	14/5	0.08	8.58	13/6	30.0
~1	1999	10/5	11.66	23.00	27/5	17.0
	2000	14/5	0.40	22.00	6/6	23.0
Rawandooz	1998	17/5	1.58	15.41	6/6	20.0
	1999	5/5	3.08	28.00	30/5	25.0
	2000	14/5	1.43	39.33	7/6	24.0
Mirkasour	1998	10/5	0.16	10.91	12/6	33.0
	1999	3/5	0.17	26.50	6/6	34.0
	2000	4/5	1.33	10.89	3/6	30.0
Sidda-Kann	1998	25/5	0.83	3.08	14/6	20.0
Stoou Humi	1999	14/5	12.83	17.00	26/5	12.0
	2000	18/5	5.11	26.67	12/6	25.0
Ioman	1998	16/5	0.33	21.50	25/6	40.0
Jonan	1999	8/5	2.00	22.50	3/6	26.0
	2000	16/5	6.22	38.00	27/6	42.0
Barzann	1998	11/5	0.22	23.16	31/5	20.0
Durbuill	1000	2/5	2.20	70.16	1/6	20.0
	1777	2/3	2.20	10.10	1/0	50.0
	2000	30/4	0.78	12.33	19/5	20.0

Table 4. Population density and existence of Sunn Pest nymphs in the north of Iraq, 1998, 1999 and 2000.

\* Average nymph existence in the field= 29.31373

following localities: in 1998; Aski-Kalak (71.25), Shorsh (38.33) and Barazan (31.0). In 1999: Harir (99.41), Shaklawa (87.18) and Barazan (59.4); in 2000: Shorsh (11.83), Koisinjack (11.25) and Tak-Tak (10.5). The higher densities of new adults caused more expensive damage to wheat and barley seeds. This was observed in all localities except in

Koshtoba in 1999. Grain damage is proportional to the Sunn Pest population. The adults completed their development at the time of harvest and aggregated in high numbers in the delayed harvested fields causing severe damage to the grains in the years 1998, 1999 and 2000.

**Table 5.** The date of first appearance of new adults of Sunn Pest *Eurygaster integriceps* Puton in 1998, 1999 and 2000.

	Temperature and date of first appearance of new adults					
Area name	1998	Temp. °C	1999	Temp. °C	2000	Temp. °C
Ain-Kawa	28/5	33.4	22/5	27.4	21/5	28.0
Binsilawa	21/5	21.3	17/5	32.1	20/5	25.4
Koshtoba	19/5	26.0	24/5	32.3	27/5	29.5
Aski-Kalak	23/5	23.2	20/5	24.6	23/5	30.0
Shorsh	20/5	26.1	20/5	24.6	18/5	25.9
Koisinjack	23/5	23.2	24/5	32.3	26/5	29.1
Tak-Tak	21/5	21.3	18/5	30.4	22/5	29.2
SalahEldin	25/5	29.1	22/5	27.4	24/5	31.3
Shaklawa	26/5	30.0	22/5	27.3	29/5	30.4
Harir	24/5	26.6	26/5	31.4	29/5	30.4
Khlaifan	27/5	32.1	24/5	32.3	25/5	31.5
Siddiq	28/5	33.4	27/5	30.7	1/6	31.0
Rawandozz	27/5	32.1	29/5	34.8	27/5	29.5
Mirgasour	30/5	28.7	25/5	33.3	3/6	31.7
Sida-kan	25/6	29.1	26/5	31.4	30/5	31.5
Joman	1/6	29.4	31/5	31.4	28/5	28.4
Barazan	31/5	30.4	25/5	33.3	4/6	31.8

**Table 6.** Density of newly developed adults of Sunn Pest, *Eurygaster integriceps* Puton in the north of Iraq in 1998, 1999 and 2000.

_	Higher density and date of newly developed adults /10 m <sup>2</sup>							
	1998	3	1999	)	2000	)		
Area Name	Density	Date	Density	Date	Density	Date		
Ain-Kawa	18.50	2/6	11.00	23/5	1.29	22/5		
Binsilawa	1.33	27/5	14.44	27/5	1.11	20/5		
Koshtoba	16.66	28/5	1.92	24/5	3.33	27/5		
Aski-Kalak	71.25	30/5	10.83	20/5	3.11	23/5		
Shorsh	38.33	20/5	29.89	25/5	11.89	10/6		
Koisinjack	29.08	7/6	26.75	27/5	22.89	6/6		
Tak-Tak	24.50	29/5	12.66	19/5	52.19	22/5		
SalahEldin	15.13	4/6	48.75	26/5	5.44	28/5		
Shaklawa	6.58	15/6	87.18	2/6	11.00	4/6		
Harir	27.50	10/6	99.41	27/6	7.50	31/5		
Khlaifan	7.37	17/6	32.83	30/5	3.56	30/5		
Siddiq	17.00	7/6	12.83	5/6	7.78	1/6		
Rawandozz	19.83	16/6	27.50	29/5	21.78	3/6		
Mirgasour	29.16	9/6	38.33	6/6	13.33	17/6		
Sida-kan	4.92	14/6	15.00	7/6	30.67	11/6		
Joman	18.75	18/6	25.00	31/5	38.89	21/6		
Barazan	31.08	10/6	59.46	1/6	19.17	4/6		

## Migration of Sunn Pest Adults from the Plains to the Mountains

The newly developed adults prepared themselves to migrate from wheat and barley fields to the hibernated areas in the mountains. Before their departure in June, they aggregated in groups on wheat spikes for feeding. Wheat harvest activities and a gradual increase in ambient temperatures induced movement of the adults from one field towards the neighboring ones. Some adults were harvested with the wheat grains and could be found in grain bushels. Other insects were not able to migrate to the mountains, instead they stayed in the same areas spent the summer and winter there. Physiological conditions of the adults, grain harvest and higher temperatures are the main reasons causing migration of the Sunn Pest adults to the mountains. Temperature increases from 25 to 43°C motivated the adults for migration. They flew 10 to 50 km to the hibernation sites in the mountains. They can fly from 2000 to 2500 m elevation in Iran (6).

The date of migration of new adults was recorded in June and the first week of July. They first migrated towards the mountain base, then stepwise moved to the upper levels of the mountainside to reach the mountain top. They spend summer aestivation underneath the plants.

The number of Sunn Pest aggregated in Seffin Mountain ranged from 284 to 331 adults/10 m<sup>2</sup> with an average 301.61 adults/10 m<sup>2</sup> (Table 7). Numbers of adults increased in August to reach 344.1 adults/10 m<sup>2</sup> and decreased in September and October to 195.75 and 130.25 adults /10 m<sup>2</sup>, respectively.

The decrease in adult populations could be due to the effect of ecological and biological conditions (parasites, predators and disease causal organisms). Sunn Pest adults enter summer aestivation on the mountain summit. They resume their movement from the top to the mountain sides and to the base of the mountain. The overwintering adults hide under the plants for protection from winter conditions. This same behavior was observed in other areas (2, 4, 12). These hibernated insects also hide under fallen leaves and rooks for months to protect from winter severe conditions (12).

**Table 7.** Accumulation of Sunn Pest adults, *Eurygaster integriceps* Puton, per  $10 \text{ m}^2$  on the top of Seffin Mountain from July to October of 1999, north of Iraq.

	Sunn Pest adults / 10 m <sup>2</sup>					
Month	Low	High	Avg. Ë SD			
July	284	331	301.66± 12.10			
August	105	612	$74.11 \pm 34.10$			
September	175	216	$195.75 \pm 7.25$			
October	80	160	$130.25 \pm 15.60$			
November	15	222	$97.75 \pm 42.10$			
December	70	121	$121.00 \pm 12.60$			
January	17	42	$29.50 \pm 8.80$			
February	20	29	$24.50\pm3.10$			
March	14	16	$15.33\pm0.50$			

Sunn Pests hide under many plants for summer aestivation and winter hibernation. These plants grow naturally on the top of the mountains at different altitudes, generally higher than 1900 meter and include: Astragalus russelii Banks & Sol., Prunus argentea (Lam.) Rehd., Euphorbia peplus L., Thymbra vulgaris L. (Thyme), Quercus aegilops L., Quercus infetoria Olivier, Pistacia atlantica Desf., Crxpis parviflora, Platanus orientalis L., Teucrium chamaevrys L. subsp. sinuatum (Celak.) Rech, Telephium oligospermum Boiss, Nepeta sp., Silene ampullata Boiss., S. stenobotrys Boiss & Hausskn, Onosma albo-roseum Fisch. et Mey. Some of these plants have been recorded previously (5, 16). They do not exist in mountainsides or at the base. In addition, to providing shelter to adults, they save the insects from critical conditions of temperature, relative humidity, winds, sunlight and natural enemies. These plants are grown in high densities and provide higher moisture favored by the hibernated adults during summer aestivation. Other Sunn Pest hides in the soil for higher moisture. These plants were also recorded in the top of Seffin Mountain. Adults preferred the dens and shaded plants which gave them lower temperatures and higher moisture (Table 8).

	Underne Pla	eath the nt	Ambient		
Month	Temp. °C	R.H.	Temp. °C	R.H.	
July	28.0	39.5	29.3	28.0	
August	26.8	42.5	30.6	32.8	
September	21.4	45.7	23.8	37.7	
October	16.6	50.1	18.0	44.2	
November	9.5	57.6	7.9	60.0	
December	9.2	61.7	5.3	68.5	
January	2.2	61.5	0.0	88.0	
February	8.7	64.0	3.8	80.7	
March	14.3	61.7	9.0	63.1	

**Table 8.** The comparison in temperature and relative humidity in air and underneath the plants in summer aestivation and hibernation areas.

#### **Insect Movement in the Mountains**

Sunn Pest accumulated in mountain top and moved toward the warmest microhabitats underneath the plants as a wind shields, therefore, the insects moved downward toward the mountainside and to the base. Insect downward movement was observed from November to March (Table 9).

Table 9. Population density of Sunn Pest,Eurygaster integriceps Puton, in the mountain top,surroundings and base from November 1999 toMarch 2000.

	Population Density (adults /10 m <sup>2</sup> )							
	Months							
	Nov. Dec. Jan. Feb. Mar.							
Mountain Top								
Low	15.0	70.0	17.0	20.0	14.0			
High	222.0	121.0	42.0	29.0	16.0			
Avg.	97.8	89.1	29.5	24.5	15.3			
Mountain Surr	ounding	S						
Low	8.0	17.0	17.0	13.0	8.0			
High	31.0	21.0	17.0	15.0	18.0			
Avg.	20.8	18.7	17.0	14.0	13.3			
Mountain Base								
Low	6.0	13.0	10.0	16.0	24.0			
High	29.0	15.0	21.0	22.0	26.0			
Avg.	11.8	14.0	15.5	19.0	25.3			

Insect population density in the mountain summit and sides decreased stepwise from November to March. The mean number were decreased gradually from 97.75 and 20.75 in November to 15.33 and 13.33 adults/10 m<sup>2</sup> in March, respectively, while the density increased in the mountain base from 11.75 in November to 25.3 adults/10 m<sup>2</sup> in March.

Sunn Pest spend summer aestivation and winter hibernation from July to March in the mountain areas, they pass from 240 to 270 days as dormant adults underneath the plants or fallen leaves. Summits of Hassan Begg, Hindrin, Klinder, Sherine, Bradost, Zozak, Kork, Biran and Seffin mountains were used for Sunn adult hibernation studies. The population density was recorded in July 1999. The numbers of hibernated adults were 916, 100, 1458, 720, 943, 25, 1095, 766, and 295 adults/10 m<sup>2</sup>, respectively. These numbers were higher than those recorded in June 2000 (Table 10).

In winter, the adults faced frosty and snowy conditions (4, 11) and resumed their activity at the end of hibernation and migrated towards the fields of wheat and barley for feeding and reproduction. The dormancy period in Iraq was different than in other areas (13).

**Table 10.** The hibernated Sunn Pest in the summit of mountains in north of Iraq during July 1999 and June 2000.

		Total Insects per 10 <sup>2</sup> meter in Mountain- Top		
		Date of	of Visit	
Mountain	Mountain High (m)	July 1999	June 2000	
Hassan Begg	2110	916	76	
Hindrin	2025	100	-	
Klinder	1823	1458	-	
Sherine	2378	720	146	
Bradost	2076	943	308	
Zozak	1863	25	-	
Kork	2125	1095	373	
Biran	2076	766	692	
Seffin	1975	295	117	

## Plants used for Oviposition by Sunn Pest Adults

The hibernated adults migrated from the mountain to the fields for egg laying and reproduction. The females laid eggs on the broad leaf plants such as *Astragalus* 

michauxianus Boiss., Bongardia chysogonum L., Cirsum rhizocephalus, Avena spp., Malva parviflora L., Saponaria vaccaria L., Lolium *rigidum* Gaudin, *Gundelia tournefortii* L., *Agropyron repens* (L.) Beauv. or on the undersides of wheat leaves.

## الملخص

أمين، عبد الباسط م.، محمد ع.ج. العزي وحسن س. الأسدي. 2007. كثافة مجتمع حشرة السونة خلال البيات الشتوى والصيفى في شمال العراق. الصفحات 79–88.

عند دراسة الظهور الفصلي لحشرة السونة في شمال العراق، أظهرت النتائج أن الحشرات المشتية أستأنفت نشاطها في النصف الثاني من آذار/مارس خلال الأعوام 1998، 1999 و 2000. ظهرت البالغات المنبثقة حديثاً في النصف الثاني من أيار /مايو والأسبوع الأول من حزيران/يونيو. تراوحت كثافة مجتمع البالغات المشتية بين 0.58–11.38 بالغة/10 م<sup>2</sup> في السنوات الثلاث. تضع الإناث البيض خلال نيسان/أبريل، وقد تراوح عدد مجموعات البيض في الحقل من 1–39، 1–10 و 1-11 مجموعة/10 م<sup>2</sup> خلال الأعوام 1998، 1999 و 2000، على التوالي. تراوحت كثافة الحوريات والبالغات الحديثة خلال السنوات الثلاث من 0.08 إلى 119.26 و 1.11 إلى 99.41 حشرة/10 م<sup>2</sup>، على التوالي. تمت مراقبة حركة الحشرات ضمن المناطق المختلفة من الجبل من تشرين الثاني/نوفمبر، 1999 إلى آذار /مارس، 2000. انخفضت أعلى كثافة للمجتمع في قمة وجوانب الجبل بشكل تدريجي من 97.75 و 20.75 بالغة/10 م<sup>2</sup>في تشرين الثاني/نوفمبر إلى 15.33 و 13.33 بالغة/10 م<sup>2</sup> في آذار /مارس، على التوالي. ازدادت كثافة الحشرات في قاعدة الجبل من 11.75 بالغة/10 م<sup>2</sup> في تشرين الثاني/نوفمبر إلى 25.3 بالغة/10 م<sup>2</sup> في آذار/مارس. كانت كثافة مجتمع البالغات في الجبال: حسن بك، هندرين، كالاندر، شيرين، بارادوست، زوزيك، كورك، كيران وسفين 916، 100، 1458، 720، 943، 25، 1095، 766، و 295 بالغة/10 م2، على التوالي. اختلفت هذه الأرقام عن تلك التي سجلت عام 2000، في حسن بك، شيرين، بارادوست، كورك، بيران وسفين والتي كانت 76، 146، 308، 373، 692 و 117 بالغة/10 م<sup>2</sup>، على التوالي. تهاجر حشرة السونة خلال البيات الشتوي والصيفي إلى النباتات التالية: Astragalus russelii Banks & Sol. (Prunus argentea (Lam.) Rehd. Quercus aegilops L. Quercus infectoria Olivier Euphorbia peplus L. Thymbra vulgaris L. Teucrium chamaevrys L. subsp. sinuatum 'Platanus orientalis L. 'Crxpis parviflora 'Pistacia atlantica Desf. S. stenobotrys Boiss (Silene ampullata Boiss, (Nepeta sp. (Telephium oligospermum Boiss, (Celak.) Rech. و Onosma albo-roseum Fisch. et Mey.

كلمات مفتاحية: Eurygaster integriceps Puton، بيات شنوي، كثافة مجتمع، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Relationship between Weed Grasses and the Life Cycle of Aelia germari Kuster in the Setif High Plains (Northeast, Algeria)

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#### Abstract

Fenni, M. and M. Bounechada. 2007. Relationship between Weed Grasses and the Life Cycle of *Aelia germari* Kuster in the Setif High Plains (Northeast Algeria). Pages 89-92.

In the Setif High Plains winter cereals are grown each year on >80% of cultivated land. Wheat and barley are frequently cultivated under rain-fed conditions. In this area the pointed wheat shield bug (*Aelia germari* Kuster) has become one of the most important insect pests and causes serious damage to wheat. Adults overwinter in the mountains which are located in the north where the altitude varies between 1200 and 1600 m. The insect moves to cereal fields in the spring. During migration (second half of March - beginning of April), *A. germari* populations feed and develop on weed grasses mainly at the edges of fields. This period corresponds to pre-ovulation, which is an essential phase of their life cycle. Graminaceous weeds such as *Hordeum, Avena, Bromus, Dactylis,* and *Lolium* spp. are intermediate hosts. The early development of these weeds is supported by bad cultivation techniques (e.g. bad ploughing, late sowing). Weeding and in particular on the edges of the fields is an effective control of the pointed wheat shield bug. It remains complementary to chemical and cultural methods that are commonly used.

Keywords: Aelia germari, life cycle, plant hosts, Setif, weeds.

## Introduction

In the Sétif High Plains, a fallow - winter cereals rotation occurs yearly on >80% of cultivated land. Grain yield average is <700 kg/ ha<sup>-1</sup>, and production is not sufficient to meet local consumption. The low yield is partly due to the erratic and low rainfall and also to the use of unimproved production practices, that do not consider plant protection needs. Chemical weed control is not widely used in this region, and herbicides are applied only on <5% of cereals.

The pointed wheat shield bug, *Aelia* germari Kuster, (Heteroptera, Pentatomidae) has become one of the most important insect pests of wheat and barley in the Setif High Plains, especially during years when frosts are not abundant and spring rainfall is delayed. High temperatures and excess of moisture constitute a favorable climate for pointed wheat shield bug. Yield loss is more important in wheat than in barley. In Algeria, few studies have been done on this insect (2, 6).

Several authors report that pointed wheat shield bug adults and nymphs feed and develop on wild plants of graminaceous species which grow near wheat and barley fields (6, 7, 11).

The main purpose of the present work was to inventory graminaceous weeds used by pointed wheat shield bug as intermediate plant hosts.

## Material and Methods

The Setif High Plains climate is characterised by a cold and wet winter and a hot summer that is generally very dry. Two bioclimatic areas are found there: the semi-arid area in the south and the sub-humid in the north. The main environmental characteristics of these two geographical regions are summarized in Table 1.

During the growing season 1999-2000, 57 surveys were done using a stratified sampling method based on agro-ecological factors. In each homogeneous area three wheat fields were chosen for sampling. The average

of the width and the length of these fields varied respectively between 100 m - 120 m and 200 - 250 m. The level of infestation for grass weed species and pointed wheat shield bug was scored on the edges and in the medium (middle) of each field. The level of infestation was estimated by using the abundance - dominance scale of Braun-Blanquet (5). According to the percent of cover the following indices were used: + (Simply present, their cover and abundance very low), 1 (Abundance low and cover < 5%), 2 (cover between 5 and 25%), 3 (cover 25 to 50%), 4 (cover 50 to 75%) and 5 (cover >75%). Weed species were identified following the « Nouvelle flore d'Algérie » (9).

## **Results and Discussion**

Fifteen species belonging to eight genera were identified (Table 2). The most abundant species on the edges of fields were *Hordeum murinum* L., *Bromus rubens* L. and *Cynodon dactylon* (L.) Pers. In the middle of the fields the most abundant were *Avena sterilis* L., *Bromus rigidus* Roth., *B. rubens* L. and *Phalaris brachystachys* Link. *A. sterilis* is common throughout the Mediterranean region. The increasing cereal monocultivation and late machine harvesting are factors favouring *Avena* spread (4).

Table 1. Main environmental characteristics of the Setif high plains.

	Mean annual rainfall (mm)	Average of min. of coldest month	Average of max. of hottest month	Climatic area	Altitude (m)	Soil characteristics	Main cereals
Zone Northern	400–700	2°C January	34°C August	Sub-humid	1000-1200	Clay soils, marly substrate, pH 7.2 – 7.5	Durum wheat, bread wheat
Zone southern	300–400	0°C December	36°C July	Semi-arid	900–1000	Calcareous soils, pH 7.8 – 8.2	Durum wheat, barley

 Table 2. Average infestation of graminaceous weeds and of pointed wheat shield bug on the edges and in the medium of field.

	Graminaceous weeds		Pointed who	eat shield bug
	Edges	Medium	Edges	Medium
Aegilops triuncialis L.	+	+	+	+
Avena sterilis L.	1	3	2	1
Bromus madritensis L.	+	1	+	+
Bromus rigidus Roth.	1	3	1	1
Bromus rubens L.	2	2	2	1
Bromus sterilis L.	+	1	+	+
Cynodon dactylon (L.) Pers.	2	+	+	+
Dactylis glomerata L.	1	+	2	1
Hordeum murinum L.	3	1	3	2
Lolium multiflorum Lamk.	+	+	+	+
Lolium perenne L.	1	+	1	+
Lolium rigidum Gaudin	1	+	1	+
Phalaris brachystachys Link.	1	2	+	+
Phalaris minor Retz.	+	1	+	+
Phalaris paradoxa L.	+	1	+	+

Compared to wheat, graminaceous weeds have an earlier cycle of development, especially on the edges of the fields where the seeds are ready to germinate as soon as conditions are favorable. Just after the first rains of autumn, seeds inflate and germinate, these two processes take place during December. The first leaf appears for the majority of species at the end of December and beginning of January. They reach their full vegetative development in the beginning of March.

The pointed wheat shield bug hibernates in the mountains (altitude varies between 1200 and 1600 m) located to the north of the Setif High Plains. When temperatures rise in spring they move to cereal fields to feed and mate (3, 8). Migration of overwintered adults from mountains to fields begins during the first week of March. During migration (second half of March - beginning of April), *A. germari* populations feed and develop on weed grasses mainly at the edges of fields. This period corresponds to the pre-ovulation and, is an essential phase of their life cycle. Highest infestation levels of *A. germari* were noted on *H. murinum*, *A. sterilis*, *B. rubens*, *D. glomerata* and *Lolium* spp. For the same weed species densities of pointed wheat shield bugs were higher at the edges than in the middle of the fields (Table 2). All pointed wheat shield bug species and Sunn Pests prefer feeding on wild graminaceous weeds that frequently grow near cultivated wheat and barley (6, 7, 11).

Weeding especially at field edges is an effective control means against pointed wheat shield bug. It remains complementary to chemical and cultural methods. In the Setif High Plains several insecticides are used against *A. germari* (4, 7). The suitable application of cultural practices such as dates of sowing and harvesting, crop rotation, early varieties and resistant varieties (1, 10) optimizes the use of insecticides, avoids their secondary effects such as soil and water pollution and limits damage caused by pointed wheat shield bug.

## الملخص

## فني، محمد ومصطفى بونشادة. 2007. العلاقة بين النباتات العشبية ودورة حياة Aelia germari Kuster في سهول سطيف المرتفعة (شمال شرق، الجزائر). الصفحات 89–92.

تزرع الحبوب الشتوية في سهول سطيف المرتفعة كل عام على حوالي 80% من الأراضي المزروعة. يزرع القمح والشعير بشكل متكرر تحت الظروف المطرية. أصبحت حشرة Aelia germari Kuster إحدى أهم الآفات الحشرية وتسبب ضرراً خطيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في الجبال الواقعة في الشمال حيث ينتوع الارتفاع بين 1200 من مرراً خطيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في الجبال الواقعة في الشمال حيث ينتوع الارتفاع بين 1200 ومرراً خطيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في الجبال الواقعة في الشمال حيث ينتوع الارتفاع بين 1200 من مرراً خطيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في الجبال الواقعة في الشمال حيث ينتوع الارتفاع بين 1200 ومرراً خطيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في الجبال الواقعة في الشمال حيث ينتوع الارتفاع بين 1200 ومراراً حفيراً للقمح في هذه المنطقة. تشتي الحشرات البالغة في البيل الهجرة (النصف الثاني من آذار/مارس – بداية نيسان/أبريل)، نتخذى مجتمعات حشرات المعروب في الربيع. خلال الهجرة (النصف الثاني من آذار/مارس – بداية نيسان/أبريل)، تتغذى مجتمعات حشرات المع ومنه لي وليبع. خلال الهجرة (النصف الثاني من آذار/مارس – بداية نيسان/أبريل)، تتغذى مجتمعات حشرات المع وول على النباتات العشبية وبشكل رئيس على حواف الحقول. تتطابق هذه الفترة مع ما قبل وضع البيض، والتي هي مرحلة أساسية في دورة حياتها. تعد الأعشاب النجيلية مثل Bromus، Avena، Avena، Avena، المعابية في دورة حياتها. تعد الأعشاب النجيلية مثل Dactor، والتي هي مرحلة أساسية في دورة حياتها. تعد الأعشاب النجيلية، الزراعة المرازعة المبكر، ولما المبكر، والتي هي مرحلة أساسية في دورة حياتها. تعد الأعشاب النجيلية، الزراعة المرازم المبكر، ولما المبكر المبكر، والم يعوم لي مراحم على حواف الحقول هي مكافحة فعالة لحشرات المرازعة المبكر (مثل الحارثة السيئة، الزراعة المبكر، وليفن هذه المبكر لمو المبكر، والم يعولي والمبكر، والغ مل المرازة وخاصة على حواف الحقول هي مكافحة فعالة لحشرات A. germari مراح المبكرة، الزراعية المبكرة المبكرة المبكر المبكر المبكر المبكرة ممامل المبكر والمبكرة، والزاعية مل مد مول المبكرة، والزراعية المبكرة، والزراعة المبكر مما مل مراح المبكرة، والزلمن مراح ممام ملمبكرة لهذه اله ممام المبة مي مرم مراح ملمبكم المبلغر المبكر ا

كلمات مفتاحية: Aelia germari، دورة حياة، عوائل نباتية، سطيف، أعشاب.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon. Bruce L. Parker<sup>1</sup>, Masood Amir-Maafi<sup>2</sup>, Margaret Skinner<sup>1</sup> and Mustapha El Bouhssini<sup>3</sup>

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### Abstract

Parker, B.L., M. Amir-Maafi, M. Skinner and M. El Bouhssini. 2007. Sunn Pest Populations Under *Artemisia, Astragalus, Quercus* and *Centaurea gaubae* in Overwintering Sites. Pages 93-98.

Sunn Pest population levels were determined beneath *Astragalus* sp., *Artemisia* sp., *Quercus* sp., and *Centaurea gaubae* (Bornm.) Wagenitz, plant species that commonly grow in overwintering sites in Iran. When comparisons were made within an overwintering site, significantly more Sunn Pest adults were found overwintering under *Astragalus* sp. than *Artemisia* sp. Sunn Pest adults were also found overwintering in the litter around *Quercus* sp. and *C. gaubae* in Iran, although populations were generally low compared to the other bushes sampled. Implications for sampling and management of overwintering Sunn Pest adults are presented.

Keywords: Sunn Pest, *Eurygaster integriceps* Puton, population distribution, overwintering sites, *Artemisia, Astragalus, Quercus, Centaurea gaubae.* 

## Introduction

Sunn Pest, Eurygaster integriceps Puton, spend ~  $2\frac{1}{2}$  to 3 mo annually feeding in wheat and barley fields and then migrate to the foothills and low mountains, where they remain in aestivation and diapause in what is commonly referred to as the overwintering period (1). During this period they generally remain in a quiescent state beneath the litter around various plants where climatic conditions are favorable for survival during the intense heat of summer and extremes of cold in winter. In southern Afghanistan the overwintering period may also be spent in grasses along river banks (G.R. Faizy, Kabul Univ., pers. comm.); in Syria beneath pine trees (MEB); in Turkey under the litter of oak trees and thorny bushes (R. Canhilal, Plant Prot. Res. Inst., pers. comm.); and in Iran beneath various species of bushes (MAM). Extensive research has been conducted in recent years on Sunn Pest population dynamics in overwintering sites in Iran (2).

Habitats that appear to be suitable as Sunn Pest overwintering sites cover thousands of hectares, and research is underway in Esfahan and Varamin to determine if Sunn Pest distribution is spread out over these vast areas or are clumped into specific focal areas within the general overwintering site regions (3). To facilitate this work, studies have included population determinations under different plant species that occur in overwintering sites. The study reported herein compared adult Sunn Pest population levels under the bushes of Artemisia sp. (Asteraceae), Astragalus sp. (Fabaceae), *Centaurea gaubae* (Bornm.) Wagenitz (Asteraceae) and *Ouercus* sp. in overwintering sites in Iran (Figure 1).

## **Materials and Methods**

Intensive sampling of Sunn Pest was completed at one overwintering site in Varamin during the period of November 2002 and January 2003. At this site an arbitrary population center was selected and from this
point, at each of the four cardinal directions, the number of live Sunn Pest were counted under five bushes (40-60 cm diam), either Artemisia sp. or Astragalus sp., at 500-m intervals to a distance of 5 km from the center point. An abundance of both plant species occurred at this site, ensuring that the number of bushes was not a limiting factor in terms of distribution. To further validate the Sunn Pest distribution relative to plant type, overwintering areas on mountainsides in Esfahan, Kordestan, Kermanshah, Golestan, Ghazvin and Azarbaija-Gharbi were selected as sample sites based on historical records of insect distribution, and 60 bushes were randomly selected at each site for sampling. The total number of live Sunn Pest under the bushes of Artemisia sp. and Astragalus sp. were counted. Additional sampling was conducted in four separate overwintering sites at Kermanshah and Golestan, where Quercus sp. were located and adult Sunn Pest were counted beneath 60 randomly selected plants of *Quercus* sp. and *Artemisia* sp. at each site. This method of sampling was repeated in three sites in Esfahan comparing Sunn Pest populations under *C. gaubae* and *Artemisia* sp. Comparisons of population density under *Quercus* sp. or *C. gaubae* with *Astragalus* sp. were not possible because these latter two species were not found growing together at the same sites in sufficient abundance to collect reliable data.

## **Results and Discussion**

When averaged over all cardinal directions, approximately 3.4 times more Sunn Pest adults per bush were found around *Astragalus* sp. (130.17) than *Artemisia* sp. (38.57) (Figure 2).



**Figure 1.** Plant species sampled to assess Sunn Pest distribution within overwintering sites in Iran: A) *Artemisia* sp., B) *Astragalus* sp., C) *Quercus* sp., D) *Centaurea gaubae* (Bornm.) Wagenitz.



Figure 2. Mean number of adult Sunn Pest overwintering under *Artemisia* sp. (A) and *Astragalus* sp. (B) by cardinal direction.

A consistent distribution pattern linked with cardinal direction was observed for both species of bushes. To the north, the greatest number of Sunn Pest were found at around 3 and 2.5 km from the center point for *Astragalus* sp. and *Artemisia* sp., respectively. To the east, Sunn Pest populations were greatest at around 2-3 km for both plant species. In general where populations were greater under *Astragalus*, more were also found under *Artemisia*, though the differences were not consistently of the same magnitude. For example, for both plant species, between 4-5 times fewer Sunn Pest were found to the south of the center point than to the north. In contrast, 3.9 times fewer Sunn Pest were found to the west than to the south under *Artemisia*, and only 2.1 times fewer Sunn Pest were found to the west under *Astragalus* than to the south. This suggests that other factors related to the site, such as aspect or elevation, affected Sunn Pest population distribution as well as the type of vegetation present.

Based on results from sampling for Sunn Pest under Astragalus and Artemisia in multiple overwintering sites around Iran (Esfahan, Kordestan, Kermanshah, Golestan, Ghazvin and Azarbaija-Gharbi), no significant difference between the two plant species was detected when data from all sites were combined. When data within a site were considered separately, significantly more Sunn Pest were found under Artemisia than Astragalus in only Site 2 (Figure 3). In three sites there was no difference in population levels between the two plant species, and two sites had more under Astragalus than Artemisia. The specific factors that a Sunn Pest uses to select a location for overwintering is unknown, though these results suggest that if both Astragalus sp. and Artemisia sp. are present, they may sometimes preferentially select the former. Sunn Pest populations at these additional sample sites were significantly lower than at Varamin, where a definite preference for *Astragalus* was detected. This suggests that a density-dependent relationship may exist relevant to the selection by Sunn Pest of an overwintering location.

Morphological differences in these two plant types may partially explain a potential preference by Sunn Pest. Artemisia sp. typically has an open form that allows sunlight and air to penetrate into the bush. This would result in higher temperatures in the warmer months, lower temperatures in the winter, and wider ranges in high and low temperatures throughout the overwintering period. Humidity levels under the bush would also be affected by the shape of the bush. These extreme conditions could have a negative impact on survival. Unlike Artemisia, Astragalus sp. typically is a bush with a dense canopy which limits sunlight penetration and air movement to the soil surface. This results in a cooler and more humid habitat beneath the bush where Sunn Pest commonly overwinter. Further research is needed to determine if the differences in Sunn Pest population levels reflect differential mortality rates due to habitat, or an actual selection preference made by the insect.



Figure 3. Mean number of live Sunn Pest per bush at six overwintering sites in Iran.

Sunn Pest populations in overwintering sites in which both Quercus sp. and Artemisia sp. grew (Kermanshah and Golestan) were <5 insects per bush, and no differences in distribution between these two plant species was observed when pest populations were this low. In the one site in which C. gaubae occurred with Artemisia, significantly more Sunn Pest were found associated with the former (115 per bush) than the latter (60 per bush). Results from this study indicate that Pest population density Sunn within overwintering sites may vary significantly depending on the plant species sampled. When populations are high and an abundance of both Astragalus and Artemisia sp. bushes are present in an overwintering site, more Sunn Pest may be found under the former bush. When Sunn Pest populations are low however, either type of bush can be sampled to determine population size. If only Quercus

sp. were present in the overwintering site, the standard sampling procedure of Parker et al. (3) has been shown to be useful and produce information with a precision of ~20-25%. The accuracy of the sampling procedure for counts of live adult Sunn Pest beneath *C. gaubae* has not been verified.

## Acknowledgments

The authors appreciate the technical assistance of the following personnel: Esfahan: A. Haghshenas, Tehran: A. Hassani, M. Safari, The University of Vermont: S.D. Costa, and ICARDA: Safaa Kumari, W. Reid. This research was funded in part by US Agency of International Development (USAID) Linkage funds and Department For International Development (DFID).

# الملخص

باركر، بروس ل.، مسعود أ. مافي، مارغريت سكنر ومصطفى البوحسيني. 2007. مجتمعات حشرة السونة تحت الأنواع النباتية Quercus ، Astragalus ، Artemisia و Centaurea gaubae في مواقع البيات الشتوي. الصفحات 93–98.

تم تحديد مستويات مجتمع حشرة السونة تحت الأنواع النباتية sp. ، Astragalus sp. ، Astragalus sp. ، astragalus sp. ، والتي تتمو عادة في مواقع البيات الشتوي في إيران. عندما أجريت مقارنات و Centaurea gaubae (Bornm.) Wagenitz، والتي تتمو عادة في مواقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي في إيران. عندما أجريت مقارنات محمن موقع البيات الشتوي، وجد عدد أكبر وبشكل معنوي من بالغات حشرة السونة المشتية تحت النوع النباتية . Astragalus sp. وبشكل معنوي من بالغات مشرة أسونة السونة المنتية حول الأنواع النباتية . ويحمن عاد معنوي من بالغات مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عدل أكبر وبشكل معنوي من بالغات مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عدل أكبر وجد عالغات السونة أيضاً مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عالغات السونة أيضاً مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عالغات السونة أيضاً مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عالغات السونة أيضاً مشتية في البقايا النباتية حول الأنواع النباتية . *Quercus* sp. وجد عالم منتية في البقارات مع الشجيرات الأخرى المفحوصة. سوف يعرض و معامين من أجل أخذ عينات وإدارة البالغات المشتية لحشرة السونة.

كلمات مفتاحية: حشرة السونة، Eurygaster integriceps، توزع مجتمع، مواقع بيات شتوي، Astragalus ، Artemisia، Centaurea gaubae ، Quercus.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Life History of Eurygaster testudinaria Geoffroy under Laboratory Conditions

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## Abstract

Mohaghegh, J. 2007. Life History of *Eurygaster testudinaria* Geoffroy under Laboratory Conditions. Pages 99-101.

*Eurygaster testudinaria* Geoffroy is one of three species of *Eurygaster* in Iran. Under laboratory conditions ( $26-27^{\circ}C$ ; 50-60% RH; L:D = 16:8) developmental periods of egg and instars 1-5 were (mean±SE):  $5.53\pm0.04$ ,  $3.21\pm0.06$ ,  $8.0\pm0.10$ ,  $5.22\pm0.07$ ,  $6.74\pm0.07$  and  $10.90\pm0.17$  days, respectively. There was a significant difference between female ( $41.63\pm0.63$  days) and male ( $38.85\pm0.54$  days) total developmental times. Females laid an average  $2.9\pm0.05$  rows of eggs in batches of  $12.1\pm0.45$ . Maximum mortality was 44% and occurred at 2nd instar. Overall survival of the immature stage was 32.2%. The life history data of *E. testudinaria*, developmental time, survival, Sunn Pest.

Introduction

*Eurygaster testudinaria* Geoffroy (Hemiptera: Scutelleridae) has sporadically been reported as one of the important bugs in cereal crops, especially in the literature of the 1960's (4, 5). It is one of three species of *Eurygaster* Laporte found in Iran, occurring in small numbers in natural and agricultural ecosystems (3, 6). However, relative population density of E. testudinaria has been considerably lower than its coexistent species E. maura L. in the studied areas (3, 6). Some information on taxonomic and morphological aspects of E. testudinaria have been given in detail (e.g., 2, 3, 6, 8, 9), but the data on its bionomics and life history has hardly been found, probably due to the lack of abundance. The aim of this research was to study the life history of E. testudinaria under laboratory conditions.

#### **Materials and Methods**

Overwintered adults of *E. testudinaria* (9 females + 1 male) were collected from a wheat field in Chamestan (Noor, Mazandaran, north of Iran) in late May. These bugs were all reproductively active, and cohabited with

relatively abundant numbers of their closest species E. maura. After introduction to the laboratory, they were placed collectively in a 2 1 Plexiglas container provided with wheat grains (variety Ghods) as a food supply and a wet cotton plug inserted in a water-tube as a moisture source. Every day, food and water were replenished and egg batches were collected. Egg batches <24 h old were collected and were kept separately in 0.5 1 containers each with wheat grain, as a substrate for first instars, and wet cotton fitted in a plastic dish. From second instar fresh wet wheat grains were added on a daily basis. Egg hatch, molting and number of dead nymphs were recorded daily until adult eclosion. The experiment was conducted in a climate chamber at 26-27°C, 50-60% RH and 16:8 (L:D).

## **Results and Discussion**

Three hundred and seventy five eggs in 31 batches were obtained within 3 weeks of their introduction to the laboratory, when 7 adult females were still alive. Females laid an average  $2.9\pm0.05$  rows of eggs in batches of  $12.1\pm0.45$ , both were higher than those of *E. maura* with 2.30 and 10.43, respectively (7).

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Egg, nymphal and the overall developmental times are presented in Table 1. There was a significant difference between female (41.63±0.63 days) and male (38.85±0.54 days) total developmental times (*t*-test, t = -3.355; df = 85; P = 0.001). This difference has not been proved for E. integriceps (1) and E. maura (7). The first instars with 3.2 days had relatively less time to develop. The 5th instars had the longest developmental time (average of 10.4 days). About 44% of the total mortality was at second instar. The fourth instars had relatively low mortality (3.3%) (Table 1).

**Table 1.** Mean  $\pm$  SE developmental times (days)and percent mortalities of different immature stagesof *Eurygaster testudinaria*.

Stage	Percent mortality	
Egg	$5.53 \pm 0.04$ (277)	9.2
1 <sup>st</sup> instar	3.21 ± 0.06 (266)	3.6
2 <sup>nd</sup> instar	$8.0 \pm 0.10$ (132)	43.9
3 <sup>rd</sup> instar	$5.22 \pm 0.07 (125)$	2.3
4 <sup>th</sup> instar	$6.74 \pm 0.07 (115)$	3.3
5 <sup>th</sup> instar	10.36 ± 0.17 (95)	6.5
Egg-to-adult	41.63 ± 0.63 (40)	
(female)		
Egg-to-adult	38.85 ± 0.54 (47)	
(male)		

The relatively long developmental time and highest mortality of second instars may partly result from their gradual adaptation to the food supply (fresh wheat grain). Likewise, Bullmann and Faber (4) reported a relatively long developmental time for second instars of *E. maura* and *E. austriaca* Schrank (Figure 1).

Overall 68.8% mortality of Ε. testudinaria is more than that reported for E. integriceps, 40% (1) and 45% (6), but like its closest species of E. maura, 65% (7), under similar experimental conditions. Overall mortality of E. testudinaria may, however, be lower under field conditions. Bullmann and Faber (3) reported higher survival of E. maura in the field. Further study on the life history of testudinaria is needed under field Е. conditions.



Figure 1. Percentage survival of different immature stages of *Eurygaster testudinaria* in the laboratory.

# الملخص

محقق، جعفر. 2007. تاريخ حياة Eurygaster testudinaria Geoffroy تحت ظروف المختبر. الصفحات 101-99.

تعد حشرة Eurygaster testudinaria Geoffroy أحدى الأنواع الثلاثة من الجنس Eurygaster في إيران. عند ظروف المختبر (26–27 °س؛ 50–60% رطوبة نسبية؛ 8:16 ضوء:ظلام)، بلغت فترات تطور البيضة والأعمار الحورية الخمسة (1-2): 5.23±0.04 نافذ 2.24±0.00، 5.22±0.00، 5.22±0.00 و 10.90±0.17 (المتوسط ± الخطأ القياسي) يوم، على التوالي. ظهر فرق معنوي بين فترات التطور الكلية للأنثى (6.14±6.00 يوم) وللذكر (3.85±2.04 يوم). وضعت الأنثى بالمتوسط 2.9±0.00 صف من البيض في مجموعات من 1.21±25%. بلغت نسبة الموت العظمى 44% وحدثت عند العمر الحوري الثاني، وبلغت نسبة البقاء الكلية للأطوار غير الكاملة 22.2%. قورنت بيانات تاريخ حياة E. testudinaria مع تلك

كلمات مفتاحية: Eurygaster testudinaria، زمن التطور، بقاء، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon. Jafar Mohaghegh

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#### Abstract

# Mohaghegh, J. 2007. Life History and Reproductive Table of *Eurygaster maura* L. (Hemiptera: Scutelleridae) in the Laboratory. Pages 103-108.

*Eurygaster maura* L. is a prevailing species of the genus *Eurygaster* in the wheat fields of the northeast of Iran (Golestan province). An experiment was carried out to determine its life history and demographic parameters under laboratory conditions (T= 26-27°C; RH= 50-60%; L:D= 16:8 h) using individuals collected from overwintering sites. Developmental periods of immature stages (egg and five nymphal instars) were:  $5.94\pm0.01$ ,  $3.51\pm0.02$ ,  $5.71\pm0.03$ ,  $5.26\pm0.03$ ,  $5.75\pm0.02$  and  $9.13\pm0.04$  days, respectively. There was no difference between female ( $35.54\pm0.18$  days) and male ( $35.12\pm0.17$  days) total developmental times. Adult females lived significantly longer than males ( $71.5\pm6.79$  vrs.  $37.33\pm7.07$  days). Preoviposition period was 11.67 days. Demographic parameters of *GRR*, *R*, *r<sub>m</sub>*, *T* and *Dt* were 90.44, 30.14, 0.0574, 1.0591, 59.33 and 12.07, respectively.

Keywords: Demography, development, Eurygaster maura, Iran, reproduction.

## Introduction

The Mauritian bug, Eurygaster maura L., is one of the three most important noxious bugs of the genus Eurygaster Laporte (7, 13, 18). Its damage to wheat crop has been reported from Russia (3), Turkey (8), Greece (19), Bulgaria (13) and Italy (17). Similar to its closely related species, Eurygaster integriceps Puton, it causes hydrolysis of gluten proteins of wheat grains via proteolytic enzymes resulting to poor dough properties (15, 20). However, relative population density of E. maura has been considerably less than two other coexistent species E. integriceps and Eurygaster austriaca Schrank in the studied areas (4, 11, 12). E. maura is a prevailing species of the genus Eurygaster in the wheat fields of the northeast of Iran, Golestan province (9, 10, 14). Although some studies have been done on development and reproduction of E. maura (4, 9, 16), less is known on its life history and reproductive characteristics.

Insect demography has been defined as study of insect populations and the processes that shape them (6). Thus it is a tool for quantifying biological events in an insect cohort giving a series of statistics concerning reproductive and survival potential of the subjected species. In this investigation information on developmental and reproductive properties of *E. maura* have been documented from demographic point of view.

#### Materials and Methods

Overwintring adults were collected at 980 m.a.s.l. height from mountainous forest of Zard-Chakal located 50 Km east of Gorgan city (northeast of Iran) in late December. Twelve pairs of them were separately confined in a plastic container of 0.5 l. Each container was provided with wheat grains (variety Ghods) as food supply and a piece of wet cotton inserted in a water-tube as moisture source. Every day, adult mortality and number of eggs were checked and food and water were replenished. Replicates produced no eggs, due to parasitism by tachinid flies or other physiological cues, were not included to demographic calculations. Egg batches were kept separately in a 0.5 l container with water, wet cotton fitted in a plastic dish, and from second instar on fresh wet wheat grains were added. Egg hatch, molting and number of dead nymphs were recorded daily till adult eclosion.

Carey's method (5) was used to calculate demographic parameters. Since the bug has obligatory reproductive diapause and consequently produced only one generation per year, specific age stage was started from overwintering adults, but as an assumption the obtained preimaginal developmental period and mortality were then considered in calculating reproductive table. The experiment was conducted under climatic conditions of 50-60% 26-27°C temperature, relative humidity and a photoperiod of 16:8 (L:D) h.

#### **Results and Discussion**

Egg, different nymphal and the overall developmental time are presented in Table 1. There was no difference between male and female total developmental times (t-test, t =1.6615; df = 360; P = 0.0975). Under the studied conditions total developmental time was 35 days. The first instar with 3.5 days has relatively less time to develop. The longest developmental time belongs to the 5th instars with average of 9.1 days. The rest of nymphal instars each took about 5 days to develop to the next stage. About one third of the total mortality related to the eggs in this study is in accordance with that of Bullmann and Faber (4) who reported 30.6% egg mortality. The late instars demonstrated less mortality. In a previous study with the same strain (9), the respective immature developmental times were 6.8, 4, 8.7, 7.4, 7.3 and 10.7 days (total of 42.5 days) where temperature  $(25\pm1^{\circ}C)$  and wheat variety (var. Karaj-1) were different. The longest developmental times in that experiment may be mainly related to the lower temperature used. In more fluctuating temperature (17-22°C) durations of the five instars were reported 6.1, 10.2, 8.1, 7.9, and 10.2 days, respectively (4). However, general pattern of immature development is similar in all above-mentioned studies. Comparing with its congeneric bug of E. integriceps (e.g., 2, 9) similar pattern of development has been determined in the present study.

**Table 1.** Mean  $\pm$  SE developmental times (days) and percent mortalities of different immature stages of *Eurygaster maura* L.

Stage	Developmental time (n)	Percent mortality
Egg	5.94±0.01 (740)	29
Nymph		
1 <sup>st</sup> instar	3.51±0.02 (656)	8
2 <sup>nd</sup> instar	5.71±0.03 (550)	10
3 <sup>rd</sup> instar	5.26±0.03 (473)	8
4 <sup>th</sup> instar	5.75±0.02 (428)	4
5 <sup>th</sup> instar	9.13±0.04 (362)	6
Egg-to-adult	35.54±0.18 (181)	
(female)		
Egg-to-adult	35.12±0.17 (181)	
(male)		

Reproductive characteristics of *E. maura* are presented in Table 2. Females lived significantly longer than males (71.5±6.79 vrs.  $37.33\pm7.07$  days; *t*-test: *t* = 3.4842, df = 10, *P* = 0.0059). After 54 days of introduction to the laboratory 67% of the original females were alive whereas the male survival was 17% (Figure 1). At that time 93.6% overall eggs had been produced. Males and females mated several times during their life spans (i.e., males are polygynous, females polyandrous), and preovoposition period was 11.67±0.71 days on introduction to the laboratory.

The gross and net fecundity rates were 180.88 and 176.5 eggs per female, respectively. This result is much higher than those reported earlier by Bullmann and Faber (4) with an average of 55 eggs per female and Stavraki (16) with about 90 eggs per female. In an earlier experiments (9), female of the same strain collected from the wheat field near to the forest in early spring, demonstrated a better fecundity of 249.6 eggs per female. This may be, in part, addressed to the relatively better fitness of the bugs emigrated from overwintering sites to the fields, compared with those collected from overwintering



Figure 1. Survival curves of overwintering Eurygaster maura L. females and males in the laboratory.

sites 3-4 months before emigration. Likewise, individuals of *E. integriceps* collected from overwintering sites laid in average 144 eggs per female (1), whereas Mohaghegh (9) reported 300 eggs per female from field collected ones, using similar experimental conditions.

 Table 2. Reproductive parameters of Eurygaster maura L.

Parameter	Value				
Life time reproductive rate (eggs)					
Gross fecundity rate (eggs)	180.88				
Gross fertility rate (eggs)	124.99				
Gross hatch rate	0.69				
Net fecundity rate (eggs)	176.50				
Net fertility rate (eggs)	123.50				
Mean age of reproduction (days)					
Mean age gross fecundity	63.25				
Mean age gross fertility	60.62				
Mean age net fecundity	62.45				
Mean age net fertility	60.24				
Mean age hatch	65.86				
Daily reproductive rate					
Mean egg per day	1.97				
Eggs/female/day	2.70				
Mean fertile eggs per day	1.36				
Fertile eggs/female/day	1.89				

Fecundity of a female that lived to her maximum life span was only 2% more than average fecundity of its cohort, and the respective fertilities were 96% and 70%. Figure 2 showed age specific survival and fecundity of a female. After oviposition period considerable portion of females was alive. Forty eight days upon introduction of original cohort to the laboratory when all female adults were still alive 91.1% of the gross fecundity was achieved. Net fertility was 32% less than gross fecundity. Of this only 2% was due to female mortality and 30% due to infertility. Number of eggs per batch, and egg rows per batch averaged  $10.43 \pm 0.37$  (*n* = 101) and  $2.30 \pm 0.05$  (*n* = 81), respectively.

Demographic parameters of *E. maura* are summarized in Table 3. Conversely to its congeneric species *E. integriceps* (2), infertility of eggs performed a major effect in reducing gross fecundity. If a female lived to her physiological maximum life it would have produced 90.4 newborn females, but because of mortality it produced 30.1 ones.

Parameter	Value	Unit
Gross reproductive rate (GRR)	90.44	female eggs/ female
Net reproductive rate $(R)$	30.14	female offspring/ female
Intrinsic rate of increase ( <i>r<sub>m</sub></i> )	0.057	female/day
Finite rate of increase ()	1.059	per day
Generation time $(T)$	59.33	Days
Doubling time ( <i>Dt</i> )	12.08	Days
Birth rate (b)	1.36	
Death rate (d)	1.30	

**Table 3.** Demographic parameters of a *E. maura*female.

Since Amir-Maafi and Parker (2) did not include the data from immature stage of E. *integriceps* in their analysis (personal communication), demographic comparison between the two studies would result biases. However, in case of excluding the immature period of E. maura from calculations somewhat better performances were obtained for almost all studied parameters. Nevertheless, comparative demography between the two species would simultaneously be conducted under similar conditions in order to avoid misleading estimates. Furthermore, under realistic field conditions the Mauritian bugs may demonstrate different reproductive performances from what they did in the laboratory. Thus the results of this study should not be extrapolated to the natural field conditions.



Figure 2. Mean reproduction (M<sub>x</sub>) and survival (L<sub>x</sub>) of a female *Eurygaster maura* L.

#### الملخص

محقق، جعفر. 2007. تاريخ الحياة وجدول التكاثر عند حشرة .Eurygaster maura L تحت ظروف المختبر. الصفحات 103–108.

يعتبر النوع Lerygaster maura L. نوعاً سائداً من الجنس Eurygaster في حقول القمح في شمال شرق (محافظة غولستان). أجريت تجربة لتحديد تاريخ حياته والعوامل الديموغرافية تحت ظروف المختبر (درجة حرارة 26-2<sup>0</sup>) س؛ رطوية نسبية 50-60%؛ ضوء:ظلام 8:16 ساعة) باستخدام أفراد تم جمعهم من مواقع البيات الشنوي. بلغت فترات التطور للأطوار غير الكاملة (البيض والأعمار الحورية الخمسة): 9.45.00 ما 25.50، 50.25±00، 50.2±00، 50.2±00، 50.2±5.75 للأطوار غير الكاملة (البيض والأعمار الحورية الخمسة): 9.45.40، 15.2±00، 50.2±5.75 ما للأطوار غير الكاملة (البيض والأعمار الحورية الخمسة): 9.45.40، 15.2±00، 50.2±5.75 ما للأطوار غير الكاملة (البيض والأعمار الحورية الخمسة): 9.45.40، 15.2±00، 50.2±5.70 ما 20.2±5.75 و 50.2±5.70 أو 20.0±2±0.00 معلى التوالي. ولم يكن هناك فرق بين فترات التطور الكلية للإناث (54±35.40 يوماً) وللذكور (25±7±0.00 ما 20.2±5.75 معلى التوالي. ولم يكن هناك فرق بين فترات التطور الكلية للإناث (54±35.40 معارة معلى وللذكور (25±5.70 معلى معنوي (20.5±5.70 معارة معلى وللذكور معلى معنوي (20.5±5.70 معارة معارة معلى وللذكور معلى معنوي (20.5±5.70 معارة معلى معنوي (20.5±5.70 معارة مع معلى معنوي (20.5±5.70 معارة مع معارة مع معارة المول من الذكور بشكل معنوي (20.5±5.70 معارة مع معارة مع معارة مع معارة مع معارة معارة معارة معارة معارة معارة معارة معارة معارة مع معارة مع معارة مع

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# A New Technique for Monitoring Sunn Pest

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#### Abstract

#### Adldoost, H. 2007. A New Technique for Monitoring Sunn Pest. Pages 109-114.

The Sunn Pest, *Eurygaster integriceps* Puton is a significant pest of wheat in Iran. Chemical spraying is the common method for its control that is conducted on all fields where Sunn Pest densities exceed the economic threshold. Chemical treatments against overwintering adults should be made before the onset of oviposition. This will stop Sunn Pest damage while protecting oophagous parasitoids. Accurate monitoring is the basis for effective control and thereby increases the efficacy of parasitoids. Some techniques have already been tested on cereals other techniques should be tested. Most of Sunn Pest eggs are laid on weeds. Some weed plants provide good protection for eggs or early immature stages from direct sunlight. During 1999–2001 the oviposition of the insect was investigated in 30 fields of West Azarbaijan, Iran. Ten samples of wheat and weeds were collected from each field using a one-m<sup>2</sup> sampling frame. Samples were transported to the laboratory in plastic bags and Sunn Pest eggs on the plants were counted. The maximum number of eggs was 30 and the mean was 4.96 clusters per m<sup>2</sup>. About 62% of the total eggs found per m<sup>2</sup> were on the *Conringia orientalis* (L.) Dumort, a shiny, smooth and slightly succulent weed plant mainly associated with wheat. Its presence in an area is considered to be an indicator for the monitoring of Sunn Pest eggs. **Keywords:** *Eurygaster integriceps*, monitoring, Sunn Pest, weed plants in wheat.

#### Introduction

Some wheat pests are of such paramount importance that they bear special mention or have the potential to spread to other similar agro-climatic zones where wheat is grown. Many of these pests, or groups of pests, typically undergo annual outbreaks in many countries and cause substantial crop losses (15). Sunn Pest and pointed wheat shield bugs are widespread throughout the rainfed grainproducing regions of southern and eastern Europe, northern Africa, and southwestern and south-central Asia (9, 11, 12). The Sunn Pest, a complex of bugs, is responsible for the application of pesticides costing US\$ 40 million on ~ 15 million ha of wheat and barley annually (5, 8). Wheat is the most common host, although barley, rye, oats, sorghum, and maize may also be attacked. Wheat plants are injured when Sunn Pest extracts fluids from stems, leaves, or developing grains thereby reducing plant vigour. When feeding on kernels, the Sunn Pest ingests the milky endosperm, which may result in a 15 to 60%

reduction in kernel weight. Both nymphs and adults inject a proteolytic enzyme while feeding that aids in dissolving plant proteins. Leaf or stem tissues surrounding the feeding site die. The enzymes remain in a dehydrated, inactive state within the kernels after the insect ceases to feed. When water is added to flour milled from infected grain, these enzymes are reactivated and destroy the dough's gluten. Such dough lacks adequate gluten strength and cannot be used to make bread (1, 2, 18). The earliest record of Sunn Pest in Iran dates from about 1862 when a severe outbreak caused panic in Fars Province. There is little doubt that the harmful activity of Sunn Pest goes back even further into Iran's history, with outbreaks occurring whenever conditions were favourable. Currently it infests about 1 million ha of wheat and barley. The average yield losses are estimated at 20 to 30% if the insects are not controlled (12). Wheat production in the Sunn Pest-affected areas of the Near East heavily dependent on chemical was insecticides (7) and without exception, countries infested by Sunn Pest rely on chemical sprays for control (16). However Sunn Pest control by chemical insecticides is virtually the sole method currently used in Romania, Europe and Asia (10). The first chemical treatment in Iran was performed about 40 years ago while there has been a nearly tenfold increase in the infested area over the last 20 years (12). In general, an application of chemical insecticides on overwintering adults negatively affects parasitism, reducing it from 86.9% (unsprayed plots) to 8.3% (sprayed plots) in some cases (14). Beneficial insects must be protected at time of egg laying in the refuge areas. The cost of putting such actions into practice is low, is economically viable and dose not require insurance policies (3). Hymenopteran egg parasitoids are among the natural enemies that contribute to the reduction of Sunn Pest populations. They are active in the spring, about 2 weeks after Sunn Pest migrates to cereal fields from its overwintering sites. The level of parasitism varies from year to year and some years it reaches 100% at the end of the season. These natural enemies seem to be playing an important role in regulating Sunn Pest populations and thus should be conserved (4).

Biological control of Sunn Pest must include all aspects of Sunn Pest biology that can be manipulated to maintain populations at economically acceptable levels. While no aspect should be neglected, egg deposition is particularly important. Eggs must be controlled, both quantitatively and qualitatively (17). Eggs are laid on wheat and weed plants (6, 18). Radjabi (13) has listed 32 species of weed plants belonging to 15 families including Gramineae, Cruciferae, Caryophyllaceae and Papsveracea that E. integriceps lays eggs on. These are not hosts but act as a shelter for the eggs. Biological control begins with an understanding of pest biology and a choice of a biological agent that is adapted to the pest and its host plant. Control agents must be released at the correct time to correspond with the presence of the insect host in the field (17). Biological control of Sunn Pest has been practiced only in Iran, owing to the ability there to collect large numbers of adults from hibernation sites. These are kept in captivity to mass-produce eggs for rearing parasitoids (3). The object of this study was to determine plant species in wheat fields which could have more Sunn Pest eggs and are distinct from graminae. They could serve as an easy indicator for monitoring Sunn Pest eggs.

#### **Materials and Methods**

During 1999-2001, 30 wheat fields in the West Azarbaidjan Province were surveyed at different locations for the eggs of Sunn Pest (Eurygaster integriceps Puton). Ten samples were taken at random from each field using a one-m<sup>2</sup> sampling frame. All wheat and weed plants inside the sampling frame were cut and adult Sunn Pest collected and transported to the laboratory in plastic bags. We recorded the number of plants of each sample and identified the plant species. Leaves of the collected plants were investigated for Sunn Pest egg clusters. Frequency of egg clusters on each plant species and the numbers of eggs in each cluster were recorded. Eggs were kept in an incubator (at  $25\pm1^{\circ}$ C and  $70\pm5^{\circ}$  RH) for the appearance of egg parasitoids.

#### **Results and Discussion**

Twenty eight rainfed wheat fields were surveyed in Orumieh, Naghadeh, Mahabad and Bookan, each at an elevation of 1300-1400 m. The dominant variety of wheat was Sardary. A total of 64,887 plants belonging to 48 spp. were sampled: 50,246 wheat, and 14,631 weed plants. From twenty three fields that *Conringia orientalis* was more prevalent a total of 1489 egg clusters were recovered: 414 clusters on wheat, 918 clusters on *C. orientalis* and 142 clusters on other plants (Table 1). Average parasitism of eggs were 12.56% and maximum was 12.53%. From the 4 wheat fields that *Papavera* sp. was prevalent a total of 120 egg clusters were recovered: 32 clusters on wheat, 69 clusters on *Papavera* sp., and 19 on other plants (Table 2). Average egg parasitism was 3.24% and maximum was 6.89%. Also from three wheat fields that *Goldbachia laevigata* (M. Bieb.) DC. was prevalent a total of 50 clusters were recoverd with 16 on wheat, 26 on *G. lavigata* and 8 were on other plants (Table 3). No egg parasitism was found in these fields.

However, during three years of survey, from each sq m of wheat fields a maximum 30

Sunn Pest egg clusters were recovered and the average was 0.5 to 11 clusters depending on location. Although the parasitism rate of eggs was not high but where the orchards or other trees was grown it was a little higher. Also there was no significant parasitism difference between eggs laid on wheat or on other plants. *Trissolcus semistriatus* Nees (Hymenoptera: Selionidae) and *Ooencyrtus nigerrimus* Ferr. & Voeg. (Hymenoptera: Encyrtidae), were the egg parasitoids identified.

			Mean egg cluster /m <sup>2</sup>			
Location	Date	Sample	Total/m <sup>2</sup>	Wheat	C. orientalis	Other plants
Naghadeh-M	02/05/1999	10	3.3	2.30	0.80	0.2
Naghadeh-H	30/04/1999	10	3.3	2.10	0.80	0.4
Balestan-A	04/05/1999	10	10.2	6.50	3.30	0.4
Balestan-2	24/04/1999	10	2.5	0.20	2.20	0.1
Balestan-3	24/04/1999	10	4.9	0.10	4.60	0.2
Balestan-4	24/04/1999	10	4.2	0.60	3.20	0.4
Balestan-B	26/04/1999	25	3.8	0.24	3.36	0.2
Balestan-5	08/05/2000	10	9.9	4.00	3.00	2.2
Jolbar-Ts1	24/04/2000	10	5.8	1.30	3.80	0.7
Jolbar-R	24/04/2000	10	4.0	2.50	0.80	0.7
Jolbar-NV	25/04/2000	10	6.8	3.50	3.10	0.2
Jolbar-Pa	26/04/2000	10	9.0	4.50	3.90	0.6
Jolbar-Ve	28/04/2000	10	8.4	1.50	5.10	1.8
Jolbar-Ts2	14/03/2001	10	6.3	1.20	4.70	0.4
Jolbar-Ch	16/03/2001	10	7.8	1.10	5.40	1.2
Jolbar-A	18/03/2001	10	7.5	1.60	5.30	0.5
Jolbar-B	19/03/2001	10	7.8	0.80	5.80	1.2
Jolbar-C	20/03/2001	10	6.8	1.00	5.00	0.8
Jolbar-Pr	20/03/2001	10	11.0	2.50	8.30	0.2
Jolbar-D	20/03/2001	10	7.1	0.80	5.60	0.7
Jolbar-E	20/03/2001	10	8.6	2.30	5.70	0.6
Naghadeh-G	23/04/2001	10	1.8	0.20	1.50	0.1
Naghadeh-L	23/04/2001	10	1.8	0.20	1.50	0.1

Table 1. Sunn Pest egg density in wheat, Conringia orientalis and other weed plants.

Table 2. Sunn Pest eggs density in wheat, Papavera sp. and other weed plants.

			Mean egg cluster / m <sup>2</sup>			
Location	Date	Sample	Total/m <sup>2</sup>	Wheat	<i>Papavera</i> sp.	Other plants
Mahabad C	02/05/1999	10	5.7	1.5	3.9	0.3
Mahabad A	28/4/2001	10	1.4	0.3	0.9	0.2
Mahabad B	28/4/2001	10	1.9	0.5	0.9	0.5
Mahabad D	28/4/2001	10	3.0	0.9	1.2	0.9

Based on results Sunn Pests prefer to lay eggs on nongramineaus plants in dry lands (Table 4). These provide good protection for the eggs and first instar nymphs from hot sunlight in wheat fields. *Conringia orientalis* may be the best one for surveying because 62% of Sunn Pest eggs were laid on it. It is a shiny, smooth and slightly succulent plant associated with wheat but well distinct from it. Its presence in an area is considered to be an indicator for the monitoring of Sunn Pest eggs.

Table 3. Sunn Pest eggs density in wheat, Goldbachia laevigata and other weed plants.

			Mean egg cluster / m <sup>2</sup>			
Location	Date	Sample	Total/m <sup>2</sup>	Wheat	G. Lavigata	Other plants
NaghadehBM	02/05/1999	10	3.3	1.4	1.5	0.4
Bookan-1	28/04/2001	10	1.2	0.1	0.7	0.4
Bookan-2	28/04/2001	10	0.5	0.1	0.4	0.0

			Plants density/m <sup>2</sup>		
Location	Date	Sample	Wheat	C. orientalis	Other plants
Naghadeh - M	02/05/1999	10	513.8	6.0	32.50
Naghadeh - H	30/04/1999	10	540.2	3.5	14.10
Balestan – A	04/05/1999	10	514.0	10.3	19.50
Balestan - B	26/04/1999	25	527.0	17.3	28.76
Jolbar - T	24/04/2000	10	250.0	84.9	124.10
Jolbar - J	24/04/2000	10	293.7	16.1	53.80
Jolbar -R	25/04/2000	10	265.9	3.1	13.80
Jolbar - P	26/04/2000	10	238.6	6.0	60.60
Balesatan C	08/05/2000	10	194.9	15.5	54.30
Jolbar - Ts	14/03/2001	10	343.5	20.1	23.60
Jolbar - Ch	16/03/2001	10	170.5	8.4	22.10
Jolbar - A	18/03/2001	10	149.6	9.3	19.60
Jolbar - B	19/03/2001	10	141.5	6.8	11.10
Jolbar - C	20/03/2001	10	195.1	7.3	18.90
Jolbar - Pr	20/03/2001	10	161.6	7.8	16.50
Jolbar - D	20/03/2001	10	186.5	8.1	36.10
Jolbar - E	20/03/2001	10	215.7	8.3	33.40
Naghadeh - G	23/04/2001	10	138.8	5.2	26.30
Naghadeh - L	23/04/2001	10	138.8	5.2	26.30

Table 4. Density of *Conringia orientalis* and other weed plants/m<sup>2</sup> in wheat fields.

#### الملخص

# عدل دوست، حيدر. 2007. تقنية جديدة لرصد حشرة السونة. الصفحات 109-114.

تعد حشرة السونة (Eurygaster integriceps Puton)، من الآفات المهمة على القمح في إيران. يعتبر الرش الكيميائي الطريقة الشائعة لمكافحتها، والذي يتم في جميع الحقول التي تتجاوز فيها كثافة حشرة السونة العتبة الاقتصادية. يجب أن تجرى المكافحة الكيميائية للبالغات المشتية قبل بداية وضع البيض، وذلك لتقليل ضرر حشرة السونة وحماية طفيليات البيض. يعد الرصد الدقيق العامل الأساس لمكافحة فعالة التي تزيد من فعالية الطفيليات. اختبرت بعض التقنيات مسبقاً على محاصيل الحبوب وهناك تقنيات أخرى يجب أن تختبر. معظم بيوض حشرة السونة توضع على الأعشاب الضارة، التي تؤمن حماية جيدة للبيض والأطوار غير الكاملة المبكرة من أشعة الشمس المباشرة. تم خلال الفترة 1999–2001 تقصي وضع البيض للحشرات في 30 حقلاً في غرب أذربيجان، إيران. تم جمع عشر عينات من القمح والأعشاب من كل حقل باستخدام إطار لأخذ العينات مساحته 1 م<sup>2</sup>. نقلت العينات إلى المختبر في أكياس بلاستيكية وتم عد بيض السونة الموجود على النباتات. كان أكبر عدد للبيض 30 والمتوسط 4.96 مجموعة بيض/م<sup>2</sup>. كان حوالي 62% من البيض الكلي/م<sup>2</sup> على النبات العشبي Conringia للبيض 30 والمتوسط 6.96 مجموعة بيض/م<sup>2</sup>. كان حوالي 62% من البيض الكلي/م<sup>2</sup> على النبات العشبي Lowror للرصد بيض حشرة السونة.

كلمات مفتاحية: Eurygaster integriceps، رصد، حشرة السونة، نباتات الأعشاب الضارة في القمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Binomial and Sequential Sampling of Adult Sunn Pest, Eurygaster integriceps Puton

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#### Abstract

# Amir-Maafi, M., B.L. Parker and M. El Bouhssini. 2007. Binomial and Sequential Sampling of Adult Sunn Pest, *Eurygaster integriceps* Puton. Pages 115-121.

During a two-year field survey of the Sunn Pest, *Eurygaster integriceps* Puton on winter wheat, data sets were generated which consisted of 100 estimates of mean density (*m* Sunn Pest per quadrate  $(=1m^2)$ ), variance (s<sup>2</sup>), and the proportion of empty quadrate (P<sub>0</sub>). Each estimate of Sunn Pest populations was based on counts per individual quadrate on each sampling occasion. Taylor's power law was used to model relationships between mean densities (per m<sup>2</sup>(=quadrate)) and variances for Sunn Pest on winter wheat in Varamin, Iran. Model slope was >1.0 for Sunn Pest, which indicated that adults showed aggregated spatial patterns. Sequential sampling plans, based on numerical (complete) and binomial (presence or absence) counts, for sunn pest were developed using Taylor's parameters and that estimated from the linear regression of ln(*m*) on ln[-ln(P<sub>0</sub>)]. Suggestions are made concerning the use of the sampling plans and the levels of precision that may be attained.

Keywords: Sunn Pest, sampling, Eurygaster integriceps, wheat pests.

#### Introduction

Sunn Pest, *Eurygaster integriceps* Puton (Hemiptera: Scutelleridae) causes considerable damage in wheat and barley in West and Central Asia. In these regions, millions of hectares of wheat must be sprayed with pesticides each year to control this insect (6). While chemical control is recommended to reduce economic losses, there is inadequate information available for accurate and efficient monitoring of Sunn Pest populations which is necessary for making control decisions.

Sampling populations of Sunn Pest is usually difficult and time consuming if estimates are based on numerical counts (i.e., completely counting all individual pests in each sample unit) (1).

For successful IPM, strategy there must be a sampling protocol to rapidly assess Sunn Pest infestations so that appropriate

management tactics can be chosen. One such method is a binomial (i.e., presence-absence) sampling technique. Binomial sampling is based on parameters which describe a relationship between mean density (m) and infestion level (denoted herein by the proportion of empty sample units,  $P_0$ ). An empirical model has been used to describe the *m*-P<sub>T</sub> relationship based on parameters from the linear regression of  $\ln(m)$  on  $\ln[-\ln(P_T)]$  (2. 8, 12, 13, 15). Other binomial models have described the relationship between m and 1-  $P_T$ (i.e., the proportion of sample units infested with more than T pests) using parameters from Taylor's power law (20) analysis or the dispersion parameter k from the negative distribution (15, binomial 22). When parameters from these models are used to determine binomial sample size, the variance associated with estimation of mean density [Var(m)] is computed in different ways by different authors, thus resulting in different sample sizes (7).

In this paper, data which include a range of population variation for Sunn Pest in winter wheat in Varamin, Iran, is presented to reveal its spatial pattern and empirical relationships between m and P<sub>0</sub>. Sequential sampling plans based on numerical (i.e., complete counting) or binomial (i.e., presence or absence) counts are also included with discussions of their precision and efficiency for Sunn Pest.

#### Materials and Methods

#### **Field Sampling of Sunn Pest Population**

Samples of Sunn Pest were taken from 5 fields of winter wheat (each field was approximately 2 ha in size), during the 2002 and 2003 growing seasons. In each field, using a standard  $1 \times 1$  m quadrate (sample unit) Sunn Pest adults were counted. Samples were taken twice a week from mid-April to mid-May, each year.

The number of quadrates (*n*, sample size) sampled from each of the fields on each sampling occasions was based on the mean number of Sunn Pest per quadrate (*m*) and its variance (s<sup>2</sup>), which was estimated from Sunn Pest counts of the first 25 quadrates sampled randomly in the field. The appropriate sample size was determined as  $n=s^2/(m^2E^2)$  where *E* represents sampling error level (18), and was set to 0.15 in this study. When the calculated *n* was larger than 25, sampling continued until the preset error level was attained. As a result, sample sizes ranged from 25 to 596 quadrates.

Population statistics, including m, s<sup>2</sup> and P<sub>0</sub> (proportion of empty quadrate) for Sunn Pest, were computed separately (on the basis of individual quadrates) for each sampling occasion.

Spatial patterns of Sunn Pest adults were described using parameters estimated from the analysis of Taylor power law ( $s^2=am^b$ ; 20) i.e.,  $ln(s^2)=ln(a)+b ln(m)$  (1)

The empirical relationship between mean Sunn Pest density, m, and the proportion of quadrates with no Sunn Pest, P<sub>0</sub>, was based on the parameters from the linear regression of ln(m) on  $ln[-ln[(P_0)]$  (2, 4, 8, 12, 13):

 $\ln(m) = + \ln[-\ln[(P_0)](2)]$ 

where and are intercept and slope parameters from the regression, respectively. In the form of the antilogarithm, the above equation becomes:

 $m = [-\ln[(P_0)] (3)]$ 

Consequently, given an estimated  $P_0$  from the field, the empirical m- $P_0$  relationship described by Eq.3 was used to obtain an estimate of m. The variance for estimation of m from  $P_0$  differs considerably among previous authors (2, 4, 8, 12, 13, 16), but was computed following Binns & Bostanian (4) and Nyrop & Binns (14):

 $\operatorname{var}(m) = \operatorname{var}_{p}(m) + \operatorname{var}_{s}(m)$  (4)

Where  $var_p(m)$  and  $var_s(m)$  are components of the variance from the prediction itself and the sampling error, respectively. These two components were estimated as:

 $var_{p}(m)=m^{2}\{mse/N+(\ln[-\ln(P_{0})]-avglnln P_{0})^{2}s^{2}+mse\} (5)$ 

 $var_s(m) = m^2 \{ \frac{2(1 - P_0)}{(n P_0[\ln(P_0)]^2)} \} (6)$ 

In equations 5-6, *n* is the number of quadrates (sample unites) desired to be taken from a population, N is the number of data points used to fit equation 2, *mse* is the residual mean squared error from the regression of  $\ln(m)$  on  $\ln[-\ln[(P_0)]$ ,  $avglnln P_0$  is the mean value of the independent variable  $\ln[-\ln[(P_0)]$  used in the regression, and s<sup>2</sup> is the variance of the regression slope (which equals the value of *mse* divided by the sum of squared deviations of the  $\ln[-\ln[(P_0)]$  from *avglnln* P<sub>0</sub>; s is the standard error of from the regression), respectively.

Sequential sampling plans were designed based on Karandinos' (11) sample size formula,

 $n = [Z_{/2}/d]^2 s^2/m^2$  (7)

where Z  $_{/2}$  is the standard normal deviate such that P(Z > Z  $_{/2}$ )= /2, *d* is the predetermined half width of confidence interval (CI) as a proportion of the mean (*d*=CI/2*m*), and s<sup>2</sup> is the variance of estimated *m*. Replacing s<sup>2</sup> in equation 7 with Taylor's variance (am<sup>b</sup>) estimated from equation 1 and *m* with  $T_n/n$  ( $T_n$ is the cumulative number of individual pests in *n* sample units) and rearrangement the terms,  $T_n$  can be solved (14) as :

 $\ln(T_n) = \{\ln[d^2/(\mathbb{Z}_{2a})]\}/(b-2) + [(b-1)/(b-2)]\ln(n) (8)$ 

which in the form of antilogarithm, generates a stop line for sequential sampling based on numerical (or complete) counts. Three stop lines for numerical sequential sampling of Sunn Pest were determined using the fixed d values of 0.2, 0.3 and 0.5 (which are equal to sampling error levels of 0.1, 0.15 and 0.25), respectively.

To develop binomial sequential sampling plans for monitoring for Sunn Pest populations, Binns and Bostanian (4) variance (Equation 4) was used to replace the  $s^2/n$  in equation 7. Then, rearranging equation 7 and incorporating the terms of equation 5 and 6 into it yielded:

 $dm = Z_{/2}[s^{2}/n]^{1/2} = Z_{/2}[var_{p}(m)+var_{s}(m)]^{1/2} (9)$ A solution to *d* was then obtained as follows:  $d = Z_{/2}\{ {}^{2}(1-P_{0})/(n P_{0}[\ln(P_{0})]^{2}) + [(\ln[-\ln(P_{0})] - avglnln P_{0})^{2}s^{2} + mse(N+1)/N] \}^{1/2} (10)$ 

Here, d is the value that can actually be reached when sampling is based on binomial counts given a desired sample size n. Note that m was canceled in equation 10 and that dvalue would change with the desired n and the proportion of empty quadrate P<sub>0</sub> related to population density. From equation 10, solving for n generated a general binomial sample size curve as:

 $n = \{ [ {}^{2}(1-P_{0})]/P_{0}[\ln(P_{0})]^{2} \} / \{ ( d/Z_{/2})^{2} - [(\ln[-\ln(P_{0})] - avglnln P_{0})^{2}s^{2} + mse(N+1)/N] \} (11)$ 

where *n* was dependent only upon the level of  $P_0$  when a minimal *d* value was determined using equation 10. Because  $T_n/n = P_0$  where  $T_n$  is the cumulative number of empty quadrate in *n* quadrates sampled, a stop line for binomial sampling was determined as  $T_n = n P_0$ . The level used for calculation of *d* and *n* was 0.05.

#### **Results and Discussion**

Data consisting of 100 estimates m,  $s^2$  and  $P_0$  for Sunn Pest adults were generated from a 2-year field study. These estimates were made from 0.1 to 2.82 Sunn Pest adults per quadrate.

Results of fitting Taylor's model to the data are listed in Table 1. The regression equation had a high coefficient of determination ( $r^2$ =0.95), indicating a good fit of the model. This supports results reported by Amir-Maafi (1). Model slope (b) was >1.00 (Table 1). According to Taylor (20), such a value indicates that the spatial pattern was aggregated. Sunn Pest adult distribution fitted Taylor's power law regression model, that is commonly employed for other insect species (5, 10, 17, 19).

The optimal sample size for a fixed precision level (for mean values around the economic threshold 2-4 adults/quadrate), was 50 samples. For low Sunn Pest densities with a desired accuracy of 0.25, (used by several authors in similar studies (e.g., 9, 18, 21)), the sampling effort needed was extensive (max 596 quadrates).

Numerical sequential sampling plans with fixed precision level (Figure 1.) were developed based on Taylor's parameters (Table 1). For scouting purposes we recommend that at least a 25% error level (d=0.5) be maintained when estimating populations of adult Sunn Pest. This should provide enough precision for management decision-making, while still being practical enough for use by field scouts and growers. However, for research, error levels of  $\leq 15\%$ would be more desirable, but consequently more costly in time and effort.



**Figure 1.** Stop lines for numerical sequential sampling of adult Sunn Pest populations with fixed d levels (=0.05).

Based on parameters from the regression of ln(m) on  $ln[-ln(P_0)]$  (Table 1), mean adult Sunn Pest density is displayed as a function of the proportion of empty quadrates (Figure 2).

Based on the parameters estimated for depicting the empirical relationship between mean density and proportion of empty quadrates (Table 1), stop lines for binomial sequential sampling were developed for adult Sunn Pest (Figure 3A). The curves shown in the graph represent sample sizes required to estimate m from P<sub>0</sub> with 95% confidence. For adults , when the desired sample size was less than 50 quadrates, the d values attained for the required sample sizes (Figure 3B) would be greater than 0.5 (which is greater than a sampling error of 0.25) and would likely lead to questionable population estimates. When the desired sample size was larger than 50, the resulting d levels were reduced and fell within a range of 0.25 to 0.5, except at extreme adult Sunn Pest densities (low and high densities) (Figure 3B).



**Figure 2.** Mean adult Sunn Pest density (*m*) as a function of the proportion of empty quadrates ( $P_0$ ).

Numerical and binomial sequential sampling plans based on the parameters determined from Taylor's power law analysis allowed sampling with a precision close to that initially selected and may provide an aid in population studies of Sunn Pest by enhancing efficiency and accuracy of estimating the densities of adults.

Binomial sequential sampling requires a larger sample size than complete count sampling. However, it may reduce the time required to process samples because checking whether a sample unit is occupied or not takes less time than counting the actual number of Sunn Pest present. Although binomial sampling may reduce the time and effort spent estimating population levels in the field, generally more samples are required and a higher variance is associated with the population estimates than are those associated with complete count or numerical sampling (3, 14). This tradeoff may make binomial sampling unattractive to those interested in its potential use for pest population monitoring. The larger variances associated with the estimates of population densities from binomial counts can be significantly decreased by using tally thresholds in sorting samples. A tally threshold is defined as the maximal number of pest individuals which may appear in a sample unit (3, 4, 14).

Therefore, to estimate adult Sunn Pest densities during spring on a weekly basis, numerical sequential sampling is recommended. This sampling plan should provide effective management of Ε. integriceps in wheat fields and minimize sampling time and cost. To use this sampling plan, the sampler accumulates adult counts as each 1 m<sup>2</sup> random quadrate is sampled. After each quadrate is sampled, the cumulative count is compared with the graph, leading to a decision to continue or not to continue sampling. When the stop line is crossed, sampling stops. The cumulative count can then be converted to a density and compared with the economic threshold.

## Acknowledgments

We thank A. Hassani and M. Safari for their technical assistance in this work. This research was supported by grants from DFID project No.R7863(C).

**Table 1.** Parameters estimated from the linear regression of  $\ln(s^2)$  on  $\ln(m)$  and  $\ln(m)$  on  $\ln[-\ln[(P_0)]$  for Sunn Pest populations (n=100) on wheat in Varamin, Iran during the growing seasons of 2002-2003.

Regression	Intercept (SE)	Slope (SE)	MSE	$r^2$
$\ln(s^2)$ on $\ln(m)$	0.355756 (0.05214)	1.137577 (0.038548)	0.06768	0.95
ln(m) on ln[-ln[(P <sub>0</sub> )]	0.110734 (0.2632989)	0.975475 (0.02014135)	0.03496	0.96



**Figure 3.** Stop lines for binomial sequential sampling of adult Sunn Pest populations (A) with the minimal *d* levels achived, (B) when sample sizes of 50,100 200 and 400 quadrates are used in the original prediction of *m* from P<sub>0</sub> respectively (=0.05).

#### الملخص

أمير-مافي، مسعود، بروس ل. باركر ومصطفى البوحسيني. 2007. أخذ العينات الثنائية والمتعاقبة لحشرة السونة البالغة (Eurygaster integriceps Puton). الصفحات 115–121.

خلال عامين من المسح الحقلي لحشرة السونة (Eurygaster integriceps Puton) على القمح الشتوي، استخرجت قواعد البيانات التي تألفت من 100 تقدير لمتوسط الكثافة (m حشرة سونة في المربع (=1 م<sup>2</sup>))، التباين (2<sup>°</sup>)، ونسبة المربعات الفارغة (P0). اعتمد كل تقدير لمجتمعات حشرة السونة على الأعداد في المربع الواحد لكل أخذ عرضي للعينة. استخدم قانون تايلور لتحديد العلاقات بين متوسط الكثافات (في م<sup>2</sup> (=مربع)) والتباينات لحشرة السونة على القمح الشتوي في فارامين، إيران. كان نموذج الانحدار > 1.0 لحشرة السونة، والذي أشار إلى أن البالغات أظهرت نماذج تجمع مكاني. تم تطوير خطط لأخذ العينات المتعاقبة لحشرة السونة، اعتمدت على الحسابات العددية (الكاملة) والثتائية (وجود أو غياب)، باستخدام مؤشرات تايلور وتلك التي قدرت من الانحدار الخطي لـ 10(m) على 10 [ ما – 10 (P0]]. اقترح استخدام خطط أخذ العينات ومستوى الدقة الذي يمكن تحقيقه.

