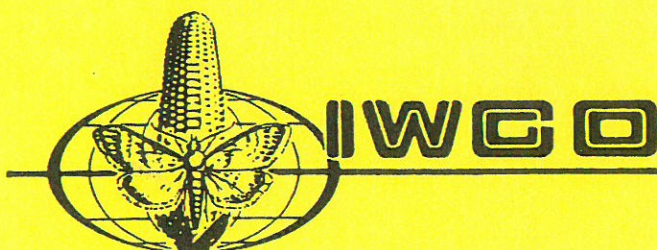


ORGANISATION INTERNATIONALE DE LUTTE
BIOLOGIQUE CONTRE
LES ANIMAUX ET LES PLANTES NUSIBLES



INTERNATIONAL ORGANIZATION
FOR BIOLOGICAL CONTROL
OF NOXIOUS ANIMALS AND PLANTS



**Proceedings of the XX Conference of
the International Working Group on
Ostrinia and Other Maize Pests**

**Adana (Turkey)
4-10 September 1999**



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Editor : Serpil Kornoşor

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PREFACE

The XXth International Working Group on *Ostrinia* and Other Maize Pests Conference was held in Adana, Turkey, September 4-10, 1999.

This conference was organised by the IOBC Global Working Group, IWGO and was by sponsored Agriculture Faculty of Cukurova University, Adana; The Scientific and Technical Research Council of Turkey and Plant Protection Research Institute, Adana.

The Conference was organised on two issues which were scientific reports sessions and documentary visits in experimental fields in the Çukurova Region.

The scientific sessions consisted of 28 scientific papers from Egypt, France, Germany, Greece, Hungary, Italy, Japan, Romania, Russia, Slovenia, Turkey, the United Kingdom, U.S.A. and Yugoslavia regarding the host specificity, mass rearing, natural enemies and biological control of ECB, Pink Stalk Borer and root worms problem in Germany and Romania. In addition, Bt-corn production, adaptation, effects of *Bacillus thuringiensis* on Stem borers, monitoring wire worm population with sex pheromone traps in different countries of Europe were discussed for the first time in XX th IWGO Conference.

The contents of scientific papers in this proceedings are under the responsibility of their corresponding authors.

As the organiser of the conference I would like to express my special thanks to all IWGO members and Organisation Scientific Committee and especially Dr. H. K. Berger for support in organising the conference and publishing the proceeding.

Serpil KORNOŞOR

XXth IWGO Meeting in Adana; Tukey

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It is the third time that an IWGO meeting has taken place outside Europe. The group previously met outside Europe in 1974 in the USA and Canada and 1986 in Beijing, China. When Prof. Dr. Serpil KORNOSOR, the Chairwomen of the Organizing Committee, invited IWGO at the XVIII meeting in Turda, Romania to come to Turkey the situation in regard to this meeting was not so clear. Things worked out for this meeting and we are all happy to be in Turkey.

A large number of participants responded to the call from Prof. Kornosor to attend the meeting in Adana. More than 16 countries are represented according to the applications received. It is a great pleasure for me to welcome also friends from Russia, and for the first time a participant from Japan. This also indicates the importance of this working group. Our numbers are growing with new members and new countries taking part.

It is remarkable that besides the meetings of our *Diabrotica* subgroup, also the "big" IWGO meetings are attended by large numbers of people. One of the reasons might be the construction of this the oldest Global - IOBC WG. Established in 1968, it is now in its 31st year of existence. Its importance is regularly acknowledged, and is considered an active and highly productive WG. It is the mixture of general and applied science related to maize production, which makes the WG so attractive to participants. And of course, the primary areas we are dealing with, that being the insects and diseases of maize represent global problems being addressed by fellow scientist.

Although IWGO also organizes meetings of the "IWGO - *Diabrotica* - subgroup" (1999: Paris, France), the subject *Diabrotica* will also be discussed during this biannually IWGO meeting. The main pests we are dealing with, at least within the last several years, *Ostrinia nubilalis* and *Diabrotica virgifera virgifera*, have become more and more important *Ostrinia* because of GMO maize varieties, which are resistant/tolerant to attack by this pest, and *Diabrotica* because of its recent appearance in Europe. IWGO - originally designed as a small group of scientists mainly involved in resistant breeding, has changed over time. As time has passed more and more entomologists have attended meetings and subsequently fewer breeders have been in attendance. Although breeders still have a part to play in IWGO, the expansion of the group into other areas of maize protection has been healthy.

Other organizations, such as EPPO (European and Mediterranean Plant Protection Organization) and FAO (Food and Agricultural Organization of the United Nations) have shown interest in the work of IWGO members on *Diabrotica*. It should be noted that research on this pest has become more and more difficult due to the well-known political situation in the Balkans. Now, after Serbia, Montenegro, Hungary, Croatia, Bosnia & Herzegovina, Bulgaria, Romania and perhaps Albania, the pest has reached Italy, the first EU country. This gives additional dimensions to this problem and adds additional challenges as to how the problem should be addressed.

Even though much of our effort have been devoted to the above pests, we must not forget that other pests and problems in maize, such as *Ostrinia furnacalis*, *Sesamia*

nonagrioides, aphids and others, are important to many of us. In some of the maize growing countries these pests are still more, or at least as important, as *O. nubilalis* and *D. virgifera virgifera*.

So I hope that this XX meeting of the International Working Group on Ostrinia and Other Maize Pests will be a successful meeting for each of you. I am sure the papers will be very interesting. As always, this is a good opportunity for friends and colleagues to get together to discuss common problems and matters of interest.

Bt Corn Production in the USA: Present Situation – Future Considerations

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Bt transgenic crops were first registered in the USA by the United States Environmental Protection Agency (USEPA) in 1995. The first commercial plantings were made in 1996 with less than 1% being planted each to corn ($\approx 161,943$ ha) and potato ($\approx 4,049$ ha) and 14% to cotton ($\approx 728,745$ ha). By 1998 these numbers had grown to 17% for corn ($\approx 4,858,300$ ha), 3% for potato ($\approx 20,243$ ha), and 17% for cotton ($\approx 931,174$ ha). USA Bt corn plantings for 1999 are projected at 20-25% ($\approx 6,283,400 - 7,854,251$ ha) of the total corn plantings (31,417,004 ha). To date, three Bt toxins have been incorporated into commercial corn and registered in the USA by USEPA. These include Cry1Ab, Cry1Ac, and Cry9C. The events associated with these incorporations include Event 176 (Novartis, KnockOut[®]; Mycogen, NatureGard[®]), BT11 (Novartis, YieldGard[®] and Attribute[®]), MON810 (Monsanto, YieldGard[®]), DBT-418 (DeKalb/Monsanto, Bt-Xtra[®]), and CBH-351 (AgrEvo/PGS, Starlink[®]).

One of the major issues associated with the extensive use of Bt corn is the potential for resistance to develop in the European corn borer (ECB), *Ostrinia nubilalis* Hübner, population. As a result of the recommendation of a Scientific Advisory Panel (SAP), the USEPA is planning to institute a 20% refuge requirement beginning in 2000 (50% if corn and cotton are planted in the same area, primarily for managing resistance in corn earworm/cotton bollworm, *Helicoverpa zea* (Boddie)). For ECB, Bt fields should have non-Bt corn within 0.8 km according to these planned regulations. This SAP panel was made up of individuals from registrants, academia, and public interest groups. Another issue of concern is the acceptance of Bt corn in foreign markets. It is still unclear as to how this will be resolved, but some markets are, and possibly will be closed to raw and processed commodities from these transgenics. Public perceptions/views related to the broader area of genetically manipulated plants and animals (GMO's) also bring a degree of uncertainty to the future of Bt corn and other transgenics. No doubt, Bt corn and on a larger scale GMO's are and will change our lives. One can only hope that these will be positive changes and that these organisms will be developed and used responsibly. It is imperative that these be adequately tested/evaluated prior to their release and use.

Key Words: Bt Corn, GMO, Transgenics, GMO/Transgenic Issues

Entomological Problems of Maize in Turkey

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Introduction

In Turkey, although it may vary each year, maize is grown in an approximate area of 600.000 ha, and is the thirdmost important crop following wheat and barley. With "The Second Crop Agricultural Research and Dissemination Project" administered by the Ministry of Agriculture in 1982, it was aimed to increase maize planting area to reach 700.000 ha by putting uncultivated irrigated land into use and starting maize production in the Southeast Anatolia (Anonymus 1987). In Turkey, maize production area have fluctuated in size between 1980 to 1995. Since the start and the spread of second crop sowing in The Mediterranean and The Aegean Regions in 1983 and the sowing of productive variety of hybrids, maize production has doubled. In Turkey, maize is mainly grown in The Black Sea, Marmara, Aegean and Mediterranean Regions. Maize has been grown in large area in The Black Sea and Marmara Regions only as a first crop, and in The Aegean and Mediterranean Regions as a first crop until 1983 and as a first and second crop since then. Although, the maize sowing area is the largest in In The Black Sea Region but it is quite low in production. On the other hand, the production is high in The Mediterranean Region but sown maize areas are 33% less in size compared to those in The Blacksea Region. After the sowing of maize in large areas as a second crop, pest population has been increased. Although some research on maize pests has been carried out before 1983 (Yürüten 1961, 1963, 1971; Teoman 1973; Özdemir 1974; Şimşek 1983), since then, a lot of research has been done mainly on the biology, population development, and the natural enemies of the major pests *Ostrinia nubilalis* Hbn. and *Sesamia nonagrioides* Lef. in Faculties of Agriculture, Plant Protection Research Institutes and Agricultural Research Institute (Kayapınar & Kornoşor 1991, 1992a,b, 1993; Kornoşor et al 1992; Kornoşor & Kayapınar, 1988, 1989; Özpınar & Kornoşor 1994 a,b, 1997; Özpınar, 1994 etc.). Pest and beneficial species have been determined in the maize agroecosystem which has a rich insect fauna in different maize growing region (Özpınar & Kornoşor 1994 a,b.; Sertkaya & Kornoşor, 1994; Özpınar & Kornoşor 1995, 1997; Melan et.al. 1996; Uzun et al. 1996; Sertkaya et al. 1999) There are differences as major pests between the first and the second crop of maize.

Main Pests on The First Crop Maize

In the seedling period, *Gryllotalpa gryllotalpa* L. (Orthoptera, Gryllotalpidae), wireworms *Agriotes lineatus* L. *A. obscurus* L. (Coleoptera, Elateridae) and cutworms *Agrotis ipsilon* Hufnagel, *Agrotis segetum* Denis & Schiffermüller (Lepidoptera, Noctuidae) which feed on the roots of the young plants, are all found to be harmful only for first crop maize, due to their seasonal appearance and climatic requirements (Table 1). Among these pests, cutworms, which are considered to be the most economically important pest, have occasionally damaged the roots of plants at the beginning of the seedling period, causing the fields to rest or crops to be re-sawn. Seed and surface insecticides are applied in order to prevent their damaging to the plants.

Table 1. Maize Pests Related to Maize's Growth Stages

Stages	Emergence-Two leaves	Early whorl (4-6 leaves)	Mid whorl (7-8 leaves)	Late whorl (10-12 leaves)	Tassel-Silk	Milky stage	Maturity
Plant parts							
Tassel					<i>Rhopalosiphum</i> spp. <i>O. nubilalis</i>	<i>O. nubilalis</i> <i>Rhopalosiphum</i> spp.	<i>O. nubilalis</i>
Ear					<i>O. nubilalis</i> <i>Sesamia</i> spp. <i>N. viridula</i> <i>H. armigera</i>	<i>O. nubilalis</i> <i>Sesamia</i> spp. <i>N. viridula</i> <i>H. armigera</i>	<i>O. nubilalis</i> <i>Sesamia</i> spp.
Stalk			<i>S. nonagrioides</i> <i>S. cretica</i>	<i>Sesamia</i> spp. <i>O. nubilalis</i> <i>N. viridula</i>	<i>O. nubilalis</i> <i>Sesamia</i> spp. <i>N. viridula</i> <i>Rhopalosiphum</i> spp.	<i>O. nubilalis</i> <i>Sesamia</i> spp. <i>N. viridula</i>	<i>O. nubilalis</i> <i>Sesamia</i> spp.
Leaves	<i>Thrips</i> spp. <i>Tetranychus urticae</i> <i>T. cinnabarinus</i> <i>Spodoptera exigua</i>	<i>Thrips</i> spp. <i>Tetranychus</i> spp. <i>S. exigua</i>	<i>Sesamia</i> spp. <i>Spodoptera exigua</i> <i>N. viridula</i> <i>Thrips</i> spp. <i>Tetranychus</i> spp.	<i>Sesamia</i> spp. <i>O. nubilalis</i> <i>A. loreyi</i> <i>P. unipuncta</i> <i>N. viridula</i> <i>Tetranychus</i> spp. <i>Rhopalosiphum</i> spp.	<i>O. nubilalis</i> <i>A. loreyi</i> <i>P. unipuncta</i> <i>N. viridula</i> <i>Rhopalosiphum</i> spp. <i>Tetranychus</i> spp.	<i>Tetranychus</i> spp. <i>N. viridula</i> <i>A. loreyi</i> <i>P. unipuncta</i>	
Root	<i>G. gryllotalpa</i> <i>Agrotis</i> spp. <i>Agriotes</i> spp.	<i>Agrotis</i> spp. <i>Agriotes</i> spp.					

Tanymechus dilaticollis Gyll. (Coleoptera, Curculionidae) was determined to be harmful only in The Black Sea Region in the seedling period in some years feeding on the leaves of the maize, and seed and surface insecticides are recommended to control this pest (Yürüten 1961).

In The Mediterranean Region, an increase in the population of *Pseudaletia unipuncta* (Haworth) and *Acantholecania loreyi* (Duponchel) (Lepidoptera, Noctuidae) which feed on the maize leaves has been observed, within the last ten years. These pests damage crops by cutting the tassels of maize cobs which have not yet completed their fertilisation. Both species have effective larva paratoids that decies 80% of their damage in the region (İkincisoay et al. 1994).

In addition, *Nezara viridula* Linnaeus (Heteroptera, Pentatomidae) which cannot feed sufficiently in wheat fields moves to maize fields and causes considerable deformations on the cops.

Pests sucking the sap of crops, such as *Rhopalosiphum maidis* Fitch., *R. padi* Linnaeus (Homoptera, Aphididae), *Tetranychus cinnabarinus* Boisduval, *T. urticae* (Koch) (Acarina, Tetranychidae) have seen but have also been kept control pressure by a rich natural enemy fauna. Problems occur in regions where insecticides are extensively used by these plant sucking insects.

Stemborers do not cause economic loss in the first crop maize in The Mediterranean and Aegean Regions due to their low population (Kavut 1985; Kayapınar & Kornoşor 1991; Derin 1992; Kornoşor et al., 1995; Özpınar & Kornoşor, 1997).

In the Marmara and the Black Sea regions, *Ostrinia nubilalis* is parasitized with the larva parasitoids of *Lydella thompsoni* Herting (Diptera, Tachinidae) at 55-62 %, and with *Trichogramma evanescens* Westwood at 97.03 % as a natural parasitization, there is no need to apply insecticides (Melan. et al. 1996) because of this chemical insecticides are not used. In conclusion, depending on the sawing time, there is no chemical application on the first crop maize in general.

Main Pests on the Second Crop Maize

Insects causing damage on the first crop maize, except pests which feed with roots of seedlings (for example *Agriotes* spp., *Agrotis* spp.), are harmful on the vegetation period of the second crop maize to a varying extent. Considering the crops, phenological development, in the Mediterranean Region, *Spodoptera exigua* are seen on maize crops at the four or five- leaf-stage, especially in those sowed with delay on the first half of July (1-15 July). In the region where intensive agriculture takes place, *S. exigua* has many hosts and its population starts increasing early June, and reaches the pick stage in July and August damaging the maize, clover, cotton, soybean, many arable plants and different weeds. Meanwhile, natural parasitism with larva parasitoids are quite high in maize in The Mediterranean Region. Damage is avoided by sowing the crops earlier than usual (for example from beginning to mid June). In regions where insecticides are widely used and sawing is late synthetic pyrethroids are applied to control these pests..

After the height of the maize reaches 40-50 cm, *O. nubilalis* starts being seen, depending on the sowing time and population density increases and it , becomes the major pest. It is kept as a major pest until milky and maturity stage. *O. nubilalis* population reaches its pick stage from late July till end of August and the population starts decreasing after early September (Kornoşor & Kayapınar, 1988, 1989; Kayapınar & Kornoşor, 1992a; Kornoşor et al., 1995)

Despite *S. nonagrioides* having a low population at the beginning of the vegetation period till early September, in other words towards the end of the season when the European Corn Borer population starts to fall, *S. nonagrioides* population reaches a high density and makes damages on the second crop maize severely, especially on the late sown maize (Kornoşor et al., 1995; Özpınar&Kornoşor, 1997; Bayram &Kornoşor, 1999).

Helicoverpa armigera Hbn. (Lepidoptera, Noctuidae) causes damage locally on the maize-cobs which have not yet fertilized by cutting their tassels, which encumbers the development of grains.

In some years *Spodoptera littoralis* Boisd. (Lepidoptera, Noctuidae) damages on maize beginning from the early August by eating the leaves and tips of the corn-cob in The Mediterranean Region.

Natural Enemies of Maize Pests

On the basis of regions the natural enemies of *O. nubilalis* and other maize pests have been studied extensively (Table 2). In these studies, the natural existence of *Trichogramma* has been noted and natural parasitization rate for each region and years, has been determined. This findings revealed that natural parasitization existed in the Black Sea Region at a level of 97.60 % (Melan et.al. 1996),in the Mediterranean Region 2.30 -51.06% depending on the year (Kornoşor & Kayapınar 1992a; Kornoşor et al., 1995) and in the Aegean Region with *T. brassicae*, 2.60-% 100 % (Uzun et al. 1994).

In the Black Sea and Aegean Regions, *L. thompsoni* has been found to be the most effective and widespread larva parasitoid. The percentage of this parasitoid is 4.54-65.34 % in the Aegean region, and 55.62 % in the Black Sea Region (Melam et al. 1996; Uzun et al.1994). As mentioned previously, due to the high percentage of parasitization by natural enemies, chemical control is not used to control *O. nubilalis* in the Black Sea Region.

The egg parasitoid of *S. nonagrioides*, *Platytenomus busseolae* (Gahan) (Hymenoptera, Scelionidae) has been commonly found in the Mediterranean region, and causing high percentage of parasitization depending on the year. Research reveals that in the years 1990, 1991, 1992 and 1993, instances of natural parasite were observed at levels of 56.80 %, 49.37 %, 38.63 % and 48.08 % , respectively (Kornoşor et al., 1992, 1994; Sertkaya &Kornoşor, 1994). At the same time *T. evanescens* was found as a egg parasitoid of *S. nonagrioides* in low percentage. Field and laboratory studies related this are in progress. In The Mediterranean Region, studies that aims to determined the natural enemies of Lepidopterous pests are also in progress. It has been found that *Meteorus oculatus*, *Cotesia ruficornis* (Holiday) (Hymenoptera, Braconidae), *Diadegma* spp, *Hyposoter deidymator* (Thbg) and *Camoplex* sp. (Hymenopters, Ichneumonidae) are involved effective parasitoids of *A. loreyi* and *P. unipuncta* (İkincisoy et al. 1994).

Apart from the previous studies in the Mediterranean Region, studies on the inundative release of *T. evanescens* and the biological control of *O. nubilalis* have started (Özpınar& Kornoşor, 1994b; Coşkuntuncel & Kornoşor, 1997).

Following the success of preliminary studies a small *T. evanescens* production unit was established in the University of Çukurova, Faculty of Agriculture, Department of Plant Production in 1990. Since 1994 inundative releases of *T. evanescens* against *O. nubilalis* have been done 50 ha maize production field of the Production and Research Farm of the Agriculture Faculty. Meanwhile, *T. evanescens* keeps the population of *Helicoverpa armigera* under pressure.

Studies on determination the resistance maize varieties against *O.nubilalis* and *S.nonagrioides*, also adaptation of Bt. maize seeds research are carried out in the Agriculture Research and Plant Protection Research Institutes.

Table 2. Natural enemies of maize pests in Turkey

Family	Species	Region
<i>Ostrinia nubilalis</i>		
Coccinellidae	<i>Coccinella septempunctata</i>	Mediterranean Black Sea Aegean
	<i>Coccinella undecimpunctata</i>	Mediterranean
	<i>Harmonia 14-punctata</i>	Black Sea
	<i>Hippodamia variegata</i>	Mediterranean Aegean
	<i>Propylae quatuordecimpunctata</i>	Aegean
Anthocoridae	<i>Orius niger</i>	Mediterranean Aegean
	<i>Orius minutus</i>	Mediterranean Aegean
Nabidae	<i>Nabis punctatus</i>	Mediterranean Aegean
Chrysopidae	<i>Anisochrysa carnea</i>	Mediterranean Aegean Black Sea
Erythraeidae	<i>Abrolophus</i> sp.	Mediterranean
Trichogrammatidae	<i>Trichogramma evanescens</i>	Black Sea Mediterranean Marmara
	<i>T. brassicae</i>	Aegean
	<i>T. maidis</i>	Marmara
Braconidae	<i>Bracon hebetor</i>	Black Sea Mediterranean
	<i>Iphialuse imposter</i>	Mediterranean
	<i>Cotesia ruficrus</i>	Mediterranean
	<i>Schizoprymnus obscurus</i>	Mediterranean
Ichneumonidae	<i>Diadegma crassicorn</i>	Mediterranean
	<i>Eriborus terebrans</i>	Black Sea
	<i>Pimpla spuria</i>	Black Sea
	<i>Phaegenes nigridentis</i>	Aegean
Pteromalidae	<i>Trichomolopsis genalis</i>	Aegean
Tachinidae	<i>Lydella thompsoni</i>	Black Sea Mediterranean Aegean
	<i>Pseudoperichaeta insidiosa</i>	Black Sea

<i>Sesamia nonagrioides</i>		
Scelionidae	<i>Platytelenomus busseolea</i>	Mediterranean Aegean
Braconidae	<i>Habrobracon hebetor</i>	Mediterranean Aegean
	<i>Cotesia ruficrus</i>	Mediterranean
Ichneumonidae	<i>Ichneumon sarcitorius</i>	Mediterranean Aegean
	<i>Pimpla spuria</i>	Mediterranean
	<i>Barichneumon</i> sp.	Mediterranean
	<i>Pimpla turionella</i>	Aegean
	<i>Syspasis rufinus</i>	Aegean
Pteromalidae	<i>Conomorium patulum</i>	Aegean
Trichogrammatidae	<i>Trichogramma evanescens</i>	Mediterranean
<i>Helicoverpa armigera</i>		
Coccinellidae	<i>Scymnus apetzoides</i>	Aegean
	<i>Scymnus interruptus</i>	Aegean
	<i>Symnus apetzi</i>	Aegean
Lygaeidae	<i>Piocoris lirudus</i>	Aegean
	<i>Piocoris erythrocephalus</i>	Aegean
	<i>Geocoris megacephalus</i>	Aegean
Nabidae	<i>Nabis pseudoferus</i>	Aegean
Anthocoridae	<i>Orius niger</i>	Aegean
	<i>Orius horwethis</i>	Aegean
Chrysopidae	<i>Anisochrysa carnea</i>	Mediterranean Aegean
Braconidae	<i>Apanteles</i> spp.	Aegean
	<i>Microplitis rufiventris</i>	Aegean
	<i>Habrobracon hebetor</i>	Aegean
Ichneumonidae	<i>Diadegma</i> sp.	Aegean
<i>Nezara viridula</i>		
Scelionidae	<i>Trisolcus</i> sp.	Aegean
Politidae	<i>Polistes gallicus</i>	Aegean
Tachinidae	<i>Escophasia crassipennis</i>	Aegean
<i>Rhopalosiphum</i> spp.		
Coccinellidae	<i>Coccinella septempunctata</i>	Aegean
	<i>Scymnus</i> spp.	Aegean
Anthocoridae	<i>Orius horwethis</i>	Aegean
	<i>Orius minitus</i>	Aegean
	<i>Orius niger</i>	Aegean

Miridae	<i>Deraecoris pallens</i>	Aegean
Chrysopidae	<i>Anisochrysa carnea</i>	Aegean
Syrphidae	<i>Episyrphus balteatus</i> <i>Ischiodon aegyptius</i>	Aegean Aegean
Nabidae	<i>Nabis pseudoferus</i>	Aegean
<i>Spodoptera exigua</i>		
Braconidae	<i>Microplitis tuberculifer</i> <i>Microplitis rufiventris</i> <i>Sinophorus xanthostomus</i> <i>Chelonus obscurator</i> <i>Meteorus ictericus</i>	Mediterranean Mediterranean Mediterranean Mediterranean Mediterranean
Ichneumonidae	<i>Hyposoter deidymator</i>	Mediterranean
<i>Acantholeucania loreyi</i> and <i>Pseudoletia unipuncta</i>		
Braconidae	<i>Cotesia ruficrus</i> <i>Meteorus oculatus</i> <i>Meteorus ictericus</i>	Mediterranean Mediterranean Mediterranean
<i>Agrotis ipsilon</i>		
Braconidae	<i>Meteorus rubens</i>	Mediterranean

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Building an Economical Refuge: Profitably Extending the Life of Bt Corn

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Planting Bt corn on large areas may lead to European corn borer (ECB), *Ostrinia nubilalis* Hübner, resistance to Bt corn. Scientists recommend planting non-Bt corn as an ECB refuge as part of a resistance management strategy. Different refuge configurations may impact farm profits differently. This paper analyzes the economics of alternative refuge configurations in Indiana.

Traditional agricultural production techniques are being altered by biotechnology. Currently, herbicide tolerant crops allow farmers to spray for weeds during the growing season without damaging yield potential. This leads to a more effective weed control program. Some examples of this technology include Roundup Ready soybeans or Liberty Link corn. Other crops can produce their own insecticide to kill particularly damaging insects. One example is Bt corn.

Bt corn has been genetically engineered to produce a protein, which becomes toxic to European corn borer (ECB) once ingested by the insect. ECB cause between \$1 and 2 billion in annual damages to corn in the USA, with most of the damage in regions with high ECB pressure. With this large potential loss in farm incomes, many farmers are quickly adopting Bt corn. In 1998, after only three years on the market, Bt corn accounted for about 19% of the total USA corn acreage.

Given that the economic benefits of Bt corn are potentially high in some areas, scientists worry that over-planting Bt corn may lead to rapid development of ECB resistance to Bt corn. To slow resistance development, scientists have recommended implementing a high-dose/refuge strategy. The idea behind this is that if the Bt is sufficiently strong to kill all ECB which possess a recessive Bt non-susceptible gene, as well as all ECB which are Bt susceptible, and if the refuge allows Bt susceptible ECB to live and mate with Bt resistant borers, then the overall ECB population will be smaller but the frequency of Bt susceptible genes in the population will remain high.

Entomologists and biologists researching the refuge strategy have recommended that 20-30% of corn acreage be planted to a non-Bt corn refuge that is not sprayed with ECB insecticides. If sprayed for ECB, a 40% refuge is recommended. Entomologists further recommend that the refuge should be planted near or within Bt fields.

Several potential configurations for the refuge are under consideration. For example, it may be planted in several strips throughout the field, in a block along the edge of the field, or as a perimeter around the field. An alternative to the non-Bt corn refuge strategy is to plant a crop that is more attractive to ECB than field corn. Recent research has shown that densely planted popcorn is much more attractive to ECB than regular corn. Using a popcorn-based refuge would allow for smaller refuges, but this type of refuge is expected to be so densely infested as to make harvesting uneconomical. Thus, this type of refuge is referred to as a sacrificial refuge.

This study looks at the net economic benefit of each potential refuge. The analysis uses detailed crop budgets to analyze potential differences in benefits or costs. The benefits are

measured by differences in corn yield between alternative refuge configurations. Yield differences may occur because some configurations are likely to provide better control of ECB. Recent research from Kansas has shown that a “halo of suppression” around Bt corn may occur as ECB population numbers are lowered in those areas. This so-called “halo effect” may differ between refuge configurations. This directly impacts corn yields.

Costs may differ because of different planting efficiencies associated with alternative configurations. For example, total planting time may increase due to the time required to adjust or change seed in the planter from Bt corn to the refuge seed. Increased time requirements may result in increased labor costs, for example. Total planting time for the farm may also increase, which may reduce yields on later planted fields due to a loss of timeliness. On average in Indiana, corn yields are estimated to fall 94.2 kg per hectare per day when delayed past May 10.

The general recommendations by many scientists studying the optimal refuge have been to plant a percentage of corn acreage to a non-Bt corn. However, given this percentage, farmers will choose to plant it in the most economical fashion.

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Key Words: *Ostrinia nubilalis* Hübner, Bt Corn, Transgenics, Refuge, Economics

Lepidopterous Pests and Their Natural Enemies on Maize in Çukurova Region*

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Abstract

This study was conducted on first and second crop maize in Çukurova Region between 1994-1998. Maize fields were surveyed once a week from seedling period to harvesting time in different locations. One hundred plants were randomly examined in a transect across each field. Fifteen parasites belong to different family were obtained from different lepidopterous pests. Among them, *Microplitis rufiventris*, *M. tuberculifer*, *Meteorus ictericus*, *Chelonus obscurator*, *Sinophorus xanthostomus* and *Hyposoter deidymator* were found to be larvae parasites of *Spodoptera exigua*. Natural parasitism was found to be higher on first crop maize than second crop maize. *Ichneumon sarcitorius*, *Pimpla spuria* and *Barichneumon* sp were determined pupa parasites of *Sesamia nonagrioides*. Larvae parasites of *S. exigua* and pupa parasites of *S. nonagrioides* were determined first time in this region and Turkey.

On the other hand 17 different predator species were also determined which might have an important role in supression of the population of the pests

Introduction

The area of maize production in Turkey has been increasing since 1982. Due to increasing maize culture, lepidopterous pests have gained important pest status. Of these, *Sesamia nonagrioides* Lef., *Ostrinia nubilalis* Hbn., *Agrotis ipsilon* Hbn., *Spodoptera exigua* Hbn., *Helicoverpa armigera* Dennis&Shiffer, *Acantholeucania loreyi* Dup., *Pseudoleatia unipuncta* Haw. have been determined as the most important pests on maize in the Southeast Mediterranean Region of Turkey (Kayapınar and Kornoşor, 1991).

In our climatic condition, insecticides are applied at least three times a season to control these pests. Knowledge of the role of natural enemies is a key factor for pest management systems (Cabello, 1989). Their identification and quantification of their effects on pest populations are essential for the development of biological control (Miller, 1983; DeBach, 1984).

The aim of this work was to determine the lepidopterous pests and their natural enemies on maize in the Southeast Mediterranean Region of Turkey.

Key words: Maize, Lepidopterous pests, natural enemies

Materials and Methods

This study was conducted on first and second crop maize in the Çukurova region during 1994-1998. Maize fields in different locations were surveyed once a week from the seedling period to harvesting time. One hundred plants were randomly examined in a transect across each field. Different stages of lepidopterous pests were collected and isolated in glass tubes and plastic vials. They were reared at 25 ± 1 °C, 65 ± 10 % relative humidity, 16h light daily

until adult or parasitoid emergence. All cultures were checked daily, and parasitized and non-parasitized eggs, larvae and pupae were recorded. In addition, adult predators were collected with an aspirator from 25 randomly chosen plants. Immature stages of predators were collected and brought to the laboratory in order to obtain their adults. Lepidopterous pests were identified in our laboratory whereas parasites and predators were kindly identified by related experts in Turkey.

Result and Discussion

In this study, a total of 15 parasites and 17 predators were identified from first and second crop maize during 1994-1998 (Table 1 and 2).

Lepidopterous pests and their parasites obtained on first and second crop maize in Cukurova region are shown in Table 1. Six larval parasites of *S. exigua* were found. These were *Microplitis rufiventris*, *M. tuberculifer*, *Meteorus ictericus*, *Chelonus obscurator*, belonging to Braconidae, and *Sinophorus xanthostomus* and *Hyposoter deidymator* belonging to the Ichneumonidae. *S. exigua* has a broad host range (Pearson, 1982). Even though it appears on first and second crop maize, it causes most economic damage on second crop maize, especially on crops sown at the beginning of July. Intensive insecticide application is needed in this situation. In 1996, larvae of *S. exigua* collected from the field were parasitized at the rate of 73.19 % and 18.22 % in first and second crop maize respectively. In 1997, these values were 90.24 % and 18.40 % respectively. Average parasitism on first and second crop maize was found as 35.06 % and 43.61 % in 1996 and 1997 respectively. Parasitism was low when sowing of second crop maize had been delayed due to late harvesting of melon, watermelon and onion because of insecticide application made against *S. exigua*. Data collected in two years showed that parasitization rate of *S. exigua* was higher in first crop than those in second crop.

Abundance of parasites were determined in 1997. The most common parasite was *H. deidymator* on first crop maize, with a frequency of 42.67 %. *Microplitis* spp. were obtained mainly on the second crop maize with a frequency of 30.57 % but *Meteorus icterus* and *Chelonus obscurator* were obtained from larvae collected on both first and second crop maize, with abundance values were 21.65 % and 4.45 % respectively. The abundance value of *S. xanthostomus* was found to be 0.63 %. Ruberson et al. (1993) reported that the parasitism rate of *S. exigua* collected from cotton fields in USA were 46.8 % and 40.2 % in 1992 and 1993 respectively, and *C. marginiventris* was found as predominant species. Cabello (1989) reported that *S. xanthostomus* and *H. deidymator* had a broad host range, and parasitized *S. exigua* larvae on different crops.

Three larva parasites, *M. oculatus*, *M. icterus* and *Cotesia ruficrus*, were obtained from *A. loreyi* that caused damage on first and second crop maize and determined as very common in the Cukurova Region. *Meteorus* sp. *Cotesia ruficrus*, *H. deidymator*, *Diedegma* sp. and *Campoplex* sp. were detected as larvae parasites of *A. loreyi* by İkincisoý et al. (1994), and 20.65 and 52.57 % total collected larvae of *A. loreyi* were parasitized by these parasites in 1992 and 1993 respectively. Of these, *C. ruficrus* was recorded as the most promising. *C. ruficrus*, a larval parasite of many species of Noctuidae (Cabello, 1989), has been released against *Mythimna seperata* in New Zealand, resulting in a parasitization rate of 50-55 % and a reduction of 54-69 % in feeding damage (Hill, 1988).

Platytelenomus busseolae and *Trichogramma evanescens* were found as egg parasitoids of *S. nonagrioides*. *Habrobracon hebetor* and *Cotesia ruficrus* were found as larvae parasites, and *Ichneumon sarcitorius*, *Pimpla spuria* and *Brachneumon* sp. as pupal

parasites in Cukurova Region. The natural effectiveness of *P. busseolae* has been known since 1990 and can be used in biological control agent for *S. nonagrioides*. High parasitism rates were recorded where insecticide application was limited or none (Kornosor et. al. 1994, Sertkaya, 1999). *H. hebetor* and *C. ruficrus* were found to be common, and parasitized *O. nubilalis* and *A. loreyi* in the region (Kayapınar, 1991; İkinçisoğul et. al., 1994). The presence of the pupal parasitoid of *S. nonagrioides* was recorded both in the region and Mediterranean countries for the first time by Sertkaya (1999). The parasitoids were obtained from the overwintered generation of *S. nonagrioides*. Although the main host of *Trichogramma evanescens* was *O. nubilalis*, it also parasitized *S. nonagrioides* at a low rate (Kornosor et. al. 1992; 1994) and biological features of the parasitoid on *S. nonagrioides* were investigated by Bayram (1999).