كلمات مفتاحية: حشرة السونة، أخذ عينات، Eurygaster integriceps، آفات قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Determination of Fat Rate of Sunn Pest to Forecast Populations in Fields in Southeast Anatolia Region, Turkey

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#### Abstract

Karaca, V., E. Kiran, S. Fidan, M. Bashan, R. Canhilal and A. Demir. 2007. Determination of Fat Rate of Sunn Pest to Forecast Populations in Fields in Southeast Anatolia Region, Turkey. Pages 123-126.

Sunn Pest is one of the most important pests of wheat and barley in the Southeast Anatolia Region, Turkey. The objective was to determine the fat rate of Sunn Pest in wheat and barley fields and overwintering sites to forecast Sunn Pest populations in fields in Diyarbakir, Mardin, anlurfa, Elazı Provinces and the overwintering site in Karacadag. The lowest fat rate (2.36%) was in overwintered adults in wheat fields in the spring. The highest fat rate (29.82%) was in new generation adults just migrated to overwintering sites in autumn. In 1998, fat rate of Sunn Pest reduced 60.1%, in Sunn Pest in overwintering sites with snow vs. those in wheat fields. The fat rate of Sunn Pest was reduced 41.9% during the overwintering period. **Keywords:** Fat rate, forecast population, Southeast Anatolia Region, Sunn Pest.

# Introduction

In 1999, in Turkey, cereals were grown on 13,925,743 ha and yields of cereals were 28,749.710 tons. In the Southeast Anatolia Region, 1.593.237 ha were cultivated with cereals and 2.340.000 tons of cereals produced (1). One of major constraints to production of cereals is insects and the most harmful insect in Turkey is the Sunn Pest.

There are three well known species of Sunn Pest in Turkey. These are *Eurygaster integriceps* Puton., *E. maura* L. and *E. austriaca* Schrank. The most widespread is *E. integriceps* (2, 9).

In the past few years Sunn Pest has caused epidemic attacks in the Southeast Anatolia Region. Damage by nymphs and adults can be 100% when populatons are high. Over time Sunn Pest has become adapted via aestivation and hibernation to the climatic conditions and the cereals grown in this region. There are also a number of other factors which favor attack by Sunn Pest including the agricultural methods used and the topography of the area. Insects usually enter diapause when ecological conditions are not adequate for survival. These adverse ecological conditions cause changes in the physiological and biochemical structure of the insect and prepare them for the hard conditions. During this stage some nutrition, especially lipids, are increased in the insect body (5, 7). Survival in harsh conditions depends on the quantity of some nutritional factors such as lipids. Some researchers suggest that Sunn Pest epidemics depend on the quantity of lipids accumulated for diapause (3).

#### **Materials and Methods**

Approximately 100 adult Sunn Pests were collected from each wheat field in Diyarbakir, Mardin, Elazi and Sanlıurfa at the beginning and end of the growing season. An equal number of adults was collected at the beginning of migration and at the start of aestivation from the overwintering site at Karacadag. Lipid rate of adult Sunn Pest was determined according to Folch *et al.* (4). Collected adults were placed in an icebox and

transported to the laboratory. Males and females were separated in each sample. Then from each sample 20 adults (10 males and 10 females from each sample) were weighted and put into a homogenizer with chloroform and methyl alcohol (2/1). After homogenization, they were filtered and put into an evaporator for 1 h at  $65^{\circ}$ C to remove the solvents. Then the solid material was put into a desiccator and the quantity of total lipid content was determined.

## **Results and Discussion**

For Sunn Pest management an early estimation of population size and an awareness program was developed in the Southeast Anatolia region of Turkey. The purpose was to determine Sunn Pest density and areas to spray the following year. The most important parameter to determine Sunn Pest density is counts of adults per plant in overwintering sites. To estimate next year adult density in the fields the lipid rate of Sunn Pest at different times was determined. The lipid rate can be used for short and long term estimations and should be determined before Sunn Pest migrates to their over-wintering sites, during September-October. It should also be determined at the beginning of migration from the overwintering sites to wheat fields (3). For long term estimations, the most important parameter is the lipid rate of adult Sunn Pest, as they are moving from wheat fields to overwintering sites (8). Information regarding collected adult Sunn Pest is given in Table 1.

The highest (10.98) and smallest (5.91) lipid rate of adult Sunn Pest under snow in overwintering sites was found in March 1995 and in April 1998, respectively.

The highest (10.13) and smallest (2.36) lipid rate of Sunn Pest, collected from wheat fields was in April 1994 in Diyarbakir, in May 1998 in Elazig respectively.

The highest lipid rate of new generation adult Sunn Pest, collected from the overwintering site "Karacadag" was 29.82 in 1999 while the smallest lipid rate of new generation adult Sunn Pest, collected from wheat fields was in June 1993 in Sanliurfa Province.

When comparing the lipid rate of adults collected from under snow in overwintering sites to wheat fields, the lipid rate decreased 42.5, 21.8, 23.9, 60.1 and 55.7% in 1993, 1994, 1995, 1998 and 1999, respectively.

The smallest lipids rate (2.36%) of adult Sunn Pest collected from wheat fields was from individuals in Elazıg, 1998. When comparing the lipid rate of adult Sunn Pest collected under snow in Karacadag with the lipid rate of adults Sunn Pest collected from wheat fields, the highest decrease (60.1%) in lipid rate was seen in Elazı, 1998 It is suggested that lipid rate of Sunn Pest collected from wheat fields in spring decreases because of the long rainy period and cool days. Sunn Pest does not feed on wheat on rainy and cool days. Fedotov (3) suggested that digestion activities of Sunn Pest increases immediately when they fly from overwintering sites to wheat fields. They immediately start to feed on new green plants, and as a result of this feeding the water content of Sunn Pest increases and the lipid rate decreases.

When comparing the lipid rate of adults collected from overwintering sites at the beginning of migration in March, April to adults in overwintering sites in September, October the lipid rate decreased over the winter months 32.3, 3.2, 33.2 and 41.9% in 1994, 1995, 1998 and 1999, respectively.

The highest decrease of lipid rate between new generation adult and overwintered adults was 41.9%. It is thought that the lipid rate decreased because of metabolic activities of adult Sunn Pest during diapause. Fedotov (3) suggested that female and male Sunn Pest use 36 and 43% of their total lipids during diapause in overwintering sites. Kılıncer et al. (6) found that lipid rate of E. maura L. varied 15.64-23.04% in females 11.55-18.65% in males in the and overwintering site "Beynam".

The highest lipid rate (29.82%) was found in new generation adults collected from

the "Karacadag" overwintering site in 1999. It is also thought that new generation adults continue to feed on wheat grown around the overwintering sites, where conditions are still cooler and wheat has not been harvested. As a result Sunn Pest lipid rates are higher. Fedotov (3) suggested that ecological conditions are optimum for Sunn Pest and lipid rates reach a maximum level just before they fly to their overwintering sites.

Long-term studies are necessary to explain some of the parameters needed for an early estimation and awareness program.

 Table 1. Lipid rate of adult Sunn Pest collected from the Southeast Anatolia region of Turkey during the period of 1993-1999.

				Lipid rate (%)
Year	Month	Place	<b>Biological stage</b>	(female + male)
1993	April	Karacada	Overwintered adult	10.64
	April	Diyarbakır	Overwintered adult	8.52
	April	Mardin	Overwintered adult	6.12
	April	anlıurfa	Overwintered adult	6.12
	June	Karacada	New generation adult	11.80
	June	Diyarbakır	New generation adult	10.83
	June	Mardin	New generation adult	8.41
	June	anlıurfa	New generation adult	6.50
1994	March	Karacada	Overwintered adult	7.99
	April	Diyarbakır	Overwintered adult	10.13
	April	Mardin	Overwintered adult	6.25
	June	Diyarbakır	New generation adult	11.78
	June	anlıurfa	New generation adult	11.31
	June	Karacada	New generation adult	11.34
1995	March	Karacada	Overwintered adult	10.98
	April	Diyarbakır	Overwintered adult	8.36
	June	Diyarbakır	New generation adult	7.60
	June	Diyarbakır	New generation adult	14.80
	June	anlıurfa	New generation adult	9.36
	July	Karacada	New generation adult	13.13
1996	June	Diyarbakır	New generation adult	10.93
	September	Karacada	New generation adult	12.12
	April	Karacada	Overwintered adult	7.22
	July	Karacada	New generation adult	8.85
1998	April	Karacada	Overwintered adult	5.91
	April	Diyarbakır	Overwintered adult	4.61
	April	anlıurfa	Overwintered adult	3.09
	May	Elazı	Overwintered adult	2.36
	June	anlıurfa	New generation adult	15.93
	July	Elazı	New generation adult	13.61
1000	September	Karacada	New generation adult	15.15
1999	March	Karacada	Overwintered adult	8.80
	April	anlıurfa	Overwintered adult	6.48
	April	Diyarbakır	Overwintered adult	3.90
	May	Elazı	Overwintered adult	5.52
	June	anlıurfa	New generation adult	17.44
	June	Diyarbakır	New generation adult	23.43
	June	Karacada	New generation adult	29.82

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كاراكا، ف.، إي. كيران، س. فيدان، م. باشان، ر. جانهلال و ع. دمير. 2007. تحديد نسبة الدهون في حشرة السونة من أجل التنبؤ بمجتمعاتها في الحقول في منطقة جنوب شرق الأناضول، تركيا. الصفحات 123–126.

تعد حشرة السونة إحدى أهم آفات القمح والشعير في منطقة جنوب شرق الأناضول، تركيا. هدف البحث إلى تحديد نسبة الدهون في حشرة السونة في حقول القمح والشعير ومواقع البيات الشتوي للتنبؤ بمجتمعات حشرة السونة في الحقول في محافظات ديار بكر، ماردين، أورفة، ألاجيك ومواقع البيات الشتوي في قره جاداك. كانت أقل نسبة دهون (2.36%) في البالغات المشتية في حقول القمح في الربيع، في حين كانت أعلى نسبة دهون (29.82%) في بالغات الجيل الجديد التي هاجرت إلى مواقع البيات الشتوي في الخريف. في عام 1998، انخفضت نسبة الدهون في حشرة السونة في مواقع البيات الشتوي في حشرة السونة مع ما كانت ألم مواقع البيات في دقرل القمح في الربيع، في حين كانت أعلى نسبة دهون (29.82%) في بالغات الجيل الجديد التي هاجرت إلى مواقع البيات الشتوي في الخريف. في عام 1998، انخفضت نسبة الدهون في حشرة السونة في مواقع البيات الشتوي المغطاة بالثلوج بنسبة فترة السونة مع ما كانت عليه في الحشرة في حقول القمح. خفضت نسبة الدهون في حشرة السونة في مواقع البيات الشتوي.

كلمات مفتاحية: نسبة دهون، تنبؤ بمجتمع، منطقة شمال شرق الأناضول، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Temperature and Rainfall: Critical Factors for Management of Sunn Pest in Overwintering Sites

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#### Abstract

Skinner, M., B.L. Parker, M. El Bouhssini, W. Reid and Z. Sayyadi. 2007. Temperature and Rainfall: Critical Factors for Management of Sunn Pest in Overwintering Sites. Pages 127-131.

Humidity and temperature are critical factors influencing the survival and efficacy of entomopathogenic fungi. These factors must be considered when determining the best time to make a fungal application to Sunn Pest, *Eurygaster integriceps* Puton, overwintering sites. At ICARDA, Tel Hadya, Syria, temperature was recorded from 2002-2004 in a typical Sunn Pest overwintering site under pine trees at the soil/litter interface, where the insect is commonly found in winter. Precipitation data from the ICARDA weather station were also compiled. Considering the response of entomopathogenic fungi to temperature and the moisture requirements for germination, recommendations are made for the most appropriate timing of applications to Sunn Pest overwintering sites in the Aleppo, Syria region, taking advantage of climatic conditions to encourage infection.

Keywords: Temperature, rain, overwintering sites, Eurygaster integriceps, entomopathogenic fungi.

#### Introduction

Sunn Pest adults migrate to the foothills near wheat fields in late June or July to escape the hot summer temperatures. In Syria they seek refuge under trees, aestivating at the soil/litter interface, where it is cool and moist. They remain in these sites over the winter, returning to the fields in the early spring as temperatures rise. These overwintering sites represent an ideal environment for Sunn Pest management with entomopathogenic fungi because there is a concentrated adult population there, and high moderate humidity and temperatures compared with conditions in fields after the wheat harvest. These fungi have specific temperature and humidity requirements to survive, germinate and grow. Laboratory trials have been conducted to determine the temperatures necessary for fungal germination and growth. Weather data from typical overwintering sites provide critical information on the most suitable time to make

fungal applications in the overwintering sites to promote infection.

#### Materials and Methods

Precipitation data collected from the ICARDA meteorological station at Tel Hadya, Syria from 1998-2004 were compiled to determine monthly cumulative rainfall amounts by year and an average over all years. Ambient temperature data were also compiled over that period by determining the average daily temperatures.

From 2002-2004 temperature at the soil/litter interface was measured 0.5 m from the bole of pine trees in a typical Sunn Pest overwintering site in Tel Hadya, Syria using a Hobo Temperature Data Logger with four probes (Hobo Data Logger Co., Pocasset, MA, USA 02559-3450). One probe was placed at each of the cardinal directions around three trees. Temperatures were automatically

recorded every 30 min. Data were averaged among probes to determine the mean temperature at the soil/litter interface each day.

#### **Results and Discussion**

Based on laboratory tests at 15, 22 and 28°C, hyphal growth of several entomopathogenic fungi (some isolated from Sunn Pest) increased with temperature. At 35°C, no growth occurred. Germination of isolates of *Beauveria bassiana* (Balsamo) Vuillemin occurred most rapidly at 22 and 28°C (1, 2). This suggests that fungi applied to the overwintering sites will be most effective when temperatures are within that range.

Ambient temperature provides a relative indication of the most appropriate time to apply fungi, as it directly influences temperature at the soil/litter interface, where infection of Sunn Pest takes place. The moderating influence of the litter is evident from the data collected. Whereas the ambient minimum and maximum temperature in May was around 12 and 32°C, respectively, under the litter, the average temperature was around 17°C (Figures 1 and 2). In October, ambient minimum and maximum temperature was around 9 and 24°C, respectively, while under the litter, the average temperature was around 23°C (Figures 1 and 2).



Figure 1. Ambient maximum and minimum temperatures at the Sunn Pest overwintering site in Tel Hadya, Syria.



**Figure 2.** Maximum and minimum temperatures at the soil/litter interface beneath pine trees at the Sunn Pest overwintering site in Tel Hadya, Syria. Arrows indicate times when conditions are ideal for fungal spore germination.

The temperature at the soil/litter interface, where Sunn Pest would be targeted in the overwintering sites, is critical for infection to occur. As indicated in laboratory trials, fungal germination and growth occur most readily between 22-28°C, and if the temperature is too low or high, germination will not take place. Based on data collected from 2002-2004, applications made in October or April/May would be the most suitable (Figure 2). Many fungi are killed when temperatures reach  $\geq$  35°C. Therefore, an application earlier in the fall would not be beneficial because temperatures may reach that level.

Though rainfall amounts varied from year to year in Tel Hadya, Syria, when averaged over multiple years, the timing of significant precipitation periods was evident. After a dry period from June–August, rainfall usually begins in September, and increases steadily through December, after which it gradually decreases until May.

When temperature and precipitation factors are considered together with the

migration patterns of Sunn Pest, the ideal time to make a fungal application would be in October. At this time the Sunn Pest is resting under the litter in the overwintering sites, where it has been since July. The winter rains begin around this time, which will increase humidity under the litter. In addition temperature at that time under the litter is adequate for spore germination and fungal growth to occur. Fungi applied in the fall would also be in place should suitable conditions for infection to take place in the early spring before Sunn Pest migrate back to the wheat fields. Adults that had not been infected and killed during the fall or winter could come in contact with spores as they crawl through the litter to leave the overwintering sites. They could be killed by the infection after reaching the wheat field, or a low-level infection could affect their feeding and reproductive capacity. In addition, they could transmit the fungus to other adults or their offspring after reaching the wheat field.
# Acknowledgments

Support for this research was provided by US Agency of International Development

(USAID) Linkage funds, the Conservation, Food and Health Foundation, and Department For International Development (DFID).



**Figure 3.** Cumulative rainfall at the ICARDA weather station, Tel Hadya, Syria. Arrows indicate months when a fungal application would be ideal relative to precipitation.

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سكنر، مارغريت، بروس ل. باركر، مصطفى البوحسيني، ويليم ريد وزياد صيادي. 2007. الحرارة والمطر: عوامل حاسمة لإدارة حشرة السونة في مواقع البيات الشتوي. الصفحات 127–131.

تعد الرطوبة والحرارة من العوامل الحاسمة التي تؤثر في بقاء وفعالية الفطور الممرضة للحشرات. يجب أخذ هذه العوامل بعين الاعتبار عند تحديد الوقت الأفضل للقيام بالتطبيق الفطري في مواقع البيات الشتوي لحشرة السونة (Eurygaster integriceps Puton). سجلت درجات الحرارة في محطة بحوث إيكاردا (تل حديا، حلب، سورية) خلال الفترة 2002–2004 في موقع نموذجي لبيات حشرة السونة تحت أشجار الصنوبر عند السطح البيني للتربة/البقايا النباتية، حيث توجد الحشرات بشكل شائع في الشتاء، كما جمعت بيانات الهطول أيضاً من محطة مناخ إيكاردا. تم وضع توصيات للتوقيت الأكثر ملاءمة للتطبيقات في مواقع البيات الشتوي لحشرة السونة في حلب، مع الأخذ بعين الاعتبار استجابة الفطور الممرضة للحشرات ملاءمة للتطبيقات في مواقع البيات الشتوي لحشرة السونة في حلب، مع الأخذ بعين الاعتبار استجابة الفطور الممرضة للحشرات معادلت مثالية المرابة المرابة من أجل الإنتاش، بالاستفادة من ميزة الظروف المناخية لتشجيع العدوى.

كلمات مفتاحية: حرارة، مطر، مواقع بيات شتوي، Eurygaster integriceps، فطور ممرضة للحشرات.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# r -Amylase Activity in Hibernating Sunn Pest

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## Abstract

# Kazzazi, M., A.R. Bandani, S. Hosseinkhani and A. Ashoori. 2007. r-Amylase Activity in Hibernating Sunn Pest. Pages 133-138.

Cereals especially wheat and barley are strategic crops in Iran and much effort has been placed to increase yields. However, insect pests, in particular Sunn Pest, are the main hurdles to crop production and many control measures have been taken to suppress populations. Sunn Pest not only causes quantitative damage but it also destroys baking properties of wheat. The aim of the current study was to take a new approach to control of Sunn Pest. We identified and characterized  $\alpha$  -amylase activity of the insect. Adults were collected from their hibernating sites during the winter. These were dissected in saline and their salivary gland and their guts were separated and collected. Alpha-amylase activity in the midguts and salivary glands was determined using starch as the substrate. In the early spring when adults became active and returned to wheat fields they were collected and their  $\alpha$ -amylase activity determined. The amounts of  $\alpha$ -amylase activity in the midgut was about the same but there were differences in salivary gland . $\alpha$ -amylase activity between those adults collected from hibernating sites and those that were collected from fields in early spring. **Keywords**:  $\alpha$ -amylase, *Eurygaster integriceps*, midgut, salivary gland.

## Introduction

Sunn Pest, *Eurygaster integriceps* Puton, (Heteroptera: Scutelleridae), is well known as a serious limiting factor for production of wheat in the wide areas of the Near and Middle East, Eastern and South Europe and North Africa. This pest accounts for annual yield losses up to 100% for wheat in some areas of Iran before the beginning of chemical control strategies (10). The economic importance of this insect has increased over the years.

Sunn Pest, like other insect pests of wheat, lives on a polysaccharide-rich diet and depends to a large extent on effectiveness of their -amylases for survival. From the enzymes known to act preferentially on long  $\alpha$ -1,4-glucan chains such as native starch or glycogen, only  $\alpha$ -amylases have been found in insects (12). Alpha-amylase (-1,4-glucan-4-glucanohydrolases: E.C.3.2.1.1) hydrolyses starch, glycogen and related polysaccharides by randomly cleaving internal -1,4-glucosidic linkages. Therefore, -amylase catalyses the

initial hydrolysis of starch into shorter oligosaccharides which is an important step towards transforming starch into single units that can be assimilated by the organism. To accomplish their enzymatic functions they have a three-dimensional structure capable of binding to substrates and by the action of highly specific catalytic groups promotes the breakage of the glycoside links (7).

The enzyme is widely distributed in various bacteria, fungi, plants and animals and has a major role in the utilization of polysaccharides. In insects -amylase has been widely distributed and its characterization has been found for some insects e.g. *Sitophilus oryzae* L., *Prostephanus truncates* (Horn), *Deraecoris nebulosus* (Uhler) (1, 4, 9).

The recent increase in the study of insect digestive enzymes seems to make sense in the realization that the gut is the major interface between the insect and its environment. Hence, an understanding of digestive enzyme function is essential when developing methods of insect control such as the use of enzyme inhibitors and transgenic plants to control phytophagous insects. The impact of *E. integriceps* on economically important crops such as wheat and barley especially in developing countries makes this insect a very important subject of study. The aim of the current study is to identify and characterize Sunn Pest  $\alpha$ -amylase activity to gain a better understanding of its digestive physiology, which hopefully will lead to new strategies of the insect control.

# **Materials and Methods**

### Insects

Adult *E. integriceps* were collected from their hibernating sites (Karaj mountains) during the winter (10-25 February) and early spring (1-10 April) when they become active and ready to return to wheat fields.

## **Sample Preparation**

Enzyme samples were prepared by the method of Cohen (6) with some modifications. Salivary gland complexes (SGC) from these individuals were dissected under a light microscope in ice-cold phosphate buffer (pH 7). The salivary gland complex, including all lobes, accessory glands, and tubules, was exposed by holding the abdomen with fine forceps and gently pulling the head and prothorax away from the abdomen with another pair of fine forceps. The midgut was exposed by holding the body with forceps, and with another pair of forceps, pulling the ovipositor and last three or four segments of the abdomen away from the rest of the abdomen.

The SGC was separated from the insect's body, rinsed in ice-cold phosphate buffer and placed in a pre-cold homogenizer. The SGCs of 10 adults from *E. integriceps* were collected and ground in a homogenizer with 1 ml of buffer. The homogenate was transferred to a 1.5 ml centrifuge tube and centrifuged at  $15000 \times g$  for 20 min at 4°C. The supernatant was pooled and stored at  $-20^{\circ}$  C for subsequent analysis. The whole midguts from these insects were dissected under a light

microscope in ice-cold phosphate buffer (pH 7). Homogenization and centrifugation of gut were carried out as mentioned for SGCs.

#### -amylase Assay

Amylase activity was measured bv dinitrosalicilic acid (DNS) procedures of (3) with slight modifications using soluble starch as substrate. One unit of amylase activity is defined as the amount of enzyme required to produce one milligram of maltose in three minutes at 30°C. The reaction mixture contained 50 µl of 1% soluble starch in 50 mm phosphate buffer (pH 7) and 50 µl enzyme solution. The reaction was stopped by adding 100 µl DNS solution after incubation for 3 minutes. Then the mixture was heated in boiling water for 10 min and the absorbance at 540 nm was measured after cooling in ice and diluting with 800 µl distilled water.

#### **Kinetic Studies**

Enzyme kinetic studies were performed in triplicate employing a range of substrate concentrations from 0.76 mg ml<sup>-1</sup> to 3.84 mg ml<sup>-1</sup> with constant enzyme levels in the reaction volume. All reactions were at pH 7 and stopped by DNS after 5, 10, 20, 30, 40, 50 and 60 min. From absorbance readings the amount of maltose per min, was calculated for each of substrate concentrations.

# **Results and Discussion**

#### -Amylase Activity

Alpha-amylase activity in the salivary gland of the overwintering adults were less than feeding Sunn Pest which were collected from wheat fields in early spring (Table 1) e.g. total amylase activity in overwintering adults were 6.16 U (unit) but in the activated adults were 27 U. The reverse trend is observed in the amylase activity of midgut preparations. So, the enzyme activity in the overwintering adults was higher than that of feeding individuals e.g. in the overwintering adults total activity was 152 U whereas in feeding adults it was 109.5 U (Table 1).

Sample	Total activity (U)	Total protein (mg ml <sup>-1</sup> )	Specific activity (U/ mg ml <sup>-1</sup> protein)
SGC1 <sup>a</sup>	6.16	18.04	0.34
SGC2 <sup>a</sup>	27.0	23.72	1.13
Midgut1 <sup>b</sup>	152.0	24.40	6.22
Midgut2 <sup>b</sup>	109.5	29.00	3.77

**Table 1.** Comparison of  $\alpha$ -amylase activity in midgut and salivary glands of Sunn Pest.

<sup>a</sup> SGC1 and SGC2 are salivary gland complexes from overwintering and activated bugs which have been collected in the wheat field, respectively.

<sup>b</sup> Midgut1 and Midgut2 are guts from overwintering and activated bugs which have been collected in the wheat field, respectively.

A consumer's ability to use plant or animal materials for food is indicated by the presence of specific digestive enzymes (5). So, the current study shows that Sunn Pest like the other phytophagous insects relies on aamylase for starch digestion. The presence of  $\alpha$ -amylase activity both in the salivary gland and gut shows that the insect digest polysaccharides partially by salivary secretions and these amylase secretions are ingested by the insect along with partially digested starches to be used in the midgut to continue the breakdown of starch (4). The presence of amylase activity in the salivary

gland of other phytophagous heteropterous has been reported (4, 5, 13).

Salivary gland amylase activity was low in the overwintering stage in comparison with that of the feeding stage. This may indicate that feeding stimulates secretion of amylase by salivary glands. On the other hand, amylase activity in the midgut of the overwintering stage was high in comparison with those of the feeding stage probably due to accumulation of enzymes in the gut.

Silva and Terra (11) questioned whether heteropterous insects produce  $\alpha$ -amylase. However, results from this study and the other showed that this enzyme is produced in the salivary gland and gut of these insects especially Sunn Pest (13).

#### **Kinetic Studies**

The effect of different substrate concentrations on -amylase was determined from which  $K_m$  and  $V_{max}$  were calculated. Table 2 shows the value for 1/S and 1/V from which the Lineweaver-Burk plots (8) were derived.

Figure 1 shows a double reciprocal plot of amylase activity of midgut in the presence of different concentrations of substrates. Calculated  $K_m$  value for the enzyme was 1.6 mg and  $V_{max}$  was 0.6 mg ml<sup>-1</sup> min <sup>-1</sup>.

As can be seen from Figure 2,  $K_m$  and  $V_{max}$  for amylase activity of salivary gland were 19.1 mg and 0.28 mg ml<sup>-1</sup> min<sup>-1</sup>, respectively.

**Table 2.** The reaction velocities [Vo] produced by incubating enzyme extracts of midgut and salivary gland complexes (SGC) at different substrate concentration [S].

[S] (mgml <sup>-1</sup> )	1/[S]	Vo <sup>a</sup> (midgut)	1/Vo (midgut)	Vo (SGC)	1/Vo (SGC)
0.76	1.31	0.213	4.69	0.007	142.85
1.53	0.65	0.309	3.23	0.024	41.66
2.30	0.43	0.357	2.80	0.030	33.33
3.07	0.32	0.425	2.35	0.040	25.00
3.84	0.26	0.455	2.19	0.047	21.27

a. mgml-1min-

The study shows that kinetic parameters ( $K_m$  and  $V_{max}$ ) of amylase activity in the salivary gland and gut are different. This indicates that different forms of enzyme (isozymes) in salivary gland and gut are involved in starch digestion. The  $\alpha$ -amylase isozymes have been reported from different species of insects (2, 9).



Figure 1. Lineweaver-Burk Plot of  $\alpha$ -amylase activity of midgut of Sunn Pest from which  $K_m$  and  $V_{max}$  were calculated.



Figure 2. Lineweaver-Burk Plot of  $\alpha$ -amylase activity of salivary gland complexes of Sunn Pest from which  $K_m$  and  $V_{max}$  were calculated.

## Acknowledgements

The authors are grateful to the University of Tehran for providing funds for the current study. We also thank Mr V. Hosseini Naveh for his technical help.

# الملخص

كازازي، مجيد، علي. ر. بانداني، س، حسينخاني وأحمد آشوري. 2007. نشاط ألفا-أميلاز في حشرات السونة المشتية. الصفحات 133–138.

تعد محاصيل الحبوب وخاصة القمح والشعير من المحاصيل الاستراتيجية في إيران، وقد بذلت جهود كثيرة لزيادة إنتاجها. وتعد الآفات الحشرية، وبشكل خاص حشرة السونة، من العقبات الرئيسة لإنتاج المحاصيل وقد اتخذت العديد من إجراءات المكافحة لقمع مجتمعاتها. لا تسبب حشرة السونة ضرراً كمياً فقط لكنها أيضاً تدمر خصائص الخبيز للقمح. كان الهدف من هذه الدراسة اتباع طريقة جديدة لمكافحة حشرة السونة. حيث تم تحديد وتوصيف نشاط الفا-أميلاز في الحشرة. جمعت الحشرات البالغة من مواقع بياتها الشتوي خلال الشتاء، وتم تشريح الحشرات في وسط ملحي ثم فصلت وجمعت غددها اللعابية وأحشائها. تم قياس نشاط ألفا-أميلاز في المعي الأوسط والغدد اللعابية باستخدام النشاء. جمعت الحشرات في الربيع المبكر عندما عادت إلى حقول القمح بعد أن أصبحت نشيطة وتم قياس نشاط ألفا-أميلاز فيها. كانت كميات نشاط ألفا-أميلاز في المعي الأوسط نفسها تقريباً لكن كانت هناك اختلافات في نشاطه في الغدد اللعابية بين تلك الحشرات البالغة التي جمعت من مواقع البيات الفسها تقريباً لكن كانت هناك المبكر عندما ماليات.

كلمات مفتاحية:  $\alpha$ -أميلاز، Eurygaster integriceps، معي أوسط، غدة لعابية.

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# **Degree Days for Forecasting Migration of Sunn Pest**

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## Abstract

Amir-Maafi, M., M. Majdabadi, F. Aghdasi, B.L. Parker and F. Parsi. 2007. Degree Days for Forecasting Migration of Sunn Pest. Pages 139-145.

Seasonal flight of Sunn Pest, *Eurygaster integriceps* Puton, was monitored from 1992-2002. First migration was related to physiological time. (degree-days [DD]). A base temperature of 0°C yielded the lowest coefficient of variation for DD summations to first migration and was therefore selected for use in a linear heat unit system. The number of DDs accumulated to the day of first migration varied between years and locations. This was likely due to a variable relationship between air temperature and temperature in the Sunn Pest's overwintering site. A more accurate predictive index could be developed through an understanding of the relationship between overwintering site and ambient temperatures. **Keywords**: *Eurygaster integriceps* Puton, forecasting, migration, Sunn Pest, wheat pest.

## Introduction

Sunn Pest, *Eurygaster integriceps* Puton (Het.:Scutelleridae), is a primary pest of wheat and barley throughout West and Central Asia. Sunn Pest has caused major concern for more than 70 y (5). To prevent economic damage, management is commonly done with insecticides. For example, in Iran1.5 million ha of wheat must be sprayed each year to control this insect (2).

Sunn Pest overwinters as an adult, at elevations between 1000 and 2000 m., usually under bushes of *Artemisia* sp. and *Astragalus* sp., or in deciduous forests, usually oak (*Quercus* spp). They have an obligatory diapause (4). The mountain hibernation sites are usually about 10-20 km from wheat fields although they can be 150 km distant (9). When temperatures reach 12-13°C in the spring, migration takes place, usually in February-May. At this time the insects move to adjacent lower lying plains where cereal crops are grown (4).

Development of effective management strategies for Sunn Pest depends on an

understanding of their life history and population dynamics. One aspect that remains poorly understood is information on the first migration of adults from their overwintering sites in spring. The first migration can vary from year to year, so that field scouting activities must be routinely conducted for long periods. The objective of this study was to develop a procedure that would precisely predict the migration date of adult Sunn Pest from overwintering sites in the spring, based upon heat-unit accumulations. Such a system would provide the manager with a predictive for determining the time tool for implementation of a control strategy.

## Materials and Methods

The first flight of Sunn Pest was monitored from 1992-2002, in Arak, Saveh, Tafresh and Shazand, in Markazi Province, Iran. Ten commercial field sites in each location were sampled daily from 1 March to the first occurrence of adult Sunn Pest. A 38-cm sweep net was used and a sample consisted of thirty 180° sweeps taken randomly in each wheat field on each sampling date.

Daily maximum and minimum ambient temperatures were obtained from a national weather station which was usually located within 10 km of each location.

Thermal accumulation expressed as degree-days (DD) was computed using three commonly employed methods:

1)  $DD_1=(T_{max}+T_{min})/2-T_h$ Where  $T_{max}$ =daily maximum temperature  $T_{min}$ =daily minimum temperature  $T_h$ = temperature threshold

2) 
$$DD_2 = (T_{max} + T_{min})/2 - T_h$$
 (where  $T_{min} = (T_{max} + T_h)/2 - T_h$  (where  $T_{min} < T_h$ )

3)  $DD_3$ = The double sine method (1) of estimating thermal accumulation using daily maximum and minimum temperatures and assuming the sine curve as an approximation

of the diurnal temperature curve Heat accumulation for a day is estimated by computing the area under the curve.

Accumulated DDs were computed from 1 January to the first occurrence of adult Sunn Pest in the field.

# **Results and Discussion**

Seasonal migration data for Sunn Pest adults in different locations of Markazi Province indicated that flight began in mid March to April. Date of first migration for different locations of Markazi Province associated DD summations at 7 base temperatures and 3 methods of computation of thermal accumulation are listed in Tables 1-4. A base temperature of 0°C gave the lowest coefficient of variation in DDs, and is therefore the best estimate of the appropriate base temperature.

**Table 1.** Comparison of DD requirements for first migration of *E. integriceps* in Arak, Markazi Province, Iran, calculated from 7 base temperatures.

$DD_1 = Relef to (1) III Waterials and Wethous Section, DD_2 = (2), and DD_3 =$	Refer to (1) in Materials and Methods Section; $DD_2 = (2)$ , and $DD_3$	= (.	3	)	•
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Rase		-			Ye	ar / Dat	e of first	Migrat	ion					
temp		1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002		
(°C)		9/4	1/4	31/3	25/3	31/3	9/4	29/3	27/3	23/3	22/3	13/3	Mean	CV
0	$DD_1$	172	227	402	399	326	423	316	439	292	353	288	331	25
	$DD_2$	232	273	469	483	352	525	379	495	356	417	336	392	24
	$DD_3$	167	228	383	395	293	455	308	432	315	366	287	330	26
2	$DD_1$	109	143	256	244	194	272	207	286	196	232	199	213	25
	$DD_2$	163	198	357	384	260	405	288	384	273	328	269	301	26
	$DD_3$	125	141	273	289	214	333	222	307	228	259	226	238	27
4	$DD_1$	57	81	140	120	86	149	127	150	112	138	125	117	26
	$DD_2$	111	140	263	295	187	299	212	291	208	255	210	225	28
	$DD_3$	81	91	185	211	143	221	173	218	160	195	169	168	28
6	$DD_1$	23	38	65	48	31	68	76	72	50	72	65	54	32
	$DD_2$	73	97	183	214	123	208	148	212	156	191	158	160	30
	$DD_3$	51	68	121	138	78	142	110	142	110	133	106	109	29
8	$DD_1$	8	9	21	12	10	26	43	19	18	32	23	20	54
	$DD_2$	46	64	119	137	71	136	97	141	110	138	116	107	31
	$DD_3$	28	40	74	77	42	85	60	84	69	89	75	66	32
10	$DD_1$	3	0	4	1	3	6	20	4	7	6	4	5	100
	$DD_2$	24	39	73	74	36	80	64	84	69	94	79	65	34
	$DD_3$	13	21	40	37	19	43	40	43	33	54	44	35	36
12	$DD_1$	1	0	0	0	0	0	4	0	4	0	0	1	197
	$DD_2$	10	21	39	35	17	39	40	42	39	60	48	35	41
	$DD_3$	5	9	18	15	8	19	24	18	19	31	23	17	44

**Table 2.** Comparison of DD requirements for first migration of *E. integriceps* in Saveh, Markazi Province, Iran, calculated from 7 base temperatures.

$DD_1 =$	Refer to (	(1) in N	Aaterials and	l Methods	Section; D	$D_2 = (2)$ , and	$DD_{3} = ($	3).
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Rase				Y	'ear / Dat	e of first ]	Migration	n				
temn		1993	1994	1995	1996	1997	1998	1999	2000	2001		
(°C)		1/4	31/1	25/3	31/3	9/4	29/3	27/3	23/3	22/3	Mean	CV
0	$DD_1$	428.8	427.9	552.2	524.5	550.4	325.8	389.3	520.9	350.2	452.2	19.3
	$DD_2$	483.4	440.8	554.5	547.0	597.5	370.0	401.0	561.4	370.5	480.7	18.4
	$DD_3$	441.1	423.0	537.9	516.7	552.5	336.1	387.7	524.3	311.5	447.9	20.1
2	$DD_1$	295.3	303.4	416.2	372.0	400.8	209.5	280.1	369.2	244.6	321.2	22.4
	$DD_2$	356.5	336.3	435.8	405.6	472.3	277.6	307.3	445.2	291.3	369.7	19.5
	$DD_3$	316.1	294.7	401.2	358.5	421.5	232.9	259.3	400.3	254.3	326.5	21.8
4	$DD_1$	182.7	187.3	280.6	243.2	264.3	120.6	182.6	238.6	144.6	204.9	26.7
	$DD_2$	252.9	254.6	340.1	293.4	361.5	202.0	233.9	345.0	228.2	279.0	20.8
	$DD_3$	207.9	217.2	285.6	245.9	292.8	165.0	200.7	292.5	179.5	231.9	21.3
6	$DD_1$	91.3	94.4	151.7	136.5	150.2	60.9	99.4	134.4	67.6	109.6	31.6
	$DD_2$	176.5	186.1	264.6	206.6	271.1	138.5	174.2	260.8	174.0	205.8	23.4
	$DD_3$	137.1	139.9	212.1	156.7	198.3	97.0	137.8	199.9	119.6	155.4	25.5
8	$DD_1$	36.0	31.8	60.2	57.3	68.5	28.3	41.4	67.0	25.9	46.3	36.8
	$DD_2$	116.4	126.0	194.9	142.7	194.7	88.1	123.8	189.3	125.3	144.6	27.0
	$DD_3$	74.2	78.7	135.1	98.9	135.7	55.5	86.8	133.3	80.5	97.6	30.7
10	$DD_1$	7.3	6.3	17.7	22.5	17.8	10.7	9.6	26.1	7.5	13.9	52.2
	$DD_2$	69.7	75.0	128.6	91.7	129.5	50.8	82.0	126.1	81.1	92.7	31.0
	$DD_3$	41.2	42.1	76.6	58.8	78.9	31.0	47.0	79.7	45.9	55.7	33.2
12	$DD_1$	1.6	0.5	2.9	5.3	2.6	1.7	0.0	8.1	3.0	2.8	87.8
	$DD_2$	36.8	36.3	73.4	50.9	76.3	26.9	49.2	77.0	46.0	52.5	35.8
	$DD_3$	18.9	17.7	37.6	29.4	40.1	15.1	25.1	44.7	23.3	28.0	38.0

**Table 3.** Comparison of DD requirements for first migration of *E. integriceps* in Tafresh, Markazi Province, Iran, calculated from 7 base temperatures.

$DD_1 =$	Refer to	(1)11	n Materials	and Me	thods S	Section;	$DD_2 =$	(2), and	$DD_{3} = 0$	(3)	•
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Base				Year	/ Date of f	ïrst Migra	tion				
temp	-	1992	1994	1995	1996	1997	1998	1999	2001		
(°C)		9/4	31/3	25/3	31/3	9/4	29/3	27/3	22/3	Mean	CV
0	$DD_1$	142.5	375.4	382.8	268.3	433.9	285.2	419.1	323.0	328.8	29.2
	$DD_2$	260.1	475.2	476.3	355.4	542.0	408.1	505.1	383.8	425.7	21.6
	$DD_3$	193.3	428.4	406.9	302.5	447.0	296.3	434.4	356.5	358.1	24.8
2	$DD_1$	81.5	232.9	230.0	145.2	277.4	184.7	270.1	212.3	204.2	32.2
	$DD_2$	177.4	359.8	377.7	260.8	413.9	304.3	394.9	282.2	321.4	24.9
	$DD_3$	125.7	268.6	272.3	203.0	345.9	232.1	303.8	241.3	249.1	26.7
4	$DD_1$	35.3	125.8	105.7	58.0	155.9	111.1	145.3	125.9	107.9	38.6
	$DD_2$	111.6	264.0	290.4	182.3	302.7	218.8	303.0	200.3	234.1	29.1
	$DD_3$	76.4	187.8	204.8	122.6	225.3	164.9	219.0	161.9	170.3	29.8
6	$DD_1$	15.3	60.9	38.1	23.0	71.0	58.1	57.9	64.8	48.6	42.4
	$DD_2$	68.8	180.8	207.7	118.3	210.7	150.0	224.2	137.2	162.2	33.0
	$DD_3$	43.3	120.0	130.6	73.5	140.5	101.4	149.8	103.7	107.8	33.1
8	$DD_1$	5.3	18.8	8.8	10.5	24.0	18.6	12.5	25.4	15.5	47.3
	$DD_2$	39.8	113.0	131.2	67.8	138.4	102.3	153.7	92.1	104.8	36.2
	$DD_3$	20.6	67.5	71.5	38.4	85.2	65.2	88.9	60.5	62.2	36.8
10	$DD_1$	1.0	3.8	1.0	3.0	3.3	2.3	1.3	3.0	2.3	47.6
	$DD_2$	20.8	63.0	70.9	36.0	80.4	66.5	92.7	54.7	60.6	38.4
	$DD_3$	10.8	32.9	32.6	19.0	43.1	37.6	46.3	31.7	31.8	37.2
12	$DD_1$	0.0	0.0	0.0	0.3	0.3	0.0	0.0	0.0	0.1	185.2
	$DD_2$	10.3	29.3	29.1	18.3	39.4	39.8	47.5	30.1	30.4	39.6
	$DD_3$	4.6	12.8	11.5	8.9	18.1	19.3	20.3	14.3	13.7	39.3

**Table 4.** Comparison of DD requirements for first migration of *E. integriceps* in Shazand, Markazi Province,Iran, calculated from 7 base temperatures.

Base					Year / Dat	te of first	Migratio	n				
temp		1992	1994	1995	1996	1997	1998	1999	2000	2001		
(°C)		9/4	31/3	25/3	31/3	9/4	29/3	27/3	23/3	22/3	Mean	CV
0	$DD_1$	172.6	355.3	558.8	300.0	392.1	306.3	307.4	267.5	431.3	343.5	31.9
	$DD_2$	268.9	430.8	652.3	389.7	508.9	404.8	439.9	416.0	531.0	449.1	23.8
	$DD_3$	214.2	390.2	598.6	326.3	435.5	354.0	353.6	324.5	475.7	385.8	28.2
2	$DD_1$	102.4	213.0	359.1	160.3	242.8	199.3	168.8	154.3	283.8	209.3	36.9
	$DD_2$	176.2	308.8	510.3	286.9	384.9	297.8	339.9	311.3	408.8	336.1	27.6
	$DD_3$	162.1	309.8	483.0	281.6	363.9	299.0	309.8	284.4	407.5	322.3	27.8
4	$DD_1$	53.6	108.8	194.9	82.5	127.6	127.5	74.3	78.5	171.0	113.2	41.5
	$DD_2$	115.9	214.5	391.6	200.6	278.2	202.8	250.1	223.5	309.5	242.9	32.1
	$DD_3$	82.1	157.4	286.7	142.7	203.6	159.4	164.5	152.7	232.0	175.7	33.3
6	$DD_1$	25.7	45.0	87.6	47.3	56.8	73.8	29.8	36.8	92.5	55.0	44.6
	$DD_2$	78.9	139.8	288.1	126.1	191.9	131.6	172.4	154.0	229.0	168.0	36.9
	$DD_3$	55.3	93.5	194.2	78.3	128.9	95.6	104.6	96.5	161.0	112.0	38.3
8	$DD_1$	13.7	11.5	29.4	24.3	16.5	28.8	10.0	13.5	36.0	20.4	46.0
	$DD_2$	50.4	78.8	189.7	75.3	125.4	83.6	107.0	96.8	157.3	107.1	40.9
	$DD_3$	27.3	44.2	110.2	48.4	74.2	57.7	57.6	54.5	99.3	63.7	41.7
10	$DD_1$	5.2	2.5	5.1	5.8	3.5	4.8	1.8	2.5	8.0	4.3	45.4
	$DD_2$	30.4	39.5	105.2	45.8	76.2	53.3	59.8	55.0	100.5	62.8	41.5
	$DD_3$	18.7	20.7	53.3	27.3	37.3	31.3	28.8	27.9	56.6	33.6	39.7
12	$DD_1$	3.0	0.0	0.0	0.8	0.0	0.0	0.0	0.0	0.0	0.4	240.0
	$DD_2$	18.9	16.3	47.7	27.3	33.4	28.8	29.3	27.5	57.8	31.9	41.4
	$DD_3$	10.4	7.7	21.5	15.2	13.1	13.9	12.3	12.7	28.6	15.0	42.1

 $DD_1 = Refer to (1)$  in Materials and Methods Section;  $DD_2 = (2)$ , and  $DD_3 = (3)$ .

Taranuha and Telenga (7) in the Ukraine and Martin *et al.* (6) in Iran, reported that when temperatures reached 12-13°C spring migration began and it lasted from February to May. This predictive index was used with local weather records to predict the day of first migration during the study at the four locations in Markazi Province. However, mean temperatures did not exceed 12-13°C before migration at any of these four locations started (Figure 1).

Degree-days accumulated from 1 January to the beginning of the period during which first spring migration occurred varied (Table 1-4). The exact day of first migration was usually unknown and the comparison between locations during one year and between years at one location is difficult. Variability between locations in accumulated DDs to first migration could be attributed to population differences, such as those found between some of Phyllonorycter blancardella (F.) (8) and between some European populations of the cherry fruit fly, Rhagoletis cerasi (L.) (3). However, variability between years at one location would suggest that an index was unreliable for forecasting first spring migration. Considerable evidence exists that daily maximum and minimum air temperatures will not provide a reliable, location specific index. The number of DDs accumulated to the estimated day of first migration also varied considerably between locations each year and between years at one location. This variation (Tables 1-4) suggests that heat unit accumulations from 1 January using daily maximum and minimum air temperatures cannot be used to forecast, first spring migration of Sunn Pest.



Figure 1. Mean air temperature 10 days before Sunn Pest first migration in four locations, Markazi province.

It is concluded that the day of first migration of Sunn Pest cannot be accurately predicted using a DD index computed using only daily maximum and minimum air temperatures. The unreliability of a forecasting index for Sunn Pest based on air temperature is not unexpected. The temperature in a Sunn Pest overwintering habitat can often differ markedly from air temperature. Snow cover and radiant heating could be responsible for a variable relationship between air temperature and temperature in the overwintering habitat. The presence of snow would insulate Sunn Pest from both convective and radiant heating and in its absence thermal accumulation due to radiant heating could occur even on days when the maximum air temperature does not exceed this insect's threshold temperature. We think an index based on temperature in the Sunn Pest overwintering habitat was more accurate than an index based on air temperature.

However, Sunn Pest overwintering habitat temperatures are generally unavailable, whereas air temperatures are available for many locations in all of Iran's wheat and barley growing areas. Therefore, a predictive DD index using air temperature would be widely applicable using available weather information. Perhaps such an index could be developed if the relationship between relevant weather factors and Sunn Pest overwintering habitat temperature was understood.

## Acknowledgments

We thank Mr. A. Hassani and M. Safari for their technical assistance in this work. This research was supported by grants from Plant Pest and Disease Research Institute and Plant Protection Organization.

# الملخص

أمير-مافي، مسعود، م. ماجد أبادي، ف. أقدسي، بروس ل. باركر، و ف. بارسي. 2007. درجات الحرارة اليومية المؤثرة للتنبؤ بهجرة حشرة السونة. الصفحات 139–145.

تمت مراقبة الطيران الموسمي لحشرة السونة (Eurygaster integriceps Puton) خلال الفترة 1992–2002. كانت الهجرة الأولى عائدة للزمن الفيزيولوجي (DD] (degree-days) [DD]. أنتجت درجة الحرارة الأساس 0°س أقل معامل تباين لمجموع درجات الحرارة المؤثرة اليومية للهجرة الأولى واختيرت لذلك لأجل الاستخدام في نظام وحدة الحرارة الخطية. اختلف عدد درجات الحرارة اليومية المؤثرة المتراكمة إلى يوم الهجرة الأولى بين السنوات والمواقع. من المحتمل أن يكون هذا عائداً للعلاقة المتغيرة بين درجة حرارة الهواء ودرجة الحرارة في مواقع البيات الشتوي لحشرة السونة. يمكن تطوير دليل تنبؤ أكثر دقة عبر فهم العلاقة بين مراقع البيات الشتوى ودرجات حرارة الوسط المحيط.

كلمات مفتاحية: Eurygaster integriceps، نتبؤ، هجرة، حشرة السونة، آفة قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Chemical and Electrophysiological Studies on Sunn Pest

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#### Abstract

# Hall, D.R., A. Cork, M.C.A. Downham, D.I. Farman, P.J. Innocenzi and M. El Bouhssini. 2007. Chemical and Electrophysiological Studies on Sunn Pest. Pages 147-155.

The Sunn Pest, *Eurygaster integriceps* Puton (Hemiptera; Heteroptera; Scutelleridae) is the most damaging insect pest of wheat and barley in countries of West Asia. Previous work has shown that adult males produce a number of chemicals, and these have been implicated in attraction between the sexes. Volatiles were collected from insects by air aeration and trapping on Porapak resin and solid-phase microextraction (SPME) fibers. Males produced large amounts of homo- -bisabolene identified previously and smaller amounts of bisabolene and vanillin. Ethyl acrylate could not be detected, although reported previously. Females did not produce any of these compounds. These compounds were produced by males only during the light period and only after reaching sexual maturity. When disturbed, both males and females produced (E)-2-hexenal, tridecane, (E)-2-octenal and (E)-4-oxo-2-hexenal, compounds shown to be produced from glands in the thorax and thought to play a role in defense against predators. In analyses of volatiles from male or female bugs by gas chromatography linked to electroantennographic (EAG) recording, no significant responses were obtained from male antennae. In most analyses, no EAG responses were obtained from the antennae of female bugs, but in a very few cases a response to the homo- -bisabolene in collections from male bugs was observed. This indicates that receptors for this component are present on the antennae of the female bugs and suggests that the compound may have a role in intraspecific communication.

Keywords: Bisabolene, *Eurygaster integriceps*, Hemiptera, intraspecific communication, pheromone, Scutelleridae, semiochemicals, Sunn Pest, homo--bisabolene, vanillin.

# Introduction

The Sunn Pest, Eurygaster integriceps Puton (Hemiptera: Heteroptera: Scutelleridae) is the most damaging insect pest of wheat and barley in countries of West Asia with infestations spreading over 15 million ha in the affected area. Both nymphs and adults cause damage by feeding on leaves, stems and grains. Currently chemical insecticides are the main approach to control of Sunn pest, costing an estimated US\$ 40 million annually in the countries concerned. There is a need to develop alternative approaches to control of this pest that minimize or avoid use of chemical insecticides and reduce the hazards to human health, water quality and the environment in general. In this context, a better understanding of the ways in which Sunn pest locate mates and host plants could provide new methods for controlling the pest by interfering with these processes which are vital for survival of both the individual and the species.

There is good evidence that Heteropteran bugs communicate using sex pheromones, aggregation pheromones and alarm pheromones (2, 3, 12). Males of E. integriceps have been reported to produce a sex pheromone that attracts adult females (10, 11, 18). Some of the chemicals involved have been identified as vanillin (16), ethyl acrylate (17) and homo--bisabolene (15), but the behavioral effects of these chemicals have not been fully evaluated and it is not even known whether all the chemicals making up the pheromone blend have been detected and identified. In the absence of this information, no work has been done to develop use of these materials in management of Sunn Pest,

although a pheromone that attracts females would be particularly useful in both monitoring and control.

The aim of these studies was to analyze volatiles collected from male or female *E. integriceps* and identify components present. Rates of production of the components were measured under various conditions in order to determine whether their production was associated with particular times or stages in the life of the adults, and hence to gain information on their possible function. Electrophysiological recording from the insect antennae was then used to determine whether the insects can detect any of these components by means of receptors on the antennae.

## Materials and Methods

#### Insects

Male and female *E. integriceps* were sent separately as live adults from ICARDA to NRI by air freight. Those sent from December – March were collected from overwintering sites and hence unmated. Those sent April – May were collected from wheat fields and were probably mated. At NRI the male and female bugs were maintained in separate rooms on wheat seedlings with L:D 16:8 hr and temperatures  $25^{\circ}$ C :  $20^{\circ}$ C.

#### **Collection of Volatiles**

Collections of volatiles were carried out in the room used to house the corresponding sex. Bugs (1-10) were placed in a glass chamber (12 cm x 4 cm dia.) containing wheat seedling stems (approx 10). A diaphragm pump (Capex Mk II, Charles Austen, UK) was used to draw air at 1 l/min into the chamber through an activated charcoal filter (20 cm x 2 cm, 6-18 mesh) and out through a collection filter containing Porapak Q (200mg, 50-80 mesh, PhaseSep), held between plugs of silanized glass wool in a Pasteur pipette. The Porapak was purified by Soxhlet extraction with chloroform for 8 hr, and filters were washed well with dichloromethane immediately before use. Adsorbed volatiles were removed from the filters with dichloromethane (Pesticide grade; 3 x 0.5 ml).

For collection of volatiles by solid-phase microextraction (SPME), a polydimethylsiloxane fiber (100  $\mu$ m coating) was used. The bug(s) was held in a glass vial (5 ml) sealed with aluminum foil and sampling was done for 15-30 min. The fiber was desorbed in the GC injector for 1 min at 220°C using conditions described below for the polar GC column.

## Analyses by Gas chromatography (GC) and Gas Chromatography linked to Mass Spectrometry (GC-MS)

GC analyses were conducted using Carlo Erba Mega Series 5300 instruments with a fused capillary column (25 m x 0.32 mm i.d.) coated with polar CPWax52CB (Carbowax 20M equivalent; Chrompack, The Netherlands) or CPSil5CB (methyl non-polar silicone. Chrompack). The carrier gas was helium at 50 kPa, and the oven temperature was held at 50°C for 2 min then programmed at 6°C/min to 240°C. Injection was splitless (200°C) and detection was by flame ionization detection (FID, 240°C). Data was captured and processed using EzChrom 6.1 software. Retention Indices (RI) were calculated relative to the retention times of straight-chain hydrocarbons.

GC-MS analyses were carried out with a fused silica capillary column (25 m x 0.25 mm i.d.) coated with polar CP Wax 52CB linked directly to a Finnigan Ion Trap Detector 700 (ITD) operated in electron impact or chemical ionization (*iso*-butane) mode. GC conditions were as above. Spectra were compared with those in the ITD NBS/NIH/EPA library, the ITD terpenoid library (1), and a library generated from samples analyzed previously at NRI.

### Electrophysiology

During 2003-2004, electroantennograms (EAG) were recorded from whole bugs. After

anaesthetisation with carbon dioxide, the insect was placed in a plasticine block, its antennae restrained with copper wire hooks. Glass capillaries (50 x 0.2 mm diameter) containing 0.5 Μ KC1 solution with 1% polyvinylpyrrolidine were placed over silver wire electrodes attached to a DC amplifier (UN06. Syntech model, Hilversum, Netherlands). The electrodes were held in place by micromanipulators (Leitz, Wetzlab, Germany). The amplifier was connected to a PE Nelson 5300 interface, and the output captured and processed using Turbochrom 4 software (Perkin Elmer-Nelson, Beaconsfield, Bucks, UK). The recording electrode was always positioned distally to the indifferent electrode either in the tip of the fifth antennal segment or in the membrane connecting the fourth and fifth antennal segments. The indifferent electrode was inserted into the intersegmental membrane of the scape and pedicel of the same antenna.

During 2006, EAG recording was carried out with a portable device (INR-02; Syntech) consisting of integrated electrode holders, micromanipulators and amplifier. An antenna was excised at the base and suspended between the glass electrodes which were cut so that they just accommodated the ends of the antenna. The signal was amplified x500 and the amplifier was connected to the GC as a detector device. Data was processed with EZChrom Elite (v3.0).

For linked GC-EAG analyses, effluent from the GC outlet was partitioned equally between the FID and the insect preparation. The GC column effluent was blown at 15-sec intervals over the EAG preparation with a three-second pulse of nitrogen (200 ml/min) (7).

#### **Post-thoracic Gland Studies**

Droplets were collected from an overwintered female Sunn Pest produced from paired orifices located on the thorax and presumably leading to the metathoracic glands. Droplets were dissolved in dichloromethane (1 ml) and analyzed by GC on the polar column.

## **Synthetic Chemicals**

(*E*)-2-hexenal, tridecane, (*E*)-2-octenal and vanillin (4-hydroxy-3-methoxybenzaldehyde) were obtained from Sigma-Aldrich (Gillingham, Dorset, UK). (*E*)-4-Oxo-2-hexenal was synthesized at NRI (9).

# Results

# Analysis of Volatiles Collected from Live Sunn Pest

GC analysis of volatiles collected from male E. integriceps on Porapak showed a single major peak accounting for over 90% of the total peak area (Figure 1). This peak had mass retention data (Table 1) and spectra (Figure 2) consistent with the (4Z,4E')-4-(1,5'-dimethyl-4'-heptenylidene)-1-methylcyclohexene (homo- -bisabolene) structure proposed by Staddon et al. (15) (Figure 3). This peak was never present in similar collections from female bugs (n = 15collections from batches of 1-10 bugs) (Figure 1).



**Figure 1.** GC-MS traces of volatiles collected from male (upper) and female (lower) *E. integriceps* on Porapak (polar GC column).

**Table 1.** GC retention data for compounds detected and searched for in volatiles from male *E. integriceps.*

	Reten	tion Index <sup>a</sup>
Compound	Polar	Non-polar
homobisabolene	1810	1619
Bisabolene	1720	1523
Vanillin	2517	1352
(E)-2-hexenal	1218	812
Tridecane	1300	1300
(E)-2-Octenal	1434	1031
(E)-4-oxo-2-hexenal	1608	837
Ethyl acrylate	1030	

<sup>a</sup> Retention Index relative to retention times of nhydrocarbons.



**Figure 2.** Mass spectra in EI (upper) and CI*iso*butane (lower) modes of major component in volatiles collected from male *E. integriceps*.

Minor components in volatiles from male bugs were identified as bisabolene and vanillin (Figure 3). When collections made from batches of 10 male bugs were analyzed (n= 8), amounts of these components relative to the major component were 5.3% (SE 1.0) and 1.4% (SE 0.2). In these analyses the amounts of homo- -bisabolene were quantified by peak area relative to dodecyl acetate and that of vanillin by peak area relative to external standard.



**Figure 3.** Structures of compounds found in volatiles from male *E. integriceps*: (I) homo--bisabolene; (II) bisabolene; (III) vanillin.

Over 50 collections were made from male bugs. Amounts of the homo--bisabolene produced were variable. During initial studies, amounts collected from 1, 3 or 10 male bugs over 24 hr periods were equivalent to 0.80, 0.67 and 0.52 µg/bug/hr respectively, suggesting that presence of other males did not influence production markedly. Analysis of collections from batches of 10 male bugs made over 24 hr periods (n = 7) gave a mean of 1.4 µg/bug/hr (SE 0.4). However, the homo- -bisabolene was produced only during the light period: collections from four separate males during the light period gave a mean of 2.54 µg/bug/hr (SE 1.06) and no detectable homo--bisabolene during the dark period. During 2006 the period of maximum production of the homo--bisabolene was narrowed down further. Using two replicates of 10 bugs over seven separate days, production during the first six hours of the light period was 0.15 µg/bug/hr (SE 0.05), during the second six hours of light 5.42 µg/bug/hr (SE 2.63) and during the 12-hr dark period 0.22 µg/bug/hr (SE 0.09). Occasionally much higher rates were recorded, e.g. 11.3 and 6.2 µg/bug/hr recorded from different bugs over 8 hr periods.

Most collections were made at NRI from January – April from male bugs collected as overwintering adults during October – December and presumed to be virgin. Collections made in May (n= 4) from bugs collected from the field and presumed to be mated still produced significant amounts of the homo- -bisabolene. However, when a batch of bugs was sent from ICARDA on 2 November 2002 and collections were made from two batches of 2 males on 5 November, 7 November and 19 November, no homo-bisabolene could be detected. Collections made on 16 December from this batch contained the homo--bisabolene but at low levels of 0.02  $\mu$ g/bug/hr measured over 24 hr, compared with the figures of 1-2  $\mu$ g/bug/hr obtained during January.

In a few collections from both males and females, four other components were detected and these were identified as (E)-2-hexenal, tridecane, (E)-2-octenal and (E)-4-oxo-2-hexenal by comparison of their GC retention times and mass spectra with those of authentic samples (Figure 4). These were subsequently shown to be produced from the metathoracic gland and are considered to be defensive compounds.



**Figure 4.** GC-MS analyses of volatiles collected from male (upper) and female (lower) *E. integriceps* showing "defensive" compounds (polar GC column: scan 429 (*E*)-2-hexenal; 542 tridecane; 729 (*E*)-2-octenal; 980 (*E*)-4-oxo-2-hexenal; 1120 bisabolene; 1232 homo-bisabolene).

Ethyl acrylate was never observed in the collections of volatiles from male or female bugs separately (n = >50 and 15 respectively) or together (n=14) trapped on Porapak. As the ethyl acrylate eluted just after the solvent front in these analyses, trapping of volatiles on an SPME fiber was also carried out with subsequent solvent-less thermal desorption in the GC injector. Ethyl acrylate could not be detected in volatiles from female (n= 3), male (n=5) or male and female bugs (n=10), although large amounts of the homo-bisabolene were observed in samples from males alone and in the presence of females (Figure 5). Similar analyses carried out in June (n= 2 for males and females) also showed no ethyl acrylate but no homo--bisabolene either.



**Figure 5.** GC analysis of volatiles collected from male *E. integriceps* by SPME (polar GC column: ethyl acrylate elutes at 4.4 min; bisabolene 18.7 min; homo- -bisabolene 20.5 min).

#### **GC-EAG Analyses**

Linked GC-EAG analyses were carried out during 2003-2004 with recording from the antennae of intact female *E. integriceps*. No significant responses (> 10% background) were observed in analyses of volatiles from male bugs collected on Porapak (n = 12) or by SPME (n = 1) or of volatiles from a male and female bug together collected by SPME (n =2). No response was observed to synthetic ethyl acrylate (n = 2).

Further studies were carried out during 2006 using isolated antennae from virgin female (n=28) or male (n=12) bugs. Samples

analyzed contained volatiles from male bugs (female preparation n=19; male n=5), volatiles from female bugs (female preparation n= 8; male n= 8) or defensive compounds derived from the metathoracic gland (see above) (female preparation n=10; male n=6). No EAG responses were ever observed from female or male antennae to components in volatiles collected from female bugs or to the defensive compounds. Similarly no EAG responses were obtained from male antennae to volatiles collected from males, and, in most cases, no EAG response was observed from female antennae to the volatiles collected from males. However, on one occasion a good EAG response was recorded from a female antenna to the homo- -bisabolene (40 ng injected; 20 ng to the EAG preparation) in volatiles collected from male bugs (Figure 6). This could not be repeated on the same preparation, although on three other occasions a much smaller EAG response was observed to this component (Figure 7).



**Figure 6.** GC-EAG analysis of volatiles collected from male *E. integriceps* on Porapak with female *E. integriceps* EAG preparation (polar column: bisabolene at 11.56 min; homo--bisabolene at 12.53 min; EAG response \*).

#### **Post-thoracic Gland Studies**

GC analysis of fluid exuded from glandular openings on the thorax of a female *E. integriceps* bug showed four major components (Figure 8). These were identified from comparison of their GC retention times and mass spectra with those of authentic standards as (E)-2-hexenal, tridecane, (E)-2-octenal and (E)-4-oxo-2-hexenal. Traces of (E)-2-octenol and (E)-2-octenyl acetate were also detected.



**Figure 7.** GC-EAG analysis of volatiles collected from male *E. integriceps* on Porapak with female *E. integriceps* EAG preparation (polar column: bisabolene at 11.56 min; homo--bisabolene at 12.53 min; EAG response \*; lower trace is expansion of upper).



**Figure 8.** GC analysis of droplets collected from thoracic glands of female *E. integriceps* (polar GC column: (*E*)-2-hexenal at 5.85 min; tridecane 7.55 min; (*E*)-2-octenal 10.40 min; (*E*)-4-oxo-2-hexenal 14.30 min).

# Discussion

Male E. integriceps bugs were confirmed to produce large amounts of the homo-bisabolene identified previously by Staddon et al. (15). This component typically constituted >90% of the volatile materials produced and amounts of up to 11.3 µg/bug/hr were recorded. The only significant minor components detected were bisabolene and vanillin, the latter having been reported by Ubik et al. (16) and Staddon et al. (15). Ethyl acrylate reported by Vrkoc et al. (17) was never detected in volatiles from either male or female *E. integriceps*. The homo--bisabolene insect is а unique compound in semiochemistry, although bisabolene epoxides are produced by males of some species of related Pentatomid bugs, e.g. Nezara viridula L. (5) and Acrosternum hilare (Say) (13).

The above compounds were not produced by female bugs. They were only produced by the male bugs on reaching sexual maturity and then only during daylight hours when the bugs are most active (6). Production was shown to continue after the males have mated, but the males are capable of mating more than once. These observations suggest that the compounds play some role in mating.

In linked GC-EAG analyses of volatiles collected from male or female bugs, no responses were ever obtained from the antennae of male bugs. Similar results were obtained in many analyses with the antennae of female bugs, using either intact insects or isolated antennae. However, in one analysis a good EAG response to the homo- -bisabolene in volatiles collected from male bugs was obtained, and in three other analyses a much smaller response was observed. To the best of our knowledge, this is the first report of such an EAG response in Pentatomid bugs. The reason for the extreme variability in responsiveness is unknown, but these results do suggest there are receptors for the homo-bisabolene produced by male *E. integriceps* on the antennae of female bugs and hence that this compound has a role in intraspecific communication in this species.

The origin of the homo- -bisabolene was not investigated, although in male N. viridula the bisabolene epoxides are produced from glands in the ventral abdominal cuticle (14). In E. integriceps secretions from glands on the thorax contain (E)-2-hexenal, tridecane, (E)-2octenal and (E)-4-oxo-2-hexenal. These are commonly found in thoracic glands of Heteroptera (2) and are generally considered to be defense compounds against arthropod predators. These compounds were only observed very occasionally in volatiles collected from both male and female bugs. This may have resulted from stress experienced by the bugs during the collection process or possibly due to the presence of one or more dead individuals from which the contents of the thoracic glands "leaked" out.

# الملخص

هول، ديفيد ر.، آلن كورك، مارك س. أ. داونهام، ديدلي آي. فارمان، باول ج. إنوسينزي ومصطفى البوحسيني. 2007. دراسات كيميائية وكهروفيزيولوجية على حشرة السونة. الصفحات 147–155.

تعد حشرة السونة (Eurygaster integriceps Puton) من أكثر الآفات الحشرية ضرراً للقمح والشعير في دول غرب آسيا. أظهرت دراسات سابقة أن الذكور البالغة تنتج عدداً من الكيماويات التي تلعب دوراً في الانجذاب بين الجنسين. جمعت المواد المتطايرة من الحشرات بواسطة إشباع الهواء واصطيادها باستخدام ألياف Porapak (مادة تستخدم لامتصاص واصطياد الغازات والمواد المتطايرة) الصمغية والصلبة للاستخلاص الدقيق (SPME). تنتج الذكور كميات كبيرة من هومو – بيسابولين المعرف مسبقاً وكميات أصغر من بيسابولين وفانيلين. لم يتمكن من اكتشاف إيشيل أكريلات، رغم تسجيله مسبقاً. لم تنتج الإناث أياً من هذه المركبات، ولكن أنتجت الذكور هذه المركبات فقط خلال فترة الضوء وبعد الوصول إلى النضج الجنسي. عندما يتم إزعاجها نتتج كل من الذكور والإناث مركبات (E) – 2-هيكسينال، ترايديكان، (E) – 2-أوكتينال و (E) – 4-أوكسو – 2-هيكسينال، وتنتج هذه المركبات من غدد في الصدر تلعب دوراً في الدفاع ضد المفترسات. عند تحليل المواد المتطايرة من الحشرات الذكور أو الإناث بواسطة غاز الكروماتوغرافي المربوط إلى مسجل إليكتروأننتوغرافي (EAG)، لم يتم الحصول على أي استجابات معنوية من قرون استشعار الذكور. في معظم التحاليل، لم يتم الحصول على استجابات (EAG) من قرون استشعار حشرات الإناث. ولكن في حالات قليلة جداً لوحظت استجابة لهومو – بيسابولين في تجمع من الحشرات الذكور. هذا يشير إلى أن مستقبلات هذا العنصر موجودة على قرون استشعار إناث الحشرة، ولهذا يمكن أن يكون لهذا المركب دور في التخاطب بين أفراد النوع الواحد. كلمات مفتاحية: بيسابولين، Scutelleridae الايمانية، حشرة السونة، هومو – بيسابولين.

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# The Role of Vibratory Signals in Mating Behavior of Sunn Pest

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#### Abstract

Green, S.V., M.C.A. Downham, D.I. Farman, D.R. Hall and M. El Bouhssini. 2007. The Role of Vibratory Signals in Mating Behavior of Sunn Pest. Pages 157-165.

Eurygaster integriceps Puton (Hemiptera; Scutelleridae) is the most damaging insect pest of wheat and barley in countries of West Asia. This study aimed to characterize vibratory signals produced by male and female E. integriceps and to investigate the role of these signals in intraspecific communication, both in the presence and absence of odor signals from the insects. The male "calling song" consists of short chirps repeated at regular intervals with a call rate of 4.6 chirps/sec, mean chirp duration of 0.085 sec and inter-chirp interval 0.17 sec. When approached, a second male typically produces a distinctive agonistic call or "rivalry song" in which chirps are repeated at a much faster rate of 10.4/sec. Females occasionally call spontaneously, but they are much more likely to do so in response to male calling song, or indeed when any other conspecific is nearby. The female song structure does not differ from the male calling song in its chirp rate, although it is generally produced at lower amplitude and the vibratory movements generating the calls are not always evident. Typically the chirp rate increases as the male gets close to the female, and once mounted with genitalia engaged, vibratory signals subside. A Y-track bioassay system was developed and when vibratory signals produced by live insects were used as source, male bugs were significantly attracted to signals from both females and males. Females were not attracted to signals from either sex. When recorded signals were used as source, rather different results were obtained in that females were significantly attracted to signals from males and males were not attracted to signals from males or females. In the presence of odor collected from male bugs on Porapak, females showed no preference for vibratory signals from live males or the recorded signal. The role of vibratory signaling in mate location in this species and the interaction of these signals with production of volatiles are discussed.

Keywords: *Eurygaster integriceps*, Hemiptera, Heteroptera, intraspecific communication, Scutelleridae, vibrational signals.

## Introduction

*Eurygaster integriceps* Puton (Hemiptera; Scutelleridae) is the most damaging insect pest of wheat and barley in countries of West Asia with infestations spreading over 15 million ha in the affected area. Currently chemical insecticides are the main approach to control costing an estimated US\$ 40 million annually in the countries concerned. There is a need to develop alternative approaches to control of this pest that minimize or avoid use of chemical insecticides and reduce hazards to human health, water quality and the environment in general. In this context, a better understanding of the ways in which Sunn Pest locate mates and host plants could provide new methods for control by interfering with these processes which are vital for survival of both the individual and the species.

The suborder Heteroptera, a group which includes the "stink bugs", is associated with production of volatile chemicals. There is good evidence that heteropterans communicate using sex pheromones, aggregation pheromones and alarm pheromones (1, 2). More recently, however, growing evidence substrate-borne suggests that vibratory signaling is also widespread in these insects (8). This mode of communication has long been recognized in the other hemipteran suborder, the Homoptera (5, 12).

In E. integriceps, males are known to produce volatile compounds, generating a characteristic aroma of vanilla. Previous research has analyzed these volatiles and suggested that certain components are active in intersexual communication (9, 10, 13, 14, 15). Preliminary observations on the behavior of E. integriceps at NRI in 2002 suggested that vibratory signals also occur in this species, since males were observed repeatedly to shake their abdomens through a narrow vertical arc of approximately 10-20 degrees. An earlier study by Zdarek and Kontev (16) noted this behavior, but apparently did not make the link between these movements and vibratory signaling through the substrate.

The function of heteropteran vibratory signals is in communicating the identity and location of the caller to potential mates or rival males. In *Nezara viridula* L. it is believed that long-range attraction to the vicinity of the caller is mediated by chemical signals, while vibratory signals direct the final stages of mate location (4, 6, 7). Recent evidence, however, further suggests that chemical and vibratory communication operate together in the later stages of mate location and/or courtship, since pheromone emission by male *N. viridula* increases when they are exposed to female vibratory signals, as occurs prior to mating (11).

This study was done to characterize vibratory signals produced by male and female *E. integriceps* and to investigate the role of these signals in intraspecific communication, both in the presence and absence of odor signals from the insects. This work was carried out in parallel with studies of behavioral responses of Sunn Pest to volatiles (4) and investigation of vibratory signals (8).

## **Materials and Methods**

#### Insects

Male and female *E. integriceps* were sent separately as live adults from ICARDA to NRI by air freight. Those sent from December – March were collected from overwintering sites

and hence unmated. Those sent April – May were collected from wheat fields and were probably mated. At NRI the male and female bugs were maintained in separate rooms on wheat seedlings with L:D 16:8 hr and temperature 25°C:20°C, respectively.

### **Recording of Vibratory Signals**

Recording of signals was hampered initially by extraneous background vibrations, but the establishment of a vibration-free recording surface greatly reduced the problem. This consisted of a slate slab resting on a semiinflated tire inner tube, mounted on a marble table. The substrate used for recording was a recently-cut stem of perennial rye grass (Lolium perenne L.) clamped vertically and held in contact with the membrane of a dynamic microphone (UHER M517) by a small strip of double-sided adhesive tape (2 mm x 2 mm). Virgin male and female insects were placed onto the stem above the microphone, approximately 50cm below a 60W light bulb at a temperature of 26.5-28°C. Vibrations were transduced by the microphone into an analogue electrical signal, recorded using an IC reel-to-reel tape recorder (UHER 4000 Report) or a cassette recorder (UHER CR210) onto a chromium oxide audio cassette. Recordings were played back for analysis using Avisoft SAS-Lab Pro acoustic software. Recordings of signals were made mostly using known virgin insects. Damaged or lethargic individuals were not used.

#### **Bioassay of Responses to Vibratory Signals**

The role of vibratory signals in locomotory behavior of *E. integriceps* was investigated in a vertical Y-type bioassay constructed from three bamboo barbecue skewers (20 cm x 2 mm dia.) with tapered ends. The stem of the Y was clamped vertically and the arms of the Y were held approximately at right angles so that the three sticks did not touch but the gaps between the ends were approximately 1.5 mm. The test insect ascending the stem stick to the junction could thus sample both choice arms for vibrations before deciding on which

direction to take. In practice this did not always take place since bugs sometimes walked up one side of the vertical stem and then climbed straight onto the nearest choice arm without touching the other.

Signals were delivered to the arms of the Y-track by live *E. integriceps* (3 males or females) placed on the end of the stick and concealed from the test insect by a filter paper (7 cm dia.) placed on the arm with the stick through its centre. Alternatively a repeated recording of male or female calling song vibrations was transmitted to the stick by means of an intermediate stick held in contact with a loudspeaker cone. In the latter case, the playback amplitude was adjusted to match vibratory signals from live males, intensities being compared by means of a microphone membrane as previously used in recording.

Bioassays were conducted in an insectary which at that time was used for these studies alone. Lighting was provided by 4 standard fluorescent tubes. Temperature was maintained at 26±1°C and relative humidity at  $40\pm15\%$ . For each bioassay, the test insect was introduced onto the vertical stem approx. 7 cm below the tip by means of a bamboo sliver such that it alighted facing upwards. Each bug was then allowed 5 min in which to approach the tip and make a choice by climbing onto one or other choice arm. Insects moving >3cm downwards from the starting position were replaced. Data were analyzed by a Chisquared test and results were considered significant if the probability of their occurrence by chance was less than 5%.

# Bioassay of Responses to Combination of Vibratory and Chemical Signals

The above Y-track apparatus was modified so that vibrations and volatiles could be delivered independently during bioassay. Two transparent Plexiglas tubes (each 20 cm long, 3 cm i.d.) were held by clamps over the arms of the Y-track with their bottom ends contacting each other at the Y-junction. The top of each tube was closed with a silicone rubber stopper with a glass tube (4 mm i.d.) through the centre attached with Tygon tubing to the sidearm of a glass Buchner flask (250 ml) containing the odor stimuli. Each flask had a glass inlet tube (4 mm i.d.) to the base of the flask connected to a charcoal filter (10 cm x 2 cm; 10-18 mesh) and then to a diaphragm pump (Charles Austin DA7C). Airflow through each flask was 1.5-2 liter/min and the flasks were positioned behind the olfactometer so that their contents were not visible to insects during the bioassays. An exhaust fan was positioned 20 cm behind the Y-junction with a hose leading to the exhaust of the bioassay room.

Both airstreams contained volatiles from leaves (approx. 20) freshly cut from wheat seedlings. Volatiles from male bugs were collected by aeration of 10 male bugs for 24 hr and trapping on Porapak resin (9). Trapped volatiles were removed with dichloromethane (1.5 ml giving approx 1.25 ml recovered). An aliquot of this solution (100  $\mu$ l) was applied to filter paper (4.5 cm dia) and, after evaporation of the solvent this was placed in the flask connected to the test arm of the bioassay. This dose was equivalent to 19.2 bug hr and was the same as that which caused significant attraction of female bugs previously (3).

Natural and recorded vibratory signals were delivered as above except that for the natural signals two bugs were used on the end of the stick inside a translucent film canister. Some bugs called almost continuously, others remained silent, while most called sporadically. Signaling was monitored with a dynamic microphone with the membrane exposed and held in contact with the relevant choice arm skewer.

In each bioassay, a female was transferred onto the lower, vertical skewer and observed closely. At the Y-junction, an assessment was made as to whether the female touched both upper skewers with her tarsi before moving onto a choice arm or climbed straight up onto one without touching the other. Only the former were recorded as a response, even though this lead to approx 70% of the data being discarded. This distinction was seen as important when vibratory signals were involved, since a real choice could only be made by females that sampled both active and inactive choice arm skewers before climbing across the gap. This did not apply in the case of bioassays involving air-borne volatiles only. Where live males were used as the source of vibratory signals, it was also noted whether the males were in fact calling at the moment the female selected a choice arm. In this case, a valid choice was recorded only when a female touched both skewers when the males were actually calling, prior to climbing across the gap onto one choice arm or the other.

The active arm (carrying volatiles and/or vibratory signals) was alternated after five consecutive tests to avoid bias due to extraneous cues, at which point the skewers were replaced with new ones and chemical sources renewed. As far as possible, individual virgin females were used on just one occasion in the bioassays. Shortage of insects, however, meant that some females were used several times, although individuals were never used twice on the same day.

## Results

#### Song Types and Behavioral Observations

Isolated Е. integriceps males call spontaneously. However, they seem much more likely to do so if another insect is present on the same stem, and especially so if it walks. Tarsal-stem contacts result in perceptible "footsteps" seen as extraneous noise on song recording traces. Males also seem stimulated to call when a female is held close (2-3 cm) in forceps but not touching the stem, in which case male antennal movement always occurs. Careful observation of calling males suggests that the abdomen does not strike the substrate, but rather that physical transmission of vibrations occurs through the male's legs.

The male "calling song" consists of short chirps repeated at a regular intervals with a call rate of 4.6 chirps/sec (mean chirp duration =c.0.085 sec, inter-chirp interval =c.0.17 sec) (Figure 1). When examined at increased temporal resolution, most chirps are composed of 2 or 3 syllables (Figure 1). The dominant frequency is c.100-200 Hz. Under laboratory conditions, males have been observed to call more-or-less continuously for periods exceeding 2 min, but such sustained calling occurred only when another insect was present on the same stem. Short song bouts with longer pauses between are more typical of solitary males. After calling, a male will usually approach any second individual present, continuing to call as he comes close.



**Figure 1.** "Calling song" of male *E. integriceps;* lower at higher resolution (time scale (sec) on x-axis, signal amplitude on y-axis).

When approached, a second male typically produces a distinctive agonistic call or "rivalry song" (Figure 2). In this case chirps are repeated at a much faster rate, more than double that of the calling song, at 10.4/sec. The chirps are slightly shorter compared to the calling song, but the inter-chirp interval is considerably reduced (mean chirp duration =c.0.06 sec, inter-chirp interval =c.0.04sec). Rivalry songs are sometimes produced in alternation by both males, in which case the insects separate soon afterwards.

Despite the distinctive structure of rivalry song, advancing males frequently seem unable

to discriminate the sex of their conspecific and often evert their genitalia (see below), sometimes even attempting to mount a second male on coming into contact.



**Figure 2.** Calling song of approaching male, followed by rivalry song of second male.

Females occasionally call spontaneously, but they are much more likely to do so in response to a male calling song, or indeed when any other conspecific is nearby. Virgin females confined together often call in closerange interactions with other females, further suggesting that intersexual discrimination on the basis of vibratory signals is problematic. The female song structure does not differ from the male calling song in its chirp rate, although it is generally produced at lower amplitude and the vibratory movements generating the calls are not always evident. The extent of female calling during intersexual interactions varies considerably. Some matings were observed in which no female call was apparent. In other cases, however, a "malefemale alternation" occurred, with both individuals signaling as they moved together (Figures 3 and 4). During this mutual approach, the male calls are generally louder and longer and he moves faster.

Typically, the chirp rate increases as the male gets close to the female. At this stage receptive females may adopt a stilt-legged posture, raising up their entire body, and this appears to facilitate genital coupling. Distinctive close-range signals were not observed. Once mounted with genitalia engaged, vibratory signals subside (Figure 5).



**Figure 3.** Male-Female Alternation. Male calling song followed by lower amplitude female calling song, a short pause, then male calling song #2, female calling song #2, start of male calling song #3. The time scale is more compressed than in previous traces.



**Figure 4.** Detail of previous trace of male-female alternation: end of male call (chirps 1-4) followed by first female call (chirps 5-13) then male call (chirps 14-20).



**Figure 5.** Male calling song just before copulation, showing increased chirp rate and amplitude during the mount, as well as noise generated by foot movements.

Figure 5 shows a male calling song just before copulation, showing increased chirp rate and amplitude during the mount, as well as noise generated by foot movements. No female signals were detected. Male chirps subside as copulation ensues.

During the close-range interaction immediately before mating, the terminal segment of the male's abdomen is everted and then rotated through 180° before the male attempts to engage genitalia. A strong odor of vanilla was detected in many male-female interactions at this stage. The exposed intersegmental membrane just in front of the genitalic segment(s) appears shiny and moist, and this seems a likely dispersal surface for involved volatiles intersexual in communication. Both show sexes а considerable amount of antennal movement during close-range interactions.

In total, 33 matings were observed. All were preceded by male calling song, but female calls could not be reliably scored. Copulation lasted for several hours. In one sample, three out of seven mated females were observed to mate again on the second day after their original mating.

#### **Bioassay of Responses to Vibratory Signals**

When vibratory signals produced by live insects were used as source, males were significantly attracted to signals from both females and males (Table 1). Females were not attracted to signals from either sex.

When recorded signals were used as source, rather different results were obtained in that females were significantly attracted to signals from males. Males were not attracted to signals from males or females in contrast to the results with signals from live insects (Table 2).

# Bioassay of Responses to Combination of Vibratory and Chemical Signals

Responses of virgin female *E. integriceps* only were recorded. In the absence of odor, attraction of females to the vibratory signals from live males was confirmed, but not to the

recorded signals from males. When the odor collected from male bugs was added, no significant preference for either arm of the Y-shaped apparatus was observed for vibratory signals from live males or the recorded signal (Table 3).