In this study, *T. evanescens* was found as an egg parasitoid of *O. nubilalis* while *H. hebetor* was a larval parasitoid of *O. nubilalis*. *T. evanescens* was the most common natural enemy of *O. nubilalis* in the region, and natural parasitism of *O. nubilalis* by *T. evanescens* ranged from 27 to 51 % (Kayapınar, 1991). It was found to be very effective if it was released against *O. nubilalis* (Özpinar and Kornosor, 1995; Coskuntuncel and Kornosor, 1996). The natural efficiency of *H. hebetor* on *O. nubilalis* larvae was 15-30 % (Kayapınar, 1991).

Agrotis ipsilon causes economic damage on the first crop maize in the region unless control measures are taken. *Meteorus rubens* was determined as a larval parasite of *A. ipsilon*, and obtained from larvae of *A. ipsilon* on the first crop maize. *M. rufiventris* was obtained from larvae of *S. exigua*, and it was found that it could parasitize *Helicoverpa armigera* larvae in maize agroecosystem.

On the other hand, 17 predators species belong to different families were represented in the maize ecosystem (Table 2). Most of them are polyphagous and feed on different prey in many agroecosystems such as cotton, citrus, vegetable etc. (Atakan, 1993; Zeren, 1989). *C. septempunctata*, *Orius niger*, *O. minutus* and *Chrysoperla carnea* were found to feed on eggs and early instars of *O. nubilalis* (Kayapınar, 1991).

In conclusion, a total of 15 parasites and 17 predators species were obtained from different lepidopterous pests on first and second crop maize in the Çukurova Region. Traditional insecticide applications are harmful to beneficial insects in Çukurova region. The population level of these pests is usually below the economic injury threshold, so insecticide applications are not essential. Insecticide use on first crop maize must be avoided. It is concluded that insecticide applications will not be needed if second crop maize is sown at the optimum date such as the beginning of June. Furthermore, *Trichogramma* releases are realized.

Table 1. Parasites of lepidopterous pests on maize in 1994-1998.

Parasitoids species	Host	Stage at sampling
<i>Meteorus rubens</i>	<i>Agrotis ipsilon</i>	Larvae
<i>Microplitis tuberculifer</i>	<i>Spodoptera exigua</i>	Larvae
<i>Microplitis rufiventris</i>	<i>Spodoptera exigua</i>	Larvae
<i>Sinophorus xanthostomus</i>	<i>Spodoptera exigua</i>	Larvae
<i>Chelonus obscurator</i>	<i>Spodoptera exigua</i>	Larvae
<i>Meteorus ictericus</i>	<i>Spodoptera exigua</i>	Larvae
<i>Hyposoter deidymator</i>	<i>Spodoptera exigua</i>	Larvae
<i>Meteorus oculatus</i>	<i>A. loreyi</i> , <i>P. unipuncta</i>	Larvae
<i>Meteorus ictericus</i>	<i>A. loreyi</i> , <i>P. unipuncta</i>	Larvae
<i>Cotesia ruficrus</i>	<i>A. loreyi</i> , <i>P. unipuncta</i>	Larvae
<i>Platytelenomus busseolae</i>	<i>Sesamia nonagrioides</i>	Egg
<i>Trichogramma evanescens</i>	<i>Sesamia nonagrioides</i>	Egg
<i>Habrabracon hebetor</i>	<i>Sesamia nonagrioides</i>	Larvae
<i>Cotesia ruficrus</i>	<i>Sesamia nonagrioides</i>	Larvae
<i>Ichneumon sarcitorius</i>	<i>Sesamia nonagrioides</i>	Pupae
<i>Pimpla spuria</i>	<i>Sesamia nonagrioides</i>	Pupae
<i>Barichneumon sp</i>	<i>Sesamia nonagrioides</i>	Pupae
<i>Trichogramma evanescens</i>	<i>Ostrinia nubilalis</i>	Egg
<i>Habrabracon hebetor</i>	<i>Ostrinia nubilalis</i>	Larvae

Table 2. Predator species which were determined on maize in 1994-1998.

Predator species	Order	Family
<i>Adonia variegata</i>	Coleoptera	Coccinellidae
<i>Coccinella septempunctata</i>	"	"
<i>Nephus nigricans</i>	"	"
<i>Scymnus quadricuttatus</i>	"	"
<i>Scymnus levaillanti</i>	"	"
<i>Scymnus pallipediformis</i>	"	"
<i>Scymnus rubromaculatus</i>	"	"
<i>Scymnus subvillosus</i>	"	"
<i>Serangium parcesetosum</i>	"	"
<i>Stethorus gilvifrons</i>	"	"
<i>Synharmonia conglobata</i>	"	"
<i>Chrysoperla carnea</i>	Neuroptera	Chrysopidae
<i>Orius niger</i>	Heteroptera	Anthocoridae
<i>Orius minutus</i>	"	"
<i>Deraeocoris pallens</i>	"	Miridae
<i>Nabis punctatus</i>	"	Nabidae
<i>Piocoris erythrocephalus</i>	"	Lygaeidae

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Results on IWGO Maize Inbred Lines Tolerance, Their Productivity and Yield Response to the ECB *Ostrinia nubilalis* Hbn. in Yugoslavia in the Period 1981 - 1994

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Abstract

The resistance of 359 maize genotypes was evaluated by artificially infesting them with egg masses of *Ostrinia nubilalis* Hbn., while yield and yield response to ECB damage were determined under chemical control. A total of 167 inbreds with average yield of above 25 g plant⁻¹ and yield reduction of below 50% was selected and classified. Twenty inbreds, satisfying the criterion of resistance/tolerance, based on the general damage rating (score up to 3.5) and yield response (reduction to 20%), were considered promising for the further practical breeding programme. The inter-relationship of resistance parameters and their accord with yield response were discussed. Significant differences in inbred responses over investigation years were determined. Their ratio amounted to 1:2 (2.87:5.98) in 1982 and 1986 for general damage rating, 1:5 (2.81:13.59plant⁻¹) in 1982 and 1987 for the number of larvae, 1:3 (25.4:76.1g plant⁻¹) in 1989 and 1986 for yield and 1:5 (11.7:58.3%) in 1982 and 1987 for yield reduction.

Key words: European corn Borer (*Ostrinia nubilalis* Hbn.), inbred lines, resistance, productivity, resistance.

Introduction

Many maize inbred lines were tested as part of the common IWGO investigation for resistance to the European Corn Borer (ECB) (*Ostrinia nubilalis*) in 14 countries of Europe, North America and Asia, and to the Asian Corn Borer (*Ostrinia furnacalis*) in China. Only 167 out of 359 inbreds were selected and analysed, as their average yield was over 25 g plant⁻¹ i.e. over 1.5 t ha⁻¹, while yield reduction was below 50%. The remaining 192 inbreds had low adaptability for the Yugoslav Corn Belt, or were observed in bad season and/or years favourable for ECB.

The aim of this choice of lines was to confirm the hypothesis that genotypes of higher genetic yield potential can tolerate infestation with 30-50 eggs plant⁻¹ without significant yield reduction.

Materials and Methods

Insect test material of ECB was collected from a wild population in Zabojnica, at about 300 m above sea level in central Serbia. This population is well known for a satisfactory level of larval viability (Bača and Hadžistević, 1986).

Maize inbreds tested in Yugoslavia during the period 1981 – 1994 were comprised of 268 European, 65 US and Canadian, 24 Chinese and 2 inbreds from Egypt. All inbreds were exchanged among countries and biannually tested by the common IWGO procedure, in Zemun Polje, Yugoslavia in the years 1981 to 1994.

The trials were set up in two variants according to the IWGO procedure, with three replications and five plants per replication:

1 - the variant "A" - plant artificial infestation with four egg masses (30-50 eggs plant⁻¹) for testing of their reaction to larval damage,

2 - the variant "B" - plant chemical protection to observe data on yield of borer free plants.

The following parameters were observed:

- Number of days from germination to silking,
- General damage rating at the harvest time, the 1-10 scale (1 tolerant, 10 susceptible (Hadžistević, 1968, Bača, 1988).
- Grain yield in grams (shelled grain at 14% moisture).
- The number of larvae, holes and tunnels and tunnel lengths by plant dissection.

The results were summarised for each inbred line over all years. Annual data were used as a replication. Average data for two successive years in which the same set of inbred lines was tested were categorised according to nine criteria. Particular importance was given to the general damage rating (GD), number of larvae, grain productivity and yield response. Multiple correlation was calculated using the annual trial average data.

Results and Discussion

Average results, summarised over all investigation years, for 369 inbred lines tested in Yugoslavia over 14 years are presented in Tables 1-6.

Table 1. Annual Average Test Results of All IWGO Inbred Lines, Zemun Polje Yugoslavia, 1981 – 1994

Year	N° of Inbr	Days Silk.	GD	Plant dissection				Yield plant ⁻¹		Yield Red.
				N° of Larva	N° of Holes	N° of Tunnels	Length cm	"A"	"B"	
		(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)
1981	58	60.0	3.06	3.34	6.73	4.00	15.83	50.5	66.3	23.8
1982	58	61.1	4.50	2.81	2.66	2.48	9.37	58.1	65.8	11.7
1983	64	62.1	2.87	3.09	5.12	3.17	19.23	34.0	45.8	25.8
1984	75	67.2	3.56	8.76	10.31	5.67	35.54	29.3	67.0	56.3
1985	54	61.8	3.74	11.44	11.10	6.27	45.09	22.8	49.7	54.1
1986	65	67.2	5.98	9.09	11.62	7.16	43.90	34.9	71.6	51.4
1987	44	66.4	3.90	13.59	14.87	6.06	46.55	23.0	55.1	58.3
1988	44	65.4	4.30	8.81	12.44	5.37	38.71	30.9	49.0	36.9
1989	47	74.9	4.86	7.08	11.77	4.76	30.62	12.8	25.4	49.6
1990	51	67.5	5.86	10.24	16.94	5.09	34.58	18.4	31.6	41.8
1991	44	77.0	4.38	8.03	17.10	5.77	36.32	24.3	50.9	52.3
1992	44	66.2	3.18	3.63	10.60	4.42	32.60	41.8	59.2	29.4
1993	29	70.9	3.43	3.38	5.18	2.80	15.30	23.7	27.5	13.8
1994	31	67.5	4.64	6.64	12.11	4.89	31.60	25.0	55.0	54.5
\bar{X}	51	66.8	4.16	7.15	10.61	4.85	31.09	30.7	51.4	40.3

Legend: (1) Number of days to silking (2) General Damage Rating (3) Number of Larvae per Plant (4) Number of Holes per Plant (5) Number of Tunnels per Plant (6) Length of Tunnels (cm plant⁻¹) (7) Grain Yield Production (g year⁻¹) in "A" (8) Grain Yield Production (g year⁻¹) in "B" (9) Grain Yield Reduction in (% year⁻¹)

Table 2. Annual Average Test Results on IWGO Inbred Lines According to Categories of Resistance in Zemun Polje, Yugoslavia, 1981 – 1994

Res. Category According	№ of Rep.	Days Silk.	GD	Plant dissection				Yield g/pl.		Yield Red.
				№ of Larva	№ of Holes	№ of Tunnels	Length cm	"A"	"B"	
Number of days to silking										
< 65 days	4	61.2	3.54	5.22	6.40	3.98	21.12	41.4	56.9	27.2
65-70 days	7	66.8	4.49	8.68	12.70	5.52	37.64	29.0	55.5	46.9
70-75 days	2	72.9	4.14	5.23	8.48	3.78	22.96	18.2	26.4	31.1
> 75 days	1	77.0	4.38	8.03	17.10	5.77	36.32	24.3	50.9	52.3
Correlation coefficient r =			0.341	0.181	0.537	0.23	0.250	-0.65	-0.49	0.372
General Damage Rating										
2.1-3.5	4	64.8	3.14	3.41	6.91	3.60	20.74	37.5	49.7	24.5
3.6-5.0	8	67.7	4.24	8.40	11.54	5.16	34.22	28.3	52.2	45.8
> 5.0	2	67.4	5.92	9.66	14.28	6.12	39.24	26.6	51.6	48.4
Correlation coefficient r =				0.426	0.500	0.448	0.362	-0.30	-0.10	0.36
Number of Larvae per Plant										
< 5.0	5	64.1	3.41	3.29	6.06	3.37	18.47	41.6	52.9	21.4
5.1-10.0	6	69.9	4.62	8.07	12.56	5.60	36.12	26.2	53.2	50.8
10-15	3	65.2	4.50	11.76	14.30	5.81	42.07	21.4	45.5	53.0
Correlation coefficient r =					0.767	0.842	0.890	-0.60	-0.02	0.827

Table 2. continued

Number of Holes per Plant										
< 5.0	1	61.1	4.50	2.81	2.66	2.48	9.37	58.1	65.8	11.7
5.0-10.0	3	64.3	3.12	3.34	5.68	3.32	16.79	36.1	46.5	22.4
10-15	8	67.1	4.27	8.63	11.85	5.58	38.08	27.6	54.0	48.9
> 15.0	2	72.2	5.12	9.14	17.02	5.43	35.45	21.4	41.2	48.1
Correlation coefficient r =					0.761		0.803	-0.65	-0.17	0.778
Number of Tunnels per Plant										
< 5.0	7	67.5	4.96	9.99	13.48	5.91	40.10	26.2	53.6	51.1
5.0-10.0	7	66.1	3.79	4.31	7.74	3.79	22.08	35.1	49.3	28.8
Correlation coefficient r =							0.943	-0.44	0.25	0.880
Length of Tunnels (cm plant ⁻¹)										
< 10.0 cm	1	61.1	4.50	2.81	2.66	2.48	9.37	58.1	65.8	11.7
10-20	3	64.3	3.12	3.34	5.68	3.32	16.79	36.1	46.5	22.4
30-40	7	69.4	4.40	7.60	13.04	5.14	34.28	26.1	48.3	46.0
> 40.0 cm	3	65.1	4.54	11.37	12.53	6.50	45.18	26.9	58.8	54.2
Correlation coefficient r =								-0.56	0.09	0.869

General damage ratings (GD) ranged over years from 2.87 (1983) to 5.98 (1986) with an average of 4.16. Comparing the GD with the other criteria, the highest correlation was detected between GD and the number of holes ($r = 0.500$), then GD and the number of larvae ($r = 0.426$) and GD and the number of tunnels ($r = 0.448$). Correlation coefficients between GD and the other criteria were close to a significant level (0.514 at 95% and 0.641 at 99% level of probability), but were never significant. The absence of significant correlation between the general damage rating and other criteria indicates that there were certain significant differences in the nature of damage caused by ECB larvae over the tested years. These differences were caused by different climatological conditions, especially variable amount of rainfall in the growing season, as well as higher temperatures in dry years. *Lydella thompsoni* Hrt. (Bača et al., 1997), as beneficial biotic organism, led to the decrease of the larval level

According to the 4-year average results (1981, 1983, 1992 and 1993) GD ranged between 2.1 and 3.5. Therefore, the majority of inbreds evaluated in the above-mentioned years were designated as resistant/tolerant. In 1986 and 1990, the majority of inbreds were evaluated as very susceptible (over 5.0). For the other 8 years, the average GD score ranged between 3.5 and 5.0.

Table 3. Productivity and Relationship between Plant Damage and Yield Response in Zemun Polje, Yugoslavia, 1981 – 1994

Resistance Category According	№ of Rep.	Days Silk.	GD	Plant dissection				Yield g plant ⁻¹		Yield Red.
				№ of Larvae	№ of Holes	№ of Tunn	Length cm	"A"	"B"	
Grain Yield Production (g year ⁻¹)										
20-30	2	72.9	4.14	5.23	8.48	3.78	22.96	18.2	26.4	31.1
31-40	1	67.5	5.86	10.24	16.94	5.09	34.58	18.4	31.6	41.8
41-50	3	63.1	3.64	7.78	9.55	4.94	33.34	29.2	48.2	39.4
51-60	4	69.3	4.02	7.97	13.67	5.28	27.62	28.5	55.0	48.2
61-70	3	62.8	3.71	5.04	6.57	4.05	20.25	46.0	66.4	30.7
>70	1	67.2	5.98	9.09	11.62	7.16	43.90	34.9	71.6	51.4
Grain Yield Reduction (% year ⁻¹)										
10-20	2	66.0	3.96	3.10	3.92	2.64	12.34	40.9	46.6	12.2
21-30%	3	62.8	3.04	3.42	7.48	3.86	22.55	42.1	57.1	26.3
31-40%	1	65.4	4.30	8.81	12.44	5.37	38.71	30.9	49.0	36.9
41-50%	2	71.2	5.36	8.66	14.36	4.92	32.60	15.6	28.5	45.3
> 50 %	6	67.8	4.37	9.59	12.85	5.97	39.83	26.6	58.2	54.3
Correlat. coeff. r =		0.37	0.36	0.82	0.77	0.88	0.869	-0.63	0.09	

The duration of the vegetation from planting to silking varied over years and ranged from 60 to 77 days in 1981 and 1991 respectively. The range was 17 days, and it is one of the reasons for different stages of organogenesis of maize plants during artificial infestation with egg masses. A positive correlation ($r=0.537$), significant at 95% probability level, was detected between the number of days to silking and the number of holes. A negative correlation was found between the number of days to silking and yield obtained in variant "A" ($r= - 0.652$). A greater number of days to silking and longer duration of certain stages of organogenesis in maize occurs with greater amount of precipitation. Precipitation favours

maize and the survival of ECB larvae, which cause damage by tunnelling into the stalk, in proportion to the number of surviving 4th instar larvae.

Data on stalk dissection were highly significantly correlated - the number of larvae with the number of holes, the number of tunnels and the tunnel length, as well as with grain yield reduction. Data variability over investigation years, as well as the two-year means for each of tested inbreds, were very pronounced. Differences over investigation years ranged from 2.81 larvae plant⁻¹ (or only 5.6% survivals) in 1982 to 13.59 larvae plant⁻¹ (or 27.2% survivals to the beginning of diapausing, in relation to the number of egg masses artificially infested).

The differences in the results obtained based on all nine recorded parameters would have been less if all the inbreds had been tested in two successive years. Table 4 presents data on the basic abiotic and biotic agents significantly affecting maize and ECB.

Table 4. Some Abiotic and Biotic agents Influencing ECB Population, Larval Harmfulness, Plant Productivity and Yield Response, Zemun Polje, Yugoslavia, 1981 – 1994

Year	∑ in mm	∑ HU	1 st Generation Moth		2 nd Generation Moth		% of <i>L.t.</i>
			№	%	№	%	
	(1)	(2)	(3)	(4)	(5)	(6)	(7)
1981	301	1464	377	23.7	1215	76.3	12,20
1982	307	1617	447	44.2	564	55.8	12,20
1983	297	1517	1341	47.7	1470	52.3	1,84
1984	288	1446	188	21.6	681	78.4	1,03
1985	308	1474	318	16.0	1665	84.0	0,23
1986	227	1385	377	9.2	3703	90.8	0,80
1987	353	1572	369	4.4	7978	95.6	0,01
1988	189	1579	653	10.3	5704	89.7	0,93
1989	311	1375	485	18.2	2173	81.8	2,26
1990	164	1480	607	23.4	1990	76.6	1,96
1991	328	1494	278	10.2	2452	89.8	0,74
1992	177	1889	849	20.6	3271	79.4	0,69
1993	194	1792	640	24.0	2029	76.0	1,60
1994	309	1761	479	10.5	4245	89.5	1,22
\bar{X}	268	1560	529	15.1	2976	84.9	2.69

Legend:

- (1) Amount of precipitation in mm; May – September.
- (2) Heat Units >10°C; May – September.
- (3 & 4) Number and percentage of the First Generation of ECB Moth in Light Trap.
- (5 & 6) Number and percentage of the Second Generation of ECB Moth in Light Trap
- (7) Percentage parasitisation of ECB Larvae by *Lydella thompsoni* Hrt.

Amounts and distribution of precipitation varied over the period May-September and ranged from 164 mm to 353 mm in 1990 and 1997 respectively. At the same time, the accumulated effective temperatures varied from 1375°C (1989) and 1385°C (1986) to 1889°C (1992).

The number and percent distribution of moths caught in a light trap, as parameters of the level and structure of the natural population, were as follows:

- A range of 188 to 1341 in 1984 and 1983, respectively, in a 1:7 ratio; percent distribution of the ECB first generation varied from 4.4% to 44.2% in 1987 and 1982, respectively. At the same time weather conditions were exceedingly favourable for the ECB second generation in 1987 and extremely unfavourable in 1982. Hence, 369 and 447 moths were trapped in 1987 and 1982, respectively.

The number of the ECB second generation varied from 546 (1982) and 681 (1984) to 5704 (1988) and as many as 7978 in 1987. The difference between the highest and lowest number amounted to 1:14.

Furthermore, summer parasitization of ECB by *Lydella thompsoni* was detected. Low larval parasitisation, determined in the majority of years, ranged from 0.01% in 1987 to 2.26% in 1989. The highest number of larvae per plant (13.59) was on average found in 1987. Moreover, the greatest loss of yield (58.3%) was recorded in 1987. Parasitisation of 2.26% and an average number of larvae per plant of 7.08 were recorded in 1989. At the same time, parasitisation of larvae amounted to 12.2% in two out of 14 investigation years (1981 and 1982). The average number of larvae in these two years, estimated by plant dissection, amounted to 3.54 and 2.81 respectively.

Table 5. Number of Inbreds according to their Productivity and Percentage of Yield Reduction in Zemun Polje, Yugoslavia during 1981 – 1994

DEGREE OF TOLERANCE over the Percentage of Yield Reduction	Grouping Interval of Productivity					
	Number & Percentage	Middle 25-50 g	High 51-75 g	Very high 76-100g	Extremely high >100 g	Sum
I Up to 5% Very Tolerant	№	10	9	1	0	20
	%	5.99	5.39	0.60	0.00	11.98
II 5.1 – 10.0 % Tolerant	№	5	0	1	3	9
	%	2.99	0.00	0.60	1.80	5.39
III 10.1 – 20.0 % Moderately Susceptible	№	6	4	2	4	16
	%	3.59	2.40	1.20	2.40	9.58
IV 20.1 – 40.0 % Susceptible	№	25	32	14	5	73
	%	14.97	19.16	8.38	2.99	43.71
V Over 40.0 % Very Susceptible	№	15	17	12	5	49
	%	8.98	10.18	7.19	2.99	29.34
Total Number	№	61	62	30	14	167
Total percentage	%	36.53	37.13	17.96	8.38	100.

Based on two-year means on the level and response of yield, only 20 out of 167 selected inbreds (Table 5) were very tolerant, 9 were tolerant and 16 were moderately tolerant. Yield reduction for the first 20 inbreds was up to 5%, for the following 9 was 5.1-10% and for the last 16 ranged from 10.1 to 20.0%. Table 6 presents 20 out of 45 inbreds with satisfactory tolerance. The complete results obtained in Yugoslavia will be issued soon together with results gained in other IWGO member countries on the behalf of all participants.

Table 6. The List of Promising IWGO Inbred Lines Tested in Zemun Polje, Yugoslavia during 1981-1994

Promising Inbreds	Days Silking	Gen. Dam	Plant dissection				Yield g/pl.		Yield Red.
			No of Larva	No of Holes	No of Tun n.	Lenght cm/pl.	"A"	"B"	
CH 581-13	73	2.0	7.2	9.0	4.9	29.2	30	31	3.2
SIPING 404	68	2.7	3.0	4.6	3.3	11.5	47	37	-27.0
TVA 2008-3	58	3.5	3.0	4.2	3.0	10.5	41	44	7.7
F 676	80	2.2	6.8	12.7	5.0	33.4	25	26	1.2
MR 17	54	3.5	4.0	5.2	4.2	17.2	27	24	-10.8
B 85	63	3.0	2.5	3.8	3.0	11.2	43	48	9.7
OH 43	52	3.3	2.4	2.8	2.6	8.6	51	47	-7.6
A 660	68	2.2	5.6	7.6	4.4	24.6	29	36	19.4
MR 11	61	3.0	2.6	4.1	2.9	11.8	61	63	2.2
HMV 410	71	3.0	5.1	12.0	4.0	28.5	46	53	14.2
LO 543	60	2.6	3.0	4.4	2.8	11.9	56	54	-3.0
EA 3101	76	2.4	10.6	9.9	6.5	48.8	63	57	-10.5
CM 105	57	2.7	4.0	6.2	3.4	11.2	74	73	-2.2
CH 591-36	64	2.0	2.8	3.8	3.0	11.1	92	93	1.7
ZPL 1189	71	2.0	2.6	2.8	2.6	9.0	88	82	-7.3
TSPT	72	2.4	3.9	4.1	3.2	9.6	113	135	16.3
KY 301	60	2.7	4.0	4.6	3.4	20.0	131	148	11.5
BSAA	68	3.4	12.4	12.9	5.8	43.8	94	102	7.8
ZPL 12221	68	2.4	2.6	3.8	3.0	10.0	109	123	11.4
BGAT 558	56	3.5	4.0	3.8	3.0	11.0	125	137	8.8

Conclusions

Based on the 14-year investigations on resistance/tolerance of 359 inbred lines to European Corn Borer (*Ostrinia nubilalis* Hbn.) it can be concluded that great diversity of obtained results over investigation years emerges from the phenomenon of maize resistance to this pest.

Considering all the three criteria: tolerance, yield and yield reduction, 20 IWGO inbreds can be classified into two groups according to their general combining ability and can be used in the breeding programmes.

Joint studies, mainly based on resistance tests of IWGO inbreds and development of synthetic populations derived from inbreds with confirmed resistance to ECB, were completed up to 1994.

Increased demand for mutual investigations on maize pests in regions of the north hemisphere, especially in Europe, appeared as a result of development of transgenic hybrids with the incorporated "Bt" gene in the USA and the occurrence of WCR in Yugoslavia and Europe.

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Data Obtained at Fundulea on the Maize Resistance to the European Corn Borer (*Ostrinia nubilalis*)

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Abstract

In Romania the most important yield losses induced by the European Corn Borer are due to attack to maize stalks. This report represents going forward with research started at Fundulea in 1975 to develop maize inbred lines resistant to this pest, also including suitable agronomical features, evolved in the recent experimental years.

After several previous cycles of crossing and trials for resistance to the insect, maize inbred lines have been obtained with marked resistance and certain agronomical characters. By crossing these inbred lines with maize forms showing acceptable degree of agronomic performance, new hybrid combinations have been achieved. Among these, more than 500 lines have been exposed to self-pollination every year, under artificial infestation conditions (10 plants for each line with 5-6 egg-masses/plant), retaining only the lines showing reduced attack level and acceptable agronomic traits. Some inbred lines having representative attack values have been infested with 20 egg-masses/plant. The attack was rated according to length of stalk tunnels (cms) and the number of larvae/plant, as established in autumn, by splitting plant stems.

It was found that after 20 years of investigations on maize resistance to *O. nubilalis*, inbred lines have been obtained, which constantly demonstrated reduced attack by the borer. Taking into consideration data in the last 3 experimental years, when the attack of the borer was very low (approximately the same as that in case of natural infestation), it was shown that the attack by the borer was 14 times lower in the inbred resistant lines, as compared to that of very susceptible lines. Some lines demonstrated a certain resistance degree, even to a very severe outbreak. By crossing some of these inbred lines, tolerant hybrids to the attack by the borer have been obtained. Though one of these hybrids had reduced attack level, nevertheless in Monsanto Dekalb 512 Bt hybrid no attack by the borer occurred.

Key words: *Ostrinia nubilalis*, European Corn Borer, maize inbred line, resistance, egg-masses.

Introduction

Its wide-spread in Romania and significant damages caused by *Ostrinia nubilalis*, which rise up to 60% from grain yield under favourable conditions (Paulian et al., 1962), determined start of numerous researches, of which those regarding its prevention and control are not less important. In this sense, particular attention was paid to investigations on maize resistance to the attack of this pest.

Various investigations on maize resistance to maize borer, approached in the United States of America more than 70 years ago, resulted in a series of syntheses, some of these being quoted by Hudon (1989).

In Romania the investigations on maize resistance to this pest have been approached mainly in the recent years, of which are worth to be mentioned: Mustea (1973), Mustea and

Greco (1989), Bărbulescu (1981,1993), Bărbulescu and Cosmin (1978, 1987,1995 and 1997).

The present work is ongoing with research beginning in 1975, referring to obtaining maize inbred lines resistant to the borer attack, outlining the achievements in the last 4 experimental years.

Material and Method

Experiments have been performed at the Research Institute for Cereals and Industrial Crops-Fundulea during 1995-1998 and are ongoing with researches initiated in 1975, carried out within several experimental cycles. Each year more than 500 inbred lines showing reduced attack level and acceptable agronomic features, derived from hybrid combinations between the most valuable lines from the previous experimental cycle, have been tested and submitted to selection. From each line, 8-12 plants have been infested with 5-6 egg-masses. In some inbred lines with representative attack values two seed times have been trialled, i.e. two infestation times with 20 egg-masses/plant, in order to check their reaction under different climate conditions and heavy outbreak.

In order to estimate the borer attack, prior to harvest plants have been splitted, recording length of stalk and ear peduncle tunnels, as well as the number of larvae/plant. In view to go on with experiments, the ears agronomically suitable only from those inbred lines in which the attack values recorded represented maximum 50% of difference between the medium and minimum value of the respective experiment have been harvested. In many cases, ears have been also harvested from plants with reduced attack values, even when the other plants from the inbred line had higher attack values.

Results and Discussion

Climate conditions, generally unfavourable to the borer during infestation and just after this, determined not too evident attacks (table 1), which sometimes made difficult evaluation of reaction in the inbred lines.

Table 1. Representative values of attack induced by *O. nubilalis* in each experiment for resistance

Value of attack	Length of tunnels cm/plant				No. of larvae/plant			
	1995	1996	1997	1998	1995	1996	1997	1998
minimum	8	1	1	0,4	0,9	0,3	0,2	0
maximum	73	51	41	17,1	14,1	9,5	7,4	4,3
average	32	18	11	6,1	5,1	3,9	2,1	1,3

Referring to inbred lines behaviour, it is to be shown that, due to segregation, in many situations a marked attack was induced, this imposing elimination of susceptible plants and lines. In table 2 maize inbred lines are presented, which had reduced attack values every years, representing maximum 50% from difference between medium and minimum value of experiment in the respective year, being rated as resistant or medium resistant.

It is worth mentioning that for each line the different code number in the two years of tests represent the same line. The 19 inbred lines in 1997 derived from the 14 lines in 1996.

Looking for behaviour of some inbred lines with a certain degree of resistance to *O. nubilalis*, as compared to some very susceptible lines under heavy infestation with 20 egg-masses/plant (table 3), it resulted that the few inbred lines maintained the resistance character in all experimental years, irrespective of seeding time and of infestation. Nevertheless, it is to be added that in all maize lines under experiment having a certain degree of resistance, plants supported well the attack, remaining green until the autumn, at the end of vegetation, and very few plants lodged, while in the susceptible lines the attack was very heavy, the plants nearly totally dried long before the resistant lines. Due to plant lodging and drying, most of larvae participating to the attack disappeared, migrated on other plants, or were destroyed by birds.

Taking into account the maize inbred lines with extreme values of reaction to maize borer in 1995 and 1998, and namely resistant and very susceptible lines (tables 4-5), it is noted that when the attack was very low (the case of 1998), the difference between the two categories of lines was very obvious.

When comparing these data, as well as those in table 6, it resulted that, in the years 1996-1998, when the attack of the borer was very low (approximately the same as that of natural infestation), in the resistant inbred lines the attack values were about 14 times lower than in the very susceptible inbred lines.

From agronomical standpoint, it is to stress that inbred resistant lines have been obtained, with high combinative value and superior agronomic traits.

Among the maize hybrids developed from these inbred lines HSF 1071-96 has been remarked, which in both experimental years showed the lowest attack values, being rated as resistant to the attack by this pest (table 7). It is worth mentioning that HSF 1071-96, though it had low attack, the hybrid from Monsanto Dekalb 512 Bt, including the Bt gene, showed but sporadically traces of attack by *O. nubilalis*.

Conclusions

-After more than 20 years of research on maize breeding for resistance to *O. nubilalis*, inbred lines have been obtained, possessing a certain degree of resistance, having high combinative value and superior agronomic traits.

-Among the maize hybrids developed from inbred lines resistant to the maize borer, the hybrid HSF 1071-96 has been remarked for reduced attack.

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Table 2. Maize inbred lines resistant to *Ostrinia nubilalis* attack

Line	Cavities				Larvae			
	Length cm/plant		Score		No. of specimens/plant		Score	
	1996	1997	1996	1997	1996	1997	1996	1997
1996								
10/96	7	6	MR	MR	0,8	0,9	R	MR
14/96	8	4	MR	MR	2,0	0,9	MR	MR
35/96	7	4	MR	MR	1,4	0,8	MR	MR
37/96	2	6	R	MR	0,6	0,9	R	MR
		5		MR		1,0		MR
38/96	1	3	R	R	0,3	0,7	R	R
		4		MR		1,0		MR
		3		R		0,4		R
63/96	5	6	R	MR	1,3	0,9	MR	MR
65/96	2	1	R	R	0,7	0,2	R	R
		1		R		0,4		R
66/96	8	2	MR	R	1,8	0,2	MR	R
68/96	7	3	MR	R	1,5	0,6	MR	MR
109/96	9	4	MR	MR	2,0	1,0	MR	MR
264/96	8	6	MR	MR	2,0	1,0	MR	MR
296/96	5	3	R	R	0,7	0,6	R	R
392/96	6	6	MR	MR	1,5	0,6	MR	R
531/96	7	2	MR	R	2,1	0,6	MR	R
		5		MR		1,1		MR
Maximum value of experiment	51	41			9,5	7,4		

R = Resistant
MR = Medium Resistant

Table 3.Maize inbred lines resistant to *O. nubilalis* infested with 20 egg-masses/plant, as depending on seed-time.