 Table 1. Responses of *E. integriceps* to vibratory signals from live insects (2003).

Test insect	Insect Source	No. of indiv- iduals	<b>P</b> ( <sup>2</sup> )
Virgin male	Males None	22 10	0.03 (4.50)
Virgin female	Males None	19 20	0.87 (0.03)
Virgin male	Females None	27 10	0.005 (7.81)
Virgin female	Females None	19 19	

**Table 2.** Responses of *E. integriceps* to recorded vibratory signals (nt not tested; 2003).

Test insect	Source of recording	No. of indiv- iduals	<b>P</b> ( <sup>2</sup> )
Virgin male	Males None	18 15	0.60 (0.27)
Virgin female	Males None	25 12	0.03 (4.57)
Virgin male	Females None	10 12	0.67 (0.18)
Virgin female	Females None	nt nt	

## Discussion

Vibratory signals would seem to play a significant role in mate location and recognition in *E. integriceps*, with male calling song invariably preceding mating and ceasing at copulation. However, some females mate

Test insect	Source	No. of individuals	$P\left(\begin{array}{c}2\end{array}\right)$
Virgin female	Vibratory signal from live males	26	0.008 (7.11)
	None	10	
Virgin female	Recorded signal from males	5	0.29 (1.14)
	None	9	
Virgin female	Vibratory signal from live males + odor from males	4	0.37 (0.82)
	None	7	
Virgin female	Recorded signal from males + odor from males	10	0.82 (0.05)
	None	9	

**Table 3.** Responses of female *E. integriceps* responses to vibratory signals from males with and without odor collected from males (2004).

without producing female calling song, while others answer male calling song and yet others engage in a vigorous alternation of calls and advance to meet the incoming male. This could be associated with the level of receptivity of the females. On the basis of these laboratory observations it would seem that males are the more active mate-seeking sex.

Calling song structure is very similar in males and females and this may contribute to the difficulty both sexes appear to experience in sex recognition when conspecifics are encountered. Despite the production of a structurally distinct rivalry song only by males and the fact that the sexes are known to differ in their chemical profiles, males frequently attempt to mate with other males. Under field conditions it may be that competition for mates is intense and that it pays males to attempt to mount any conspecific in the first instance.

The novel bioassay methodology developed here to investigate *E. integriceps* response to vibratory signals shows promise, but results were inconsistent, even when care was taken to ensure that the stimulus insects were actually producing vibratory signals during the test and that the test insects did actually sample both arms of the Y-track. The use of repeated calling song playback from a looped recording ensured that test insects experienced calling song as they decided on which way to go, although the quality of recordings is likely to be inferior to natural song and signal delivery is unnaturally sustained.

When live insects were used as a stimulus, males were attracted to the calling song from both males and females. Attraction of males towards female vibratory signals is believed to be the commonest way in which pair-formation is initiated in pentatomid bugs (8). Although males were attracted to spontaneous intermittent vibratory signals from live insects, they did not respond to recorded continuous calling song, while the opposite was true with females. Addition of the olfactory stimulus of volatiles collected from male bugs appeared to remove the responses to vibratory signals, although probably too few insects were tested to consider this particularly conclusive.

Recent research suggests that output of volatiles by *N. viridula* males increases on exposure to female vibratory signals (11). It was suggested that the ability of the males to modulate pheromone emission in this way may reduce metabolic costs while mate-seeking. This ability would also be particularly advantageous if the timely production of high concentrations of pheromone inferred a

competitive advantage on males during closerange courtship (11). Although production of -homobisabolene by male *E. integriceps* ceases during copulation (9), males often generate a strong vanilla smell during courtship and it is possible that modulated pheromone production also occurs in this species.

Thus although it seems clear that vibratory signals are used in mate location in

*E. integriceps*, the exact role of these signals and how they interact with any volatile signals is still not clear. Further research should investigate whether the vibratory signals have an effect on production of volatiles by males or females, and it is also vital to clarify the function of the volatiles themselves in mating behavior.

# الملخص

غرين، إس. ف.، مارك س. أ. داونهام، د.آي. فارمان، ديفيد ر. هول ومصطفى البوحسيني. 2007. دور الإشارات الاهتزازية في سلوك تزاوج حشرة السونة. الصفحات 157–165.

تعد حشرة السونة (Eurygaster integriceps Puton) من أكثر الآفات الحشرية ضرراً للقمح والشعير في دول غرب آسيا. هدفت هذه الدراسة إلى توصيف الإشارات الاهتزازية التي يقوم بها ذكر وأنثى حشرة السونة وللتحري عن دور هذه الإشارات في التخاطب بين أفراد النوع الواحد، وذلك بوجود أو غياب إشارات الرائحة من الحشرات. تتألف "أغنية نداء" الذكر من أصوات قصيرة مكررة بفواصل منتظمة وبنسبة نداء 6.6 صوت/ثانية. بلغ متوسط فترة الصوت 0.085 ثانية والفاصل بين الصوت 7.10 ثانية. عندما يقترب ذكر ثاني ينتج نداء متوتراً مميزاً أو "أغنية تنافس" التي يتكرر فيها الأصوات بنسبة أسرع بلغت 10.4/ثانية. أحياناً نتادي الإناث بشكل عفوي، لكنهن غالباً ما يفعلن ذلك استجابة لأغنية نداء الذكر، أو في الواقع عند وجود أي فرد آخر من نفس النوع بقربهن. لا تختلف بنية أغنية الأنثى عن أغنية نداء الذكر في نسبة أصواتها، رغم أنها عموماً تنتج في نطاق أضيق، واهتزازات الحركة المنتجة للأصوات ليست واضحة دائماً. تزداد نسبة الصواتها، رغم أنها عموماً تنتج في نطاق وعندما نتشابك أعضاء المتنجة للأصوات ليست واضحة دائماً. تزداد نسبة الصوات عندما يصبح الذكر قريباً من الأثنى، وعندما استخدمت الإشارات الاهتزازية المنتجة من قبل الحشرات الاهتزاز تتوقف. تم تطوير نظام اختبار لتعقب الأثر بشكل حرف Y من يقس النوع بقربين. لا تختلف بنية أغنية الأنثى عن أغنية نداء الذكر في نسبة أصواتها، رغم أنها عدوجود أي فرد آخر وعندما استخدمت الإشارات الاهتزازية المنتجة من قبل الحشرات الحية كمصدر ، انجذبت ذكور الحشرة بشكل معنوي للإشارات وعندما استخدمت الإشارات الاهتزازية المنتجة من قبل الحشرات العية كمصدر ، انجذبت ذكور الحشرة بشكل معنوي للإشارات من كل من الذكور والإناث. في حين لم تنجذب الإناث للإشارات عند استخدام كلا الجنسين. عندما استخدمت الإشارات المسجلة وعندما استخدمت الإشارات الاهتزازية المنتجة من قبل الحشرات الحية كمصدر ، انجذبت ذكور الحشرة بشكل معنوي للإشارات من كل من الذكور والإناث. في حين لم تنجذبت الإناث للإشارات من الذكور ولم تنجذب الإشرانات من الذكور أو من كل من الذكور والإناث. في حين لم تنجذبت الإناث للإشارات من الذكور ولما متجذب الإشارات من الذكور أو من كل من الذكور والإناث. في حين لم تنجذب الإناث للإشارات من الذكور ولما متجذم الاشارات من الذكور أو الإناث. بوجود الرائحة المجموعة من الحشرات الاهترازاة من

Scutelleridae ، التخاطب بين أفراد النوع الواحد، Heteroptera ، Hemiptera ، Eurygaster integriceps، التخاطب بين أفراد النوع الواحد، Scutelleridae، التخاطب بين أفراد النوع الواحد،

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.
# Behavioral Responses of Sunn Pest to Volatiles from Conspecific Insects and Host Plants

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### Abstract

Athanassios, S., M.C.A. Downham, D.I. Farman, D.R. Hall, S.J. Phythian and M. El Bouhssini. 2007. Behavioral Responses of Sunn Pest to Volatiles from Conspecific Insects and Host Plants. Pages 167-180.

The Sunn pest, Eurygaster integriceps Puton (Hemiptera; Heteroptera; Scutelleridae), is the most damaging insect pest of wheat and barley in countries of West Asia with infestations spreading over 15 million ha in the affected area. The aim of these studies was to develop methods for bioassaying the responses of *E. integriceps* to volatile signals from live insects and host plants and also to collections of volatiles from the insects. In a slow-speed windtunnel with the bugs confined in longitudinal channels, there was some evidence for attraction of female bugs to male bugs on wheat seedlings and for repellency of females by female bugs on the seedlings. Bugs were found to move most easily along tubular devices 3-5 mm dia, and they are probably negatively geotropic and positively phototropic. A vertical Y-tube, linear-track olfactometer was developed which allowed comparison of responses of bugs to two stimuli in terms of number responding and time taken to respond. Females did not respond to volatiles from wheat or wheat with live conspecific males, but were attracted to wheat with live females. Males were not attracted to volatiles from wheat or wheat with live bugs of either sex. Females were significantly attracted to wheat with volatiles collected from male bugs on Porapak when an amount equivalent to 19.2 bug hr was used but not with one quarter this amount. More females went to the arm with odor from wheat and vanillin  $(1 \mu g)$  than to the arm with odor from wheat only, but the difference was not significant with the number of insects used. A preliminary experiment indicated that females were repelled by a blend of synthetic components of the metathoracic gland exudate. The significance of these results in terms of mate location in this species is discussed.

Keywords: Eurygaster integriceps, Hemiptera, Heteroptera, intraspecific communication, pheromone, Scutelleridae, semiochemicals, Sunn Pest.

## Introduction

The Sunn Pest, *Eurygaster integriceps* Puton (Hemiptera: Heteroptera: Scutelleridae), is the most damaging insect pests of wheat and barley in countries of West Asia with infestations spreading over 15 million ha in the affected area. Both nymphs and adults cause damage by feeding on leaves, stems and grains. Yield loss is commonly estimated at 20-30% in barley and 50-90% in wheat. Apart from the direct reduction in yield, the insects also inject chemicals that greatly reduce the baking quality of the dough (9). Currently chemical insecticides are the main approach to control of Sunn Pest, costing an estimated US\$

40 million annually in the countries concerned. There is a need to develop alternative approaches to control of this pest that minimise or avoid use of chemical insecticides and reduce the hazards to human health, water quality and the environment in general. In this context, a better understanding of the ways in which Sunn Pest locate mates and host plants could provide new methods for controlling the pest by interfering with these processes which are vital for survival of both the individual and the species.

Sunn Pest have a remarkable life cycle, closely adapted to the ecosystem they exploit (3). Adults overwinter in higher, mountainous regions in a state of diapause. As temperatures rise in spring they migrate to lower areas where cereals are grown and mating takes place. The eggs produce nymphs which pass through five instars to the adult bugs, and it is these nymph and adult stages which feed and are responsible for the damage. As temperatures rise further, the adults go back to the higher, cooler regions and diapause until the following spring, so that the pest only spends two to three months of the year on the cereal crop. Clearly Sunn Pest must have highly efficient methods of locating both mates and suitable host plants to survive and propagate. Exploitation of these processes could provide highly effective methods for monitoring and control of Sunn Pest and make major contributions to an IPM program.

There is good evidence that Heteropteran bugs communicate using sex pheromones, and aggregation pheromones alarm pheromones (1, 2, 15). Males of *E. integriceps* have been reported to produce a sex pheromone that attracts adult females (14, 22). Some of the chemicals involved have been identified as vanillin (20), ethyl acrylate (21) and homo--bisabolene (19), but the behavioral effects of these chemicals have not been fully evaluated and it is not even known whether all the chemicals making up the pheromone blend have been detected and identified. In the absence of this information, no work has been done to develop use of these materials in management of Sunn Pest, although a pheromone that attracts females would be particularly useful in both monitoring and control.

The aim of these studies was to develop methods for bioassaying the responses of *E. integriceps* to volatile signals from live insects and host plants and also to collections of volatiles from the insects.

## **Materials and Methods**

#### Insects

Male and female *E. integriceps* were sent separately as live adults from ICARDA to NRI by air freight. Those sent from December –

March were collected from overwintering sites and hence were unmated. Those sent April – May were collected from wheat fields and were probably mated. At NRI the male and female bugs were maintained in separate rooms on wheat seedlings with L:D 16:8 hr and temperature 25°C:20°C.

### Windtunnel Bioassay

Initial laboratory bioassays at NRI were done in a windtunnel (2 m long x 30 cm wide x 30 cm high ) with six length-wise plastic channels (2 cm wide) along the floor. Unless otherwise stated, windspeed was 20 cm/sec. Test sources of volatiles were two pots of wheat seedlings alone, with male or female E. integriceps adults (5 on each plant) or with a collection of volatiles from male *E. integriceps* (20 µl; approx 0.5 bug/hr) on filter paper. The test source was placed at the upwind end of the tunnel. Bugs were released at the centre of the tunnel, one per channel, and their behavior observed for 10 min. Movement was scored on a scale of 0 (no movement) to 5 (reaches source). Movement away from the source was scored on a scale of 0 to -5 (reaches end of tunnel away from source). Temperature and lighting were as in the rearing rooms, and experiments were carried out from 0900-1300 hr

### **Behavioral Observations**

Observations were made of the ability and willingness of bugs to move on various substrates. These included a Plexiglas Y-tube olfactometer (3 cm i.d.), filter paper strips (Whatman No 1), pipe cleaners (4 mm OD), plastic-covered wire (1 mm o.d.), wooden sticks (8 mm o.d.) and brass tubing (3 mm o.d. or 5 mm o.d.).

#### Y-tube, Linear-track Olfactometer

As a result of the above observations, subsequent bioassays used a vertical, linear-track, Y-tube approach. A Y-shaped brass stem was made from three brass welding rods (20 cm long, 3 mm o.d.) welded together with the Y-junction at  $90^{\circ}$  and carefully smoothed.

The Y-stem was held vertically by a clamp at the bottom end of its central branch. Two transparent Plexiglas tubes (each 20 cm long, 3 cm i.d.) were held by clamps over the arms of the Y-stem with their bottom ends contacting each other at the Y-junction. The top of each tube was closed with a silicone rubber stopper with a glass tube (4 mm i.d.) through the centre attached with Tygon tubing to the sidearm of a glass Buchner flask (250 ml) containing the odor stimulus. Each flask had a glass inlet tube (4 mm i.d.) to the base of the flask connected to a charcoal filter (10 cm x 2 cm; 10-18 mesh) and then to a diaphragm pump (DA7C; Charles Austin, Bylfleet, Surrey, UK). Airflow through each flask was 1.5-2 litre/min and the flasks were positioned behind the olfactometer so that their contents were not visible to insects during the bioassays.

Bioassays were carried out in the room used to house the female bugs between 0900-1300 hr, the main period of activity (3). The olfactometer system was placed on a table lit from above with a fluorescent light source. Additionally, an exhaust fan was positioned 20 cm behind the Y-junction with a hose leading to the exhaust of the bioassay room. Sunn Pests were collected from the wheat seedlings each day and only apparently healthy individuals with legs and antennae intact were used in bioassays. One adult was gently placed at the base of the central stem and generally proceeded to walk up the stem to the Yjunction. A response was recorded if the bug made a choice and passed beyond the midpoint of that stem (i.e. a distance of 10 cm). Both direction and time (seconds) to make the choice were recorded. The assay was terminated after 10 min and those that had not made a choice were classified as nonresponders.

Different Buchner flasks and Plexiglas tubes were used for each stimulus. The positions of the stimulus sources were reversed every five replicates and the Y-stem cleaned with ethanol at this time. Bugs were used only once and 20-40 were bioassayed in a day, using at least two pairs of treatments and/or sexes. Experiments were continued until 30-80 of one sex had been bioassayed.

Wheat used as odor sources was approximately 20 freshly cut seedling leaves (winter wheat; E.W. King, Essex, UK). Sources with live insects contained five of the stated sex. Collections of insect volatiles were made from 10 bugs for 24 hr on Porapak and eluted with dichloromethane (1.5 ml) (8). Four such collections were combined (5 ml) and 25  $\mu$ l (4.8 bug hr) or 100  $\mu$ l (19.2 bug hr) were used on a filter paper (Whatman No 52; 4.5 cm dia.). Analyses of the collections from male bugs by gas chromatography showed the male-specific -homobisabolene at 186 ng/µl and vanillin at 3.2 ng/µl, i.e. 4.7 µg and 18.6  $\mu$ g of the -homobisabolene and 0.08  $\mu$ g and 0.32  $\mu$ g of vanillin in the 25  $\mu$ l and 100  $\mu$ l samples respectively (8). Vanillin was tested as 100  $\mu$ l of a 10 ng/ $\mu$ l solution (1  $\mu$ g) on filter paper as above. Synthetic odor sources were renewed every five replicates. Compounds isolated from the metathoracic gland of female bugs, (E)-2-hexenal, (E)-4-oxo-2-hexenal, (E)-2-octen-1-ol, and (E)-2-octenyl acetate were made up at approximately 5 ng/µl each in dichloromethane. (E)-4-oxo-2-hexenal was prepared at NRI (11) and other compounds purchased from Sigma-Aldrich were (Gillingham, Dorset, UK).

Data were analysed by Chi-squared test of the numbers of insects responding to each source, and non-responders were excluded. The mean times for responses to the two sources were compared by a t-test. In addition, results were analysed by a one-sample Signtest in Genstat (Release 8; Lawes Agricultural Trust, Rothamsted, UK) which uses both the number of insects responding and the times and is thus an inherently more powerful single test. However, the probability values observed by these analyses were similar to those obtained by the Chi-squared test and only the former results are given here so as to be able between the to distinguish numbers responding and the time taken. Differences were considered significant if the probability of their occurrence by chance was less than 5%.

## Results

### Windtunnel Bioassay

In the first experiment, the responses of females to wheat seedlings with and without males were compared. Results (Figure 1) showed some upwind movement to the sources, but no difference between responses in the presence or absence of males.

In a second experiment, responses of females were compared to wheat seedlings alone, seedlings with males and seedlings with volatiles from males collected on Porapak. The results (Figure 2) showed little evidence of upwind movement, in contrast to the previous experiment, and no obvious differences between the three treatments.

In a third experiment, responses were compared of both males and females to wheat seedlings alone or with males or females. The results (Figure 3) showed some evidence for more upwind movement of females than males, but no obvious differences between responses to the treatments.

Finally the above experiment was repeated with a slower windspeed (2 cm/sec reduced from 20 cm/sec) and also with the pots of seedlings placed lower so that the middle of the plants was at the same level as the channels on the floor of the windtunnel containing the insects. Again there was more marked upwind movement of the females, and the response to the seedlings with males was greater than those to the other two treatments (Figure 4). Pairwise <sup>2</sup>-tests on the numbers of females scoring +5 indicated that significantly more females responded to wheat with males than to wheat with females (P=0.003), but differences between responses to wheat with males and wheat alone (P=0.24) or between wheat alone and wheat with females (P=0.052) were not significant. If numbers of bugs scoring 3-5 were considered, then the apparent repellent effect of the presence of females on the wheat was significant (P=0.03).

## **Behavioral Observations**

Unreplicated observations showed that E. integriceps adults had difficulty walking on the smooth surface of the Plexiglas tubes used to construct an olfactometer, particularly if it was orientated vertically or even at an angle of 45°. When the tubes were lined with filter paper strips, the Sunn Pest tended to walk along the edges, gripping either side of the strip. Subsequently various tubular structures were examined. The bugs had difficulty moving along fine, plastic-covered wire (approx 1 mm dia.) presumably because it was too narrow for them to grip properly. They also had difficulty on pipecleaners (approx 4 mm dia.) because the bristles made it difficult to grip the central wire. Wooden dowelling (approx 8 mm dia.) was probably a little too wide to grip properly, and the Sunn Pest seemed to move best on metal tubes or rods (3-5 mm dia.) even though they were quite smooth. The bugs were positively phototropic and/or negatively geotropic, generally moving rapidly up vertical rods or tubes. This behavior is presumably similar to that of bugs moving up wheat stems that are about 3-5 mm dia. in the field.

## Y-tube, Linear-track Olfactometer

Based on the above behavioral observations, a Y-tube olfactometer was constructed with a vertical linear track constructed from brass welding rods. Previous results showed that only males produce significant amounts of sex-specific volatiles, and it has been reported that females are attracted to the males (14, 22). Thus, in a first series of experiments, responses of virgin females were compared to volatiles from wheat with five live males and wheat alone, to wheat with live males and clean air and to wheat and clean air. There were no significant differences in numbers of insects responding in any of these pairs of treatments (Table 1). Females responded significantly faster to odor from wheat with males than to odor from wheat alone in the first series, but when odor from wheat with

males was compared with clean air, there was no difference in response time (Table 1).

Before pursuing further the anticipated attraction of females to host plants and/or volatiles from the males, it was thought prudent to check for the existence of any intraspecific response by testing all possible combinations of male and females as odor sources and responding insects. Thus responses of both virgin males and females to odor from males or females on wheat were measured. There were no significant responses of male or female bugs to odors from males or from wheat seedlings, and no response of males to odor from females (Table 2.) However, there was a significant response of females to odors from females. There were no significant differences in response times in any of the pairs of treatments, although in this series of experiments all insects seemed to respond faster than in the previous series and there were also fewer non-responders (Table 2).

**Table 1.** Comparison of responses of virgin female *E. integriceps* to volatiles from wheat with live males, wheat and an empty blank in a Y-tube linear-track olfactometer.

Test insect	n	Number of	Source individuals/Time	(SE) (secs)	<b>P</b> ( <sup>2</sup> )
Virgin female	80	Wheat with males 40 225.9 (25.1)	Wheat 28 345.4 (37.6)	Non-responders 12	0.15 (2.12) 0.008
Virgin female	80	Wheat with males 34 249.3 (24.9)	Blank 24 244.4 (30.9)	Non-responders 22	0.19 (1.72) 0.90
Virgin female	80	Wheat 31 222.6 (24.5)	Blank 28 262.9 (28.2)	Non-responders 21	0.70 (0.15) 0.28

**Table 2.** Comparison of responses of virgin male and female *E. integriceps* to volatiles from wheat with live insects, wheat and an empty blank in a Y-tube linear-track olfactometer.

			Odor Source		
Test insect	n	Number of in	dividuals/Time (	(SE) (secs)	<b>P</b> ( <sup>2</sup> )
		Wheat with males	Wheat	Non-responders	
Virgin male	30	11	15	4	0.43 (0.62)
		85.1 (20.1)	128.7 (27.0)		0.24
Virgin female	30	16	13	1	0.58 (0.31)
		77.6 (16.3)	61.1 (16.5)		0.49
		Wheat with females	Wheat	Non-responders	
Virgin male	30	15	10	5	0.32 (1.00)
-		64.1 (12.5)	103.3 (26.8)		0.15
Virgin female	30	23	7	0	0.003 (8.53)
		69.8 (13.5)	80.9 (24.6)		0.70
		Wheat	Blank	Non-responders	
Virgin male	30	13	12	5	0.84 (0.04)
C		86.4 (15.6)	66.1 (10.6)		0.30
Virgin female	30	11	16	4	0.34 (0.93)
-		71.5 (15.6)	54.8 (10.2)		0.36



Figure 1. Responses of female *E. integriceps* to wheat seedlings with or without males in a windtunnel.



Figure 2. Responses of female *E. integriceps* to wheat seedlings with or without males or volatiles collected from males in a windtunnel.



Figure 3. Responses of female (upper) and male (lower) *E. integriceps* to wheat seedlings with or without Sunn Pest in a windtunnel.



Figure 4. Responses of female (upper) and male (lower) *E. integriceps* to wheat seedlings with or without Sunn Pest in a windtunnel with a low wind speed.

A similar series of experiments was then run to test responses of virgin males or females to volatiles from the male or female odors collected on Porapak. The amount of collected volatiles used was nominally equivalent to 4.8 bug hr. No significant response of males or females to volatiles collected from either males or females was recorded, and there were no differences in time of response in any of the pairs of treatments (Table 3). More females were attracted to wheat with volatile collections from both males and females, although the differences were not significant. It is also worth noting that in both this and the previous experiment fewer males went to the wheat with volatiles from males than to wheat alone, suggesting some repellent effect of volatiles from males on males, but none of these differences were significant.

This series of experiments was then repeated using four times the amount of volatiles collected from males. Unfortunately by that time the supply of bugs collected from overwintering sites and assumed to be virgin had ended, and so experiments were run with field collected bugs which, although the same generation, were probably mated. Attraction of females to the lower quantity of volatiles was less than in the previous experiment, but significant attraction to the higher amount was observed (Table 4). Significant attraction was observed after 40 replications and, in view of the importance of this result, a further 40 replicates were run after which the significance was even greater. No significant effects were observed on the males and the repellent effect suggested by previous experiments was not observed, even at the higher dose. There were no differences in times of response among any of the pairs of treatments (Table 4).

Analysis of the filter papers (n = 2) after use in these experiments showed 6.8% and 5.3% of the original dose of the homobisabolene remaining for the 25  $\mu$ l and 100  $\mu$ l doses respectively.

In a final experiment during 2003, the response of field-collected females to vanillin was tested. More bugs went to the arm with the odor from vanillin and wheat seedlings than to the arm with odor from wheat alone, but this difference was not significant (Table 5). Similarly there was no significant difference between the mean times of response to the two arms.

**Table 3.** Comparison of responses of virgin male and female *E. integriceps* to odors from wheat with volatile collections from live insects and wheat in a Y-tube linear-track olfactometer.

			Odan Saunaa		
Test insect	n	Number of	<b>P</b> ( <sup>2</sup> )		
		Wheat with male volatiles <sup>a</sup>	Wheat	Non-responders	
Virgin male	40	11 106.2 (22.3)	16 129.3 (25.7)	13	0.34 (0.93) 0.52
Virgin female	40	19 88.9 (19.7)	11 120.8 (30.7)	10	0.14 (2.13) 0.37
		Wheat with female volatiles <sup>a</sup>	Wheat	Non-responders	
Virgin male	60	27 93.8 (13.1)	24 93.4 (17.5)	9	0.67 (0.18) 0.98
Virgin female	60	31 101.7 (16.3)	21 91.4 (18.2)	8	0.17 (1.92) 0.68

 $^{a}$  (25  $\mu l$  of volatile collections equivalent to 4.8 bug hr).

			Odor Source		
Test insect	n	Number of i	P(2)		
		Wheat with male	Wheat	Non-responders	
		volatiles (25 µl)			
Mated male	40	16	15	9	0.86 (0.03)
		97.9 (17.6)	81.1 (21.0)		0.54
Mated female	40	19	16	5	0.61 (0.26)
		62.3 (10.2)	85.2 (17.8)		0.26
		Wheat with male volatiles (100 µl)	Wheat	Non-responders	
Mated male	40	15	13	12	0.71 (0.14)
		48.9 (18.6)	62.1 (13.2)		0.98
Mated female	40	23	11	6	0.04 (4.24)
		52.0 (11.5)	36.6 (5.7)		0.37
Mated female	80	45	23	12	0.008 (7.12)
		53.6 (7.2)	54.4 (9.1)		0.95

**Table 4.** Comparison of responses of field-collected male and female *E. integriceps* to odors from wheat with different amounts of volatile collections from male insects and wheat in a Y-tube linear-track olfactometer.

**Table 5.** Comparison of responses of field-collected female *E. integriceps* to odors from wheat with vanillin  $(1 \mu g)$  and wheat in a Y-tube linear-track olfactometer.

			Odor Source		
Test insect	n	Number of in	ndividuals/Time	(SE) (secs)	<b>P</b> ( <sup>2</sup> )
		Wheat and Vanillin	Wheat	Non-responders	
Mated female	40	20	13	7	0.22 (1.48)
		74.4 (11.1)	83.9 (14.1)		0.59

During 2004, the response of females to volatiles collected from males was assessed using females collected from overwintering sites and assumed to be virgin. One set of replicates was carried out exactly as previously with the brass rod linear track. A second set utilised a linear track made up of three wooden sticks (3 mm dia). No significant attraction of the females to volatiles collected from the males was observed and more Sunn Pest went to the arm with volatiles from wheat seedlings alone than to the arm with volatiles from wheat seedlings and the volatile collection (Table 6).

A preliminary experiment was also carried out to examine the effect of compounds isolated from the metathoracic gland (MTG) of female *E. integriceps*. In these experiments, the wooden linear track was used and the distal ends of the sticks forming the arms of the Y were simply dipped into the test solution. Females collected from overwintering sites and assumed to be virgin and those collected from fields later in the season and presumed to be mated were tested. With both groups, significantly fewer went to the arm with the odor of the MTG components than to the arm without odor. Mated males were unaffected. In this experiment, neither virgin nor mated females were attracted to odor from collections of volatiles from males (Table 7).

## Discussion

Previous bioassay work on Sunn Pest by Litvenkov (14) and Vrkoc *et al.* (21) involved holding a filter paper impregnated with the test material 10 mm in front of the test insect. Scoring was 0 for no response, 1 for antennal fencing and walking towards source, 2 for precopulatory postures and elevated abdomen and 3 for rhythmic motions of abdomen and hind end directed towards odor source. The objective of the work described here was to devise a laboratory bioassay that was more closely related to field conditions and that made possible more detailed study of the behavioral responses of Sunn Pest to potential semiochemicals.

Various laboratory bioassay systems have been used for other Heteropteran bugs. For *Nezara viridula* L., Pavis and Malosse (18) used a glass tube olfactometer divided into ten sections. Borges *et al.* (4) used olfactometers constructed from plastic sandwich boxes connected by glass tubes, and Brezot *et al.* (5, 6) used a windtunnel 2 m long. Leal *et al.* (13) used a windtunnel bioassay with *Piezodorus hybneri* (Gmelin), and James *et al.* (12) used a standard Plexiglas Y-tube olfactometer with *Biprorulus bibax* Breddin. In recent work on other Pentatomid bugs, a vertical Plexiglas Ytube olfactometer has been used for *Chlorochroa sayi* (Stal) (10) and *Acrosternum hilare* (Say) (16).

During our studies, two bioassay systems were developed that seemed to give reasonable approaches for assaying responses of E. integriceps to odors from conspecific insects and host plants. The first was based on a windtunnel. It was found necessary to provide tracks along the floor of the tunnel to confine the movement of the bugs to up or down the tunnel. Furthermore, a relatively slow windspeed seemed to give more reproducible results and it is probably important to ensure that the odor source is at the same level as the bugs. Although six bugs could be tested in one run with this system, only one odor source could be tested. After 10 min rather high proportions of the bugs remained in the starting zone and statistical interpretation of the results was not always clear.

**Table 6.** Comparison of responses of virgin female *E. integriceps* to odors from wheat plus volatile collections from male insects and wheat in a Y-tube linear-track olfactometer.

		Odo	or Source		
Test insect	n	Number	of individua	als	P(2)
		Wheat with male	Wheat	Non-	
		volatiles (100 µl)		responders	
Virgin female (brass track)	92	41	51		0.30 (1.09)
Virgin female (wooden track)	66	33	36		0.72 (0.13)

**Table 7.** Responses of *E. integriceps* to odors from components of the metathoracic gland (MTG) and to odors from volatile collections from male insects in a Y-tube linear-track olfactometer.

Test insect	n	Nur	$P(^{2})$		
		MTG solution	Blank	Non-responders	
Virgin female	20	4	16	1	0.007 (7.20)
Mated female	10	1	9		0.01 (6.41)
Mated male	15	6	7	2	0.78 (0.08)
		Male volatiles	Blank	Non-responders	
Virgin female	10	5	5		
Mated female	10	5	5		

The second approach used a vertical linear track Y-tube olfactometer. This was behavioral observations based on of E. integriceps which showed that, although they were probably negatively geotropic and positively phototropic, they were unable to move along a vertical Plexiglas tube, as used by Ho et al. (10) and McBrien et al. (16) for other species. Sunn Pest moved most rapidly along tubular devices 3-5 mm dia. which they could grasp with their legs. This system allowed two odor sources to be compared directly and gave good levels of response (> 80%) within 10 min. Results could be assessed in terms of comparisons of numbers of individuals responding and the times to respond using simple Chi-squared and t-tests or a Signtest making use of both parameters.

Many experiments were run in both systems, but few consistent and statistically significant results were obtained. In the most convincing series of experiments in the windtunnel, more females were attracted to an odor source of wheat seedlings with males than to the wheat seedlings alone, although the difference was not significant. Significantly fewer females were attracted to the wheat with females than to wheat alone.

With the vertical linear track Y-tube olfactometer, virgin females collected from overwintering sites did not respond to volatiles from wheat or wheat with live conspecific males, but were attracted to wheat with live females. Males were not attracted to volatiles from wheat or wheat with live bugs of either sex. Virgin or mated females were not attracted to wheat with volatiles collected from male bugs on Porapak when an amount equivalent to 4.8 bug hr was used, but fieldcollected females which were presumed to be mated were significantly attracted to wheat with four times this amount of volatiles collected from males during 2003. However, this result could not be repeated with virgin females during the following season in 2004. More females went to the arm with odor from wheat and vanillin  $(1 \ \mu g)$  than to the arm with odor from wheat only, but the difference was not significant with the number of insects used. A preliminary experiment indicated that females were repelled by a blend of synthetic components of the metathoracic gland exudate.

Good levels of response (> 80%) were obtained in these latter experiments, and parallel observations showed that the experiments were performed with insects that were capable of mating and at the times of day when this occurred. Although these bioassays are by no means exhaustive, the absence of consistently significant responses might suggest that semiochemicals alone do not elicit strong anemotactic behavior in either sex of *E. integriceps*, at least when walking. We have shown that both males and females produce vibratory signals transmitted through the hostplant stem (7) and made preliminary investigations of the behavioral effects of these signals alone and in the presence of potential semiochemicals. Miklas et al. (17) suggested that male *N. viridula* modulate pheromone emission in response to vibratory signals from conspecifics, although the effects observed were relatively small. Nevertheless, the vertical linear-track Y-tube olfactometer would seem to have value for assaying behavioral responses of walking E. integriceps to semiochemicals. Male bugs produce large amounts of a characteristic homo- -bisabolene that seems to be associated with mating (8), and male-produced pheromones that attract females have been demonstrated in several related Pentatomid Heteropterans (15). The results suggest that the above experiments may have been carried out with levels of semiochemical stimuli that were too low, and future work should possibly start with investigating responses to higher doses of both natural and synthetic potential semiochemicals.

# الملخص

أثاناسيوس، إس.، م. س. أ. داونهام، د. آي. فارمان، د. ر. هول، إس. ج. فيثيان ومصطفى البوحسيني. 2007. الاستجابات السلوكية لحشرة السونة للمواد المتطايرة من حشرات نفس النوع والنباتات العائلة. الصفحات 167–180.

تعد حشرة السونة (Eurygaster integriceps Puton) من أكثر الآفات الحشرية ضرراً للقمح والشعير في دول غرب أسيا مع انتشار الإصابات على أكثر من 15 مليون هكتار في المنطقة المصابة. هدفت هذه الدراسات إلى تطوير طرائق لاختبار استجابات حشرة السونة لإشارات المواد المتطابرة من الحشرات الحبة والنباتات العائلة وأبضاً لمواد متطابرة مجموعة من الحشرات. في أنبوب الهواء بطيء السرعة مع الحشرات محجوزة في قنوات طولية، كان هناك بعض الأدلة على انجذاب الحشرات الإناث إلى الذكور على بادرات القمح وعلى طرد الإناث بواسطة حشرات الإناث على البادرات. وجد أن الحشرات تتحرك بسهولة أكثر على طول الأدوات الأنبوبية بقطر 3–5 مم، وقد يكون لها انتحاء سلبي للجاذبية الأرضية وانتحاء إيجابي للضوء. تم تطوير أنبوب عمودى بشكل حرف Y كجهاز طولى لتعقب أثر الروائح ليسمح بمقارنة استجابات الحشرات لنوعين من المنبهات من حيث عدد الاستجابات والوقت المستغرق للاستجابة. لم تستجب الإناث للمواد المتطايرة من القمح أو القمح مع ذكور حية من نفس النوع، ولكنها انجذبت إلى قمح مع إناث حية. لم تتجذب الذكور للمواد المتطايرة من القمح أو القمح مع حشرات الحية من أي من الجنسين. انجذبت الإناث بشكل معنوي للقمح مع مواد متطايرة مجموعة من الحشرات الذكور على مادة Porapak (مادة صلبة تستخدم لامتصاص وإصطباد الغازات والمواد المتطايرة) عندما استخدمت كمية تعادل 19.2 حشرة ساعة، ولكن ليس مع القمح الذراع إلى الإناث اتجهت الكمية. والفانيلين من الرائحة مع هذه ربع (1 ميكروغرام) أكثر منها إلى الذراع مع الرائحة من القمح فقط، لكن الفرق لم يكن معنويا مع عدد الحشرات المستخدم. أشارت تجربة أولية أن الإناث طردت بواسطة مزيج من المركبات المصنعة من مفرزات غدد الصدر الخلفي. يناقش البحث أهمية هذه النتائج من حيث موقع التزاوج في هذا النوع.

كلمات مفتاحية: Heteroptera ، Hemiptera ، Eurygaster integriceps، التخاطب بين أفراد النوع الواحد، فرمون، مواد ناقلة للرسائل الكيميائية، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# The Impact of Sunn Pest Density on Grain and Flour Quality

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## Abstract

Jaby El-Haramein, F., M. El Bouhssini, M. Amir-Maafi, R. Canhilal and H. Kutuk. 2007. The Impact of Sunn Pest Density on Grain and Flour Quality. Pages 181-186.

Wheat grain and flour quality deterioration caused by Sunn Pest (*Eurygaster integriceps* Puton) are major concerns for wheat growers and processors. A total of 44 naturally infested wheat fields in Iran (2003) and Turkey (2002, 2003) were sampled just before harvest. Collected kernels were assessed for thousand kernel weight, percent kernel damage, total protein content, and gluten quality using a modified SDS-sedimentation method and farinograph tests. Sunn Pest densities ranged from 1.0 to 57.1 nymphs/m<sup>2</sup>, which yielded 0.3-49% damaged kernels, respectively. In Iran and Turkey, wheat fields with densities less than 7 and 10 nymph/m<sup>2</sup>, respectively, caused no significant damage on flour gluten strength as measured by the sedimentation and farinograph testing. These results need to be taken into consideration in determining the economic injury level for Sunn Pest in those countries.

Keywords: Gluten strength, grain quality, Sunn Pest, wheat.

## Introduction

The Sunn Pest (*Eurygaster integriceps*, Puton) is a threat to wheat yield and grain quality. The last two nymphal stages are the most vigorous in feeding on the kernels (2). Currently, chemical insecticides are the main method of Sunn Pest management, with approximately 1.5 million hectares sprayed annually in Iran and Turkey. The decision to spray is based on the economic threshold; a level of insect infestation where the cost of treatment is justified to avoid economic crop loss. A major parameter in this model is the value of the wheat grain itself and its quality. Sunn Pest affects wheat quality by injecting saliva containing proteolytic and amylolytic enzymes into the grain during feeding. The proteolytic enzymes persist in the kernel and can cause extensive breakdown of gluten. If the infestation is severe, entire lots of grain may be unsuitable for bread making. Various thresholds for Sunn Pest are used in each country, and they are influenced by site characteristics, temperature, natural enemies, etc. As a result of this variation, the estimation of any parameter involved in the economic threshold must be robust to the abovementioned influences. The present study was conducted to determine the effect of natural Sunn Pest infestation levels on grain quality, and to identify key methodologies that can be correlated to Sunn Pest damage and used for assessment of its impact on wheat/flour quality.

# **Materials and Methods**

#### Wheat Fields and Sunn Pest Density

In Iran, 26 wheat fields were selected in 2003 within Azarbijan Gharbi, Kermenshah, Markazi, Esfahan and Lorestan provinces. In Turkey, 18 wheat fields were selected in 2002 and 2003 in Gaziantep and Kilis provinces. All fields were one ha in size and no chemical insecticide applications were made during the study. Sunn Pest densities were estimated at the initial observation of Sunn Pest adults, continuing until harvest. All fields in Iran and Turkey were divided into five equally-sized subunits. Within each subunit, five  $0.25m^2$  frames were thrown and the number of Sunn Pest adults and nymphs within the frame were counted, averaged, and recorded as number per m<sup>2</sup>. The nymphal density one week prior to wheat maturity was considered as the Sunn Pest density. At the harvest, a volume of 0.5-1 kg was taken from each unit and sent to ICARDA for grain and flour quality analysis.

## **Grain Quality Analysis**

Three analyses were conducted, the weight of 1000 kernels (TKW), the percentage of kernels with signs of Sunn Pest damage (%DK) and total protein content.

- Thousand kernel weight Collected grains were transported to ICARDA and held at 22±2°C until analysis. One hundred kernels were counted from each batch in three replicates, weighed and the mean of 1000 kernel weight calculated.
- Percent damaged kernel When Sunn Pest feeds on wheat it leaves an opaque patch on the kernel with a small dark penetration site. The average count of kernels with this characteristic from among three replicate sets of 100 kernels was taken.
- Protein content -Wheat protein content was determined using a NIRSystems model 5000 scanning monochromator. Ten percent of the samples were also analyzed by the Kjeldahl method (4) to check NIR accuracy.

#### **Flour Quality Analysis**

**Flour milling -** All milling was conducted at 23°C, 60% R.H. Wheat samples were cleaned and tempered overnight to optimum moisture (5). Tempered wheat was milled using a Buhler laboratory mill model MLU-202, break roll gaps adjusted to  $B_2 = 3/1000$  inch,  $B_3 = 2/1000$  inch,  $C_1 = 3/1000$  inch and  $C_3 = 2/1000$  inch. Medium hard soft-wheat clothing was used. Buhler bran finisher MLU-302 was used to extract "bran flour" which was combined with all six flour streams.

- SDS sedimentation test Based on Cressey and McStay (1), the Modified Sodium Dodecyl Sulphate (SDS) Sedimentation Test was used to evaluate Sunn Pest damage in wheat. Five g of sieved flour (using UDY cyclone mill and 100 mesh sieve), was transferred to a 100ml stoppered measuring cylinder. Fifty ml of indicator solution (0.100g bromophenol blue per 10 liters demineralized water) was added. Samples were incubated for 30 min at 37 °C with shaking 15 times every 5 min. After 30 min, 50 ml of 3% SDS solution was added, inverted 4 times every 2min, after 3 trials the samples were incubated for 10 min at 37 °C then sediment volume was recorded.
- Farinograph test The AACC Method No. 54-21 (3) was followed using a 50 g mixing bowl.

## **Results and Discussion**

Table 1 summarizes the grain and flour quality of Iranian bread wheat samples. The occurrence of 6.3% damaged kernels among the wheat samples resulted in poor gluten strength. Kernel size was not noticeably affected by high Sunn Pest densities under all conditions, but the gluten strength was degraded significantly at 7.4 nymphs/m<sup>2</sup> (p<0.01). We found a significant correlation between Sunn Pest density and both damaged kernels and sediment volume (r=0.59 and 0.506, respectively). Protein content was not correlated with Sunn Pest density. This was expected since the main damage of Sunn Pest is related to protein quality not quantity.

Inter-correlations between grain/flour quality tests indicated that sedimentation volume is highly correlated with Farinograph development time, stability time and mixing tolerance (r = 0.893, 0.768 and 0.933, respectively).

Area	Variety	Sunn Pest density (nymphs/ m <sup>2</sup> )	DK* (%)	TKW (g)	Protein (%)	Sediment (ml)	FAB (%)	FDT (min)	FST (min)	FMT (BU)
Rainfed-Kermamshah	Unknown	3.5	1.3	42.6	14.0	91	62.5	3.0	4.3	80
Rainfed-Kermamshah	Unknown	3.6	2.3	30.7	12.9	89	58.0	5.0	11.2	40
Rainfed-Lorestan	Sardary	4.0	6.3	37.8	11.8	39	57.0	1.8	2.0	160
Rainfed-Kermamshah	Unknown	4.5	2.0	27.5	13.2	90	58.0	5.0	13.1	20
Irrigated-Esfahan	Mahdavi	5.0	4.7	35.6	11.7	33	65.0	2.2	1.9	175
Irrigated-Esfahan	Roshan	5.3	0.7	38.4	11.2	55	63.0	2.5	2.3	170
Rainfed-Kermamshah	Unknown	5.4	9.3	40.9	13.8	89	62.0	3.8	5.8	60
Rainfed-Kermamshah	Unknown	5.8	6.3	38.1	13.7	86	61.5	3.9	6.2	70
Rainfed-Kermamshah	Unknown	7.2	8.3	44.2	12.8	88	61.5	3.3	4.8	80
Rainfed-Lorestan	Sardary	7.4	29.3	40.1	8.9	7	55.0	1.3	1.5	235
Rainfed-Lorestan	Sardary	7.7	27.0	39.8	11.1	10	57.5	1.3	1.2	260
Rainfed-Markazi	M.V.17	8.0	25.3	33.5	10.9	9	59.5	1.7	1.5	270
Rainfed-Lorestan	Sardary	8.2	12.0	39.0	11.6	38	58.0	2.0	1.5	180
Rainfed-Azar/Gharbi	Sardary	8.3	3.3	26.5	13.5	74	56.0	2.8	3.5	115
Rainfed-Lorestan	Sardary	8.6	9.3	40.7	9.3	12	55.5	1.2	1.6	220
Rainfed-Lorestan	Jin-Alta	9.3	36.0	38.3	11.2	13	56.5	1.4	1.4	235
Rainfed-Markazi	Back Cross	9.4	5.3	39.4	11.5	53	59.5	2.5	2.4	120
Rainfed-Markazi	M.V.17	9.9	25.3	33.9	11.5	9	58.5	1.6	1.4	260
Irrigated-Esfahan	Ghods	10.7	9.3	38.0	12.8	15	64.0	2.0	1.7	240
Rainfed-Markazi	Back Cross	11.0	2.0	39.0	10.7	54	59.0	2.2	2.8	120
Irrigated-Esfahan	Roshan	11.1	3.7	37.3	12.1	43	60.5	2.1	2.0	110
Rainfed-Azar/Gharbi	Sardary	13.1	7.0	32.6	10.8	44	54.0	2.0	3.4	110
Rainfed-Markazi	Back Cross	18.2	39.3	36.0	10.4	7	56.0	1.5	1.4	280
Rainfed-Markazi	Back Cross	26.6	37.3	35.4	10.5	7	55.0	1.2	1.3	310
Irrigated-Esfahan	Roshan	26.9	49.0	41.2	13.7	11	64.0	2.0	1.1	310
Irrigated-Esfahan	Roshan	44.0	26.3	50.9	9.2	8	59.0	1.5	1.6	200

**Table 1.** Grain and flour quality of Iranian wheat samples in the 2003 harvest season.

\* DK=damaged kernel, TKW=thousand kernel weight, Protein as is moisture basis, FAB=Farinograph water absorption, FDT=Farinograph development time, FST=Farinograph stability time, FMT=Farinograph mixing tolerance.

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The grain/flour quality of the Turkish wheat samples is presented in Table 2. No effect of Sunn Pest density on gluten strength up to 10.4 nymphs/m<sup>2</sup> was detected among all fields. This supports the Turkish decision for initiating Sunn Pest control at the economic threshold level of 10 nymphs/m<sup>2</sup>. Of the durum lines tested, most yielded low sedimentation values and were of poor quality. Only one sample was acceptable, sampled from a field with 5.3 nymphs/m<sup>2</sup>. As the

remaining durum wheat samples had densities of 12.6 nymphs/m<sup>2</sup> or higher, we cannot consider this density the true limit for Sunn Pest impact on flour quality. The anticipated nymphal density that is of economic importance to wheat farmers and processors is between 5.3 and 12.6 nymphs/m<sup>2</sup>. The presence of 4.5% damaged kernels was found to weaken gluten strength.

<b>Table 2.</b> Grain and flour quality of Turkisn wheat samples in 2002 and 2003 harvest seasons.
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		Sunn Pest								
Site	Variety	(nymphs/ m2)	DK* (%)	TKW (g)	Protein (%)	Sediment (ml)	FAB (%)	FDT (min)	FST (min)	FMT (BU)
Maras-Tigem (03)	Golye-BW	1.0	0.8	25.9	12.2	89	60.0	1.5	4.9	80
K. Maras-Tigem (02)	Golye BW	1.9	0.70	26.9	11.2	77	57.5	1.8	4.2	85
Islahiye-Hanagazi (02)	Golye BW	2.9	0.30	34.4	9.6	64	57.5	1.4	3.1	110
Islahiye-zincirli (02)	Golye-BW	4.0	1.40	29.6	11.8	82	57.5	2.0	6.0	65
Nurdagi-Ciftlik (03)	Basri bey-BW	6.2	1.3	35.9	11.3	69	59.0	2.5	4.8	120
Islahiye-Kozdere (03)	Golye-BW	8.5	2.6	26.6	12.2	63	58.5	2.0	5.1	80
Araban-Baspinar (02)	Golye-BW	9.1	3.30	31.6	11.9	57	58.0	1.5	5.0	100
Nurdagi-Ciftlik (02)	Golye-BW	10.4	2.90	35.0	10.5	52	58.0	1.2	3.5	115
Nurdagi-Sakcagozu (02)	Ozdemir bey-BW	34.0	7.40	33.5	10.5	18	58.5	3.0	2.1	260
Gaziantep-Degirmen (03)	Unknown BW	57.1	23.0	29.3	12.8	7	57.5	1.8	0.8	360
Zihni Kutlar-Ciftligi (03)	Ege 88-Durum	5.3	1.0	33.2	12.0	28	65.5	1.6	1.5	145
Kilis-Yavuzlu (02)	Ege 88-Durum	12.6	4.50	42.7	11.3	13	64.0	1.3	1.5	230
Kilis-Yavuzlu (03)	Ege 88-Durum	13.0	2.2	33.0	14.6	17	67.0	2.1	1.6	220
Havaalani-Fistikcilik (03)	Akcakale 2000-	14.3	5.2	41.8	11.0	10	63.5	1.8	1.3	260
Sanko-Holding (02)	Ege 88-Durum	14.8	3.25	44.5	11.5	22	63.5	1.6	1.3	220
Yavuzeli (03)	Zenit-Durum	27.2	5.4	45.5	10.4	9	65.5	1.4	1.3	250
Kutlar-Ciftligi (02)	Ege 88-Durum	29.6	9.90	51.8	8.2	10	54.5	0.8	0.8	260
Havaalani-Fistikcilik (02)	Akcakale 2000- Durum	38.7	19.60	43.9	9.2	9	62.0	1.0	1.2	290

\*DK=damaged kernel, TKW=thousand kernel weight, Protein as is moisture basis, FAB=Farinograph water absorption, FDT=Farinograph development time, FST=Farinograph stability time, FMT=Farinograph mixing tolerance.

Significant correlations (p<0.01) were found between Sunn Pest density and damaged kernels, sediment volume, Farinograph development time, stability time and mixing tolerance (r = 0.965, -0.937, -0.840 and 0.97, respectively). In Turkish bread wheat samples, there is a high correlation between sediment volume and both Farinograph stability time and mixing tolerance (r = 0.873 and -0.933 respectively).

The mean nymphal density in Iran and Turkey were similar (10.9% in Iran and 13.5% in Turkey) but, the percent damaged kernels were different (14.9% in Iran and 4.4% in Turkey). This could have been a varietal or environmental effect.

# Conclusions

Results support the reasoning that Sunn Pest densities under 10 nymphs/m<sup>2</sup> cause no significant damage to Turkish wheat quality, while in Iran 7.2 nymphs/m<sup>2</sup> results in gluten

degradation. Correlation to the percent kernel damage indicates that if this level is 4% or higher, further quality tests should be conducted to verify gluten strength. In all samples, the modified SDS-sedimentation and Farinograph measurements were directly correlated to Sunn Pest density and percent kernel damage, and were equally robust.

The assessment of percent kernel damage provides a rapid assessment of grain quality, which, together with the modified SDSsedimentation methodology, gives an overall estimate of gluten damage. Further field monitoring of Sunn Pest damage to determine the robustness of these tests among different varieties, areas, seasons and growing conditions is required. As grain quality has been shown to be correlated to Sunn Pest density, a post-harvest assessment of the grain quality could serve as feedback into the economic threshold model, allowing for periodic revision as required.

# الملخص

جابي الحرمين، فؤاد، مصطفى البوحسيني، مسعود أمير مافي، رمضان جانهلال وخليل كتك. 2007. تأثير كثافة حشرة السونة في الحبوب ونوعية الدقيق. الصفحات 181–186.

إن الضرر الذي تسببه حشرة السونة (Eurygaster integriceps Puton) لحبوب القمح ونوعية الدقيق هما مصدر القلق الرئيس لزراع ومصنعي القمح. أخذت عينات من 44 حقل قمح مصاب طبيعياً في إيران (خلال عام 2003) وتركيا (خلال عامي 2002 و 2003) قبل الحصاد مباشرة. اختبرت الحبوب المجموعة لتحديد وزن الألف حبة، نسبة ضرر الحبوب، محتوى البروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS المعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS المعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS والمعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS والمعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS والمعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS والمعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ماروتين الكلي ونوعية الغلوتين باستخدام طريقة ترسيب SDS والمعدلة واختبار الفارينوغراف. تراوحت كثافة حشرة السونة بين ورية/م<sup>2</sup> في حقول القمح في إيران وتركيا، على التوالي، ضرراً معنوياً لقوة غلوتين الدقيق عند قياسه باختباري الترسيب والفارينوغراف. يجب أخذ هذه النتائج بعين الاعتبار عند تحديد مستوى الضرر الاقتصادي لحشرة السونة في هاتين الدولتين. كلمات مقتاحية: قوة غلوتين، نوعية حبوب، حشرة السونة، قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Damage Loss Assessment of Sunn Pest on Wheat in Turkey

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## Abstract

Canhilal, R., H. Kutuk, M. Islamoglu, A.D. Kanat and A. Gul. 2007. Damage Loss Assessment of Sunn Pest on Wheat in Turkey. Pages 187-190.

Studies were conducted in cages to determine the damage of adults and nymphs of Sunn Pest on wheat in two different locations in 2002 and 2003. Adult density in the cages was 1, 2, 3, 5 and 10 adults per  $m^2$ . Nymphal density was 5, 10, 15 and 20 nymphs per  $m^2$ . Spike damage caused by adults depended upon the number of Sunn Pest released into cages and was 0.072-0.878% and 0.000-0.256% in 2002 and 2003, respectively. Total yield was 956-1150 g and 309-1165 g in two  $m^2$  in 2002 and 2003, respectively. While differences between spike damage rates were statistically significant in 2002, it wasn't statistically significant in 2003. In both years, there was no difference among total yields harvested from cages. Kernel damage caused by nymphs depended on the number of nymphs released into cages and varied from 0.287 to 0.625% in 2003.

**Keywords:** Cages, economic threshold, spike and kernel damage, Sunn Pest.

## Introduction

In Turkey, wheat and barley are the most important crops and are grown on approximately 14 million ha. Sunn Pest is the major pest of wheat and barley. One million nine hundred thousand ha of wheat and barley were sprayed against Sunn Pest by aerial and ground equipment in 2003 (1). Both nymphs and adults cause damage by feeding on leaves, stems and grains. Yield loss caused is estimated at 50-90% in wheat and 20-30% in barley. In addition to direct reduction in yield, the insect also injects chemicals while feeding and this greatly reduces the baking quality of grain. If as little as 2-3% of the grains in a given lot have been fed upon, the entire lot may not be used for baking (6).

Sunn Pest control has been managed by the government of Turkey since 1927. After overwintering under the litter in the highlands, Sunn Pest migrates to cereal fields in spring. It is stated that migration continues 7-10 d under normal weather conditions (1). After migration, technical consultants start surveys in the fields. Adult Sunn Pests are counted in  $0.25 \text{ m}^2$  frames, and their density are determined. The fields where the density exceeds 0.8 adults/m<sup>2</sup> were monitored for egg parasitism when 20-30% of the Sunn Pest eggs are in the anchor sign stage. If the egg parasitism rate is high enough, spraying is not conducted. Nymphal density is determined the same as overwintered adult density. If nymphal density reaches 10 nymphs per m<sup>2</sup>, fields are sprayed (1).

One of the key factors affecting the success of an IPM program is economic threshold (ET). The economic threshold used for Sunn Pest control was established about 40 years ago in Turkey. There is a need to recalculate the ET because of changes in climatic conditions, wheat varieties used, agronomical practices and crop diversity etc.

The aim of this study was to determine the yield loss caused by Sunn Pest adults and nymphs on wheat in cages to provide data to be used in recalculating the ET.

# **Materials and Methods**

Studies were conducted in cages (2x1x1.5m) covered with cheesecloth. The number of wheat plants in each cage was approximately 500-600; 1100-1200 plants/cage in Oguzeli, Nurdagi, respectively. Two separate experiments were designed to determine the yield loss due to adult and nymphal feeding on kernels.

# Yield Loss Caused by Overwintered Adults (Spike Damage)

Studies were started at the end of migration. A completely randomized plot design with 6 treatments (1, 2, 3, 5, 10 adults/m<sup>2</sup> and control) and 4 replications was used. The adults were collected from wheat fields at the beginning of migration by using a sweep-net and held in a small cage until they were transferred experimental to cages. Overwintered adults were collectively placed in the cage with fresh wheat and allowed to feed for 2-3 days. They were then placed in experimental cages. This was done on April 22, 2002, in Nurdagi and on April 18, 2002, in Oguzeli. The percent spike damage for each cage, called white-spike damage, was calculated. Overwintered adults were allowed to feed on wheat until May 10, 2002, which was the beginning of the milky period and damaged and undamaged spikes were recorded in each cage separately. When the wheat reached harvesting stage, all plants in each cage were cut and put in a bag separately. The damage of various Sunn Pest densities on spike and the total yield were evaluated by analysis of variance (ANOVA) for each site. The same study was repeated in Tigem and Sanko on April 17, 2003, and evaluation for spike damage was made on May 17, 2003.

# Kernel Damage Caused by Nymphs and New Generation Adults

To determine the damage caused by nymphs and new generation adults on kernels, studies were started in cages on May 17, 2003, in Sanko. A completely randomized plot design with 5 treatments (5, 10, 15, 20 nymphs/m<sup>2</sup> and control) and 4 replications was used. At the beginning of the milky period of wheat, second or third instars were released into cages. When the plants reached the harvesting stage, all plants in each cage were cut and put in a bag and brought to the laboratory.

To determine percentage of kernel damage caused by nymphs and new generation adults, 100 kernels were randomly selected from each cage and inspected under a dissecting microscope. This was repeated 20 times. Damaged and undamaged kernels were counted and damage calculated based on Sunn Pest density in each cage (2).

# **Results and Discussion**

# Yield Loss Caused by Overwintered Adults (Spike Damage)

During our studies, when Sunn Pest migrated from overwintering sites to wheat fields, wheat plants were 15-20 cm in height. Therefore there was no stem damage. When wheat plants reached 10-15 cm high, damage by adult Sunn Pest on leaves and stems is not important, but they continue to feed and cause spike damage (5).

No differences were found between spike damage rates in 2003, while some differences between spike damage rates were statistically significant in 2002. In both years, there were no statistical differences among total yields harvested from cages (Tables 1 and 2).

Ten adult Sunn Pest per m<sup>2</sup> caused <1% spike damage. In wheat fields with high populations, the spike damage rate was 10-90% (8). Kılıç *et al.* (3) found that 0.4, 1.0-1.5, 1.6-2.0, and 2.1-2.3 adult Sunn Pest per m<sup>2</sup> caused 1.1, 3.6, 4.2 and 6.6% spike damage in wheat fields, respectively. im ek *et al.* (7) stated that when overwintered adult density was one adult per m<sup>2</sup>, 7% stem damage and 1.9% spike damage resulted.

Tab	le 1	. Spike d	lamage rate (%	), tota	l yield (g	/ 2
m²)	at	various	overwintered	adult	densities	in
Nurc	lagi	and Ogu	zeli, 2002.			

Density	Spike damage	Total yield
(adults /m <sup>2</sup> )	(%)	$(g/2 m^2)$
<u>Nurdagi</u>		
Control	0.000±0.000 a	$968.0 \pm 42.60$
1	0.072±0.036 ab	$1049.0 \pm 78.34$
2	0.104±0.040 abc	$1061.0 \pm 72.10$
3	0.266±0.065 bc	1124.0±61.03
5	0.250±0.066 bc	$1084.0 \pm 42.37$
10	0.312±0.122 c	956.0±37.04
<u>Oguzeli</u>		
Control	0.188±0.060 a	1099.0±20.22
1	0.228±0.090 a	$1150.0\pm 66.41$
2	0.562±0.090 ab	1033.0±64.26
3	0.570±0.120 ab	$1142.0\pm 32.06$
5	0.752±0.070 b	1066.0±23.99
10	0.878±0.270 b	$1064.0\pm24.26$

df= 5,24 *P*<0.05; Means followed by different letters within a column are significantly different at Duncan test.

**Table 2.** Spike damage rate (%), total yield (g/2 m<sup>2</sup>) at various overwintered adult densities in Tigem and Sanko, 2003.

Density	Spike damage	Total yield
(adults /m <sup>2</sup> )	(%)	$(g/2 m^2)$
Tigem		
Control	0.000±0.000 a	911.20±02.00
1	0.045±0.030 a	980.00±73.35
2	0.160±0.130 a	920.00±39.70
3	0.120±0.070 a	$880.00 \pm 62.77$
5	0.235±0.010 a	$1165.00 \pm 102.1$
10	0.256±0.150 a	973.70±168.4
<u>Sanko</u>		
Control	0.000±0.000 a	$432.50 \pm 52.97$
1	0.000±0.000 a	415.00±53.30
2	0.017±0.017 a	$452.00 \pm 47.80$
3	0.000±0.000 a	$308.75 \pm 58.32$
5	0.000±0.000 a	$357.50 \pm 38.32$
10	0.070±0.070 a	331.25±36.65

df= 5,18 P < 0.05; Means followed by different letters within a column are significantly different at Duncan test.

# Kernel Damage Caused by Nymphs and New Generation Adults

The percent kernel damage depended on the nymphal density in each cage (Table 3). Kernel damage caused by nymphs varied from 0.287 to 0.625%. There were no statistical differences among kernel damage rates. When nymph density reached 32.6 per  $m^2$ , Sunn Pest caused 15% kernel damage in the south Anatolia Region of Turkey in field conditions (4). Ten nymphs per  $m^2$  caused ~2-3% kernel damage (1). Our results do not match these findings.

**Table 3.** Kernel damage (%) at various nymphal densities in Sanko, 2003.

Density	
(nymphs /m <sup>2</sup> )	Kernel damage (%)
Control	0.00±0.00 a
5	0.52±0.15 a
10	0.28±0.01 a
15	0.62±0.20 a
20	0.55±0.27 a

df= 4,15 P<0.05; Means followed by different letters within a column are significantly different at Duncan test.

# Conclusions

Small cheesecloth cage studies may not give reliable results for determining ET of adult Sunn Pest. Cages may change feeding behavior and shorten the lifetime of adults.

Most of the nymphs released into cages might not have reached adulthood, because of cage conditions. Therefore expected kernel damage based on nymphal density was not seen.

## Acknowledgement

This study was a part of the DFID project on Integrated Pest Management of Sunn Pest in West Asia. We would like to thank ICARDA and the project leader, Dr. M. El Bouhssini for their support of our work.

## الملخص

جانهلال، رمضان، خليل كنك، محمود إسلام أوغلو، أ. ديران كانات وأيكوت غول. 2007. تقديرالخسارة الناتجة عن ضرر حشرة السونة على القمح في تركيا. الصفحات 187–190.

أجريت دراسة في أقفاص لتحديد ضرر بالغات وحوريات حشرة السونة على القمح في موقعين مختلفين خلال عامي 2002 و 2003. بلغت كثافة البالغات في الأقفاص 1، 2، 3، 5 و 10 بالغات/م<sup>2</sup>، وكثافة الحوريات 5، 10، 15 و 20 حورية/م<sup>2</sup>. إن الضرر الذي تسببه البالغات على السنابل يعتمد على عدد حشرات السونة التي أطلقت في الأقفاص، والتي بلغت 0.072– 1150–90.00% و 0.000–250.0% خلال عامي 2002 و 2003، على التوالي. بلغت الغلة الكلية 656–1150 و 2003–1165 غ /2 م<sup>2</sup> خلال عامي 2002 و 2003، على التوالي. بلغت الغلة الكلية م566–1050 و 2003–2003 على السنابل معنوية إحصائياً في عام 2002 فقط، ولم يكن هناك اختلاف بين الغلة الكلية المحصودة من الأقفاص في كلا العامين. اعتمد ضرر الحبوب الذي تسببه الحوريات على عدد الحوريات التي أطلقت في الأقفاص في كلا العامين. 2003

كلمات مفتاحية: أقفاص، عتبة اقتصادية، ضرر حبوب وسنابل، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.



# Timing as a Tactic of Ecological Selectivity for Chemical Control of Sunn Pest

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## Abstract

Sheikhi Garjan, A., A. Mohammadipour, Gh. Radjabi and Gh. Sabahi. 2007. Timing as a Tactic of Ecological Selectivity for Chemical Control of Sunn Pest. Pages 193-201.

Nine insecticides were tested against overwintered adults and nymphs in Iran. Two years of experimentation showed that spraying against overwintered adults had no significant effects on reducing punctured and damaged wheat kernels. The quantity of rainfall and movement of overwintered adults among the fields were effective factors in resulting in low efficacy of the chemical treatments against overwintered adults, and also this application had negative effects on natural enemies. Trichlorfon was the only selective insecticide for chemical control of overwintered adults and nymphs. Lambda-cyhalothrine, deltamethrin, and esfenvalerate were more efficient compared to other insecticides for control of nymphs. Applications targeting 3<sup>rd</sup> instar nymphs of Sunn Pest significantly reduced punctured and damaged kernels (>2%). Applications against overwintered adults of had less efficacy compared to applications against nymphs. **Keywords**: *Eurygaster integriceps*, insecticides, overwintered adults, natural enemies, nymphs.

## Introduction

Wheat is a fundamental and staple crop in the economy and agricultural activities of Iran. It ranks first among cultivated crops in terms of both production and area under cultivation. More than 5.73 million ha. of wheat were planted in Iran in 2003 (7).

Sunn Pest, *Eurygaster integriceps* Puton is the most important pest of wheat in Iran. The problem surfaced and became economically important after 1980 in the former Union of Soviet Socialist Republics, Afghanistan, Iraq, Syria, Turkey, Bulgaria and Romania where the increasing use of chemical controls was accompanied by an increase of new cereal pests and the repetition of research activities on subjects often studied before (19).

In Iran, in 2003, approximately 1.6 million hectares were sprayed against Sunn Pest. Eighty percent of the total area was treated by ground application and the rest by aerial application. Seventy to eighty percent of the applications were against nymphs and 20-30% against overwintered adults (7). A broad spectrum insecticide, fenitrothium, is used for chemical control of overwintered adults and

new generation Sunn Pest. Other insecticides such as trichlorfon and fentithion may also be used (1).

Overwintered adults attack leaves, central shoots or ears. The color of attacked parts of wheat changes to brown or white. When the damage takes place at the early stage of wheat growth, shoots die before ear formation but the plant responds by producing new shoots which develop ears with fewer grains. High population of overwintered adults can reduce yields considerably (3, 4, 11).

In general, application of insecticides on overwintered adults negatively affects natural enemies' populations, because it can reduce parasitism percentage of Sunn Pest egg from 86.9% to 8.3% in some cases (12). Chemical control also disrupts the beneficial insects which maintain Sunn Pest populations at acceptable levels under normal conditions (22).

Applications against overwintered adults are commonly done with spring rainfalls and this adversely affects the deposition of the insecticide and thus reduces mortality of the pest (20, 24). The objectives of this study were to evaluate efficacy of candidate insecticides to overwintered adults based on the resultant density of the new generation population. The main goal was to compare efficacy and selectivity of new insecticides with fenitrothion, as a traditional insecticide.

# Materials and Methods

From, 1999-2000 two field trials were conducted at the Sunn Pest Research Department, Plant Pests and Diseases Research Institute (PPDRI) located at Varamin. For field trial I, treatments included Phosalone (Zolone, 35 EC, formulated in Iran) at 1400 ×0.35 g (ai)/ha; fenvalerate (Sumicidin 20 EC, formulated in Iran) at 115 x 0.2 g (ai)/ha; diazinon (diazinon 60 EC, formulated in Iran ) at 1500 x 0.6g (ai)/ha; fenitrothion (fenitrothion 50 EC, formulated in Iran) at 1000 x 0.5 g (ai)/ha; pirimicarb (Primor 50 WP, Zeneca) at 280 x 0.5g (ai)/ha and trichlorfon (Dipterex 80 SP, Bayer) at 1250 x 0.8 g (ai)/ha. Trial I had 7 treatments, 6 insecticides and a check, arranged in a randomized complete block design (RCBD) with 4 replications. In the second year in trial I, pirimicarb was replaced by lambdacyhalothrine (Karate 5 EC, Zeneca) at 150 x 0.05 g (ai)/ha. For trial II, treatments included fenitrothion (fenitrothion 50 EC, formulated in Iran) at 1000 x 0.5g (ai)/ha; deltamethrin (Decis 2.5 EC Agroevo) at 300 x 0.025 g (ai)/ha; esfenvalerate (Sumi-alpha 5 EC, Sumitomo) at 150 x 0.05 g (ai)/ha, fenitrothion (fenitrothion 20 MC, Sumitomo) at 2500 x 0.2 g (ai)/ha. In trial II these 5 treatments included 4 insecticides and check, arranged in a randomized complete block design (RCBD) and each treatment had 4 replications. Each plot was 25 x 20 m<sup>2</sup>, separated by 3 m as a buffer row. Treatments were applied with a lance sprayer mounted tractor as usual sprayer calibrated to deliver 250 L/ha. Treatments were applied at wheat growth stage 61 (Zodaks) in the morning, when more overwintered adults were settled in the field.

At the time of application, average ambient temperature and relative humidity were 23°C and 60%, respectively.

Overwintered adults, new adults, nymphs and natural enemies were collected and counted in each sample unit, which consisted on 5 sweepings with a net over the tops of wheat plants. In each plot, 3 sample units were taken. All treatments were evaluated 1 day before and 2, 5, 8, 14 and 24 days after application. Before harvest, ten 1/4 m<sup>2</sup> quadrants were evaluated in each plot counting the total number of ears and selecting two ears randomly from each quadrant for evaluation. Therefore, 20 ears were randomly collected to determination Sunn Pest damaged kernel rate in each plot.

Total pest mortality data was transformed by arcsine and subjected to one way analysis of variance (ANOVA) and treatment means were compared using Duncan's multiple range test (P=0.05) (14).

This study was also done to find the effect of insecticides against nymphal stage on third instar Sunn Pest. The methods used were as described above.

## Results

## **Application Against Overwintered Adults**

All insecticides in each of the two trials caused 20-50% and 30-60% mortality on overwintered adults and the new generation population respectively (Figures 1 and 2). Deltamethrin had the most (~90% mortality) and phosalone the least (~35% mortality) on overwintered adults. At 22 d post application, trichlorfon and pirimicarb treatments had the least efficacy on new generation (~30% mortality), whereas deltamethrin had the most efficacy on new generation (~75% mortality) (Figures 1 and 2).

Lambda-cyhalothrine had the most (~75% mortality) and phosalone had the least efficacy (~38% mortality) at 22 days after application among tested insecticides in trial I and II (Figures 3 and 4).



Figure 1. Efficacy means  $(\pm SE)$  of different insecticides on overwintered adults at 3 & 5 days (a) and on nymphs at 14 & 22 days (b) after application against overwintered adults in trial I in the first year. Treatments with the same letter are not significantly different in each time after application (P=0.05; Duncan's [1995] multiple range test).

The results of trial II revealed that fenitrothion 20 MC had more efficacy than fenitrothion 50 EC at 22 d after application in both years. Results obtained from both trials (I, II) indicated that all insecticides had 30-80 mortality on overwintered adults but only synthetic pyrethroid insecticides and fenitrothion 20 MC could have more effect on population density of new generation compared with other insecticides.

Observations of damaged kernels in all treatments showed that treatments against overwintered adults did not reduce damage below 2% (the accepted damage rate in Iran) (Tables 1 and 2).

**Table 1.** Means (±SE) of Sunn Pest damaged kernelrate as percentage in treatments of trial I(application against overwintered adults).

	Year	
Treatments	First	Second
Fenitrothion	2.8±0.5	3.8±2.0
Control	6.4±2.3	8.0±3.0
Trichlorfon	3.1±0.6	4.1±2.0
Phosalone	6.1±1.4	6.1±2.0
Diazinon	3.0±0.5	$4.0{\pm}1.8$
Fenvalerate	7.1±1.1	7.1±2.8
Pirimicarb A*	6.2±2.0	3.0±2.0

\* was pirimicarb in first year and lambda-cyhalothrine in second year.



**Figure 2.** Efficacy means  $(\pm SE)$  of different insecticides on overwintered adults at 5 days (a) and on nymphs, 28 days (b) after application against overwintered adults in trial II in the first year. Treatments with the same letter are not significantly different in each time after application (P=0.05; Duncan's [1995] multiple range test).

 Table 2. Means (±SE) of Sunn Pest damaged kernel

 rate as percentage in treatments of trial II

 (application against overwintered adults).

	Year	
Treatments	First	Second
Control	$15.0 \pm 2.5$	12.0±3.5
Esfenvalerate	$11.8 \pm 2.0$	8.5±2.0
Fenitrothion EC	$3.8{\pm}1.8$	2.8±0.6
Fenitrothion MC	7.0±2.0	5.2±1.5
Deltamethrin	3.8±1.5	2.5±1.0



**Figure 3.** Efficacy means  $(\pm SE)$  of different insecticides on overwintered adults at 8 days (a) and on nymphs at 22 days (b) after application against overwintered adults of Sunn Pest in trial II in the second year. Treatments with the same letter are not significantly different in each time after application (P=0.05; Duncan's [1995] multiple range test).

### **Application Against New Generation**

All insecticide treatments had a high efficacy (80-98%) on new generation Sunn Pest on 2 days after application, except phosalone and pirimicarb (40%). Deltamethrin had the most efficacies ( $98\pm2\%$ ) and phosalone had the

least ( $40.2\pm3\%$ ) on Sunn Pest (Figures 5 and 6). Sunn Pest is very susceptible to synthetic pyrethroid insecticides, fenitrothion, diazinon and trichlorfon but very tolerable to phosalone and pirimicarb. Except phosalone and pirimicarb treatments, the insecticides tested lowered the damaged kernel rate below 2% (Tables 3 and 4).

# Discussion

None of the insecticides used against overwintered Sunn Pest were effective in reducing the rate of damaged kernels. Deltamethrin and lambda-cyhalothrine decreased new generation density lower than control at 22 days post application. Previous studies have documented that chemical control of *E. integriceps* on wheat in Ukraine has been based on controlling overwintered adults (2).



Figure 4. Efficacy means  $(\pm SE)$  of different insecticides on overwintered adults at 7 days (a) and on nymphs at 22 days (b) after application against over wintered adult. Treatments with the same letter are not significantly different in each time after application (P=0.05; Duncan's [1995] multiple range test).

**Table 3.** Means (±SE) of Sunn Pest damaged kernelrate as percentage in treatments of trial I(application against new generation of Sunn Pest).

	Year	
Treatments	First	Second
Fenitrothion	$0.8\pm0.4$	1.5±0.6
Control	4.0±0.6	6.3±3.0
Trichlorfon	1.5±0.6	1.5±0.5
Phosalone	3.8±0.3	7.0±3.5
Diazinon	0.3±0.8	2.0±1.0
Fenvalerate	0.5±0.4	0.2±0.1
Pirimicarb A*	2.3±0.8	0.3±0.3

\* pirimicarb in the first year and lambda-cyhalothrine in the second year.



**Figure 5.** Efficacy means  $(\pm SE)$  of different insecticides on nymphs and young adults in application against new generation in trials I (a) & II (b) in the first year. Treatments with the same letter are not significantly different in each chart (P=0.05; Duncan's [1995] multiple range test).

**Table 4.** Means (±SE) of Sunn Pest damaged kernelrate as percentage in treatments of trial II(application against new generation of Sunn Pest).

	Year	
Treatments	First	Second
Control	6.6±1.5	8.0±2.0
Esfenvalerate	1.8±0.5	2.0±0.5
Fenitrothion EC	0.3±0.8	2.0±0.8
Fenitrothion MC	1.8±0.6	0.6±0.3
Deltamethrin	$0.5\pm0.5$	0.3±0.2



Figure 6. Efficacy means  $(\pm SE)$  of different insecticides on nymphs in application against new generation in trials I (a) & II (B) in the second year. Treatments with the same letter are not significantly different in each chart (P=0.05; Duncan's [1995] multiple range test).

Applications against new generation Sunn Pest showed that chemical control can affect density especially by use of synthetic pyrethroids. In this period, natural enemies have less contact exposure to leaf deposits because their production has ended and they have started to leave the wheat fields (Figure 7) (15) and also, diversity of field canopies and habitats of natural enemies are more than in the period of application against overwintered adults. Therefore applications against new generation populations have a lower impact on natural enemies (5, 9).

The egg parasitoids play a major role in reducing Sunn Pest populations. Early in the growing season 70-80% of all Sunn Pest eggs are parasitized by overwintered parasitoids. Only 15 to 25% of the first adult parasitoids emerge from the overwintered generation and are able to parasitize Sunn Pest eggs (10, 11, 13, 23). Therefore, the application against new generation can not be coincided with activity of overwintered parasitoids. For this reason this period of chemical control has less of an adverse effect on parasitism and mortality of parasitoids compared to the application against overwintered adults.

The beginning and duration of migration of overwintered adults from mountains to wheat fields depends on climatic conditions. Investigations during 12 years showed that the mean length of this migration was nearly 30 d. Decrease of temperature and high frequency and quantity of rainfall can lengthen that period (Figure 8) (6, 11). The overwintered adults can move from one field to another shortly after the main migration from the mountains. These behaviors of overwintered adults can decrease efficacy of chemical control and sometimes it may seem that some wheat fields with a low density of overwintered adults were sprayed (11).

Investigating the effect of artificial rain on the efficacy of insecticides on the pest in wheat and barley fields showed that rain had no adverse effect on the efficacy of deltamethrin, whereas the efficacy of pirimicarb was reduced slightly by the rain. Early and heavy rain after a few days application has a greater impact on insecticide efficacy than late and light rains (20). Literature review showed that organophosphorus and carbamate insecticides were toxic for short periods (8 days) (16). In during the time of chemical application against overwintered adults, the mean quantity of rainfall was usually more than in that of application against the new generation (Figure 8) therefore, rain may reduce the efficacy of chemical application against overwintered adults.

Estimations of total percentage mortality were similar to results of bioassay of insecticides against overwintered adults and new generation (17). Trichlorfon had high efficacy on overwintered adults at 5 d post treatment and low efficacy on new generation at 22 days. These results apparently reflected a rapid decline of trichlorfon residues or its halflife in the field. High efficacy of synthetic pyrethroid insecticides both on overwintered adults and new generations indicated that these insecticides were long-lasting in the field. These results were the same as results of Sheikhi *et al.* (16) and Theilling & Croft (21).

Application against overwintered adults could not reduce density of new generation, because population density of new generation (nymphs and new adults) in all treatments was above the ET. Insecticide applications against overwintered adults often can not reduce the need for applications against new generation Sunn Pest which are primarily responsible for damage to kernels. The chemical control of overwintered adults is not efficacious because of rainfall and long periods of Sunn Pest migration from the mountains, but having a good monitoring system and information about spatial and temporal distribution of Sunn Pest, topography and climatic conditions of the region can increase the efficacy of insecticides and lead to avoiding additional applications against the nymphal stage.

Application against overwintered adults is usually coincide with the immigration and reproduction of beneficial arthropods, thus, use of synthetic pyrethroid insecticides such as, deltamethrin and lambda-cyhalothrine, which have good toxicity on adults and high residual activity on nymphs, can not be recommended for chemical control of overwintered adults, because they cause mortality of natural enemies and outbreak of secondary pests such as cereal aphids (5, 16).

To control overwintered adults, use of trichlorfon is more reliable compared to synthetic pyrethroid insecticides, because this insecticide has high toxicity but short residual activity. It allows natural enemies to immigrate again to sprayed fields, to begin their reproduction and to build up their population.

Additional studies to develop a Sunn Pest management model regarding timing of application and use of selective insecticides are needed. The ovicidal effects of insecticides should be determined. In conclusion, results confirmed that the performance of pyrethroid insecticides on overwintered adults can be attributed to mortality of both overwintered adults and nymphs. Control of overwintered adults could be effective only if landing or settling of overwintered adults is complete and no rainfall occurs after application (5, 8) and also it can have important role in chemical control of Sunn Pest if we restrict it only on the first landing spots of overwintered adults.



Figure 7. Population dynamism of aphid and natural enemies in wheat fields (15).



Figure 8. Average precipitation of different months in Tehran province, Iran, 2004 (24).

# الملخص

شيخي كرجان، عزيز . أ.، أ. محمد يبور، غ، راجابي، و غ. ساباهي. 2007. التوقيت كوسيلة للانتقائية البيئية للمكافحة الكيميائية لحشرة السونة. الصفحات 193–201.

اختبرت تسعة مبيدات حشرات ضد بالغات حشرة السونة المشتية والحوريات في إيران. أظهرت نتائج عامين من التجارب أن رش البالغات المشتية لم يكن له أي تأثير معنوي في تخفيض نسبة حبوب القمح المتقوبة والمتضررة. كانت كمية الهطول المطري وانتقال البالغات المشتية بين الحقول عوامل فعالة في تخفيض فعالية المعاملات الكيميائية للبالغات المشتية. كما كان لهذا الرش تأثير سلبي على الأعداء الحيوية. كان الترايكلوروفون المبيد الحشري الانتقائي الوحيد للمكافحة الكيميائية للبالغات المشتية والحوريات. كانت المبيدات لامبدا-سيهالوثرين، دلتامثرين، إسفنفاليرات أكثر فعالية في مكافحة الحوريات بالمقارنة مع مبيدات أخرى للحشرات. خفض الرش ضد حوريات العمر الثالث ويشكل معنوي الحبوب المتقوبة والمتضررة (> 2%). كانت فعالية الرش ضد البالغات المشتية أقل بالمقارنة مع الرش ضد الحوريات.

كلمات مفتاحية: Eurygaster integriceps، مبيدات حشرات، بالغات مشتية، أعداء طبيعية، حوريات.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.
# Properties and Regulation of Activity of Cholinesterases and Carboxylesterases on Sunn Pest

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#### Abstract

# Kushiev, H., I. Allaeva and Z. Tilyabaev. 2007. Properties and Regulation of Activity of Cholinesterases and Carboxylesterases on Sunn Pest. Pages 203-208.

This work is dedicated to the study of catalytic properties of metabolic enzymes of *Eurygaster integriceps* Puton - basic pest agricultural plants by the method of substrate-inhibitor analysis in connection with the problem of selectivity action insecticides. In this work it was defined localization and dynamic activity of cholinesterase and carboxylesteras of *E. integriceps* and was shown antiesteras properties of a number of O,O-dialkyl-S-epilupinantiophosphat ( $R = C_2H_5$ ,  $C_3H_7$ ,  $C_4H_9$ , i-C<sub>4</sub>H<sub>9</sub>) in their interaction with cholinesterase and carboxylesteras were established.

Keywords: Acetylcholinesterase, butirylcholinesterase, carboxylesteras, epilupinantiophosphat, *Eurygaster integriceps*, onthogenes, O,O-dialkyl-S-epilupinantiophosphat.

## Introduction

Chemical preparations are important as a means of combating agricultural pests. But the existing number of these materials does not answer the needs of agriculture as many pests quickly develop resistance to them.

The toxicity of phosphororganic and carbamate insecticides is caused by their ability to inhibit the catalytic activity of acetylcholinesterases (AchE) (5, 7, 9) and the resistance of pests to such insecticides is caused by the induction of some metabolic ferment systems such as carboxylesterases (CBE), glutation-S-transpherases, oxidas of mixed functions, etc. That's why combating agricultural pests, especially those resistant to insecticides, depends on an all-inclusive investigation of the properties of metabolic ferments, localization and dynamics of their activity, and the chemical nature of inhibitors which can influence effectively the functional properties of these ferments.

We report on substrate specificity and perceptibility of cholinesterases (ChE) and CBE of *Eurygaster integriceps* Puton to phosphororganic compounds (PhOC) at different stages of their development.