Line	1996						1997						1998					
	5 may		3 june		6 may		6 june		7 may		3 june		7 may		3 june			
	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae	Cavities	Larvae		
	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2		
301/95	6	R 1,1	7	R 1,3	2	R 0,6	M	0,8	M	0,2	M							
302/95	6	R 1,4	16	M 3,3	2	R 0,5	R	0,6	R	0,2	M							
308/95	8	M 1,9	9	R 1,7	1	R 0,3	R	1,2	M	0,2	M							
309/95	7	R 1,5	10	R 2,0	3	M 0,5	R	1,2	M	0,2	M							
310/95	5	R 1,1	12	M 2,5	2	R 0,4	R	0	R	0	R							
338/95	9	M 1,9	12	M 2,9	3	M 0,6	M	1,2	M	0,2	M							
341/95	11	M 1,9	12	M 3,2	3	M 0,6	M	0	M	0	M							
448/95	6	R 1,9	15	M 2,8	2	R 0,3	R	0	R	0	R							
449/95	10	M 1,7	12	M 2,3	1	R 0,2	R	0	R	0	R							
453/95	9	M 1,6	15	M 2,5	4	M 0,8	M	0	R	0	R							
910/95	10	M 1,8	16	M 2,5	4	M 0,2	R	0	R	0	R							
447/95	9	M 1,4	12	M 2,5	0,3	R 0,2	R	0	R	0	R							
527/95	11	M 1,9	9	R 2,0	3,0	M 0,5	R	0	R	0	R							
66/97																		
67/97																		
91/97																		
94/97																		
229/97																		
259/97																		
276/97																		
298/97																		
Max. Value of exp.	46,0	7,0	65,0	14,9	24,0	3,6		22,2	3,2			46,7	9,1	44,1	6,3			

1 = Length of cavities cm per plant/no. of larvae per plant 2 = Score R = Resistant M= Medium resistant

Table 4. Maize inbred lines with extreme values of reaction to *O. nubilalis* in 1995 when an evident attack of the borer was registered

Line	Cavities		Larvae	
	Length cm/plant	Score	No. of specimens/plant	Score
Lines with low attack				
308/95	14	R	1,9	R
311/95	10	R	1,6	R
453/95	13	R	1,9	R
486/95	12	R	1,9	R
487/95	13	R	1,8	R
558/95	13	R	1,9	R
834/95	12	R	1,6	R
893/95	12	R	1,4	R
894/95	12	R	1,9	R
909/95	8	R	0,9	R
MEAN R	11,9		1,7	
Lines with heavy attack				
71/95	58	VS	9,7	VS
258/95	63	VS	10,3	VS
260/95	69	VS	12,4	VS
263/95	65	VS	11,6	VS
265/95	59	VS	10,5	VS
266/95	59	VS	10,6	VS
267/95	62	VS	11,0	VS
268/95	58	VS	12,2	VS
270/95	64	VS	14,1	VS
349/95	57	VS	10,2	VS
374/95	70	VS	10,2	VS
422/95	61	VS	10,6	VS
473/95	64	VS	9,8	VS
574/95	67	VS	10,3	VS
MEAN VS	62,6		10,2	

R = Resistant

VS = Very susceptible

Table 5.Maize inbred lines with extreme values of reaction to *O. nubilalis* in 1998, when a very low attack of the borer was registered

Line	Cavities		Larvae	
	Length cm/plant	Score	No. of specimens/plant	Score
Lines with low attack				
166/98	1,0	R	0,2	R
172/98	1,6	R	0,3	R
219/98	0,6	R	0,1	R
237/98	1,7	R	0,2	R
572/98	1,4	R	0,3	R
627/98	1,3	R	0,2	R
657/98	0,6	R	0,1	R
658/98	0,4	R	0,2	R
664/98	0,4	R	0,2	R
MEAN R	1,0		0,2	
Lines with heavy attack				
146/98	15,8	VS	3,4	VS
148/98	15,4	VS	3,6	VS
180/98	17,1	VS	3,0	VS
191/98	14,6	VS	3,0	VS
222/98	14,5	VS	3,5	VS
225/98	17,0	VS	3,7	VS
228/98	16,2	VS	4,3	VS
593/98	17,0	VS	3,3	VS
MEAN VS	15,9		3,5	

R = Resistant

VS = Very susceptible

Table 6.Maize inbred lines with extreme values of reaction to *O. nubilalis* in the last four years of testing for resistance

Lines with extreme attack values	1995		1996		1997		1998	
	Length of tunnels cm/plant	No. of larvae/plant	Length of tunnels cm/plant	No. of larvae/plant	Length of tunnels cm/plant	No. of larvae/plant	Length of tunnels cm/plant	No. of larvae/plant
Resistant	11,9	1,7	3,1	0,7	2,3	0,4	1,0	0,2
Very susceptible	62,6	10,2	40,0	7,9	32,2	6,0	15,9	3,5
Ratio resistant/very susceptible lines	5,2	6,0	12,9	11,3	14,0	15,0	15,9	17,5

Table 7. Behaviour of some maize hybrids to the *O. nubilalis* attack

Hybrid	Cavities				Larvae											
	Length cm/plant		Score		No. of specimens/plant		Score									
	1995	1996	1997	1998	1995	1996	1997	1998								
F 376	9	25	10,8	9,1	LR	LR	S	S	1,6	4,0	2,0	2,2	LR	LR	S	
Florencia	9	22	8,9	3,6	LR	MR	LR	LR	1,3	3,4	1,8	0,7	MR	R	LR	MR
HSF 737-94	6	19	7,2	-	R	R	LR	-	1,0	3,5	1,4	-	R	R	LR	-
HSF 749-94	10	28	-	-	LR	VS	-	-	1,8	4,7	-	-	LR	S	-	-
HSF 785-94	8	29	-	-	MR	VS	-	-	1,5	4,9	-	-	LR	VS	-	-
HSF 799-94	11	28	9,1	-	S	VS	S	-	2,1	4,9	1,6	-	S	VS	LR	-
HSF 853-94	11	28	8,7	-	S	VS	LR	-	2,1	5,2	1,6	-	S	VS	LR	-
HSF 857-94	9	23	7,2	-	LR	LR	LR	-	1,5	4,3	1,4	-	LR	LR	LR	-
HSF 321-94	15	-	-	-	VS	-	-	-	2,7	-	-	-	VS	-	-	-
HSF1057-96	-	-	9,3	6,1	-	-	S	S	-	-	1,8	1,3	-	-	LR	S
HSF 1071-96	-	-	5,4	1,3	-	-	R	R	-	-	1,2	0,2	-	-	MR	R
HSF 1083-96	-	-	4,7	3,6	-	-	R	LR	-	-	1,1	0,6	-	-	MR	MR
HSF 1135-96	-	-	8,9	5,3	-	-	LR	LR	-	-	1,6	0,7	-	-	LR	MR
HSF 1147-96	-	-	12,2	9,0	-	-	S	S	-	-	2,1	1,6	-	-	S	S
HSF 1151-96	-	-	5,7	4,6	-	-	R	LR	-	-	1,2	1,0	-	-	MR	LR
HSF 1231-96	-	-	15,5	15,4	-	-	VS	VS	-	-	3,0	3,2	-	-	VS	VS
HSF 1233-96	-	-	5,7	3,9	-	-	MR	LR	-	-	1,1	0,9	-	-	MR	LR
Dekalb 512 Bt	-	-	-	0,7	-	-	-	R	-	-	-	0,1	-	-	-	R

R = Resistant; MR=Medium resistant; LR= Low resistant; S= Susceptible; VS= Very Susceptible

Bionomy and Strains of European Corn Borer (*Ostrinia nubilalis* Hb.) in Slovenia

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Abstract

During several years of studying bionomy and strains of European corn borer (*Ostrinia nubilalis* Hb.) by light traps, pheromone traps and visual examination of development stages on maize, considerable differences have been observed in the time of its appearance and in the number of its generations in Slovenia. In Nova Gorica (near the Italian border) two generations per year were observed during a three-year study. European corn borer shows a very similar bionomy in the Southern part of Slovenia, where equally two generations develop each year. In the central and Eastern part of Slovenia European corn borer develops only one generation per year, this being rather long, as moths appear unevenly from the end of May till August. In order to determine if the number of generations depends on climatic conditions two populations of European corn borers - one from Prekmurje (near Hungarian border) and one from Nova Gorica - were observed under the same conditions in greenhouse with net cover on the Biotechnical Faculty in Ljubljana. During this two-year study of bionomy under natural conditions it was found out that the population from Nova Gorica developed two generations per year also under somewhat colder conditions, while the population from Prekmurje had only one generation yearly. The bionomy of the population from Primorje coincided in time with the bionomy of the same population in the natural habitat which is an even greater surprise. The up to now three-year study of European corn borer strains with pheromone traps did not give satisfactory results. In Bilje, where two generations develop each year, the E and EZ strains are equally common, in central Slovenia the E strain is more usual, while the Z strain is the only one found in the Eastern part of Slovenia along the Hungarian and Austrian border. The Z strain develops only one generation under the conditions in Slovenia, the same is true for the E strain in the central part of Slovenia. The results on the European corn borer strains in Slovenia are identical to those obtained by researchers in the neighbouring countries.

Key words: European corn borer, *Ostrinia nubilalis*, bionomy, strains, monitoring, Slovenia

Introduction

Slovenia is a small middle-European country, growing of maize plays an important role in its agriculture. It is grown on 44 % of arable land (i.e. on 77.343 ha) and is the most important crop in Slovenia.

European corn borer is the most important maize pest in Slovenia. It is spread over entire country up to 1200 m above the sea level. Owing to different climatic conditions in Slovenia the attacks of this pest differed considerably, but the reasons for these differences were not fully understood. So, lately more thorough investigations on bionomy and strains of European corn borer in Slovenia were undertaken on selected areas in Slovenia, chosen according to means available. European corn borer in Slovenia has been investigated by quite numerous researchers (Milevoj, L., Vrabl, S., Žolnir, M., Matjaž, K., Kač, M.). Their

investigations were mostly limited to the areas covered by the institutions where they worked. So, Vrabl (1983, 1986, 1992) determined that the pest has one generation in the SE part of Slovenia, the investigations of Milevoj (1991) showed one generation for the Ljubljana region and those of the researchers in Žalec (Matjaž, 1989; Kač, 1983, 1985) confirmed the existence of two generations of European corn borer. No comparative data existed for the entire country of Slovenia, especially some non central regions were complete blanks, so investigations taking place over some years should fill these blanks. At the same time one could get also insight into various strains of European corn borer in Slovenia which up to now were not known. Considering data on the strains found in the neighbouring countries (Rauscher *et al.*, 1991; Maini *et al.*, 1978; Bartels *et al.*, 1997), it seemed reasonable to assume that all three known strains meet in Slovenia. The investigation was performed to determine the strains of European corn borer that occur in Slovenia and the areas of their occurrence in this country.

Material and Methods

The strains of European corn borer were studied in the period 1997-1999 on some locations in Slovenia. The type of the strains was determined by the use of pheromone traps, which were placed on the fields among the plants (these being maize plants or hop plants, according to the area in question). Four types of pheromone traps were used in the experiments (E strain, Z strain, EZ strain and control), the experiments were performed in three repetitions. The traps were positioned on plastic 1.6 m high posts, paced 15 m from each other. Initially, they were checked every fortnight, later on every three to four weeks, as not too many specimens were collected. The entire traps were replaced by new ones every three to four weeks. They were placed when the first moths appeared (end of May) and removed at the end of their appearance (end of August). The pheromone traps used in years 1997 and 1998 were produced in Holland (DLO Research Institute of Plant Protection, Wageningen), while those used in 1999 were produced in Hungary (Plant Protection Institute of the Hungarian Academy of Science, Budapest). All traps used were of the sticky trap type.

The bionomy of European corn borer was followed on selected locations characterised by different climatic conditions. Constant monitorings of bionomy were performed in Kostanjevica on Krka (during 1994), in Bilje near Nova Gorica in the Primorje region (during 1996, 1997 and 1998) and in Radlje on Drava (during 1998). To catch moths on these locations standard light traps with 150 W fluorescent bulbs (otherwise standard in street lamps) were used. On each location the catching of moths was followed by one trap placed in the field of the crop where the monitoring was performed. In Kostanjevica the trap was in the orchard next to the field, in Bilje in a corn field, in Radlje in a hop garden. At this monitoring the counting of the trap catch was performed daily, the only exception being Kostanjevica, where the traps were checked every 2-3 days. Random monitoring was performed on those localities where a permanent monitoring was impossible because of great distances or insufficient means. At this inventory, several occurrences of European corn borer moths were described at the time favourable for its occurrence. Special tents designed for moths inventorying were used (fluorescent tents of pyramidal shape). Fluorescent bulbs of Philips TLD type were used as light sources. Each time the number and the development stages of European corn borer specimens that were captured on three such tents were inventoried. All such inventories were performed in corn fields. Results are given for Prekmurje (for the 1998 period) and for Ljubljansko Barje (for the 1997 period).

To check the theory that the climatic conditions also influence the bionomy of European corn borer, two populations of this pest were grown for one year in Ljubljana under identical conditions. One originated from Bilje in Vipavska dolina and the other one from Mala Polana in Prekmurje. The growing took place in the open air in a greenhouse with net cover and in the laboratory. For overwintering, both populations were raised in the greenhouse with net cover during the previous year, they overwintered there and the observations of bionomy began the next year. In the laboratory, the bionomy of the specimens raised from the eggs in the same year was observed. In order to get more reliable results for the population from Bilje, the growing in the greenhouse was continued also in 1999.

Results and Discussion

The results of the determination of the European corn borer strains in Slovenia during a three-year period are presented in Tables 1-3 and in Fig. 1. As evident from the Tables 1-3, the results of the catch on the pheromone traps vary a great deal and the number of the specimens caught was generally speaking relatively small. Nevertheless an acceptable variability of the results was achieved in the three-year period and the results allow an estimation of the occurrence of various European corn borer strains in Slovenia.

Table 1: Results of the studies on the European corn borer strains in 1997

locality	duration of the experiment	the number of changing of the pheromone baits	the total number of the specimens of <i>O. n.</i> caught.			
			E	EZ	Z	Control
Bilje	May 27 - Sept. 18	4	0	2	0	0
Gančani	June 01 - Sept. 22	4	0	0	9	0
Pacinje	June 03 - Sept. 23	3	4	0	1	0
Moste near Kamnik	June 10 - Sept. 03	3	2	0	0	0

Table 2: Results of the studies on the European corn borer strains in 1998

locality	duration of the experiment	the number of changing of the pheromone baits	the total number of the specimens of <i>O. n.</i> caught.			
			E	EZ	Z	Control
Bilje	June 04 - Aug.25	4	23	6	4	0
Gančani	June 05 - Aug. 30	4	0	0	23	0
Pacinje	June 09 - Aug. 26	3	3	2	0	0
Kamnik	June 09 - Aug. 26	3	2	1	0	0
Žalec	June 02 - Aug. 16	3	5	2	0	0
Radlje	June 02 - Aug. 13	3	0	0	2	0

Table 3: Results of the studies on the European corn borer strains in 1999

locality	duration of the experiment	the number of changing of the pheromone baits	the total number of the specimens of <i>O. n.</i> caught.			
			E	EZ	Z	Control
Bilje	May 27 - Aug. 25	4	15	0	0	0
Gančani	May 29 - Aug. 28	4	0	0	4	0
Pacinje	June 12 - Aug. 28	4	2	0	0	0
Križ near Kamnik	June 15 - Aug. 25	4	51	2	0	0
Žalec	June 09 - Aug. 19	4	23	0	0	0
Radlje	June 09 - Aug. 19	4	0	0	2	0
Krško	June 04 - Aug. 25	4	9	1	0	0

As the results show, there are three strains of European corn borer in Slovenia. The areas under investigation show different occurrence of these strains. In the Eastern part of Slovenia the Z type occurs, in the central, Western and South Slovenia predominates the E strain. Details are shown in Fig. 1.

According to our experiences with the pheromone traps, we can conclude that they are not selective enough for European corn borer. The catch with these traps suffered from high variations and was considerably lower compared to the light traps. It is probable, that due to rather incomplete knowledge on the sexual pheromonic substances of European corn borer, the attraction of males by such a pheromone trap is not effective enough. So, a weekly check of the traps was not necessary and they were checked only every 3-4 weeks, which showed to be optimal as well from the practical as also from the economical point of view. For the reasons given above, the results of the catch on the pheromone traps can not give a genuine picture of the European corn borer bionomy. Many times the number of specimens on pheromone traps differed from that obtained by the light traps and the results were contradictory. So we decided to investigate the bionomy of European corn borer using only the results obtained by the light trap catch.

The regular monitoring of the occurrence of European corn borer gave some interesting results which contradict the till now generally accepted assumption that there is only one European corn borer strain in Slovenia. For Bilje (Western Slovenia) and for Kostanjevica on Krka (Southern Slovenia) it has been proven that European corn borer in these areas has two generations yearly (Graphs 1, 2, 3). The first generation in Bilje in Vipavska dolina appears at the end of May and reaches its maximum during the first half of June, the second appears at the end of July and reaches its maximum at the beginning of August. The flight of the second generation can proceed into autumn, till October. The bionomy of European corn borer in Kostanjevica on Krka is very similar, but compared to Bilje the population here is considerably less numerous. Comparing Graphs 1 and 2 shows that the dynamics differs considerably from year to year and depends strongly on the weather conditions of each season; temperature, rainfall and relative humidity in the period between laying eggs and appearance of the moths being the most crucial factors. The second generation in 1997 was rather more numerous compared to the same generation of the previous year, apparently due to more favourable weather conditions during its development. This was clearly

during its development. This was clearly observable also from the results of the estimation of the damage in the maize fields caused by this pest. The loss was monitored in autumn. For 1998 this discrepancy was even more pronounced, when - during the peak season of the second generation of European corn borer - more than 400 specimens of this pest were caught during one night only. The autumn monitoring of the damage in the maize fields showed not one single plant which would not have been attacked by this pest. The plants suffering from only one hole in the stem were very rare. This speaks for a very massive occurrence of the pest in 1998, the maize crop was seriously damaged.