# **Materials and Methods**

*Eurygaster integriceps* were grown in laboratory conditions. To determine the activity of ChE and CBE, homogenates from the pectoral and cerebral parts were prepared at a rate of 1 g of tissue per 9 ml of 0.05 M phosphate buffer (pH 7.5).

The ferment activity was determined at 25°C pH 7.5 using thiocholinesterases of acetic, propionic and butric acids as substrates for ChE and S-ethylic ether of buteric acid for CBE. Thiocholine ethers were purchased "Chemapol" (Czechoslovakia). from Proserineis, a commercial preparation, was purchased from Germany. S-ethylic ether of butyric acid and PhOC were synthesized in the laboratory at the Institute Bioorganic Chemistry (10).

The catalytic activity of ChE and CBE was Ellman's modified method (2), based on the measurement of the speed of forming yellow coloring at ferment hydrolysis of substrates with the help of 5,5 dythiobis of nitrobenzous acid.

The kinetic parameters of the substrate hydrolysis ( $V_{max}$  and  $K_m$ ) were determined by the method of Lynuiver-Berk (3). Bimolecular

constants (k<sub>2</sub>) of inhibiting ferments of PhOC were calculated by using the formula:

$$k_2 = \frac{2,3}{t \cdot [1]} \lg \frac{v_0}{v_t}$$

The experiments were replicated 3 or 4 times. Statistical analyses were performed using the method of Muller (4).

## **Results and Discussion**

The presence of ChE in different stages of development in ova, nymphs and adults of *E. integriceps*, was investigated. ChE activity was determined by hydrolysis of thiocholine ethers, acethylthiocholine (ATCh), buthyrylthiocholine (BuTCh), and propionylthiocholine (PTCh).

Table 1 gives results of ChE activity of *E. integriceps* at different stages in the pectoral and cerebral parts of the body. An increase of ChE activity happens when nymphs transform to adults. The ferments activity in the pectoral part is more intensive than in the cerebral part. Consequently, ChE activity depends on the development phase. But the degree of activity of ChE and CBE of adults in their cerebral and pectoral parts is equal.

Besides cholinesterase it is possible to determine CBE activity which is the ability of homogenates to break up the specific substrate of CBE S-ethylic ether of butyric acid (KS-14) which was synthesized in the laboratory (Small Organic Synthesis of Physiologically

 $0.40\pm0.02$ 

1.0

4-5 instar

Adult

Active Substrates at the Institute of Bioorganic Chemistry of the Academy of Sciences of Uzbekistan) by B.N. Babaev) (10). The activity of ferments was discovered in nymphs and adults. CBE and ChE are localized in the pectoral and cerebral parts of insects. CBE was mostly active in adults, and less active in the 2-3 instars (Table 1).

ChE and CBE are both localized in the cerebral and pectoral sections of Sunn Pest. It is characteristic of both ferments that their activity increases in the process of development from nymph to adult.

The speed of the ferment hydrolysis of ATCh, PTCh and BuTCh was determined in a broad interval of concentrations from 0.5x10<sup>-2</sup> to 5.0x10<sup>-5</sup> M in the pectoral part of Sunn Pest (Table 2). The homogenate, prepared from the cerebral part differed from the homogenate prepared from the pectoral part by its capacity to break up ATCh, PTCh and BuTCh (Figure 1). The cerebral ferment of E. integriceps hydrolyzes ATCh, with a high speed that is 2.5-3.5 times faster than it hydrolyzes PTCh (which we call a poor speed) or BuTCh, (which we call a weak speed). The results of the investigation demonstrated that various types of cholinesterase are localized in these parts and that the ferment breaking up ATCh with a high speed can be referred to as AChE (K.F.3.1.1.7) and the ferment, that is localized in the pectoral section that hydrolyzes BuTCh more quickly than ATCh and PTCh was BuTCh (K.F.3.1.1.8).

 $0.59 \pm 0.06$ 

1.0

 $0.61 \pm 0.06$ 

1.0

according to the c	evelopment phases.				
		Activity in re	elative units		
	Cerebr	al part	Pectorial part		
The	ChE	CBE		С	
uevelopment	(substrate ATCh)	(substrate:KS-14)	(substrate: ATCh)	(substrate:KS-14)	
Ova	-	-	-	-	
2-3 instar	0.21±0.03	$0.08\pm0.04$	$0.35 \pm 0.04$	$040\pm0.04$	

 $0.12 \pm 0.03$ 

1.0

**Table 1.** The activity dynamics of cholinesterase and carboxylesteras of *Eurygaster integriceps* Puton according to the development phases.

The			Cerebral part				Pectorial part		
develo- pment	Para- meters	ATCh	PTCh	BuTCh	KS-14	ATCh	PTCh	BuTCh	KS-14
2-3 instar	m	14.2±0.23	$4.4 \pm 0.24$	4.1±0.31	0.5±0.03	3.9±0.1	4.0±0.45	2.9±0.11	2.7±0.32
	$V_{\text{vax}}$	3.9±0.10	3.7±0.34	2.0±0.4	0.1±0.23	2.2±0.37	2.01±0.12	4.1±0.13	2.9±0.07
4-5	m	13.8±0.27	13.0±0.55	2.9±0.41	1.2±0.5	5.5±0.31	4.7±0.42	6.9±0.13	3.8±0.23
ilistai	$V_{\text{vax}}$	9.8±0.13	10.0±0.40	4.0±0.1	0.8±0.5	3.8±0.1	3.8±0.33	9.8±0.55	8.02±0.36
Adult	m	5.9±0.32	5.8±0.41	1.6±0.61	-	6.7±0.22	6.02±0.51	4.4±0.37	-
	$V_{\text{vax}}$	17.9±1.4	17.9±0.6	4.7±0.31	-	11.1±0.4	$10.01 \pm 0.05$	7.7±0.23	-

**Table 2.** The kinetic parameters ( $_{\rm m}$  10<sup>-3</sup> and V<sub>max</sub> 10<sup>-6</sup> . mg<sup>-1</sup>.min<sup>-1</sup>) of the hydrolysis of substrates under the influence of esterases *Eurygaster integriceps* Puton.



**Figure 1.** The dependence of activity of cholinesterase and carboxylesteras of the 4-5 instar of *Eurygaster integriceps* Puton on the substrates concentration. A-the pectorial part; B-the cerebral part: (1)- ATCh; (2)-BTCh; (3)- PTCh; (4)- KS-14. V-the speed of the ferment hydrolysis of substrates (mmol/mg); pS-negative logarithm of the mole concentration of substrates.

For greater confidence than when we investigated one ChE in the cerebral part and the other in the pectoral part of Sunn Pest we determined the constant quantities of the speed of interaction of ChE with proserine and some PhOC which are different in their structures (Table 3).

The choice of these inhibitors was made because proserine and Gd-7 are selective inhibitors of AChE (1), and O-ethyl-S- $\beta$ -ethylanabasinylphnylthiophosphate and

O,O-dibutyl-S-butynpyperidylthiophosphate display more specificity with respect to BuChE (8).

From Table 3 it is clear that ChE localized in the cerebral part is inhibited by small concentrations of the inhibitor Gd-7 and correspondingly has high constants of inhibition in comparison with ChE of the cerebral part (AChE). The high values of  $k_2$  for ChE of the pectoral section when interacting with O-penthyl-S- $\beta$ -ethylanabasinylphenylthiophosphate and O,O-dibutyl-S-buinpiperidylthiophosphate showed

that this ferment is apparently BuChE. CBE of insects did not practically display perceptibility to the selected PhOC. Such phenomenon was fixed in respect of CBE of cockroaches, aphids and the nervous and adipose tissues of the desert locust (6, 9, 10). The acnive centure of CBE was the only etherizing point (location).

Anticholinesterasing activity of O,Odialkyl-S-epyilupinanthiophosphates with respect to AChE and BuChE of nymphs and adults of *E. integriceps* has been studied (Table 4).

**Table 3.** The perceptibility  $(k_2, -1 \text{ min}^{-1})$  of cholinesterase of the larvae of the 4-5 age of *Eurygaster integriceps* Put. to inhibitors of various chemical nature inhibitors of different chemical nature.

	Chalingstangs of the combrol part		Cholinesterase of the pectorial			
	Cholinester	ase of the ce	rebral part		part	
Inhibitors	ATCh	BuTCh	PTCh	ATCh	BuTCh	PTKh
Proserin (CH <sub>3</sub> ) <sub>2</sub> NC(O)O H <sub>3</sub> C-N -CH <sub>3</sub> CH <sub>3</sub>	I <sub>50</sub> =8.2 x 10 <sup>-4</sup>	I <sub>50</sub> =1.5 x 10 <sup>-5</sup>	I <sub>50</sub> =1.7 x 10 <sup>-5</sup>	I <sub>50</sub> =3.01 x 10 <sup>-4</sup>	I <sub>50</sub> = 3.7 x 10 <sup>-4</sup>	I <sub>50</sub> =3.8 x 10 <sup>-4</sup>
Gd-7 $CH_3$ $P$ $C_2 H_5$ $S-C_2 H_4-S-C_2 H_5$	(6.6±0.12) x 10 <sup>5</sup>	(7.8±0.4) 10 <sup>5</sup>	(6.1 ±0.6) x 10 <sup>5</sup>	(1.6±0.6) 10 <sup>4</sup>	(3.1 ±0.71) x 10 <sup>4</sup>	(3±0.3) x 10 <sup>4</sup>
O-ethyl–Sethyl- anabasinylthiophos- phanat $C_2 H_5$ $C_2 H_{11}$ P $C_2 H_4$ -N $C_2 H_4$ -N	(7.09±0.5) 10 <sup>4</sup>				(1.4± 0.21) x 10 <sup>7</sup>	
0,0-dibutyl–S-butynpiperidylthio- phosphat $C_{4}H_{9} \xrightarrow{O} C_{4}H_{9} \xrightarrow{S} CH_{2}-C=C-CH_{2}-N$	(2.8±0.39) 10 <sup>6</sup>				$(2.9 \pm 0.32) \ge 10^8$	

	2-3 Instar		<b>4-5 I</b>	4-5 Instar		Adult	
	AKhE	BuKhE	AkhE	BuKhE	AKhE	BuKhE	
C <sub>2</sub> H <sub>5</sub>	0.41±0.11	0.15±0.12	0.31±0.22	0.16±0.37	0.11±0.51	0.11±0.43	
$C_3H_7$	0.59±0.12	0.19±0.23	$0.76 \pm 0.05$	$0.18\pm0.21$	0.31±0.19	$0.08 \pm 0.21$	
C <sub>4</sub> H <sub>9</sub>	3.9±0.31	3.2±0.25	3.1±0.21	1.9±0.36	$1.5\pm0.28$	$1.35 \pm 0.39$	
$i - C_4H_9$	$0.32\pm0.21$	$0.15 \pm 0.21$	$0.21 \pm 0.05$	$0.07 \pm 0.48$	$0.1\pm0.11$	$0.05 \pm 0.48$	

**Table 4.** Anticholinesterasing activity  $(k_2, 10^6 - 1 \text{ min}^{-1})$ , -dialkyl-S-epilupinanthiophosphate.

The lengthening of the alkyl radical from  $C_2H_5$  to  $C_4H_9$  increased the activity of inhibitors. The greatest activity was observed for O,O-dibutyl-S-epilupinanthiophosphate.

When studying biochemical properties of ChE and CBE of *E. integriceps* with the help of substrate-inhibitoring methods, we found the localization of these ferments in various tissues and that CBE is localized mainly in the pectorial part of the body and slightly in the cerebral part, and AChE and BuChE are accordingly in the pectorial and cerebral parts.

Anticholinesterasing activity of some PhOC with different chemical structures was studied. We observed an increase of activity to the butyl radical of O,O-dialkyl-S-epilupinanthiophosphates.

But, when transforming into  $i-C_4H_9$  derivatives the value  $k_2$  drops abruptly, which proves the high degree of affinity of O,O-dibutyl-S-epilupinanthiophosphate with the active center of AChE and BuChE of *E. integriceps*.

AChE of nymphs possesses a greater susceptibility to the influence of PhOC than AChE of adults. So, on the basis of the substrate-inhibitor analysis we can state that there compounds of nymphs and adults of *E. integriceps* are both localized in their cerebral and pectorial parts.

# الملخص

كوشيف، حبيب، ل. اللايفا وز. تيليابايف. 2007. خصائص وتنظيم نشاط انزيمات كولين استر وكاربوكسيل استر في حشرة السونة. الصفحات 203–208.

يهدف البحث لدراسة الخصائص التحفيزية للأنزيمات الاستقلابية لحشرة السونة (Eurygaster integriceps Puton.)، الآفة الأساسية للنباتات الزراعية، بطريقة تحليل مثبط مادة التفاعل الأنزيمي المرتبط بفعل مبيدات الحشرات الانتقائي. تم تحديد التمركز والنشاط الحركي لاتزيمات كولين استر وكاربوكسيل استر عند حشرة السونة، كما تم إظهار خصائص مضاد انزيم الاستر عند عدد من (O,O-dialkyl-S-epilupinantiophosphat (R = C<sub>2</sub>H<sub>5</sub>, C<sub>3</sub>H<sub>7</sub>, C<sub>4</sub>H<sub>9</sub>, i-C<sub>4</sub>H<sub>9</sub>) مع انزيمات كولين استر وكاربوكسيل استر.

كلمات مفتاحية: انزيم أسيتيل كولين استر، انزيم بوتيريل كولين استر، انزيم كاربوكسيل استر، إيبيلوبينانتيوفوسفات، • G,O-dialkyl-S-epilupinantiophosphat، أونثوجينات، Eurygaster integriceps،

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Aerial vs. Ground Applications for Sunn Pest Management

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#### Abstract

Erman, A., Y. Sabaho lu, N. Babaro lu and R. Canhilal. 2007. Aerial vs. Ground Applications for Sunn Pest Management. Pages 209-213.

Cereals are the most important crops for internal consumption and export in Turkey. Wheat and barley are grown on 13,765,000 ha. The use of insecticides is a common method for protection against Sunn Pest. Commonly liquid pesticides have been applied by air and until 2000 treatments were conducted by the government. With a new approach, farmers will spray their fields using ground sprayers. In this study, for more effective spraying and less non-target contamination, advantages and disadvantages of aerial spraying vs. ground spraying are examined.

Keywords: Aerial spraying, ground spraying, management, Sunn Pest.

#### Introduction

In Turkey 39.3% of the agricultural and forest area (18,123,000 ha) is cultivated. Cereals are grown on 13,765,000 ha and annual production is 30,161,500 tons. The area devoted to wheat and barley production and yield between 1995 and 2002 are presented in Table 1. Wheat and barley are produced on 9,300,000 ha and 3,600,000 ha respectively; 19,000,500 tons of wheat and 8,300,000 tons of barley are produced (6). Cereals are the staple food for the population. Insect pests are among the most important factors limiting production.

Sunn Pest is one of the most important pests of cereals (wheat, barley) in Turkey; there are three economically important species, *Eurygaster integriceps* Puton., *E. maura* L. and *E. austriaca* Schrank. The dominant species is *E. integriceps* in South and Southeast Anatolia and Thrace, while *E. maura* dominates in Central Anatolia and the Aegean Region. Sunn Pest became the most important pest in 1927-1929 in South Anatolia, in 1939-1941 in Southeast Anatolia and since the 1980's in Thrace, Central Anatolia, Aegean and Marmara Regions. When the population density is high and control measures are neglected, damage levels may reach 100%.

#### Sunn Pest Outbreaks

The first outbreaks of Sunn Pest were recorded in 1927-1929 in South Anatolia and in 1939-1941 in Southeast Anatolia. Outbreaks also occurred 1955-1959 and 1965-1973 in South Anatolia. These outbreaks recurred in 1977 in these regions and continue to the present day.

The first Sunn Pest outbreaks in the Aegean Region started in 1987, in Thrace in 1982, in Central Anatolia in 1988, and in Marmara Region in 1990. These outbreaks are still going on. Overwintering and infestation rates of Sunn Pest in the 1950's and 2000's showed variation respectively. If we make a comparison between these years, we see that, in the past, Sunn Pest was present only in South and Southeast Anatolia, but now it also causes significant damage in Central Anatolia, Aegean, Marmara and Thrace. So 76% of the area sown with barley and 71% of the wheat area are infested with Sunn Pest. Generally, about 75% of the cereal production area is under threat of attack in Turkey (7).

The total sprayed area was 973,181 ha and 1,882,893 ha in 1997 and 2003 respectively. The duration of the outbreaks and the area treated has gradually increased (9).

# Sunn Pest Control Program in Turkey

#### **Surveys and Treatment Activities**

Before chemical control is applied surveys are done to assess Sunn Pest population levels. Then Sunn Pest mortality is compared with previous years.

Counts are done in certain overwintering areas in the spring and autumn of each year to assess the populations for the following year. Results are compared with those of previous years. The migration from hibernation areas to the field is observed to determine the spring population density of Sunn Pest in the field. For this, one or two overwintering areas are sampled. The starting date of migration from overwintering areas to fields is determined by observing the activity of the pest when the maximum daily temperature reaches or exceeds 15°C, usually in March and April. Successive counts are done from the beginning of migration until 90% of the overwintered population has left the overwintering area.

Surveys are conducted by trained personnel to determine areas for treatment. At the end of the surveys, if the population level of overwintered adults is equal to or exceeds 0.8 insects per square meter, an egg parasitoid survey is conducted. If the percentage of parasitoid eggs is low chemical control is used. Chemical control begins when the percentage of 2nd instar nymphs in the population reaches 40%. However, if chemical control can't be completed during this period, the insecticide applications may be continued through the 4th or 5th instars. Chemical application for young nymphs begins 5 to 6 d after eggs hatch in infested areas (4).

#### **Pesticide Applications**

Pesticide application is a delivery process of agents to control populations of organisms that endanger the viable production of a crop. These agents have to reach the target with as little waste as possible by means of an efficient application. If the intended target is missed there is a hazard of human poisoning or environmental contamination (1). Proper adjustment of equipment and selection of product is critical for satisfactory performance. It can be assumed that about 50% of pesticides used are wasted by bad application (11).

Usually liquid pesticides are applied to large areas of crops by air for Sunn Pest management in Turkey. Until 2000 applications have been conducted using planes rented from the Ministry of Agriculture and Rural Affairs. When there are irregular landscapes and small fields, ground sprayers have been used by farmers. In 2000 the Ministry began to gradually change aerial applications to ground applications. Farmers have been spraying their areas using ground equipment. They have been supplied with insecticides and equipment by the Ministry.

Table 1. Growing area, production and yield for wheat and barley in Turkey from 1995 to 2002

		Wheat		Barley			
			Yield			Yield	
Years	Area (ha)	Production (ton)	(kg/ha)	Area (ha)	Production (ton)	(kg/ha)	
1995	9,400,000	18,000,000	1,915	3,525,000	7,500,000	2,128	
1996	9,350,000	18,500,000	1,979	3,650,000	8,000,000	2,192	
1997	9,340,000	18,650,000	1,997	3,700,000	8,200,000	2,216	
1998	9,400,000	21,000,000	2,234	3,750,000	9,000,000	2,400	
1999	9,380,000	18,000,000	1,919	3,650,000	7,700,000	2,101	
2000	9,400,000	21,000,000	2,234	3,629,000	8,000,000	2,204	
2001	9,350,000	19,000,000	2,032	3,640,000	7,500,000	2,060	
2002	9 300 000	19,000,500	2,101	3,600,000	8,300,000	2,307	

Between 1955-2000 most of areas of the country were sprayed by air (5). Some of the advantages of aerial applications are (i) the ability to cover large acreages rapidly (which may be critically important during pest outbreaks), (ii) the ability to treat fields during wet conditions and (iii) the elimination of soil compaction (no wheel traffic) caused during application. Over the years some economical, efficiency and environmental pollution problems have occurred.

The cost of application equipment is relatively high because of spare parts and repairs. Pilots should be sufficiently well informed on the pest, pesticide and other related subjects. Therefore training programs are needed and the establishment of an aerial application department is essential. In Turkey areas are not only flat but also with hills and valleys which are not suitable for aerial applications. This increases cost and risk. High voltage lines, telephone lines and other cables also cause insufficient treatments. Such areas form an important nucleus for Sunn Pest spread in the following years.

In general big, monoculture and planned crop blocks are suitable for aerial applications. But wheat fields are usually small and scattered in the country. Different crops exist dispersed among these fields. For example in the Thrace Region sunflower and Southeast Anatolia Region lentil fields are found between wheat fields. This causes low efficiency and use of excessive product.

Drift is undesirable for economic. environmental and safety reasons. Unsatisfactory pest control could result if a significant portion of the chemical is lost in drift. It could result in respraying. Recently spray drift incidents have increased to the extent that spray drift is cited as the number one problem for agricultural aviation (10). Aerial spraying remains the most highly wasteful, polluting practice still being used in agriculture today. An estimated 85 to 90% of the pesticide drifts of target and <1% is necessary to control the target insect. Pesticides can drift for as far as 80 km from the site of application depending on droplet size, nozzle type and size, spray height and weather conditions (2). Because of the topographic situation of the fields aircrafts cannot fly as low needed. This also causes drift.

In Turkey since small droplets are produced with ULV applications high drift has occurred. Small droplets have low inertial energy making them susceptible to drift. The effect of aerial applications can be seen in different regions of the country. Southeast Anatolia Region provides 15% of Turkey's cereal production and in this region egg parasitoids have a large effect on Sunn Pest populations. Today these parasitoids are scarce. In other regions depending on the year, spray applications were timed to avoid peak of parasitism.

Over the years, while problems of aerial application are increasing, they were needed to place pesticide on target with uniform distribution, good penetration of the canopy and low drift. One of the most important factors controlling deposition within the canopy is droplet size. Suitable droplets with maximize deposition on the pest, improve the level of control, reduce pesticide costs and minimize ground contamination. For these reasons usage of ground sprayers is seen as an alternative to aerial sprayers. Areas for Sunn Pest management by different application methods from 1997-2003 are shown in Figure 1 (9). There is about a 50% increase in the usage of ground sprayers. In 2003, 55% of the total treated area was spayed by ground sprayers.

There are also some problems with ground sprayers. There may be an incorrect setting of equipment, wrong direction of the spray, uneven distribution over the target areas which may leave untreated areas or an overdose of material in other spots of the crop. Calibration of sprayers is very important and education in this regard is needed in Turkey. Only a few technical staff has a clear idea about the correct use of pesticide equipment.



Figure 1. Treated area against Sunn Pest between 1997-2003 in Turkey.

Some equipment that is being used by farmers is unsafe and of poor quality. They are built by farmers, village mechanics or small manufacturers in the absence of safety standards. On the other hand, while the importance of ground spraying is growing some sprayer manufacturers are now producing new models which meet the needs of local farmers (suitable boom length, efficient pump, stable boom etc.) In addition, unsafe and insufficient tractor equipment is a hazard for the environment.

With ground applications there is a drift problem similar to aerial applications. Drift can range from 5 to 60 m depending on treatment conditions (3). If Turkish farmers were educated about boom height, nozzle pattern and type, flow rate and ground speed then pesticides would be applied correctly.

# Conclusions

Since 1955 all pesticide applications for Sunn Pest management were aimed to increase the biological efficiency and reduce spraying costs and environment pollution in Turkey. Both treated areas and populations of Sunn Pest show that aerial applications were insufficient. Pesticides were wasted; they cover nontargeted parts of plants, or drift outside the crop by wind or suspended in the air or dropped into sources of water or were deposited on the ground. As a result of this environment pollution occurs and the overall impact of natural enemies is reduced. Using ground sprayers for Sunn Pest control is efficient. But more research and educational programs are required. Information supporting production of suitable equipment is needed to optimize pesticide delivery under country conditions.

# الملخص

إرمان، أرزو، ياسمين صباح أوغلو، نيمان بابار أوغلو ورمضان جانهلال. 2007. الرش الجوي بالمقارنة مع الرش الأرضى لإدارة حشرة السونة. الصفحات 209–213.

تعتبر محاصيل الحبوب من المحاصيل الأكثر أهمية للاستهلاك الداخلي والتصدير في تركيا. يزرع القمح والشعير على مساحة 13,765,000 هكتار. إن استخدام مبيدات الحشرات هي الطريقة الشائعة للوقاية من حشرة السونة. طبقت معاملات مبيدات الآفات السائلة عن طريق الجو من قبل الحكومة حتى عام 2000. بوجود الطرائق الجديدة، سيقوم الزراع برش حقولهم باستخدام المرشات الأرضية. من أجل رش أكثر فعالية وتلوث أقل للمناطق غير المستهدفة، قام هذا البحث باختبار ميزات وسلبيات الرش الجوي بالمقارنة مع الرش الأرضي.

كلمات مفتاحية: رش جوي، رش أرضى، إدارة، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Effects of Insecticides on Sunn Pest Egg Parasitoids, Trissolcus grandis Thomson and T. semistriatus Nees

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#### Abstract

Saber, M. 2007. Effects of Insecticides on Sunn Pest Egg Parasitoids, *Trissolcus grandis* Thomson and *T. semistriatus* Nees. Pages 215-219.

Sunn Pest, *Eurygaster integriceps* Puton is the most important pest of wheat and barley in Iran. Effects of fenitrothion and deltamethrin, the most commonly used insecticides in Iran for controlling the pest, were assessed on the Sunn Pest egg parasitoids *Trissolcus grandis* Thomson and *T. semistriatus* Nees. The field recommended concentrations of the insecticides caused 100% mortality of adults within 24 h. LC<sub>50</sub> values of fenitrothion and deltamethrin for *T. grandis* and *T. semistriatus* were 16.2, 7.9 and 155 and 145.6 ppm, respectively. Life table parameters of adult females exposed to LC50s of the insecticides within the first 24 h after emergence were evaluated. Longevity and reproductive capacity of females were not significantly reduced for both species. The sex ratio of the progeny of both parasitoids was not affected significantly. The intrinsic rates of increase of *T. grandis* were 0.298, 0.289 and 0.286 and for *T. semistriatus* 0.297, 0.293 and with 0.295 female offspring per female per day in the control, fenitrothion and deltamethrin treatments, respectively.

Keywords: egg parasitoids, insecticides, scelionid, side effect, Sunn Pest, Trissolcus spp.

#### Introduction

Sunn Pest, Eurygaster integriceps Puton is the most important pest of wheat and barley in Iran and other middle eastern countries. Egg parasitoids (Scelionidae) are the most important biological control agents of Sunn Pest (8). Early attempts to achieve biological control using these parasitoids were never successful owing to difficulties of rearing them under controlled conditions and problems in getting them established in agricultural fields due to the widespread applications of broad spectrum insecticides (3). It is estimated that egg parasitoids reduce *E. integriceps* populations by ca. 23% yearly in Iran (1). Fenitrothion and deltamethrin are the most commonly used insecticides to control E. integriceps in Iran (1, 11). There are a few detailed studies on the effects of conventional insecticides on Sunn Pest egg parasitoids (5, 7, 11, 13).

As part of our efforts to develop an IPM program for wheat and barley in Iran, we are especially concerned with side effect of

commonly used insecticides on *Trissolcus* grandis Thomson and *T. semistriatus* Nees, scelionid egg parasitoids of *E. integriceps*.

#### **Materials and Methods**

#### **Cultures and Insecticides**

The *T. grandis* and *T. semistriatus* colonies used in all experiments originated from overwintering adults collected in October 1999 from a cherry orchard in Fashand-Karaj, Iran. The wasps were reared on *E. integriceps* eggs for two generations in a growth chamber at  $25\pm1^{\circ}$ C,  $60\pm10^{\circ}$  RH and a photoperiod of 16:8 (L: D). Adult wasps were provided with honey as a food source. The second-generation parasitoids (F2) were used in all experiments.

The insecticides used were fenitrothion (Fenitrothion 50EC, Gyah Co., Iran) and deltamethrin (Decis, 2.5EC, Hoechst, AgrEvo).

#### Lethal Concentration Bioassay

A stock solution at a concentration of 500 ppm was prepared for each insecticide. Aliquots

were taken from the stock solution and mixed with water to prepare 6 concentrations determined after an initial dose setting experiment. Fifteen glass plates (14×15 cm) were sprayed with 1 ml of each insecticide dilution at 14 mbar using a Potter Spray Tower (Burkard Mfg. Co. Ltd. Uxbridge, UK). Control plates were sprayed with distilled water. The plates were placed in the laboratory for 1 h and allowed to dry. The exposure cages were assembled after drying the plates. The exposure cages consisted of a frame and 2 glass plates as floor and ceiling. The frame was  $13 \times 14$  cm long, 2.5 cm high, and 0.5 cm wide and was fitted with an adhesive tape with foam material to provide pads for the two glass plates. Each of the 4 sides of the frame contained six ventilation holes 1.5 cm in diameter. The inside surface of the frame was lined with black and tight net to cover the holes. One of the holes was used to introduce honey on strips of paper as food for the parasites. Before completely assembling the cages, 30 female adults (<24 h old) were anesthetized with CO<sub>2</sub> and placed in each exposure cage. The number of dead and live wasps in each cage was counted 24 h after initial exposure to the insecticide residue. The data were analyzed using PROC PROBIT procedures (10) to compute LC10, LC50 and LC90 values on a standard and log scale with an associated 95% fiducial limit.

#### **Life Table Parameters**

Adult wasps (<24 h old) were exposed to LC50 concentrations of the insecticides for 24 h. Then twenty-five randomly chosen alive females were transferred individually to a Plexiglas cage ( $16 \times 10 \times 5$  cm). The cages were transferred to a growth chamber maintained as stated above. Each female was presented 3 *E. integriceps* fresh egg masses (42 eggs) and honey as food on a strip of white paper.

The egg masses were changed daily until the female died. The parasitized eggs were stored at  $25\pm1^{\circ}$ C,  $60\pm10\%$  RH with a photoperiod of 16:8 (L: D) and allowed to emerge for 18 days. The total numbers of

eggs, the numbers of black eggs (parasitized eggs) and emerged wasps, the sex of emerged wasps and the number of eggs containing dead adults were recorded. Emergence, sex ratio, survival, longevity and fecundity data were analyzed by analysis of variance (ANOVA) with mean separation at the 5% level of significance by the Fisher protected least significance difference (LSD) using SAS. Daily schedules of mortality and fecundity were integrated into a life table format (2) and used to calculate net reproductive rate (R0), mean generation time (T), and intrinsic rate of increase (rm) (Table 1). The Jackknife technique was used to calculate the variance of rm and other life table parameter estimates (6).

#### Results

#### LC50 Bioassay

The LC<sub>50</sub> values based on formulated insecticides indicated that fenitrothion was more toxic than deltamethrin for the egg parasitoids *T. grandis* and *T. semistriatus* (Table 1). According to the amount of active ingredient per ml solution, there was no difference in toxicity between the insecticides (Table 1).

#### Life Table Parameters

Sublethal effects of the insecticides on life table parameters of *T. grandis* and *T. semistriatus* are summarized in Tables 2 and 3.

The insecticides did not significantly affect the mean longevity of *T. grandis* (F=0.52; df=2, 27; P=0.6) and *T. semistriatus* (F=2.62; df=2, 57; P=0.82). Analysis of the reproductive activity of females exposed to  $LC_{50}$  for 24 h, revealed significant treatment effects on the rate of parasitization for *T. semistriatus* (F=6.29; df=2, 57; P=0.003) but did not for *T. grandis* (F=0.5; df=2, 27; P=0.612) (Tables 2 and 3). The mean number of female offspring per female did not differ significantly compared to the control for *T. grandis* (F=0.2; df=2, 27; P=0.81) and

*T. semistriatus* (F=0.58; df=2, 57; P=0.56) (Tables 2 and 3). The sex ratio of the progeny was not significantly affected for *T. grandis* (F=0.07; df=2, 27; P=0.93) and *T. semistriatus* (F=0.41; df=2, 57; P=0.67) (Tables 2 and 3). The intrinsic rates of increase of *T. grandis* 

were 0.298, 0.289 and 0.286 and *T. semistriatus* were 0.297, 0.293 and 0.295 female offspring per female per day in the control, fenitrothion and deltamethrin treatments, respectively (Tables 2 and 3).

**Table 1.** Toxicity of fenitrothion and deltamethrin to the adult egg parasitoids *Trissolcus grandis* and *T. semistriatus*.

Insecticide/			Lethal concentrations (ppm) or [µg a.i./ml]				
Parasitoid species	n	Slope±SE	LC10 (95% FL)	LC50 (95% FL)	LC90 (95% FL)		
fenitrothion		•	× /	```'			
T. grandis	800	6.01±0.45	(9.9 (8.8-10.8)) [4.9 (4.4-5.4)]	(16.2 (15.4-16.8)) [8.1 (7.7-8.4)]	(26.4 (24.8-28.6)) [13.2 (12.4-14.3)]		
T. semistriatus	960	8.42±0.68	(5.6 (5.2-5.9)) [2.8 (2.6-2.9)]	(7.9 (7.7-8.1)) [3.9 (3.8-4.1)]	(11.2 (10.6-12.2)) [5.6 (5.3-6.1)]		
deltamethrin							
T. grandis	600	4.15±0.53	(76.3 (60.6-7.9)) [1.9 (1.5-2.2)]	(155 (145.6-166.9)) [3.9 (3.6-4.2)]	(315.8 (276.2-414.9)) [7.89 (6.9-10.4)]		
T. semistriatus	560	5.31±0.62	(83.6 (70.8-93.1)) [2.1 (1.8-2.3)]	(145.6 (138.3-153.8)) [3.6 (3.5-3.8)]	(253.7 (225.5-304.4)) [6.3 (5.6-7.6)]		

Lethal concentrations and 95% fiducial limits (FL) were estimated using logistic regression (10).

**Table 2.** Sublethal effects of the insecticides on life table parameters of *T. grandis* exposed to LC50 value of fenitrothion and deltamethrin.

Treatment	Longevity (day)	Mean no. of progeny per female (Mx)	Mean no. of female progeny per female (mx)	Sex ratio m/(m+f)	Intrinsic rate of increase (rm)
fenitrothion	33.7±4.9 a	168.3±10.87 a	102.2±14.5 a	0.35±0.09 a	0.289
deltamethrin	31.8±4.1 a	168.1±19.20 a	94.7±09.9 a	0.39±0.05 a	0.286
Control	38.6±5.4 a	185.4±10.30 a	105.8±12.9 a	0.37±0.08 a	0.298

Means within a column followed by different letters are significantly different (Fisher protected least significant difference (LSD), *P*=0.05).

**Table 3.** Sublethal effects of the insecticides on life table parameters of *T. semistriatus* exposed to LC50 value of fenitrothion and deltamethrin.

Treatment	Longevity (dav)	Mean no. of progeny per female (Mx)	Mean no. Of female progeny per female (mx)	Sex ratio m/(m+f)	Intrinsic rate of increase (rm)
fenitrothion	31.5±2.3 a	239.65±8.6 a	117.8±12.55 a	0.35±0.09 a	0.293
deltamethrin	28.1±2.5 a	206.60±7.9 b	103.4±09.20 a	0.39±0.05 a	0.295
Control	36.3±2.8 a	192.60±11.9 b	105.1±08.90 a	0.37±0.08 a	0.297

Means within a column followed by different letters are significantly different (Fisher protected least significant difference (LSD), *P*=0.05).

# Discussion

Fenitrothion and deltamethrin were both highly toxic to adult T. grandis and T. semistriatus (Table 1). Neither of these insecticides is compatible with Trissolcus adults. Overall, these studies suggest that insecticide applications should be avoided early in the season when adult parasitoids migrate to wheat and barley fields. Based on dose-response results, LC50 values were about 1/100 of the field concentration of formulated fenitrothion and 1/3 of the field concentration of deltamethrin (Table 1). The use of deltamethrin may be preferred to fenitrothion. When most of the parasitoids are in the preimaginal stage, they may be subject to minimal adverse effects of the insecticides (9).

Sublethal effects of insecticides (4) can have profound effects on surviving parasitoids (12). In the present study, female adult longevity, fecundity, number of female offspring per female and sex ratio of offspring were used to assess sublethal effects of the insecticides.

Longevity of adult females of both parasitoids was not significantly affected (Tables 2 and 3). Also total number of female offspring per female was not affected significantly by fenitrothion and deltamethrin in both parasitoid species. The insecticides had significant effects on fecundity of *T. semistriatus* (Table 3). It seems that fenitrothion had an hormolygosis effect on *T. semistriatus* and increased fecundity by 24.4%

compared to the control (Table 3). Sex ratio of progeny of both parasitoids was not affected significantly by insecticides but it was malebased. The reduced numbers of males in the fenitrothion and deltamethrin treated populations could have an impact on the subsequent parasitoid generation. Fewer males may result in a reduced number of mated females thereby reducing the number of female progeny produced and lowering the population's reproductive potential. Α preponderance of male progeny produced in T. grandis insecticide-treated and Τ. semistriatus populations is an important finding, and previously was reported by Novozhilov et al. (7) and Smilanick et al. (12). Insecticide-induced distortion of sex ratio (in favor of males) not only slows the recovery of the parasitoid population following treatment, but under certain circumstances it may lead to local extinction (12). More attention should be devoted to field experiments to more clearly determine the effects of insecticides on egg parasitoids under agricultural conditions.

## Acknowledgments

We thank Dr. Masoud Amir Moafi and Dr. Aziz Sheikhi for help in analyzing data. The technical assistance of A. A. Hassani and M. Safari is greatly appreciated. The facilities of the Sunn Pest Research Department of the Plant Pest and Disease Institute (IRAN) were used to conduct this study.

# صابر، موسى. 2007. تأثيرات مبيدات الحشرات على طفيليات بيض حشرة السونة Trissolcus grandis صابر، موسى. Tomson و Trissolcus grandis. الصفحات 215-219.

تعد حشرة السونة (Eurygaster integriceps Puton) من أهم الآفات الحشرية على القمح والشعير في إيران. تم تقييم تأثيرات مبيدي الحشرات فينيتروثيون ودلتامثرين (الأكثر استخداماً لمكافحة الآفة في إيران) على طفيليات بيض حشرة السونة Trissolcus grandis Thom. و Trissolcus grandis Nees. سببت التراكيز الحقلية المنصوح بها من مبيدات الحشرات نسبة موت T. grandis ضمن 24 ساعة. بلغت قيم LC50 للفينيتروثيون والدلتامثرين له 24 ساعة التي الم و 100% للبالغات ضمن 24 ماعة. إلمانيون، على التوالي. تم تقييم مؤشرات جدول الحياة للإناث البالغة التي عرضت لـ LC<sub>50s</sub> من مبيدات الحشرات ضمن الـ 24 ساعة الأولى بعد الانبثاق. لم تنخفض طول فترة الحياة والقدرة التكاثرية للإناث بشكل معنوي عند كلا النوعين. ولم نتأثر النسبة الجنسية معنوياً عند نسل كلا نوعي الطفيليات. بلغت نسبة الزيادة الطبيعية في النوع T. grandis (0.298 وفي النوع 0.286 وفي النوع 0.297 T. semistriatus ومع 0.295 نسل

أنثى لكل أنثى في اليوم في معاملات الشاهد، فينيتروثيون، ودلتامثرين، على التوالي.

كلمات مفتاحية: طفيليات بيض، مبيدات حشرات، Scelionid، تأثير جانبي، حشرة السونة، .Trissolcus spp.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Chemical Control with Emulsifiables in Water using ULV Ground Applications

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#### Abstract

Al-Gamali, N.A. and A.A. Al-Kafaji. 2007. Chemical Control with Emulsifiables in Water using ULV Ground Applications. Pages 221-224.

This study was carried out in wheat field at Al-Qush region (Mousl province). It used new pesticides called Beticol 20% SL, Trebon 7.5% ULV, Ofunack 25% ULV, Cyperlod 7.5% ULV with concentrate 0.25 cm<sup>3</sup>/L to every one, compared to sumialpha 2.5% EC and Diazinon 60% EC with concentrate 1 cm<sup>3</sup>/L. Results showed that all pesticides was high efficiency against the Sunn Pest (*Eurygaster integriceps* Puton). The killing percent was between 89.26-95.60% in the first year and 96.54-98.52% the second year. **Keywords:** ULV applications, Sunn Pest, ground spraying.

#### Introduction

Wheat was the first among grain crops and the most important elements of economics in the world. One of the important factors limiting the productivity of wheat was Sunn Pest, Eurygaster integriceps Puton (Scutelleridae: Hemiptera), which also attack barley, oats, and rye. It also sometimes attacks corn and sorghum (10). Sunn Pest became economically important in Northern Iraq in 1985; it is controlled every year by using insecticides. In 1991, it was found in most wheat planting regions in Iraq but under economic injury levels (1). Since 2002 it has become an economic pest in other regions in Middle Iraq especially in Dewania & Najaf provinces where its control is absolutely necessary to reduce economic damage (2). The infestation intensity of this insect is different from region to region in Iraq. The population density was higher in the north compared with middle regions, and was non economic pest in southern regions. The insect behavior during hibernation and aestivation is different in the middle regions compared to the northern regions of Iraq; in the middle regions it migrates to orchards close to fields and under non irrigated deciduous trees (4) or between the base fronds in the top of palm trees (3). While in the northern regions it migrates to mountains at elevations of 750-2300 m (6). Chemical method is still used against this pest in Iraq and in the world. In Syria, they use the pesticides Diptrex, Zolon, Decis, Sumithion and Fastac (8); while in Turkey they use Dursban, Lebaycid and Actellic and they are conducting research on other pesticides as Fenitrothion, Cypermethrin, Cyfluthrin etc. (10). Specialists confirmed that the pesticides Sumi-alpha, Fastac, Decis, Karate, Sinoratox and Onefon were the most used in Romania with about 91-100% efficiency (9). In Iran, Sumithion is used on a large scale in addition to Dursban, Zolone and Actellic (7). In Iraq, we have used Diptrex, Lebaycid, Sumithion, Dursban, Karate and Nogos (1, 5, 11) and Chemocidin, Vapcocidin and Decis (A.A. Al-Khafaji and N.A. Al-Gamali, unpublished data). The objective of this study was to evaluate the efficacy of pesticides which belong to different chemical groups applied as an emulsion in water using ground spraying systems.

# **Materials and Methods**

This study was conducted in a wheat field in Al-qush region, 60 km north of Mosul, using following pesticides Ofunck the (Pyridaphenthion) 25% ULV, cyperlod (Cypermethrin) 7.5% ULV. Beticol (Acetamiprid) 20% SL & Trebon (Etofenprox) 7.5% Each pesticide was applied using ULV and compared to Sumi-alpha (Esfenvalerat) 2.5% EC and Diazinon 60% EC Treatments, and control (only water), were distributed in a randomized complete design (RCD), four replicates to every treatment. The treatment area was equal to one donum (1000 sq. m or 0.247 acres) Spraying was done after maximum egg hatch using a knapsack sprayer with as capacity of 18 liters. Estimation of Sunn Pest density was conducted by using a sweep net, 20 sweeps /one replicate. Sunn Pest populations were determined this way one day before spraying and 1, 3, 7 days after spraying. The experiment was repeated for two years consecutively. Results were analyzed statistically. The relative efficacy of pesticides was calculated by using Henderson-Telton equation.

# **Results and Discussion**

The average number of Sunn Pest declined on the first day after spraying to about 1.2 insects/20 sweeps on day 6 compared to 20.5-21.0 insects/20 sweeps before spraying, while the average of insect numbers in control treatment (water only) increased from 18.2-24.2 insects/20 sweeps (Table 1). Basically the same trend occurred in year (Table2).

Table 1. The average number of Sunn Pest./20 sweeps before & after spraying (first year).

	Average number of	Average number of Sunn Pest after spraying (days)		
Treatment	Sunn Pest prespray	1	3	6
Ofunack 25% ULV	20.5 a	8.8 b	4.5 c	1.2 b
Cyperiod 7.5% ULV	23.0 a	7.2 b	4.2 c	1.5 b
Beticol 20% SL	22.5 a	12.2 b	7.5 b	2.8 b
Trebon 7.5% ULV	22.8 a	9.5 b	4.5 cb	1.5 b
Diazinon 60% EC	21.0 a	10.8 b	8.5 b	3.0 b
Control	18.2 a	22.2 a	24.5 a	24.2 a
LSD at 0.05 probability level	5.020	5.29	3.67	3.29

Numbers in columns followed by like letters are not significantly different

Table 2. The average number of Sunn Pest./20 sweeps before & after spraying (second year).

	Average number of	Average number of Sunn Pest after spraying (days)			
Treatment	Sunn Pest prespray	1	3	6	
Ofunack 25% ULV	32.25 a	9.50 b	4.00 b	1.50 b	
Cyperiod 7.5% ULV	35.75 a	8.75 b	4.25 b	1.50 b	
Beticol 20% SL	33.50 a	8.50 b	3.75 b	0.75 b	
Trebon 7.5% ULV	31.75 a	9.00 b	4.00 b	0.75 b	
Sumi-alpha 2.5% EC	30.25 a	9.25 b	5.50 b	1.50 b	
Control	28.75 a	32.50 a	38.25 a	41.25 a	
LSD at 0.05 probability level	11.061	6.797	6.658	3.075	

Numbers in columns followed by like letters are not significantly different

Tables 1 and 2 showed significant differences between treatment (pesticides) and control at 0.05 probability level, but it was not significant among pesticides. The results indicate that the pesticides tested had a high efficiency against Sunn Pest; mortality 6 d postapplication was 95.60, 95.10, 90.64 and 95.05% for Ofunack, Cyperlod, Beticol and Trebon respectively in the first year (Table 3). But in the second year it was 96.76, 97.08, 98.52 and 98.44%, respectively (Table 4). The results showed also that control pesticides Sumi-alpha and Diazinone were not different than other used pesticides, they gave high efficiency, 96.54 and 89.26% respectively after 6 days (Tables 3 and 4). In conclusion, we can use Ofunack, Cyperlod, Beticol and Trebon against Sunn Pest because of their high efficiency and they can be applied as emulsifiables in water from the ground of by ULV from the air. Spraying must target Sun Pest nymphs and adults when they are present in fields and conserve egg parasites.

**Table 3.** The percent mortality of Sunn Pest/dayspostapplication using Henderson equation (firstyear).

	Percent mortality of Sunn Pest (days)					
Treatment	ent <u>1 3 6</u>					
Ofunack 25% ULV	64.81	83.69	95.60			
Cyperiod 7.5% ULV	74.34	86.44	95.10			
Beticol 20% SL	55.55	74.58	90.64			
Trebon 7.5% ULV	72.36	85.34	95.05			
Deazinon 60% EC	57.84	69.33	89.26			

 Table 4. The percent mortality of Sunn Pest/days

 postapplication using Henderson equation (second year).

	Percent mortality of Sunn Pest (days )					
Treatment	1 3 6					
Ofunack 25% ULV	73.74	90.68	96.76			
Cyperiod 7.5% ULV	78.35	91.07	97.08			
Beticol 20% SL	77.56	91.59	98.52			
Trebon 7.5% ULV	74.93	90.53	98.44			
Sumi-alpha 2.5% EC	72.95	86.33	96.54			

#### الملخص

الجمالي، أ. ناصر وعبد الستار أ. الخفاجي. 2007. المكافحة الكيميائية بمبيدات قابلة للاستحلاب في الماء باستخدام مرشات ULV أرضية. الصفحات 221–224.

نفذت هذه الدراسة في حقل قمح في منطقة الكوش (محافظة الموصل)، حيث استخدمت مبيدات الآفات الجديدة التالية: 0.25 (ULV)، Cyperlod (ULV)، 25 Ofunack (ULV)، 25 Trebon (SL)، 20 Beticol سم<sup>2</sup>/ليتر لكل مبيد، بالمقارنة مع EC) (EC) (EC) و 60 Diazinon (EC) بتركيز 1 سم<sup>2</sup>/ليتر. أظهرت النتائج أن جميع مبيدات الآفات كانت عالية الفعالية ضد حشرة السونة (EC) و Eurygaster integriceps Puton). تراوحت نسبة القتل بين 29.56-89.26% في العام الأول و 26.54-96.52% في العام الثاني. كلمات مفتاحية: تطبيقات ULV، حشرة السونة، رش أرضي.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.





# Egg Parasitoids of Sunn Pest in Turkey: A Review

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## Abstract

#### Koçak, E. 2007. Egg Parasitoids of Sunn Pest in Turkey: A Review. Pages 227-235.

Sunn Pest is the most important pest of wheat and barley in Turkey, and approximately 75% of the cereal areas are under threat of this pest. Species of *Trissolcus* Ashmead, 1893 are the most effective egg parasitoids of Sunn Pest. The natural effect of *Trissolcus* sp. can reach 100% mortality in some localities and that's why spray applications are avoided. *Trissolcus* sp. exhibit differences in distribution, density, effectiveness according to the geographic areas they are found in. Much research on *Trissolcus* sp. distribution, taxonomy, biology-bioecology, hosts, mass rearing, releasing, effectiveness, and side effects of insecticides has been done in Turkey. Results of this work are presented herein. **Keywords:** *Eurygaster*, *Trissolcus*, egg parasitoids, Sunn Pest.

#### Introduction

Sunn Pest, Eurygaster sp., is the most important pest of cereals, especially wheat, in Turkey. The first outbreaks were recorded in 1927-1929 and subsequently 1939-1941, 1955-1959, 1965-1973 and 1977- to date. First chemical control application was applied to 24.660 ha in 1955 and in the following years the sprayed area increased consistently and reached approximately a half million ha in 1985. Approximately 1.9 million ha were sprayed in 2003, mostly by aerial application. Aerial applications negatively affect the egg parasitoids and natural balance because of drift. The egg parasitoid Trissolcus sp. has a more important role than any other natural enemy of this pest. Sunn Pest control strategies are based on egg parasitoids except in Southeastern Anatolian region in Turkey. In some years no spraying is done against the pest because of the parasitoid activity, especially in south, west and northwest regions. For example, approximately fifty percent of the cereal areas were not sprayed because of egg parasitoid activity in Western Anatolian region between 1987 and 1997 (4). In the world, the first biological control application was done with T. vassilievi Mayr against Eurygaster sp. in 1903. In Turkey, the first study of Trissolcus sp. was done in 1928 (41). Because the pest is very important and their parasitoids have a noteworthy effect on control of this pest, forty three different research manuscripts were studied and reviewed for this paper.

#### Literature Review

#### **Faunistic Studies**

The first study of *Trissolcus* sp. was started by Zwölfer (41) who determined that T. semistriatus Nees and T. vassilievi were present in Adana Province. Seventeen Trissolcus sp. parasitizing Eurygaster eggs have been identified (Table 1). T. manteroi Kieffer and T. djadetshko Rjachovsky have been recently recorded as new specimens on Sunn Pest eggs in Turkey (16). A total of 2493 parasitized Eurygaster egg batches were collected across Turkey. The most common effective egg parasitoid was and *T*. semistriatus (69.3%) followed by T. simony Mayr (16.9%), T. grandis Thomson (9.4%), T. vassilievi (3.6%), T. pseudoturesis Rjachovsky (0.4%), T. djadetshko (0.1%) and T. manteroi (0.04%)(17).

*Trissolcus semistriatus* and *T. simony* were obtained from each of the 76 egg batches of the 2493 egg batches collected. Five *Trissolcus* sp. were identified from the same egg masses: *T. semistriatus*, *T. simony*, *T. grandis*, *T. vassilievi* and *T. pseudoturesis* (18).

Species*	Regions	Reference
15, 17	Mediterranean	41
15, 17	Mediterranean, Aegean, East and Southeastern Anatolia	21
7, 11, 15, 17	Mediterranean and Southeastern Anatolia	9
12, 15	Central Anatolia	2
2, 3, 7, 12, 15, 16, 17	Mediterranean and Southeastern Anatolia	39
7, 11, 17	Mediterranean and Southeastern Anatolia	3
2, 15, 17	All Regions	21
1, 14	Aegean	21
4	East and Southeastern Anatolia	21
7, 12, 16	Southeastern Anatolia	21
3, 15, 17	Mediterranean	27
15, 17	Southeastern Anatolia	31
7, 11, 13, 14	Thrace	1
7, 8, 10, 15, 16	Thrace	23
2, 3, 15, 17	Central Anatolia	25
7, 14, 15, 16, 17	Mediterranean	33
2, 3, 15	Thrace	26
2, 6, 10, 12, 15	Mediterranean	34
2, 7, 12, 14, 15	Aegean	4
6, 12, 15, 16	Mediterranean	37
15	All Regions	17
7	All Regions except Blacksea Region	17
17	East and Southeastern Anatolia, Central Anatolia, Mediterranean	17
16	Mediterranean, Aegean, Marmara, Blacksea and Central Anatolia	17
10	Mediterranean, Aegean, The Marmara and Central Anatolia	17
12	Central and Southeastern Anatolia	17
5	Mediterranean and Central Anatolia	17
9	Central Anatolia	17

**Table 1.** The distribution of the Sunn Pest egg parasitoid *Trissolcus* sp. (17 species) according to regions in Turkey.

\* 1) Trissolcus anitus (Nixon); 2) T. basalis (Wollaston; 3) T. choaspes (Nixon); 4) T. culturatus (Mayr); 5) T. djadetshko (Rjachovsky); 6) T. festivae (Viktorov); 7) T. grandis (Thomson); 8) T. histani (Voegele); 9) T. manteroi (Kieffer); 10) T. pseudoturesis (Rjachovsky); 11) T. reticulatus (Delucchi); 12) T. rufiventris (Mayr); 13) T. rungsi (Voegele); 14) T. scutellaris (Thomson); 15) T. semistriatus (Nees); 16) T. simony (Mayr); 17) T. vassilievi (Mayr)

#### **Taxonomy and Morphology**

There has been only one study on *Trissolcus* taxonomy and morphology. This was done on *T. semistriatus, T. grandis, T. simony, T. vassilievi, T. pseudoturesis, T. rufiventris, T. djadethsko,* and *T. manteroi,* all Sunn Pest egg parasitoids. An identification key was prepared with descriptions and scanning electron microscopic photographs to show specific morphological characters (19).

Some *T. semistriatus* specimens have morphological abnormalities such as shortened and fused antenna segments from the third

flagellum segment only in males; a separated thorax in two parts from the middle with vertical convex shape in two males and one female. In male as the right antennal segment is normal shape, the first two segments of the left antennal flagellum are fused and thus there appears to be 9 flagellum segments instead of 10. In another male both flagellum segments are fused from third flagellum segments, so the flagellum segments seem as eight segmented instead of 10 (15).

#### **Biology and Bioecology**

The first studies on biology and bioecology in Turkey were carried out in Adana Province (42). Many studies have been done to determine adult longevity, parasitization capacity, emergence ratio, sex ratio, developmental period, developmental threshold and thermal requirements (Table 2).

Trissolcus semistriatus on Sunn Pest eggs in the Southeastern Anatolia region has 2 (32) or 3 generations (21); in the Çukurova region there are 3 generations on Sunn Pest but a total of 16 generations on other hosts per year are possible (42); 9 generations with other hosts in the Thrace region (11), and 2 generations on E. maura L. in the Central Anatolia region (25, 30). The parasitoid overwinters under the bark of different trees. Trissolcus sp. are seen in nature; from March 18 in zmir, March 20 in Adana, and April 15 in Divarbakır (21). The temperature at the time of appearance was ~15 C in the Thrace region (38). Sunn Pest eggs and the parasitoids were found together when the mean temperature was 14 C and rH 55.5% in the Aegean region (4). Trissolcus sp. were found one week before the appearance of Sunn Pest when the mean temperature was 16.9 C and rH 42.3% in the Central Anatolia region (24). In the Southeastern Anatolia region, they became active 22 days before Sunn Pest when the mean daily temperature was 13 C and they moved to cereal fields on May 7 to parasitize Sunn Pest eggs after existing on various weeds. They left the cereal fields in the yellow ripening stage of wheat. Then the parasitoids migrated to their overwintering sites, generally in October, when the average temperature fell to 10 C (29).

#### Studies on Effectiveness of Trissolcus sp.

*Trissolcus* sp. generally parasitizes approximately 80 eggs (Table 2). Low temperatures increase adult longevity but decrease the parasitoid egg numbers (11). *Trissolcus semistriatus* parasitized an average 6.8 (range 0-33) *E. maura* eggs, and a maximum parasitisation of an average 12.6 were realized in the first three days of oviposition (24); *T. semistriatus* parasitized an average 6.7 (range 0-27) *E. integriceps* eggs, with a maximum parasitisation of an average 10.5 were realized in the first two days (11). Depending on the increasing temperature, female longevity, length of the oviposition period, egg blackening and development duration of *T. semistriatus* on the Sunn Pest eggs decreased (37).

As a result of high parasitisation of Sunn Pest eggs and low adult populations, nymphal populations have been under the economic threshold (6-10 nymph/m<sup>2</sup>) and no chemical applications have been performed in the Thrace region (22). If overwintered adult densities were 0.8, 1.0 and 1.5 adults/m<sup>2</sup> and the parasitism rates were 40%, 50% and 70%, respectively, then parasitoids would suppress Sunn Pest (27). Maximum Sunn Pest nymphal density was 6 nymphs/ $m^2$  when the parasitation rate was 86% (32). When there was 65.7% egg parasitism and 1 Sunn Pest adult/m<sup>2</sup>, the egg parasitoids were effective in controlling the pest population (33). However it was concluded that when the Sunn Pest adult number was 1.6/m<sup>2</sup>, 66.7% parasitation rate was not sufficient to keep the pest population under the economic threshold (30). The number of overwintered adults was found to be 2.3 and  $3.5/m^2$  and the rates of parasitized eggs were 56.8-100% respectively; so the pest population was kept under the economic damage level of 10 nymph/m<sup>2</sup> in the Thrace region in 1990-1991 (26).

In Turkey, Sunn Pest control in the Southeastern Anatolia region now places emphasis on the protection of egg parasitoids until 40% of the nymphs reach  $2^{nd}$  instar (29). When 71.4 and 51.8% of Sunn Pest were  $2^{nd}$  instar on barley and wheat respectively, the percentage of the first generation parasitoid emergence was 66.7% on barley and 50.0% on wheat in the Central Anatolia region (30). Parasites depended on suitable wooded areas within 1500-3000 m from the wheat fields to be used as hibernation and aestivation sites (31).

Table 2.	Biological	data on	Sunn	Pest	egg	parasitoid,	Trissolcus	spp.

Species/ Hosts	Optimal temperature and RH conditions in laboratory (L) or field (F)	Adult longe- vity (Day) Male/ Female	No. of eggs parasit- Ized	Adult emer- gence (%)	Sex ratio Male/ Female	Develo- pment period (days) egg to adult Male - Female	Develop- ment threshold (°C)	Thermal requir- ement (day degrees)	Refer- ence
Trissolcus sem	istriatus								
C. pudicus	26±0.5°C 60±10% RH (L)	-	87.3	95.9	0.30/0.70	-	-	-	12
D. baccarum	26±0.5°C 60±10% RH (L)	-	80.0	98.2	-	10.1-11.6	-	-	13
D. baccarum	26±0.5°C 60+10% RH (L)	-	83.6	97.6	0.40/0.60	-	-	-	12
E. integriceps	24.6°C 67.5% RH (L)	18.4-25.5	101.5	88.5	0.47/0.53	-	-	-	11
E. integriceps	22.6°C 55.1% RH (L)	20.8-37.8	57.4	95.3	0.83/0.17	-	-	-	11
E. integriceps	26±0.5°C 60±10% RH (L)	-	88.0	98.6	0.40/0.60	-	-	-	12
E. integriceps	26±0.5°C	-	90.0	100.0	-	9.7-10.9	-	-	13
E. integriceps	20.4°C (L) 27°C (F)	-	90.0	-	0.20/0.80	12.0-16.0	-	-	21
E. integriceps	18.0-34.0°C (L)	-	-	-	-	-	11.79	138.8	37
E. integriceps	18-20°C (L)	-	-	-	-	30.0-34.0	15.5	123	42
E. ornatum	26±0.5°C 60+10% RH (L)	-	28.5	71.9	-	12.5-13.3	-	-	13
E. maura	26±1°C 65+5% RH (L)	8.5-12.4	85.4	60.4	0.42/0.58	14.7-12.4	-	-	24
E. ornatum	26±0.5°C 60±10% RH (L)	-	24.0	45.8	0.80/0.20	-	-	-	12
G. lineatum	26±0.5°C	-	94.8	100.0	0.60/0.40	-	-	-	12
G. lineatum	26±0.5℃	-	82.5	97.2	-	9.5-11.6	-	-	13
H. vernalis	60±10% KH (L) 26±0.5℃	-	80.8	98.6	0.70/0.30	-	-	-	12
H. vernalis	60±10% RH (L) 26±0.5°C 60±10% RH (L)	-	86.5	95.7	-	10.5-11.7	-	-	13
Trissolcus sim	onv								
C. pudicus	26±1°C 60±10% RH (L)	-	84.0	-	0.20/0.80	9.6-10.8	-	-	14
D. baccarum	26±1°C 60±10% RH (L)	-	81.6	-	0.50/0.50	9.2-10.3	-	-	14
E. integriceps	$26\pm1^{\circ}C$	-	86.8	-	0.30/0.70	12.1-12.4	-	-	14
G. lineatum	60±10% RH (L) 26±1°C 60±10% RH (L)	-	82.8	-	0.20/0.80	11.3-11.5	-	-	14
<b>Trissolcus vass</b> E. integriceps	<i>ilievi</i> 20.4°C (L)	-	28.0	-	-	-	-	-	21

Parasitized *E. maura* eggs were 200-300 m from trees (25). Parasitoid activity changed according to regions and especially to climatic factors. Polyculture farming positively affected parasitoid activity (10).

In the laboratory, an average of 75 *E. integriceps* eggs was parasitized by *T. semistriatus* females during their average of 26 days longevity (35). For succesfull parasitization 1-3 days old eggs of appropriate hosts was important (13).

#### Hosts, Mass Rearing and Releasing

The first study on alternate hosts was done by Zwölfer (43). Alternate hosts are vitally important relevant to the effectiveness and population development of Trissolcus sp. Twenty two alternate hosts have been identified. These include Aelia acuminata L., A. rostrata Boh., Carpocoris iranus Tam., C. fuscispinus Boh., C. mediterraneus Tam., C. pudicus (Pd.), Codophila pusio Kol., Dolycoris baccarum (L), Eurydema festivum L., E. ornatum L, Eysarcoris inconspicuus (H.S.), Graphosoma italicum (Muell.), G. lineatum L., G. semipunctata F., G. stali Horv. Holcostethus vernalis (Wolff.), Nezara viridula L., Odontotarsus plicatulus Horv., O. purpureolineatus (Rossi), Piezodorus lituratus F., Psacasta exanthematica (Scopoli) and Raphigaster nebulosa (Pd.) (1, 11, 21, 29, 36, 43).

Lodos (21) recorded that T. semistriatus parasitized Nezara viridula L. eggs in the laboratory. However others reported that N. viridula eggs were not parasitized by and not a host to T. semistriatus (12). T. semistriatus was reared successfully from C. pudicus, D. baccarum and E. ornatum (11). Eurydema ornatum was not a suitable host for mass production of T. semistriatus because of the low rate of parasitism, adult emergence and a long developmental time. It was suggested that G. lineatum was the most suitable host and D. baccarum the second. Although C. pudicus and *H. vernalis* were also suitable hosts, mass production of these species was more difficult than others (12, 13).

*Trissolcus semistriatus* prefers *E. integriceps* eggs to *E. ornatum* eggs (11). *Eurydema ornatum* is the most reliable host for the mass production of Sunn Pest egg parasitoids (34). However, in another study, about mass rearing of *T. semistriatus*, it was determined that *E. integriceps* had more hopeful values according to *E. ornatum* (7).

Using field-collected eggs whereas T. rufiventris preferred Aelia eggs to Eurygaster eggs (2), T. semistriatus preferred Eurygaster eggs to Aelia eggs in the Central Anatolia region (5). The study to determine the preferences of T. semistriatus in terms of host species and age of various heteropteran host eggs (E. integriceps, D. baccarum, G. lineatum, E. ornatum, H. vernalis) indicated that a small portion of parasitism of E. ornatum eggs was obtained (28.5%), although high parasitism rates were found in other host eggs (90.0-80.0%). A high percentage of adult emergence was also recorded. When tested for its ability to parasitize host eggs of different ages, T. semistriatus was found to prefer younger hosts: it showed parasitism rates greater than 50% with up to 3 days old E. integriceps and D. baccarum, and with up to 4 days old G. lineatum and H. vernalis. However, the parasitism rates in all the ages of E. ornatum eggs were much lower than for other host species. The developmental times in all host species at different ages was extended with increased host age. It was concluded that G. lineatum and D. baccarum could be used for mass production of egg parasitoids (13). In another study, parasitism rates of E. integriceps, D. baccarum L., G. lineatum L. and C. pudicus (Pd.) averaged 86.8%, 81.6%, 82.8% and 84.0%, respectively. The parasitoid sex ratio and the percent of adult emergence did not differ significantly among the four hosts. The average development period was shorter in D. baccarum and C. pudicus, with respective mean times of 10.3 and 10.8 days for females, and 9.2 and 9.6 days for males, than in E. integriceps and G. lineatum. According to these results, E. integriceps,

# D. baccarum, G. lineatum and C. pudicus are adequate hosts for T. simony (14).

For mass rearing of T. semistriatus, the embrional development of the host eggs (E. *integriceps* and *E. ornatum*) were inhibited by exposing them to -20 C for 1, 2 and 3 h before the parasitisation process. When the host eggs were stored for a month at 7 C, the parasitisation rates were found to be ~ 60% for E. integriceps and 3.3% for E. ornatum, respectively. Pupal storage of the parasitoids is not convenient (7). When D. baccarum eggs were stored at -18 C for 5-245 days, parasitization rate and adult parasitoid emergence of *T. grandis* from the eggs were >50%. The longer the eggs were stored, the longer the parasitoid development and emergence period, but the fewer female parasitoid emerged. As a result of storage, the female and male longevities shortened 10 and 6 days, respectively (20).

Parasitoid release studies were started in the Thrace region. The first releases were unsuccesful because of incorrect timing. Egg parasitoids should be released before the main oviposition period of Sunn Pest, because old eggs are not preferred by female parasitoids (13). After first observing Sunn Pest in the field, releases should be made as soon as possible. Information on mass production and releasing of parasitiods is available (6). In another report, T. semistriatus was released for management of E. integriceps at four population densities (650, 1300, 1950, 2600/da) in wheat fields of Gaziantep Province. In 1950, there was an 8-16% increase in T. semistriatus populations on first generation on Sunn Pest eggs on one decare of wheat (37).

#### The Side Effects of Insecticides

The first study on the side effects of insecticides was done in 1985 (27). They

determined the residual effects of insecticides on Trissolcus adults. All insecticides tested killed parasitoid adults in 0.5-4.5 h after application. The side effects of Cypermethrin, Cyfluthrin, Deltamethrin, Cyhalothrin and Fenthion, commonly used in Sunn Pest control, on pupae (spraying and dipping) and adults (spraying) of Trissolcus sp. were determined. The insecticides inhibited (25.9 -93.0%) emergence of adult parasitoids. Deltamethrin showed the highest effect, Cyfluthrin and Cyhalothrin had the least effect on parasitoid emergence. Each insecticide when sprayed directly on parasitoid adults killed 100% of them (40). Similar results were reported (except Deltamethrin) when Cypermethrin, Cyfluthrin and Fenthion were sprayed on the larval stage of the parasitoid. Deltamethrin caused 29.16% mortality, and the others 56.2 - 67.7%. According to IOBC evaluation categories (8), the insecticides were grouped as Deltamethrin was harmless and others were slightly harmful. Each insecticide effected parasitoid development in the eggs (10).

It is recommended that mass production facilities capable of producing billions of parasitoids be constructed. Research on the use of artificial eggs should be initiated. Efforts should be made to preserve and enhance parasitoid overwintering sites by planting trees, organizing polyculture farming, and supporting parasitoids in fields with introductions of other species like *T.vassilievi* which is more adapted to hot and arid areas as in Southeastern Anatolia and *T. simony* which is better adapted to hot and moist areas as in Çukurova.

#### Acknowledgment

I wish to thank Birol Akbas (PPCRI-Ankara) for his contributions to the text.

#### الملخص

# كوجاك، ارهان. 2007. طفيليات بيض حشرة السونة في تركيا: نظرة عامة. الصفحات 227-235.

تعد حشرة السونة من أهم الآفات على القمح والشعير في تركيا، وتقريباً 75% من مناطق الحبوب واقعة تحت تهديد هذه الآفة. تعتبر أنواع الجنس Trissolcus Ashmead من أكثر أنواع طفيليات بيض حشرة السونة فعالية. يمكن أن يصل التأثير الطبيعي لأنواع . Trissolcus sp. إلى نسبة موت 100% في بعض المواقع، ولهذا السبب يتم تجنب تطبيق الرش. تبدي أنواع Trissolcus sp. اختلافات في التوزع، الكثافة، الفعالية تبعاً للمناطق الجغرافية التي توجد فيها. أجريت في تركيا بحوث كثيرة عن توزع، تصنيف، حياتية-بيئية، عوائل، التربية الكمية، إطلاق، وفعالية أنواع . الحشرات فيها. سوف يتم عرض نتائج هذا العمل في هذه الورقة.

كلمات مفتاحية: Eurygaster، طفيليات بيض، حشرة السونة.

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# Progress in the Development of a Mycoinsecticide for Biological Control of Sunn Pest

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#### Abstract

# Edgington, S., D. Moore, H. Kutuk, H. Satar and M. El Bouhssini. 2007. Progress in the Development of a Mycoinsecticide for Biological Control of Sunn Pest. Pages 237-243.

The Sunn Pest complex causes significant losses of yield and quality of wheat and barley in West Asia. One potential control strategy is to use entomopathogenic fungi, such as *Beauveria bassiana* (Balsamo) Vuillemin, as mycoinsecticides to control both the overwintering adults and the summer generation in the crop. However, success with a mycoinsecticide requires the optimization of a series of steps, beyond the biological efficacy of the fungal pathogen. Summer generation trials in Syria and Turkey in 2003 and 2004 have resulted in significant progress in understanding aspects of the pest/pathogen interaction that will result in effective control. One trial at ICARDA in 2004 resulted in up to 93.5% mortality of nymphs that were field treated then lab-maintained (compared with 35% of control nymphs). Implications for the development of a mycoinsecticide are discussed.

Keywords: Beauveria bassiana, isolate, mycoinsecticide, oil formulation, Sunn Pest, ULV application.

#### Introduction

A collaborative project is developing control measures against the Sunn Pest complex. One aspect being examined is the use of an entomopathogenic fungus, Beauveria bassiana (Balsamo) Vuillemin (Deuteromycotina: potential Hyphomycetes) as а mycoinsecticide. Isolates of Beauveria bassiana had been obtained from overwintering adults (15). One of these was mass produced to a high quality specification and, in the summer of 2003, oil based formulations of this isolate were applied to mature wheat crops in Turkey and Syria using an Ultra Low Volume hand-held sprayer. Despite good coverage of both crop and insect, no significant adult mortality, attributable to B. bassiana, was obtained.

At the time of the trials, samples of summer populations of adult Sunn Pest were collected, transferred to the UK and, as the insects died, they were examined for pathogens. Seven isolates of *B. bassiana* were obtained and laboratory bioassays at ICARDA confirmed their virulence to Sunn Pest (William Reid, *pers. comm*). One isolate was selected, based on virulence, ease of production and temperature tolerance. The effectiveness of this isolate for controlling summer populations of Sunn Pest was then examined. This paper summarizes the results of a preliminary set of trials conducted at ICARDA in April 2004 and discusses requirements for the successful development of a mycoinsecticide.

#### Materials and Methods

#### Fungal Isolate

Seven strains of *Beauveria bassiana* obtained from summer generation Sunn Pest samples collected from Syria and Turkey in June 2003 were placed in the Genetic Resource Collection at CABI Bioscience UK. One isolate (IMI 391044) was subsequently grown in a two-stage system (10, 11), the first stage of which was production of *B. bassiana* biomass in liquid culture; the second stage used rice as a solid substrate to promote
conidiation. Once the moisture content of conidiated rice was <10% the conidia were harvested using a specialized cyclone harvester (the "Mycoharvester" see www.mycoharvester.info). Conidia were then dried over silica gel to 5% moisture content, then hermetically sealed in tri-laminate aluminum-foil sachets and stored at 4 °C until the investigation began.

### **Mortality Assessments**

The trials were conducted during April and May 2004 at ICARDA, located approximately 40 km south of Aleppo. An area of wheat (*Triticum aestivum* L. var. Fadda 98) of approximately 5 ha was selected, within which 36 rectangular plots (20 m x 50 m) were marked out in 4 groups of 9 plots. The wheat had been sown on 13th December 2003 at a row spacing of 20 cm, and had received 2 fertilizer treatments of ammonium nitrate (November 2003 and February 2004) and 2 herbicide treatments of Topik then Duplosan (March 2004). Five plots received *B. bassiana* while five received a blank control.

Locally purchased kerosene and sunflower oil were mixed on a 1:1 basis to provide the formulating medium. Conidia of B. bassiana were added at a concentration of 5 x  $10^9$  conidia /ml. The control treatment consisted of the formulating medium but no conidia. Both treatments contained the fluorescent tracer Lumogen® (BASF) at 3%. The tracer enabled spray efficiency to be determined; the material fluoresced under UV light showing where droplets had impacted on insect or vegetation. Spraying was carried out using a hand-held Micron-Ulva + spinning disc sprayer (Micron, Herefordshire, UK), with 6 D-Cell batteries and a red restrictor. Treatments were applied at a rate of 2 L /ha with the 5 control plots sprayed first. Spraying took place on 18th April and was repeated on 29<sup>th</sup> April, commencing after 1700 h on both occasions to avoid high sunlight intensities and gusty winds. Adult Sunn Pest were collected 18 h after the first spray. Immediately prior to the second spray the

plots were seeded with Sunn Pest nymphs collected from untreated areas of wild wheat, attaching the wild ears on which the nymphs were residing, onto ears within the trial plots. Nymphs were then collected immediately after spraying. One assessor collected from control plots, another collected from B. bassiana plots, to avoid contamination. Sunn Pest were maintained in ventilated plastic boxes (15 x 10 x 5 cm) in the laboratory at room temperature (30°C max., 26°C min.), with a supply of untreated fresh wheat ears. They were examined daily to determine mortality, and every 4 days the wheat was changed. Dead Sunn Pest were surface sterilized in baths of isopropanol (50%) and NaOCl (1%) then washed three times in sterile distilled water. They were placed onto sterilized paper toweling (Kimwipe<sup>TM</sup>), moistened with sterile distilled water and incubated in 9 cm Petri dishes at 27±2°C. The dishes were examined daily for 5-10 days for B. bassiana infection.

### **Spray Coverage**

Sunn Pest collected from each *B. bassiana* treated plot, following the first spray, were examined under UV light to determine spray coverage before being incorporated into the mortality assessments. Droplet counts were made on the dorsal and ventral surfaces and on combined legs + antennae. Two days after the first spray, 20 wheat tillers were collected from each *B. bassiana* plot and the ear and flag leaf examined under UV to determine droplet coverage. Deposition on the wheat was ranked, with 0 =none, 1 =low, 2 = medium and 3 = high.

### Statistics

Mortality data and spray coverage results were arcsin transformed prior to analysis in order to improve homogeneity. Student's *t*test was used in evaluating the results. The means and SE displayed in the tables and figures are based on data prior to transformation.

### Results

#### **Mortality Assessments**

After 5 days incubation, following the first spray, mortality ( $\pm$  SE) of adult Sunn Pest was 15.0±4.18% from B. bassiana exposure, against 23.0 $\pm$ 6.82% from the controls (t = 0.9, df. 8, n.s.). After 10 days incubation mortality rose to 100% from B. bassiana exposure, against 97.0 $\pm$ 2.0% from the controls (t = 0.1, df. 8, n.s.). Final mortality (after 5 days) of nymphs collected after the second spray was 93.5±3.73% from B. bassiana exposure and 34.9±10.63% from controls, the difference was highly significant (t = 5.33, df. 8, p < 0.01) (Table 1). There were no visible signs of mycosis on dead adults from B. bassiana plots following the first spray. Whilst there were signs of mycosis on some of the dead nymphs following the second spray, it was impossible to quantify as many of the cadavers were quick to decompose and/or too small to surface sterilize sufficiently well.

**Table 1.** Summary of Sunn Pest mortality (%) following two applications of a *B. bassiana* and control spray ( $18^{th}$  and  $29^{th}$  April 2004). Figures are shown at 5 and 10 days after the first spray and 5 days after the second (when the study was terminated). The insects were maintained in ventilated sandwich boxes at room temperature.

		Mortality% (± SE)		
Sunn Pest	Incubation (days)	B. bassiana	Control	
Spray 1				
Adults	5	$15.0{\pm}4.18$	$23.0\pm6.82$	
Adults	10	$100\pm0.00$	$97.0 \pm 2.00$	
Spray 2				
Nymphs	5	93.5±3.73	34.9±10.63	

### **Spray Coverage**

Droplet coverage of Sunn Pest following the first spray can be seen in Table 2. The proportion of insects receiving direct drops was  $59.0\pm2.45\%$ . The concentration (mean  $\pm$  SE) of drops on the dorsal surfaces was  $2.7\pm0.48$  /insect, significantly higher than on the ventral or legs/antennae surfaces (t = 4.65

and 4.60, df. 8, p <0.05). The ventral and legs/antennae surfaces of each insect received on average only  $0.39\pm0.12$  and  $0.43\pm0.116$  drops respectively (t = 0.25, df. 8, n.s.). Droplet coverage of the wheat can be seen in Figure 1. All of the tillers examined had droplets. The mean level of coverage was 'low' for both ear and flag leaf.

 Table 2.
 Summary of droplet coverage on Sunn

 Pest collected immediately after the first spray.

	Number of drops (mean /insect ± SE)	
Treatment	B. bassiana	
Insect hit ( $\% \pm SE$ )	59.00±2.45	
Dorsal	2.70±0.48 a	
Ventral	0.39±0.12 b	
Legs + antennae	0.43±0.12 b	

Figures with different letters were significantly different (p < 0.05)



Figure 1. Proportion of sampled tillers with 'none', 'low', 'medium' and 'high' droplet coverage of ear and flag leaf, 48 h after *B. bassiana* spray. Data combined from all sprayed plots.

### Discussion

The results following the second spray suggest that there may be potential for developing B. *bassiana* as a control for summer generation Sunn Pest. If there is good coverage of the insect and the wheat, as was partially demonstrated by the fluorescent tracer, this potential would be enhanced. The trials were

only preliminary though, with mortality assessments carried out in the controlled/artificial conditions of a laboratory. Hence, there is clearly a need for further trials, in particular trials based solely in the field.

The planning and execution of the study gave some fundamental information necessary for mycoinsecticide research in the field, helping to illustrate the step-wise nature of developing an effective mycoinsecticide. Since the 1990s and principally since the development of a project on the biological control of locusts and grasshoppers, known as LUBILOSA (12), there has been a greater appreciation of how interactions between the host, pathogen, environment and application strategy can effect the development of epizootics within insect populations (2, 12). Focus on just one element, for example the isolate, will usually result in failure. Each component, and any interactions between the major components needs optimization to develop a product which will be biologically successful (at which point, commercial requirements become critical). To illustrate this, the findings from the trial at ICARDA will be discussed in the context of the development of a mycoinsecticide.

### Isolate

Detailed screening incorporating a number of parameters is imperative to isolate selection (14). Selecting an isolate solely on virulence can lead to variability in results that can mycoinsecticide ultimately impede development. Other features to consider include stability, production characteristics, sporulation capabilities and persistence. Interactions with environmental factors, such as solar radiation, temperature and relative humidity can dramatically influence entomopathogen efficacy (9). Tolerance of the mycoinsecticide to sub-optimal conditions will be of critical importance in the field, pest particularly as control by an entomopathogenic fungus is dose-related.

Since the inception of this project a number of overwintering and summer isolates,

obtained from Sunn Pest, have shown good virulence in laboratory bioassays (15). Unfortunately when applied to summer wheat fields, little or no infection has been recorded. The only exception was significant mortality following the second spray in this present investigation, even this is no evidence of field efficacy as insects were examined under controlled laboratory conditions. Molecular characterization of B. bassiana isolates obtained from overwintering Sunn Pest showed no significant relationships between fungus and insect (1). Therefore, would a summer isolate be more adapted to summer generation Sunn Pest, having successfully attacked a presumably healthy individual? Furthermore, would such an isolate be more suited to the environmental conditions during the summer (1)? These questions need to be examined more closely.

### **Isolate and Formulation**

It would be advantageous to have the isolate persisting in the crop for a significant time. Isolates vary in their environmental longevity (8), with persistency critical for maximizing host kill. If no persistent isolates are available, formulation can improve field efficacy by protecting against factors such as desiccation and UV radiation. The addition of sunscreens and/or the type of carrier oil may, for example, increase UV tolerance of fungal conidia (6). Genetic manipulation and specific production systems can help increase the ecological fitness of conidia and in doing so provide increased tolerance to a wider range of RH and temperature (7, 16).

#### **Isolate and Application**

Some of the fungus will be destroyed by UV, washed away by rain or lost by the insect through preening and moulting (4). Protection of the mycoinsecticide could be increased by applications of more conidia at lower levels in the crop, protecting against the effects of UV irradiation. This could, for example, involve the use of a motorized mist blower (13). Depositing spray on the underside of the

leaves would also offer some protection against UV as well as rainfall, as would timing the application of the spray to avoid the sunniest, hottest and/or wettest parts of the day. Spray timing could also be coordinated with periods of least moulting in the life cycle of the insect.

### **Isolate and Ecology**

There is surprisingly little quantified information on the Sunn Pest and its behaviour in the crop, particularly in English (but see 3 and 5). From what is available, there is evidence to suggest that the majority of Sunn Pest spend the hottest times of the day away from the top of the crop, below the level of the flag leaf and often at the very base of the tiller Circumstantial evidence (3).from observations made during this present study suggests that the insects are quite active during the day. There were no observations made at night and there appears to be no information on night behaviour. How often will Sunn Pest climb up and down the crop in a given time? The more active the insect is, the greater the exposure of the insect to spray deposits. If direct spray hit on the insect causes 90% mortality then secondary uptake may not be necessary, but if this is not the case then secondary uptake from sprayed plant surfaces becomes crucial. Hence, so does isolate persistency.

Isolate IMI 391044 clearly displayed significant levels of virulence, but the susceptibility of adult and nymph stages may be variable. Only adults were collected after the first spray and there was no significant mortality from the *B. bassiana* treatment. Only nymphs were collected after the second spray and mortality was significant. The lack of adult mortality may have been an experimental artifact, or an indication that the adult stages were less susceptible to the isolate vis a vis nymphs because of, for example, body size or are adults just more difficult to kill? Adults do have very thick cuticles and most spray lands on the dorsal surface, although secondary uptake is likely through the ventral surface. Other vital biological data is lacking. What is the behaviour of a Sunn Pest infected with an entomopathogen? Does it move to the top of the crop (and be more vulnerable to predation by birds) or maybe to the bottom of the crop (and be more vulnerable to predation by ants)? Does it eat and move less, lay down less fat and not be able to migrate? Can the Sunn Pest bask in the sun and control the progress of the disease?

A successful move from this period of research to one of product development will require a greater understanding of any factors limiting fungal efficacy. Obtaining 93.5% kill from a preliminary trial, all be it under (part) artificial conditions, does provide some indication that an effective mycoinsecticide product could be a possibility (12). Nevertheless there is a need for considerable amounts of fieldwork to build on the results of 2004, together with an optimization of both product and usage. With background knowledge on mycoinsecticide development and good field results, it is feasible that a practical product could be developed in 3-4 years, but skimping on the development studies will result in failure.

Since these preliminary trials in April 2004, additional summer treatments have been carried out at ICARDA and these will be described in subsequent papers.

### Acknowledgements

The authors wish to thank Ziad Sayyadi and his co-workers at ICARDA for their help in the field, and Emma Thompson (CABI, Ascot, UK) for mass production of the isolate. William Reid (University of Vermont) gave invaluable assistance without which the work could not have been accomplished. This publication is an output from a research project funded by the United Kingdom Department for International Development (DFID). The views expressed are not necessarily those of DFID (R7863 Crop Protection Programme). With fond memory of Hatice Satar.

### الملخص

إيدينغتون، إس.، ديف مور، خليل كتك، هـ. ساتار ومصطفى البوحسيني. 2007. التقدم في تطوير مبيد حشرات فطرى من أجل المكافحة الحيوية لحشرة السونة. الصفحات 237–243.

يسبب معقد حشرة السونة خسائر هامة في غلة ونوعية القمح والشعير في غرب آسيا. إحدى استراتيجيات المكافحة الممكنة هي استخدام فطور ممرضة للحشرات مثل Balauo) Vuillemin (Balsamo) كمبيدات حشرات فطرية لمكافحة كل من البالغات المشتية والجيل الصيفي على المحصول. ولكن يتطلب نجاح مبيد الحشرات الفطري تحسين سلسلة من الخطوات علاوة على الفطري. أحدثت نتائج تجارب مكافحة الجيل الصيفي في سورية وتركيا خلال الخطوات علاوة على الفعالية الحيوية للممرض الفطري. أحدثت نتائج تجارب مكافحة الجيل الصيفي في سورية وتركيا خلال الخطوات علاوة على الفعالية الحيوية للممرض الفطري. أحدثت نتائج تجارب مكافحة الجيل الصيفي في سورية وتركيا خلال عامي 2003 و 2004 تقدماً هاماً في فهم أوجه تفاعل الأفة/الممرض الذي سيعطي مكافحة فعالة. أعطت نتائج إحدى التجارب في إيكاردا عام 2004 نسبة موت وصلت حتى 93.5% من الحوريات التي تمت معاملتها في الحقل ثم حفظت في المخبر. (مقارنة بنسبة موت حريات الشاهد ك5%). في هذه الورقة سيتم مناقشة مضامين تطوير مبيد حشرات فطري. ولمائلة من معاملتها في الحقل تلال معنوي مكاردات الذي سيعطي مكافحة فعالة. أعطت نتائج إحدى التجارب في إيكاردا عام 2004 نوعالة موت وصلت حتى 93.5% من الحوريات التي تمت معاملتها في الحقل ثم حفظت في المخبر. (مقارنة بنسبة موت حوريات الشاهد 35%). في هذه الورقة سيتم مناقشة مضامين تطوير مبيد حشرات فطري. كمائلة مفارية أولي ميائلة منائلة مولي ميد حشرات فطري. مستحضر زيتي، حشرة السونة، تطبيق ULV.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Infectivity of Steinernema feltiae Filipjev (Rhabditida: Steinernematidae) to Eurygaster maura L.

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### Abstract

Koçak, E., A. Gokce and I. Kepenekci. 2007. Infectivity of *Steinernema feltiae* Filipjev (Rhabditida: Steinernematidae) to *Eurygaster maura* L. Pages 245-249.

Entomopathogenic nematodes as *Steinernema* (Rhabditida: Steinernematidae) and *Heterorhabditis* (Rhabditida: Heterorabditidae) are important biocontrol agents and their usage has recently increased. *Eurygaster maura* L. is one of the most important pests of cereal in Turkey. Two *Steinernema feltiae* Filipjev isolates were tested against adult *E. maura* in laboratory conditions. Each isolate was tested at [25, 50 and 100 infective juveniles (IJs)/0.2 ml water per adult] and at 10, 15 and 25°C. The mortality was recorded after 72 h and 96 h of each incubation. Mortality of *S. feltiae* (All type); at 96 h and a dose of 100 IJs/0.2 ml water per adult was 50% at 10°C; 58.3% at 15°C and 63.8% at 25°C; for *S. feltiae* (S3); 58.3% at 10°C and 15°C, and 74.9% at 25°C. Mortality was lower at the other doses. When the temperature rises, mortality also rises. These results indicated that entomopathogenic nematodes may be used to control *E. maura*. **Keywords:** Entomopathogenic nematodes, *Eurygaster maura*, *Steinernema feltiae*.

### Introduction

Sunn Pests are serious damagers of cereal throughout the Balkans, the Middle East and the former USSR (3). *Aelia* (Pentatomidae) and *Eurygaster* (Scutelleridae) are the two important genera within these families. *Eurygaster integriceps* Puton and *E. maura* L. (Hemiptera: Scutelleridae) are the most important and common species in Turkey (16, 19).

Sunn Pest control is based on activity of the egg parasitoids which are the only natural enemies of this insect in Turkey. If they can not suppress the pest then chemical control is started.

Entomopathogenic nematodes can be very effective biological control agents against a number of insect pests and possess several advantages over chemical pesticides (11). They can actively find their hosts, can recycle in the soil environment (11), and are environmentally safe (1).

The economic importance of entomopathogenic nematodes (EPNs) of the families Steinernematidae and Heterorhabditidae is increasing as a result of intensive studies of their potential as biological control agents (2, 17). Species in these two families have been tested on various insect hosts belonging to different orders (5, 9, 23). Nematodes search for a suitable insect host, enter not only natural openings but also through the cuticle. After entering the host they release their symbiotic bacteria, Xenorhabdus and Photorhabdus into the haemolymph (6). Proliferation of the bacteria leads to death of the insect within 24-48 h, followed by nematode development and reproduction (10).

Sunn Pest suffers high levels of natural mortality and this has encouraged research on the use of these natural enemies as biological control agents. The natural enemies of Sunn Pest include a range of predators, including the carabids, *Pterostichus*, *Calosoma*, *Bembidion* and *Pterostichus*, egg parasitoids, belonging to genera *Trissolcus* (Hymenoptera), microorganisms, *Beauveria bassiana* (Balsamo) Vuillemin, *Bacillus thuringiensis*, and nematodes, *Mermis* spp. (3, 18, 20).

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It was reported that the nematodes caused about 16% Sunn Pest mortality in the overwintering sites in Ankara province (21). Promising results in controlling *E. integriceps* using entomopathogenic nematode *Steinernema carpocapsae* (Weiser) has also been reported (4). Pathogenicity of the entomopathogenic nematodes *S. carpocapsae* (Anamur), *Heterorhabditis bacteriophora* Poinar (Tur-H1), *H. bacteriophora* (Tur-H2) was tested on *E. maura* and the mortalities were 55%, 69% and 95% respectively (15).

The first *Steinernema* record of *S. feltiae* was from Rize province on the Black Sea coast and *H. bacteriophora* was first found in a population of stink bug, *A. rostrata* Boh. in Turkey (12, 22).

The goal of the present study is to ultimately extend the entomopathogenic nematode successes noted in Europe and the U.S. to Turkey. The first step in achieving this end has been achieved by the isolation of several indigenous species and strains (7, 8, 13, 15, 22, 25). We report the virulence of two isolates of *S. feltiae* from Turkish soil against Sunn Pest adults at different concentrations and temperatures.

### **Materials and Methods**

Entomopathogenic nematodes and wax moth larvae, *Galleria mellonella* L., were obtained from stock cultures maintained at the Plant Protection Central Research Institute of Ankara. *S. feltiae* (All type), used in the experiments was isolated from soil samples taken from the Black Sea coast of Turkey (22). The other *S. feltiae* (S3) isolate was obtained from soil samples taken at the Agricultural Faculty Campus in Ankara. (14, 25). Nematodes were reared on last-instar wax worms at 25°C according to methods of Woodring and Kaya (27). After harvesting the nematodes were stored at  $5\pm1^{\circ}$ C for 2 weeks.

*E. maura* adults were collected from an aestivating area in Haymana (Ankara, Turkey). The soil was clay loam with the percentage of sand: silt: clay =27%: 44%: 29%

respectively. The pH =7.7 and organic matter =1.22% by weight. The soil samples were taken where the insects were collected (soil from the aestivating area of the pest). The soil was autoclaved and then dried at room temperature.

Experiments were done in plastic plates which had 12 wells, 2.2 cm dia., and 2 cm deep according to the procedure described by Shapiro et al. (24). One *E. maura* adult was placed at the bottom of each well and then each well was filled with approximately 7-8 cm<sup>3</sup> autoclaved soil.

series of Α three nematode concentrations, 25, 50 and 100 infective juveniles (IJs)/0.2 ml water, was prepared from the stock suspension by serial dilution. The sand-well bioassay method was used in dose-mortality experiments. Each well contained 7-8 cm<sup>3</sup> loan sand soil. Sunn Pest adults were selected at random and placed individually to the 2 mm depth of the soil in each well. Each nematode isolate was tested on 36 adults at dose rates of 25, 50 and 100 IJs per insect. Nematodes were applied in 0.2 ml sterile-distilled water to each of 12 wells onto 7-8 cm<sup>3</sup> of soil (repeated 3 times for 12 replicates). For the control, 0.2 ml sterile distilled water was applied. The plates were labeled and incubated at 10, 15 and 25°C for 72 h and 96 h. Dead insects were recorded after 72 h and later 96 h. Dead insects after 96 h were incubated individually at 25°C by the "White traps" technique (26).

### **Results and Discussion**

The effectiveness of two entomopathogenic nematode isolates, *S. feltiae* (All type) and *S. feltiae* (S3) used against Sunn Pest adults in laboratory conditions is shown in Figures 1 and 2.

The results were found hopeful when 50 and 100 nematodes were exposed to each adult. Mortality of adults has been increased and the highest mortality was observed at 25°C. When different entomopathogenic nematode isolates were taken into consideration the highest mortality was observed when adults were infected with S3. S3 was more effective than A11 type. The mortality was recorded separately after 72 h and 96 h of each incubation. The following results of mortality at 96 h and a dose of 100 IJs/0.2 ml water per adult were obtained for *S*. *feltiae* (All type); 50.00% at 10°C; 58.30% at  $15^{\circ}$ C and 63.86% at 25°C; for *S. feltiae* (S3); 58.30% at 10°C and 15°C, and 74.96% at 25°C. Mortalities were lower at the other doses. When the temperature rises, the mortality also rises.

Although many studies with EPNs against economically important pest groups have been recorded, only two, *S. carpocapsae* and *H. bacteriophora* have been studied on *Eurygaster* spp. To date there has been no literature record about *S. feltiae* on *Eurygaster* spp.



Figure 1. The average mortality (%) of Sunn Pest adults exposed to infective juvenile *S. feltiae* (All type).



Figure 2. The average mortality (%) of Sunn Pest adults exposed to infective juvenile *S. feltiae* (S3).

The first study reported was with *S. carpocapsae* against *E. integriceps* and had promising results for management (4). The other study included *S. carpocapsae* (Anamur), *H. bacteriophora* (Tur-H1) and *H. bacteriophora* (Tur-H2). They caused Sunn Pest mortality of 55%, 69% and 95% respectively (15). The study reported herein with *S. feltiae* (All type) and *S. feltiae* (S3) supports previous work and indicates the potential of nematodes for control of *E. maura.* Field studies are now needed.

### Acknowledgement

We thank to Dr. N. Özer (Department of Biology, Faculty of Science, Hacettepe University, Ankara, Turkey) who supplied the nematode isolate *S. feltiae* (All type).

### الملخص

كوجاك، إرهان، أيهان غوكسي وآيلكير كيبينكسي. 2007. مقدرة النيماتودا الممرضة Steinernema feltiae كوجاك، إرهان، أيهان غوكسي وآيلكير كيبينكسي. 2007. مقدرة النيماتودا الصفحات 245–249. Eurygaster maura L. على إصابة حشرة Rhabditida: Steinernematidae) و Rhabditida: Steinernematidae) و Heterorhabditis

(Rhabditida: Heterorabditidae) من عوامل المكافحة الحيوية الهامة والتي ازداد استخدامها مؤخرا. تعتبر حشرة Eurygaster maura L. من أهم آفات الحبوب في تركيا. اختبرت عزلتين من Steinernema feltiae ضد بالغات حشرة E. maura في ظروف المختبر. حيث اختبرت كل عزلة بثلاثة تراكيز [25، 50 و 100 يرقة معدية (0.2/(IJs مل ماء لكل حشرة بالغة]، وعند ثلاث درجات حرارة (10، 15 و 25°س). سجلت نسبة الموت بعد 72 و 96 ساعة من كل تحضين. بلغت نسبة الموت التي سببتها S. feltiae (20 مل ماء للبالغة 50% عند درجة نسبة الموت التي سببتها S. feltiae (20 مل ماء للبالغة 50% عند درجات حرارة 00°س، 88.3% عند 50°س؛ أما عند (33) S. feltiae فبلغت نسة الموت 88.5% عند درحات حرارة 10°س، 88.3% عند 51°س و 86.3% عند درحات الحراة 10 و 15°س و 74.5% عند 25°س؛ أما عند (33) S. feltiae (33) فبلغت نسة الموت 88.5% عند درحات حرارة 10°س، 87.3% عند 51°س و 86.3% عند درحات الحراة 10 و 10°س، 88.5% عند 51°س و 86.3% عند درحات الحرارة 10 و 10°س، 88.3% عند 51°س و 86.3% عند درحات الحرارة 10 و 10°س، 88.5% عند 51°س و 86.5% عند درحات عند الحرارة 10 و 10°س، 88.5% عند 52°س في حين كانت نسبة الموت 10 و 10°س و 74.5% عند 52°س. في حين كانت نسبة الموت أخفض عند الجرعات الأخرى. ارتفعت نسبة الموت 10 و 10°س و 74.5% عند 52°س. في حين كانت نسبة الموت أخفض عند الجرعات الأخرى. ارتفعت نسبة الموت 10°س، 10 و 10°س و 74.5% عند 52°س. في حين كانت نسبة الموت أخفض عند الجرعات الأخرى. ارتفعت نسبة الموت 10°س، 10 و 10°س و 74.5% عند 52°س. في حين كانت نسبة الموت 10°س، 10°س و 74.5% عند 52°س. في حين كانت نسبة الموت أخفض عند الجرعات الأخرى. ارتفعت نسبة الموت عند ارتفاع درجة الحرارة. أشارت هذه النتائج إلى أنه يمكن استخدام النيماتودا الممرضة للحشرات لمكافحة حشرة E. maura عند ارتفاع ديمة يماتودا مرضة للحشرات، 10°س و 10°س، 20% عند 52°س. كلمات مفتاحية المعرض قد الحقرات مرضية للحشرات، 10°س و 10°س.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

### Survey of Syrian Soils for Entomopathogenic Nematodes and Efficacy vs. Sunn Pest

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### Abstract

Abdulhai, M., Z. Sayyadi, W. Reid and M. El Bouhssini. 2007. Survey of Syrian Soils for Entomopathogenic Nematodes and Efficacy vs. Sunn Pest. Pages 251-257.

Four laboratory trials were conducted using seven species of entomopathogenic nematodes (EPN). These included *Heterorhabditis indica* (Poinar, Kanunakar & David), *H. bacteriophora* (Poinar), *H. zealandica* (Poinar), *Steinernema riobravae* (Cabanillas, Poinar & Raulston), *S. carpocapsae* (Weiser), *S. glaseri* (Steiner), and *S. feltiae* (Filipjev). Adult overwintering *Eurygaster integriceps* were challenged with three concentrations of infective juveniles (IJ). Initial mortality results were low, and only *H. indica*, *S. riobravae*, and *S. carpocapsae* were capable of reproducing in *E. integriceps*, and only on insects collected immediately prior to migration. Further surveying to identify candidate strains adapted to Syria was conducted in the spring months in northern, north-east, central, southern and coastal areas of Syria. Six isolates of EPN within the genus *Heterorhabditis* indicated mortality averaging around 60% for treatments ranging for all concentrations of IJ tested for all isolates tested. These six, along with three of four isolates previously isolated from Syrian soils were found to be capable of using *E. integriceps* for development. **Keywords:** Entomopathogenic nematodes, soil survey, Sunn Pest, Syria.

### Introduction

Integrated pest management strategies are constantly improving and the search for novel methods of control not based on synthetic organic chemicals are always under testing (8). Parasites, predators and pathogens are among those components of an IPM package considered as biological control agents. Among the pathogens, many have limited host specificity, while some are broad-spectrum generalists, and can induce mortality among many different insects, even across orders. One type of pathogen is the group of entomopathogenic nematodes (EPN) in the Rhabditia. Two of the major genera in this Order are Steinernema and Heterorhabditis. These genera most commonly affect immature insects in the Coleoptera and the Lepidoptera, however reports of efficacy have been reported among other orders including Isoptera and Diptera (2, 8). These pathogens

can be isolated from soil, mass-produced in the laboratory and are not known to be harmful to vertebrates (1, 4). The first objective of this study was an initial investigation into the ability of EPN to demonstrate efficacy against Eurygaster integriceps and to determine if they can develop and pass through a life cycle in this host. The second objective was to isolate and conduct efficacy tests on EPN isolated from Syrian soils against E. integriceps. Standard surveys for EPN are typically performed using a highly susceptible host such as larvae of the greater wax moth, Galleria mellonella (L.). This host is particularly suitable as it is highly susceptible to all currently known EPN (6). Surveys of these soils for all possible nematodes, and efficacy testing of recovered isolates was the first step in the determination of the potential use of EPN as a management tactic for *E. integriceps*.

### **Materials and Methods**

## Rearing and Adjustment of Infective Juveniles

Nematodes were *in vivo* reared in late stage *G. mellonella* larvae at 27°C in Petri dishes lined with two layers of moistened filter paper. Third stage infective juveniles were harvested using the White trap collection method and stored at 10°C until use (7). Suspensions were prepared in tap water and adjusted to the appropriate concentrations by adjusting for the average direct count of three 50µl drops.

#### **Efficacy Testing of Known Species**

A total of six species were tested for efficacy against adult overwintering *E. integriceps*. A single strain per species was used to allow for a larger number of species for testing. Three concentrations were tested, 50, 100 and 200 IJ per adult to determine if efficacy has a dose-dependence. Individual adult *E. integriceps* were collected from overwintering sites in Tel Hadya, Syria and placed singly into wells of 24-well tissue culture plates (5ml well volume) that had been lined with two layers of filter paper and moistened with 150µl of appropriate suspension volume. A total of 15 adults were tested per replicate with a total of four replicates.

### **Testing of Syrian-Collected Species**

Adult *E. integriceps* were collected from the field and sorted for live and dead in the laboratory, grouped into sets of five and transferred to 9-cm-diam Petri dishes lined with two layers of filter paper to which a total of 1ml of an appropriate suspension concentration was added. Dishes were sealed with Parafilm and held at  $27\pm2^{\circ}$ C for 5d. Insects were assessed for viability by prodding. Dead insects were transferred to White traps and held at  $27\pm2^{\circ}$ C for 2wks, or until nematode emergence occurred. Five replicates were conducted per isolate at 50, 100, 200 and 400 IJ per ml and experiments were repeated twice.

### Survey for Entomopathogenic Nematodes

Five soil samples, each of about 400 g, were collected at a depth of 10 cm at each sampling site, with an area of approximately 100 m<sup>2</sup>. Samples from each site were homogenized by shaking in a polypropylene bag and transported to the laboratory and placed into 400-ml containers, on the bottom of which five late instar Galleria mellonella were placed. The Galleria trap bioassays were incubated at 27°C and checked for larval mortality after 3 and 6 d. Dead G. mellonella larvae from the soil bioassays were placed in separate Petri dishes with moistened filter paper and held in an incubator at 27°C then transferred to White traps. Soil samples were examined from 148 sampling sites in Syria. Isolates were identified to genera using a compound microscope at 40x magnification.

### **Statistical Analysis**

Mortality of *E. integriceps* was expressed as a percentage and arcsin square root transformed to satisfy the assumption of homogeneity of variance. Data were analyzed as a one-way factorial design using proc glm in SAS (11). Means were separated using the Student-Newman-Keuls protocol.

### **Results and Discussion**

Mortality was low and no effect of concentration was observed. Mortality did not differ significantly between 5 and 10 d, with most mortality occurring by day five. Statistically significant mortality (Figure 2) was only detectable for isolates C3B, S. Carpocapsae (Weiser), and Texas, S. riobravae (Cabanillas, Ponar & Raulston), for 50 IJ and 50 and 100 IJ respectively. When cadavers were placed on White traps to check for nematode emergence, only three isolates, C3B, Texas, and Venezuela, H. indica (Poinar, Kanunakar & David), were capable of developing through *E. integriceps*. The results from these trials suggest that while some species of EPN are capable of using E. integriceps as a host, the actualized efficacy of the treatments was low. Once a treatment effect had been demonstrated and it had been determined that EPN were capable of development inside *E. integriceps*, further work to identify suitable candidate strains isolated from local conditions was needed to identify candidate sources of material for testing for the use as a biological control agent.

Entomopathogenic nematodes are typically found in low abundance in nature, and their isolation requires that searches be conducted on a large scale, with typical success rates ranging from 0.5 to 10% recovery. Results from the soil baiting identified two sites within Syria positive with EPN (Figure 1). Recovered isolates are listed in Table 1. All isolates recovered were determined to be in the genus *Heterorhabditis* owing to their distinctive apical crenulation and large dorsal tooth (9).

To determine the efficacy of these isolates, laboratory trials were set up using a standard filter paper Petri dish assay (6). This assay was chosen because of its ease of replication, low cost and ability to determine isolates which are capable of using *E. integriceps* as a host and those that are not capable of this.



Figure 1. Distribution of entomopathogenic nematodes in Syria.

Isolate	Collection location	Site characteristics	Isolation date
Do-1	East Douma, Damascus	Grape vineyard	8 May 2004
Do-2	East Douma, Damascus	Alfalfa field	8 May 2004
Do-3	Douma, Damascus	Orchard	8 May 2004
DZ-1	Al Mreaia, Dier Ezzor	Alfalfa field	29 Mar 2004
DZ-2	Sfira Tahtani, Dier Ezzor	Grassland	29 Mar 2004
DZ-3	Bukros Fokani, Dier Ezzor	Sugar beet field	1 May 2004

**Table 1.** Entomopathogenic nematodes isolated from Syria. All strains are *Heterorhabditis* sp. Host bait insect used was late instar *Galleria mellonella* (L.) in all isolations.



Treatments

**Figure 2.** Screening of different species of entomopathogenic nematodes for efficacy against *Eurygaster integriceps*. Error bars superceded by the same letter are not significantly different at the =0.05 level of significance when tested using the Student-Newman-Keuls means separation procedure. Sc: *Steinernema carpocapsae* (Weiser), Sf: *Steinernema feltiae* (Filipjev), Sg : *Steinernema glaseri* (Steiner), Hb: *Heterorhabditis bacteriophora* (Poinar), Sr: *Steinernema riobravae* (Cabanillas, Poinar & Raulston), Hi: *Heterorhabditis indica* (Poinar, Kanunakar & David).

The highest observed mortality ranged between 50 and 60% (Figure 3). No effect of concentration within isolate was detected, however, at the 400 IJ per dish rate, six isolates demonstrated a significant treatment effect, DZ-3, Do-1, RC-3, RC-5, Texas and C3B. Only two isolates demonstrated this at each of 50 and 200 IJ per dish while no isolate demonstrated this at 100 IJ per dish. It is likely that this increase in observable treatment

effect is due to the increased chance of *E. integriceps* to encounter an infective juvenile within the test dish. This suggests that the use of a standard filter paper assay typically used for testing of efficacy against eruciform larvae, which maintain abdominal surface contact with the filter paper may not be suitable for the estimation of the efficacy of EPN against *E. integriceps*. While this method is a suitable test to determine isolates capable of using *E. integriceps* as a host, it is not very robust to isolate efficacy separation. Revision of these assays using simulated field conditions may provide better results in future studies for the purposes of isolate selection.



Figure 3. Mortality of Eurygaster integriceps tested against Syrian isolates of entomopathogenic nematodes.

Given that all isolates recovered were within the genus *Heterorhabditis* and that all were capable of inducing mortality and capable of development within *E. integriceps*, it is most likely that the future direction of this work is dependent on identifying strains with suitable environmental tolerance. Future, and repeated, surveys of areas within Syria should hopefully identify candidate strains for further development. In addition, testing that incorporates field conditions into the system, such as laboratory-controlled microcosom study is needed.

### Acknowledgements

The authors would like to thank Drs. Bryron J. Adams and Khuong B. Ngyuen for graciously providing the nematode strains for testing and Mr. Fawzi Rihawi and Zaccarhia Ibrahim for their technical assistance.

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عبد الحي، محمد، زياد صيادي، وليم ريد ومصطفى البوحسيني. 2007. تقصي الترب السورية لأجل النيماتودا الممرضة للحشرات ودراسة فعاليتها ضد حشرة السونة. الصفحات 251–257.

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### The Present Situation of Some Alternative Hosts (Dolycoris baccarum L., Piezodorus lituratus F.) of the Natural Enemies of Sunn Pest in Southeastern Anatolian Region of Turkey

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### Abstract

Akkaya, A. 2007. The Present Situation of Some Alternative Hosts (*Dolycoris baccarum* L., *Piezodous lituratus* F.) of the Natural Enemies of Sunn Pest in Southeastern Anatolian Region of Turkey. Pages 259-263.

The Sunn Pest, *Eurygaster integriceps* Puton, is the most important pest of wheat and barley in Southeastern Anatolia. This pest has many natural enemies in cereals but also in lentil areas due to the presence of the pentatomid bugs, *Dolycoris baccarum* L. and *Piezodorus lituratus* F. that damage lentils and serve as hosts of these natural enemies. These pentatomid species were researched from point of view their occurrence, distribution, hosts and importance as potential hosts of the natural enemies including mainly scelionid egg parasitoids and tachinid adult parasitoids in the lentil agroecosystem. The data obtained on some biological and ecological properties of these pentatomid species were evaluated from an associate lentil-cereal IPM aspect taking into consideration previous data obtained on Sunn Pest and its natural enemies. **Keywords**: *Dolycoris baccarum*, *Eurygaster integriceps*, *Piezodorus lituratus*, Scelionidae, Sunn Pest.

### Introduction

The Sunn Pest, Eurygaster integriceps Puton, is the major pest of wheat and barley in Southeastern Anatolian Region of Turkey. Chemical control has been used for management and large cereal areas in the region have been sprayed with insecticides every year. Among the biotic factors that have limited populations of this pest, natural enemies have an important place. Scelionid egg parasitoids and tachinid adult parasitoids are the most important ones. These parasitoids have a rich host complex including most of the pentatomid species. Aelia acuminata L., Aelia rostrata Boh., Dolycoris baccarum L., Carpocoris fuscispinus Boh., Graphosoma italicum (Muell.). Piezodorus lituratus F., Eurydema festivum L., and Nezara viridula L. species were determined as hosts of egg parasitoid Trissolcus semistriatus Nees, besides *Eurygaster* spp in the Region (2).

In recent years, due to an increase in irrigated areas of Southeastern Anatolian

Region, as parallel changing climatical conditions and plant patterns, the populations of some pentatomids has increased and they have become main pests for some crops. For instance, while the berry bug, *D. baccarum* and gorse shield bug, *P. lituratus* are pentatomids normally neutral in the agroecosystem, now they are among the important harmful pests of lentil.

To determine distribution, hosts and some biological properties of these pentatomid species biological and ecological studies were done in Southeastern Anatolia from 1997-2000. The results were evaluated from the point of view not only of their control in lentil areas but also their importance as hosts of parasitoids of Sunn Pest in the development of an integrated pest management system against these pests in fields of winter lentil and wheat.

### Material and Methods

To obtain distribution areas of *D. baccarum* and *P. lituratus*, survey studies were done in

lentil fields of Adıyaman (Center, Gölba 1, Kahta), Diyarbakır (Center, Bismil, Çınar, Ergani), Mardin (Center, Derik, Midyat, Kızıltepe) and anlıurfa (Merkez, Hilvan, Siverek, Bozova, Viran ehir) provinces. Sampling studies were done in 98 lentil fields between April 5-June 11,1998. For sampling methods, sweeping and direct observations of plants were done.

To investigate hosts, their seasonal occurrence and biological properties of D. baccarum and P. lituratus, field studies were conducted in lentil and in other crop production areas in the region during the period between their coming from overwintering areas to production areas in early spring and migration to overwintering areas in autumn. For biological studies, adult activities were investigated in lentil fields during the vegetation period. To provide information on seasonal occurrence and hosts of the bugs, different biological stages were studied on different cultural plants and weeds.

### **Results and Discussion**

### **Distribution Areas**

Lentil production areas of Adıyaman, Divarbakır, Mardin and anlıurfa provinces were infested with both stink bugs. D. baccarum and P. lituratus were observed together in each lentil field, but percentages of each species relevant to the total pentatomid population indicated differences according to provinces. These rates were determined for *D*. baccarum as 29.9%, 82.9%, 90.0%; for P. lituratus 70.1%, 17.1% and 10.0% in Divarbakır, anlıurfa and Mardin provinces respectively. Regionally, the infestation rate in the lentil fields (with both of the stink bugs) averaged 57.5%. Lodos (3) recorded that D. baccarum and P. lituratus showed wide distribution in all regions of Turkey and the ratios of one to the other varied.

### Biology

Dolycoris baccarum overwinters under perennial plants, in soil, in caves of stones as

adults on mountains with intermediate elevations of 1700 to 2300 m. P. lituratus mostly overwinters under the bark of the trees. At the first week of April (6.4.1998; 8.4.1999) with increasing temperatures overwintered adults of both species start to migrate from mountain areas to fields. Temperatures were recorded as 12.3°C and 11.8°C when the bugs were seen for the first time in lentil fields in 1997 and 1998 respectively. After feeding for a few days, at the second week of April (10.4.1998; 9.4.1999) when temperatures were 15.6°C and 12°C respectively, mating activity started. The first eggs were seen the third week of April (24.4.1998; 20.4.1999) and the temperatures in the beginning oviposition stages for both of the bugs were 11.7°C and 14.2°C, respectively. The oviposition period continued 5-11 days according to temperature. The egg incubation period lasted for about a week. Nymphs of D. baccarum and P. lituratus have 5 instars that lasted for 4-5, 4-13, 6-11, 9-11, 5-8 and 5, 8-12, 3-7, 4-8, 4-6 days, respectively (1). At lentil harvest the nymphs of both of species became adults.

### Hosts

The hosts of *D. baccarum* in Southeastern Anatolian Region are given at Table 1.

*P. lituratus* feed on Leguminosae plants. When they migrate from overwintering areas, they first feed on *Vicia narbonensis* var. *narbonensis* L.. At this time lentil is not flowering. After lentil harvest, the last stage nymphs and new generation adults feed on Compositeae weeds and *Verbascum* spp. (Scrophullariaceae) to supply their water needs. During June-September, the bugs were seen in clover fields near overwintering areas.

### **Seasonal Occurrences**

*D. baccarum* is a highly polyphagous pentatomid. When they migrate from overwintering areas, they first feed as aggregates on mainly Brassicae and Labiate weeds. When the flowers of the weeds start to open, they move to lentil plants and feed on the generative parts of the plants. After lentil

harvest, the last stage nymphs and new generation adults feed on Compositeae weeds and Verbascum spp. (Scrophullariaceae) not only to feed but to supply their water needs. Then they migrate to overwintering areas. The bugs congregate in large numbers on Pistacia spp. and Cerasus mahalep L. before they reach the hibernation areas. New generation adults prefer aestivating near the top of the mountains under perennial plants. At the end of the observations made in agricultural areas, from June-August, the bugs were seen on chickpea, sunflower, melon plants and the weeds in these areas. Their populations were higher on the weeds than on the crops.

In autumn, the bugs caused important damage to maize, tomato, grape, tobacco, clover, beet, sunflower, fig, pomegranate near overwintering areas. They preferred maize, tobacco, sunflower and tomato plants. They were found on tobacco pods in very high populations, but due to harvesting economic damage wasn't observed.

Piezodorus lituratus hasn't a rich host complex like D. baccarum. This species feeds on Leguminosae plants. In early spring, after they migrate from overwintering areas, they first feed on Vicia narbonensis var. narbonensis before the flowering stage of lentil. Until lentil is harvested they feed on lentil pods and seeds. After lentil harvest, the last stage nymphs and new generation adults move to wild Compositeae and Verbascum spp. (Scrophullariaceae) together with D. baccarum for their water needs. P. lituratus were found in clover fields near overwintering areas at high population densities during June-September.

Considering the egg parasitoid (*Trissolcus semistriatus* Nees) of Sunn Pest their alternative hosts were present in nature and they had a wide distribution in the region.

**Table 1.** The plant hosts of *Dolycoris baccarum* L.in the Southeastern Anatolian Region.

Family	Plant species
Amaranthaceae	Amaranthus retroflexus L.
Boraginaceae	Echium italicum L.
C C	Teucrium polium L
Capparaceae	Capparis spinosa L.
Compositae	Centaurea calcitropa L.
	Centaurea balsamite L.
	Centaurea kurdica Reichardt
	Centaurea sclerolepis Boiss.
	Centaurea solstitialis L.
	Cichorium intybus L.
	Picnomon acarna (L.)
	Compositae polypodiifolia Boiss. var.
	pseudobehen (Boiss.)
	<i>Cynare</i> sp.
	Echinops nitro L.
	Echinops sp.
	Gundelia tournefortii (L.)
	Helianthus annuus L.
	Lactuca sp.
a	Notobasis syriaca L.
Caryophylaceae	Silene sp.
Chenopodiaceae	Beta vulgaris L.
Cruciferae	Brassica elongata Enrn.
D:	Sinapis sp.
Dipsacaceae	Scabiosa sp.
Euphorofaceae	Euphorbia sp Ou chryskie sp
Fabaceae	Onobrychis sp.
	Vicia narbonensis var narbonensis I
	Vicia norana var norana Reut Ex
	Roiss
Gramineae	Triticum spp
Grannieae	Zea mays L.
Labiatae	Lamium amplexicaula L.
Luciuue	Molucella laevis L.
	Phlomis sp.
	Salvia svriaca L.
Leguminosae	Astragalus spp.
8	Cicer arietinum L.
Malvaceae	Alcea sp.
Plumbaginaceae	Acantholimon acerosum var.
Ū.	acerosum Boiss.
Portulacaceae	Portulaca oleracea L.
Punicaceae	Punica granatum L.
Rosaceae	Cerasus mahalep L.
Scrophullariaceae	Echium sp.
	<i>Kickxia</i> sp.
	Verbascum spp.
Solanaceae	Capsicum annum L.
	Lycopersicum esculentum Mill.
	Nicotiana tabacum L.
Vitaceae	Vitis vinifera L.

Dolycoris baccarum and P. lituratus leave overwintering areas before Sunn Pest. While the temperature was  $11.8^{\circ}C$  when D. baccarum and P. lituratus were first observed in lentil fields, E. integriceps adults and T. semistriatus were first seen in wheat fields when the temperature was 17°C and 13°C, respectively (4). Sunn Pest migrated last. After a few days feeding on these wild plants, the stink bugs mate and lay eggs. The first eggs were found the second week of April, about 12-15 days after the stink bugs migrated to lentil fields. Temperature was recorded as 14°C in the beginning of the oviposition stage. Oviposition period continued an average 7.4 (5-11 days). D. baccarum eggs were found on Petroselinum crispum Mill. (Umbelliferae), Capsella sp. (Cruciferae), Triticum vulgare L. (Gramineae), Lamium amplexicaula L.

(Labiate) and Orobancheae weeds in lentil fields. Piezodorus lituratus eggs were found only on lentil plants. im ek (4) recorded that T. semistriatus adults feed on some weeds including Capsella bursapastoris (L.). Myagrum perfoliatum L., Sisymbrium altissimum L., Erysimum smyrnaeum Boiss et Bal., Neslia paniculata (L.) Desv., Ranunculus arvensis L., Scandix pectenveneris (L.), Ranunculus sp., Gallium aperine L., Asperula arvensis L., Fumaria oficinalis L., Arthemisia hyalinae during April. These results showed that D. baccarum and P.lituratus eggs found at nature during the last half of April. Some weeds are important not only for adult egg parasitoid feeding but also for carrying the eggs of hosts. D. baccarum is an important host for *T.semistriatus* before *E. integriceps* is present and available for parasitization.

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أكايا، عائشة. 2007. الوضع الحالي لبعض العوائل البديلة (Dolycoris baccarum L.، الوضع الحالي لبعض العوائل البديلة (F. يالأعداء الطبيعية لحشرة السونة في منطقة جنوب شرق الأناضول من تركيا. الصفحات 259–263.

تعد حشرة السونة (Eurygaster integriceps Puton) أهم آفة على القمح والشعير في جنوب شرق الأناضول. لهذه الآفة العديد من الأعداء الطبيعية في مناطق الحبوب، لكن أيضاً في مناطق العدس نظراً لوجود حشرات البق من فصيلة Dolycoris baccarum L. ، Pentatomidae و Piezodorus lituratus F. توزعها، عوائلها وأهميتها كعوائل لهذه الأعداء الطبيعية. درست هذه الأنواع من فصيلة Pentatomidae من ناحية تواجدها، توزعها، عوائلها وأهميتها كعوائل ممكنة تما عداء الطبيعية متضمنة بشكل رئيس طفيليات البيض من فصيلة Scelionidae من ناحية تواجدها، توزعها، عوائلها وأهميتها كعوائل ممكنة في النظام البيئي الزراعي للعدس. تم تقييم البيانات التي تم الحصول عليها حول بعض الخصائص الحيوية والبيئية لهذه الأنواع من فصيلة عميلة علي المنات البيئية لهذه الأنواع من أهميتها للقوة لترافق العدس الحيوية والبيئية لهذه الأنواع من فصيلة علي متضمنة من أوجه الإدارة المتكاملة للآفة لترافق العدس-الحبوب آخذين بعين الاعتبار البيانات التي تم الحصول عليها سابقاً على حشرة السونة وأعدائها الطبيعية.

كلمات مفتاحية: .Scelionidae ، Piezodorus lituratus ، Eurygaster integriceps ، Colycoris baccarum ، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

### Role of Biocontrol Agents in Decreasing Populations of Sunn Pest in Northern Iraq

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### Abstract

Al-Izzi, M.A.J., A.M. Amin and H.S. Al-Assadi. 2007. Role of Biocontrol Agents in Decreasing Populations of Sunn Pest in Northern Iraq. Pages 265-271.

The infection of Sunn Pest populations, *Eurygaster integriceps* Puton by *Beauveria bassiana* (Balsamo) Vuillemin, egg parasites *Trissolcus* spp. and *Ooencyrtus* sp., the adult parasite *Phasia* sp. and predators were recorded in the top, mid and foothills of mountains in Erbile. Fungal infection in the top of Seffin Mountain was recorded in July, August, September, October, November, and December, 1999. It was 58.78%, 44.19%, 20.82%, 38.96%, 14.89% and 40.48%, respectively. During January, February and March, 2000, infection rates were 32.26%, 31.58% and 2.16%, respectively. However, on the mid-mountain it was 52.94%, 17.86% and 2.5%, respectively and in the foothills it was 49.15%, 16.33% and 13.04%, respectively. Parasitism of Sunn Pest eggs by *Trissolcus* spp. and *Ooencyrtus* sp. was recorded in the fields of Koshtapa, Ashkawsaka, Koysinjak during April and Smak-shereen and Erbil during May, 1999. Percent parasitism was 100%, 76.92%, 85.71%, 100% and 100%, respectively. Adults parasitized by *Phasia* sp. was recorded in Seffin Mountain. The number of dead adults per 10 m<sup>2</sup> was observed during July, August, September, October, November and December, 1999. The percentages were 8, 21, 67, 43, 2 and 6. Adult parasitism decreased to 1% in March, 2000. Number of predators (*Calosoma, Carabidae, Coccinella septempunctata* L., spiders, and birds) was observed feeding on different developmental stages of Sunn Pest.

Keywords: Beauveria bassiana, biological control, egg and adult parasites, Sunn Pest.

### Introduction

There are a number of insect pathogens that effect populations of Sunn Pest during hibernation and aestivation. *Beauveria bassiana* (Balsamo) Vuillemin and *Aspergillus candidus* Link. (21), killed 80% of the adult population in 1973 in Turkey (11). Parasites of Sunn Pest eggs include *Trissolcus* spp., *Telenomus* spp. and of adults *Phasia* spp. (13, 24).

Parasite populations differ from year to year (9). *Telenomus chloropus* Thomson prefer cold and humid weather whereas *Trissolcus grandis* Thomson prefers irrigated soils (2). Egg parasitization by *T. grandis* and *T. vassilievi* Mayr increased to 50% during the oviposition season (22). There are 15 tachanid parasites (Phasiinae) infesting Sunn Pest adults (4). Some of these lay their eggs on the cuticle and the larvae feed inside the host.

Predator numbers increased in barley and wheat fields to 40-60 insects/sq m during the growing season (17). carabid beetles prey on Sunn Pest nymphs (12). They decreased populations by 24% in Hungary. There are 57 species belonging to 26 genera of carabid beetles that feed on Sunn Pest during the winter in wheat (20) and there are 85 species of carabid beetles in the north of Quokkas feeding on one or more stage of Sunn Pest (14). The green lace wing and spiders are other predators that attack eggs and nymphs of Sunn Pest and with other beetles reduce populations (14) The most abundant spiders found in the soil are Pardosa monticola (Clerck) and Trochosa terricola Thorell (23).

### **Materials and Methods**

The research was done in wheat and barely fields in the plains and hibernation areas in the mountains. The influence of factors effecting the survival of Sunn Pest during its migration or hibernation were studied. Mortality factors were either identified in the Iraqi Atomic Energy Commission (IAEC) or in the Iraqi Natural History Museum and separated according to the followings: those killed by *Beauveria bassiana*, parasitoids, or predators.

## The Entomopathogenic Fungus Beauveria bassiana

The adults of Sunn Pest covered by a layer of overgrown white mold with external mycelium and white conidia or those adults filled with fungus mycelium and conidiophores growing inside the insect skeleton were separated from underneath the plants on the mountains during the hibernated season.

The infected insects were left inside a plastic container and transferred to the laboratory to calculate the infestation percentage in the area.

The fungal growth was isolated from the infected insects and transferred to potato dextrose agar medium in glass Petri-dishes under aseptic conditions, the dishes were then placed inside an incubator  $(25\pm1^{\circ}C)$  for growing the fungus. The white growth of *Beauveria bassiana* was isolated and transferred to new growth dishes for purification from other fungi.

Purification and growth of *Beauveria* bassiana was continued to have a pure culture of the fungus.

The isolated pure culture of *Beauveria* bassiana was compared with the fungus pure culture stored in the biotechnology laboratory in IAEC.

### **Parasitoids**

Sunn Pest eggs, nymphs and adults were collected from the wheat plants in the field and transferred separately to small screen cages

inside the laboratory, the cages contained wheat plants grown in pots. Parasitoid adults were collected from the cages and identified by scientists at the Iraqi Natural History Museum.

### Predators

Sunn Pest predators were collected when they were found feeding on one or more of Sunn Pest stages (eggs, nymphs and adults). The predators were collected either from infested wheat plants by Sunn Pest stages or during hibernation of Sunn Pest in the mountains. Predators were identified at the Iraqi Natural History Museum.

### **Results and Discussions**

### Infection of Sunn Pest by the Entomopathogenic Fungus Beauveria bassiana

Field visits showed that the entomopathogenic fungi *Beauveria bassiana* invaded Sunn Pest adults during hibernation season in the mountains. Mortality by *B. bassiana* was different between July 1999 and June 2000 on the following mountains: Hassan Begg, Hindrin, Klinder, Sherine, Bradost, Zozak, Kork, Biran and Seffin (Table 1). Mortality by the fungus is not related to the height of the Mountains.

From July to October Sunn Pest aggregated, at the top of Seffin Mountain. The highest number was recorded in August. The number decreased in the following months to reach its minimum during January, February and March. The highest number of adults infected by *Beauveria bassiana* was recorded in August and decreased in the following months to reach its minimum number in March 2000 (2.86%). The highest rate of infection was 58.78% during July 1999 (Table 2).

The infected adults were characterized by inactivity and/or slow movement. When we dissected these adults we noted fungal mycelia throughout the insect cuticle with permeation into the insect viscera. In advanced stages of infection the fungus appeared outside the insect skeleton and caused death of the insect.

Temperatures ranged between 16.6 and 28.0°C during July to October which kept the hibernated insects underneath the plants and allowed fungal development to continue. When temperatures fell to 9°C in November and December the insect population at the top

migrated to the sides of the mountain. The relative humidity (RH) in hibernated areas ranged from 39.5 to 64.0% from July to March (Table 2). Temperature  $<10^{\circ}$ C and RH >50% decreased the insect population at the top of the mountain with variability in amount of fungal invasion.

**Table 1.** Sunn Pest mortality at the top of different mountains in the Northern Iraq during July 1999 and June 2000.

			Total insects per 10 <sup>2</sup> m at	
Mountain	Elevation (m)	Date of visit	mountain-top	Mortality%
Hassan Begg	2110	1/7/1999	916	55.56
		26/6/2000	76	89.47
Hindrin	2025	7/7/1999	100	81.30
Klinder	1823	8/7/1999	1458	28.53
Sherine	2378	20/7/1999	720	36.94
		29/6/2000	146	30.83
Bradost	2076	22/7/1999	943	59.07
		25/6/2000	308	71.10
Zozak	1863	6/7/1999	25	40.00
Kork	2125	21/7/1999	1095	66.30
		27/6/2000	373	33.78
Biran	2076	22/7/1999	766	79.63
		24/6/2000	692	35.26
Seffin	1975	5/7/1999	295	61.27
		30/6/2000	117	80.34

**Table 2.** Infection of hibernated Sunn Pest by the entomopathogenic fungus *Beauveria bassiana* at the top of Seffin Mountain, Iraq.

	No. of	No. of insects	% of	Hibernation Conditions	
	hibernated	infected by	infected	Temperature	<b>Relative Humidity</b>
Month	insects	B. bassiana	insects	° C	%
July	905	532	58.78	28.0	39.5
August	1824	806	44.19	26.8	42.5
September	783	163	20.82	21.4	45.8
October	521	203	38.96	16.6	50.1
November	47	7	14.89	9.5	57.6
December	42	17	40.48	9.2	61.6
January	31	10	32.26	2.3	61.5
February	38	12	31.58	8.8	64.0
March	35	1	2.86	14.3	61.7

In November, when hibernated adults moved from the top to the sides of the mountain they carried B. bassiana to these new locations and then to the mountain base. The rate of infection at the side of the mountain was 83.13-85.7% and 85.17-71.43% in November and December (Table 3). Most of the infected insects died by the end of December which left a small population of hibernated insects. The percentage of infected insects on the mountain sides decreased gradually from 85.71 in December to 52.94 in January to 17.86 in February and to a minimum 2.5 in March. The percentage of infected individuals at the mountain base decreased from 85.17 in November to 49.15 in January to 16.33 in February and to 13.04 in March.

**Table 3.** Infestation of hibernated Sunn Pest by theentomopathogenic fungus *Beauveria bassiana* inthe slope and base of Seffin Mountain, Iraq.

Month	No. of hibernated insect	No. of insects infested by fungus	% of insects infested by fungus
Mountain s	ide / slop		
November	83	69	83.13
December	56	48	85.71
January	43	18	52.94
February	28	5	17.86
March	40	1	2.50
Mountain b	ase		
November	391	333	85.17
December	273	195	71.43
January	59	29	49.15
February	49	8	16.33
March	46	6	13.04

The activity of *B. bassiana* was responsible for killing 80% of Sunn Pest adults in overwintering sites in mountain areas in Turkey (10). This fungus was produced commercially under the trade name Boverin and used under laboratory conditions against Sunn Pest in Romania (6) and in the Soviet Union and Vietnam against 70 species of insects including Sunn Pest (1).

### **Egg Parasitoids**

Egg parasitoids were observed on Sunn Pest eggs in wheat and barely fields during April and May. The female of *Trissolcus* spp. (Hymenoptera: Scelionidae) and *Ooencyrtus* sp. (Hymenoptera: Encyrtidae) (first recorded in Iraq) insert their eggs inside Sunn Pest eggs. The female can recognize parasitized eggs from non parasitized eggs (8). The period for egg insertion until adult emergence of *Trissolcus* spp. takes two weeks. The parasitization percent was calculated at different times and locations (Table 4).

The distribution of *Trissolcus grandis* and *Telenomus chloropus* has been recorded on Sunn Pest eggs in different countries (16, 19). In some locations they caused a higher percentage of egg parasitization, infestation reached 70% (7), 80% (14, 18, 19, 5) and 90% in the irrigated fields and in mountains (8).

 Table 4. Field observation of the activity of Trissolcus spp. infesting Sunn Pest eggs in the field.

Date 1999	Location	No. of host eggs	No. of parasite adults produced	Parasiti- zation %
6/4	Koshtoba	14	14	100
18/4	Ashkosaqa	13	10	77
21/4	Koisinjack	14	12	86
5 /5	Smaq Shirin	14	14	100
17/5	Erbile	13	13	100

### **Sunn Pest Adult Parasitoids**

During the hibernation period in the mountain regions and plains areas, infected Sunn Pests were observed by a holes on the lateral sides of abdominal segments or at the abdominal tip as a result of parasitization by *Phasia* spp. The number of parasites increased gradually in the field from July to September then decreased in March of the following season (Figure 1). The growth of the parasitoid immature stages inside the host depends on the environmental conditions and the host hibernation sites. These factors can cause an adverse effect on adults of Sunn Pest.

Parasite emergence was observed from adults migrated from hibernated areas to the fields of wheat and barley. Larvae emerged either from the abdominal segments or from the thorax of the host and then pupated on the ground. Pupation lasted two weeks for development to adults. Parasitoid adults emerged in April and May of 1999 and 2000. The parasitoid female inserts a single egg into the dorsal area between the head and thorax of Sunn Pest adults. The parasite continues its development inside adults during hibernation (3) and infects new adults before migration to the mountains.

### Effect of Predators on Sunn Pest Populations

There are many different predators feeding on Sunn Pest in wheat and barely fields. Spiders were observed feeding on Sunn Pest nymphs. The adults of coccinellids with seven spots were observed feeding on adults and coccinellid larvae feeding on eggs. Calosoma and other carabid beetles were found feeding on hibernated adults. Spiders and arthropod predators were found in different numbers feeding on Sunn Pest eggs and 1<sup>st</sup> and 2<sup>nd</sup> instars during April and May (15, 12, 17).



Figure 1. Sunn Pest adults parasitized by Phasia sp. from July 1999 to March 2000 in Northern Iraq.

### الملخص

العزي، محمد.ع. ج.، عبد الباسط. م. أمين، وحسن. س. الأسدي. 2007. دور عوامل المكافحة الحيوية في تخفيض مجتمعات حشرة السونة في شمال العراق. الصفحات 265–271.

سجلت إصابة مجتمعات حشرة السونة Phasia sp. بالفطر Phasia sp. بالفطر Phasia sp. بالفطر Phasia sp. في قمة، منتصف Phasia sp. وسفج الجبال في إربيل. سجلت الإصابة الفطرية في قمة جبل سفين في تموز /يوليو ، آب/أغسطس ، أيلول/سبتمبر ، تشرين وسفح الجبال في إربيل. سجلت الإصابة الفطرية في قمة جبل سفين في تموز /يوليو ، آب/أغسطس ، أيلول/سبتمبر ، تشرين الأول/أكتوبر ، تشرين الثاني/نوفمبر وكانون الأول/ديسمبر ، 1999. بلغت نسبة الإصابة 8.78 ، 44.19 ، 20.20 ، 38.96 ، الأول/أكتوبر ، تشرين الثاني/نوفمبر وكانون الأول/ديسمبر ، 1999. بلغت نسبة الإصابة 8.78 ، 44.19 ، 20.20 ، 38.96 ، 14.89 ، 20.20 ، 38.96 ، 14.89 ، 20.20 ، 38.96 ، 14.89 وكانون الثاني/نيابير ، شباط/فبراير وآذار /مارس ، 2000 بلغت نسبة الإصابة 32.26 ، 32.96 ما وعند سفح 31.58 و 10.2% ، على التوالي . خلال كانون الثاني/ينابير ، شباط/فبراير وآذار /مارس ، 2000 بلغت نسبة الإصابة 32.26 ، 32.96 ما وعند سفح 31.58 و 10.5% ، على التوالي . خلال كانون الثاني/ينابير ، شباط/فبراير وآذار /مارس ، 2000 بلغت نسبة الإصابة 32.26 ما 32.58 ما 32.58 ما 32.56 ما 32.56

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon. 271

### Sunn Pest Overwintering Sites, Parasitoids and Effects on Cereal Lines and Varieties

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### Abstract

## Yildirim, A.F., E. Kinaci and M. (Elmali) Uysal. 2007. Sunn Pest Overwintering Sites, Parasitoids and Effects on Cereal Lines and Varieties. Pages 273-277.

Sunn Pest is one of the most important cereal pests that affects both grain yield and quality of cereal in Central Anatolia, Turkey. In this study, overwintering sites, death rate in overwintering sites of Sunn Pest, and their migrations to cereal fields in Konya Province were investigated. *Eurygaster maura* L.and *E. austriaca* Schrank are prevalent. The rate of egg parasitism was between 4 and 93.8% and the parasitoids were more abundant in irrigated and bushy areas. *Trissolcus grandis* Thomson was the most common egg parasite in the region. Cage experiments were conducted and field screenings were done to determine the varietal preferences of Sunn Pest. In the cage experiments, it was found that there was variation among varieties in rate of grains damaged by the pest, and hard red grains like Bezostaja-1 were less damaged than white soft variety Gerek-79. In the fields, nymphs and new adults were abundant in some lines and varieties for example line Arina and variety Gerek-79.

Keywords: Cage, Eurygaster austriaca, Eurygaster maura, natural death, overwintering sites, parasitism, tolerance, Trissolcus grandis.

### Introduction

In agriculture, producing high quality products is as important as increasing yields. One of the factors making it difficult to reach this goal is the Sunn Pest of cereals. Characteristically insects in the family Scutelleridae produce a bad odor and cause damage by piercing and sucking plants. Grains damaged by Sunn Pest lose weight, grain quality is lowered and it becomes difficult to make bread out of these damaged grains (1, 11).

Although it has been recognized in the Central Anatolian Plateau of Turkey for years, it has not been given enough importance because its damage was below the economical threshold until 1989, when it caused significant damage in cereal fields of Konya Province. Populations of and damage by Sunn Pest varies from region to region and year to year, depending on ecological conditions and if they are controlled by egg parasitoids. Sunn Pest damage in Central Anatolian Plateau is lower than in Southeastern Anatolia and the Thrace.

In this study, to gather basic information for integrated control systems to keep Sunn Pest damage below the economic threshold, species of Sunn Pest in Konya, their overwintering sites, the vegetative cover in these sites, egg parasitoids and their impact were investigated. Sunn Pest damage on wheat cultivars grown intensively in the region was examined in cage experiments and field observations.

### **Material and Methods**

## Overwintering Sites and Species Identification

To estimate Sunn Pest population density in overwintering sites, and to observe their migration from these sites to the plains, counting of live and dead Sunn Pest were made at overwintering sites using 25 x 25 cm. frames. Twenty separate counts were made in
an area of 0.2 ha. Species identifications and sex ratios were done in the laboratory of Selcuk University, Faculty of Agriculture, according to Memisoglu (7) and Paulian and Popov (8) using adult Sunn Pests collected from fields. To understand the timing of Sunn Pest activation and the start of their migration in the spring, temperatures of air, below cover (oak leaves, *Astragalus*), and soil at 3-5 cm depth were measured.

#### **Field Studies**

With the beginning of Sunn Pest migration observations and counts of Sunn Pest populations and egg parasitoids were started in fields on the base of the mountains where overwintering sites were located. A Sunn Pest egg parasitoid survey was done when ~70% of the Sunn Pests had migrated to the plains and 20-30% of the eggs were one week old. The egg parasitoid ratio in the region was calculated by dividing the total number of eggs in egg packages collected from fields by the total number of eggs with parasitoids. The Sunn Pest eggs with parasitoids were put into glass jars with the plant part they were found on, and the jar covered with tulle. Jars were held in the laboratory at 25±2°C for parasitoid emergence. Emerging parasitoids were put into glass bottles containing 70% alcohol and identified by Professors Ne et Kılınçer and Miktat Do anlar. Sunn Pest densities were observed and wheat varieties and lines and their hybrids damaged by Sunn Pest were studied in yield trials in Konya (901 entries) and Çumra (321 entries) to understand whether the pest had selectiveness on varieties under natural conditions.

Cage experiments were established to determine the level of Sunn Pest damage on wheat varieties under controlled conditions. In the first year of these experiments (1990), registered varieties Kırac-66, Bezostaya-1, Seri 82, Gerek-79, Bolal-2973, Atay-85, Es-14, Kırkpınar-79 and 4 promising lines from Bahri Dagdas Winter Cereal Research Center, BDME-112, BDME-128, BDME-156 and BDME-157 were planted in 3 rows of 2 m long with 3 replications. In May, all entries in all replications were covered with cages measuring 60x60x90 cm. The sides of the cages were covered with a dense textured tulle 250 µm. Second year cage experiments (1991) were established with Odeskaya-51, Es-14, Bezostaya-1, Atay-85, Gerek-79, Kıraç-66 varieties and a promising line, BDME157 (Da da -94) on seed multiplication plots. Three pairs of adult Sunn Pest were put into each cage whereas control cages had no adult Sunn Pests. Cages were checked every week and dead Sunn Pests were replaced by new field-collected ones. At the end of the first year's harvest, damaged grains were evaluated whereas in the second year's experiment numbers of Sunn Pest and wheat spikes in parcels and 1000 grain weights were evaluated for loss weight of grain that it is compare with control parcel. Grains obtained from each cage were divided into 10 groups of 100 grains and the numbers of sucked grains were taken as the varietals average of damage (rate).

#### **Results and Discussion**

#### Sunn Pest Overwintering Sites

Ten overwintering sites were found in Konya Province. Sunn Pest live from autumn to spring under leaves of oak., Pinus spp., Thymus spp., Acantholiman spp. and Astragalus spp. and in soil cracks, in diapause. The most important overwintering site is the Karada site in Cumra, Eurygaster maura L. and E. autriaca Schrank. were found in and around Konya. The majority were E. maura (96.2%) with E. autriaca (3.8%). E. maura had the higher density in Karada site in Cumra, constituting 43.9% of the total regional population. Altinayar (2) found only E. maura in Karaman and Ere li, while Lodos (5), Lodos and Önder (6) reported that E. maura and E. austriaca existed in the Central Anatolian Plateau but they did not find any E. integriceps. In Ankara Province, Memi o lu (7) found E. maura and E. autriaca and reported that *E. integriceps* constitutes 99.28% of the total Sunn Pest population in the region.

Examination of genital organs of collected adults showed that the male-female sex ratio was 1:1.2 (44.7% male-55.3% female). The natural death rate was 5-10% at the overwintering sites. In years of dry springs and summers, this rate was higher due to insufficient nutrition of new generation adults. Karman (4) reports that Sunn Pests returning to overwintering sites without having sufficient nutrition are more sensitive to chemicals and adverse environmental conditions. Memi o lu (7) reports that the death rate at overwintering sites varies between 6 and 13% and this rate is higher on the northern parts of these sites. Migration of Sunn Pest to plains started when below-cover temperature was 11-13°C and the air temperature was 20-22°C. Yüksel (11) reports that Sunn Pests fly when the temperature beneath overwintering site vegetation is 20°C, flights decrease with lowering temperatures and stop below 10°C.

#### Egg Parasitoid Ratios and Some Parasitoid Species Found in the Region

The numbers of Sunn Pest eggs with parasitoids in Konya Province are given in Table 1.

**Table 1.** The percentage of Sunn Pest parasitizedeggs in Konya in 1990.

County	Lowest ratio (%)	Highest ratio (%)
Ak ehir	84.2	93.8
Çumra	15.0	91.0
Konya-Merkez	4.0	53.8
Karapınar	9.4	52.0
Kadınhanı	64.0	75.0
Sarayönü	57.0	88.0

Parasitoid ratios are higher in Ak ehir, Çumra and Sarayönü counties where irrigated land and woods cover large areas. Radjabi Amir-nazari (9) found in Iran that parasitoid ratios can be as high as 90% in irrigated cereal fields and high rainfall areas. As a result of diagnostic studies on the egg parasitoids collected from the region, it was found that *Trissolcus grandis* Thomson, *T. vassilievi* Mayr., *Oencyrtus telenomicida* (Vassiliev), *Oencyrtus* spp. exist in the region. *Trissolcus grandis* was the most predominant species. Gültekin (3) reported that *T. grandis*, *T. reticulates* Delucchi, *T. scutellaris* (Thomson), *T. rungsi* Voegele, *Telenomus* sp., *O. telenomicide* existed and *T. grandis* was the predominant species in the Thrace region.

#### **Field Works**

Observations on the experimental plots in Konya and Çumra under natural conditions showed that Sunn Pest adults and nymphs existed intensively on some varieties and lines but not on the others. Pedigrees and Sunn Pest counts of some promising lines are given in Table 2.

**Table 2.** Pedigrees and numbers of Sunn Pestfound on promising lines in Konya in1989-1990.

	No. of Sunn Pest adults, 4 <sup>th</sup> and 5 <sup>th</sup>
Pedigree	instar (8.4m <sup>2</sup> )
Hys"s"/7c/5/kvz/4/inia/3Cno//Elgav/Son64	12
Hys/7c//Tosun21	12
Hys/7c//Krc/3/Hno	12
No57//093-44/No57/3/TRS237	16
Qt254/Ravi66/mxp65/47Menk"s"/My48//4	22
-14/3/Yy 305	
Ksk//In1a/lfn/37Çalıbasan	15
Hys77c//kç66/3/Mu2	17
Pj62/Lut/3/D229-22/Nio//No64	14
Bl"s"/Au/3/A ri//Hys//c	11
Kç66/3/Au/Pj"s"//Gb/ykf	34
K66//Dibo/mfo	41
Au//yt54*2/N10B/3/II8260/4/Hys/7c//kç66	25
Au//yt54/N10B/3/ Grk	12
Tam102//Rd1/Sulu/Lom10/Co*2//co/3/San	19
tecatalina//Kenya125/R12-4-11	
Pato//63.130/67,2/3/p221-35/7907//Au	24
Arina	38
Odeskaya51/4/Son64//Ske/An <sup>2</sup> E/3/5x	18
Kç66//Skp35/B1	15
TamW108	22
Jar"s"Co652142//Lcr/3/zincirli/Au//Preska	26
C126-15/Cofn"s"/N10B/p14//	21
p101/4/Kç66	
Edch/Lifn"s"//Br <sup>2</sup> /3/Grk79	16
Osaga/Liakafen	11

#### **Cage Experiments**

Sunn Pest damage was observed on every variety but the rate of damage to grains varied with varieties. Proportions of damaged grains were 3.4% in Seri-88, 4.1% in Bezostaja-1, 7.1% in Atay-85, 7.2% in Kırkpınar-79, 8.37% in Ak-702, 9.3% in Kırac-66, 10.2% in Bolal-2973, 8.3% in BDME-128, 0.8% in BDME-157 (Da da ) and 11.4% in Gerek-79. Rate of damage was lower in hard red wheat than white soft. Paulian and Popov (8) reported that Sunn Pest attacked hard wheat less frequently than soft. Swallow and Cressey (10) reported that Sunn Pest damage in all samples of Karamu, which constituted 25% of the total wheat area in New Zealand in 1979, but they had not seen any damage on many other lines.

In conclusion, the following measures are recommended to keep Sunn Pest populations under control:

1. Forested areas, which provide natural protection to Sunn Pest egg parasitoids,

should be protected against deforestation for fuel or farming purposes.

- 2. To protect beneficial insects, extensive use of chemicals should be prevented. Chemical treatments should be considered only after determination of egg parasitoid ratios results of economic threshold surveys.
- 3. Multi crop agriculture should be given more importance.
- 4. Experiments showed that Sunn Pest is selective on varieties and the rate of damage varies from variety to variety. Hence, the entire region should not be planted with only one wheat variety.
- 5. Sunn Pest egg parasites, which are abundantly found in the region, should be multiplied in laboratories and released.
- 6. Varieties and lines that are extremely sensitive to Sunn Pest damage should not be used extensively in crossing (breeding) programs.

#### الملخص

يلديريم، أ. ف.، إ، كيناسي، و م. (إلمالي) يوسال. 2007. مواقع البيات الشتوي لحشرة السونة، الطفيليات والتأثير على سلالات وأصناف الحبوب. الصفحات 273–277.

تعد حشرة السونة إحدى أهم الآفات التي تؤثر على غلة ونوعية الحبوب في وسط الأناضول، تركيا. تم في هذه الدراسة تقصى مواقع البيات الشتوي، نسبة موت حشرات السونة في مواقع البيات الشتوي، وهجرة الحشرات إلى حقول الحبوب في محافظة قونيا. يسود النوعان Eurygaster maura و Eurygaster interوحت نسبة التطفل على البيوض بين 4 و 93.8% وكانت الطفيليات أكثر وفرة في المناطق المروية والكثيفة الأشجار. كان النوع Trissolcus grandis أكثر طفيليات البيض شيوعاً في المنطقة. أجريت تجارب ضمن الأقفاص وتجارب غربلة في الحقل لتحديد نفضيل حشرة السونة للأصناف. في تجارب الأقفاص وجد أن هناك اختلافاً بين الأصناف في نسبة الحبوب المتضررة بالآفة، وكانت الحبوب الحمراء القاسية مثل بعض الأصناف والسلالات مثل السلامة الحرية والحديث كانت كثافة الحوريات والحشرات البالغة الحديثة أعلى على بعض الأصناف والسلالات مثل السلامة مالمري و30-Gerek.

كلمات مفتاحية: قفص، Eurygaster maura، Eurygaster austriaca، موت طبيعي، مواقع البيات الشتوي، تطفل، تحمل، Trissolcus grandis.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

#### Effect of Temperature on the Development, Reproductive Potential, and Longevity of Trissolcus semistriatus Nees (Hymenoptera: Scelionidae) under Constant Temperatures

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#### Abstract

Tarla, S. and S. Kornosor. 2007. Effect of Temperature on the Development, Reproductive Potential, and Longevity of *Trissolcus semistriatus* Nees (Hymenoptera: Scelionidae) under Constant Temperatures. Pages 279-285.

Egg parasitoids are the most important natural enemies of Sunn Pest. *Trissolcus semistriatus* Ness is the most common species attacking eggs of *Eurygaster integriceps* Puton. (Heteroptera: Scutelleridae) in the south Anatolia. The effects 18, 22, 26, 30, and  $34\pm1^{\circ}$ C, L:D 16:8 photoperiod and  $65\pm5$  % rH. on the development, reproductive potentials, and longevity of *T. semistriatus* were evaluated, using *E. integriceps* eggs as the host. The developmental time was found to be inversely related to temperature. From egg to adult emergence varied from  $7.1\pm0.01$  to  $22.6\pm0.09$  days. for females and from  $6.1\pm0.03$  to  $20.7\pm0.08$  days for males. The theoretical lower developmental threshold and thermal requirement was estimated as  $11.8^{\circ}$ C, 138.8 degree-days (DD) for males and  $11.6^{\circ}$ C, 161.3 DD for females via linear regression. In all temperatures male parasitoids emerged one or two days before females. The mean number of progeny ranged from  $91.6\pm5.46$  to  $120.9\pm5.13$  offspring per female during their lifetime. Females lived significantly longer than males in all temperatures. Female longevity ranged from  $10.9\pm0.73$  days. at  $34\pm1^{\circ}$ C to  $67.9\pm2.44$  days. at  $18\pm1^{\circ}$ C, while males lived from  $8.8\pm0.87$  days. at  $34\pm1^{\circ}$ C to  $34.8\pm3.85$  days. at  $18\pm1^{\circ}$ C. Temperature did not significantly affect the sex ratio and emergence rate of *T. semistriatus*.

Keywords: Development, egg parasitoid, *Eurygaster integriceps*, temperature, thermal constant, *Trissolcus semistriatus*.

#### Introduction

The Sunn Pest, Eurygaster integriceps Puton, (Heteroptera: Scutelleridae), is the most important pest of wheat and other cereal crops causing severe losses in Turkey. Both nymphs and adults cause damage to plants, and reduce yields by feeding on leaves, stems and grain. Besides the direct reduction in yield, the insect also injects a salivary secretion into the grain. This secretion contains proteolytic enzymes that greatly reduce dough-baking quality (2, 20). In a batch if only 2-3% of the grains are contaminated by Sunn pest feeding the lot can be unacceptable for baking purposes (7). The critical threshold is ~3-5% (18, 21) and damage in excess of 2% cannot be acceptable for exporting products.

Parasitoids could play an important role in reducing Sunn Pest populations. Conservation and supportive augmentation of populations of egg parasitoids appears to offer high potential for biological control of Sunn pest in an integrated pest management (IPM) system (26). Obtaining *Trissolcus semistriatus* Nees from Sunn Pest eggs in the laboratory, release in different densities and evaluation of their effectiveness in the biological control of Sunn pest in wheat fields were described by Tarla and Kornosor (22).

Of all the climatic factors, temperature has probably the greatest effect on insect developmental rates (16, 23). Studies have shown that temperature influences several biological characteristics of insects such as adult life span, survival, fecundity, fertility (6, 27) and sex ratio (10). It is therefore necessary to determine how various environmental parameters affect the development of *T. semistriatus*.

The research herein was aimed at a better understanding of the influence of temperature on the duration of development, oviposition period, reproductive potential, longevity, emergence and sex ratio of *T. semistriatus* for use in mass rearing this insect.

#### **Materials and Methods**

Obtaining the host and eggs. Overwintered adult Sunn Pest were collected in wheat fields (37° 02' N 35° 21' W, and 93 m elevation) in Adana, Turkey in 2001. They were released into standard glass-topped, wooden sleeve cages (50 x 50 x 70 cm) on wheat planted in the pots and also released on cut off wheat plants in 8-liter plastic rearing cups covered by organdy cloth tops. Sunn Pest preferred to lay their eggs on napkin strips rather than on the leaves of wheat plants in the laboratory. To collect egg masses, napkin strips were removed from the plastic cups every day. Wheat plants were removed from the plastic cups and replaced with newly cut ones every other day. To mate and obtain enough eggs from adult Sunn Pest, the cages and plastic cups were placed in a chamber at 29±2°C, 16:8 (L:D) and 65±10 % RH.

#### **Obtaining Parasitoids**

Parasitized Sunn Pest egg masses were collected in wheat fields in late March and early April in the same province and years and incubated at  $26\pm1^{\circ}$ C, L:D 16:8, and  $65\pm5$  % rH. After parasitoids emerged from the eggs, they were removed and reared one generation on Sunn Pest eggs.

## Effect of Temperatures on Development of *Trissolcus semistriatus*

To determine the duration of development of *T. semistriatus*, 1-day-old egg mass (one egg mass=14 eggs) of *E. integriceps* were glued to

a paper index card strip (1 x 3.5 cm) and were exposed to 1-day-old copulated female parasitoids (F1) in 1.6 x 10 cm test tubes held at 26±1°C. This procedure was repeated 100 times, and after 12 h the adult parasitoids were discarded and 20 tubes were randomly assigned for placement in each of the temperatures studied. The egg masses from each tube were observed under a stereoscopic binocular microscope twice a day until parasitoid emergence. The time between oviposition and adult emergence of T. semistriatus females and males was recorded at the five constant (18, 22, 26, 30, and 34±1°C, 65±5% RH, and L:D 16:8) temperatures. The mean development times was established for each temperature treatment. For estimations of the thermal constant and lower developmental threshold of T. semistriatus females and males, a linear regression y=a+bx, was used, where y is the developmental rates (1/ developmental time), a and b are constants of the regression, and x is the temperature in °C. The lower temperature threshold (t) was estimated in a subsequent computation as t=-a/b and the result was confirmed with the regression equation, solving for t when y=0 (1). The thermal constant, DD, was determined as1/b (3).

# Effect of Temperatures on Reproductive Potentials and Longevity of *Trissolcus semistriatus*

*T. semistriatus* were maintained at the same five constant temperatures. For each temperature, 20 pairs (females + males) of parasitoids were used and each pair was isolated in test tubes. Before the experiment was conducted, individual female and male parasitoids were collected at emergence and kept together one day for mating in the test tubes at  $26\pm1^{\circ}$ C,  $65\pm5$  % RH and L:D 16:8. Females mate immediately after emergence, and are capable of successful oviposition a few days after emergence. Because *T. semistriatus* laid more eggs in the beginning of their oviposition period, in the first two days four (and in the other days two) egg masses of

E. integriceps, glued to a labeled index card strip, were offered to a pair of parasitoids (F1) in the test tubes for 24 h or until female parasitoids died. These were then placed in adjusted incubators (18, 22, 26, 30, and 34±1°C, L:D 16:8 photoperiod and 65±5% RH.). Upon emergence, nymphs of E. integriceps were removed. Young eggs (<24 h old) were used because eggs of this age are suitable for parasitoid oviposition. Trissolcus grandis Thomson prefers to oviposit in hosts in the early stages of their embryo (14). 10 % honey solution was streaked on the inside of tubes to provide a food source for the parasitoids. After all parasitoid emergence was completed; black eggs that failed to produce parasitoids were dissected. Male and female parasitoid mortality was recorded twice a day throughout the experiment (until all parasitoids had died). As a result the longevity of male and female, the lengths of oviposition period, fecundity, emergence rate, and sex ratio of T. semistriatus were recorded at each temperature.

#### **Data Analysis**

The data were tested using one-way analysis of variance (ANOVA) and means were separated by using Duncan's Multiple Range Test (< 0.05). Percent emergence of adult parasitoids and sex ratio were transformed to arcsin x before analysis.

#### **Results and Discussion**

## Effect of temperature on the development of *Trissolcus semistriatus*

*T. semistriatus* successfully completed its development (oviposition to adult emergence) in temperatures of 18-34°C. Male parasitoids developed faster than female at all five temperatures. Males emerged 1.0-2.2 days earlier than females. There was a significant difference in the developmental time of females (F=8603.74; df=4, 699; P<0.05) and males (F=980.59; df=4, 488; P<0.05) among temperatures.

From the calculated regression lines theoretical lower (Figure 1), the developmental threshold and thermal requirement was estimated at 11.8°C, 138.8 DD for males and 11.6°C, 161.3 DD for females. Male achieved higher rates of development at constant temperatures as described by equations of regression: The calculated regression for male and female were y=0.0072x-0.0849, R<sup>2</sup>=0.9715 and y=0.0062x-0.0717, R<sup>2</sup>=0.9786, respectively (Figure 1).



**Figure 1.** Developmental rate of eggs to adult of *Trissolcus semistriatus*. Male and female represents the linear regression analysis of developmental rate at five constant temperatures.

Determination of the thermal requirement of this parasitoid is important in estimating the number of generations per year. The theoretical lower developmental threshold and thermal requirement was calculated by linear regression for males and females. Many empirical and biophysical models describe the time versus temperature or the rate versus temperature curve of insect development. temperature Models of rate versus relationships are used widely to estimate insect developmental times because mean daily rates can be accumulated under fluctuating temperature environments. Linear regression of developmental rates vielded different estimates for the lower developmental

threshold and thermal requirement. Theoretically the more elaborate curvilinear fitting to developmental rate data yields the more accurate estimate of the relationship (11). However, the linear degree-day model has the advantage of simplicity and, on a practical level; linear models often seem to perform adequately in forensic entomological casework (12).

Voronin (24) reported that adults of T. grandis, simony Mayr, Telenomus Τ. *chloropus* Thomson, Trissolcus basalis Wollaston and T. histani Voegelé became inactive at 9.0°C, increasingly active from 10.0 to 11.0°C and start to parasitize their host eggs at 13.0°C. Telenomus remus Nixon became inactive at 10°C (19). There are many studies to determine the effect of temperature on scelionid egg parasitoids. The lower developmental threshold and thermal requirements were calculated as 13.7°C and 204.0 DD for *Telenomus busseolae* (Gahan) (15), and 12.1°C and 158.7 DD for T. lobatus Johnson and Bin (17), 11.9°C and 129.5 DD for *T. chrysopae* Ashmead (17). The thermal requirement of males and females was 126.5 and 139.4 DD for *Telenomus podisi* Ashmead (28), 118.2 and 142.2 DD for *Trissolcus euschisti* (Ashmead) (29), 157.0 and 163.0 DD for *Telenomus reynoldsi* Gordh & Coker (4).

# Effect of Temperatures on Reproductive Potential and Longevity of *Trissolcus* semistriatus

The study of the effect of temperature on reproductive potentials, and longevity of *T. semistratus* lead to a better determination of the most favorable thermal condition for the mass production of this species. *Trissolcus semistriatus* females began ovipositing on the first day after their emergence. Oviposition period varied from  $10.1\pm0.66$  to  $37.9\pm2.14$  with higher values for  $18\pm1^{\circ}$ C and lower values for  $34\pm1^{\circ}$ C and the difference was statistically significant (Table 1, F=97.59; df=4, 95; P<0.05).

	Oviposition	Adult long	gevity (days)			Sex ratio <sup>a</sup> (Females/ females +
°C	period (days)	Females	Males	Progeny	Emergence <sup>a</sup>	males)
18±1	37.9±2.14a	67.9±2.44a	34.8±3.85a	111.0±3.93a	81.9±0.91ab	51.5±1.44a
N	20	20	20	20	20	20
22±1	24.9±1.15b	31.4±1.74b	20.5±2.02b	120.9±5.13a	79.8±0.61b	52.3±1.89a
N	20	20	20	20	20	20
26±1	15.3±0.73c	16.2±0.72c	11.7±0.70c	117.5±3.99a	82.7±0.81a	55.2±1.72a
N	20	20	20	20	20	20
30±1	10.7±0.50d	14.9±0.97c	12.9±1.43c	114.2±4.92a	80.8±1.02ab	54.5±1.41a
N	20	20	20	20	20	20
34±1	10.1±0.66d	10.9±0.73c	8.8±0.87c	91.6±5.46b	82.6±0.60a	53.4±0.94a
N	20	20	20	20	20	20

**Table 1.** Duration of oviposition, adult longevity, progeny, emergence and sex ratio of *Trissolcus semistriatus* on the Sunn pest eggs (Mean±SE) at five different constant temperatures.

Means in column followed by the same letter are not statistically different by Duncan's Multiple Range Test (p<0.05). <sup>a</sup>Data transformed using arcsin x.

The longevity of adult females decreased with increasing temperature (Table 1). There was a significant difference at five temperatures for longevity of adult females (F=252.36; df=4, 95; P<0.05) and males (Table 1, F=25.35, df=4, 95; P<0.05). Males of *T. semistriatus* are not overwintered only mated and unmated females are overwintered. Adult longevity of this parasitoid female is longer overall than males at all temperatures. A long life span is important for egg parasitoids of pentatomids because their hosts move often from one habitat or host plant to another throughout a season (8).

The total number of progeny produced was lowest at  $34\pm1^{\circ}$ C and there was no significant difference except at  $34\pm1^{\circ}$ C among temperatures (Table 1, F=5.87; df=4, 95; P<0.05).

Parasitoid emergence success was high. The difference was not significant statistically in relation to emergence recorded in the five temperatures (F=2.27; df=4, 95; P>0.05). Black eggs that failed to emerge were dissected and it was observed that parasitoids had completed their development but did not emerge and died because of the upside down head position in the host eggs. The emergence was high because of using 1-day-old host eggs (<24 h old). The emergence on the *E*.

*integriceps* eggs was calculated as 98.6% at the same temperature (13) and also the emergence of *T. grandis* from *E. integriceps* eggs was calculated as 98.7 % (5).

The sex ratio was not significantly affected by temperatures (F=0.98; df=4, 95; P>0.05) and these results are similar to those recorded by Foerster and Butnariu (9) for *Telenomus cyamophylax* Polaszek. The sex ratio of *T. semistriatus* was distinctly femalebiased. Knowing what factors influence the sex ratio is important for rearing parasitoids successfully, either in quarantine or in commercial insectaries for their subsequent release (25).

These data presented here combined with field studies should be valuable to elucidate the factors influencing the population dynamics of this species in the field and in any future attempts at mass production for biological control of Sunn Pest.

#### Acknowledgements

University of Cukurova Research Fund financially supported this research. I thank M. R. Ulusoy for permission to use many of the facilities within their laboratories.

تارلا، إس. و إس. كورنوسر. 2007. تأثير الحرارة على التطور، القدرات التناسلية، وطول فترة الحياة للطفيل -279 (Hymenoptera: Scelionidae) تحت درجات الحرارة الثابتة. الصفحات 279- 285.

تعد طفيليات البيض من أهم الأعداء الطبيعية لحشرة السونة (Eurygaster integriceps Puton)، ويعد الطفيل Trissolcu semistriatus Nees النوع الأكثر شيوعاً الذي يهاجم بيوض حشرة السونة في جنوب الأناضول، تم تقييم تأثيرات درجات الحرارة 18، 22، 26، 30 و 34±1° س، الفترة الضوئية 16: 8 (ضوء : ظلام) والرطوبة النسبية 65±5% على التطور، القدرات التتاسلية، وطول فترة الحياة للطفيل T. semistriatus الضوئية 16: 8 (ضوء : ظلام) والرطوبة النسبية التطور مرتبط عكسياً بالحرارة. حيث اختلف من البيضة حتى انبثاق البالغة من 1.1±00 إلى 22.6±00 يوم للإناث ومن 1.0±00 إلى 20.7±0.00 يوم للذكور. قدرت عتبة التطور الدنيا النظرية والمتطلبات الحرارية به 11.8° س، 138.8 درجة حرارة مؤثرة يومية (DD) للذكور و 16.1°س، 16.3 (DD) للإناث بواسطة الانحدار الخطي. تتبثق الطفيليات الذكور قبل الإناث بيوم واحد أو يومين عند جميع درجات الحرارة. تراوح متوسط عدد النسل من 91.6±5.46 إلى 120.9±5.13 فرداً للأنثى من خلال فترة حياتها. عاشت الإناث لفترة أطول بشكل معنوي من الذكور عند جميع درجات الحرارة. تراوحت فترة حياة الأنثى من 0.15±5.13 يوم عند درجة الحرارة 81±1 °س، بينما عاشت الذكور من 0.8±8.8 يوم عند درجة الحرارة 18±1 °س، بينما عاشت الذكور من 8.8±8.8 يوم عند درجة الحرارة 18±1 °س. لم تؤثر درجة الحرارة بشكل معنوي في عند درجة الحرارة 11.5 فرداً للأنثى من 10.5 عنوي من الذكور عند جميع درجة الحرارة 11.5 فرداً للأنثى من 10.5 عنوي من الذكور عند حميع درجة الحرارة 12.5 فرداً للأنثى من 10.5 عنوي من الذكور عند حميع درجة الحرارة 11.5 للذكور من 10.5 عند درجة الحرارة 12.5 الأورة من الذكور من 10.5 عنوي من الذكور من معنوي من الذكور من الذكور من معنوي من الذكور من الذكور من الذكور من 10.5 عند درجة الحرارة 11.5 من الذكور من الذكور من 10.5 عنوي من الذكور من 10.5 عنوي من الذكور من الذكور من 10.5 عنوي من الذكور من الذكور من الذكور من 10.5 عنوي من الذكور من معنوي من الذكور من الذكور من 10.5 عنوم عند درجة الحرارة 15±1 من الذكور من الذكور من معنوي من الذكور من 10.5 عنوم عند درجة الحرارة 15±1 من الذكور من 10.5 عنوي من الذكور من الذكور من الذكور من 10.5 معنوي من الذكور من 10.5 معنوي من الذكور من 10.5 من الذكور من الذكور من الذكور من 10.5 من الذكور من 10.5 من الذكور من الذكور من 10.5 معنوي من الذكور من الذكور من الذكور من الذكور من 10.5 معنوي من الذكور من الذكور من الذكور من الذكور من 10.5 معنوي من الذكور من من الذكور من

كلمات مفتاحية: تطور، طغيل بيض، Eurygaster integriceps، حرارة، ثابت حراري، Trissolcu semistriatus.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

#### Mass Production of Entomopathogenic Fungi for **Biological Control of Insect Pests**

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#### Abstract

#### Jenkins, N.E., B.S. Ali and D. Moore. 2007. Mass Production of Entomopathogenic Fungi for **Biological Control of Insect Pests. Pages 287-293.**

A research program to develop a mycoinsecticide to control Sunn Pest has made good progress and it is necessary to plan ahead for some aspects of practical use. One of the most important of these is mass production. Control over the areas susceptible to Sunn Pest damage will require large quantities of high quality fungal material, suitably formulated for long term storage and ease of use. The scale-up of production beyond experimental facilities brings many challenges. CABI Bioscience has extensive experience derived from setting up production facilities, in Benin and Trinidad, providing consultancy support for commercial producers in South Africa, France and USA and working with many small scale producers in the Americas, Africa, Asia and Europe. Lessons from this experience, in terms of quantity of production and quality assurance procedures will be reported. Long term storage stability can be achieved through a combination of downstream processing measures and the selection of suitable packaging, thus ensuring predictable product viability even under fluctuating temperature conditions. Attention to all aspects of the production process will ensure that consistent and effective products are reliably available for sustainable pest management. Keywords: Beauveria bassiana, mass production, mycoinsecticide, quality control, Sunn Pest.