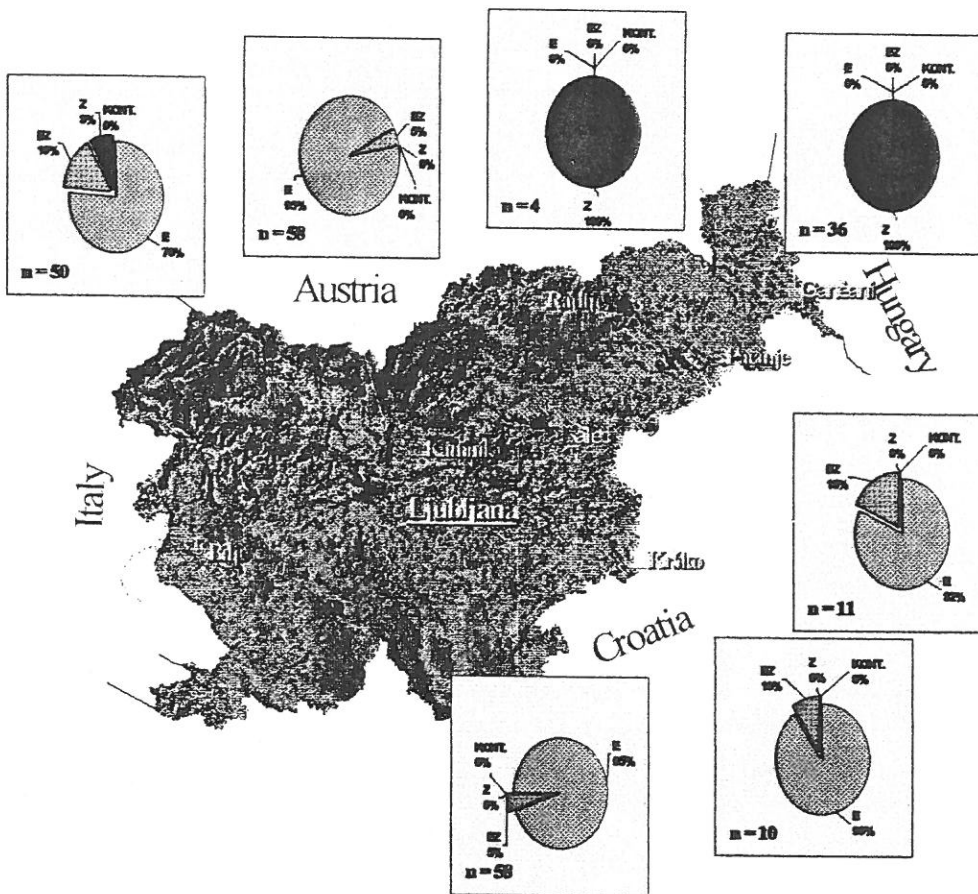
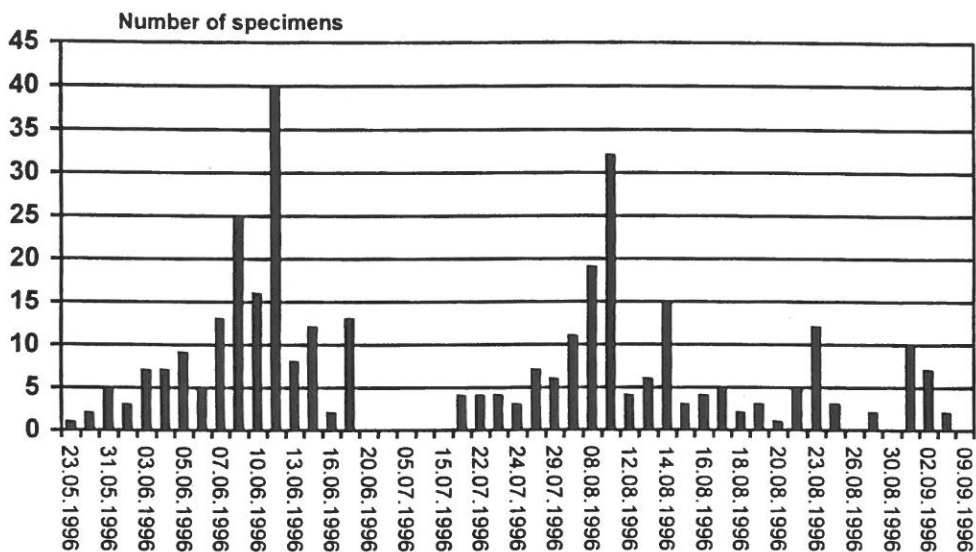
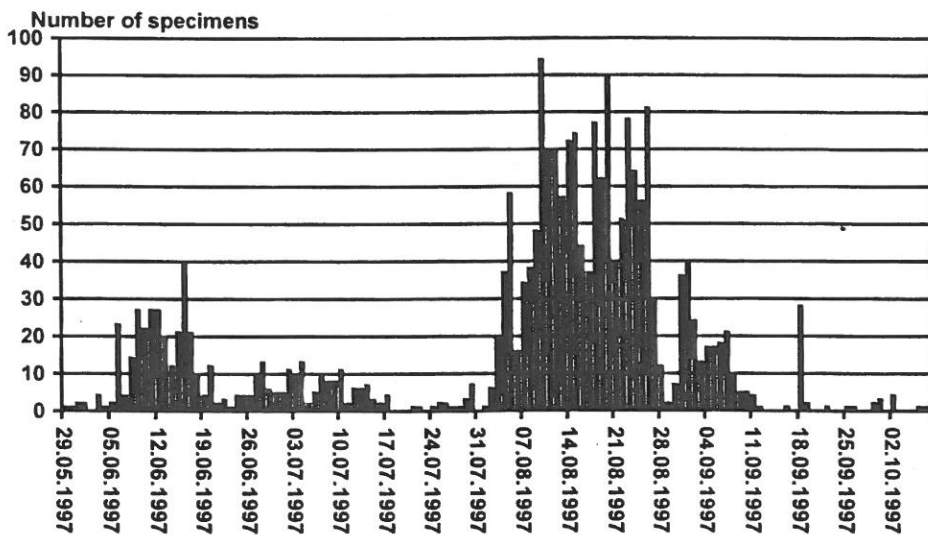


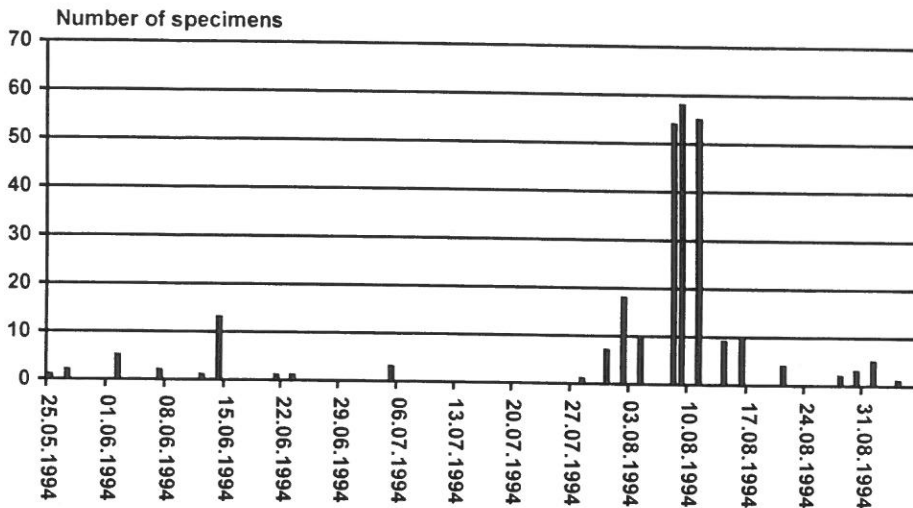
Figure 1. Strains of European corn borer on various locations in Slovenia as determined from the results of the pheromone trap catch in the 1997 - 1999 period



Graph 1: Bionomy of European corn borer in Bilje in 1996 (light trap)



Graph 2: Bionomy of European corn borer in Bilje in 1997 (light trap)



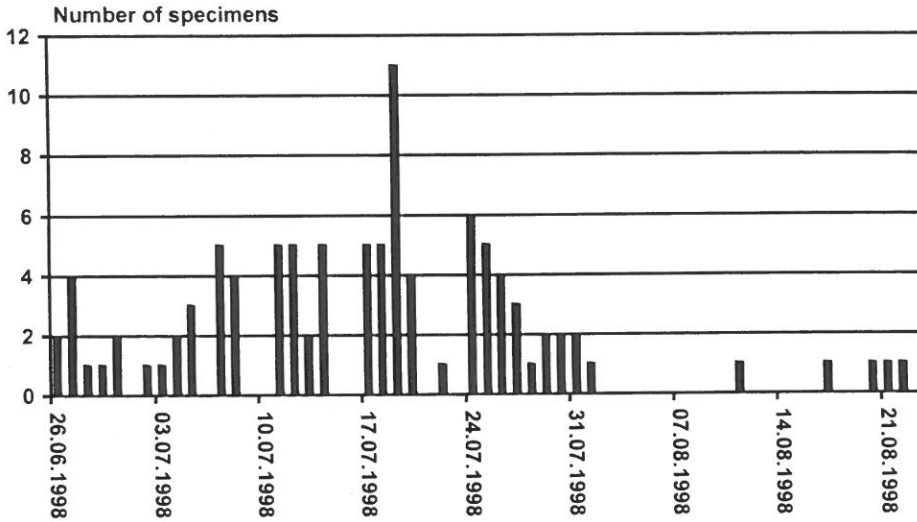
Graph 3: Bionomy of European corn borer in Kostanjevica on Krka in 1994 (light trap)

In other parts of Slovenia only one generation of European corn borer yearly was found. One generation was observed in Radlje on Drava (Graph 4) in Prekmurje (Graph 5) and on Ljubljansko Barje (Graph 6). Following the developmental stages of the insects, their attack as well as the catch on the pheromone traps one generation only was confirmed also for Kamnik, Pacinje at Ptuj and Žalec. Graphs 4-6 show pronounced differences in occurrence of the first moths for various locations. In Radlje, first moths do not appear before the end of June, on Ljubljansko Barje this event takes place a little earlier, in Prekmurje it occurs at the end of May. This depends on the climatic conditions in which the populations are established as well as on the weather conditions of any single year.

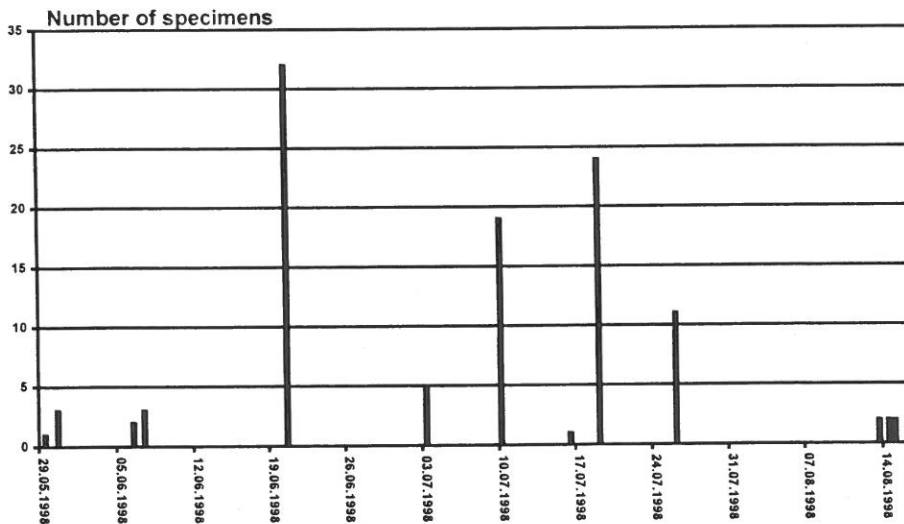
Also the dynamics of the appearance of the moths of this only generation can last for a longer period. This has to do with the climatic characteristics of the areas in question. So the flight of the moths stops in Radlje at the earliest date, the location on Ljubljansko Barje follows and in Prekmurje it terminates on the latest date. These conclusions are not based on the data presented in the Graphs in this contribution alone, they include also other data in the personal data base. From the data one can conclude that the period of the flight of the moths is the longest in the area with the longest warm period and *vice versa*.

Additionally, some differences in the appearance of the moths in Prekmurje (Eastern Slovenia) due to microclimatic differences were observed. It was established that on wet regions (Mala Polana and Muriša) they appear earlier and exhibit an earlier maximum of the flight period compared to drier, more agricultural districts with more arable land. On these latter locations moths can be found till the beginning of September. On arable lands in this part of Slovenia the maximum of the flight period (*maksimum izleta*) takes place at the beginning of June and the season lasts, though sporadically, till the end of August. Following the dynamics of European corn borer population for some years allows the conclusion that the flight period during the season varies a lot and occurs during a long period. The dynamics of the catch in the light traps during the season varies a lot, so the results can be misleading in the sense that they may indicate more generations that actually present. To avoid this possible misinterpretation, two populations of European corn borer

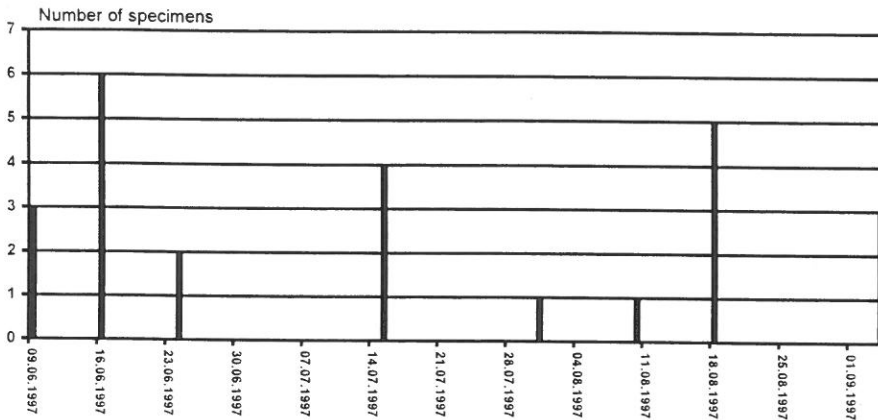
were grown in the greenhouse with net cover on the Biotechnical Faculty in Ljubljana for close examination of their development.



Graph 4: Bionomy of European corn borer in Radlje on Drava in 1998 (light trap)



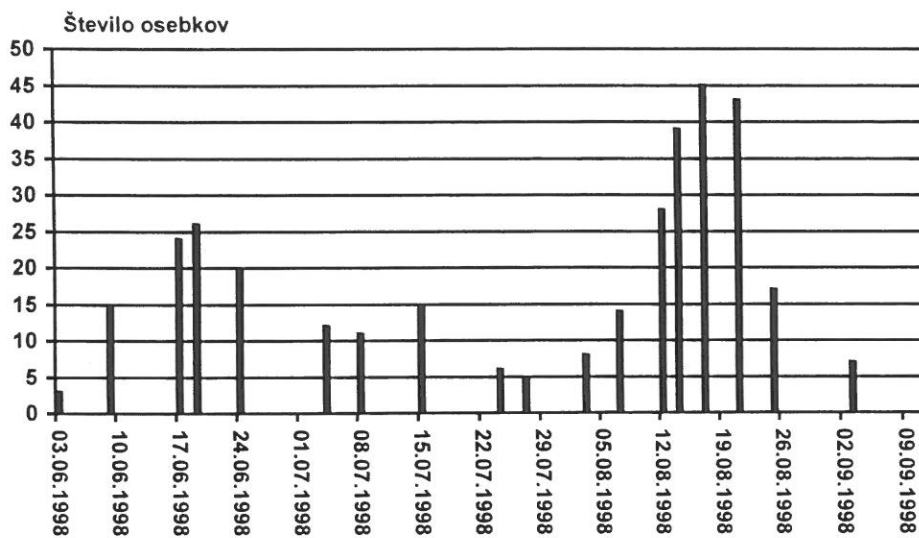
Graph 5: Bionomy of European corn borer in Prekmurje in 1998 (fluorescent light tent)



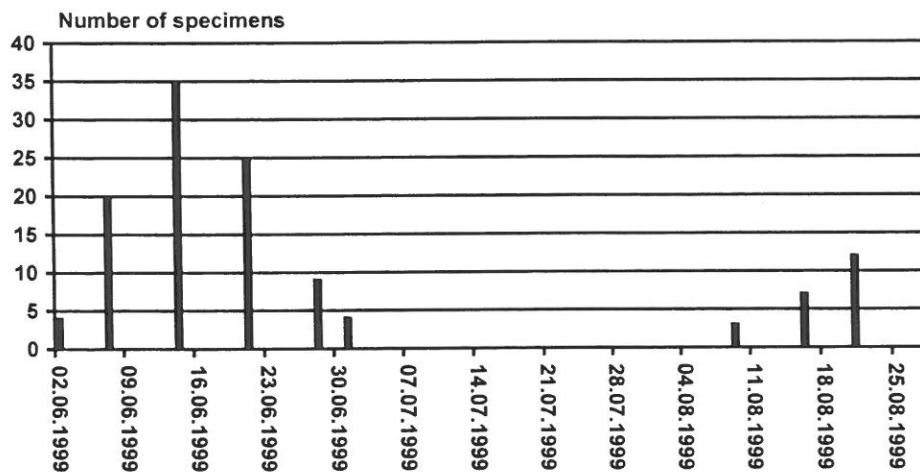
Graph 6: Bionomy of European corn borer on Ljubljansko Barje in 1997 (fluorescent light tent)

Two populations of European corn borer, one from Vipavska dolina and one from Prekmurje, were included into a comparative study in Ljubljana. Grown under controlled conditions in a laboratory at constant temperature of 21 °C, feed on maize plants, the population from Bilje developed two generations during one season, while the population from Prekmurje developed only one generation during the same period. The caterpillars of the first generation from the Bilje population pupated in the middle of July, the moths appeared at the end of July and at the beginning of August. From this generation the second generation caterpillars developed, but these did not pupate during the same season. The caterpillars from the Prekmurje generation did not pupate during the same season, they fed for some time and entered the phase of diapause. The caterpillars from the Prekmurje generation were bigger compared to the caterpillars from the Bilje generation. This growing showed that the temperature alone does not influence the number of generations developed per season and that many additional factors, some including also the genetic traits play a decisive part.

The same results were obtained by growing both populations under natural conditions, on the trial field of the Biotechnical Faculty in Ljubljana, during 1998 and 1999 (Graphs 7, 8). After the overwintering of the caterpillars of both populations, the caterpillars from the Bilje population pupated in the period from middle May till the end of May, those of the second generation from the middle of July till the end of July, the moths appeared in June and in the beginning of August, the maximum being in the middle of June and in the middle of August. The number of the flying moths was determined by frightening them from the plants and counting them in the air. The caterpillars of this population overwintered in different developmental stages (L2 - L4). These caterpillars overwintered in the pith of the maize pant stem and many of them on the ears. The population from Prekmurje had only one generation of moths in one season, these were appearing from the middle of June till the middle of August. The caterpillars of this population did not pupate during this season, they overwintered as adult caterpillars in the stem pith.