#### Introduction

Research has shown good potential for using Beauveria bassiana (Balsamo) Vuillemin as a mycoinsecticide to control Sunn Pest. An oil formulation, sprayed onto a maturing wheat crop, caused significant mortality of the pest during trials conducted at ICARDA in 2004 (4). This increases the possibility of developing a product which can be used on a sufficiently large scale to be economically viable. However, the scale of production is often underestimated; for example, application over one million hectares may require 50-100 tons of dry spore powder depending on dose used; produced from 2,000 tons of substrate. An effective mass production and quality control system is essential if the fungal agent is to be produced as a viable and consistent product. Production under stringent quality control protocols will maximise product

performance. ensure product safety, standardise manufacturing costs and reduce the risks for supply failure, thus building user confidence (9). If these are lacking, output is uncontrolled and the final product can have variable concentrations of active agent, resulting in variable product performance. Erratic and inconsistent performance of mycoinsecticides is a significant factor in their limited successful commercialisation (11). Mycoinsecticides have not achieved a consistent level of efficacy comparable with that of their chemical counterparts, or with that of the leading bacterial agent, Bacillus thuringiensis despite potential superiority in terms of insect control (12).

Modern advances have allowed industry to produce entomopathogens in specially designed, sealed aerated bioreactors with automated process control along similar principles to those of liquid fermentation. This

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is capital expensive but reduces labor costs and allows close control and standardization of the process. In other countries different economic constraints exist and the capital costs of automated processes may be unacceptable. There are many labor intensive production systems worldwide, particularly in Latin America and China (5), supplying sufficient quantities of product for niche markets in their immediate area.

Two production systems are described in this paper; both are of an intermediate technology level, producing high quality material in sufficient qualities to satisfy local demand. These may be suitable models for the Sunn Pest, where local isolates may be used for individual countries or even for regions with specific environmental conditions.

#### The LUBILOSA System

The LUBILOSA project developed a successful mycoinsecticide to control locusts and grasshoppers, specifically for Africa, although the technology is now in practical use in Australia. *Metarhizium anisopliae* var. *acridum* Driver & Milner was shown to be effective under desert conditions, when formulated, and mass production was required to produce enough inoculum for experimental and small scale field needs.

The LUBILOSA production facility is housed in a purpose-built wing of the International Institute of Tropical Agriculture (IITA). Plant Health Management Division in Cotonou, Republic of Benin. A feature of the LUBILOSA system is that the purchase of expensive capital equipment has been avoided wherever possible and low-cost, locally available alternatives are used in preference. The production process is a typical two stage system in which liquid inoculum is produced in shake flasks and used to inoculate autoclaved rice, on which conidiation occurs. This combines the benefits of high biomassproduction in liquid fermentation and production of stable, hydrophobic, aerial conidia on a solid substrate. The technology, although specifically for *M. anisopliae* var *acridum*, is suitable for a range of mitosporic fungi and requires modestly equipped laboratories (8).

Fungal biomass is produced in a simple liquid medium of waste Brewer's Yeast and sucrose in shake flasks. After 2-3 days of fermentation, the liquid biomass is transferred to par-boiled autoclaved rice. Water is added to give a 35-40% moisture content, after inoculation with the fungal biomass. One kilogram of rice treated as above is placed in locally produced autoclavable plastic bags (obtained from the local market at a fraction of the cost of laboratory quality autoclavable bags) and placed in stackable plastic bowls. After 10 days of fermentation, the bowls are opened and re-stacked to allow the contents to air-dry in a dehumidified room for at least 48 hours. Conidia are extracted from the rice using a cyclone extractor and further dried to 5% moisture content. These are packed into tri-laminate (aluminium:plastic:aluminium) packets and will remain viable at 30°C for over a year and, refrigerated, for six years or more.

The LUBILOSA production system represents one of the more complicated mycopesticide production scenarios. The conidia must be completely separated from the substrate, in order to meet the requirements of formulation for controlled droplet application, as may be applicable to Sunn Pest control.

The concentration (density) of conidia in the end product (pure conidial powder) is  $5x10^{10}/g$  and the field application rate is  $5x10^{12}$  conidia/ha. Average yields are 31.1g of dry conidia powder/kg of rice substrate and annual production is 300-350 kg dry powder (3) equivalent to 3000-6000 ha depending on host target and hence dose.

The LUBILOSA Program's laborintensive model and a capital intensive model (developed by Mycotech Corporation) have been compared (13). The capital intensive system required an output sufficient for at least 80,000 ha per year to reach economic viability. The labor intensive production system was more economical at output levels equivalent to 20,000 ha per year. Consequently a simpler, labour intensive production system may prove appropriate, particularly for niche market products or for pests occurring over a range of environments where particular isolates may be needed.

#### The CARONI System

Metarhizium anisopliae (Metschn.) was developed for the management of the sugar cane froghopper, Aeneolamia varia saccharina Distant, a serious wet season pest of sugar cane in Trinidad. This project was conducted at the Caroni Research and Development Division - the national sugar cane research institute in Trinidad and Tobago.

The production facility is located in the central region of Trinidad and Tobago and consists of a single-storey concrete facility, two annexes and a cold storage container. The main building includes an enclosed production area, laboratory and administrative facilities. The annexes house a loading and silo storage area, harvesting and distribution areas and utility buildings. The main plant equipment includes 2 steam-reliant type autoclaves, a diesel-fuelled boiler with the accompanying water treatment system, incubators, clean benches, microscopes, an in-house designed and built spore-dryer and a CAB International designed and constructed cyclone harvester. All of the production operations and related laboratory activities are conducted in-house.

The production process is similar to the LUBILOSA system and includes the discrete activities of sterilisation, inoculation, incubation under strain-specific environmental conditions, drying, cyclone extraction of conidia, packaging and storage/distribution. However, the scale of the operation is quite different. In this system there is a substrate handling capacity of 250 kg per batch and a weekly throughput of 3000 kg. The production unit operates in 2-12 hour shifts with each shift having a complement of 20 persons

including supervisory, technical and skilled labour. Therefore while the processes are similar and follow the same flow, the standard different operating practices are and encompasses the transition of basic insect pathology laboratory procedures and techniques into a successful scaled up commercial operation. The transition of these activities required careful revision of inputs and outputs, development of an efficient system flow and the implementation of a detailed quality control program that encompasses all activities.

A combination of unpolished rice and tap water is packaged in autoclavable plastic bags. These bags are manufactured within the facility. The bags are partially sealed and stacked in the autoclave and sterilized for 1 h after which they are removed, shaken to disperse clumps and cooled. Three to 5 d old liquid inoculum, produced in yeast and glucose, is used to inoculate the sterilized rice within the plastic bags. The bags or inoculated rice are shaken to facilitate distribution of the inoculum and then resealed with micropore tape to allow airflow. For conidia production, the inoculated rice is incubated for 14 d during which the bags are intermittently shaken to break up clumps and encourage aeration. For harvesting, one face of the bag is cut away and the bags are then placed in a cool dehumidified room for 48 h. Conidia are extracted with the industrial model cyclone extractor and then placed for further drying in the dryer. The conidia are then sealed in plastic bags and stored at 4°C.

Prior to 1995, the laboratory scale production system was being used and there was an annual production of 10-40 kg of spore powder with varying viability and purity. At the conclusion of the 10-year period of R&D (1991-2001) the annual production increased to 1500-2000 kg of spore powder of a high viability, virulence and purity. The rice substrate yields an average of 45 grams of dry conidia per kilogram of rice with an end product concentration of  $5 \times 10^{10}$  conidia per gram.

# Production and Quality Control of Mycoinsecticides

An effective isolate is essential for the development of a mycoinsecticide. Not only must the fungal spore successfully complete the complex process of infection, it must also be capable of storage before use and, perhaps, of persistency in the environment. The latter requires resistance to environmental influences, of which ultra-violet irradiation is probably the most serious. Efficacy, at each stage, is influenced by production system. Quality control systems for biological agents are often more comprehensive and specialised than those used for synthetic chemical insecticides where a simple chemical analysis can be used (9).

#### **Culture Maintenance**

Fungal isolates should be lodged with a recognised culture collection and formally identified, preferably both on morphological and molecular characters. A limited number of species are likely to be considered for mycoinsecticide use, but the number of isolates of each species is, in effect, infinite and there is a need to identify the individual isolate. This provides a mechanism for tracking an agent in the environment (2), to check the purity of formulations, and to provide a reference for registration.

Careful management of fungal material from the point of original isolation can reduce the danger of character loss through serial subculturing. Sub-cultures from a clean, uncontaminated culture should be stored, using as many long-term storage methods as possible, ideally including lyophilisation (freeze drying), cryopreservation (using liquid nitrogen) and storage at -80°C. Techniques requiring less sophisticated equipment include silica gel storage, storage under mineral oil, soil storage and water storage can also be effective and can be used as a back-up to the above techniques. The regular passaging of the fungal isolate through the host or closely related species reduces the risk of loss of valuable characteristics, although this carries a risk of contamination. Another technique is the taking of single spore isolation.

# Production Process and Record Keeping

procedures Standardised for process monitoring and recording should be applied to ensure consistent production of reliable, high quality products. In countries where product registration is enforced, a complete description of the process used for production of the agent is generally required. In-process monitoring and quality control should be an integral part of this description. Culture conditions affect many aspects of pathogen performance, including virulence, storage longevity and durability (1, 6, 7); records should be kept for each production batch and maintained on file so that the information gathered can be backchecked according to batch number as required (9).

Important parameters include temperature, both for yield during production and durability during the drying process, moisture content adjusted to give optimum production (10), pH and contamination monitoring. The first three have some error for margin, although yield loss may occur, but contamination may cause the loss of a whole production run if it occurs early in the process. Careful monitoring for contamination, at each stage, reduces lost production time required to eradicate the problem (9). If contaminants have entered the system, the batch should be discarded immediately, reducing the risk of introducing high levels of contamination into the production environment.

#### Safety

The intrinsic safety of a mycoinsecticide can be evaluated by carrying out the appropriate mammalian and ecotoxicological safety tests; in countries where registration is not required, microbial products may be produced without such tests. In addition to the safety aspect of the fungal isolate itself, a number of safety issues arise with respect to microbial contamination of the final product. Carrying out specific tests for all such organisms on a batch by batch basis is impractical but standards for biological purity should be set.

#### Efficacy

Efficacy is obviously critical; high viability and virulence/pathogenicity, increase the chances of successful field performance. Viability is easily measured in fungi; with percentage germination of the conidia being Virulence/pathogenicity determined. is generally verified with a standard bioassay using the target insect. The dose/dosage used should be low to enable differences in virulence/pathogenicity to be detected and bioassays should always be run against a standard batch of product as the quality of the test organism can vary over time. Changes in the production process require further series of assays to identify resultant changes in product efficacy. То ameliorate variability, performance of a test sample should be considered in relation to that of the standard, using a ratio rather than absolute comparisons. Bioassay data should be archived to assess the temporal variation in assays.

#### Storage

Shelf life is critical in the use and acceptance of microbial biocontrol products. Applying equations and experimental protocols used for estimating seed longevity, it is possible to define constants for conidia of individual species of mitosporic fungi and thus predict the longevity of fungal products in storage (7). Packed in hermetically sealed, foil lined sachets at low moisture content (<5%), conidia of *M. anisopliae* var. *acridum* can be stored for up to a year at 30°C and >6 years under refrigeration. Conidia of different fungal species and of isolates of a single species, can vary in their absolute longevity, but the relative effect of moisture content and temperature on the life span of conidia is similar across species.

Most important with regard to shelf life is that each batch of product should perform similarly to the next during long-term storage. To ensure that this is the case, nutritional conditions, method of down stream processing, final moisture content of the product and packaging should be standardised. Providing these parameters are consistent for all batches, storage temperature becomes the primary factor in determining likely shelf life.

#### **Specifications**

Specifications over and above the standards laid out above include information such as the amount of active ingredient (a.i.) per gram or millilitre of product (e.g. conidia  $g^{-1}$  dry powder or ml<sup>-1</sup> formulation). Further specifications might include particle size spectrum, which is critical for many application techniques and other physical properties related to the formulation such as suspensibility, wettability, flowability and emulsion stability.

#### Conclusions

Experience has shown that many commercial products are of poor quality. High levels of contamination were found in five of six products from China, Indonesia, India and Columbia, there was low content of active ingredient in two and viability of less than 10% in three. None matched the standards of the LUBILOSA "Green Muscle" (9, 10). However, high quality products can be produced in less wealthy countries using low technology systems, as illustrated by the production of the *Metarhizium* product 'Green Muscle<sup>®</sup>' in west Africa (3, 10). This requires the establishment of strict, internal quality standards and appropriate protocols, which are

enforced by the production manager as part of the production process.

The effective mass production of mitosporic fungi is possible under relatively low technology conditions, to exploit situations where labour costs are low. Such mass production sites could be a valuable source of employment. The production, for perhaps 20,000-40,000 hectares for local use, would also allow economies in terms of transport. However, although the necessary

systems for effective production are straight forward, their strict observance is often neglected, leading to failures.

#### Acknowledgment

The UK Department for International Development funded work on Sunn Pest control.

#### الملخص

جينكينز، ن. ي.، ب. إس. علي وديف مور. 2007. الإنتاج الكمي للفطور الممرضة للحشرات من أجل المكافحة الحيوية للآفات الحشرية. الصفحات 287-293.

حقق برنامج بحثي لتطوير مبيدات فطرية لمكافحة حشرة السونة تقدماً جيداً ومن الضروري وضع خطة متقدمة لبعض أوجه الاستخدام التطبيقي. أحد أكثر هذه الأوجه أهمية هو الإنتاج الكمي. سوف نتطلب مكافحة المناطق الحساسة لضرر حشرة السونة كميات كبيرة من مواد فطرية ذات نوعية عالية، محضرة بشكل مناسب للتخزين لفترة طويلة وسهلة الاستخدام. زيادة الانتاج أكثر من الحدود التجريبية سيجلب تحديات عديدة. لدى CABI للعلم الحيوي خبرة واسعة نشأت من تشبيد وحدات إنتاج، في بينين وترينيداد، نقديم دعم استشاري للمنتجين التجاريين في جنوب أفريقيا، فرنسا والولايات المتحدة الأمريكية والعمل مع العديد من المنتجين الصغار في الأمريكيتين، أفريقيا، آسيا وأوربا. سيتم تدوين ما تم تعلمه من هذه الخبرة من حيث كمية الإنتاج ونوعية إجراءات السلامة. يمكن تحقيق الثباتية عند التخزين طويل الأمد عبر الجمع بين توجيه ضوابط العملية واختيار التعليب المناسب، وبالتالي ضمان القدرة على التنبؤ بحيوية المنتج حتى تحت ظروف الحرارة المتذبذبة. سيضمن الاهتمام بجميع أوجه

كلمات مفتاحية: Beauveria bassiana، إنتاج كمي، مبيدات حشرات فطرية، تحكم بالنوعية، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

#### Mass Production and Storage of Trissolcus grandis (Thomson) (Hymenoptera: Scelionidae)

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#### Abstract

# Kodan, M. and M.O. Gurkan. 2007. Mass Production and Storage of *Trissolcus grandis* (Thomson) (Hymenoptera: Scelionidae). Pages 295-301.

In this study, mass production of *Trissolcus grandis* (Thomson) on stored eggs of *Dolycoris baccarum* L. (Heteroptera: Pentatomidae) alternative host of the parasitoid, was investigated. Fresh eggs of *D. baccarum* were stored at  $-18\pm1^{\circ}$ C for 8 month. These eggs were parasitized and placed in a chamber maintained at  $25\pm2^{\circ}$ C,  $65\pm5^{\circ}$  RH and 16:8 L:D for parasitoid emergence. Parasitization rate and adult parasitoid emergence from the eggs were 50%. The longer the eggs were stored, the longer the parasitoid development and emergence periods were, but the less the emergence rate of female parasitoids was. The longevities of parasitoids obtained from stored eggs were not different when compared with the controls. Parasitized 6-8 day old and 10-12 days old *D. baccarum* eggs were stored at  $5\pm1^{\circ}$ C in a refrigerator for 5-60 days and parasitoid emergence on the  $25^{\text{th}}$  day. The emergence period was similar in stored eggs and controls. Longevity was shorter in parasitoids obtained from stored eggs when compared with controls. All examined parameters were similar both in 6-8 day old and 10-12 day old parasitised and stored eggs.

Keywords: Dolycoris spp., mass rearing, storage, Trissolcus spp.

#### Introduction

Sunn Pest have been recognized as serious pests of wheat and other cereal crops. The most important biological control agent of Sunn Pest is the egg parasitoid, *Trissolcus* spp. Egg parasitoids of Sunn Pest can cause high levels of mortality under natural conditions, and this makes them good candidates for augmentation. It is necessary to mass produce *Trissolcus* spp. and release them to increase effectiveness. The parasitoid is effective in some provinces in Turkey.

*Trissolcus grandis* Thomson mass rearing research was done on the eggs of the alternative host *Dolycoris baccarum* L.

#### **Material and Methods**

*Dolycoris baccarum* was reared on seed of tobacco, soybean, peanut, sunflower, and *Diplotaxis tenuifolia* (L.) Dc. Emphasis was

placed on the storage of parasitized and fresh eggs.

#### Storage of Fresh Eggs of D. baccarum

Fresh eggs of *D. baccarum* were stored in a deep freezer at  $-18\pm1^{\circ}$ C for 8 months. Eggs were put out each five days and then exposed to two pairs of female adult and one male adult *T. grandis* in a glass tube hold in incubator. Eggs were inspected daily and after 2 days, parasitoids were removed. The duration of parasitoid development, parasitism rate, emergence rate, sex ratio and longevity were recorded.

#### Storage of Parasitized D. baccarum Eggs

Six to eight days old and 10-12 days old *D.* baccarum eggs parasitized by *T. grandis* were stored at  $5\pm1^{\circ}$ C in a refrigerator for 2 months. The parasitized eggs were removed at 5 different time periods and placed in a chamber. With daily inspections the length of parasitoid development, emergence rate, sex ration and longevity were recorded.

Experiments were conducted in an environmental chamber maintained at 25±2°C, 65% RH and 16:8 L:D. Statistical analysis was performed according to the Duncan test.

#### **Results and Discussion**

#### Storage of Fresh Eggs of D. baccarum

Following storage of the eggs the parasitization rate was 50%, except those eggs stored for 35-60 days (Table 1, Figure 1). As storage periods were increased, parasitization of the eggs decreased. Popov (4) found that parasitism rates were 94-54% on eggs of *Dolycoris baccarum* stored at  $-20^{\circ}$ C for 30, 90, 180, and 270 days and he explained

that these eggs might be stored 180-270 days and still be useful. Correa-Ferreira *et al.* (1) stored eggs of *Nezara viridula* (L.) (Het.: Pentatomidae) at -15°C for 0-360 days. From zero to 180 days storage the average parasitization of eggs was 63.75% and after 180 days it decreased to <39%. Safavi (6) emphasized that eggs of *Eurygaster* spp. were stored for12 months, and 8 months later 50% of these eggs lost property of parasitisim.

Adult parasitoid emergence was 50%, except when eggs were stored for 35-60 days. When the storage period of eggs was increased, parasitoid emergence decreased. These data agree with information of Popov (4) that parasitoids emergence from D. *baccarum* eggs stored at -20°C decreased as the storage period increased.

Table 1. Development of Trissolcus grandis on different period storaged Dolycoris baccarum eggs.

	Main ± SEM (Min-Max)							Main ± SEM	(Min-Max)
	Development time of	Development		Emergence	Parasitoio	l rate (%)	Sex	Longevi	ty (day)
Storage period (days)	female parasitoid (day)	time of male parasitoid (day)	Parasitism rate of eggs (%)	rate of parasitoids (%)	Female	Male	rate Female -Male	Female	Male
5-30	20.1±0.27 ab (16-27)	17.7±0.19 b (12-25)	71.3±4.9 b (21.43-100)	69.2±4.93 b (16.7-100)	22.5±4.8 b (0-100)	77.5±4.8 (0-100)	1:3.13	35.2±1.7 c (7-58)	19.2±1.3 d (4-49)
35-60	19.6±0.34 bc (16-24)	16.2±0.16 d (12-23)	48.8±4.6 c (10-100)	47.2±4.50 d (0-100)	15.9±3.4 b (0-61.5)	80.7±4.3 (0-100)	1:4.6	38.3±1.1 bc (22-51)	27.9±1.2 a (9-46)
65-90	18.7±0.24 d (15-26)	15.9±0.11 d (11-22)	55.9±4.6 bc (5.88-100)	52.3±4.39 bcd (5.9-100)	25.5±4.6 b (0-75)	74.5±4.6 (25-100)	1:2.65	40.8±1.3 b (20-63)	(29.9±1.2 a (9-47)
95-120	19.1±0.26 cd (17-26)	17.0±0.15 c (13-25)	54.8±3.9 bc (10-100)	53.9±3.98 bcd (25-100)	14.3±2.9 b (0-60)	85.7±2.9 (40-100)	1:4.35	39.4±1.6 bc (17-57)	30.4±1.0 a (15-46)
125-150	19.5±0.29 bcd (16-27)	16.3±0.12 d (13-24)	66.9±4.5 b (10.7-100)	66.7±4.10 bc (15.8-100)	18.0±4.2 b (0-100)	82.0±4.2 (0-100)	1:5.02	48.2±2.3 a (16-68)	28.3±1.4 a (7-56)
155-180	(10.27) 19.9±0.31 bc (15-25)	17.4±0.20 bc (12-28)	64.4±3.8 bc (18.2-100)	60.7±4.10 bcd (18.2-100)	23.6±4.3 b (0-81.8)	76.4±4.3 (18.2-100)	1:2.94	39.8±1.7 bc (16-62)	22.9±1.0 bc (6-41)
185-210	20.3±0.54 a (0-25)	17.4±0.16 bc (13-29)	58.6±5.3 bc (6.7-100	51.7±5.12 cd (6.7-92.9)	12.8±4.2 b (0-71.4)	87.3±4.2 (28.6-100)	1:4.5	34.5±1.8 c (0-65)	20.8±1.1 (7-39)
215-245	21.0±0.24 a (16-24)	18.5±0.13 a (14-25)	64.6±4.5 bc (11.1-100)	60.4±4.42 bcd (11.1-100)	17.3±3.9 b (0-81.8)	82.7±3.9 (18.2-100)	1:3.77	34.5±1.5 c (12-63)	$20.5\pm1.1$ cd (6-38)
Control (0 day)	16.7±0.29 e (14-22)	12.8±0.14 e (11-16)	97.6±1.5 a 92.6-100	95.8±1.87 a (90.9-100)	46.0±6.3 a (28-60.7)	53.9±6.3 (39.3-72)	1:1.17	44.3±1.3 ab (37-50)	$26.7 \pm 2.8$ ab (15.40)

P<0.05 values in lines followed by the same letters are significantly different and statistically are important. Note: The column values aren't comparison each other.



**Figure 1.** Parasitism and emergence rate of *Trissolcus grandis* from *Dolycoris baccarum* eggs stored for different periods at  $-18\pm1$  °C.

The sex ratio of individuals parasitizing fresh (not stored) eggs was 1:1.17. This ratio changed as storage time increased. As it increased more males and fewer females appeared (Table 1, Figure 2). But this was not consistent. This result may be caused by properties of eggs and their storage time. Correa- Ferreira *et al.* (1) reported a decrease in numbers of emerging females from eggs of *N. viridula* stored at -15°C for 0-360 days.

The shortest parasitoid development period was  $16.70\pm0.29$  days. This period was for females from the control eggs (not stored). Male development took an average  $12.77\pm0.14$ days. The longer the eggs were stored, the longer the parasitoid development period (Table 1, Figures 3 and 4). Similar result w obtained by Safavi (6) who reported development of *T. grandis* at 25°C took 14-16 days. Development of *T. basalis* (Wollaston) at  $26\pm3$ °C took  $13.5\pm1.6$  days (5).

Parasitoid development period on stored eggs, is importent for any release strategy.

There were differences in male and female longevity from eggs of *D. baccarum* 

not stored and stored for 8 mth. (Table 1). This difference was 10 days for females and 6 days for males. The longevity of *T. grandis* was 44.27 days for control females and 26-70 days for males. Correa Ferreira *et al.* (1) reported that the longevity of *T. basalis* was 42.6 days. Porta (5) implied that longevity of *T. basalis* was 31-6 days. The longevity of females and males obtained from stored eggs was shorter than obtained from nonstored eggs. This is important for future biological control studies.

## Storage of *D. baccarum* Eggs Parasitized with *T. grandis*

Parasitized 6-8 days old and 10-12 days old *D*. baccarum eggs were stored at  $5\pm1^{\circ}$ C in a refrigerator for 5-60 days. Parasitoid emergence did not occur until 20 days and no emergence occured on 25 days.

#### Parasitized 6-8 day old D. baccarum Eggs

The emergence period was different in stored eggs. This period was 9-11 days for females and 7-11 days for males (Table 2).



**Figure 2.** Sex ratio resulting from growing *Trissolcus grandis* on *Dolycoris baccarum* eggs stored for different periods at  $-18\pm1$  °C.

Table 2. Development of Trissolcus grandis on parasitized 6-8 day old Dolycoris baccarun	<i>i</i> eggs
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		Ma	_	Main (Min	± SEM -Max)			
Storage	Emergenc parasito	e period of ids (day)	Emergence rate of Parasitoid rate (%)			Sex rate	Longevity (day)	
period (days)	Female	Male	parasitoids (%)	Female	Male	Female- Male	Female	Male
5	10.3±0.3 c (9-13)	7.7±0.2 b (5-12)	94.4±7.8 a (57.1-100)	27.7±13.0 (0-83.3)	72.3±13.0 ab (16.7-100	1:3.25	23.5±2.3 b (8-28)	12.4±2.4 b (6-27)
10	11.1±0.4 b (8-14)	7.7±0.3 b (5-17)	70.2±11.6 a (27.8-100)	20.7±6.4 (0-40)	79.3±6.4 ab (60-100)	1:4.85	26.4±1.8 b (22-33)	13.0±2.3 b (6-27)
15	12.1±0.3 a (11-14)	9.2±0.2 a (6-13)	80.7± 4.2 a (71.4-100)	21.5±9.7 (0-62.5)	87.8±5.3 a (66.7-100)	1:6.33	27.3±3.8 b (3.77)	10.1±2.0 b (4-18)
20	9.5±0.3 d (8-12)	7.4±0.3 c (6-9)	35.2±17.1 b (0-100)	14.8±11.3 (0-68.7)	35.2±18.2 c (0-100)	1:1.38	13.4±2.4 c (6-24)	17.3± 3.9 b (6-26)
0	11.1±0.3 b (8-16)	7.5±0.3 c (5-11)	100± 0.0 a (100-100)	51.4±3.6 (36.4-63.6)	48.6±3.6 bc (36.4-63.6)	1:1.06	42.2±1.5 a (35-48)	25.5±2.8 a (14-38)

P<0.05 values in lines followed by the same letters are significantly different and statistically are important.

Adult emergence rate was effected by the 20d storage period. The emergence rate was the highest in the control, and the lowest on stored eggs stored for 20 days (Table 2, Figure 5). Six day-old *Ephestia kuehniella* Zell. eggs parasitized by *Trichogramma embryophagum* Htg. and *T. turkeiensis* Kostadinov might be stored at 8°C for 2 mths (2).

The storage period has also effected sex rate. When storage period increased numbers of female decreased. During the different storage period, female rate were found between 27.70-14.80% and female rate of control was 51.40%.

Longevity was shorter for parasitoids obtained from stored eggs when compared

with the control. When fresh eggs of *D*. *baccarum* were given these adults, parasitism was not different.



**Figure 3.** Interaction of the development period of female parasitoids grown on *Dolycoris baccarum* eggs. storaged for different periods at  $-18\pm1^{\circ}$ C.



**Figure 4.** Interaction of the development periods of male parasitoids grown on *Dolycoris baccarum* eggs. storaged for different periods at  $-18\pm1^{\circ}$ C.



**Figure 5.** Emergence rate from stored eggs of 6-8 days old *Dolycoris baccarum* parasitized with *Trissolcus grandis*.

**Parasitized 10-12 day old** *D. baccarum* eggs The parasitoid emergence period from 10-12 day old eggs stored at  $5\pm1^{\circ}$ C was 5-7 days for females and 2-4 days for males (Table 3).

Adult emergence rate from parsitized 10-12 days old *D. baccarum* egg decreased as the storage period increased (Table 3, Figures 6 and 7). The lowest adult emergence was 45.1% from eggs stored 20 days.



Figure 6. Emergence rate from stored eggs of 10-12 days old *Dolycoris baccarum* parasitized with *Trissolcus grandis*.



**Figure 7.** The relationship between storage period and emergence rate of from stored eggs of 10-12 days old *Dolycoris baccarum* parasitized with *Trissolcus grandis* 

Storage period effected parasitoid emergence. Talhouk (7) determined that parasitized Sunn Pest eggs stored at 6°C had a 90% emergence rate up to 3 weeks and that no emergence occured on the 8th weeks. Uzun (8) determined that emergence from *E. kuehniella*  eggs parasitized with *T. embryophagum* and stored at 4-9°C for 67 days decreased. At day 67 there was no emergence. The sex ratio obtained from 10-12 day old parasitized *D. baccarum* eggs changed favoring males after storage (Table 3). There were female parasitoids decreasing on stored 10 days. There was a little increasing on stored 10-20 days. As a result, we can say there might be an increase hatching of female numbers on the eggs stored 15-20 days in comparison with 510 days storage. In addition the male parasitoid number increased when stored 5-10 days. Longevity was shortest in parasitoids obtained from 20 days stored eggs when compared with the controls. When fresh eggs of *D. baccarum* were presented to adults they became parasitized and adult parasitoids emerged. The longevity of parasitoids is an important consideration for biological control strategies.

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		Mai	_	Main ± SEM (Min-Max)				
Storage	Emergence period of Storage parasitoids (day)		Emergence rate of	Parasitoi	d rate (%)	Sex rate	Longevi	ty (day)
period			parasitoids			Female-		
(day)	Female	Male	(%)	Female	Male	Male	Female	Male
5	5.3±0.2 c	3.71±0.1 b	97.8±1.4 a	34.46±9.51	65.54±9.51	1:3.27	25.33±1.05 b	11.14±1.47c
	(4-8)	(3-5)	(92.9-100)	(0-66.67)	(33.33-100)		(17-27)	(6-15)
10	6.6±0.3	4.7±0.2 b	73.6±11.8 b	19.27±7	80.73±7	1:4.69	25.33±1.05 b	15.89±1.69
	ab (5-9)	(1-6)	(45.5-100)	(0-40)	(60-100)		(25-27)	bс (7-21)
15	6.0±0.2 b	3.1±0.3 c	69.4±7.3 bc	31.2±13.8	(68.8±13.8	1:1.68	21.38±1.29 bc	18.29±33.04c
	(4-9)	(1-8)	(40-88.9)	(0-81.2)	(18.7-100)		(14-25)	(5-15)
20	7.0±0.5 a	2.9±0.2 c	45.1±9.1 c	29.1±11.9	70.9±11.9	1:1.38	18.73±2.95 c	18.29±3.04 b
	(5-11)	(1-4)	(14.3-72.7)	(0-81.2)	(21.4-100)		(0-78.6)	(6-26)
0	5.1±0.2	3.1±0.2 c	100±0.0 a	46.26±6.39	52.22±7.69	1:1.10	43.20±2.05 a	27.73±2.89 a
	bc	(1-5)	(100-100)	(26.32-	(18.18-		(32-51)	(14-41)
	(4-9)		. ,	72.73)	73.68)			

P<0.05 values in lines followed by the same letters are significantly different and statistically are important.

#### الملخص

كودان، م. و م. أو. غوركان. 2007. الإنتاج الكمي والتخزين للطفيل (Trissolcus grandis (Thomson). الصفحات 295-301. (Hymenoptera: Scelionidae)

تم في هذه الدراسة، بحث إمكانية الإنتاج الكمي للطفيل Trissolcus grandis Thomson على البيض المخزن للعائل البديل حشرة. الدراسة، بحث إمكانية الإنتاج الكمي للطفيل Pentatomidae). خزن البيض الطازج لحشرة للعائل البديل حشرة. للعائل البديل حشرة عرارة – 1±1 °س لمدة 8 أشهر. تم التطفل على هذا البيض ووضع في حجرة ضبطت على حرارة 2±25 °س، رطوبة نسبية 65±5% و 16: 8 (ضوء: ظلام) من أجل انبثاق الطفيل. كانت نسبة التطفل وانبثاق بالغات الطفيل من الطفيل 2±25 °س، رطوبة نسبية 65±5% و 16: 8 (ضوء: ظلام) من أجل انبثاق الطفيل. كانت نسبة التطفل وانبثاق بالغات الطفيل من البيض الفيل أقل. لم تختلف فترات حياة الطفيليات التي تم الحصول عليها من البيض المخزن عند مقارنتها مع الشواهد. بيض

D. baccarum المتطفل عليه بعمر 6-8 و 10-12 يوم تم تخزينه عند درجة حرارة 5±1 °س في البراد لمدة 5-60 يوم وتم تحديد نسبة انبثاق الطفيل. حصل انبثاق الطفيل حتى اليوم العشرين من التخزين ولكن لم يحصل انبثاق على اليوم الخامس والعشرين. كانت فترة الانبثاق متشابهة بين البيض المخزن والشواهد. كان طول فترة حياة الطفيليات التي تم الحصول عليها من البيض المخزن أقصر بالمقارنة مع الشواهد. كانت كل المؤشرات المدروسة متشابهة في البيض المخلف عليه والمخزن بعمر 6-8 يوم تم تحريم من التحريم وتم تحريم وتف الطفيليات التي تم الحصول عليها من والعشرين. كانت فترة الانبثاق متشابهة بين البيض المخزن والشواهد. كان طول فترة حياة الطفيليات التي تم الحصول عليها من البيض المخزن أقصر بالمقارنة مع الشواهد. كانت كل المؤشرات المدروسة متشابهة في البيض المتطفل عليه والمخزن بعمر 6-8 يوم.

كلمات مفتاحية: .Dolycoris spp، تربية كمية، تخزين، .Trissolcus spp.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

#### Mass Rearing of the Sunn Pest Egg Parasitoid, Trissolcus grandis Thomson (Hymenoptera: Scelionidae): a Demographic Framework

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#### Abstract

Amir-Maafi, M. 2007. Mass Rearing of the Sunn Pest Egg Parasitoid, *Trissolcus grandis* Thomson (Hymenoptera: Scelionidae): a Demographic Framework. Pages 303-307.

Mass production of most parasitoid species requires host rearing. Therefore, one of the objectives for rearing is to optimally balance the number of hosts available for parasitization with the number of adult parasitoid females held. In this study, periodic mass rearing and harvesting of the host, eggs of *Graphosoma lineatum* L., and the parasitoid, *Trissolcus grandis* Thomson, are analysed based on the demographic method. Life history data of the host and parasitoid were used to estimate the proportion of individuals that can be harvested from a rearing colony to keep the population size constant. The harvest rate for host (hh) and parasitoid (hf) are 0.98 and 0.99, respectively. The per female production rate of the host target stage (Ph) is 7.4 (egg/female), and the per capita adult female parasitoid rates for production of harvestable daughters (Pf) and sons (Pm) are 13.2 and 3.96 (female/day), respectively. The use of these findings in the harvesting schedule of the parasitoid is discussed.

Keywords: Eurygaster integriceps, Graphosoma lineatum, mass rearing, Scelionidae, Sunn Pest, Trissolcus grandis.

#### Introduction

Sunn Pest, *Eurygaster integriceps* Puton (Heteroptera: Scutelleridae), is one of the most important cereal pests in Iran. An integrated management program is currently being developed for Sunn Pest on utilization of a complex of hymenopteran egg parasitoids. The dominant species in this complex is *Trissolcus grandis* Thomson (Hymenoptera: Scelionidae) which comprised about 70% of the parasitoids collected from Sunn Pest eggs from 1995-2002 in Iran (1).

Research on Scelionid egg parasitoids of Sunn Pest provides a general understanding of the factors that influences the productivity of mass production units including physiological factors such as temperature, host exposure time and frequency, and host age (2, 7, 13). Complex behavioral factors such as the effect of parasitoid and host semiochemicals on host discrimination, longevity and sex ratio have been reported (4, 9, 8, 11). While this knowledge provides a basis for maximizing the efficiency of production, it remains expensive because of the cost of producing host eggs. This can be reduced by using alternative hosts. *T. grandis* and *T. semistriatus* Nees will readily develop on alternative hosts and there is considerable literature on rearing them on the pentatomid species *Graphosoma semipunctata* F., *G. lineatum* and *Eurydema* spp. (3, 7, 13, 14, 15).

Efficient and economic production is a major factor determining the success of a biological control program. Many insects and mites have been reared successfully in small numbers in the laboratory, but mass producing them requires a specific approach (10, 12). When a small laboratory culture has been enlarged to a mass-rearing system, efficiency may be enhanced using life history data. Carey and Vargas (6) used life table techniques to determine the fraction of the population

replaced and to calculate per-female production rate of the stage that is harvested.

In this paper we optimize the production of *T. grandis* using life tables and determine the optimal age to discard individuals and the harvest rates using the method of Carey and Vargas (6).

#### **Materials and Methods**

*Trissolcus grandis* and *G. lineatum* were obtained from a laboratory culture, maintained at the Sunn Pest Research Department. *Trissolcus grandis* came from Karaj, Fashand and *G. lineatum* from Karaj, Khor. The following data were collected: 1) life-cycle survivorship, and 2) fecundity and fertility. Standard life table parameters were calculated from daily records of mortality, fecundity and fertility.

#### **Harvest Rates**

During the time course of Sunn Pest egg parasitoid, T. grandis and its laboratory host, G. lineatum the following events are relevant to mass rearing: birth at age 0, egg hatch at age , harvesting at age , adult eclosion at age , first reproduction at age , and discarding from the colony at age (following demographic notations of (5)). It is assumed that the colony is kept at a fixed size. The objective of the rearing system is then to harvest a fraction h, leaving a fraction 1-h for colony maintenance (6). Survival and fecundity schedules are required to calculate *h*: *lx* denoting the number of days lived by the average individual in the age class from x to x+1 and mx denoting the average number of female offspring produced by a female during the interval x to x+1.

Thus the fraction of the host target stage that must be harvested to confer zero population growth in the rearing system, denoted hh (host harvest), is:

$$h_h = 1 - (R_0)^{-1}$$

where the harvest age of the target stage is less than . We assume that the sex-specific vital rates of hosts are identical. Therefore, we don't model each host sex separately.

We treat parasitoid males and females separately in the model since their primary sex ratios vary with maternal age. Let and denote survival to age x of parasitoid females and males, respectively, and let and denote a female parasitoid's production of female and male offspring at age x, respectively. Therefore, the per female production of daughters, denoted and sons, denoted, from their age of first reproduction to their age () is given as:

$$R_0^f = \sum_{x=r}^{u} l_x^f m_x^f$$
$$R_0^m = \sum_{x=r}^{u} l_x^m m_x^m$$

Hence the harvest rate of female parasitoids required to confer zero population growth in their population must be the solution to the equation,

$$h_f = 1 - \left(\sum_{x=x}^{u} R_0^f\right)^{-1}$$

and the primary sex ratio (male:female), denoted *S*, is

$$S = R_0^m / R_0^f$$

#### **Per Female Production Rates**

The per female production rate of the host target stage, denoted Ph, is

$$P_{h} = 2(R_{0} - 1)l_{.} / \sum_{x=v}^{u} l_{x}$$

where denotes the age at which the target stage is harvested and represents eclosion age. The factor '2' accounts for the production of males, under the assumption of 1:1 sex ratio. Following the same reasoning, the per capita adult female parasitoid rates for production of harvestable daughters and sons, denoted *Pf* and *Pm*, respectively, are:

$$P_{f} = [R_{0}^{f} - 1]l_{.}^{f} / \sum_{x=v}^{u} l_{x}^{f}$$
$$Pm = PfS$$

#### Results

#### Harvest Rates and Discard Ages

The harvest rates necessary to confer zero population growth for both *G. lineatum* and *T. grandis* for different age classes are given in Table 1. Perhaps the most revealing aspect of these relations is the difference between the changes in the net reproduction rates with age between the species. From age class 20 to 70 net reproduction for *T. grandis* increases <2-fold from 126.002 to 201.19 while net reproduction for *G. lineatum* from age class 40 to 150 by almost 10-fold, from 3.75 to 39.68 the specific harvest rates do not reflect the optimal discard ages since the objective in mass rearing is not to maximize the fraction

harvested but rather to maximize the per capita production rates for adult female. Harvest rates, *hh* and *hf* simply give the numerical requirements for exact replacement.

#### **Per Capita Production**

Production rates of *G. lineatum* egg per adult *G. lineatum* female ranged from  $\sim 12$  at early *G. lineatum* discard ages to 4 at older, *G. lineatum* discard ages.

The trade-offs between discard age and per capita female parasitoid production rates of harvestable male and female offspring are given in Figure 1. Two aspects merit comment. First, the maximum total production rates between ages 10 to 25 d are 33.4 to 9.53 (both sexes) per female/d. This is higher than the per capita rate for the *G. lineatum* target stage, which was about 7 (both host sexes) per female/d. These differences are due entirely to the differences in net reproduction between the species. Second, the optimal adult *T. grandis* per capita production is sensitive to discard age as reflected in the sharp peak at age 10 days.

**Table 1.** Net reproduction (female offspring) by age for *G. lineatum* and *T. grandis* and the associated harvest rates.

Age	G. linea	ıtum	T. grandis		
class	Net reproduction	Harvest rate	Net reproduction	Harvest rate	
20			126.002	0.992	
30	0.00	0.00	193.450	0.994	
40	3.75	0.73	201.190	0.995	
50	14.32	0.93	201.190	0.995	
60	23.25	0.96	201.190	0.995	
70	29.79	0.97	201.190	0.995	
80	34.23	0.97			
90	37.29	0.97			
100	38.27	0.97			
110	38.59	0.97			
120	39.11	0.97			
130	39.46	0.98			
140	39.62	0.98			
150	39.68	0.98			



Figure 1. Per capita adult female *T. grandis* production rates of female and male offspring reared on *G. lineatum.* 

#### الملخص

أمير-مافي، مسعود. 2007. التربية الكمية لطفيل بيض حشرة السونة . Trissolcus grandis Thomson. أمير-مافي، مسعود. (Hymenoptera: Scelionidae): إطار عمل ديموغرافي. الصفحات 303-307.

يتطلب الإنتاج الكمي لمعظم أنواع الطفيليات تربية العائل. لذلك، فإن أحد أهداف التربية هو الموازنة المتلى بين عدد العوائل المتاحة للتطفل مع عدد الإناث البالغة للطفيل المحنفظ بها. تم في هذه الدراسة تحليل التربية الكمية الدورية وحصاد العائل، بيض Graphosoma lineatum، والطفيل Strissolcus grandis، اعتماداً على الطرائق الديموغرافية. استخدمت بيانات تاريخ الحياة للعائل والطفيل لتقدير عدد الأفراد التي يمكن الحصول عليها من مستعمرة مرباة للمحافظة على حجم ثابت للمجتمع. نسبة حصاد العائل (hh) والطفيل (hh) هي 9.08 و 0.99، على التوالي. معدل الإنتاج للأنثى لطور العائل المستهدف (hh) منببة حصاد العائل (hh) والطفيل (hh) هي 13.2 و 0.99، على التوالي. معدل الإنتاج للأنثى لطور العائل المستهدف (h) هو 7.4 (بيضة/أنثى)، والمعدلات القصوى لأنثى الطفيل البالغة لإنتاج إناث يمكن حصادها (fh) وذكور (m) هي 13.2 و 3.96 (أنثى/يوم)، على التوالي. سوف يتم مناقشة استخدام هذه النتائج في جدول حصاد الطفيل. كلمات مفتاحية: Scelionidae، تربية كمية، تربية مية منية، تربية علية من متواتية، معنه، الطفيل، علي والونة، السونة، كلمات مفتاحية، التوالي. سوف يتم مناقشة استخدام هذه النتائج في جدول حصاد الطفيل.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

#### Survey of Egg Parasitoids of Sunn Pest in Northern Syria

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#### Abstract

### Trissi, A.N., M. El Bouhssini, J. Ibrahem, M. Abdulhai and W. Reid. 2007. Survey of Egg Parasitoids of Sunn Pest in Northern Syria. Pages 309-314.

Surveys of Sunn Pest eggs and egg parasitoids were made in three northern provinces of Syria, Idleb, Aleppo and Al Hassakeh in the 2003 field season. Egg laying occurred from 7 April until 30 May, with parasitized eggs present within the first week of egg observance and continuing throughout the season. Parasitoids were recovered from all provinces and sites, with the exception of one site that had been aerially treated with Decis for Sunn Pest management. Species abundance was correlated (r=0.496, p=0.0001) with total percent parasitism. In total, five species were recovered, *Trissolcus grandis* Thomson, *T. semistriatus* Nees, *T. vassilievi* Mayr, *T. festivae* Viktorov and *Ooencyrtus fecundus* (Ferrier & Voegele). This is the first record of *T. semistriatus* and *T. festivae* in Syria. The most abundant species were *T. semistriatus* which occurred in all provinces and 26.98% of all sites overall. *O. fecundus* was recovered only from Idleb, *T. festivae* from Idleb and Aleppo provinces, and *T. grandis* from Aleppo and Al Hassakeh provinces. All species of *Trissolcus* were recovered throughout the season with the exception of *T. grandis* which was first observed 4 May, three weeks post first egg observation. Overall parasitism rates reached 58% at the end of the season. Rates within the first month of egg observance ranged from 10 to 25% where *T. vasselievi* was present and 6 to 20% in areas with *T. semistriatus*.

Keywords: Biological control, egg parasitoids, Eurygaster integriceps, Sunn Pest.

#### Introduction

The Sunn Pest, Eurygaster integriceps Puton (Hemiptera: Scutelleridae), is a serious pest of wheat and barley, causing widespread crop losses in the cereal-growing regions of Central and West Asia, Eastern Europe and Russia (8, 19, 22). Damage is caused by both the immature and adult stages (5), severely reducing yields and grain quality (7, 16, 17, 18). Surveys of natural enemies have been conducted throughout the affected region, and include predators, insect-killing fungi and adult and egg parasitoids (11, 13, 14, 17, 18). Parasitism rates in Eastern Europe averaged 57.8%, and have been recorded as high as 100% (18), which is likely an overestimation biased by late season sampling when healthy eggs have already hatched (2). All egg parasitoid species recorded, to date, are within the Hymenopteran families Scelionidae and Encyrtidae. Their distributions are summarized in Table 1.

The role of these egg parasitoids as an integrated pest management (IPM) tactic for control of E. integriceps populations is viewed from augmentative, inoculative and conservational perspectives. Studies on parasitoid augmentation have been conducted using Trissolcus spp. with mixed results. Field control levels using release rates of 15,000 adult females per hectare in Iran and 50,000 per hectare in Russia gave 80-95% and 27.9% parasitism rates, respectively (10, 15). An alternative approach to the role of these parasitoids in the system as a single species augmentation is the approach of their use as a complete guild; however, problems with multiple species releases can arise. Interspecific competition between the egg Telenomus parasitoids T. grandis and chloropus Thomson has been previously
recorded in Russia (12), while additive effects of parasitoids by multiple egg parasitoid species has been recorded in Iran (3), and additive effects of a guild of egg parasitoids of the related bug Nezara viridula (L.) has also been recorded (4). To date, all recorded egg parasitoids are generalist species, parasitizing eggs of various insects within the Hemipteran families Scutelleridae and Pentatomidae. Inoculative release of absent species coupled with preservation of their natural habitats may serve to be an important tactic in the IPM of E. integriceps. This study investigates which species are present in Northern Syria, and approximates their abundance, and estimates the impact of these species as a guild and its effect on the percentage of parasitized eggs.

## Materials and Methods

## Surveys of Eggs

Egg masses of *E. integriceps* were sampled from one-hectare fields in nine sites in Northern Syria. Each field was divided into five even blocks, and within each block, five  $0.25 \text{ m}^2$  frames were randomly thrown, and all *E. integriceps* egg masses within the frame were collected and recorded.

#### **Emergence and Identification**

Egg masses were returned to the laboratory and transferred singly into 10-ml glass tubes held at  $25\pm1^{\circ}$ C and  $60\pm5\%$  RH under a 16:8 L:D photoperiod. Parasitoid adults were killed and preserved in 70% ethanol containing 5% glycerol. Preliminary identifications were made at ICARDA and confirmed by Dr. Miktat Donglar (Department of Entomology, Mustafa Kamal University, Antakia, Turkey).

#### Statistical analyses

The levels of parasitism were averaged by site throughout the season to reflect the total impact of all parasitoids on *E. integriceps*. Percent parasitism was regressed against total number of species for all sites using a general linear model in SAS with the percent parasitism weighted for total egg abundance (21).

 
 Table 1. Egg parasitoids observed on Eurygaster integriceps
 Puton and recorded rates of field parasitism.

	Field notes	
Country	rielu l'ales	
Species recovered	norociticm	Dof
Inon	parastusii	Kel.
Iran Tuisgoloug onge dig	00.00/	(20)
Trissolcus granais	90.0%	(20)
Trissolcus semistriatus	22.9%	(3)
Trissoicus vassilievi		
Trissolcus basalis		
Trissoicus rujiventris		
	07 10/	(02)
Trissolcus granais	87.1%	(23)
Trissolcus flavipes		
Trissolcus rufiventris		
Trissolcus scutellaris		
Trissolcus simony		
Russia	264 40 50	(0)
Trissolcus grandis,	36.4-48.5%	(9)
Trissolcus simony		
Telenomus chloropus		
Syria		
Trissolcus grandis	36.0-50.0%	(1)
Trissolcus vassilievi		
Trissolcus simony		
Ooencyrtus fecundus		
Ooencyrtus telenomicida		
Turkey		
Trissolcus grandis	46.4%	(6)
Trissolcus choaspes		
Trissolcus rufiventris		
Trissolcus basalis		
Trissolcus semistriatus		
Trissolcus reticulates		

# **Results and Discussion**

Sunn Pest egg masses were attacked by five species. Trissolcus semistriatus Nees. Trissolcus vassilievi Mayer, Trissolcus Thomson, grandis Trissolcus festivae Viktorov and Ooencyrtus fecundus Ferriere & Voegele. This is the first record of T. semistriatus and T. festivae in Syria. All parasitoids were recovered between late April and late May.

The species and percent parasitism of Sunn Pest eggs collected from wheat fields are shown in Table 2.

		Field rates			
		of			
Province/Region	%	parasitism			
Species recovered	parasitism	(%)			
Aleppo/Azaz					
Trissolcus semistriatus	9.25	28.57			
Trissolcus vassilievi	14.29				
Trissolcus grandis	4.76				
Aleppo/Yahmool					
T. semistriatus	10.71	25.00			
T. vassilievi	8.36				
Trissolcus festivae	8.93				
Aleppo/Nadda					
T. grandis	5.00	5.00			
Idleb/Benesh					
T. semistriatus	12.5	22.92			
T. festivae	4.17				
Ooencyrtus fecundus	6.25				
Idleb/Sraqeb					
T. semistriatus	17.78	17.78			
Idleb/Sarmeen					
T. semistriatus	14.58	14.58			
Al-Hassakeh/Al-Malk	ia				
T. semistriatus	17.64	58.82			
T. vassilievi	29.41				
T. grandis	11.76				
Al-Hassakeh/Al-Qameshleh					
T. vassilievi	21.43	21.43			
Al-Hassakeh/Tall-Shaeer					
No. egg parasitoids	0.00	0.00			

**Table 2.** Egg parasitoids observed on *Eurygasterintegriceps*Puton and recorded rates of fieldparasitism in Syria.

Egg parasitoids were recovered from all sites in Aleppo with initial recovery oneweek- post Sunn Pest oviposition with the exception of Nadda where no parasitoids were observed until four-weeks-post Sunn Pest oviposition on 22 of May. Total percent parasitism reached 28.57%, 25% and 5% in Azaz, Yahmool and Nadda, respectively. In Idleb, egg parasitoids were recovered from all sites, and the total percent parasitism reached 22.92%, 17.78% and 14.58% in Benesh, Sraqeb and Sarmeen, respectively. Female egg parasitoids were present in wheat fields oneweek-post Sunn Pest oviposition in Benesh site and one week later in the other sites.

In Al-Hassakeh, the abundance of parasitized egg masses peaked by the second

week of May. Total percent parasitism reached 59% and 22% in Al-Malkia and Al-Qameshleh, respectively. No parasitoids were recovered from Tall Sheer where chemical control against Sunn Pest was conducted on 7<sup>th</sup> and 14<sup>th</sup> May, 2003.

Throughout the second week of Sunn Pest oviposition, percent parasitism reached 23.53, 11.76 and 10.53% in Aleppo, Al-Hassakeh and Idleb, respectively. These results shadowed the maximal abundance of healthy egg masses by one week, and are comparable to results from Iran 1997, 1998 (3) reflecting the results that the naturallyoccurring levels of egg parasitism is low during the initial stages of Sunn Pest oviposition.

*T. semistriatus* was ubiquitous species to all provinces and 50.79% of all sites, the first observation of this species was throughout the second week of April in Idleb fields and was present for more than one month and had the highest percent parasitism, 14.89%, by the end of the season. In Al- Hassakeh, *T. vassilievi* was dominant, present in 26.98% of all sites. It was the first species recovered and persisted in the field until the end of May. *T. grandis* was recovered from Aleppo and Al-Hassakeh provinces through the first week of May, with total percent parasitism 6.35% of all sites. The occurrence of egg parasitoids species are shown in Figure 1.

Multi species effect was detected by weighted regression of parasitism rate for all samplings combined against total number of species, slope was positive (0.1956), and significantly deferent from zero (p=0.0001), These results indicate a positive association between the number of species and percent parasitism, although the correlation was not strong (r=0.49), but the intercept was very close to zero (0.01932). The multi species benefit was evident as percent parasitism increased when more species were obtained. Further study that spam additional levels of species diversity are needed in order to estimate this effect.



Figure 1. Occurrence of egg parasitoids in Northern Syria.

These results suggested that any augmented use of egg parasitoids for management of Sunn Pest in Northern Syria should consider the use of locally abundant species, as there was a correlation between the number of species and the percent parasitism. This demonstration of the complimentary effects of a guild of egg parasitoids in the total percent parasitism showed that under natural conditions this effect occurs. Further research is needed to determine if this case holds true under single or multiple-species augmented releases.

# Acknowledgment

The authors appreciate Dr. Miktat Donglar (Department of Entomology, Mustafa Kamal University, Antakia, Turkey) for identifying the Sunn Pest egg parasitoids.

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تريسي، عبد الناصر، مصطفى البوحسيني، جمعة إبراهيم، محمد عبد الحي ووليم ريد. 2007. حصر طفيليات بيض حشرة السونية في شمال سورية. الصفحات 309–314.

كلمات مفتاحية: مكافحة حيوية، طفيليات بيض، Eurygaster integriceps، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Survey of Sunn Pest Adult Parasitoids in Syria

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### Abstract

# Abdulhai, M., R. Canhilal, M. El Bouhssini, W. Reid and F. Rihawi. 2007. Survey of Sunn Pest Adult Parasitoids in Syria. Pages 315-318.

A survey of adult parasitoids of Sunn Pest, *Eurygaster integriceps* Puton, was conducted in Syria in 2003 and 2004. Four species of Phasiine flies were found: *Phasia subcoleoptrata* (L.), *Heliozeta helluo* (F.), *Ectophasia oblonga* (Robineau-Desvoidy) and *Elomyia lateralis* (Meigen). In 2003, parasitism rates in all of the sampled overwintering sites were negligible. In-wheat-field rates in the early spring were 1.3% in Hamaa, 2.7% in Swedaa and not detected in the other areas. Towards the end of the spring, the percent in-wheat-field parasitism rates increased dramatically and were 6.47, 4.0, 5.85 and 13.25% in Azaz, Idlib, Al Kamishli and Al Swidaa, respectively. In 2004, the levels of parasitism in the overwintering sites were again negligible, except in Azaz (1% parasitism). In-wheat-field rates in the early spring reached 1.9% in Al Malkia and were not detected in the other areas. Late-spring parasitism rates reached 5.88% in Al Kamishli and 12.45% in Al Malkia.

Keywords: Adult parasitoids, biological control, Sunn Pest, Tachinidae.

## Introduction

Sunn Pest (Eurygaster integriceps Puton) is a very destructive insect pest of wheat and barley in Syria and in central and west Asia, Caucasus, Bulgaria and Romania. Both nymphs and adults cause damage to plants and reduce yield and quality by feeding on leaves, stems, and grains. Apart from the direct reduction in yield, the insects also inject chemicals that greatly reduce the baking quality of the dough. If as little as 2-5% of the grain is affected, the whole grain lot might be rendered unacceptable for baking (2, 4). Sunn Pest infestations, which can lead to 100% crop loss in the absence of control measures, affect 15 million ha. annually. About US\$ 40 million is spent each year on pesticides in the Sunn Pest-prone areas (5). In 2004 more than 277,000 ha. were sprayed against the pest in Syria (1). The present insecticide-based strategy must be replaced with an integrated pest management approach comprising of biological control, host plant resistance, and cultural practices. Dipteran adult parasitoids from the family Tachinidae are important natural enemies of Sunn Pest, and seem to contribute to a reduction of populations.

Previous studies in Syria have mentioned that one parasitoid species belonging to family Tachinidae in the order Diptera attacks Sunn Pest adults. That species was *Phasia crassipennis* F. (3). We hypothesized that additional adult parasitoids were active in Syria because we had observed significant infestation of Sunn Pest adults by parasitic flies in previous seasons. Therefore, the objectives of this study were to 1) identify additional adult parasitoid species of Sunn Pest, and 2) estimate their level of parasitism in Sunn Pest prone areas.

# **Materials and Methods**

#### **Adult Parasitoid Species in Syria**

Field collections of parasitized adults of Sunn Pest were made during 2003-2004 seasons twice in winter from the overwintering sites in Azaz, Ariha, Ksabia and Tel Hadya, and twice in spring in wheat fields in Al-Hassaka, Al-

Ghab, Azaz and Aleppo. In each overwintring site, two sets of samples from each site at two week intervals before migration starts were taken. In the field, sampling was done twice; 1) when the migration to fields started (around the end of March- beginning of April) and 2) at the middle of the season (about one month after the first sampling). Field sites were Hama, Idlib, Al Swedaa, Azaz, Kamishli and Dier Ezzour. Two hundred Sunn Pests were collected for each sample. After bringing them to the lab, they were separated into males and females, which were placed separately in different small plastic containers and closed with cheese cloth. The containers were placed in a rearing room at 23±2°C, 70% RH. and 16:8 LD. A layer of paper was put at the bottom of the container with some wheat leaves to feed Sunn Pest. Adults were checked 2x/weeks. and numbers of dead, alive and parasitized individuals recorded. Parasitoid pupae were held in Petri dishes for adult emergence. The adults were pined and sent to Prof. Mektat Dognlar (specialist in insect Identification) at Plant Protection Department, Faculty of Agriculture, Mustafa Kemal University, Turkey, for identification.

# **Results and Discussion**

Four parasitoid species belonging to the family Tachinidae were found attacking Sunn Pest adults. They were: *Phasia subcoleopturata* (L.), *Heliozeta helluo* (F.), *Elomya lateralis* (Meigen) and *Ectophasia oblonga* (Robineau-Desvoidy). These adult parasitoids are reported as Sunn Pest adult parasitoids for the first time in Syria (Figure 1).



Figure 1. Distribution of Sunn Pest adult parasitoids in Syria.

*Phasia subcoleopturata* was the dominant species in Azaz, Idlib, and Al-Kamishli, while *Heliozeta helluo* was the dominant species in Aleppo (Tel Hadya), Al-Malkia and Al-Swedaa. The four species are among seven species reported on Sunn Pest in Turkey, the nearest country to north Syria (6). The first three species were reported on Sunn Pest in Iran, in addition to *Ectophasia crassipennis* (Fabricius) (7).

In 2003, the % parasitism of overwintering Sunn Pest adults was <0.5% in all sites; and 1.3%, 2.7% in Hama and Swedaa respectively, and less than 0.5% in other sites in early spring; and 6.74%, 4%, 5.85% and 13.25% in Azaz, Idlib, Al Kamishli and Al Swedaa respectively in the late spring of 2003 (Figure 2).

In 2004 the % parasitism of overwintering Sunn Pest adults was 1% in Azaz, while it was <0.5% in the other sites; and 1.9% in Al Malkia, and <0.5% in other sites in early spring; and 5.88%, 12.45% in Al Kamishli, Al Malkia in the late spring of 2004 (Figure 3).

This study showed that adult parasitoids become active in the spring about 2 weeks after the Sunn Pest migrates to cereal fields from the overwintering sites and starts laying eggs. The level of parasitism varies from year to year. Parasitoids could contribute to reducing Sunn Pest populations provided they are not disturbed by the use of broad-spectrum insecticides for control, usually by aerial spraying covering large and continuous areas.



Figure 2. Parasitism levels of Tachinid flies on Sunn Pest in Syria, 2003.



Figure 3. Parasitism levels of Tachinid flies on Sunn Pest in Syria, 2004.

# الملخص

# عبد الحي، محمد، رمضان جانهلال، مصطفى البوحسيني، وليم ريد وفوزي ريحاوي. 2007. حصر طفيليات الحشرة البالغة للسونة في سورية. الصفحات 315–318.

أجري حصر لطفيليات الحشرة البالغة للسونة (Eurygaster integriceps Puton) في سورية عامي 2003 و 2004. وجدت أربعة أنواع من ذباب الفازيا هي: (L.) Phasia subcoleoptrata (L.) Robineau- Isvoidy) و (Robineau-Disvoidy) و (Robineau-Disvoidy) في عام 2003، كانت نسب التطفل جديرة بالإهمال في جميع مواقع البيات الشتوي التي أخذت منها العينات. كانت نسب التطفل في حقول القمح في الربيع المبكر 1.3% في حماه، و 2.7% في السويداء، بينما لم يلاحظ التطفل في المناطق الأخرى. ازدادت نسب التطفل في حقول القمح في الربيع المبكر 1.3% في عام 2003، كانت نسب التطفل في حماه، و 2.7% في السويداء، بينما لم يلاحظ التطفل في المناطق الأخرى. ازدادت نسب التطفل في حقول القمح في الربيع المبكر 1.3% في حماه، و 2.7% الربيع وكانت 6.47% منها العينات. كانت نسب التطفل في حقول القمح في الربيع المبكر 1.3% في حماه، و 2.7% الربيع وكانت 6.47%، منها العينات المناطق الأخرى. ازدادت نسب التطفل في حقول القمح بشكل ملحوظ باتجاه نهاية مستويات التطفل في مواقع البيات الشتوي جديرة بالإهمال أيضاً باستثناء إعزاز (التطفل 1.1%). وصلت نسب التطفل في حقول المتوجر إلى 2.58% في المالكية بينما لم يسجل التطفل في المناطق الأخرى. وصلت نسب التطفل في حقول المتأخر إلى 5.88 في القامشلي و 2.1% في المالكية بينما لم يسجل التطفل في المناطق الأخرى. وصلت نسب التطفل في الربيع

كلمات مفتاحية: طفيليات الحشرة البالغة، مكافحة حيوية، حشرة السونة، Tachinidae.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Entomopathogenic Fungi for Sunn Pest Management: Efficacy Trials in Overwintering Sites

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### Abstract

Skinner, M., B.L. Parker, S. Gouli, W. Reid, M. El Bouhssini, M. Amir-Maafi and Z. Sayyadi. 2007. Entomopathogenic Fungi for Sunn Pest Management: Efficacy Trials in Overwintering Sites. Pages 319-328.

The cadavers of Sunn Pest, *Eurygaster integriceps* Puton, infected with entomopathogenic fungi have been observed in overwintering sites, suggesting that fungi are a natural mortality agent in this environment. Over 220 strains have been isolated from Sunn Pest overwintering sites in West and Central Asia and Russia. Based on results from laboratory bioassays and studies on germination and sporulation characteristics, several promising isolates were selected for testing against Sunn Pest in overwintering sites in northern Syria and north-central Iran. From 2002-2003, fungal treatments were made using different formulations and application times. Results from these tests are reported and recommendations for future directions in the development of entomopathogenic fungi for Sunn Pest management are proposed.

Keywords. overwintering sites, Eurygaster integriceps, entomopathogenic fungi, persistence.

## Introduction

Sunn Pest, Eurygaster integriceps Puton, is found in parts of Eastern Europe and West and Central Asia, causing significant damage to winter wheat and barley (2). Biological control research has been underway for many years, but insecticides remain the most common means of management, which is commonly applied by air over large areas throughout the infested regions (6). Recognizing the importance of this insect and concerns about extensive pesticide use, the Food Agriculture Organization convened a meeting of experts to review the Sunn Pest problem, recommending development of an integrated pest management (IPM) approach with a particular focus on biological control (3). Evaluation of the potential for maximizing on the naturally-occurring diseases in overwintering sites was one priority identified. Reports of Sunn Pest infected

with the entomopathogenic fungus, *Beauveria bassiana* (Balsamo) Vuillemin, in overwintering sites had been made previously, but until recently, isolates from this insect had not been recovered (7). Because little information was available on fungal pathogens attacking Sunn Pest and none were available for use, a search for pathogens in their overwintering sites was conducted from 1999-2002. More than 200 strains have now been isolated and placed in permanent collections (8).

The biology of Sunn Pest offers several different opportunities for management using fungi. One is to target them in their overwintering sites. In June and July, adults leave wheat and barley fields and fly to the surrounding foothills (1). There they seek shelter from the heat of summer and cold of winter and protection from predators under litter around trees and shrubs. Tree plantations established for reforestation are commonly maintained in many of these regions and provide relatively easy access to the resting sites. Populations as high as 1,000 Sunn Pest per tree have been found in northern Syria (BLP, pers. comm.). Sunn Pest remain dormant in these overwintering sites for about 8 mo until early spring when they migrate to fields of young, succulent winter wheat. The moist conditions occurring under the litter where the Sunn Pest are found are ideal for establishing a fungal epizootic. It may be possible to apply fungi around the trees and shrubs where they congregate, which, once established, would provide an on-going source of inoculum.

Alternatively, Sunn Pest could be targeted in the early spring when they return to wheat fields. Fungal applications could be applied to the perimeters of the wheat fields to reduce adult populations before egg laying begins. It is believed that Sunn Pest fly to the field edges first and then slowly colonize the rest of the field. A similar approach is being tested in IPM pilot sites in Iran, where insecticides are applied only around field borders early in the growing season when adults gather there to mate and populations reach 2-3 adults/m<sup>2</sup> (A. Abdollahi, Ministry of Agriculture, Tehran, pers. comm.). This strategy minimizes the cost of application and the impact on nontarget beneficial organisms.

Entomopathogenic fungi offer several advantages that favor their use as a biological control agent. Many species are easy to mass-produce on solid or liquid media, and standard techniques for their commercialization have been developed. Some species are facultative saprophytes and have the ability to maintain viable inoculum in the absence of an insect host. They occur in the environment naturally, and have a demonstrated ability to kill Sunn Pest in overwintering sites. Many isolates have a fairly limited host range and thus pose little or no risk to humans or the environment.

The primary objective of the research reported herein was to determine if selected

fungal strains could kill Sunn Pest adults in their overwintering sites. In addition, we sought to determine if fungi persisted within these overwintering sites after application. This information is critical for further development of entomopathogenic fungi for Sunn Pest IPM.

# Materials and Methods

## Spring 2002 Trial: Syria

Pine trees (Pinus brutia Tenore) at Tel with Hadya, Syria, overwintering populations of Sunn Pest in the litter were randomly selected. Beneath each tree a plot (0.5 x 0.5 m) was marked out with a wooden frame. Treatments, each replicated 5 times, were randomly assigned to individual trees. Two fungal isolates, B. bassiana (SPT22) and Metarhizium anisopliae (Metschnikoff) Sorokin (ERL500B) were selected for field testing based on results from previous laboratory bioassays (8). These isolates were cultured in 9-cm diam Petri dishes on quarter-strength Sabouraud dextrose agar with 0.25% w/v yeast extract (SDAY) supplemented with 0.001 g/l penicillin G and 0.005 g/l streptomycin sulfate. They were held at 20°C in the dark for 14 days then conidia were harvested in 0.02% Tween 80. Conidial viability was assessed using methods described by Goettel and Inglis (4). Only batches that were >90% viable after 24 h were used in the assays. Spore suspensions were prepared in sterile distilled water with 0.02% Tween 80.

The following seven treatments were tested: fungi, B. bassiana (SPT22) and M. (ERL500B) anisopliae prepared as in horticultural suspensions oil or water/0.02% Tween 80 applied at a rate of 1.77 x 10<sup>10</sup> conidia/plot; formulation blank controls (oil and Tween) and an untreated control. Treatments were made on 28 February 2002. After 7 days, all Sunn Pest adults were collected from the test plots. Dead Sunn Pest were surface sterilized, and placed in a Petri dish to allow for fungal outgrowth to determine if they were

infected. *B. bassiana* and *M. anisopliae* were readily discernable based on colony form and color. Live Sunn Pest were placed in a plastic screw-top jar (5.5 cm inside diam x 5.7 cm high), with fresh wheat grass (ICARDA variety Cham2). Mortality was assessed after 5 and 10 days, at which time dead insects were assessed for fungal infection as described above.

Fungal persistence in the plots was assessed over a 9-d period. Litter samples (~15 g) were taken with an 8-cm diam corer from each treatment plot on the day of application, and at 3, 6, and 9 days post application and placed in plastic jars. In each jar, 10 Sunn Pest collected from another overwintering site were added and the jars were held in the laboratory at 20°C under natural light conditions. Every other day the jars were inverted to encourage the Sunn Pest to crawl through the litter and come in contact with the treatment. On Day 5 adults were removed; and mortality counts were taken, placing live insects in a clean jar with fresh wheat grass. Mortality was assessed again at Day 10, at which time dead insects were removed and fresh wheat was added. A final mortality count was taken 5 days later. Dead Sunn Pest were assessed for fungal infection as described previously.

### Fall 2002 and Spring 2003 Trials: Syria

Trials were conducted at a Sunn Pest overwintering site at Tel Hadya, Syria to evaluate differences in efficacy of multiple isolates and several formulations at two treatment times. Using the same methods as for the Spring 2002 trial, several *B. bassiana* isolates and one *M. anisopliae* isolate were tested (Table 1); treatments were made on 30 Oct 2002 (fall) and 10 Feb 2003 (spring). The specific treatments tested changed from 2002 to 2003 based on results from earlier trials. Each treatment was replicated 5 times.

Sunn Pest samples were taken from the plots of the Fall and Spring trials on 11 Feb. 2003 and 12 Mar. 2003, respectively, to determine mortality, as described previously. For the Fall 2002 trial, fungal

persistence was assessed by taking separate samples of soil and litter within each treatment plot about 3 mo after treatment and exposing Sunn Pest as described previously. For the Spring 2003 trial, persistence was assessed immediately before treatment, and 10 and 20 days post treatment.

## Spring 2003 Trial: Iran

Several fungal isolates, including one *Lecanicillium lecanii* (Zimm.) Zare and Gams, were tested in an overwintering site near Esfahan, Iran (Table 1). Treatments were made on 2 March 2003 to litter beneath *Centaurea gaubae* (Bornm.) Wagenitz bushes (40-60 cm diam canopy), under which an average of 60 Sunn Pest were found. Insects were sampled to assess fungal efficacy ~1 month after treatment. Methods described above were followed for this trial.

## **Statistical Analysis**

Similar methods were used to analyze data from all of the field trials. For each experiment, analysis of variance models were run to test whether the proportion of sunn pest dead at day 10 following sampling differed by treatment type (alpha = 0.05) (10). Based on previous experimentation, mortality 10 days after collection is the best time period to assess treatment effect. It takes several days for the fungi to kill the insect, yet, after 10 days, natural mortality in increasedayssignificantly, the controls masking treatment effects. Differences at a level of P 0.05 were considered significant. For the ANOVA models, an arc sine transformation was applied to the proportion of dead bugs, and this transformed variable was used as the dependent variable in the models. The following formula was used for the transformation:

Arcsin proportion = (sqrt (T+1)) \* (2 \* arsin (sqrt((d10+1)/(T+1))))

where T = total number of Sunn Pest at start of experiment, d10 = number of Sunn Pest dead at day 10.

	Fungal	Treatment	Year and	
Code/Treatment	Species	Site	Treatment Time	Formulation
Untreated Control	None	Syria	All years and times	None
		Iran	Spring 2003	
Tween 80 blank (Twn)	None	Syria	All years and times	Water/Tween 80
		Iran	Spring 2003	suspension
Oil blank	None	Syria	Spring 2002	Horticultural oil
				suspension
NBG blank	None	Syria	Fall 2002, Spring 2003	Millet-based granular
		Iran	Spring 2003	
22/NBG	B. bassiana	Syria	Fall 2002, Spring 2003	Millet-based granular
		Iran	Spring 2003	
22/Tween 80	B. bassiana	Syria	All years and times	Water/Tween 80
22/1 (A1)		Iran	Spring 2003	suspension
22/alginate (Alg)	B. bassiana	Syria	Fall 2002, Spring 2003	Alginate granular
22/011	B. bassiana	Syria	Spring 2002	Horticultural oil
23/Tween 80	R hassiana	Svria	Spring 2002	Water/Tween 80
25/ I ween 60	D. Dassiana	Sylla	Spring 2002	suspension
566/Tween 80	B. bassiana	Svria	Fall 2002, Spring 2003	Water/Tween 80
		Iran	Spring 2003	suspension
200/Tween 80	R hassiana	Surio	Fall 2002 Spring 2003	Water/Tween 80
200/ I weell 80	D. Dassiana	Sylla	1 ali 2002, Spring 2003	suspension
493/Tween 80	B. bassiana	Svria	Fall 2002. Spring 2003	Water/Tween 80
		Iran	Spring 2003	suspension
$2/T_{\rm Wear}$ 80	P hassiana	Suria	Eall 2002 Spring 2003	Water/Tween 80
2/1 ween 80	D. Dassiana	June	Fair 2002, Spring 2003	suspension
<b>500 F</b>			Spring 2005	
502/Tween 80	B. bassiana	Syria	Fall 2002, Spring 2003	Water/Tween 80
		Iran	Spring 2003	suspension
603/Tween 80	B. bassiana	Syria	Fall 2002, Spring 2003	Water/Tween 80
		Iran	Spring 2003	suspension
GHA/Tween 80	B. bassiana	Syria	Fall 2002, Spring 2003	Water/Tween 80
				suspension
		Iran	Spring 2003	Commercial product
		T	Spring 2000	W
151/Tween 80	L. lecanıı	Iran	Spring 2003	Water/Tween 80
Green Musele	M anisoplias	Suria	Spring 2002	Suspension Water/Twaan 80
(GM)/Tween	m. anisopiae	Sylla	Spring 2003	suspension
				Commercial product
500B/Oil	M. anisopliae	Svria	Spring 2003	Horticultural oil
		~ ,	-r	suspension
500B/Tween 80	M. anisopliae	Syria	All years and times	Water/Tween 80
	-	Iran	Spring 2003	suspension

Table 1. Treatments for trials conducted in the Spring and Fall 2002 and Spring 2003 in Sunn Pest overwintering sites

# **Results and Discussion**

#### Spring 2002 Trial: Syria

Significant treatment effects were detected at Day 10 for all fungal treatments (Figure 1). Isolate 500B formulated with Tween 80 provided the highest Sunn Pest mortality (100%) mortality compared with 67.8% mortality in the formulation blank. Because of the relatively high rate of natural mortality in the controls and formulation blank treatments, all cadavers were plated in moist conditions to determine if there was a relationship between the high mortality and the fungi applied. Results from these evaluations showed that over 60% of the dead Sunn Pest from plots treated with isolate 22 showed evidence of internal infection from B. bassiana (Figure 2). Similarly, M. anisopliae was isolated from >70% of the Sunn Pest from plots treated with 500B. In contrast, <7% of the cadavers in the control or formulation blank plots produced fungal outgrowth of B. bassiana, and no M. anisopliae was recovered. This clearly demonstrates that using mortality as the only means of evaluating fungal treatment effect, underestimates the fungal effect when compared with the controls. Methods used to shold Sunn Pest after treatment should be improved to reduce the high natural mortality commonly observed.

When field-collected Sunn Pest were exposed to litter taken from the test plots 9 days post-application, a significant treatment effect was observed for isolates 23 and 500B, with ~70% mortality compared with <30% mortality for all other treatments and the control (Figure 3). No enhancement of efficacy was observed from using an oil formulation. When fungal outgrowth from the cadavers was assessed, for all but one fungal treatment (isolate 23) >90% of the cadavers developed signs of growth of the fungus that had been applied (Figure 4). Less than 20% of the cadavers from the untreated or formulation blanks exhibited fungal outgrowth of either B. bassiana or M.

(Figure 4). This again anisopliae demonstrates that the effect of a fungal be underestimated if treatment may determined only based on the number of Sunn Pest that died. Though assessment of fungal infection rates using outgrowth from cadavers takes considerable time and expertise, it definitely provides a more accurate estimate of the impact of a fungal treatment on Sunn Pest. These results provide clear evidence of fungal persistence in overwintering sites at 9 days postapplication.



Figure. 1. Percent mortality of Sunn Pest collected from treatment and control plots 7 days after treatment and held for 10 days (Spring 2002 trial in Syria). Bars with the same letters are not significantly different at P < 0.05. Treatments are: 1= Untrt., 2= Tween, 3= Oil, 4= 22/Twn, 5= 22/Oil, 6= 500B/Twn, 7= 500B/Oil.

## Fall 2002 and Spring 2003: Syria

Several isolates and formulations were tested in an overwintering site to determine if a fungal application made in the spring (before Sunn Pest migrated to wheat) resulted in higher mortality than one made in the fall (after adults returned to

overwintering sites and before the rainy season). For all treatments, Sunn Pest mortality was higher in the plots treated in the spring than in the fall, though differences were not consistent among all treatments (Figure 5). It is possible that this reflects lower vigor among Sunn Pest as a result of depleted energy reserves during the >8-mo non-feeding period in the overwintering sites. The relatively high mortality in the controls and treatment blanks supports this hypothesis. A fall application of a granular application might be particularly effective as it could provide a sustained inoculum throughout the hibernation period, enhancing the opportunity for infection. Based on the results from these trials, three isolates, 22, 566, and 493 were found promising for further study. Other isolates were eliminated either because they were difficult to mass produce (e.g., 2) or were not indigenous to the region (e.g., 500B).



**Figure. 2.** Percentage of Sunn Pest cadavers with evidence of internal fungal infection after surface sterilization (Spring 2002 trial in Syria). Treatments are: 1= Untrt., 2= Tween, 3= Oil, 4= 22/Twn, 5= 22/Oil, 6= 500B/Twn, 7= 500B/Oil.



**Figure. 3.** Percent mortality of Sunn Pest 9 days post treatment 10 days after collection (Spring 2002 trial in Syria. Bars with the same letters are not significantly different at P < 0.05. Treatments are: 1= Untrt., 2= Tween, 3= Oil, 4= GHA/Twn, 5= 22/Twn, 6= 23/Twn, 7= 200/Twn, 8= 22/Oil, 9= Ma500/Twn, 10= GM/Twn.



**Figure. 4.** Percentage of Sunn Pest cadavers with evidence of fungal infection after surface sterilization (Spring 2002 trial in Syria). Treatments are: 1= Untrt., 2= Tween, 3= Oil, 4= 726/Twn, 5= 22/Twn, 6= 23/Twn, 7= 200/Twn, 8= 22/Oil, 9= Ma500/Twn, 10= MaGM/Twn.



**Figure. 5.** Percent mortality of Sunn Pest in overwintering sites after treatment in the Fall 2002 or Spring 2003. Bars with the same letters are not significantly different at P < 0.05 within a treatment time period. Treatments are: 1= Untrt., 2= Tween, 3= Millet, 4= 22/Nbg, 5= 22/Twn, 6= 22/alg, 7= 566/Twn, 8= 200/Twn, 9= 493/Twn, 10= 2/Twn, 11= 502/Twn, 12= 603/Twn, 13=GHA/Twn, 14= 500B/Twn.

Three months after the 2002 Fall application, evidence of fungal mycelial growth in the plots treated with the granular formulation (isolate 22) was visible, despite the dry climatic conditions and cold temperatures common to these overwintering sites from October to March. Because Sunn Pest commonly overwinter beneath the litter at the soil/litter interface, both soil and litter samples were taken to evaluate where in the overwintering site ecosystem fungal inoculum occurred and if it was at levels adequate for infection. The granular formulation clearly demonstrated a capacity to persist and remain virulent in overwintering sites. Mortality of 65.8 and 71.4% was obtained when Sunn Pest were exposed to soil and litter samples. respectively (Figure 6). No significant differences between the soil and litter samples in the infective potential against

Sunn Pest. The other treatments showed no significant evidence of persistence after 3 mo.



**Figure. 6.** Percent mortality of Sunn Pest 3 mo post treatment (Fall 2002 trial in Syria). Bars with \*\* indicate significant differences from other treatments. Treatments are: 1= Tween, 2= 22/Nbg, 3= 22/Twn, 4= 22/Alg, 5= 566/Twn, 6= 200/Twn, 7= 493/Twn, 8= 2/Twn, 9= 502/Twn, 10= 603/Twn, 11= GHA/Twn, 12= 500B/Twn

Fungal persistence was also assessed on the day of application and 10 and 20 days after application in the Spring 2003 trial. The nutrient-based granular formulation of isolate 22 demonstrated a significantly greater ability to remain persistent for 20 days after application (Figure 7). One other isolate (566), formulated as a spore suspension in Tween 80, also demonstrated a capacity to persist for at least 20 days after application. This is encouraging because the application was made just prior to the emigration of adults to wheat fields and would allow for effective control of Sunn Pest for an extended period of time prior to their migration to wheat fields. Even if the fungal treatment didn't kill the adults immediately, the migrating adults could serve as a source of inoculum once they reach the fields.



**Figure 7.** Percent mortality of Sunn Pest before treatment and 10 and 20 days post application (Spring 2003 trial in Syria). Treatments are: 1= Tween, 2= Millet, 3= Alg, 4= 22/Nbg, 5= 22/Twn, 6= 22/alg, 7= 566/Twn, 8 = 500B/Twn

#### Spring 2003: Iran

When several fungal isolates were tested in a Sunn Pest overwintering site in Iran, some that had shown efficacy in Syria also performed well in this location. In particular, the nutrient-based granular formulation (isolate 22) showed a relatively high level of mortality. Interestingly, mortality was also high in plots treated with millet alone (Figure 8). It is possible that the millet provided a nutrient-rich substrate which promoted growth and sporulation of fungi that occur naturally at the site.

# **General Conclusions**

The results from this series of field experiments demonstrate the potential of using entomopathogenic fungi to manage Sunn Pest in their overwintering sites. Fungi formulated as a nutrient-based granule showed an ability to persist for several months following application, despite extremely adverse conditions. This type of formulation is clearly the most appropriate to consider for treating Sunn Pest overwintering sites. As a result of this

research, all future trials have been with granular rather than liquid formulations. The fungal isolates tested varied in their apparent virulence to Sunn Pest, and three isolates of B. bassiana (22, 566 and 493) appear to hold the greatest promise for further development. Assessment of fungal efficacy remains a challenge as Sunn Pest mortality data alone was found unreliable, and often underestimated infection rates. However, because processing treated insects to detect fungal outgrowth as evidence of infection is extremely time-consuming and requires specialized skills to handle the specimens and correctly identify emerging fungi, it is unrealistic to use this more precise evaluation method for all field-based fungal trials. Many questions remain as to how best to use entomopathogenic fungi to maximize impact against the Sunn Pest under natural conditions, but these experiments provide a foundation upon which future work can proceed.



**Figure 8.** Percent mortality of Sunn Pest following application in Spring 2003 in Iran. Bars with the same letters are not significantly different at P < 0.05. Treatments are: 1= Untrt., 2= Tween, 3= Millet, 4= 22/Nbg, 5= 22/Twn, 6= 566/Twn, 7= 493/Twn, 8= 2/Twn, 9= 502/Twn, 10= 603/Twn, 11=GHA/Twn, 12= 500B/Twn, 13= 151/Twn.

# Acknowledgments

The authors appreciate the many hours of work collecting Sunn Pest by the ICARDA technical staff and assistance with the Iran trials by personnel from the Plant Protection Organization and the Plant Pests & Diseases Research Institute, Varamin, Iran. Statistical analyses were completed by Drs. Takamaru Asikaga and Kyndaron Reinier, University of Vermont, Medical Biostatistics. Support for this research was provided by US Agency of International Development (USAID) Linkage funds, the Conservation, Food and Health Foundation, and Department For International Development (DFID).

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تمت مراقبة جثث حشرات السونة، Eurygaster integriceps Puton، المصابة بالفطور الممرضة للحشرات في مواقع البيات الشتوي، مع اقتراح أن تلك الفطور هي عامل موت طبيعي في هذه البيئة. تم عزل أكثر من 220 عزلة من مواقع البيات الشتوي لحشرة السونة في وسط وغرب آسيا وروسيا. اعتماداً على النتائج من الاختبارات الأحيائية المخبرية و الدراسات على خصائص الإنبات والتبوغ، تم اختيار عدة عزلات واعدة لاختبارها ضد حشرة السونة في مواقع البيات الشتوي في شمال سوريا وشمال وسط إيران. خلال الفترة مابين 2002–2003، أجريت المعاملات الفطرية باستخدام مستحضرات وأوقات تطبيق مختلفة. دونت نتائج هذه الاختبارات ووضعت مقترحات للاتجاهات المستقبلية في تطوير فطور ممرضة للحشرات من أجل إدارة حشرة السونة.

كلمات مفتاحية: مواقع بيات شتوى، Eurygaster integriceps، فطور ممرضة للحشرات، مثابرة.

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# Shifting from Aerial to Ground Spraying for Sunn Pest Control: Farmers' Perceptions and Problems

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### Abstract

Gul, A., A. Aw-Hassan, H. Kutuk, R. Canhilal, A. Mazid, M. Hasani Moghaddam, H. Haj Hamoud and M. El Bouhssini. 2007. Shifting from Aerial to Ground Spraying for Sunn Pest Control: Farmers' Perceptions and Problems. Pages 331-339.

Turkish government has been changing Sunn Pest control policy by shifting from aerial to ground spraying. The Thrace Region is a major cereal growing area and in 2002 and 2003 no aerial spraying for Sunn Pest was done. Our objective was to investigate the impact of this policy change in the region in terms of wheat production, grain quality and what farmers' perceptions of this change were. To achieve this a survey with farmers was done and the results evaluated. The new Sunn Pest management policy received a favorable perception from farmers and government officials in the Thrace region. As a result of two years experience, the adoption of ground spraying became easier than expected although there were some problems like lack of equipment for spraying, tractor crushing of wheat while spraying and inadequate training of personnel.

Keywords: Control methods, Eurygaster integriceps, policy, socioeconomics, Sunn Pest, Thrace Region.

## Introduction

Wheat and barley are very important food crops in the Near East, Middle East, and South-western Asian countries. They are attacked by several species of bugs including Sunn Pest, which is a major threat to wheat and to a lesser extent, barley production. Over 15 million hectares can be affected annually and during outbreaks, infestations may result in 100% crop loss. Damage commonly results in yield losses of 20-30% in barley and 50-90% in wheat. Sunn Pest also injects chemicals into the grain that destroys the gluten and reduces the baking quality of the flour (8, 12).

Presently, chemical control is the main method of protection against Sunn Pest. Within the entire region affected by Sunn Pest about 8 million hectares of wheat are infested annually, of which more than 2 million hectares are treated with insecticides at an estimated cost of 42 million dollars (U.S.). There has been an over-use of insecticides over the past and the shortcomings of pesticide usage in Sunn Pest control and its effect on the environment (particularly reductions of the beneficial entomofauna) have now become evident. Chemical treatment is costly, hazardous to human health and environmental safety, and has a negative impact on agricultural sustainability (2).

Moreover, even without these shortcomings, opportunities for chemical control are likely to become more restricted as costs increase and pesticide subsidies are reduced. The development of new, sustainable and more environmentally benign alternatives is a major priority (12). Sunn Pest populations have increased the last decade and the number of research activities on Sunn Pest has increased in Turkey and most neighboring countries (1, 5, 10, 9, 11, 14). Wheat and barley are strategic crops for Turkey and many other countries. Total cultivated area in Turkey is 26.4 million ha and 69% of it is cultivated field area (18.2 million ha) (11). Wheat is grown on approximately 9.4 million ha in Turkey. Total production of wheat is 19 million tons and the average annual yield is 2,021 kg/ha. Barley is also an important crop for animal feed. The average cultivated area of barley is 3.45 million ha and the total production is 8 million tons (7).

Sunn Pest is one of the most important pests of wheat and barley in Turkey. There are three economically important Sunn Pest species, Eurygaster integriceps Puton, E. maura L., and E. austriaca Schrank (6, 15). Sunn Pest first reached economically damaging levels in South and Southeast Anatolia in 1927, in Thrace in 1982 and in Central Anatolia in 1988. The population density of the pest increased again in South and Southeast Anatolia in 1977 and at present, about 75% of the wheat and barley areas in Turkey are threatened by Sunn Pest (15). In 2002 ~1,492,122 ha of wheat were sprayed against Sunn Pest using both aerial and ground equipment. This sprayed area was increased to 1.882.493 ha in 2003. The purpose of spraving was to prevent damage and to keep populations under the economic threshold, without causing damage to natural enemies.

Organization of Sunn Pest control has been implemented by the Ministry of Agriculture and Rural Affairs (MARA) and all requirements (planes, insecticides) have been met by the Government in Turkey. Pest surveys, egg parasitization and nymphal populations) have been carried out by Sunn Pest experts of MARA (13).

The mission of the government is Sunn Pest management by performing the proper control methods at the right time, and conserving the natural balance to support farmers by preventing damage (3). The government has conducted Sunn Pest management, mainly based on chemical control since 1927. Neither farmers nor technical consultants have been satisfied with their programs.

The area sprayed for Sunn Pest management is given in Table 1. This area has almost doubled from 1997 to 2003. The use of ground equipment as opposed to aerial application has increased rapidly from 8% in 1997 to 56% in 2003.

Sunn Pest spraying methods in Turkey can be seen in Figure 1. According to 2002 data, 39% of the total sprayed area is sprayed with ground equipment. The Thrace Region was important because no aerial spraying was done in 2002. In 2003, spraying by ground increased beyond the government's target (the target was 35% ground spray for 2003) and it was 56% including activities in the Central Anatolia Region.

One goal of the government is to transfer their Sunn Pest management programs to farmers by providing technical information and equipment. Ultimately farmers will conduct Sunn Pest management programs without any help. MARA, Ministry of Finance, State Planning Organizations, Treasury and Office for Field Crops are the main public institutions dealing with Sunn Pest management in Turkey.

These institutions have advantages in Sunn Pest management such as: experience, strong organization, powerful management, good impact on farmers, technology use, and adaptation to plant protection standards in the world. Insufficiency in research & development and experts are the weak sides of the system. There are also some opportunities like supporting the establishment of farmers' unions and assigning sufficient budget. There are also a lot of weaknesses in the system. These are as follows (2):

- Lack of coordination among public institutions;
- Farmers are irrelevant to Sunn Pest management;
- Climatic and geographical structure;
- Farmers' Unions couldn't be established;

- Unconscious intervention of politicians, farmers' associations, large land owners and local administrators;
- Sunn Pest is a big threat in neighboring countries.

The Turkish government has been changing Sunn Pest control policies by shifting from aerial spraying to ground spraying.

The Thrace Region was firstly discussed in Integrated Sunn Pest Management in the West Asia Workshop held in Tehran, ran in October, 2002. A decision was made to conduct a study in the region to determine how much farmers accepted spraying against Sunn Pest by using ground equipment and how successful they were. The purpose of this study is to investigate the impact of this policy change in the region in terms of wheat production and grain quality and farmers' perception of the change To achieve this goal a survey with farmers was done and results were evaluated.

# **Materials and Methods**

The region of Thrace in the Marmara region, has 3,310 sq mi (8,575 sq km), and is southeast of Europe. It occupies the southeastern tip of the Balkan Peninsula and is northeast of Greece, and south of Bulgaria and European Turkey. Its boundaries have varied in different periods. It is bordered by the Black Sea in the northeast and by the Sea of Marmara and the Aegean Sea in the south (4).

In Thrace Region, three major wheat producing provinces were selected Edirne, Tekirdag and Kirklareli for study. Then 141 farmers were randomly selected; 77 from Edirne, 22 from Kirklareli and 42 from Tekirdag. Questionnaires were prepared and completed with face-to-face interviews. The survey was conducted from 13-19 July, 2003. For some of the tables' the Likert scale was used to calculate the points. Besides the survey with farmers, some interviews were done with the technical staff at the Research Institute of Thrace and the Extension Services of MARA. Secondary data related to Sunn Pest were obtained from various units of MARA.

# **Results and Discussion**

## Views and Opinions Gathered from Technical Staff at the Research Institute of Thrace and the Extension Services of MARA

From 1983-2001 farmers and technical staff working for MARA noticed that spraying against Sunn Pest with airplanes was not successful. As a result of this, authorities of MARA decided that spraying against Sunn Pest must be performed with ground equipment by the farmers in the Thrace Region. MARA personnel in the agreed and they persuaded and educated farmers to make this change.

One of the reasons that the farmers accepted spraying with ground equipment, instead of airplane, was kernel damage. In areas sprayed by air kernel damage was high. In the Thrace Region a lot of farmers' wheat was sold through a Commodity Exchange and the higher the quality the greater the price of the wheat. First year spraying with ground equipment, some farmers did not spray against Sunn Pest and they had difficulty selling their wheat. The following year they realized that spraying was necessary. In the second year (2003) almost all farmers sprayed against Sunn Pest with ground equipment. Another reason that the farmers accepted spraying with ground equipment was that it was more effective than spraying with an airplane. Farmers realized this situation while selling their wheat.

There are some difficulties in spraying with ground equipment and many advantages. The most important difficulty is the crushing of wheat by the tractor while spraying. But farmers know very well how much Sunn Pest cause kernel damage when they do not spray. Because of that farmers say they will spray even though crushing damage occurs. Farmers are looking for measures to reduce crushing damage. For this they use narrower wheels on their tractors, they leave gaps in the fields for their tractors and they increase the capacity of their equipment. Farmers complain about the prices of narrower wheels and as a result they sow their seed by drilling which leaves gaps in the field for their tractors. However, in some villages one or two farmers bought narrower wheels and sprayed all the fields in the area. Crushing damage was reduced. Almost all of the farmers in the region accepted the sowing procedure. They believe that leaving some gaps reduces crushing damage and fungal diseases.

In the Thrace Region, MARA technicians have been working there many years and they know the region very well and have a good relationship with the farmers. They are not appointed frequently to other provinces. Thus, they are more effective on changing the habits of the farmers.

Another important point is that farmers have enough ground equipment to spray against Sunn Pest and adhere to changes. However some farmers in some villages of Edirne Province are still opposing ground applications because tractors damage the wheat and they insist on the use of aerial application. Some farmers complained about not receiving pesticides on time.

Most of the farmers do not know about the safety measures for spraying Sunn Pest and number of extension staff is not enough to give the necessary training.

Survey staff of MARA must be very careful about surveys of Sunn Pest to decide on spraying. Because we had some complaints from some villagers saying that officials told them there was no need for spraying against Sunn Pest but after harvest they got wheat with very high percentage of Sunn Pest damage. As a result MARA has lost some of its credibility. As a result of this some farmers obtained their pesticides from the free market and sprayed even though it was not needed. They decided to spray because the price of pesticide was low and they didn't want to take the risk of severe Sunn Pest damage. Eventually this causes the deterioration of the environment because of overuse of pesticides. Thus the government must be more careful about losing its reliability/credibility.

#### **Results of Farmers' Survey**

The average age of the farmers is 48.1 years, share of agricultural income is 86.1% and the ratio of membership to an agricultural organization is 80.1%. Of the investigated farmers, 80.4% graduated from primary school and the remaining from high school or higher education.

Chamber of Agriculture, Agricultural Credit Cooperative, Agricultural Sale Cooperative of Thrace, Irrigation Union, Commodity Exchange and Leader Farmers' Union are the major agricultural organizations that the investigated farmers have membership.

Average farm size was 17.2 ha and 12% of land was irrigated; 50.3% of farmers cultivated land was used for wheat production (8.7 ha).

95.0% of the farmers observed worms and bugs in their fields; 82.3% of farms had weeds and 22.7% had fungi; 97.2% of the farmers were able to recognize Sunn Pest; 82.3% recognized adult Sunn Pest while 61.7% could identify Sunn Pest eggs and 64.5% the nymphal stage. Farmers expressed that they have had Sunn Pest problems for an average of 8.3 years.

In Table 2 data related to farmers' evaluation of Sunn Pest are given. Farmers have good knowledge about Sunn Pest damage (66%). But farmers have very little knowledge of parasitic wasps (22%). Farmers do not feel they have enough information about Sunn Pest (47%).

Farmers (47%) responded that they do not have enough knowledge about the negative effects of aerial spraying on the environment and human health.

Farmers see Sunn Pest in the field for the first time in mid-April. For the question "in your opinion, should Sunn Pest be sprayed as soon as it appears in the field?" 30.5% of the

respondents answered yes. 76.6% of the respondents said that they informed extension service when they see Sunn Pest in the field. Most of the respondents (78.0%) said that they considered some other kind of Sunn Pest control except chemical. Among these they considered biological control (54.5%), parasitic wasps (36.4%) and resistant varieties (9.1%) as other control methods. A large proportion of the respondents (80.1%) want to get training on Sunn Pest.

74.4% of the respondents have difficulties in marketing their wheat. Sunn

Pest damage was the number one wheat marketing problem (70.9%). Structural marketing problems were second (4.3%).

When the farmers were asked about the timing of aerial spraying in previous years, only 33.3% said spraying (aerial) was done on time. Farmers' evaluation about the timing of ground spraying for the last 2-3 years was a good indicator for the success of ground spraying. 83.0% of the farmers noted that spaying (ground equipment) was done on time. The farmers were in favor of ground spraying (Table 2, last row).

		Years						
Region	Methods	1997	1998	1999	2000	2001	2002	2003
Southeastern	Aerial	563.86	941.01	922.63	579.10	529.62	574.36	770.27
Anatolia	Ground	17.29	20.99	21.70	13.73	26.77	43.87	88.10
Southern	Aerial	82.56	213.43	26.67	47.37	71.73	80.10	41.11
Anatolia	Ground	178.00	1.14	400.00	3.22	14.80	25.17	40.24
Central	Aerial	87.25	5.77	39.31	77.43	285.09	254.18	31.69
Anatolia	Ground	5.13	160.00	787.00	1.61	17.63	35.82	205.60
Aegean	Aerial	500.00	22.17	89.86	-	-	-	-
C	Ground	-	2.45	3.36	25.20	60.26	100.53	184.47
Thrace	Aerial	178.71	361.42	212.44	7.57	42.95	-	-
	Ground	48.60	56.78	49.62	16.240	39.26	357.30	487.71
Marmara	Aerial	-	-	8.53	-	-	-	-
	Ground	3.40	-	1.50	9.70	24.03	19.90	33.15
Total		973.18	1.625.31	1.375.79	803.06	1.112.13	1.492.12	1.882.49

Table 1. Sunn Pest sprayed area by regions in Turkey (ha).

Table 2. Farmers' evaluation on Sunn Pest and its control.

Evaluation	Likert scale*
Farmers' knowledge about damages caused by Sunn Pest	2,63
Farmers' knowledge about parasitic wasps**	0.87
Sufficiency of farmers' information about Sunn Pest	1.87
Farmers' satisfaction with Sunn Pest spray conducted by MARA	2.60
Farmers' knowledge about the negative effects of aerial spraying on environment and human health	1.89
Farmers' opinion about the continuation of the current policy (ground spraying)	2.85

\* Questions were asked to indicate on a 5-point Likert scale to which the statements indicate sufficiency of the criteria. Likert scale: 4=very sufficient...0=definitely insufficient.

\*\* 30% of the respondents gave reasonable answers for parasitic wasps

In ground spraying, availability of equipment is very important for success: 78.7% of the farmers said that they have enough equipment for spraying Sunn Pest with ground equipment. Some of the farmers also told that the extension services provided equipment for Sunn Pest control (2.1%). Some also expressed that the availability of tractors with narrow wheels was necessary.

90.8% of the respondents said that government officials gave them pesticides on time. The remaining had problems obtaining pesticides from the government. Some farmers (5.0%) expressed that using ground equipment harmed wheat production so they tended to use the pesticide given to them for other crops. But it is not a common practice.

A significant proportion of the farmers (55.3%) expressed that they noticed some positive environmental change (like increasing number of snakes, rabbits, turtles etc.) after ground spraying started.

In general 65% of farmers are satisfied with Sunn Pest control conducted by MARA. Some farmers were not satisfied with it and gave reasons why. Insufficient pesticide dose, no spraying, late spraying, ineffectiveness of pesticides and lack of agricultural extension are the major reasons in sequence.

Another important key point was the effectiveness of ground and aerial spraying according to farmers' opinion. Ground spraying was expressed as more effective by 68.1% of the respondents while aerial spraying by 27.7%. That is a good indicator for new policy acceptance and success by the farmers. Aerial spraying was found more effective because (1) no crop lost because of spraving. (2) more effective and (3) the whole and wider area is sprayed. The reasons for ground spraying effectiveness are expressed as (1) more effective (spraying lower elevation, only target crop is effected), (2) better timing, (3) much care on own field and (4) less environmental damage. Farmers' negative opinions for ground spraying can be listed in order of importance: (1) High crop damage, (2) lack of equipment, (3) negative effect on human health and (4) higher cost.

# Changes in Spraying Cost as a Result of Different Spraying Methods

The cost which is calculated in this section refers to the additional cost that farmers should bear as a result of shifting from aerial to ground spraying. Average cost of ground spraying was calculated 14,780,000 TL/ha (10.56 USD/ha). Labor, fuel, machinery and equipment are the major cost items. Besides this cost, there is another important one called opportunity cost, which is the crop lost due to ground spraying. In this case three scenarios should be considered:

- For the current situation nearly all the farmers do not leave unplanted space for tractor tires. If normal tractor tires are used, the crop lost will be 5%.
- 2) It will be 2 percent if narrow tractor wheels are used.
- 3) Very few farmers leave lines for tractor to work, in this case there will be no crop lost.

The average wheat yield is 3000 kg/ha in the region and the average wheat price is 330,000 TL/kg according to the Edirne Commodity Exchange. Using these data, additional opportunity cost can be calculated for each of the three scenarios (49.500.000 TL/ha -35.36 USD/ha-, 19.800.000 TL/ha -14.14 USD/ha- and no cost). If we add the machinery related cost given at the beginning that means 45.92, 24.70 and 10.56 USD in scenario order.

According to MARA, average airplane hiring cost is 344.000 TL/da/airplane (2.46 USD/ha/airplane) for ULV and 625.000 TL/da/airplane (4.46 USD/ha/airplane) for conventional (plus V.A.T.). This is the government saving as a result of no aerial spraying.

# Conclusions

Spraying with ground equipment is essential in terms of social, environmental, cost and health aspects. Therefore, the policy the current government and the previous one is following is correct. Another important factor is that this policy has become a national policy rather than a political matter. Current government's policy is the continuation of the previous one and this is a new progress in that respect because the Sunn Pest control issue has always been a way of pleasing voters.

The major finding of this study is that the new Sunn Pest management policy gives good results in terms of farmers' perception and government officials in Thrace Region of Turkey. As a result of two years experience, the adoption of ground spraying has become easier than expected although there were some problems such as the lack of equipment for spraying, tractor crushing while spraying and training.

The educational level of the farmers in the region is higher than other parts of the country. Also, most of the farmers are willing to get training on Sunn Pest control. Therefore, extension services should work harder and be well-organized in the region. The farmers in the region are aware of how Sunn Pest damage is important to get good quality wheat yield. Even there were unexpected problems (organization, providing equipment and pesticide on time and etc.) at the beginning stage of policy change; the farmers were in favor of ground spraying.

Shifting from aerial spraying to spraying with ground equipment causes some problems because farmers and technical staff are not trained and lack of equipment forces farmers to make new organizations to control Sunn Pest.

Aerial spaying must be ended unless there is an obligation in terms of technical,

economical and geographical reasons. Field surveys must be done correctly and in time. Staff dealing with Sunn Pest business should be trained and paid for their extra work.



**Figure 1**. Sunn Pest spraying methods in Turkey in 2002 and 2003.

# Acknowledgment

The authors wish to express their gratitude to the Research Institute of Thrace and Extension Service of MARA for providing the data related to Sunn Pest control and damage, transportation and technical assistance. The authors also would like to thank ICARDA for providing some funds to accomplish this study.

# الملخص

غول، أيكوت، آدن أو –حسن، خليل كتك، رمضان جانهلال، أحمد مزيد، مجيد حسني مقدم، حمود حاج حمود ومصطفى البوحسيني. 2007. التحول من الرش الجوي إلى الرش الأرضي لمكافحة حشرة السونة: إدراك الزراع ومشاكلهم. الصفحات 311–339.

غيرت الحكومة التركية سياسة مكافحة حشرة السونة بالتحول من الرش الجوي إلى الرش الأرضي. تعد منطقة ثراس (Thrace) في اقليم مرمرا من أهم مناطق زراعة الحبوب، في عامي 2002 و 2003 لم يطبق الرش الجوي لمكافحة حشرة (Thrace) السونة. هدف هذا البحث إلى تقصي أثر هذا التغيير في سياسة المنطقة من حيث إنتاج القمح، نوعية الحبوب وكيف كان إدراك الراع لهذا التغيير. لتحقيق ذلك أجري مسح للزراع وتم تقييم النتائج. تم تلقي السياسة الجديدة لإدارة حشرة السونة بإدراك إيجابي من قبل الزراع لهذا التغيير. لتحقيق ذلك أجري مسح للزراع وتم تقييم النتائج. تم تلقي السياسة الجديدة لإدارة حشرة السونة بإدراك إيجابي من قبل الزراع وموظفي الحكومة في منطقة ثراس. وبمحصلة نتائج خبرة عامين، وجد بأن تنبني الرش الأرضي أصبح أسهل من من قبل الزراع وموظفي الحكومة في منطقة ثراس. وبمحصلة نتائج خبرة عامين، وجد بأن تنبني الرش الأرضي أصبح أسهل من المتوقع رغم وجود بعض المشاكل كنقص معدات الرش، سحق الجرارات للقمح خلال الرش، والتدريب غير الكافي للموظفين. كلمات مقتاحية: طرائق مناحية التقاعيم مناحقة من معدات الرش، سحق الجرارات القمح خلال الرش، والتدريب عبر الكافي الموظفين.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Positive Effects of Commodity Exchanges in Sunn Pest Control in Turkey

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## Abstract

## Gul, A., A. Aw-Hassan, H. Isik, R. Canhilal, A.D. Kanat, A. Mazid and M. El Bouhssini. 2007. Positive Effects of Commodity Exchanges in Sunn Pest Control in Turkey. Pages 341-348.

Wheat is a strategic and basic food for Turkey. Sunn Pest is the major damaging insect of wheat. Since 1927 Sunn Pest management has been done mainly with agricultural chemicals. In recent years the government Sunn Pest control policy has been changing. They are shifting aerial spraying to ground spraying. In year 2003 Central Anatolia and Thrace Regions were excluded from aerial spraying and only spraying with ground equipments was applied. Konya and Edirne Commodity Exchanges are the two major Commodity Exchanges in Turkey in terms of wheat marketing. A study was conducted to determine the effect of Sunn Pest damage on the wheat market and to understand the positive effects of commodity exchanges on Sunn Pest management by farmers. In this context the operations of these two commodity exhanges were investigated and a survey was done with merchants in the Konya Commodity Exchange and we investigated how the commodity exchanges operated. Establishing a free market is the number one priority to evaluate Sunn Pest damage and commodity exchanges have an important role in Sunn Pest control.

Keywords: Commodity exchange, pest control, Sunn Pest, Turkey, wheat price.

# Introduction

Sunn Pest has increased in importance over the last decade and thus the number of research projects on Sunn Pest has increased in Turkey and most neighboring countries (2, 7.11.10.12.14).

Chemical control is the main method of protection against Sunn Pest. In the region about 8 million ha of wheat are infested annually, and >2 million ha are treated with insecticides at an estimated cost of 42 million dollars (US). There has been an overuse of insecticides and the shortcomings of pesticide usage in Sunn Pests control and their effect on the environment (particularly reductions of beneficial entomofauna) have now become evident. Chemical treatment is costly, hazardous to human health and environmental safety. It has a negative impact on agricultural sustainability. Opportunities for chemical

control are likely to become more restricted as costs increase and pesticide subsidies are reduced. The development of new, sustainable and more environmentally benign alternatives is a major priority (13).

Wheat and barley are strategic crops for Turkey and many other countries. The total cultivated area in Turkey is 26.4 million ha and 69% is cultivated field crops' area (18.2) million ha) (16). Wheat is grown on approximately 9.4 million ha. Total production of wheat is 19 million tons and the average yield is 2,021 kg/ha. Barley is also an important crop for animal feeding. Average cultivated area of barley is 3.45 million ha and production is 8 million tons (9).

Sunn Pest is one of the most important pests of cereals (wheat, barley) in Turkey. There are three economically important species, Eurygaster integriceps Puton., E. maura L., and E. austriaca Schrank (8, 15).

Sunn Pest first reached economically damaging levels in South and Southeast Anatolia in 1927, in Thrace in 1982 and in Central Anatolia in 1988. The population density of the pest increased again in South and Southeast Anatolia in 1977 and at present, about 75% of the wheat and barley areas of Turkey are under threat from Sunn Pest (15).

In 2002, 1,492,122 ha of wheat area were sprayed against Sunn Pest using aerial and ground equipment. This sprayed area was expanded to 1,882,493 ha in 2003. Spraying was done to prevent damage, and to suppress Sunn Pest populations under the economic threshold, without causing any damage to the natural balance.

Organization of Sunn Pest control has been implemented by the Ministry of Agriculture and Rural Affairs (MARA) and all requirements (planes, insecticides) have been arranged by the Government of Turkey. Pest surveys (prespray, postspray evaluations, egg parasitization and nymph population determinations) have been done by Sunn Pest experts of MARA (12).

The mission of the government for Sunn Pest management is to ensure that the correct control methods are used at the right time, and to conserve the natural balance and to support farmers by preventing damage by Sunn Pest (3).

Total area sprayed for Sunn Pest has almost doubled from 1997 to 2003. Area sprayed by ground equipment instead of by plane has increased from 8% in 1997 to 56% in 2003). According to the 2002 figures, 39% of the total sprayed area was done with ground equipment. No aerial spraying was done in Thrace Region in 2002. In 2003 the area sprayed from the ground increased beyond the government's target of 35%.

The government wants to transfer Sunn Pest management to the farmers by gradually providing technical information and equipment. Ultimately farmers will conduct Sunn Pest management without help, like other pest management programs.

Establishing free market conditions is essential in terms of sustainability and effective use of resources. In recent years commodity exchanges are becoming more and more important for agricultural crops, especially wheat. Wheat has been supported by the government for about 60 y. TMO (Fields' Crops Purchasing Organization) has been the most powerful organization in the wheat market but its importance has been reduced because it purchases small quantities from the Turkish wheat market. In previous years TMO bought wheat even with high damage by Sunn Pest. Therefore, wheat producers did not care about the quality of their wheat. In 2003 TMO bought very little wheat and producers had to sell it on the free market.

Until recently wheat prices were mainly determined by the government in Turkey. As a result of an agreement with the World Trade Organization, the government's effect on wheat prices has been rapidly decreased for the last few years. This situation has made commodity exchanges more important to providing free market conditions.

Commodity exchange can be expressed as an environment in which demand and supply are met (1).

Commodity exchanges are available in almost every province in Turkey and they play a significant role in price determination of wheat in Turkey. The functions of a commodity exchange can be itemized as follows (4):

- To arrange and register the trade of commodities and to establish and announce daily prices,
- To create general rules and decisions, with the acceptance of TOBB (the Union of Chambers of Commerce, Industry, Maritime Trade and Commodity Exchanges), about obligations of delivery, taking delivery and payment, and arbitration procedures in conflicts,
- To follow and communicate with domestic and foreign exchanges and markets,

- To construct laboratory and technical offices to establish types and qualities of materials that are traded in the exchange,
- To arrange and announce the custom and practices of exchange and make duties given by other regulations and Ministry of Industry and Trade.

The Turkish Government has been changing Sunn Pest control policies by shifting from aerial spraying to ground spraying. Thrace and Central Anatolia Regions are the major cereal growing areas and they have an important contribution in increasing the share of ground sprayed area in the country. In 2003 no aerial spraying was done in Thrace and Central Anatolia. Four commodity exchanges (Edirne, Konva. Eski ehir, and Polatlı) are located in these regions. Konya and Edirne Commodity Exchanges were established as a result of a World Bank Project. Konya is one of the top exchanges which determine wheat prices at the country level and has about 4000 member merchants. That is why the members of this exchange were selected to survey.

A study was conducted to determine the effect of Sunn Pest damage on the wheat market and to understand the positive effects of commodity exchanges on Sunn Pest management by farmers. In this context a survey was done with merchants in KCE and we investigated how the KCE and Edirne Commodity Exchanges operated.

# **Materials and Methods**

Information about the role and operations of the commodity exchanges was obtained from administration, staff and members of Konya and Edirne Commodity Exchanges. Records and printed materials from these institutions were also used in this study.

A survey was conducted with 47 randomly selected members of KCE. Each was interviewed right after their auctions in KCE. The study was conducted in August, 2003. For some of the tables the Likert scale was used and the points were calculated in the analysis.

Surveys were designed to determine the importance of the commodity exchanges, their operations and the members understanding of Sunn Pest and wheat price formation.

# Results

## **Commodity Exchanges**

The first commodity exchange was established in 1891 in Izmir, followed by one in Adana in 1913, Antalya in 1920, Edirne, Konya, Bursa in 1924 and Istanbul in 1925. By 1950 a total of 24 commodity exchanges were established. After 1950, with more common and detailed legislative regulations, the Union of Chambers of Commerce, Industry, Maritime Trade and Commodity Exchanges (TOBB) in Turkey were established and today almost every province has at least one commodity exchange (total number is 110) (5, 6).

In the competitive atmosphere that is created by the commodity exchanges, agricultural products find their real prices and there are heavy charges on the budget of the government because their support of agricultural products decreases. The development of the commodity exchanges is important for the application of the Premium System, wide spreading documentation system and increasing the tax income of the state.

In the markets, with the competitive atmosphere created by the commodity exchanges, the producer will understand that the product is traded on its real value, the quality is more important than the quantity and he will produce according to the productivity of seed, fertilizer, medicine and similar production factors. The income of the producer will increase; he will have new credit and insurance opportunities, so this will increase the welfare level of the country.

The circulation of information, which is provided by the competitive atmosphere, makes producers behave as associations to maximize their advantages. The merchant determines prices by estimating supply and demand and projecting these in the market according to quality and quantity factors and this decreases the costs which are then reflected to the government from the point of storage.

With the development of the commodity exchanges, the Agriculture Based-Industries will have the chance to have income in market conditions that will enable them to become competitive worldwide. With increased competitive power, the Agriculture-Based Industry will be saved from instability by making long-term production and sales plans.

The prices of the products form in the free competition conditions in the exchange. So these prices are the most acceptable prices in the country for both sellers and buyers.

In the exchange, by registering the purchases and sales, the documentation system and the registered economy are developed.

The people that sell and purchase materials have to register with the commodity exchanges in their regions. Those who don't register are considered ex-officio by the exchanges. The producers or the agents that register to the exchanges can sell their products without registering to the exchanges (4).

#### **Operations in KCE**

Basic operations for a given crop in KCE can be expressed as follows (4):

- 1. Sampling Building
  - 1.1. Applications of producers who come to the exchange to sell their products are accepted here.
  - 1.2. Samples are taken from trucks.
  - 1.3. Place to enter the information about the products of which sample is taken.
- 2. Laboratory Operations
  - 2.1. Laboratory in which physical and chemical analyses are done.
  - 2.2. Analysis operations in the lab.
  - 2.3. Modern equipment is found in the laboratory.
  - 2.4. Wheat, corn and cumin are analyzed before selling and other crops on request.

- 2.5. If the sold product and the sample in the exchange don't match, required analyses are done and the results are reported.
- 3. Producer Waiting Hall
  - 3.1. Producer can view and witness the selling of their products in a comfortable ambiance.
- 4. Trading Floor Activities
  - 4.1. Samples of which procedures are completed are ready for selling.
  - 4.2. Officers show samples to all buyers.
  - 4.3. Buyers examine samples.
  - 4.4. After the examination and decision to buy, discussions (negotiations) occur between rival buyers.
  - 4.5. This procedure continues until a maximum price of product is decided.
  - 4.6. Lastly, an officer announces the completion of the sale.
- 5. After the sale

The cashier in the trading floor announces the name of the owner of the product and asks for the decision of the producer relevant to the to the price. If the producer decides to sell his product, registration occurs. The producer is free to sell or not. The results are posted on boards.

# Results of the Survey with Merchants in KCE

A total of 47 members of KCE were surveyed. Educational levels of the members were as follows: university graduates (34.04%), high school (34.04%) and primary school (31.9%). Average age was 43 and average experience in wheat trading 15.7 years.

The primary crops that merchants trade were wheat (87.2%), barley (10.6%) and beans (2.1%). As secondary crops, barley (42.6%), beans (8.5%) and other cereals (8.5%).

Of the respondents, 57.5% preferred buying crops in a commodity exchange, only 4.3% bought crops directly from producers and 38.3% preferred both methods.

Knowledge level of the merchants about Sunn Pest and its damage was above average; 2/3 of the merchants expressed that they had good or very good knowledge while 1/4 said it was moderate and 10% had little or no knowledge (Figure 1).

Seventy five percent of the respondents said that they bought wheat with Sunn Pest damage. Figure 2 shows the highest percentage of Sunn Pest damage that the merchants considered to buy. Twenty five percent of the merchants said they bought only damage-free wheat. Fifteen percent said wheat with up to 2% kernel damage was acceptable to buy. About 40% of the merchants would not buy wheat with kernel damage >2%.



Figure 1. Merchant's knowledge of Sunn Pest and its damage.



Figure 2. The highest percentage of wheat with Sunn Pest damage that merchants considered to buy.

Laboratory results were critical for wheat price formation in the commodity exchanges. Sixty six percent of the merchants found the results reliable. Nineteen percent said the results were unreliable. The remaining 15% either have no idea about it or did not answer this question (Figure 3).

Figure 4 shows the criteria the merchants considered when they bought wheat. Sunn Pest damage (1.07) was the most important determinant. Variety and the region where the wheat was produced were the other important factors.



**Figure 3.** Merchants' opinion about the wheat laboratory results of KCE.



**Figure 4.** Criteria merchants considered when they bought wheat. Questions were asked to indicate on a 4-point Likert scale to which the statements indicate sufficiency of the criteria where 0=very important...4=not important.

Another significant result of the survey was the merchants' evaluation for wheat varieties in the region. Bezostaja (1.78) and Kiziltan (1.81) were the best varieties to buy (Figure 5). Another question was related to the wheat price formation and the factors affecting it. Quality of crops (1.32) was the most
important price determinant. Number of buyers (1.60) and amount of crop (1.78) were the other important factors (Figure 6).

Lastly, Figure 7 indicates the merchants' evaluation of KCE. In general, most of the indicators were good or around average. In terms of the number of buyers (1.43), amount of commodity sold (1.44) and speed of processing (1.52), KCE received favorable evaluations from the merchants.



**Figure 5.** Merchant's evaluation of wheat varieties in the region. Questions were asked to indicate on a 4-point Likert scale to which the statements indicate sufficiency of the criteria where 0=very good... 4=not good.

## Conclusions

The two important policy changes have been realized in terms of wheat production and marketing. Sunn Pest control policies have been changing from aerial spraying to ground spraying and the government's effect on wheat price has been rapidly decreased. These two policy changes should be performed simultaneously.

Commodity exchanges are becoming more and more important because merchants and producers did not know about Sunn Pest damage and how important it was. Commodity exchanges provided sampling units and laboratories to measure Sunn Pest damage and informed buyers and sellers of results. This was essential to establish free market conditions. If TMO was powerful in the wheat market as in previous years, the rate of farmers who take Sunn Pest serious would be very low. Nobody would care about Sunn Pest control. But now because of commodity exchanges, the wheat price is clearly affected by Sunn Pest damage and so the farmers are more conscious about its control.

Because farmers are now aware of the situation and ready to receive information about this issue from the extensionists and any other organizations, MARA and other organizations must provide this information and establish the required infrastructure.



**Figure 6.** Merchants' opinion of factors affecting wheat price. Questions were asked to indicate on a 4-point Likert scale to which the statements indicate sufficiency of the criteria where 0= very important........ 4= not important.



**Figure 7.** Merchants' evaluation of KCE. Questions were asked to indicate on a 4-point Likert scale to which the statements indicate sufficiency of the criteria where 0=very good..... 4=not good.

### Acknowledgment

The authors wish to express their gratitude to the administration of Konya and Edirne Commodity Exchange for providing the information related to Sunn Pest control and damage, operations of the system and technical assistance. The authors also would like to thank ICARDA for their support.

## الملخص

غول، أيكوت، آدن أو-حسن، ه. إيسك، رمضان جانهلال، أ. د. كانات، أحمد مزيد ومصطفى البوحسيني. 2007. التأثيرات الإيجابية لمراكز تجارة السلع في مكافحة حشرة السونة في تركيا. الصفحات 341-348.

يعد القمح غذاء استراتيجيا وأساسيا لتركيا، وتعد حشرة السونة الآفة الحشرية الضارة الرئيسة للقمح. منذ 1927 تمت إدارة حشرة السونة بشكل رئيس بالكيماويات الزراعية. تغيرت سياسة الحكومة في مكافحة حشرات السونة في الأعوام الأخيرة، وتحولوا من الرش الجوي إلى الرش الأرضي. في عام 2003 تم استثناء منطقتي وسط الأناضول و تراس من الرش الجوي وطبق الرش بالمرشات الأرضية فقط. مركزا تجارة قونيا وإدرن هما مركزا التجارة الرئيسان لتجارة القمح في تركيا. أجريت دراسة تأثير ضرر حشرة السونة على تجارة القمح ولفهم التأثيرات الإيجابية لمراكز التجارة في إدارة حشرة السونة من قبل الزراع. تم في هذا السياق تقصي عمليات الشراء في هذين المركزين كما تم تتفيذ استبيان مع التجارة وي مركز تجارة قونيا وتم تقصي عمل مراكز التجارة. إن إقامة سوق حرة هي الأولوية رقم وإحد لتقييم ضرر حشرة السونة وتسهم مراكز التجارة بدور هام في مكافحة حشرة السونة.

كلمات مفتاحية: مركز تجارة السلع، مكافحة الآفة، حشرة السونة، تركيا، سعر القمح.

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(Hemiptera, Scutelleridae): the role of mycoinsecticides in management schemes. Proceedings of the First Workshop of Integrated Sunn Pest Control. January 6-9, 1998. Ankara, Turkey.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Loss Assessment and Economic Levels for Sunn Pest in Qazvin Province, Iran

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#### Abstract

Nouri, H. 2007. Loss Assessment and Economic Levels for Sunn Pest in Qazvin Province, Iran. Pages 349-354.

Sunn Pest is the most destructive and damaging pest of wheat and barley in most parts of Iran. It reduces yield and quality of the crop. To develop an IPM strategy for the management of Sunn Pest, it was necessary to determine the economic injury level (EIL) of the pest. For over wintered Sunn Pest, a factorial experiment based on a randomized complete block design was done under cage conditions at Feiz-Abad Research Station of Qazvin Province in 1998 and 1999. Wheat cultivars, Alvand and Zarrin and different densities of over wintered Sunn Pest (2, 4, 6, 8, 10, 12, 15, 17 and 20 insect per m<sup>2</sup> and an insect free check) were tested. Using Pedigo's (1986 and 1989) equations, the calculated mean of EIL for cultivars in fields with aerial chemical control was 10.65 and 10.94, and in fields with ground applied chemical control was 13.63 and 14.01 over-wintered Sunn Pest per m<sup>2</sup>.

Keywords: Economic injury level (EIL), Eurygaster integriceps Puton, Iran, wheat, Sunn Pest.

#### Introduction

Wheat is the most strategic field crop in Iran and it plays an important role in the agricultural sector of the country. Wheat and barley are attacked by many different insect pests, but the Sunn Pest, Eurygaster integriceps Puton, is the most destructive and damaging pest throughout the country. Sunn Pest causes a reduction in yield and quality. Therefore, management of this pest is deemed the most important plant protection program of the government and private sector. Its biology, ecology and control methods have been the subject of numerous research projects, and from the standpoints of research effort and resource allocation it surpasses any other insect pest of the country (1). Recently efforts have been initiated to develop а comprehensive IPM strategy for Sunn Pest.

According to Volddichev (14), the EIL for spring wheat of *Triticom aestivum* L. is 0.5 and 1.5 over wintered Sunn Pest per m<sup>2</sup> in dry years and wet years, respectively. The EIL for winter wheat varied from 0.3 over wintered Sunn Pest per m<sup>2</sup> in dry years to 1.0 in wet years. Popov *et al.* (8, 9) estimated the EIL of

the pest to be 5 over wintered Sunn Pest per  $m^2$  in Romania. In Turkey, the EIL was 3 over wintered Sunn Pest per  $m^2$  in a cage study (15).

In Iran, different studies have given different results (5). The EIL varied from 0.63 to 1.0 over wintered Sunn Pest per  $m^2$  in Kordestan. In Kermanshah dry-land crops it was 1.4 over-wintered Sunn Pests per  $m^2$ (3).

In a two-year pilot IPM project done 1997-1998 in different parts of the country, the estimated EIL ranged from 2-3 over wintered Sunn Pest per  $m^2$  in dry land wheat with an expected yield of 2-3 ton per ha (1).

Since the availability of an accurate EIL is important to the development of an IPM program the purpose of our research was to conduct an extensive study with different densities of the pest under large walk-in cages to determine the EIL with higher precision.

#### Materials and Methods

The EIL of the Sunn Pest was studied in large screen cages. The quantitative damage of the over wintered Sunn Pest was studied in 1998 and 1999 at Feiz-Abad Research Station,

Qazvin. A factorial experiment with two variables based on a randomized complete block design (RCBD) with five replications was used. Wheat varieties used were Alvand and Zarrin, and adult insect densities were 2, 4, 6, 8, 10, 12, 15, 17 and 20 over wintered Sunn Pest per m<sup>2</sup> and an insect free check. Winter wheat was planted in the fall of 1997 and 1998. Plot preparation, sowing, fertilizers used and other agronomic practices in the experiment were similar to those in commercial wheat fields of Qazvin, except that no insecticides were applied. Each plot consisted of 9 wheat rows, 2 m long, with 15 cm between rows.

About two weeks before the immigration of the over wintered Sunn Pest to the wheat fields, a  $2.00 \times 1.50 \times 1.80$ -m cage with 28 mesh screening was placed over each plot. One hundred cages were used in this study. Adult insects were collected from their over wintering sites on March 1, and cages were infested with predetermined number 2, 4, 6, 8, 10, 12, 15, 17 and 20 adults of equal sex ratio. After 20 days, the number of plants with damaged terminals and the number of plants with dried spikes (white heads) were counted. In each plot, the yield loss was estimated using the following equations (12):

$$\begin{split} I_t &= (I_s + I_h) / N_s \qquad (I) \\ D_s &= I_t \times Y_s \qquad (II) \end{split}$$

In the above formula  $(I_t)$  is the number of damaged terminal buds and dried spikes per over wintered Sunn Pest per m<sup>2</sup>,  $(I_s)$  is the number of damaged terminal buds per m<sup>2</sup>,  $(I_h)$  is the number of dried spikes per m<sup>2</sup>,  $(N_s)$  is the number of over-wintered Sunn Pest per m<sup>2</sup>,  $(D_s)$  is the yield loss per over wintered Sunn Pest per m<sup>2</sup> in grams and  $(Y_s)$  is the mean yield per spike in grams.

Using the data acquire as above, the economic injury level of the pest was estimated using the following formula of Pedigo *et al.* (7) and Pedigo (6):

$$EIL = \frac{C}{(V.I.D.K)}$$
(III)

$$GT = \frac{C}{V}$$
(IV)  
$$EIL = \frac{GT}{b}$$
(V)

(EIL) is the number of over wintered Sunn Pest per m<sup>2</sup>, (C) is the cost of chemical control of the pest in Rials per ha., (V) is the market value of the crop in Rials per Kg, (I) is the injury units per over-wintered Sunn pest per ha., that is expressed as the proportion of damaged terminal buds and dried spikes, (D) is damage per unit injury (kg yield loss per ha. per percentage of damaged terminal buds and dried spikes), (K) is the proportionate reduction in potential injury or damage, (b) is yield loss per over-wintered Sunn pest and (GT) is the ratio of control cost to market value.

#### **Results and Discussion**

The mean of damaged terminal buds and dried spikes, yield loss, yield loss percentage and weight of 1000 grains corresponding to different number of over wintered Sunn Pest per m<sup>2</sup> is revealed in Table 1. In both cultivars, yield loss was highly correlated with over wintered Sunn Pest per m<sup>2</sup> (r = 0.975, P<0.01for Alvand and r = 0.988, *P*<0.01 for Zarrin) (Table 2). In the first method Pedigo et al. (7) was calculated using the formulae (III) for each over wintered Sunn Pest in aerial and land chemical controls (Table 3). Correlation between insect number (as log) and EIL for aerial and land chemical control were 0.958 and 0.899 on Alvand and Zarrin varieties, respectively and are given in Table 4.

The logarithmic regression between EIL and insect density on Alvand variety at aerial and land chemical controls with equations Y=1.519+9.815log10(X),  $R^2 = 0.935$ was: and Y=1.946+12.564log10(X),  $R^2=0.935$ . respectively. Between economic injury level and Sunn Pest adult density on Zarrin variety at aerial and land chemical controls it Y=5.151+6.222log10(X),  $R^2 = 0.731$ was; Y=6.608+7.954log10(X),  $R^2 = 0.732$ , and respectively.

No. of adult insects	Yield (kg/ha)		of lt Yield Yield loss ects (kg/ha) (kg/ha) % Yield loss		Weig 1000 g	ht of grain	No. of damaged terminal bud & dried spikes			
per m <sup>2</sup>	Alv.	Zar	Alv.	Zar.	Alv.	Zar.	Alv.	Zar.	Alv.	Zar.
0	6242.7	7100.2	0.0	0.0	0.0	0.0	32.5	34.2	0.0	0.0
2	5995.7	6933.2	247.0	167.1	4.0	2.4	32.1	34.3	14.6	9.1
4	5922.6	6827.8	320.1	272.4	5.1	3.8	32.2	34.1	19.0	14.6
6	5921.7	6796.2	321.0	304.0	5.1	4.3	32.7	33.7	19.2	16.4
8	5835.2	6776.8	407.5	323.4	6.5	4.6	32.6	33.5	24.3	17.2
10	5787.7	6652.2	455.0	448.0	7.3	6.3	32.0	34.1	27.4	24.0
12	5783.4	6605.1	459.3	495.1	7.4	7.0	32.6	33.7	32.7	26.4
15	5629.7	6464.2	613.0	638.0	9.8	9.0	32.2	34.0	36.7	34.0
17	5529.2	6365.9	713.5	734.3	11.4	10.3	33.0	33.4	43.0	40.0
20	5469.3	6176.6	773.1	923.6	12.4	13.0	32.0	33.8	47.1	50.1

Table 1. Mean number of damaged terminal buds and dried spikes per  $m^2$ , yield loss and yield loss percentage on wheat varieties.

**Table 2.** Correlation coefficients between yield loss percentage, yield loss, number of damaged terminal buds and dried spikes with the adult Sunn Pest density.

	%Yield loss		Yield	loss	No. of d terminal dried	amaged bud and spikes
	Alv.	Zar.	Alv.	Zar	Alv.	Zar.
% Yield loss in Zar.	$0.982^{**}$					
Yield loss in Alv.	0.993**	$0.979^{**}$				
Yield loss in Zar	$0.981^{**}$	$1.000^{**}$	$0.979^{**}$			
No. of damaged terminal buds and dried spikes in Alv.	0.994**	0.982**	1.000**	0.982**		
No. of damaged terminal buds and dried spikes in Zar.	0.981**	1.000**	0.977**	1.000**	0.980**	
No. of adult Sunn pest	0.971**	0.989**	$0.975^{**}$	$0.988^{**}$	$0.977^{**}$	$0.987^{**}$

\*\* Significant at ph 0.01

**Table 3.** EIL for different densities of over wintered Sunn Pest on Alvand and Zarrin varieties at aerial and land chemical controls.

Insect	Aerial c con	hemical trol	Land chemical control		
No./m <sup>2</sup>	Alvand	Zarrin	Alvand	Zarrin	
2	4.28	6.34	5.48	8.11	
4	6.61	7.78	8.47	10.00	
6	9.9	10.44	12.66	13.37	
8	10.37	13.10	13.28	16.77	
10	11.36	11.81	14.88	15.12	
12	13.83	12.83	17.71	16.42	
15	12.94	12.44	16.57	15.92	
17	12.59	12.25	16.12	15.69	
20	13.67	11.45	17.52	14.66	
mean	10.65	10.94	13.63	14.01	

Logarithmic equations were calculated for EIL and yield loss per over wintered Sunn Pest on Alvand variety at aerial and land chemical controls including: Y=24.791-19.358log10(X), R<sup>2</sup>=0.933 and Y=31.73-24.772log10(X),  $R^2 = 0.933$ respectively. Logarithmic regression between EIL and yield loss per over wintered Sunn Pest on Zarrin variety at aerial and land chemical controls was: Y=25.932-21.533log10(X),  $R^2 = 0.933$ Y=33.17-27.521log10(X), and  $R^2 = 0.994$ respectively.

The mean of EIL for different over wintered Sunn Pest densities was 10.65 and 10.94 insects per m<sup>2</sup> on Alvand and Zarrin

varieties in aerial chemical control and 13.63 and 14.01 insects per m<sup>2</sup> on Alvand and Zarrin varieties in land chemical control, respectively.

Over wintered Sunn Pest EIL was calculated using Pedigo's formula (IV) and (V). Linear regression was observed between yield  $(g/m^2)$  and insect density on both cultivars (Figures 1 and 2). In the regression equations they were b=3.380 and b=4.121 on Alvand and Zarrin varieties, respectively.



**Figure 1.** Linear regression between over wintered Sunn Pest density and yield on Alvand variety.



**Figure 2.** Linear regression between over wintered Sunn Pest density and yield on Zarrin variety.

The cost of aerial and land chemical controls was 50,000 and 64,000 Rial/ha respectively, and the market value of wheat was 1,050 Rial/kg. The gain thresholds were obtained for aerial and land chemical controls 47.6 and 61 kg/ha, respectively. The EIL was calculated for aerial and land application at14.1 and 18 on Alvand variety and 11.6 and 14.8 over wintered Sunn Pest per m<sup>2</sup> on Zarrin variety, respectively.

 Table 4. Correlation coefficients between log insect density yield loss and EIL in aerial and land chemical controls for Sunn Pest on Alvand and Zarrin varieties.

	EIL on Alv. in aerial control	EIL on Zar. in aerial control	EIL on Alv. in land control	EIL on Zar. in land control	Log of insect No.	Log of yield loss on Zar.
EIL on Zar. in aerial control	0.952**					
EIL on Alv. in land control	1.000**	0.950**				
EIL on Zar. in land control	0.952**	1.000**	0.952**			
Log of insect	0.958**	0.934**	0.958**	0.933**		
Log of yield loss on Zar.	-0.899**	-0.992**	-0.899**	-0.433**	-0.953**	
Log of yield loss on Alv.	-0.990**	-0.833**	-0.990**	-0.833**	-0.994**	0.871**

\*\* Significant at ph 0.01

The results of the quantitative loss assessment by different densities of over wintered Sunn Pest in this experiment were similar to results of Barbulescu (4), using cages (dimension  $1 \text{ m}^2$ ) with Sunn Pest densities 2, 5, 10 and 20. Yield loss and damaged terminal buds in 3 and 5 paired densities under cage conditions were 3.6, 2.65 and 3.3, 2.4, respectively (12).

Dependent factors to density such as yield loss, percentage of yield loss and number of damaged terminal buds and spikes were differed at Alvand and Zarrin varieties. This subject showed that it is necessary to research on quantitative loss assessment in over wintered Sunn Pest at different varieties of wheat. Climatic conditions influence Sunn Pest damage (10, 11, and 13). Developmental stage of wheat affects the rate of Sunn Pest damage (13). Thus, these factors must be considered in investigations on economic levels of Sunn Pest.

As this study was done under cage conditions, we need to calibrate sampling method for adult Sunn pest under actual field conditions to determine the relationship between caged and field studies.

#### Acknowledgement

We thank the head of Plant Pests and Diseases Institute of Iran (PPDRI) and the research assistant director of Tehran University for financial support of this experiment.

### الملخص

نوري، حسين. 2007. تقدير الخسائر والمستويات الاقتصادية لحشرة السونة في محافظة قزوين، إيران. الصفحات 349-344.

تعد حشرة السونة الآفة الأكثر ضرراً للقمح والشعير في معظم أجزاء إيران، حيث تخفض غلة ونوعية المحصول. لتطوير إستراتيجية إدارة متكاملة لحشرة السونة، كان من الضروري قياس مستوى الضرر الاقتصادي (EIL) للآفة. من أجل بالغات السونة المشتية، نفذت تجربة عاملية معتمدة على تصميم القطاعات العشوائية الكاملة تحت ظروف الأقفاص في محطة بحوث فايز –أباد في محافظة قزوين في 1998 و 1999. اختبرت أصناف القمح ألفاند وزارين وكثافات مختلفة من بالغات السونة المشتية (2، 4، 6، 8، 10، 12، 15، 17 و 20 حشرة/م<sup>2</sup> وشاهد خالي من الحشرات). باستخدام معادلة معادلة والاقلام (1989، كان المتوسط المحسوب له 13.61 و 14.01 حشرة سونة مشتية/م<sup>2</sup>.

كلمات مفتاحية: مستوى الضرر الاقتصادي (EIL)، Eurygaster integriceps Puton، إيران، قمح، حشرة السونة.

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## Screening of Wheat Varieties in Nineveh Province, Iraq

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#### Abstract

#### Abdullah, S.I. 2007. Screening of Wheat Varieties in Nineveh Province, Iraq. Pages 357-361.

Screening of wheat varieties for resistance to Sunn Pest (*Eurygaster integriceps* Puton), was done in Alkosh fields, near Mosul, in two seasons 2000/2001 and 2001/2002. A total of five wheat varieties were planted (Intesar, Om-rabee, Makciback, Abu-graib and Karonia) under the natural conditions for screening. All five wheat varieties were infested by Sunn Pest. The most susceptible variety was Intesar, with an average of 4.9 and 6.1 adults/m<sup>2</sup> during the two seasons, respectively, the average of Om-rabee and Makciback were 3.2 and 3.5 in the first season and 3.7 and 4 adults/m<sup>2</sup> in the second season, respectively. Karonia variety was less susceptible in two seasons than the other varieties with an average of 0.6 and 0.5 adults/m<sup>2</sup>. The yield of this variety was more than the other varieties with an average of 5.8 and 6.1 ton/ha, respectively. **Keywords**: *Eurygaster integriceps*, Iraq, screening, Sunn Pest, varieties.

#### Introduction

Sunn Pest is a group of species which has economical importance and is popular (common) in the north of Iraq namely. One species Eurygaster integriceps Puton, (Hemiptera: Scutelleridae) causes great loss in wheat ranging between 75-90%. This pest has a great significance for wheat and barley in the West and Center of Asia (10). This pest has caused major concern for more than 70 years (7). Early studies of the biology and ecology of this pest were conducted in Iran, Syria and Turkey in the early part of the 20<sup>th</sup> century (1, 2). In Turkey, Sunn Pest was known since 1927 (9). There was one generation per year: it attacks the leaves of plants, stems and kernels (12). Early in the growing season adults feed on the stem which reduces sap flow for the outer parts of plants. As a result, the color of stem appears white and dried. Adults and nymphs cause direct damage and they inject chemicals into kernels at the time of feeding. Direct feeding in kernels causes what is called "white head". Flour which is produced from the infected grain is of poor quality (6). The objective of this paper is to make screening of five wheat varieties in order to get more tolerance against this pest.

## **Materials and Methods**

This study has been conducted in two growing seasons 2000/2001 and 2001/2002 in wheat fields in Alkosh which is part of (Nineveh Province) in northern Iraq. The grounds is plowed and made smooth (which is represented by clay soil). It is divided into four equal blocks and split in five experimental units. The unit size is 5 x 5 m using Randomized Complete Block Design (RCBD). The five varieties used were Intesar, Omrabee, Makciback, Abu-graib and Karonia (Check).

These treatments were distributed randomly in the experimental units. Each unit is planted with 100 g seeds in fifty lines: the distance between each line is 25 cm in 15/1/2000 at the first growing season, in the second growing season, in 17/1/2001. A space is left between treatments to avoid any interference. These agricultural operations (fertilization and irrigation) are made on the experimental fields. Some paths are left between blocks, so that the edges are clear to count the Sunn Pest. This experiment encompasses the following:

# Food Preference for the Sunn Pest and the Sensitivity of Wheat Varieties Infected

The first appearance of Sunn Pest by the weekly visits with outset of March is recorded. The plants are examined in the morning to make sure that it is infected by using sweeping net. It is done by having random dual strike. Sweeps were done by swinging in an arc motion with the net held so it contacted the tops of wheat plant, from the beginning of the field till the end of it, and also examined the insects on the soil surface (6). The beginning of the appearance of the adults in the field was in 20/3/2000 in the first growing season and in 16/3/2001 in the second growing season. A wooden frame (1 x 1 m) was constructed and put at random to keep the plants in every experimental unit and on the surface of the soil weekly. This continued for four weeks till the spring migration and the descending of the adults from Alkosh Mountain to the farms in 10/4/2000 in the first growing season and 6/4/2001 in the second.

#### The Effect of the Infection of Sunn Pest on the Productive Features of Wheat During the Harvest Season

One square meter of plants was randomly cut in each experimental unit. This was done during the first week of June in the two seasons. They were tied and put in marking sacks and taken to laboratory. In the laboratory the number of spikes/m was counted, and also counted was the average number of grains in one spike and the thousand kernel weight. Analysis of variance was made for all the studied features with the adults in each square meter, testing LSD was applied between treatments at probability level 5%.

## **Results and Discussion**

The results showed that adults when seen in the early morning, from a distance, they are actively feeding on the spike; adults are very sensitive to any disturbance and drop to the ground and appear dead when they are in danger. This observation is similar to those reported by (3, 6).

Adults were counted as they appear on the plants and drop to the ground in one square meter and is repeated five times in each experimental unit. The results in Table 1 clarify that the Sunn Pest adults appeared in the experimental field on each variety (20/3/2000) in the first growing season, whereas the appearance was preceded one week in the second growing season. The first adult appeared on (16/3/2001) and its appearance was delayed on two varieties (Abu-Graib, and Garuniya) until 27/3/2000 23/3/2001 for the two seasons and respectively. The adults were found in low numbers, with an average of (0.8, 0.4)adults/m<sup>2</sup> on the last variety in each growing season respectively. The adults disappeared in the second week of April in the first growing season and the first week of April in the second growing season and no adults were seen in the field on the varieties (Intesar, Um-Rabee. Mackiback and Abu-Graib) and its existence was not found on 10/4/2001 on the Karonia variety in the first growing season and on 6/4/2002 in the second growing season. A large number of adults were found on the Makciback variety. There were fewer Sunn Pests on the Karonia variety in the two growing seasons at all times of sampling. The maximum appearance of Sunn Pest adults on the varieties tested was in the third period of each growing seasons with an average of (5.12, 5.8) adults/m<sup>2</sup> for each respectively. The variety Intesar showed an attraction of Sunn Pest adults in the two growing seasons with an average of (7.6, 9.0) adults/m<sup>2</sup>, respectively.

The difference in duration of the appearance and disappearance and the maximum appearance of the Sunn Pest adults may be related to the difference of temperature, relative humidity, speed of wind and the amount of rainfall.

The results in Table 2 indicate the significant difference at probability of 5% between varieties in the number of  $adults/m^2$  and some productive features such as: number

spikes/m<sup>2</sup>, number of grains/spike, the 1000 kernel weight and the crop yield. The most infected variety was Intesar in each of the two growing seasons (2000/2001, 2001/2002) with an average of (4.9, 6.15) adults/m<sup>2</sup>, respectively. This was passively reflected in crop yield with an average of (2.58, 2.52)ton/ha respectively. With the increase of population density of adults there was a decrease in the spike number/m<sup>2</sup> with an average of (142, 138) spikes/m<sup>2</sup>, respectively; the 1000 kernel weight with an average of 35, 34.5 g respectively. The number of grains/spike averaged 52, 53, respectively. But the loss of the milky substance is the result of the number and greediness of adults which caused a reduced the 1000 kernel weight and consequently the yield was reduced. On the other hand no significant difference in crop yield appeared between two varieties (Intesar and Om-rabee) with an average of (2.93, 2.94) ton/ha, respectively. But the preference of the pest was reduced in Om-rabee. There the number of adults was lower with an average of (3.2, 3.7) adults/m<sup>2</sup> in each of the two growing seasons (2000/2001, 2001/2002), respectively. This led to an increase of spikes with an average of 180, 173 spikes/m<sup>2</sup>, respectively.

But the number of grains/spike was reduced due to the feeding with an average of 37, 39 grains/spike, respectively; this was reflected passively on the crop yield and caused a reduction. The distinctive feature of Karonia variety was less susceptible to Sunn Pest in two growing seasons with an average of (0.5,0.6) adults/m<sup>2</sup>, respectively. This is positively reflected in the number of spikes and the number of grains/spike: the 1000 kernel weight with an average of 240, 245 spikes/m<sup>2</sup>; 40, 42 and 41 grains/spike and 61, 61.4 g/spike), respectively in each of the two growing seasons (2000/2001, 2001/2002): this goes with the increase of yield with an average of 5.85, 6.31 and 6.08 tons/ha, respectively. The variation of the adult number on testing varieties could belong to the adult preference for this variety and not to the other varieties because of the difference in its content which was in response to their tasting (4, 8) or it will affect the characteristics of plants to the behavior of the insect. This will attract the pest. As Miller et. al (11) and Blum (5) indicated the accumulation of silica in the cells wall and the thickness of walls had its influence and increased plant resistance to avoid the infection by the pests.

**Table 1.** Number of adults/m<sup>2</sup> of Sunn Pest (*Eurygaster integriceps* Put.) on five varieties of wheat during the seasons 2000/2001 and 2001/2002.

Wheat varieties	Duration of sampling/adults/m <sup>2</sup>							
2000/2001 growing season	20/3/2000	27/3/2000	3/4/2000	10/4/2000				
Intesar	3.6 a	6.6 a	7.6 a	1.8 a				
Om-rabee	2.4 b	3.6 bc	4.8 b	2.0 a				
Makciback	1.2 c	4.6 b	6.8 a	1.4 a				
Ab-Garib	0.0 d	2.4 c	5.2 b	0.2 b				
Karonia (Check)	0.0 d	0.8 d	1.2 c	0.0 b				
2001/2002 growing season	16/3/2001	23/3/2001	30/3/2001	7/4/2001				
Intesar	4.0 a	7.2 a	9.0 a	4.4 a				
Om-rabee	2.4 ab	4.0 b	6.0 bc	2.4 b				
Makciback	2.0 bc	5.4 b	7.0 b	2.6 c				
Ab-Garib	0.4 cd	4.2 b	5.0 c	0.2 d				
Karonia (Check)	0.0 d	0.4 c	2.0 d	0.0 d				

Values followed by the same letter (vertically) for each season are not significantly different at P=0.05

Wheat varieties	No. of Adults/m <sup>2</sup>	No. of spikes/m <sup>2</sup>	No. of grains/ spike	1000 KW(g)	GY ton/ha
2000/2001 growing	season				
Intesar	4.9 a	142 e	52 a	35.0 d	2.58 c
Om-rabee	3.2 b	180 d	37 c	44.0 c	2.93 c
Makciback	3.5 b	195 c	35 c	48.0 b	3.27 b
Ab-Graib	2.0 c	230 b	40 b	60.0 a	5.52 a
Karonia (Check)	0.5 d	240 a	40 b	61.0 a	5.85 a
2001/2002 growing	season				
Intesar	6.2 a	138 c	53 a	34.5 d	2.52 c
Om- rabee	3.7 b	173 b	39 c	43.6 c	2.94 c
Makciback	4.3 b	189 b	33 d	46.0 b	2.86 c
Ab-Graib	2.5 c	218 a	41 b	59.7 a	5.33 b
Karonia (Check)	0.6 d	245 a	42 b	61.4 a	6.31 a

**Table 2.** Effect of adult numbers of Sunn Pest (*Eurygaster integriceps* Put.) on the productivity characters of five wheat varieties in 2000/2001 and 2001/2002 seasons.

Values followed by the same letter (vertically) for each season are not significantly different at P=0.05

#### الملخص

عبد الله، سعاد أرديني. 2007. غربلة أصناف القمح في محافظة نينوى، العراق. الصفحات 357-361.

أجريت غربلة لأصناف القمح لاختبار مقاومتها لحشرة السونة (Eurygaster integriceps Puton)، في حقول الكوش، قرب الموصل، في موسمي 2001/2000 و 2002/2001. زرعت خمسة أصناف قمح (انتصار، أم-ربيع، مكسيباك، أبو-غريب وكارونيا) تحت الظروف الطبيعية لأجل الغربلة. أصيبت جميع أصناف القمح الخمسة بحشرة السونة. كان الصنف انتصار أكثر الأصناف حساسية، بمتوسط 4.9 و 6.1 بالغة/م<sup>2</sup> خلال الموسمين، على التوالي، وكان متوسط أم-ربيع و مكسيباك 3.2 و 3.5 في الموسم الأول، و 3.7 و 4.1 بالغات/م<sup>2</sup> في الموسم الثاني، على التوالي. كان الصنف كارونيا أقل حساسية من الأصناف الأخرى في كلا الموسمين بمتوسط 6.0 و 5.0 بالغة/م الأصناف الأخرى بمتوسط 5.8 و 1.6 طن/هكتار، على التوالي.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Evaluation of Wheat and its Wild Relatives for Resistance to Sunn Pest under Artificial Infestation

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#### Abstract

El Bouhssini, M., M. Nachit, J. Valkoun, M. Moussa, H. Ketata, O. Abdallah, M. Abdulhai, B.L. Parker, F. Rihawi, A. Joubi and F. Jaby El-Haramein. 2007. Evaluation of Wheat and its Wild Relatives for Resistance to Sunn Pest under Artificial Infestation. Pages 363-368.

There has been very little progress towards the use of host plant resistance against Sunn Pest worldwide. Besides the narrow genetic base in cultivated wheat for resistance to this pest, there are great difficulties in screening in hot spots in the field because of aerial insecticide sprays. Also, there was no reliable screening technique under artificial infestation. For the last six years ICARDA, in collaboration with its NARS partners in West Asia, has placed special effort on the development of a screening technique under artificial infestation. The method consists of using mesh screen cages (6 x 9 x 3 m) constructed directly in the field. The test is done in two stages, initial and advanced evaluation. In the initial screening wheat entries are planted in hill plots at the usual planting time in the fall. Plants are infested at the time insects migrate to wheat fields using six adults/m<sup>2</sup>. The parameters used in the evaluation for resistance are to rank shoot, leaf damage and plant stunting using two scales from 1-6 based on visual damage and infestation. In the advanced screening, the selected wheat lines from the initial test are planted in rows 2-m long, and one cage with uninfested plants is used as a control. The method of infestation and evaluation is based on the grain quality analysis. Out of 1097 lines/accessions tested using this new screening technique, several lines of wheat and its wild relatives showed good level of vegetative stage resistance. Based on quality analysis, none of the wheat lines tested showed resistance. The sources of resistance at the vegetative stage are being used in the breeding programs to develop wheat resistance varieties for the overwintered Sunn Pest.

Keywords: Sunn Pest, Triticum, Aegilops, plant resistance.

#### Introduction

Sunn Pest (*Eurygaster integriceps* Puton) is the most important insect pest of wheat in West and Central Asia and East Europe. Nymphs and adults cause damage to plants and reduce yields by feeding on leaves, stems and grains. Through their feeding on the grains, the insects inject proteolytic and amylolytic enzymes. The proteolytic enzymes cause an extensive breakdown of the gluten, which greatly reduce the baking quality of the dough. If as little as two to three percent of the grain has been fed upon, the entire grain lot may be rendered unacceptable for baking purposes because the flour is of poor quality (3). The use of insecticides has been the only means of controlling Sunn Pest. This, however, has been quite expensive with an annual cost in West Asia estimated at 40 million \$ US (5). In addition to the high cost of chemical control, insecticides are hazardous to human health, natural enemies and the environment as a whole. The present insecticide-based strategy must be replaced with an integrated pest management approach comprising biological and biotechnical control, host plant resistance and cultural practices. The most important natural enemies of Sunn Pest are entomopathogenic fungi often found in the overwintering sites (4) and egg parasitoids in cereal fields (2, 7).

There has been very little progress towards the use of host plant resistance against Sunn Pest worldwide. Besides the narrow genetic base in the cultivated wheat for resistance to this pest, there are great difficulties in screening in hot spots in the field because of aerial insecticide sprays. Also, there was no reliable screening technique under artificial infestation. The objectives of the present study were to: 1). develop a screening technique for resistance to Sunn Pest under artificial infestation, and 2). identify sources of resistance to Sunn Pest in wheat and its wild relatives.

### Materials and Methods

The screening test was conducted at ICARDA experimental station, Tel Hadya under artificial infestation during 1999-2004. The first three years were devoted mainly to the development and refinement of the screening technique. This method consists of using mesh screen cages of 6 x 9 x 3 m. The test is carried out in two stages, initial and advanced evaluation. In the initial screening test entries are planted in hill plots at the usual planting time in the fall. About 10 seeds of each entry are used for each plot. The design used in this test is an augmented one with two replications. Plants are infested at the time of insects' migration to wheat fields, around mid-March, using six adults/m<sup>2</sup>. This number of Sunn Pest adults represents about 2-3 times the economic threshold for overwintered adults used in the region. Two scales from 1-6 are used, one for visual infestation and one for damage, to assess vegetative stage damage (the% shoot and leaf damage and plant stunting) (Table 1). Entries with a score of 2 or less are classified as resistant to Sunn Pest feeding at the vegetative stage. The total number of accessions/lines screened for Sunn Pest resistance was 282 Aegilops spp., 93 Triticum spp., 288 bread wheat and 434 durum wheat.

**Table 1.** Visual damgge scale to evaluate leaf, shoot and plant stunting caused by Sunn Pest feeding.

Scale	Visual damge on leaf and shoot	Visual damage on plant stunting
1	No damage	No stunting
2	1-5% damage	Very little stunting
3	6-25% damage	Low level of stunting
4	26-50% damage	Moderate level of
	-	stunting
5	51-75% damage	High level of stunting
6	> 75% damage	Severe level of stunting

In the advanced screening, the selected wheat lines from the initial test are planted in rows 2-m long. A separate cage is kept uninfested as control. The design used in this test is a randomized complete bloc with three replications. The method of infestation consisted of adding two adults/m<sup>2</sup> at the time of insect migration to wheat fields, and later the number of nymphs is adjusted to  $8-10/m^2$ .

The evaluation is based on grain quality analysis (8). Three analyses are conducted, the weight of 1000 kernels (TKW), the percentage of kernels with signs of Sunn Pest damage (% DK) and total protein content.

- a) 1000 kernel weight. One hundred kernels were counted from each batch of the three replicates, weighed and mean of 1000 kernel weight calculated.
- b) Percent damaged kernels: When Sunn Pest feeds on wheat it leaves an opaque patch on the kernel with a small dark penetration site. The average number of kernels with this characteristic was determined by examining a random sample of 100 kernels from each replicate.
- c) Protein content: Wheat protein content was determined by NIRSystems model 5000 scanning monochromator instrument. Ten percent of the samples were also analyzed by Kjeldahl method (AACC method No. 46-12) to check NIR accuracy.

The flour quality analysis was based on the SDS (Sodium Dodecyl Sulphate) sedimentation test (1).

## **Results and Discussion**

# Initial Screening for Vegetative Stage Resistance

Resistance reaction of *Aegilops* and *Triticum* species accessions and bread wheat and durum wheat lines are summarized in Tables 2 and 3. It appears from the screening results that only a very limited number of wheat lines and wild relative accessions showed resistance to Sunn

Pest feeding at the vegetative stage. Only 2% of the *Aegilops* and 5% of the *Triticum* accessions tested were resistant. Out of the total number of wheat lines tested, the percentage of resistant lines identified for Sunn Pest was 2% and 3% for bread wheat and durum wheat, respectively. Four out of eight sources of resistance in bread wheat are synthetic wheat.

**Table 2.** List of wheat lines that showed resistance reaction to Sunn Pest feeding at the vegetative stage, under artificial infestation at ICARDA.

Plant species	Accession number/cutlivar	Origin
Triticum durum Desf.		<u>u</u>
	ICDW-12839	Afghanistan
	ICDW-13529	Afghanistan
	DCCIIRIR01-108	Syria
	DCCIIRIR01-623	Portugal
	Sebah	ICARDA
	Sebou	ICARDA
	GRU SP DW 02-991330	ICARDA
	GRU SP DW 02-991340	ICARDA
Triticum aestivum L.		
	Synthetic A02-55	ICARDA/CIMMYT
	Synthetic A02-49	ICARDA/CIMMYT
	Synthetic A02-52	ICARDA/CIMMYT
	Synthetic AYTSYN-19 (PYT-IR-A-03 No.336)	Turkey/CIMMYT/ICARDA
	Synthetic AYTSYN-28 (PYT-IR-A-03 No. 373)	Turkey/CIMMYT/ICARDA
	Synthetic AYTSYN-111 (PYT-IR-B-03 No.4185)	Turkey/CIMMYT/ICARDA
	ICBW-209284	Afghanistan
	ICBW-209270	Afghanistan
	ICBW-209273	Afghanistan
	Falat	Iran

**Table 3.** List of *Aegilops* and *Triticum* species that showed resistance to Sunn Pest feeding at the vegetative stage, under artificial infestation at ICARDA.

Species	Accession number	Origin
Aegilops umbellulata Zhuk.	IG-48404	Syria
Aegilops biuncialis Via.	IG-47891	Turkey
Aegilops triuncialis L.	IG-47893	Turkey
Aegilops speltoides var. ligustica (Savign.) Fiori	IG-46593	Syria
Aegilops speltoides var. ligustica	IG-46814	Turkey
Triticum urartu Thum.	IG-116199	Turkey
Triticum aestivum L. sub. compactum Host.	IG-45481	Iran
Triticum turgidum L. sub. dicoccon Schrank.	IG-45380	Armenia
Triticum monococcum L. sub. boeoticum Bois.	IG-118185	Syria
Triticum timopheevi Zhuk. sub. araraticum Jakubz	IG-45216	Azerbaijan

	1000 Kernel Weight	Damaged kernel	Protein	Sedimentation
Mean of infested lines	37.10	18.20	12.20	12.40
Mean of uninfested lines	38.70	1.10	12.70	65.20
LSD	1.44	6.48	0.26	10.05
CV%	1.10	19.10	0.60	7.40

Table 4. Analysis of variance of grain quality tests for wheat lines selected from initial screening.

**Table 5.** Grain quality analyses for the wheat lines selected from the initial screening.

		1000 kernel weight (g)		Damaged kernel (%)		Protein content (%)		Sediment height (ml)	
Crop/ Entry	Origin	Inf.	Uninf.	Inf.	Uninf.	Inf.	Uninf.	Inf.	Uninf.
Bread wheat									
AWYRGP00-5	ICARDA/ CIMMIT	36.5	36.2	26	2	12.1	12.4	9	76
AWYRG00-16	ICARDA/ CIMMIT	34.0	39.6	20	2	12.0	12.5	9	69
AWYRG00-19	ICARDA/ CIMMIT	35.1	37.2	17	2	13.4	14.2	9	65
AWYRG00-27	ICARDA/ CIMMIT	31.9	34.8	28	2	12.0	12.7	12	56
Synthatic-A02-49	ICARDA/ CIMMIT	38.8	40.5	19	0	12.1	12.5	10	78
Synthatic-A02-52	ICARDA/ CIMMIT	34.1	38.8	11	0	11.8	12.0	25	89
Synthatic-A02-55	ICARDA/ CIMMIT	34.6	41.2	16	0	11.2	12.0	21	84
Tabasi	Iran	44.8	41.7	34	2	12.4	13.0	9	54
Gaphgas	Iran	38.7	41.2	14	1	12.8	13.4	10	68
Falat	Iran	31.6	28.5	13	1	12.6	13.8	13	79
Cham 6	Local Check	33.6	32.3	21	2	11.8	11.6	11	48
Durum wheat									
Sebou	ICARDA/ CIMMIT	47.1	49.8	14	0	11.9	12.3	10	43
Sebah	ICARDA/ CIMMIT	42.1	41.3	7	1	12.3	12.5	14	39

Most of the identified sources of resistance of wheat, *Aegilops* and *Triticum* originated from Sunn Pest prone areas in West and Central Asia. These are the first sources of resistance identified against Sunn Pest feeding at the vegetative stage. Earlier screening studies concentrated efforts mainly on trying to find resistance at the reproductive stage (grain) (6). The reason behind separating the screening test into two steps (initial for vegetative stage damage and advanced for grain damage) is related to the biology of the Sunn Pest itself. The overwintered adults migrate to cereal fields in spring when the average temperature is about 15 C (5), feed on leaves, stems and shoots for almost two months before they die. In the first two weeks after their arrival to cereal fields, they mate and lay eggs. The nymphs and new generation adults feed only on the spikes (grain) (5). It is clear from the biology of the pest that screening needed to be done at the vegetative stage only for the ovewintered adults and at

the reproductive stage (grain quality) for the nymphs and new generation adults.

# Advanced Screening at the Reproductive Stage

For all the lines tested, there is no significant difference in kernel size and protein content between infested and uninfested lines (Table 5). Although the damaged kernel percentages varied from 7 to 34% between infested lines, but the lower damage percentage should not be considered as a criterion for resistance to Sunn Pest since the flour quality of these lines is extremely low. The weakening of gluten strength in all infested lines as measured by sedimentation test clearly indicates that there is no resistance among the material tested to Sunn Pest feeding at the reproductive stage (grain) (Table 4).

## Conclusion

Our results showed that an effective screening methodology for resistance to Sunn Pest has been developed, and is routinely used at ICARDA to identify sources of resistance to this pest. As a result several sources of resistance to Sunn Pest feeding at the vegetative stage have been identified in wheat and its wild relatives. However, none of these sources of resistance provide resistance at the grain level (quality). These sources of resistance are being used in breeding programs to develop resistant varieties against the overwintered Sunn Pest adults feeding, which cause damage to wheat at the vegetative stage.

## الملخص

البوحسيني، مصطفى، ميلودي نشيط، جان فالكون، موسى مسعد، حبيب قطاطا، عثمان عبد الله، محمد عبد الحي، بروس ل. باركر، فوزي ريحاوي، عبد الله جوبي وفوًاد جابي الحرمين. 2007. تقييم مقاومة القمح وأقاربه البرية لحشرة السونة تحت ظروف الإعداء الاصطناعي. الصفحات 363–368.

كان هناك تقدم ضئيل باتجاه استخدام مقاومة العائل النباتي ضد حشرة السونة على مستوى العالم. إضافة إلى القاعدة الورائية الضيقة في القمح المزروع لمقاومة هذه الآفة، هناك صعوبات كبيرة في الغريلة في المناطق الموبوءة في الحقل بسبب الورائية الضيقة في القمح المزروع لمقاومة هذه الآفة، هناك صعوبات كبيرة في الغريلة في المناطق الموبوءة في الحقل بسبب الرش الجوي لمبيدات الحشرات. أيضاً، لم تكن هناك تقنية غربلة موثوقة تحت ظروف الإعداء الإصطناعي. خلال الأعوام الستة الماضية بذلت إيكاردا، بالتعاون مع شركائها أنظمة البحوث الزراعية الوطنية في غرب آسيا، جهوداً خاصة لتطوير تقنية غربلة موثوقة تحت ظروف الإعداء الإصطناعي. خلال الأعوام الستة تحت ظروف الإعداء الإصطناعي. خلال الأعوام الستة متت ظروف الإعداء الإصطناعي. تتألف الطريقة من استخدام أقفاص غربلة غربلة غربولية (3 × 2 × 3 م) مشيدة مباشرة في الحقل. يجرى الاختبار في مرحلتين، تقبيم أولي ومتقدم. في الغربلة الأولية تزرع مدخلات القمح في مجموعات بطريقة عامالة المقل. يوقت الرقاعة العرقة من استخدام أقفاص غربلة غربولية (3 × 2 × 3 م) مشيدة مباشرة في في وقت الزراعة الاعتباري في مرحلتين، تقبيم أولي ومتقدم. في الغربلة الأولية تزرع مدخلات القمح في مجموعات بطريقة عامالة المؤرث في وقت الزراعة الغربلة الأولية تزرع مدخلات القمح في مجموعات بطريقة عاملية في وقت الزراعة الغتيرة في ملحيف نعدى النباتات عند هجرة الحشرات إلى حقول القمح باستخدام ست حشرات بالغة/م<sup>2</sup>. المؤشرات التي تستخدم لتقبيم المقاومة هي لقياس ضرر الورقة العلمية والورقة وتقزم النبات باستخدام متياسين من 1–6 معتمدين ويوقت المرئي والإصابة. في الغربلة المتقدمة، تزرع أصناف القمح المنتخبة من الغربلة الأولية في خطوط بطول 2 م، ويستخدم قفص واحد بنباتات غير معداة كشاهد. تعتمد طريقة العدوى والتقبيم على تحليل نوعية الحديدة ماصل ويستخدم قليول معنوى مالعالي والغولية من المالغا ومية علمين وألولية في ملول بولي أسئون ما معاد كربلة، أظهرت عدة أصناف من الغمر وأقراب البرية، أصل وألي أسلول أصل وألي أصناف مالغربلة، أظهرت عدة أصناف مالم وأولية الميوبي، ماصل وأصل أصناف مالغم وأمناف مالقمح وأماني أولية في خطوط بطول وأصنافي أمداف أصر وألور أملي مالتوى جيدمان ويستخدم قي برماي النولية، أطهرت عدة أصناف من القمح وأقاربه البري مالي وربد، المقاومة. ممادر المغاومة، مالغولي أم

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

## Comparison of Resistance to Sunn Pest in Bread and Durum Wheat

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#### Abstract

Rezabeigi, M. 2007. Comparison of Resistance to Sunn Pest in Bread and Durum Wheat. Pages 369-374.

Twenty one bread and 23 durum wheat lines were compared to study their resistance to the nymphal stages of Sunn Pest under field conditions in Varamin. For artificial infestation of treatments 40 nymphs were released in small cages installed on 40-60 spikes in each replication. Comparing six measured characteristics, there were not significant differences among weight reduction percentage per 50 grains, mortality of nymphs and total yield; but the percentage of damaged grains per nymph and adult and the total percentage of damaged grains in durum wheat lines were lower than those of bread wheat and significant differences existed among them. The durum wheat lines were more resistant than those of bread wheat to nymphal stages and newly emerged adults of Sunn Pest. Therefore we are able to increase the rate of economic injury level (EIL) of nymphs in durum wheat fields.

Keywords: Bread wheat, Eurygaster integriceps Puton, durum wheat, resistance, Sunn Pest.

### Introduction

Resistance in wheat varieties to Sunn Pest (Eurygaster integriceps Puton) has been evaluated by some researchers and resistant varieties introduced by them (2, 4, 5, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18). So far the information about resistance of durum varieties to Sunn Pest is scarce. Mohammadi and Najafi (8) studied resistance to Sunn Pest in 21 advanced lines of durum wheat and the results suggested that durum wheat is more resistant to Sunn Pest than bread wheat. Ghadiri (3) was studied resistance to Sunn pest in advanced durum lines and found some differences among them. For the reason that there was not more information about resistance to Sunn Pest in durum wheat, this investigation was done from 1998-1999 to compare resistance to Sunn Pest among two groups of bread and durum wheat lines.