Graph 7: Bionomy of European corn borer, population from Bilje (greenhouse with net cover in Ljubljana in 1998)



Graph 8: Bionomy of European corn borer, population from Bilje (greenhouse with net cover in Ljubljana in 1999)

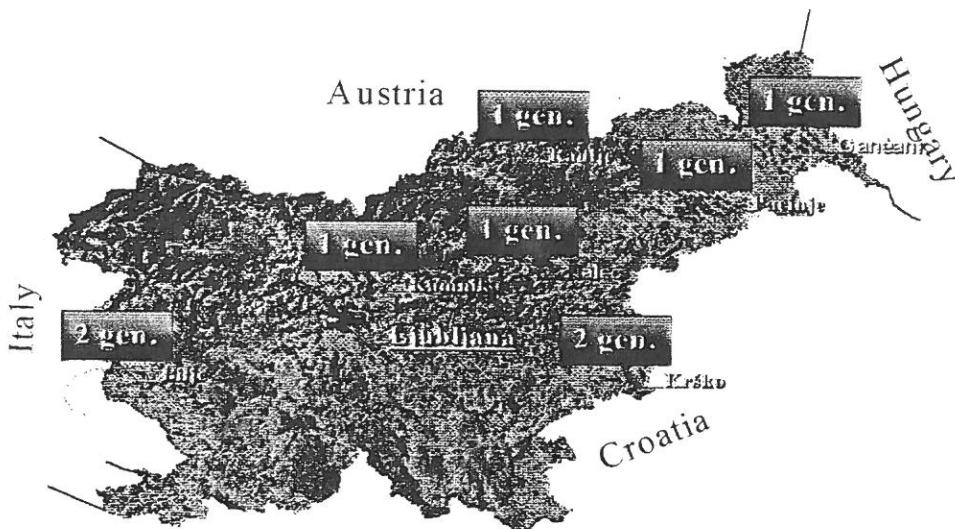
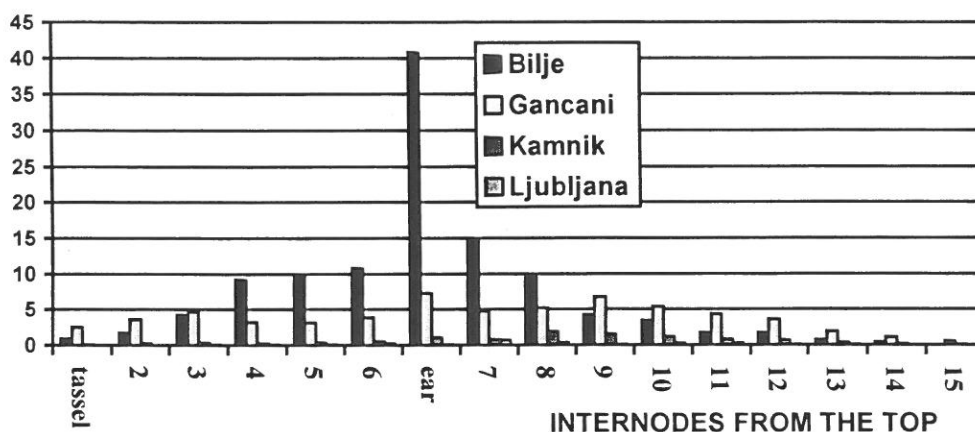


Figure 2. Number of generations of European corn borer in Slovenia, on locations investigated

If we combine the data on strains and bionomy of European corn borer, some interesting conclusions can be made. These show that the Z strain develops one generation yearly, while the E and the EZ strains develop one or two generations yearly depending on climatic conditions. So, in Slovenia two generations of European corn borer appear in warmer regions. As we were classifying the attack of European corn borer according to regions, intensity and allocations on the stem, differences between the regions were observed. In the regions where the European corn borer has two generations, the attack is stronger and the damage is greater, the allocations of the attack on the stem also show differences, as the second generation preferably attack the ear and the parts of the stem immediately at the ear (Graph 9).

The results of our investigations are in good agreement with results of the investigations conducted by other researchers on the same subject (Bača, 1995; Rauscher *et al.*, 1991; Lagenbruch, Szewczyk, 1995; Tanasijevič, Simova-Tošič, 1987; Greatti, Zandigiacomo, 1995). The set of results is too limited for more general conclusions, so it will be necessary to continue investigations on the bionomy of European corn borer, possibly on more locations. In spite of that the results collected till now during the research on European corn borer brought quite some new findings which were not known till now. The investigations confirmed some former studies, that European corn borer develops one generation per season in Eastern and central part of Slovenia, but they did not agree with the statement that it has two generations per year in Savinjska dolina. The finding that European corn borer develop two generations also in Vipavska dolina and on Krško polje is completely new.



Graph 9: Attack of European corn borer on the stem and according to regions, calculated on 100 plant examined in 1996 (n = 5640 plants)

Conclusions

- Three strains of European corn borer were found in Slovenia, they occupy geographically different areas. In the Western and in the Southern part of Slovenia the E strain prevails over the EZ strain, in the central part of Slovenia the E strain prevails, while in the Eastern part of Slovenia only Z strain of European corn borer can be found.
- The number of generations of European corn borer is different in different parts of Slovenia.
- In Eastern and Southern Slovenia the European corn borer develops two generations yearly, while it has just one generation in Eastern and central Slovenia.
- The number of the generations depends on climatic areas where the pest lives as well as on the strain of European corn borer. The Z strain develops only one generation, while the E and the EZ strains can have either one or two generations.
- The growing of natural populations of European corn borer from Bilje and from Prekmurje for two years in greenhouse with net cover and in laboratory showed that the number of generations remains the same also under changed ecological conditions.
- The bionomy of European corn borer can be reliably followed only with light traps or with accurate monitoring of development stages, the prognosis with pheromone traps is not reliable.
- The allocation of the damages caused by the caterpillars on the maize stems depend also on the number of generations of the European corn borer. The attack of the second generation is the strongest on the ear as well as on the internodes near the ear. This increases the damage on the harvest.

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Population Dynamics of *Ostrinia nubilalis*: Specificity in Key Factors for One- and Two-Generation Zones of Russia

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Abstract

Ecological factors critical for the European corn borer, *Ostrinia nubilalis* (Hbn.), (Lepidoptera: Pyralidae) were studied intensively, but their actual influence on population dynamics was not sufficiently understood yet. During 1994-1998 life tables were arranged for two local populations inhabiting eastern part of the Krasnodar Territory (two-generation zone) and southern part of the Belgorod Province (one-generation zone). By using graphic K-factor analysis (Varley, Gradwell, 1960) and regression component analysis (Harcourt, 1966) it was established that at the Krasnodar Territory a key factor performs during I-II instar larval feeding of the 1st generation, while in the Belgorod Province it operates during the period of moth activity. Just as density-independence, so density-dependence in operation of key factors was displayed. It is supposed that the behavioural responses of I-II instar larvae to aggregation may play an important role for density-dependent population decline. The density-dependence of egg realisation by females most likely based also on behavioural reactions of adults leaving overpopulated places.

Key words: European corn borer, *Ostrinia nubilalis*, population dynamics, Russia

Introduction

Ecological factors critical for the European corn borer (ECB), *Ostrinia nubilalis* (Hbn.), (Lepidoptera: Pyralidae) were studied quite intensively (Shchoegolev, 1934; Kozhanchikov, 1935; Sparks et al., 1967; Barlow, 1971; Chiang, Hodson, 1972; Hudon, LeRoux, 1986, Kornoşor, Kayapinar, 1988, etc.), but their actual influence on population dynamics was not sufficiently understood yet. Wide growing of genetically engineered maize could occur in the very near future demanding to study the ECB population dynamics just now before significant alterations happen. Furthermore, the ECB population dynamics has been properly investigated only in North America, while the pest being of the European origin. Up to date, thorough analysis of the ECB population dynamics has never been fulfilled in Europe.

The Krasnodar Territory and the Belgorod Province are the main zones of grain and seed production of maize in Russia. Both regions are characterised by climatic conditions favourable for the ECB propagation. However, these regions differ in surroundings critical for the ECB ecology. Thus, in the former zone two annual generations develop and a trace of the 3rd generation appears almost each year, while in the latter only one generation develops per year. Besides, at the Krasnodar Territory maize had long-standing grown for grain and seed production, contrary to the Belgorod Province where it had almost exclusively cultivated for silage even about 20 years ago. In so doing, the ECB is known at the Krasnodar Territory as a prime pest of maize since the first quarter of the 20th century, whereas in the Belgorod Province the insect became attacking maize seriously only about

10-15 years ago. Furthermore, the ECB at the Krasnodar Territory is capable to infest actively sorghum aside maize in contrast with the Belgorod Province where the insect is able to develop on millet.

Final goal of the work is to construct an appropriate model for the ECB population dynamics in its homeland territories, including southern (Krasnodar) and northern (Belgorod) parts of the Russian "Corn Belt". First results of the study obtained in 1994-1995 were presented at the 18th IWGO Conference held in Romania (Frolov et al., 1995). The present paper reports on life-table analysis of the ECB population fluctuations during 1994-1998 with the aim to detect key factors responsible for the insect population dynamics. The study is going to be continued and during the next stage we will assess quantitatively effects of density-dependence and density-independence as well as to start investigation of a factor responsible for cyclicity in population dynamics of the pest at the Krasnodar Territory.

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Materials and Methods

The ecological study plots were located within the scientific crop rotations of the Kuban Experimental Station (KES) of the All-Russian Institute for Plant Industry, located at eastern part of the Krasnodar Territory, and the Belgorod State Agricultural Academy (BSAA), located at southern part of the Belgorod Province. All the usual tillage cultural practices were used including cultivation, herbicide treatments, and mechanised cultivation, mowing and threshing at harvest. During 1994-1998 we made counts of insect densities on crops actively infested by the ECB, namely, maize (KES, BSAA) and sorghum (KES). Other host plants of the ECB, including millet (BSAA) or panicle cereal weeds (KES) usually occupied spaces much less than 5% of those occupied by maize, and thus were excluded of observation.

The samples were periodically collected to estimate insect densities and related mortality factors. Direct sampling was performed during certain time intervals when the specific stage of insect development (egg, larva, pupa, adult) prevailing on 0.02-22 ha fields (6-21 fields at KES and 2-4 in BSAA each year). The total number of surveys was 21-23 at KES and 12-14 in BSAA. Randomised sample units chosen were 0.1-1 m² plots for overwintering larvae in remnants, 1-3 m² plots for attacking plants larvae, and 0.8-5 m² plots for egg counts; the number of sample units on each field varied to assess statistically significant estimates (usually of 10-50 plots per field). All insect numbers were calculated per 1000 m² of maize sowing.

Spatially fixed plots were used to count egg densities. Each egg cluster found on foliage was marked and a hand lens was used to count the number of eggs as either hatched successfully, parasitised, predated, dislodged, or dead due to fail to develop embryo (infertility) or larval inability to hatch (possibly from desiccation). Each plot was examined for 6-8 times in 5-7 day intervals during the period of moth flight. To estimate the whole number of eggs laid we summed up the numbers of eggs found during each successive examination of a plot.

The summer larvae and pupae (in case of KES) were sampled on random plots usually twice by 5-10 days after finishing of egg laying. The leaf and stalk damage was used only as a good visual clue to locate presence of insects. The estimate of adult density was obtained

from the number of pupal cases. To estimate female fecundity, insects were kept under insectary conditions. The total decrease in adult numbers during their egg laying was assessed indirectly on the basis of difference between actual and expected numbers of oviposited eggs.

For the Krasnodar population insect numbers were estimated for the following age intervals of the 1st generation: 1. total eggs (eggs laid either on maize or sorghum), 2. maize eggs (eggs laid on maize), 3. I-II instar larvae, 4. III-V instar larvae, 5. pupae, 6. adults, 7. females (= adults with sex ratio of 1:1), 8. normal females (= adults with sex ratio of 1:1 and 450 eggs deposited per one gravid female), and 9. ovipositing females (the number of eggs of the next generation divided on one-half of the mean number of eggs deposited per one gravid female, that is in this case — on 225). During the development of the 2nd generation insect numbers were estimated for the following age intervals: 1. total eggs, 2. maize eggs, 3. I-II instar larvae, 4. III-V instar larvae, 5. diapausing larvae before harvest, 6. diapausing larvae after harvest before hibernation, 7. overwintered larvae, 8. pupae, 9. adults, 10. females, 11. normal females, and 12. ovipositing females. For the Belgorod population insect numbers were estimated for the following age intervals: 1. eggs, 2. I-II instar larvae, 3. III-V instar larvae, 4. diapausing larvae before harvest, 5. diapausing larvae after harvest before hibernation, 6. newly overwintered larvae (April), 7. overwintered larvae before pupation (June), 8. pupae, 9. adults, 10. females, 11. normal females (the average level of fecundity was assumed 360 eggs deposited per one gravid female), and 12. ovipositing females.

Density drop for each age interval was assessed by $K = \log N_t - \log N_{t+1}$, population trend index, $I = \frac{N_{t+1}}{N_t} \times 100$, was calculated after R. Morris (1957), where N_t and N_{t+1} =

insect densities before and after an action of mortality factor. Key factor analysis (Varley, Gradwell, 1970) and component regression analysis (Harcourt, 1969) were used to detect age intervals with the most significant insect mortality.

Results and Discussion

Analysis of age and total mortality in two European corn borer populations

More than 10-fold fluctuations in total egg numbers were observed for the Krasnodar population during 1994-1998: from 6.9 m⁻² to 109.0 m⁻² (1st generation) and from 12.3 m⁻² to 191.2 m⁻² (2nd generation). Fluctuations in egg numbers of the Belgorod population were significant as well and varied from 15.5 m⁻² to 70.8 m⁻². Changes in insect densities during specific age intervals are presented for both populations (fig. 1, 2).

Fluctuations in age mortalities along with a fluctuation in total mortality were presented also both for the Krasnodar (fig. 3, 4) the Belgorod population (fig. 5) with a view to perform K-factor analysis.

It is obvious that the age interval of I-II instar larvae is a critical period in population dynamics of the Krasnodar population during the development of the 1st generation. The early instar larval mortality in the shape of curve of k follows the same fluctuating course as K (total mortality) (fig. 3). Besides, the late instar larval mortality influenced significantly the total mortality during the period of population depression (1994-1995), but was of minor importance when the insect density increased. Fluctuations in k -values for the other age intervals (eggs, pupae, adults) seemed to have low impact on total K .

As far as the 2nd generation is concerned, neither mortality factor is capable to vary through a sufficient range of magnitude to affect fluctuating course as K (fig. 4).

An adult mortality (normal females) influenced most significantly population dynamics of the Belgorod population (fig. 5).

The component regression analysis substantiates that the early instar survival is a critical factor for the Krasnodar population dynamics during the 1st generation development (table 1). Despite a high correlation and t-value significance, both the mortality of laid on maize eggs and the mortality of I-II instar larvae seem to affect the total mortality of insects during the 2nd generation less than some other mortalities, e. g. during harvest, overwintering, or egg laying (table 2). All it means that the mortality of I-II instar larvae should consider after Morris (1959) as the only principal key factor affecting population dynamics of the pest at the Krasnodar Territory.

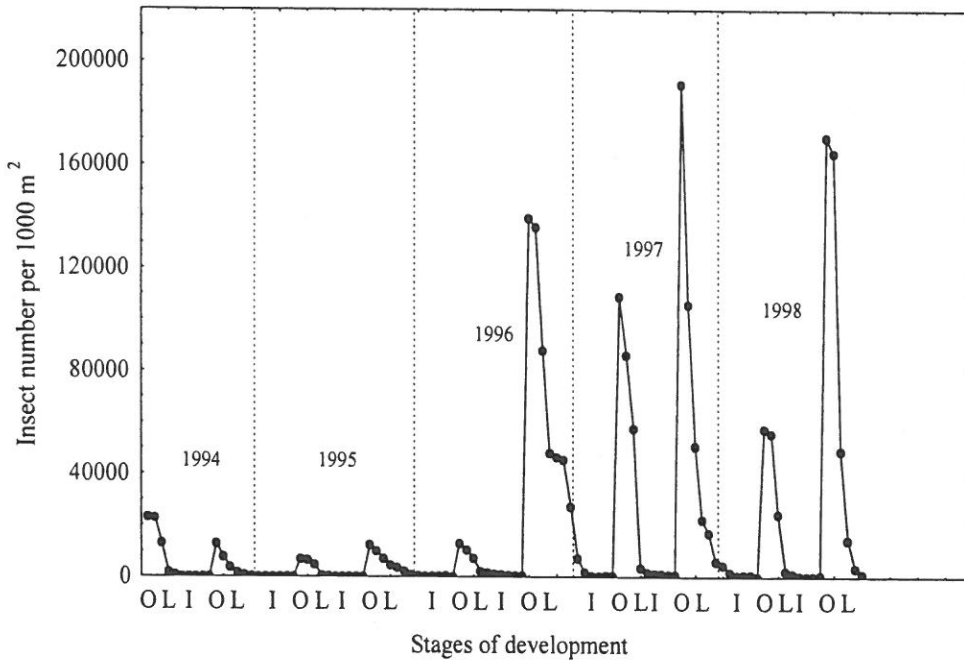


Figure 1. Population dynamics of the ECB at KES during 1994-1998. Letters O, L, I mark densities of total eggs, III-V instar larvae, and adults correspondingly.