#### **Material and Methods**

#### **Field Study**

To evaluate the resistance of 21 bread wheat and 23 durum wheat lines to the nymphal stages of Sunn Pest (Eurygaster integriceps Puton), two experiments were carried out in 1999-1998 under the same condition at the Varamin station. We used a randomized complete block design (RCB) with 4 replications for our studies. For artificial infestation of each replication, 40 third and fourth instars of Sunn Pest were released in cages installed on 40-60 wheat spikes (Figures 1 and 2). In both experiments, characters studied included percent mortality of nymphs, percent of grain damaged, percent of grain damaged per nymphs, percent of grain damaged per newly emerged adults, weight reduction percentage in 50 damaged grains and total yield.

#### Data Analysis

Analysis of variance of data was done by MSTAT-C software and means comparison was conducted by Duncan's multiple range tests. Resistance indices of wheat cultivars were calculated by ranking data by normal distribution (1). In this regard, we used indices 2, 4, 6 and 8 for susceptible, semi-susceptible, semi-resistant and resistant cultivars. Finally we used t-test analysis to compare characters studied in two groups of bread and durum wheat lines by MINITAB software.

## Results

# Resistance Indices of Bread Wheat Lines to Sunn Pest

Significant differences existed among bread wheat lines. Table 1 shows grouping of wheat lines based on studied characters. According to calculated resistance indices, Line 6 a check durum line (Chen/Alter-S-72-20) and line 11 (Kal//Bb/Kal/3/Au/50 E/3\* Kal/4/Brochis/..) were ranked as a resistant group and line 8 (Agri/Vkr) and line 7 (Alvand//Aldan''S''/ IAS58, 40072-48) were ranked as susceptible one.

# Resistance Indices of Durum Wheat Lines to Sunn Pest

Based on data analysis, there were significant differences among durum wheat lines (Table 2). Line 16 (Chahba88/..ICD-O664-ABL-OAP-4AP-...) and line 20 (Stork) were grouped as resistant and line 18 (MG-122/T.Aes/Ente/...) and line 22 (Chamran) a bread wheat check line, were ranked as susceptible ones.

Table 1. Indices of resistance of bread wheat lines to Sunn Pest and their yields.

			% of		% of		
		%	damaged	% of	weight		
	% of	of	grains	damaged	reduction in	Yield	Resist-
	nymphs	damaged	per	grains	50 damaged	(ton/	ance
Sources	mortality	grains	nymph	per adult	grains	ha)	Index
Mahdavi	82.8 a	11.0 b-d	0.28 b-d	1.42 a	16.9 b	5.10 c	3.7 c
90, Zhong, 87	52.8 e	12.3 bc	0.31 bc	0.58 b	10.0 с-е	5.87 c	3.7 c
1-23-1317//II-5017//Y50M-	63.9 cd	10.0 b-f	0.25 b-f	0.62 b	8.7 d-f	5.59 c	4.3 c
/3/Cno/Jar/On/							
Bloudan/3/Bb/7C*2//Y50E/	68.3 cd	10.7 b-e	0.27 b-e	0.75 b	0.0 i	5.98 c	5.0 c
Kal*3							
Stm/3/Kal//V534/Jit1716	70.0 b-d	7.3 e-h	0.18 e-h	0.54 b	11.8 b-d	6.01 c	5.3 b
Chen/Alter-S-72-20,	85.0 a	5.0 hi	0.13 hi	0.74 b	14.6 bc	6.74 a	7.0 a
(durum check)							
Alvand//Aldan''S''/IAS58,	62.2 d	17.7 a	0.44 a	1.04 ab	11.5 b-d	6.49 a	3.3 d
40072-48							
Agri/Vkr	67.2 cd	12.6 b	0.32 b	0.85 ab	15.9 b	5.47 c	3.0 d
Qt/ravi66/MX,P,65/3/Rsh/	68.3 cd	10.7 b-e	0.27 b-e	0.75 b	3.2 g	6.01 c	4.7 c
4/Zar75,,1-69-42					e		
GV/D630//Ald"S"/3/Azd	72.8 bc	11.8 bc	0.29 bc	0.96 ab	12.9 b-d	6.98 a	4.7 c
Kal//Bb/Kal/3/Au/50E/3*	82.2 a	5.6 gi	0.14 g-I	0.96 ab	1.4 gh	5.59 c	6.7 a
Kal/4/Brochis/		•					
4778(8)//Fkn/Gb/3/	78.9 ab	9.9 b-f	0.25 b-f	1.04 ab	22.8 a	6.74 a	4.7 c
Vee"s"/4/Buc"S"							
Kayson/Glenson,1-69-48	86.1 a	8.6 c-g	0.22 c-g	1.37 a	1.4 gh	6.04 c	5.7 b
R,16043/4/*Nac	82.8 a	5.8 g-i	0.15 g-I	0.74 b	10.5 cd	5.49 c	6.0 b
Jup/Biy//Kauz,1-73-96	86.7 a	3.5 i	0.09 Ĭ	0.58 b	13.1 b-d	5.90 c	6.3 b
Inia/A.Distichum//Inia/3/	70.6 b-d	8.0 d-g	0.20 d-g	0.60 b	6.3 ef	6.29	5.7 b
Vee''S''/4/Kauz		C	C			b	
Attil50y=,S-72-5	77.8 ab	6.8 f-h	0.17 f-h	0.68 b	6.1 f	7.15 a	6.3 b
Alborzx,K6290914/Cno//	70.6 b-d	9.2 b-f	0.23 b-f	0.69 b	5.9 f	6.15	5.7 b
K58/Tob/3/Wa,						b	
Pfau/Seri//Bow''S''	68.3 cd	8.0 d-g	0.20 d-g	0.56 b	12.6 b-d	6.67 a	6.0 b
A-12-1-32-438//Rsh/Bb/Kal	71.7 bc	9.7 b-f	0.24 b-f	0.76 b	1.7 gh	6.49 a	5.3 b
/Ald''S'',					U		
Mahdavi	65.6 cd	10.7 b-e	0.27 b-e	0.69 b	0.5 h	5.10 c	4.7 c

Same letters are not significantly different (P=0.05, Duncan's multiple range test).

Resistance Index is ranked by normal distribution. a, b, c and d are Resistant, Semi-resistant, Semi-susceptible and Susceptible, respectively.



**Figure 1.** A cage installed on the 40-60 wheat spikes for artificial infestation.



**Figure 2.** Experimental fields and the installed cages for artificial infestation.

Table 2. Indices of resistance of durum wheat lines to Sunn Pest and their yields.

			%	%	%		
			of	of	of weight		
		%	damage	damaged	reduction		
	% of	of	d grains	grains	in 50	Yield	Resist-
	nymphs	damaged	per	per	damaged	(ton/	ance
Sources	mortality	grains	nymph	adult	grains	ha)	Index
Stn/3/Hui/Care/Chen/	71.7 а-е	6.7 b-g	0.17 b-g	0.53 a-e	11.4 bc	5.80 c	5.0 c
Ento/4/Altar84							
Bashen-4	69.4 a-e	8.2 a-c	0.21 a-c	0.59 a-d	2.2 ig	6.11 c	4.3 c
Tarro-3	65.0 de	5.9 d-g	0.15 d-g	0.38 e-f	6.7 e-g	6.35 c	5.7 b
Tarro-3	70.6 a-e	5.1 b-g	0.13 f-g	0.39 d-f	5.7 f-h	6.46 c	6.7 a
Omruf-1	73.9 a-d	5.2 fg	0.13 f-g	0.44 c-f	7.0 d-g	6.35 c	6.0 b
Aco-89	72.8 a-d	7.4 fg	0.19 b-d	0.62 a-c	8.0 c-f	5.80 c	4.0 c
MRB 5895	70.6 a-e	8.8 ab	0.22 ab	0.69 a	1.7 ј	6.74 c	3.7 c
Yazi 38	75.6 a-c	7.7 b-d	0.19 b-d	0.74 a	0.0 k	6.70 c	5.0 c
Garavito-2	67.8 b-e	5.3 e-g	0.13 e-g	0.38 e-f	6.1 fg	6.29 c	6.0 b
Exp-2Pee-91,205-408	69.4 a-e	8.5 a-c	0.21 a-c	0.63 a-c	8.6 c-f	6.25 c	3.7 c
Exp-2Pee-91-138-619	79.4 a	5.4 e-g	0.14 e-g	0.61 a-c	13.5 b	6.25 c	5.0 c
Bronte	62.2 e	4.9 g	0.12 g	0.29 f	8.9 c-f	5.80 c	5.3 b
Co7784/wi22A18	74.4 a-d	7.1 b-e	0.18 b-e	0.63 a-c	0.0 k	5.87 c	5.0 c
Gentil-6	70.0 a-e	8.3 a-c	0.21 a-c	0.62 a-c	3.2 h-j	6.70c	4.7 c
Waha	75.6 a-c	7.6 b-d	0.19 b-d	0.71 a	10.7b-d	6.56 c	4.3 c
Chahba88/ICD-O664-	73.9 a-d	5.1 fg	0.13 f-g	0.42 c-f	8.5 c-f	6.67 c	6.7 a
ABL-OAP-4AP							
A63040/Sentty/Gdiz	73.9 a-d	6.5 c-g	0.16 c-g	0.56 a-e	4.1 g-I	7.33 a	6.3 b
MG-122/T.Aes/Ente/	67.2 с-е	10.1 a	0.25 a	0.75 a	22.9 a	5.83 c	2.3 d
Krf/T.dic-sy-26/2/Oak	75.6 a-c	4.9 g	0.12 g	0.46 b-f	5.7 f-h	6.22 c	6.3 b
Stork	75.6 a-c	3.4 h	0.09 h	0.42 c-f	3.4 h-j	5.63 c	6.7 a
Shwa/Mald//AAZ/3/Syn	70.6 a-e	8.4 a-c	0.21 a-c	0.67 ab	8.5 c-f	6.88 b	4.7 c
Chamran (Bread Wheat)	72.8 a-d	8.8 ab	0.22 ab	0.72 a	18.1 a	5.87 c	3.0 d
Mahdavi (Bread Wheat)	77.8 ab	6.8 b-f	0.17 b-f	0.73 a	10.4b-e	6.01 c	4.7 c

Same letters are not significantly different (P=0.05, Duncan's multiple range test).

Resistance Index is ranked by normal distribution. a, b, c and d are Resistant, Semi-resistant, Semi-susceptible and Susceptible respectively.

#### Comparison Resistance to Sunn Pest Among Bread and Durum Wheat Lines

In the first experiment, durum check line (Chen/Alter-S-72-20) was more resistant than the bread wheat lines and in the second experiment, bread wheat check line (Chamran) was more susceptible than the durum wheat lines and Mahdavi (another bread wheat check line) was semi-susceptible. The results of t-test analysis (Table 3) showed that there were not significant differences among mortality of nymphs, weight reduction percentage per 50 grains, and total yield, but the percentage of damaged grains per nymph and adult and the total percentage of damaged grains in durum wheat lines were lower than those of bread wheat and significant differences existed among them.

Percentage of damaged grains per nymph for bread and durum wheat liens were calculated 0.230 and 0.170 respectively and it was showed that the coefficient of damage by nymphs for bread wheat lines was higher (1.35 times) than the durum ones. On the other hand if the economic injury level (EIL) for bread wheat lines are 7 nymphs per m<sup>2</sup> this rate could be 9.5 nymphs per m<sup>2</sup> for durum wheat lines.

## Discussion

As a result, we found that the durum wheat lines were more resistant than those of bread wheat to the nymphal stages of Sunn Pest and our results confirm the gained data by previous researchers (8), but we need to do a complementary study about their resistance to overwintered adults of Sunn Pest. Tolerance of durum wheat is higher than that of the bread wheat and if the risk of pest population increasing were not high, we could be able to increase the EIL of nymphs (1.35 times) in durum wheat fields. This strategy helps us to reduce the chemical control in low infested area that natural enemies of Sunn Pest are active.

We do not have more information about the resistance mechanisms of durum wheat lines to Sunn Pest. Susidko and Fel'ko (13) found hard wheats were more heavily infested with Sunn Pest than soft wheats (39 as compared with 20%), but we did not found significant correlation between grain hardness and resistance or susceptibility to Sunn Pest in bread wheat lines (10). Mikhailova and Shurovenkov (6) studied resistance of 6 wheat species to E. integriceps and other insect pests. Among wheat species, Triticum monococcum L. and T. dicoccoides Koern. Ex Schweinf were most resistant to E. integriceps and T. persicum (Boiss.) Aitch. & Hemsly was the most susceptible. Resistance was associated with compact ears and large hard glumes and lemmas firmly attached to the carvopsis. characters not very marked in T. aestivum L. and T. durum Desf., which tend to be susceptible.

**Table 3.** The results of t-test analysis of resistance indices and yield in two groups of bread and durum wheat lines.

	Bread wheat lines		Durum wheat lines		Т	Probability
Studied characters	Mean	Sd	Mean	Sd	value	( <b>P</b> )
% of nymphs mortality	73.08	9.01	71.99	4.09	0.51	0.620
% of damaged grains	9.28	3.15	6.79	1.69	3.32	0.003
% of damaged grains per nymph	0.23	0.08	0.17	0.04	3.23	0.003
% of damaged grains per adult	0.81	0.25	0.56	0.14	3.94	0.000
% of weight reduction in 50	8.94	6.23	7.62	5.46	0.75	0.460
damaged grains						
Yield (ton/ha)	6.08	0.56	6.28	0.43	1.35	0.190

Mikhailova and Krasnych (7) found same results about the role of those factors on resistance of wheat to Sunn Pest. In this regard, we found that the spikes of durum wheat lines are more compact than those of the bread wheat lines, but in our previous studies compactness of spikelets had not relationship with resistance of bread wheat cultivars (9, 10). So we need to do complementary studies on the role of grain hardness and spike compactness on resistance of wheat varieties, especially in durum ones. Furthermore, comparative studies on grain structure and its components in bread and durum wheat lines are necessary.

#### Acknowledgments

We thank the Scientific Researches Council of Iran for its financial support to this project. So we thank A. Abdi, P. Zamani, and A. Matin for collaborations in field studies.

## الملخص

ريذابيجي، مانوشهر. 2007. مقارنة المقاومة لحشرة السونة في القمح الطري والقاسي. الصفحات 369–374.

تمت مقارنة 21 صنف من القمح الطري و 23 صنف من القمح القاسي لدراسة مقاومتها للأعمار الحورية لحشرة السونة تحت ظروف الحقل في فارامين. من أجل الإعداء الاصطناعي للمعاملات أطلقت 40 حورية في أقفاص صغيرة نصبت على 60-40 سنبلة في كل مكرر. بمقارنة ست صفات مدروسة، لم تكن هناك فروق معنوية بين النسبة المئوية لتخفيض وزن 50 حبة، نسبة موت الحوريات والغلة الكلية؛ لكن النسبة المئوية للحبوب المتضررة بحورية وبالغة والنسبة المئوية الكلية للحبوب المتضررة في أصناف القمح القاسي كانت أقل منها في أصناف القمح الطري ووجدت فروق معنوية بينها. كانت أصناف القمح القاسي أكثر مقاومة للأعمار الحورية ولبالغات حشرة السونة المنبثقة حديثاً من أصناف القمح الطري. لذلك يمكن زيادة درجة مستوى الضرر الاقتصادي (EIL) للحوريات في حقول القمح القاسي.

كلمات مفتاحية: قمح طري، Eurygaster integriceps Puton، قمح قاسي، مقاومة، حشرة السونة.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# Host-Plant Preference of Developmental Stages of Sunn Pest in Wheat Cultivars

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#### Abstract

# Arzani, A., A. Mohammadi and B. Hatami. 2007. Host-Plant Preference of Developmental Stages of Sunn Pest in Wheat Cultivars. Pages 375-380.

Sunn Pest, *Eurygaster integriceps* Puton (Hemiptera; Scutelleridae), is one of the most destructive insect pests of wheat in Iran. Breeding of resistant cultivars is the most economical and environmentally safe method to eliminate the use of insecticides and to reduce crop losses due to this pest. This study was conducted to evaluate the host preference and performance of Sunn Pest by different developmental stages including adults, 4th and 5th instars and newly emerged adults using nine wheat cultivars. Four durum wheat cultivars comprising 'Shadas', 'Altar-84', 'Dipper-6' and 'Massara-1' and five bread wheat cultivars comprising 'Moghan', 'Tabasi', 'Niknejad', 'Anza' and 'Tajan' were grown in Abarkoh, Yazd, a cntral part of Iran that is a highly infested area. A randomized complete block design with four replications in a fan layout was used. Cultivars had different reactions to the different developmental stages of Sunn Pest. Overwintered adults showed the lowest and the highest preference to the 'Tabasi' and 'Niknejad bread wheat cultivars, respectively. Fourth and 5th instars and the newly emerged adults had the highest preference to 'Tabasi' and the lowest to 'Tajan'.

Keywords: Eurygaster integriceps, preference, Sunn Pest, wheat.

## Introduction

Insect pests are the major biotic constraints in crop production. Sunn Pest [Eurygaster] integriceps Puton (Hemiptera, Scutelleridae)] is the most serious insect pest of wheat and other small grains in Iran, Iraq, Turkey, Syria, Lebanon and much of former USSR (1, 4, 10, 11). It is also found in many countries in eastern Europe and western Asia (13). In Iran, the infested area of about 1 million ha is treated annually with pesticides. Both nymphs and adults of the Sunn Pest cause damage and reduce yield and quality by feeding on leaves, stems and grain. It also greatly reduces bread baking quality due to grain shriveling and injection of a salivary secretion containing proteolytic enzymes that destroys the gluten in the grain (8). The use of resistant cultivars is an efficient, economical, and environmentaly safe method to protect plants from insect pests while minimizing the use of insecticides.

Biological control strategies have been neglected in favor of chemical methods due to relative ease, obvious and immediate effects and general availability of a wide range of pesticides. However, it is now generally recognized that the sole use of pesticides for controlling pests should be avoided as much as possible in favor of a more integrated pest management (IPM) approach. Breeding for host plant resistance has been one of the most important strategies in the effort to reduce losses by the Sunn Pest. Skachkova and Kolyabina (17) have tested over 800 hexaploid and octaploid triticales under artificial field conditions and shown that triticale is more resistant than wheat to Sunn Pest attack. Growing winter wheat varieties that are resistant to E. integriceps on the basis of the energy that needs to be expanded by the pest to utilize their available protein is regarded as an efficient measure (15, 16). The response of some special bread wheat cultivars to the Sunn

Pest was evaluated in Iran (7) and Turkey (9). They observed a variable response of wheat genotypes to Sunn Pest. Michailova and Krasnych (12) considered the glume hardiness as a factor affecting relative resistance to Sunn Pest in wheat. Arzani et al. (2) evaluated the resistance of bread, durum and Aegilops germplasm to Sunn Pest and observed a significant variation among genotypes for their response to Sunn Pest. Quantitative losses are generally less important than qualitative ones where low infestations may not be serious enough to reduce yield significantly. It has been shown that only as little as 2-3 % of the grain needs to be damaged for it to be rendered unacceptable for baking purposes (6).

The preference of larvae and newly emerged adults of the Sunn Pest to different winter wheat cultivars has been investigated (5, 15). They observed the highest preference for 'Bezostaia' and the lowest preference for 'Michorinica' and 'Ocraeinica' cultivars. Although much research has been done on this pest over the past 75 y., more effort still needs to be expanded to put together the best options for IPM. Our previous study indicated that *E. integriceps* has a preference for feeding on some wheat genotypes and in particular a native bread wheat cultivar ('Roshan'). The purpose of this research was to evaluate the host preference of Sunn Pest at different developmental stages including adults, 4<sup>th</sup> and 5<sup>th</sup> instars and newly emerged adults using nine wheat cultivars.

#### **Materials and Methods**

#### **Experimental Site**

Abarkoh, located in Yazd Province, is a highly infested area within central Iran and was used as the experimental site. The study was conducted in 1999-00 growing season at  $31^{\circ}$  08' N and  $53^{\circ}$  17' E, 1510 m asl.

#### **Plant Materials**

Four durum wheat cultivars comprising 'Shadas', 'Altar-84', 'Dipper-6' and 'Massara-1' and five bread wheat cultivars comprising

'Moghan', 'Tabasi', 'Niknejad', 'Anza' and 'Tajan' were used in this study.

#### **Field Experimental Evaluation**

Plant materials were grown in the field using a randomized complete block design with four replications in a fan layout. Each circle block was 4 m dia. and contained 9 plots which were allocated to the nine genotypes used. Between each pie a 30 cm row was used as a buffer area. Four hundred seeds were planted for each genotype in each of the experimental plots. In May when overwintered adults arrived, nylon gauze cages  $(4 \times 4 \times 1.8 \text{ m})$  were set up over each block. Then overwintered female adults were collected, starved for 24 h and then placed in the center of each circle blocks. The number of adult Sunn Pest in each plot was counted after 24 and 48 h. When 4<sup>th</sup> instars were present in the fields of the susceptible cultivar "Roshan" they were collected and a sample of 120 were placed in the center of each circle block. The number of 4<sup>th</sup> instars in each plot was counted after 24 and 48 h. A similar procedure was carried out to assess the preference of 5<sup>th</sup> instars and newly emerged adults on wheat genotypes.

#### Statistical Analysis

The data obtained were based on the number of Sunn Pest per individual plant and transformed by  $(X + 1/2)^{-2}$  due to their distribution between 0 to 10 (18). Analysis of variance and mean comparison using protected least significant differences (LSD) were conducted by SAS (14).

## **Results and Discussion**

Results of analysis of variance indicated a significant difference (P<0.01) between wheat genotypes for their preference for attracting Sunn Pest at the four examined developmental stages. Sunn Pest at the developmental stages studied showed different preference for feeding on the wheat genotypes.

Overwintered adults had the highest preference for 'Niknejad' bread wheat cultivar

and the lowest preference for 'Moghan-1' and 'Tabasi' bread wheat cultivars (Figure 1). Four durum wheat genotypes revealed a medium type of preference when compared to the bread wheat genotypes.



**Figure 1.** Mean comparison of the preference of overwintered Sunn Pest on nine wheat genotypes using LSD at 5%. Wheat cultivars are: 1= Moghan-1, 2= Tabasi, 3= Anza, 4= Altar-84, 5= Dipper-6, 6= Massara-1, 7= Shahdas, 8= Tajan, 9= Niknejad.

Figure 2 shows mean comparison of the genotypes for their preference to 4<sup>th</sup> instars of Sunn Pest. 'Tajan' and 'Tabasi' bread wheat cultivars were the most and the least attractive genotypes for the 4<sup>th</sup> instars, respectively. 'Anza', 'Altar-84', 'Niknejad', 'Dipper-6', 'Moghan-1', 'Massara-1' and 'Shahdas' had a moderate preference for 4<sup>th</sup> instars.

Mean comparison for the preference of wheat genotypes for the 5<sup>th</sup> instars revealed the greatest preference for 'Tabasi' cultivar and the lowest preference of 'Tajan' cultivar (Figure 3).

Bread and durum wheat cultivars also significantly varied for their attracted number of newly emerged Sunn Pest under field conditions. The lowest preference of newly emerged Sunn Pest allocated to the 'Tajan', 'Altar-84', 'Niknejad', 'Dipper-6', 'Moghan-1', 'Massara-1', 'Shahdas' and 'Tabasi', respectively (Figure 4).



**Figure 2.** Mean comparison of the preference of 4<sup>th</sup> instars of Sunn Pest on nine wheat genotypes using LSD at 5%. Wheat cultivars are: 1= Tajan, 2= Anza, 3= Altar-84, 4= Niknejad, 5= Dipper-6, 6= Moghan-1, 7= Massara-1, 8= Shahdas, 9= Tabasi.



**Figure 3.** Mean comparison of the preference of  $5^{th}$  instars of Sunn Pest on nine wheat genotypes using LSD at 5%. Wheat cultivars are: 1= Tajan, 2= Anza, 3= Altar-84, 4= Niknejad, 5= Dipper-6, 6= Moghan-1, 7= Massara-1, 8= Shahdas, 9= Tabasi.

Overall results from the evaluation of the preference of Sunn Pest at the three developmental stages revealed that 'Tabasi' cultivar was a superior genotype at overwintered developmental stage while it was inferior at the 4<sup>th</sup> and 5<sup>th</sup> instars and newly emerged adult stages. The different feeding behavior of Sunn Pest at the developmental

stages is based upon the interaction of the insect's physiological and metabolic states with the host and environmental conditions (3). Antixenosis is a host plant mechanism which includes morphological, physical or structural qualities that interfere with insect behavior such as mating, oviposition, feeding and feeding ingestion. As a resistance mechanism, antixenosis acts as structure barriers which affect the insect behavior in selecting their hosts.



**Figure 4.** Mean comparison of the preference of newly emerged adults of Sunn Pest on nine wheat cultivars using LSD at 5%. Wheat cultivars are: 1= Tajan, 2= Anza, 3= Altar-84, 4= Niknejad, 5= Dipper-6, 6= Moghan-1, 7= Massara-1, 8= Shahdas, 9= Tabasi.

According to Figures 1 to 4, feeding behavior of Sunn Pest at the three important developmental stages (4th and 5th instars and newly emerged adults) indicated the order of low preferable to high preferable genotypes as 'Tabasi', 'Shahdas', 'Massara-1', 'Moghan-1', 'Dipper-6', 'Niknejad', 'Altar-84', 'Anza' and 'Tajan' genotypes. This result is in agreement with that of Kinaci et al. (9) who reported the preference of Sunn Pest for particular wheat genotypes under natural conditions. The preference of nymphs and newly emerged adults of the Sunn Pest to different winter wheat cultivars was investigated by Shapiro They observed the and Bartoshko (15). highest preference for 'Bezostaia' and the lowest preference for 'Michorinica' and 'Ocraeinica' cultivars.

Damage of 4<sup>th</sup> and 5<sup>th</sup> instars and newly emerged adults to grain can range from complete destruction to slightly shriveled. However, the toxic saliva injected is important and destroys the gluten of the grain so that the quality reduces dramatically. baking According to our results, cultivation of 'Tajan' cultivar having the least preference for the three latest developmental stages of Sunn Pest is suggested. Furthermore, plantation of a cultivar with highly preference to Sunn Pest ('Roshan'/ 'Tabasi') as marginal or buffering rows where they should be considered as the trape rows for the pests and could be treated with pesticide, may be suggested.

#### الملخص

أرزاني، أ.، أ. محمدي و ب. هاتامي. 2007. تفضيل العائل النباتي من قبل أطوار حشرة السونة في الأقماح المزروعة. الصفحات 375–380.

تعد حشرة السونة (Eurygaster integriceps Puton) من أهم الآفات الحشرية الضارة بمحصول القمح في إيران. تعتبر تربية الأصناف المقاومة الطريقة الأكثر اقتصادية وأماناً للبيئة للتخلص من استخدام المبيدات الحشرية وتخفيض خسائر الغلة العائدة لهذه الآفة. أجريت هذه الدراسة لتقييم تقضيل العائل وأداء حشرة السونة بأطوارها المختلفة التي تتضمن الحشرات البالغة، عمري الحورية الرابع والخامس والحشرات البالغة المنبثقة حديثاً باستخدام تسعة أصناف من القمح المروع. زرعت أربعة أصناف من القمح القاسي (Massara-1 و Dipper-6 ، Altar-84 ، Shadas) وخمسة أصناف من القمح الطري (Moghan) Anza ، Niknejad و Tajan و Tajan) في Yazd ، Abarkoh ، الجزء الوسطي من إيران كمنطقة مصابة بشدة. استخدم تصميم القطاعات العشوائية الكاملة بنموذج مروحي ويأربعة مكررات. أبدت الأصناف ردود فعل مختلفة للأطوار المختلفة لحشرة السونة. أظهرت البالغات المشتية أقل وأعلى تفضيل لصنفي القمح الطري Tabasi و Niknejad على التوالي. أبدت أعمار الحورية الرابع والخامس والبالغات المنبثقة حديثاً أعلى تفضيل للصنف Tabasi وأقل تفضيل للصنف Tajan.

كلمات مفتاحية: Eurygaster integriceps، تفضيل، حشرة السونة، قمح.

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# Development of an Efficient Wheat Genetic Transformation System for the Creation of Transgenic Wheat Resistant to Sunn Pest

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#### Abstract

Fadeev, V.S. and A.K. Gaponenko. 2007. Development of an Efficient Wheat Genetic Transformation System for the Creation of Transgenic Wheat Resistant to Sunn Pest. Pages 381-389.

Sources of resistance to the Sunn bug (Eurygaster integriceps Puton) have not been found in the Triticum genera. It may not be possible to develop wheat resistance to Sunn Pest through common plant breeding procedures. Recent achievements of genetic engineering have shown that usage of transgenic insecticide crop varieties are the environmentally safest way of controlling herbivorous pests. This approach allows the use of a wide range of genes, isolated from different plant species and microorganisms. These genes have to possess a few properties: their products must be harmless to mammals do not affect bread making properties and be toxic to the different development stages of the pest. The creation of a transgenic plant requires availability of three basic components: insecticide genes, an efficient genetic transformation system and the ability to achieve a high level of transgenic expression in specific tissues. We are planning to use four groups of genes: 1) genes encoding the plant protease inhibitors and  $\alpha$ -amylase inhibitors; 2) plant lectins genes [garlic (Allium sativum L.) leaf lectin (ASAL) and snow drop (Galanthus nivalis agglutinin) GNA gene lectin]; 3) genes from the bacterium Bacillus thuringiensis - Bt-genes, which encode insecticide proteins, known as delta-endotoxins; 4) maybe synthetic genes. For this we have used a psGFPBAR vector specially designed for biolistic transformation of cereal. Choice of the selection mode of the transgenic wheat plants, was made on the basis of the bar gene action. The established wheat transformation protocols for Russian wheat varieties has an efficiency of  $\sim 2-3\%$  and allow us to start engineering wheat resistant to the main domestic wheat pest - E. integriceps.

Keywords: Genetic transformation, Sunn Pest resistance, wheat.

#### Introduction

Gene transfer techniques have been developed previously for certain wheat (*Triticum aestivum* L.) cultivars (10, 11, 2, 5) and (*Triticum durum* Desf.) (7), but problems persist in Russian lines because of low transformation rates and in vitro callus cultures. We have adopted our earlier developed method of wheat regeneration from the callus, obtained from immature embryos (4) for the Lada, Mis and others Russian varieties, that are grown in the dry and hot areas of the Russian Federation, where there are major problems with Sunn Pest and could get up to 90% plant regeneration frequency *in vitro* for these varieties. We have optimized

different bombardment parameters of Particle Inflow Gun (3) for the wheat biolistic transformation procedure such as: the pressure used, distance from the source of micro projectiles, gas volume used and types of particles. Transformation of wheat (Triticum aestivum L.) via particle bombardment of immature embryo-derived material has become standard practice in many laboratories at a reported frequency of 1-5% of bombarded embryos yielding transformed plants. Due to the independent site of transgenes integration in the genome of the transformed plant, different genetic processes are taking place during and after transformation: insertion mutagenesis, gene silencing and different levels of transgenic expression. For selection
of the proper transgenic event and the elimination of the influence of these undesirable genetic processes in genetically modified plants, breeders have to have a large sample of transgenic plants obtained from independent transgenic events. For the creation of a large amount of independent transgenic events it is necessary to have an efficient transformation system that is, based on the selection, in vitro, of high-morphogenic callus lines. For this purpose a large amount of material must be carried through the regeneration and transformation systems. These processes are time-consuming and involve significant manual labor in culturing embryos, bombarding embrygenic calli, transferring callus selection to and regeneration media and then growing putatively transformed shoots to maturity and finally confirming transgenic status of obtained plants. Final frequency of transformation will be the product of the multiplication of frequencies: 1). morphogenic callus induction; 2). gene delivery to the morphogenic competent for the regeneration of shoot cells; 3). integration of the foreign DNA into the genome of the recipient cell; 4). selection of transgenic cells; 5). plant regeneration in vitro: 6), seedling rooting: and 7). plantlet adaptation to the green house condition.

The optimization of the processes mentioned above, most of which belong to the tissue culture phase of transformation important to procedure, is very the improvement of the transformation system efficiency. Another important stage of improving the biolistic-mediated transformation system is optimization of parameters affecting transformation frequency: distance from the source of DNA microcarriers to the target cells, pressure used for micro carrier acceleration, tissue preculture treatments and the mode of selection systems. These parameters would be more easily and effectively optimized if all stage of the transformation process could be visually controlled.

For this goal screenable or marker genes are used. The most often used are the bacterial gene of  $\beta$  -glucuronidase - *uidA* (GUS) (6) and the GFP gene from the jellyfish Aequorea victoria (Murbach & Shearer) (1). The green fluorescent protein has the advantage over other reporter proteins in that the formation of a fluorescent chromophore is self-catalyzed and requires only excitation under ultra-violet or blue light to emit a bright green fluorescence, which can be observed with a fluorescent microscope equipped with the appropriate filters in vivo. Usage of GFP as the marker gene allows determination of the transformed shoots soon after bombardment and saves transgenic tissue for further plant regeneration and usage.

Selectable bar and pat genes have been used successfully in transferring herbicide resistance to phosphinothricin (PPT) in a number of crops. In wheat the bar gene has been used successfully as a selectable marker. Gene *bar* (bialaphos resistance) acetylates the free NH<sub>2</sub> group of phosphinothricin and thus inactivates it from binding to glutamine synthethase. Optimal selection systems may improve transformation frequency also and help to avoid so called "escapes". Following we have summarized and presented the results of two experiments of establishing wheat transformation systems and in the obtaining transgenic wheat with GST, Gly I and Gly II genes, which are involved in the determination of resistance to drought and salinity.

#### Material and methods

Donor plants were grown in greenhouse conditions 16/8 day/night, 25/15°C. Caryopsis were harvested from the main spikes 10-12 d after pollination, surface-sterilized for 7 min. in 70% ethanol and washed three times in sterile distiller water. Immature embryos, 0.5-1.5 mm diam. (Figure 2-A) were aseptically excised from caryopses and placed with the scutelum upwards on a solid medium in sterile petri dishes for 14 d. at 25+/- 1°C, in darkness.

Name of media	Macro salts	Micro salts	Vitamins	Type & concentr phytohormone	ation of	Sucrose (g/l)
CIM-Callus Induction Media	MS	MS	5	2,4-D - 2 mg/l		30
SIM-Shoot Induction Media	MS	MS	5	Kinetin-1 mg/l, NAA-0,5 mg/l	-1 mg/l,	30
RIM-Root Induction Media	½ MS	MS	1/2 5	NAA-0,5 mg/l		15
HOPM- High Osmotic Pressure Medium	MS	MS	5	2,4-D - 2 mg/l		140

 Table 1. Composition and name of media used for culturing somatic cell callus, plant regeneration, and rooting and pre-and post bombardment treatment.

Composition and name of media used for culturing somatic cell callus, plant regeneration, rooting and pre and post bombardment treatment, see Table 1. The pH of each medium was adjusted to 5.8 before autoclaving.

Wheat Shoot Regeneration *in vitro*, was induced by transfer morphogenic calli (Figure 2-B) on the culture media without 2.4 D and containing: 1 mg/l Kinetin, 1 mg/l and 0.5 mg/l NAA. For root induction, shoots have been transferred to the Root Induction Media (RIM), which has ½ the basal MS salts, B5 vitamins, and 30 g/l sucrose and 0.5 mg/l NAA (Figure 2-C). After 4 wks, plantlets with well developed roots were transferred form culture to perlite in a mist chamber for a one-two wks, than put in pots with soil in the grow chamber and finally transferred to soil in standard greenhouse conditions.

#### **Plasmid Used**

The plasmid psGFPBAR used in this experiment had a gene *gfp* driven by the rice actin (Act1) promoter and gene bar driven by the maize ubiquitin (Ubi1) promoter (Figure 2-E). To enhance both genes expression, introns were used (10). The selectable bar gene of Streptomyces hygroscopicus (Jensen) Waksman and Henrici encodes bialophos resistance protein (BAR), which inactivates phosphinothricin (PPT), the active component bialaphos. Plasmids of psGSTBAR, psGlyIBAR, psGlyIIBAR, have been made on the base psGFPBAR plasmid where GFP gene was changed by GST, Gly I and Gly II genes. These genes are involved in encoding drought and salinity résistance.

Transformation procedure. Thirty-fifty pieces of 2x2 mm. 14-d old calli, initiated from immature embryos were placed as the circle (or ring, for avoid wounding effect in the center of circle by high pressure of the shot) leaving an empty space 2-3 cm dia in the center of 90-mm-dia Petri dishes and precultured on High Osmotic Pressure (HOPM) Media, containing 2 mg/l 2,4-D, and 0.4 M sucrose as osmotic agents for 4-6 h prior to bombardment. A particle inflow gun (3) was used for delivery of M10 type tungsten microprojectiles, coated with psGFPBAR plasmid. DNA precipitation on the particles was made by using calcium/spermidine methods. The tissues were placed 12 cm from the syringe filter with 2-5 µl of the particle suspension placed on the center of filter holder, and a baffle (500-mm mesh nylon screen) was positioned 9 cm above the tissue for proper deflection of the particles (Figure 2-D). A vacuum pressure of 30 mm Hg was drawn in the chamber, and a helium pressure of six atmospheres, which when opened for 50 ms, was used to propel the particles. After bombardment, the tissues were maintained on HOPM during 24 h. Then calli were transferred to MS medium with 2 mg/l 2,4-D and 1mg/l phosphinotricin (PPT) or on calli induction medium for one week. Then bombardment tissue was transferred to the MS medium with 3 or 5 mg/l PPT and held in the dark for 2 weeks. Then the calli were transferred to MS medium with 5 mg/l PPT for

an additional 2 weeks. The cultures selected were held in the dark at the 25+/-1°C. At the regenerating and rooting stages tissue cultures were transferred under light. Surviving plants with well-developed roots were grown in a greenhouse until full maturity was reached.

For a general scheme of wheat *in vitro* regeneration and wheat *in vitro* biolistic genetic transformation see Figure 1.

#### **Analyses of GFP Gene Expression**

Evaluation of the GFP expression (as transient and after stable gene integration) was made with a stereo dissection microscope equipped with a fluorescence module consisting of a 100-w mercury lamp and GFP excitation and emission filters (Leica, Heerbrugg, Switzeland). Evaluation of expression was made on the 2<sup>nd</sup>, 7<sup>th</sup> and 35<sup>th</sup> d after biollistic transformation (Figures 2-B, 2-F, 2-G, 2-I).

#### PCR Analyses

For PCR analyses DNA of putative transformed plants was isolated from young leaves. (Figure 2-J).

# **Results and Discussion**

We obtained a high level of morphorgenic calli induction (44.9-95.1%), shoots

regeneration up to 88.7%, rooting and regeneration of whole fertile plants for all used genotypes (87.2% for the Lada variety) (Table 2 and Table 3). On the basis of these results we compared two different ways of in vitro selection of the transgenic plants: gradual selections (2) and delayed selections (9) (Table 4). We concluded that the gradual way of selection of transgenic plants decreased the number of escapes that came through the selection process (Table 5). For example, in the 7 putative transgenic plants of Lada variety, obtained by delay selections only 3 of them were real transformed plants. For Mis varieties delay selections gave 1 putative transformed plant, but it was an escape (Table 5, and Figure 2-B). This conclusion helped us in the second round of experiments to obtain ~ 100 putative transformed plants with the GST gene (Table 6) and to reach the level of wheat transformation of ~ 2-3%. This allowed us to obtain a hundred real transgenic wheat plants per person per season and subsequently to start work on the creation of transgenic wheat resistant to the Sunn Pest (Figure 2-K).

Table 2. Genotype effect on the frequency of morphohenic calli induction of bread wheat (*Triticum aestivum* L.)

Variety	No. of immature embryo used	No. of calli obtained	No. of morphohenic calli obtained	Frequency of morphohenic calli induction,%
Lada	800	628	597	95.1
Ester	300	213	170	79.8
Mis	380	192	86	44.8
Norris	900	714	525	73.5
Ampir	1100	820	473	57.7

Table 3. Frequency of shoots regeneration and rooting.

Variety	No. of morphohenic calli used	No. of calli, produced shoots (%)	No. of shoots rooted, (%)	No. of fertile plants obtained, (%)
Lada	156	138 (88.7)	136 (87.2)	136 (87.2)



To maturity plant growing in greenhouse condition

**Figure 1.** General scheme of wheat *in vitro* regeneration and wheat *in vitro* biolistic genetic transformation. **CIM**-Callus Induction Media, **SIM**-Shoot Induction Media, **RIM**-Root Induction Media, **HOPM**-High Osmotic Pressure Medium, **S**-selection, **MC**-morphogenic callus, **NMC**-nonemorphogenic callus, **PPT**-phosphinotricin (selection agent).



**Figure 2.** Wheat regeneration *in vitro* and wheat biolistic transformation. (A) Immature embryo on the third day in culture; (B) Morphogenic calli; (C) Shoot and root regeneration; (D) Calli, prepared for the transformation; (E) psGFPBAR; (F) Transient expression of the GFP genes: A, B - 1 d. after bombardment. C - 2 d. after bombardment. D - Control (morphogenic callus without bombardments); (G) Part of the transgenic wheat shoot. *GFP* gene expression observed after 35 d. of bombardment; (H) Schema of the Particle Inflow Gun; (I) Root of transgenic wheat, showing high level of *GFP* gene expression, observed after 35 d of bombardment; (J) PCR analysis of putative transgenic plants with *bar* and *nos 3* praimer; (K) Mature transgenic wheat (*Triticum aestivum* L.) plants growing in greenhouse condition.

Mode of selection	Callus induction	Plantlets regeneration	Rooting
Gradually selections <sup>1</sup>	*S1-1 mg/l S2-3 mg/l S3-5 mg/l	5 mg/l	5 mg/l
Delay selections <sup>2</sup>	After 7 days of callus induction Without selection *S1-5 mg/l S2-5 mg/l	5 mg/l	3 mg/l

**Table 4.** Comparison of the modes of selection on the (
 ).

\*S1, S2, S3 - Round of selection.

Table 5. Transgenic wheat plants, expressing GFP and BAR.

			Days after b	ombardm	ent	After coloci	ion and creation	PCR analyses of obtained		
			7		35	Anter select	seeds	plants has shown		
Variety	No. of callus used	No. of <i>GFP</i> + calli	Frequency of <i>GFP</i> + Calli (%)	No. of <i>GFP</i> + calli	Frequency of <i>GFP</i> + Calli (%)	No. of putative transgenic plants	Preliminary calculated transfor- mation Frequency (%)	No. of independent obtained transgenic plants	Real Transfor- mation frequency (%)	
Lada test 1**	466	186	40	51	11	4	0.85	4	0.85*	
Lada test 2***	446	214	48	57	13	7	1.57	3	0.65	
Mis test 1**	106	34	32	9	8	2	1.89	2	1.89*	
Mis test 2***	104	44	42	10	10	1	0.96	-	0	

\*\*test 1 - Gradually selections

\*\*\*test 2 - Delay selections

test 2 Benay selections

**Table 6.** Obtaining transgenic plans with the *GST* gene (in progress).

Variety	Lada
Number of immature embryo used	2680
Number of calli used for biolistic transformation	2348
Selection 1	2348
Selection 2	912
Selection 3	420
Plantlets regeneration	306
Putative transgenic plantlets rooted	91
Preliminary estimated the Frequency of transformation	3.39%

#### Acknowledgments

We acknowledge Prof. B.V. Conger and Prof. P. Quail, Department of Plant and Soil Science, The University of Tennessee, who kindly provided the enhanced psGFPBAR expression vector for cereal transformation. Thanks to Prof. John Finer, The Ohio State University, for the generous gift of the Particle Inflow Gun. We acknowledge Prof. Sudhir Sopory (IGGEB, New-Delhi, India) for kindly providing *GlyI*, *Gly II* and *GST* genes. We acknowledge Dr. Boris Kuznetchov and Natalya Slobodova, College of the Centre "Bioengineering" RAS, for PCR analyses of the transgenic wheat plants and our postgraduate student Yaroslav Neskorodov for help preparing pictures *via* the computer.

This work was supported in part by a grant from the Ministry of Science and Technology of Russian Federation.

#### الملخص

فادييف، ف. س.، و أ. ك. غابونينكو. 2007. تطوير نظام نقل جينات فعال للقمح لاستنباط قمح محور وراثياً مقاوم لحشرة السونية. الصفحات 381–389.

لا توجد مصادر مقاومة لبقة السونة (Eurygaster integriceps Puton) في الجنس Triticum. قد لا يكون من الممكن تطوير مقاومة القمح لحشرة السونة عبر إجراءات تربية النبات الشائعة. أظهرت الانجازات الحديثة للهندسة الوراثية أن استخدام أصناف محصول محورة وراثياً كمبيدات حشرات هي الطريقة الآمنة بيئياً لمكافحة الآفات الضارة بالنباتات. تسمح هذه الطريقة باستخدام مدى واسع من الجينات، معزولة من أنواع نباتية وكائنات حية دقيقة مختلفة. يجب أن تمتلك هذه الجينات بعض الخصائص: يجب أن تكون منتجاتها غير. ضارة للثدييات ولا تؤثر. على خصائص صناعة الغبز. وأن تكون سامة للأطوار المختلفة للآفة. إن تطوير نبات محور وراثياً يتطلب توفر ثلاث مكونات رئيسة: جينات مبيدات الحشرات، نظام نقل جينات فعال والقدرة على تحقيق مستوى عال من التحوير الوراثي في نسيج معين. نحن نخطط لاستخدام أربع مجموعات من الجينات: 1) جينات مرمزة لمثبطات بروتياز النبات ومثبطات ألفا أميلاز؛ 2) جينات لكتينات النبات [الثوم (Allium sativum) ليكتين الورقة (ASAL) وقط\_\_\_رات ال\_\_\_ثلج (ASAL Galanthus) جـــين اللكتــين GNA]؛ nivalis 3) جينات من البكتريا Bacillus thuringiensis - جينات الـ Ht، التي ترمز بروتينات مبيدات الحشرات، المعروفة به delta-endotoxins؛ 4) ربما جينات مصنعة. استخدم من أجل ذلك ناقل psGFPBAR مصمم خصيصاً لنقل القائمة الحبوبة للحبوب. تم اختيار نمط الانتخاب لنباتات القمح المحورة ورائياً على قاعدة عمل شريط الجينات. إن طرائق تحوير القمح المعروفة لأصناف القمح الروسي تملك فعالية تصل إلى 2-3% تقريباً وتسمح لنا بالبدء بهندسة قمح مقاوم لآفة القمح الوطنية الرئيسة -.E. integriceps

كلمات مفتاحية: تحوير وراثى، مقاومة حشرة السونة، قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# The Effect of Starch Granule Size of Grain's Endosperm on the Resistance of Wheat Cultivars to Sunn Pest (Eurygaster integriceps Puton)

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#### Abstract

# Rezabeigi, M., Gh. Radjabi and G. Nouri Ganbalani. 2007. The Effect of Starch Granule Size of Grain's Endosperm on the Resistance of Wheat Cultivars to Sunn Pest (*Eurygaster integriceps* Puton). Pages 391-397.

The abundance of small (1-10 micron) and large (10-30 micron) starch granules in damaged and nondamaged flour of 12 wheat cultivars with different level of resistance to Sunn Pest were measured. The results showed that there are significant differences between the rate of small and large starch granules in wheat cultivars. Based on this cue, it has been found that Sunn Pest inflicts damage mostly to the small size granules, and as the percentage of large granule increases the rate of susceptibility of wheat decreases. The correlations between the percentage of the large-size granules, the relative abundance of large granules to the small ones and resistance indices of wheat varieties have proved to be significant and positive. Therefore, measuring starch granules has shown to be a good criterion in evaluating the resistance level of wheat varieties to the Sunn Pest and it may be possible to evaluate and screen wheat varieties based on the granule size of endosperm starch. Multiple regression equations have been developed to predict the indices of resistant lines of wheat to Sunn pest.

Keywords: Eurygaster integriceps Put., resistance, starch granules, Sunn Pest, wheat.

#### Introduction

The size of starch granules of grain's endosperm is introduced as a good indicator to select wheat varieties resistant to Sunn Pest in the earliest stages of breeding. Evaluating wheat grains being damaged by Eurygaster integriceps Put. has showed that the insect feeds preferably on fine-grained starch and a correlation has been found between the composition of the middle part of the endosperm, as reflected in the proportions of starch-grain sizes, and the degree of damage caused by this pest (8). The large starch grains in the endosperm succumbed to hydrolysis by the enzymes of the Sunn Pest more slowly than the small grains and were found to be a good criterion for evaluating the resistance of a given variety (6). Different degrees of susceptibility to hydrolysis by digestive enzymes of Sunn Pest were found to exist in

the starch complex in different varieties of wheat, and in resistant varieties this complex was the most resistant to hydrolysis. The degree of availability of the carbohydrate in the starch complex to E. integriceps can be used as a criterion of varietal resistance to this pest (5). Some methods are described for determining the degree of resistance of wheat to E. integriceps based on measuring the degree of starch breakdown from the grain affected by salivary-glands hydrolyses of the insect under standardized conditions (3, 4). An analysis of the starch grains in the endosperm of a number of varieties has indicated that the different starch grains size could be used as indicators of resistance. The endosperm of resistant varieties contained the largest starch grains, while susceptible varieties had mainly small starch grains (7).

This investigation was carried out to compare the abundance of small and large

granules in damaged and non-damaged kernels of 12 wheat cultivars with different level of resistance. Studying correlation between the rate of the small and large-size granules and resistance indices of wheat cultivars to Sunn Pest was another objective of this research.

#### Material and Methods

#### **Field Study**

To evaluate the resistance of 12 wheat cultivars to the nymphal stages of Sunn Pest (Eurygaster integriceps Put.), three experimental methods were used during 1995-97. All experiments were carried out by using factorial design based on randomized complete block design (RCB) with 4 replications. The first factor was 12 wheat cultivars and the second one was 2 different densities of pests. In the first method conducted under the field conditions, Sunn Pest eggs obtained in the rearing of 3 and 5 pairs of overwintered adults under greenhouse conditions were used to infest the plants in the field. In a similar experiment, eggs laid under field conditions were used to infest plants. Finally, in the third approach, experiments were conducted in cages installed in the field using two different population density of the pest (50 and 70 eggs per cage). In all three approaches, characters studied, included percent mortality of nymphs, weight of newly emerged adults, dry weight and fat body percentage of newly emerged adults, percent of grain damaged, percent of grain damaged per newly emerged adults, and weight reduction percentage in 50 damaged grains.

#### Laboratory Study

The abundance of small (1-10 micron) and large (10-30 micron) starch granules in damaged and non-damaged kernels of wheat cultivars were measured by microscopic examination (Figure 1). In this regard, 10 mg flour of each sample was transmitted to a microscope slide. After staining of flour by Lugol's solution (0.3 gr I + 3 gr KI + 100 ml Dis. water), the abundance of starch granules

measured by a graticule eyepiece in 400 X magnification (10 views in 4 replications). After measuring the number of starch granules, the percentage of small and large granule sizes, the relative of large granules to the small ones and the reduction percentage of small granules were calculated.



**Figure 1.** The small and large size starch granules in Sardari cultivar flour. S, small size granules (1-10  $\mu$ ) and L, large size granules (10-30  $\mu$ ).

#### Data Analysis

Analysis of variance of the all collected data in the field and laboratory were done by MSTAT-C software and means comparisons were conducted by Duncan's multiple range and Tukey's W-procedure. Resistance indices of wheat cultivars were calculated by ranking of data by normal distribution (1). In this regard, we used 2, 4, 6 and 8 indices for susceptible, semi-susceptible, semi-resistant and resistant cultivars. The correlations and multiple regression analysis of resistance indices and the other collected data were computed by MINITAB software.

#### Results

# Resistance Indices of Wheat Cultivars to Sunn Pest

In all three evaluating methods, significant differences have been shown among wheat cultivars. Similar results were obtained in three different approaches (Table 1). According to data analyses, Falat cultivar was

ranked as resistant and Azadi, Golestan, Ghafghaz, and Navid were grouped as semiresistant ones. Other cultivars including Bistoon, Sabalan, Zardak, Tabassi, Rashid, Bezeostaya, and Sardari were ranked as semisusceptible to Sunn Pest. No significant differences were observed regarding to density levels of the pest.

#### Abundance of Starch Granules in Grains of Wheat Cultivars

There were significant differences among starch granules in wheat cultivars (Table 2). Rashid (a susceptible cultivar) had the highest rate of small size granules and Falat (a resistant one) showed the lowest rate. Falat ranked as having the highest rate of large size granules and Bistoon (semi-susceptible cultivars) ranked as having the lowest. Bezostava and Navid had the lowest level percentage of small size granules reduction and Sardari (a semi-susceptible cultivars) had the highest level.

There were significant differences among small and large sizes starch granules in damaged and non-damaged grains using t-test analyses (P=0.01). The abundance of small size granules was 44.1 and 27.8 in nondamaged and damaged grains respectively but those for large size ones were 5.4 and 7.9, respectively (Figure 2). Decreasing of small size granules and increasing of large size ones in damaged kernels show that the nymphs and newly emerged adults of Sunn Pest can hydrolyse only the small size granules. Storage of large size granules in grain endosperm continues during feeding period of pest and the rate of these granules increase in damaged kernels.



Figure 2. The abundance of starch granules in damaged and non- damaged grains.

Table 1.	The	means	of resistance	indices of	of wheat	cultivars t	o nymphs	and	newly	emerged	adults	of Sunn
Pest (Va	ramin	station	ı, 1995-97).									

	First ev	valuating	method	Seco	Second evaluating method		Third evaluating method			Mean of all evaluating methods		
	Dens adu	sity of ults		Dens ad	Density of adults		Density of eggs			Density of pest		
Sources	3 pairs	5 pairs	Mean	3 pairs	5 pairs	Mean	50	70	Mean	Low	High	Mean
Sabalan	4.0 c	5.0 b	4.2 c	3.4 d	5.0 bc	4.6 c	4.6 c	5.2 b	4.6 c	4.0 c	5.1 b	4.5 c
Falat	7.6 a	7.2 a	7.4 a	7.2 a	7.2 a	7.6 a	7.8 a	7.8 a	7.8 a	7.5 a	7.4 a	7.6 a
Sardari	5.4 b	3.8 c	4.4 c	3.8 c	3.8 c	3.6 c	4.0 c	3.4 d	3.4 d	4.4 c	3.7 c	3.8 c
Zardak	5.0 bc	4.6 c	5.0 bc	3.8 c	4.2 c	3.6 c	4.2 c	5.6 b	4.6 c	4.3 c	4.8 c	4.4 c
Bezostaya	3.8 c	4.0 c	3.8 c	4.8 c	4.8 c	4.6 c	3.8 c	3.2 d	3.8 c	4.1 c	4.0 c	4.1 c
Tabassi	5.2 b	4.2 c	4.6 c	4.0 c	3.8 c	4.0 c	4.4 c	2.8 d	3.2 d	4.5 c	3.6 c	3.9 c
Rashid	2.8 d	3.2 d	2.6 d	4.2 c	4.4 c	4.2 c	4.6 c	4.6 c	4.8 c	3.9 c	4.1 c	3.9 c
Bistoon	4.8 c	3.6 c	4.2 c	5.2 b	5.0 bc	5.0 bc	4.8 c	5.0 b	5.0 bc	4.9 c	4.5 c	4.7 c
Azadi	5.4 b	6.4 b	6.0 b	5.6 b	6.0 b	5.8 b	6.0 b	5.6 b	5.8 b	5.7 b	6 b	5.9 b
Golestan	6.4 b	7.0 a	7.0 a	6.4 b	4.8 c	5.8 b	6.2 b	5.2 b	5.4 b	6.3 b	5.7 b	6.1 b
Ghafghaz	5.2 b	5.0 bc	5.2 b	5.6 b	5.6 b	6.0 b	4.4 c	5.6 b	5 bc	5.1 b	5.4 b	5.4 b
Navid	4.8 c	6.8 a	5.8 b	5.4 b	5.0 bc	5.0 bc	4.0 c	5.2 b	4.6 c	4.7 c	5.7 b	5.1 b

Ranking of treatments were done by normal distribution. a, b, c and d are Resistant, Semi-resistant, Semi-susceptible and Susceptible respectively.

	Non-dama	ged grains	Damage	ed grains	Percentage of
Sources	Small size granules (1-10 ~)	Large size granules (10-30 ~)	Small size granules (1-10 ~)	Large size granules (10-30 ~)	reduced small size granules in damaged grains
Sabalan	44.2 a-c	4.4 de	20.8 de	5.8 ef	53.0 ab
Falat	42.2 a-c	8.6 a	26.8 cd	9.0 ab	36.1 b-d
Sardari	46.1 ab	5.4 b-d	18.0 e	5.3 f	60.7 a
Zardak	36.9 c	5.2 b-e	21.0 de	9.4 ab	43.0 a-c
Bezostaya	43.1 a-c	5.0 de	33.5 ab	6.4 d-f	22.2 d
Tabassi	39.8 bc	6.4 b	29.8 bc	6.2 ef	25.1 cd
Rashid	48.6 a	5.0 с-е	35.3 ab	10.0 a	27.3 cd
Bistoon	44.2 a-c	4.2 e	26.8 cd	9.7 a	39.4 b-d
Azadi	43.9 a-c	4.6 de	34.8 ab	6.8 de	24.1 cd
Golestan	46.4 ab	4.2 e	31.3 a-c	9.9 a	37.4 cd
Ghafghaz	46.6 ab	4.5 de	29.2 bc	7.5 cd	37.4 cd
Navid	47.3 ab	6.8 bc	36.4 a	8.5 bc	22.5 d

**Table 2.** Abundance of starch granules in damaged and non-damaged grain's flour ( $10 \times 10$  micron in  $400 \times$  magnification).

Same letters are not significantly different (P=0.01, Duncan's multiple range test).

#### **Correlation and Regression Analyses**

Among characters studied, weight and dry weight of newly emerged adults, fat body percentage of adults, percentage of damaged grains per newly emerged adults and weight reduction percent per 50 damaged grains had significant correlation with resistance index, but correlations between percentage of nymphs mortality, percentage of damaged grains and resistance index were not significant (Table 3). Significant correlations were found among the abundance of large size granules, weight and dry weight of newly emerged adults, weight reduction percent per 50 damaged grains and resistance index.

The results showed that the abundance of large size starch granules has a positive effect on resistance of wheat cultivars to Sunn Pest, but there were not any significant correlation between small size starch granules and resistance components. Percentage of nymphs mortality had a significant correlation with the abundance of small size granule starch, but it is not important because the nymphs mortality has no relationship with other resistance components such as resistance index. We created some regression equations to predict components according resistance to measurement of starch granules (Table 4). Coefficient of determination  $(R^2)$  to predict resistance indices according to abundance of large size of starch granules was 14.5 and showed that we can predict only 14.5 percentage of resistance index alternation according to measurement of large size starch granules in wheat grains.

#### Discussion

As a result, we found that the nymphs and newly emerged adults of Sunn Pest feed on only small size starch granules and probably could not hydrolyse large size ones. Our results confirm the gained data by previous researchers (3, 4, 5, 8). We need to find out the reasons of stability of large size starch granules to salivary gland enzymes of Sunn Pest. Burinskaya (2) found that there was no difference in the amylose and amylopectin contents of the starch of ripe grains of wheats Ukrainka (resistant to *Eurygaster integriceps*) and Bezostaya 1 (susceptible), but we suggest that those components have to be measured in two groups of resistant and susceptible wheat varieties. As Gapanova (6) and Shapiro and Nefedova (7) recommended, the abundance of

	Abundance of small size starch granules	Abundance of large size starch granules	The relative abundance of large gran ules to the small ones	% of small size starch granules reduction	% of nymphs mortality	Weight of newly emerged adults of Sunn pest (mg)	Dry weight of newly emerged adults of Sunn pest (mg)	Fat body % of newly emerged adults	% of damaged grains	% of damaged grains per newly emerged adults	Weight reduction percent per 50 amaged grains
Abundance of large size starch granules	-0.23										
The relative abundance of large granules to the small ones	-0.50*	0.96**									
% of small size starch granules reduction	-0.03	-0.21	-0.17								
% of nymphs mortality	-0.73**	0.19	0.39	0.12							
Weight of newly emerged adults of Sunn pest (mg)	0.22	-0.51**	-0.53**	0.25	-0.15						
Dry weight of newly emerged adults of Sunn pest (mg)	0.12	-0.56**	-0.53**	0.01	-0.18	0.72**					
Fat body % of newly emerged adults	-0.24	-0.33	-0.23	0.25	-0.05	0.59**	0.77**				
% of damaged grains	0.24	0.20	0.09	-0.23	-0.49*	0.12	0.23	0.09			
% of damaged grains per newly emerged adults	-0.24	-0.16	-0.08	-0.02	0.16	0.63**	0.62**	0.68**	0.20		
Weight reduction percent per 50 damaged	0.24	-0.40*	-0.42*	-0.30	-0.20	0.62**	0.50*	0.32	0.13	0.44*	
grains Resistance Index	0.01	0.43*	0.38	-0.09	-0.04	-0.82**	-0.91**	-0.84**	-0.16	-0.81**	-0.53*

Table 3. Correlation coefficients between resistance indices of wheat cultivars to Sunn Pest and the abundance of starch granules in their grains.

\* and \*\* are significantly differences in P=0.05 and P=0.01, respectively.

	Abundance of small size starch granules	Abundance of large size starch granules	The relative abundance of large granules to the small ones	Percent of small size starch granules reduction	Intercept	Coefficient of determination	
Resistance Indices	(X1)	(X2)	(X3)	(X4)	( <b>C</b> )	( <b>R</b> <sup>2</sup> <b>Adj.</b> )	Р
Weight of newly emerged adults of Sunn pest (mg)		-1.68			133	23	0.01
Dry weight of newly emerged adults of Sunn pest (mg)		-1.78			64.5	26.7	0.005
Weight reduction percent per 50 damaged grains	-0.122		0.561		30.9	26.3	0.016
Weight reduction percent per 50 damaged grains		-1.17			26.0	11.9	0.06
Resistance Index		0.357			3.06	14.5	0.04

**Table 4.** The results of regression analysis to predict resistance indices of wheat cultivars to Sunn Pest regarding abundance of starch granules in their grains.

Regression equations are  $R = C + a X_1 + bX_2 + ... nX_n$ . R, resistance index; C, intercept of R and a, b and n are regression coefficient for variables X1, X2 and ...Xn.

large size starch granules is a good index to select wheat varieties resistant to Sunn Pest in the earliest stages of breeding, but evaluating of selected varieties in greenhouse and field conditions is necessary. Our complementary studies on relationship between resistance to Sunn Pest in other wheat genotypes and their starch granules showed same results (9). So we need to do some heritability tests on wheat cultivars with a high rate of large size starch granules.

#### Acknowledgments

We thank Dr. R. Heydari for technical advising and Dr. G.A. Abdollahi and Dr. M. Amirmaafi for financial support of this project. We also thank A. Matin for some services in the field.

# الملخص

ريذابيجي، م.، غ. راجابي، وغ. نوري غانبالاني. 2007. تأثير حجم حبيبات نشاء سويداء الحبوب في مقاومة أصناف القمح المزروعة لحشرة السونة (Eurygaster integriceps Puton). الصفحات 391–397.

تم قياس وفرة حبيبات النشاء الصغيرة (1–10 ميكرون) والكبيرة (10–30 ميكرون) في الطحين المصاب وغير المصاب لـ 12 صنف مزروع من القمح بمستويات مختلفة من المقاومة لحشرة السونة. أظهرت النتائج أن هناك فروقاً معنوية بين نسبة حبيبات النشاء الصغيرة والكبيرة في أصناف القمح المزروعة. اعتماداً على ذلك، فقد وجد أن حشرة السونة توجه الضرر على الأغلب للحبيبات صغيرة الحجم، وبزيادة النسبة المئوية للحبيبات الكبيرة تنخفض درجة حساسية القمح. ثبت أن علاقات الارتباط بين النسبة المئوية للحبيبات كبيرة الحجم، الوفرة النسبية المعربيات كبيرة الحجم بالنسبة للحبيبات صغيرة الحجم ومؤشرات مقاومة بين النسبة المئوية للحبيبات كبيرة الحجم، الوفرة النسبية للحبيبات كبيرة الحجم بالنسبة للحبيبات صغيرة الحجم ومؤشرات مقاومة أصناف القمح معنوية وإيجابية. لذلك، ظهر أن قياس حبيبات النشاء هو مؤشر جيد في تقييم مستوى مقاومة أصناف القمح لحشرة السونة وقد يكون ممكناً تقييم وغريلة أصناق القمح اعتماداً على حجم حبيبات نشاء سويداء الحبوب. تم تطوير معادلات انحدار متعددة للتنبؤ بمؤشرات أصناف القمح المقاومة لحشرة السونة. كلمات مفتاحية: Eurygaster integriceps Puton، مقاومة، حبيبات نشاء، حشرة السونة، قمح.

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Bruce L. Parker, Margaret Skinner, Mustapha El Bouhssini & Safaa G. Kumari (Eds.): Sunn Pest Management: A Decade of Progress 1994-2004 © 2007 Arab Society for Plant Protection, ISBN 978-9953-0-1063-2, Printed in Beirut, Lebanon.

# 2<sup>nd</sup> International Conference on Sunn Pest ICARDA, Aleppo, Syria, July 19-22, 2004

# **Conference Committees**

#### **Conference Coordinators**

Mustapha El Bouhssini (Chairperson) International Center for Agricultural Research in the Dry Areas (ICARDA Aleppo, Syria

Margaret Skinner Entomology Research Labortory University of Vermont Burlington, Vermont, USA

#### **International Organizing Committee**

Lukas Brader (Chairperson) International Center for Agricultural Research in the Dry Areas (ICARDA Aleppo, Syria

Ghulam Abbas Abdollahi Plant Pests and Disease Research Institute Tehran, Iran

David Hall Natural Resources Institute Kent, Unted Kingdom

#### **ICARDA Local Organizing Committee**

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Aden Aw-Hassan International Center for Agricultural Research in the Dry Areas (ICARDA Aleppo, Syria

Ramazan Canhilal Plant Protection Research Institute Adana, Turkey

Peter Kenmore Food and Agriculture Ogrnazation Rome, Italy

William Reid Fadel Afandi Ahmed Shahbandar

# 2<sup>nd</sup> International Conference on Sunn Pest

ICARDA, Aleppo, Syria, July 19-22, 2004

# **Conference Program**

# Sunday, 18 July 2004

19:30-22:00 Cocktail & Registration, at ICARDA office I, Aleppo city

# Monday, 19 July 2004

08:00	Transport to Tel Hadya (ICARDA Headquarters)
08:15-9:00	Registration
9:00-9:30	Opening Statements
	University of Vermont, by Prof. Dr. Bruce L. Parker Arab Society for Plant Protection, by Dr. Wafa Khoury ICARDA, by Prof. Dr. Adel El-Beltagy - Director General
9:30-10:00	Coffee Break and Group Photo
10:00-11:30	KEYNOTE PRESENTAT ONS Chairperson: Dr. William Erskine, ICARDA, Syria
10:00-10:45 10:45-11:30	<b>IPM in the CGIAR,</b> <i>by Khaled Makkouk, Al-Manar University of Tripoli, Lebanon</i> <b>Lessons Learned from IPM Programmes: Ecology, Policy, and Community</b> <b>Empowerment,</b> <i>by Peter Kenmore, FAO, Italy</i>
11:30-15:00	SESS ON 1: SOC OECONOM CS OF SUNN PEST Chairperson: Dr. Majd Jamal, GCSAR, Syria
11:30-11:45	The basic role of extension, farmer grouping and participation in Sunn Pest management, by Gholam Abbas Abdollahi, PPDRI Iran
11:45-12:00	Analysis of policies on Integrated Pest Management on Sunn Pest in Iran, Syria and Turkey, by Aden Aw-Hassan, ICARDA, Syria
12:00-12:15	Shifting from aerial to ground spraying in Sunn Pest control in the Trace region of Turkey: Farmers' perception and problems, by Aykut Gul, University of Cukurova, Turkey
12:15-12:30	<b>Positive effects of commodity exchange in Sunn Pest control in Turkey in recent</b> <b>vears</b> , by Avkut Gul, University of Cukurova, Turkey
12.30-12.45	The role of Socio-economic characteristics in the holding of farmer field school and
12.30 12.43	acceptance of participation in IPM of Sunn Pest, by Majid Hassani Moghaddam, PPDRI, Iran

13:00-14:00	Lunch
	SESS ON 1 (Continued)
14:00-14:15	The loss assessment and economic levels for Sunn Pest, <i>Eurygaster integriceps</i> Puton, through cost-benefit methods in Qazvin province of Iran, by Hossein Noori, Oazvin Agricultural and Natural Research Center Iran
14:15-14:30	Determination and comparison of allocative, technical and economic efficiency in outside and inside of integrated Sunn Pest management pilot sites in Iran, by Majid
14:30-14:45	<b>Evaluating Sunn Pest management in Turkey in terms of food security, human health, environmental conservation and economic returns,</b> by Hasan Akca,
14:45-15:00	Gaziosmanpasa University, Turkey Modified Farmers' Field Schools as a new option in IPM of Sunn Pest Eurygaster itegriceps Puton (Hemiptera: Scutelleridae), by Mohammed Abdulhai, GCSAR, Syria
15:00-18:30	SESS ON 2: SUNN PEST B OLOGY AND ECOLOGY Chairperson: Dr. Gholam Abbas Abdollahi, PPDRI, Iran
15:00-15:15	Sunn Pest population distribution in overwintering sites in Iran, by Bruce L. Parker,
15:15-15:30	Damage loss assessment of Sunn Pest, <i>Eurygaster integriceps</i> Puton (Hemiptera: Scutelleridae) on wheat in Turkey, by Ramazan Canhilal, Adana Plant Protection
15:30-15:45	Investigation of mechanisms of mate-finding in Sunn Pest, by David Hall, University of Greenwich, UK
15:45-16:15	Coffee Break
	SESS ON 2 (Continued)
16:15-16:30	Mitochondrial DNA variations in Iranian geographic populations of Sunn Pest,
16:30-16:45	Morphometric studies on Iranian geographic populations of Sunn Pest, Eurygaster integricens Puton by Alimorad Sarafrazi PPDRI Iran
16:45-17:00	The role of biocontrol agents in decreasing the population of Sunn Insect Eurygaster integriceps Puton in wheat fields in the north of Iraq, by Mohammed A. J. Al-Izzi, $AOAD$ Sudan
17:00-17:15	Entomophilous and entomopathogenic fungi associated with Sunn Pest in overwintering sites. by W Reid ICARDA Syria
17:15-17:30	Sunn Pest, Eurygaster integriceps Puton, status in Iraq, by Nazar N. Hama, SBAR, Iraa
17:30-17:45	The impact of Sunn Pest density in wheat fields on grain and flour quality, by Fouad laby Fl-Haramein ICARDA Syria
17:45-18:00	A new technique for monitoring of the Sunn Pest, <i>Eurygaster integriceps</i> Puton (Hemiptera: Scutelleridae), by Heidar Adldoost, Agricultural Research Center of West Azarbaijan Iran
18:00-18:15	Life history and reproductive table of <i>Eurygaster maura</i> L. (Het.: Scutelleridae) in the laboratory, by Jafar Mohaghegh, PPDRI, Iran
18:30	Transportation to hotels in Aleppo

# Tuesday, 20 July 2004

08:00	Transport to Tel Hadya (ICARDA Headquarters)
08:30-10:00	KEYNOTE PRESENTAT ONS Chairperson: Dr. Lukas Brader, Netherlands
08:30-09:15	Green Muscle, from isolate to commercial product, by Christiaan Kooyman, IITA, Republic of Benin
09:15-10:00	<b>Biorational Insecticide use in integrated pest management,</b> by Charles Vincent, Agriculture and Agri-Food Canada, Canada
10:00-10:30	Coffee Break
10:30-12:30	SESS ON 3: SUNN PEST B OLOGY AND ECOLOGY Chairperson: Dr. David Hall, University of Greenwich, UK
10:30-10:45	Host plant-associated variation and sexual dimorphism in size and shape in Iranian geographic populations of Sunn Pest, Eurygaster integriceps Puton, by
10:45-11:00	Biological parameters of Sunn Pest, Eurygaster integriceps Puton, in wheat fields in the north of Iraq, by Mohammed Al-Izzi, AOAD, Sudan
11:00-11:15	Population density of Sunn Pest, <i>Eurygaster integriceps</i> Puton, during hibernation and aestivation in the North of Iraq, by Hassan S. Al-Assadi, University of Baghdad, Iraa
11:15-11:30	Survey of egg parasitoids of Sunn Pest in Northern Syria, by A. N. Trissi, Aleppo University Syria
11:30-11:45	Survey of Sunn Pest adult parasitoids in Syria, by Mohammed Abdulhai, GCSAR, Syria
11:45-12:30	Discussion
12:30-13:30	Lunch
13:30-15:30	TOUR OF CARDA

- 15:30-16:45 SESS ON 4: IPM: CHEM CAL CONTROL Chairperson: Dr. Ramazan Canhilal, GDPC, Turkey
  - 15:30-15:45 Timing as a tactic of ecological selectivity for chemical control of Sunn Pest, *Eurygaster integriceps* Puton, by A. Sheikhi Garjan, PPDRI, Iran
  - 15:45-16:00 The influence of insecticide residues on the foraging behaviour of the Sunn Pest egg parasitoid (*Trissolcus grandis*) in the laboratory, by A. Sheikhi Garjan, PPDRI, Iran
  - 16:00-16:15 Effects of conventional insecticides on Sunn Pest egg parasitoids *Trissolcus grandis* and *T. semistriatus* (Hymenoptera: Scelionidae), by Moosa Saber, Maragheh College of Agriculture, Iran
  - 16:15-16:30 Chemical control of Sunn Pest with pesticides (ULV) applicable for emulsion in water by using ground spraying system, by Nassir A. Al-Gamali, State Board of Plant Protection, Iraq

16:30-16:45 **The effects of application methods in chemical treatments for Sunn Pest control in Turkey,** by Arzu Erman, Plant Protection Central Research Institute, Turkey

# 16:45-18:15 Coffee Break & POSTERS SESS ON I: IPM

- 1. Outbreaks of cereal Sunn Pests and solutions for sustainable control, by M. Javahery, Macdonald Campus of McGill University, Canada
- 2. The effect of temperature on the development, reproductive potentials, and longevity of *Trissolcus semistriatus* Nees (Hymenoptera: Scelionidae) under constant temperatures, by S. Tarla, University of Mustafa Kemal, Turkey
- 3. Binomial and sequential sampling of adult Sunn Pest, *Eurygaster integriceps* Puton (Heteroptera: Scutelleridae), *by M. Amir-Maafi, PPDRI, Iran*
- 4. Effects of indoxacarb to a predacious mirid and two species of predatory mites found in orchards, by Charles Vincent, Agriculture and Agri-Food Canada, Canada
- 5. UDA-245: a botanical insecticide/acaricide for worldwide horticultural uses, by Charles Vincent, Agriculture and Agri-Food Canada, Canada
- 6. Evaluation of wheat cultivars and advanced lines for resistance to Sunn Pest (*Eurygaster integriceps* Puton) in Iran, by A. R. Haghshenas, Isfahan Agricultural and Natural Resources Research Center, Iran
- 7. Study on predatory and parasite mite associated with overwintering sites of sunn pest in western Iran, by Mohammad Khanjani, Faculty of Agriculture, Hamada, Iran
- 8. The present situation of some alternative hosts (*Dolycoris baccarum L., Piezodorus lituratus* F.) on the natural enemies of Sunn Pest (*Eurygaster integriceps* Puton) in the Southeastern Anatolian region of Turkey, by Ayse Akkaya, Plant Protection Research Institute, Diyarbakır, Turkey
- 9. Screening of some wheat varieties to Sunn Pest in Nineveh province, Iraq, by Suaad Irdeny Abdullah, Mosul University, Iraq
- 10. Temperature and Rainfall: Factors for fungal success in Sunn Pest overwintering sites, by Margaret Skinner, University of Vermont, USA
- 11. Comparative study of the properties and regulation of the activity of cholinesterases and carboxylesterases of Sunn Pest, *Eurygaster integriceps* Puton, by Habib H. Kushiev, Gulistan State University, Uzbekistan
- 12. **Observations on important wheat pests in Tajikistan,** by Munira Otambekova, Tajikistan
- 18:15 Transportation to hotels in Aleppo
- 20:00 Dinner Reception

# Wednesday, 21 July 2004

08:00	Transport to Tel Hadya (ICARDA Headquarters)
08:30-10:00	<b>KEYNOTE PRESENTAT ONS</b> <i>Chairperson: Dr. Bruce L. Parker, University of Vermont, USA</i>
08:30-09:15	Miracle Economics and IPM: Success in developing countries, by Douglas Gollin,
09:15-10:00	<b>How can scientists and scientific programmes connect with public and private</b> <b>enterprises to further the cause of IPM,</b> by Lukas Brader, Herent, Belgium
10:00-10:30	Coffee Break
10:30-12:45	<b>SESS ON 5: IPM: B OLOG CAL CONTROL / IPM: GENERAL TOP CS</b> <i>Chairperson: Dr. Charles Vincent, Agriculture and Agri-Food Canada</i>
10:30-10:45	<b>Cereal Sunn Pests and sustainable management in the 21<sup>st</sup> century</b> , by M. Javahery, Mandonald Commun of MaCill University, Conada
10:45-11:00	Sunn Pest management in Romania, by Felicia Mure anu, Agricultural Research and Development Station Turda, Romania
11:00-11:15	Entomopathogenic fungi for management of Sunn Pest: efficacy trials in overwintering sites by Magaget Skinger, University of Vermont USA
11:15-11:30	<b>The development of a mycoinsecticide for the biological control of Sunn Pest</b> , by Dave Moore CABL Bioscience, UK
11:30-11:45	Mass production of entomopathogenic fungi used for the biological control of insect pests. by Dave Moore, CABI Bioscience, UK
11:45-12:00	Initial studies on the potential use of an entomopathogenic fungal granular formulation along the edges of wheat fields, by W. Reid, ICARDA, Syria
12:00-12:15	Studies on the egg parasitoid ( <i>Trissolcus</i> spp., Hymenoptera: Scelionidae) of Sunn Pest ( <i>Eurygaster</i> spp., Hemiptera: Scutelleridae) in Turkey, by Erhan Koçak, Central Plant Protection Research Institute Turkey
12:15-12:30	Infectivity of two isolates of <i>Steinernema feltiae</i> (Rhabditida: Steinernematidae) in relation to <i>Eurygaster maura</i> L. (Heteroptera: Scutelleridae) adults, by Erhan Kustel, Plant Partenting Carteril Property Institut, Tarkan
12:30-12:45	Initial studies on the efficacy of entomopathogenic nematodes against Sunn Pest and survey of Syrian soils for locally adapted isolates, by Mohammed Abdulhai, General Commission for Scientific Agricultural Research, Syria
12:45-13:45	Lunch
13:45-16:45	SESS ON 6: IPM HOST PLANT RESISTANCE / IPM GENERAL TOP CS Chairperson: Dr. Khaled Makkouk, MUT, Lebanon
13:45-14:00	Evaluation of wheat and its wild relatives for resistance to Sunn Pest under artificial infestation. by M. El-Bouhssini, ICARDA, Syria

- 14:00-14:15 Comparison of resistance to Sunn Pest (*Eurygaster integriceps* Puton) in some bread and durum wheat lines, by Manouchehr Rezabeigi, PPDRI, Iran
- 14:15-14:30 Effect of different cereals (Wheat and Barley) on the population of Sunn Pest, Eurygaster integriceps Puton (Heteroptera: Scutelleridae), in the South East Anatolia region of Turkey, by Vedat Karaca, Diyarbakir Plant Protection Research

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Institute, Turkey

14:30-14:45	Host-p	olant prefer	ence and pe	rformar	ice ł	oy diffei	ent develo	pm	enta	l stages	of Sunn
	Pest,	Eurygaster	integriceps	Puton,	on	wheat	cultivars,	by	Α.	Arzani,	Isfahan
	Univer	sity of Tech	nology, Iran								

- 14:45-15:00 The effect of the starch granule sizes of grain endosperm on the resistance of wheat cultivars to Sunn Pest (*Eurygaster integriceps* Puton), by Manouchehr Rezabeigi, PPDRI, Iran
- 15:00-15:15 **Development of an efficient wheat genetic transformation system for the creation** of the transgenic wheat resistant to the Sunn Pest (*Eurygaster integriceps* Puton), by A. K. Gaponenko, "Bioengineering" RAS, Russian Federation
- 15-15-15:30 **The Sunn Pest in former USSR countries: history and current status,** by Galia Zharmukhamedova, Institute for Plant Protection, Kazakhstan
- 15:30-15:45 **Resistance to Sunn Pest (Eurygaster integriceps Puton) in advanced lines of durum** and bread wheat, by Tohid Najafi Mirak, Seed and Plant Improvement Institute, Iran

#### 15:45-17:15 Coffee Break & POSTERS SESS ON II: BIOLOGY AND ECOLOGY

- 1. The investigation of -amylase activity in hibernating Sunn Pest (Eurygaster integriceps Puton), by Majid Kazzazi, University of Tehran, Iran
- 2. Relation between weed grasses and life cycle of Sunn Pest in the Setif high plains (North-East of Algeria), by Mohamed Fenni, University Ferhat Abbes, Algeria
- 3. Biology, ecology and chemical control of Sunn Pest, Aelia germari Kuster (Heteroptera: Pentatomidae), in the Setifian high plains (North-East of Algeria), by Mustapha Bounechada, University Ferhat Abbes, Algeria
- 4. A study of the Sunn Pest species on cereal crops in the Setifian High Plains (North-East of Algeria), by Adel Nadjib Chaker, University Ferhat Abbes, Algeria
- 5. Prediction of Sunn Pest life cycle using environmental data over the North-East of the I.R. of Iran, by A. Movaghar Moghaddam, Climatologic Research Institute, Iran
- 6. Morphometric analysis of two populations of *Eurygaster maura* L. (Heteroptera Scutelleridae) from Iran, by Jafar Mohaghegh, PPDRI, Iran
- 7. Studies on the determination of the fat rate of Sunn Pest, *Eurygaster integriceps* Puton, (Heteroptera: Scutelleridae) to forecast Sunn Pest populations in fields in the South East Anatolia Region of Turkey, by Abdullah Demir, Diyarbakir Plant Protection Research Institute, Turkey
- 8. Ecological principles of Sunn Pest monitoring in Kazakhstan, by Amangeldy Sarbaev, Research and Production Center for Agriculture and Plant Growing, Kazakhstan
- 9. Studies on Sunn Pest overwintering sites, parasitoids and effects of Sunn Pest on Cereal Lines and varieties in Konya province, Turkey, by A. Faik Yildirim, Anatolian Agricultural Research Institute, Turkey
- 10 **Harmfulness of Sunn Pest in grain crops of Azerbaijan**, by Sardar Ibragimov, Azerbaijan Research Institute of Agriculture, Azerbaijan
- 11. Sunn Pest populations under different plant species in overwintering sites in Iran, by Bruce L. Parker, University of Vermont, USA
- 12. Investigations on the mass production and storage possibilities of the egg parasitoid, *Trissolcus grandis* (Thomson) (Hymenoptera: Scelionidae), by Münevver Kodan, Central Plant Protection Research Institute, Turkey

17:15-18:30	CLOSING SESSION Chairperson: Dr. William Erskine
	Wrap-up comments- by Dr. Lukas Brader Closing - by Prof. Dr. Adel El-Beltagy

18:30 Transportation to hotels in Aleppo

# Thursday, 22 July 2004

Touristic Trip

07:30	Departure to Aleppo Citadel
08:00-09:15	Citadel visit
09:30	Departure to Carpet Workshop
10:30-11:45	Carpet workshop visit
11:45-12:30	Visit to Ain Dara Temple
13:00-14:00	Visit to Qalaat Seman (St. Simeon)
14:00-16:00	Lunch (st. Someon restaurant)
16:30	Visit the Roman Road
17:30	Arrival to Aleppo
18:30-20:30	Visit the old Souk (traditional shopping area)

# 2<sup>nd</sup> International Conference on Sunn Pest

ICARDA, Aleppo, Syria, July 19-22, 2004

# **List of Participants**

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# **External Morphological Differences Between Male and Female Sunn Pest**



Posterior portion of the abdomen of a male Eurygaster integriceps adult.



Posterior portion of the abdomen of a female Eurygaster integriceps adult.

# Where Not to Plant Wheat

In recent years farmers have cultivated marginal, less fertile lands located beside Sunn Pest overwintering sites. This leaves the wheat vulnerable to migrating adults in the spring and disrupts valuable habitats for beneficial insects.



As a component of IPM, farmers are now encouraged not to cultivate these areas, providing a buffer between overwintering sites and productive farm land, and enhancing the natural enemy complex.