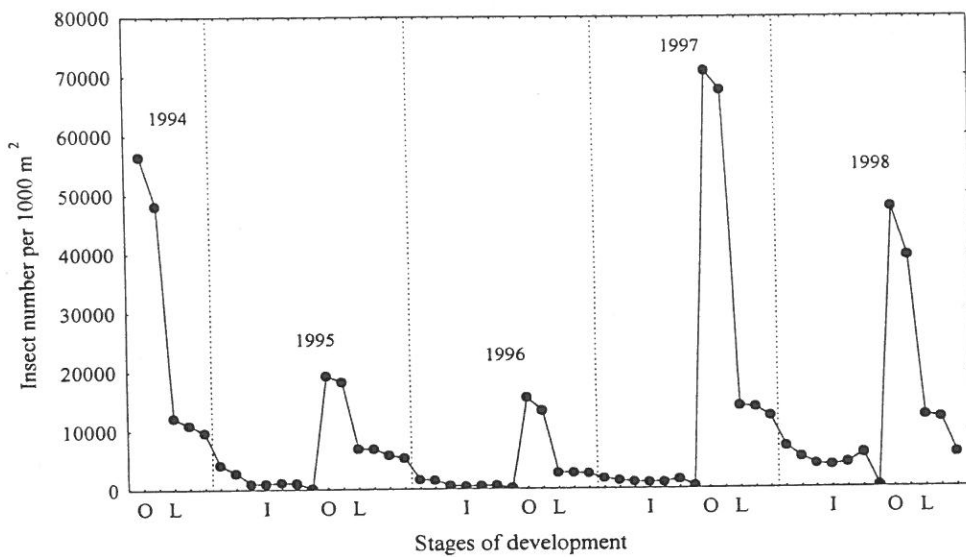


Figure 2. Population dynamics of the ECB in BSAA during 1994-1998. See legend of fig. 1 for explanation of O, L, and

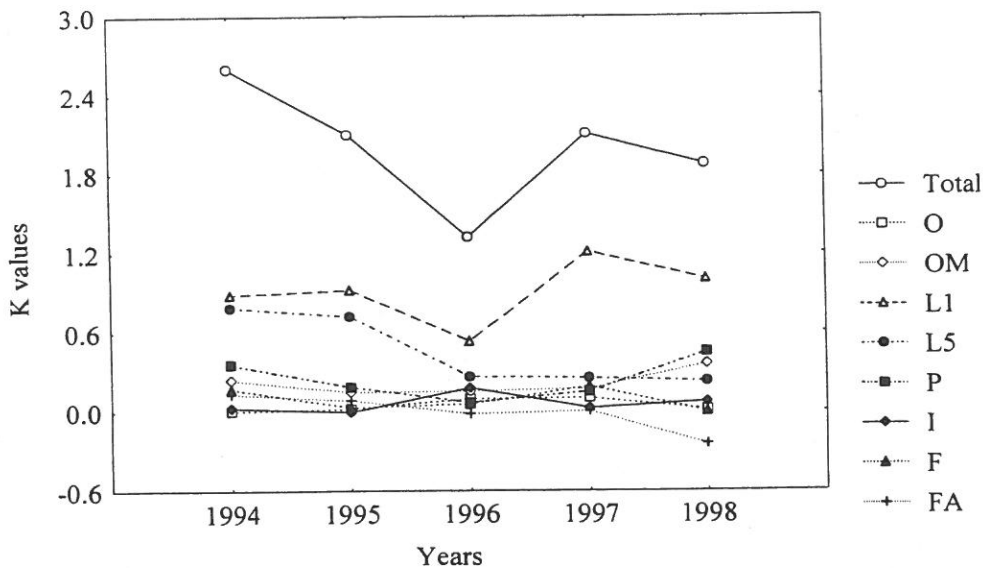


Figure 3. Curves of age and total mortalities of the ECB during the 1st generation at KES in 1994-1998. Age intervals: O – total eggs, OM – eggs laid on maize, L1 – I-II instar larvae, L5 – III-V instar larvae, P – pupae, I – adults, F – females, FA – normal females. Total – total K for a generation.

	1st generation		2nd generation	
Population trend index	3.564	4.105	0.667	0.324
Total eggs	0.903	0.097	0.737	0.196
Maize eggs	0.620	0.115	0.582	0.115
I-II instar larvae	0.141	0.090	0.529	0.087
III-V instar larvae	0.410	0.214	0.674	0.223
Pupae	0.602	0.205	0.466	0.106
Adults	0.883	0.135		
Females	0.828	0.160		
Normal females	1.073	0.408		
Population trend index	0.667	0.324	0.667	0.324
Total eggs	0.737	0.196	0.737	0.196
Maize eggs	0.582	0.115	0.582	0.115
I-II instar larvae	0.529	0.087	0.529	0.087
III-V instar larvae	0.674	0.223	0.674	0.223
Diapausing larvae before harvest	0.466	0.106	0.466	0.106
Diapausing larvae after harvest before hibernation	0.498	0.240	0.498	0.240
Overwintered larvae	0.561	0.279	0.561	0.279
Pupae	0.626	0.314	0.626	0.314
Adults	1.012	0.173	1.012	0.173
Females	1.129	0.116	1.129	0.116
Normal females	0.405	0.378	0.405	0.378

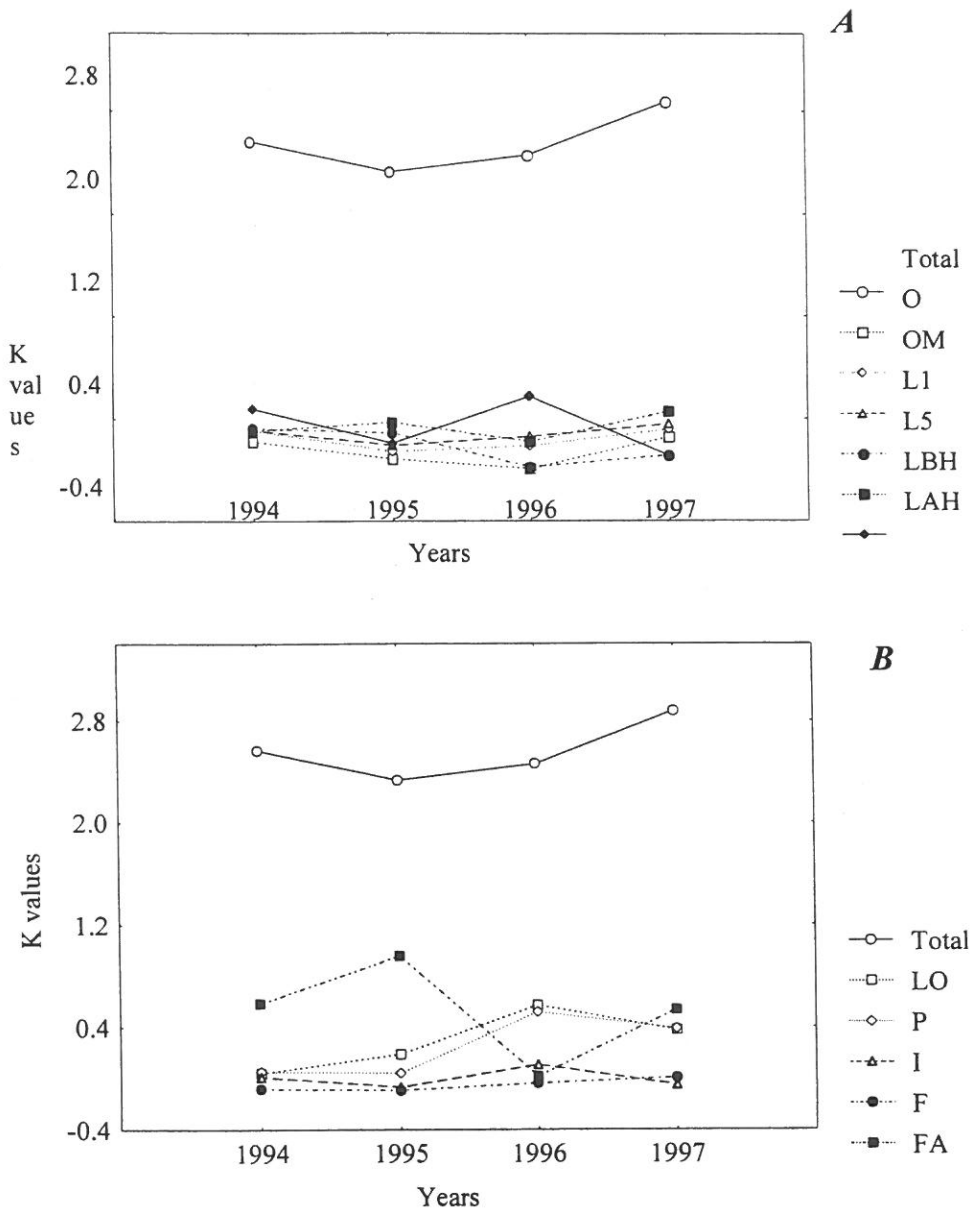


Figure 4. Curves of age and total mortalities of the ECB during the 2nd generation at KES in 1994-1998. *A* (before hibernation): O – total eggs, OM – eggs laid on maize, L1 – I-II instar larvae, L5 – III-V instar larvae, LBH – larvae before harvest, LAH – larvae after harvest, before hibernation; *B* (after hibernation): LO – overwintered larvae, P – pupae, I – adults, F – females, FA – normal females. Total – total K for a generation.

In spite of a high correlation and t-test significance, the larval mortality of the Belgorod population during harvest appeared to influence the total mortality less than some other

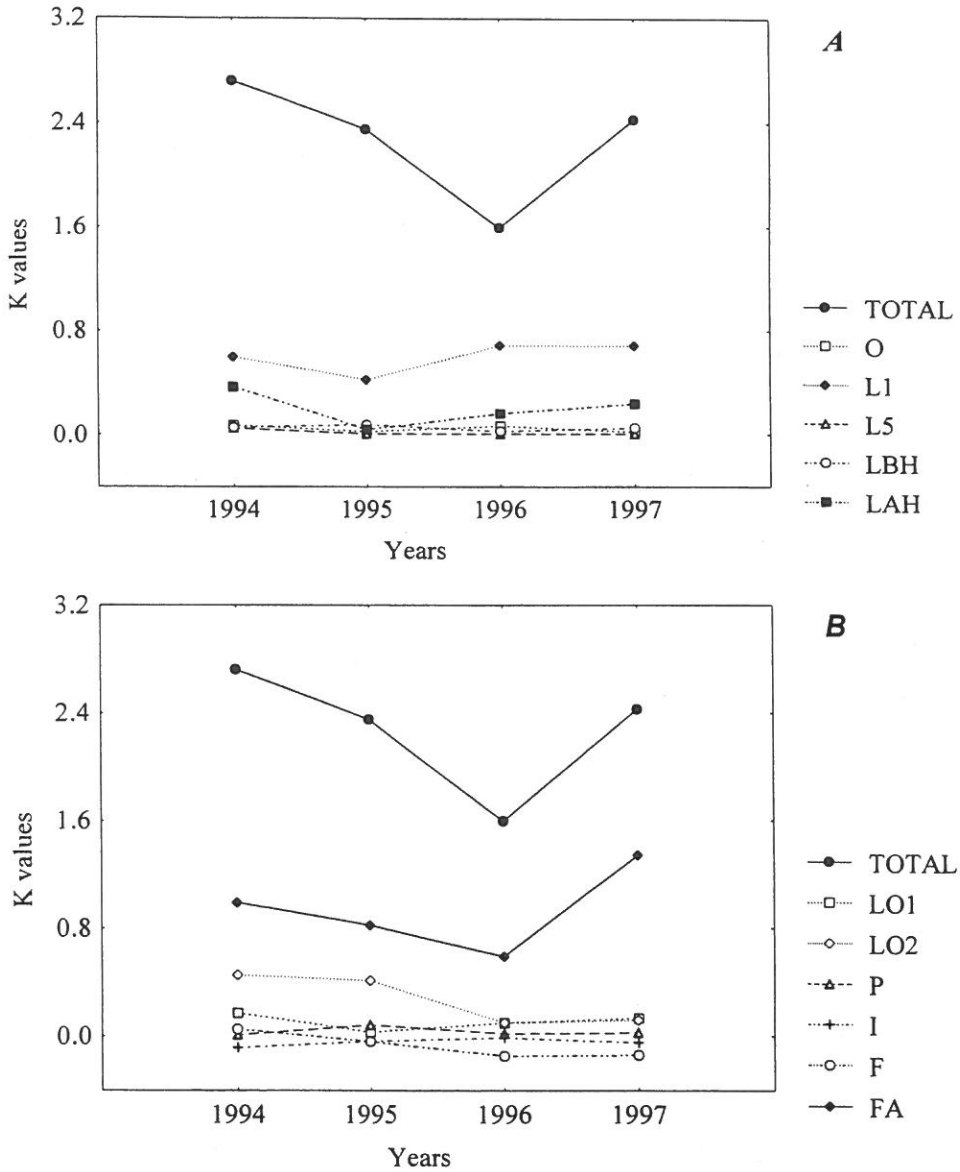


Figure 5. Curves of age and total mortality of the ECB in BSAA in 1994-1997. **A** (before hibernation): O – eggs, L1 – I-II instar larvae, L5 – III-V instar larvae, LBH – larvae before harvest, LAH – larvae after harvest, before hibernation; **B** (after hibernation): LO1 – newly overwintered larvae in April, LO2 – overwintered larvae before pupation in June, P – pupae, I – adults, F – females, FA – normal females. Total – total K for a generation.

factors, such as the larval death during hibernation or before pupation in April – June. The adult mortality (normal females) is the only factor, which affects a population dynamics dramatically (table 2).

It is important to mention that at the Krasnodar Territory the ECB population numbers declined seriously during the oviposition by overwintered (= 2nd generation) adults as well (table 1). For the Belgorod a population death of early instar larvae is detected being rather high too (table 2).

In other words, insect numbers of both populations decreased most significantly during the same age intervals (= critical periods in general cycle), as follows: 1) the early instar larval feeding within the maize leaf whorl, and 2) the activity of overwintered adults when egg laying. However, graphic K-factor analysis and regression component analysis have proved their specific role in regional population dynamics: the former factor determines total mortality level at the Krasnodar Territory, the latter one – in the Belgorod Province.

Table 2. Means, variances, correlation coefficients, and t-tests of significance of age survivals and population trend index at BSAA (1994-1997)

Survival estimate	Mean	Variance	$r_{(Y,X)}$	$r^2_{(Y,X)}$	t	p_a
Population trend index (I)	1.598	1.994				
Maize eggs	0.903	0.054	-0.453	0.205	-0.719	0.547
I-II instar larvae	0.261	0.082	-0.403	0.163	-0.623	0.597
III-V instar larvae	0.966	0.045	0.403	0.163	0.623	0.597
Diapausing larvae before harvest	0.888	0.040	0.827	0.683	2.078	0.173
Diapausing larvae after harvest before hibernation	0.494	0.179	0.702	0.492	1.393	0.298
Overwintered larvae (April)	0.787	0.110	0.167	0.028	0.242	0.831
Overwintered larvae before pupation (June)	0.575	0.238	0.646	0.417	1.196	0.354
Pupae	0.926	0.068	0.209	0.044	0.302	0.791
Adults	1.112	0.078	-0.794	0.630	-1.840	0.206
Females	1.192	0.245	0.612	0.374	1.094	0.388
Normal females	0.139	0.894	0.883	0.780	2.662	0.117

Key factors in the European corn borer population dynamics

The mortality of I-II instar larvae during the 1st generation development at the Krasnodar Territory

The average survival of I-II instar larvae varied from 6.2 to 29.4% during 1994-1998. The early instar larval survival was confirmed to correlate with the host plant resistance in maize (fig. 6). However, neither meteorological factor measured (mean, maximum, and minimum daily air temperature; maximum and minimum soil surface temperature; the least relative air humidity; precipitation) influenced significantly on the early instar larval survival. Perhaps, it was due to a short duration of larval stay outside the host plant. Nevertheless, it is not improbable that weather/climate can influence the larval survival by means of changing host plant suitability for insect feeding.

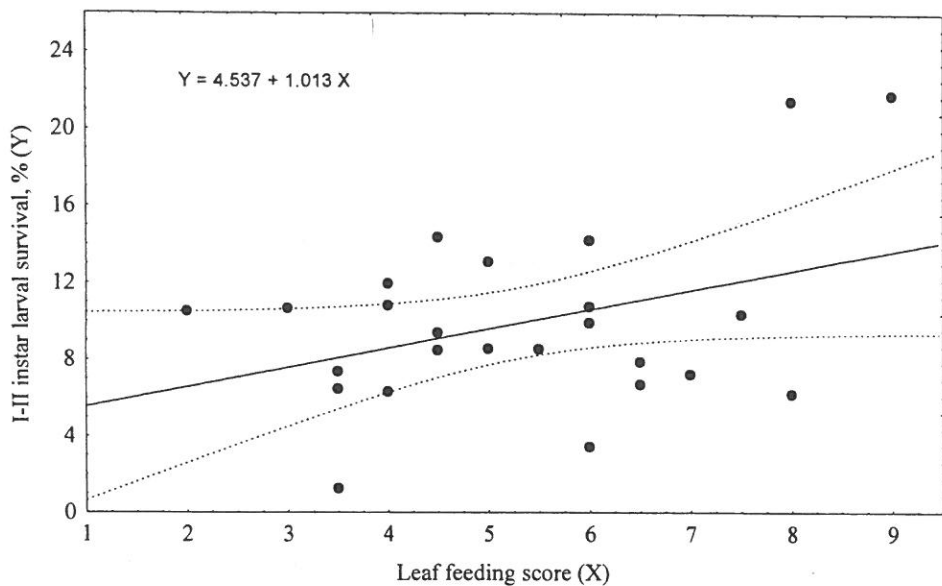


Figure 6. Relationship between the I-II instar larval survival and the host plant resistance in maize varieties measured by leaf feeding scores at KES (1994-1998)

On the other hand, the early instar larval survival demonstrated significant density-dependence proved at $p_a = 0.0001$ (fig. 7).

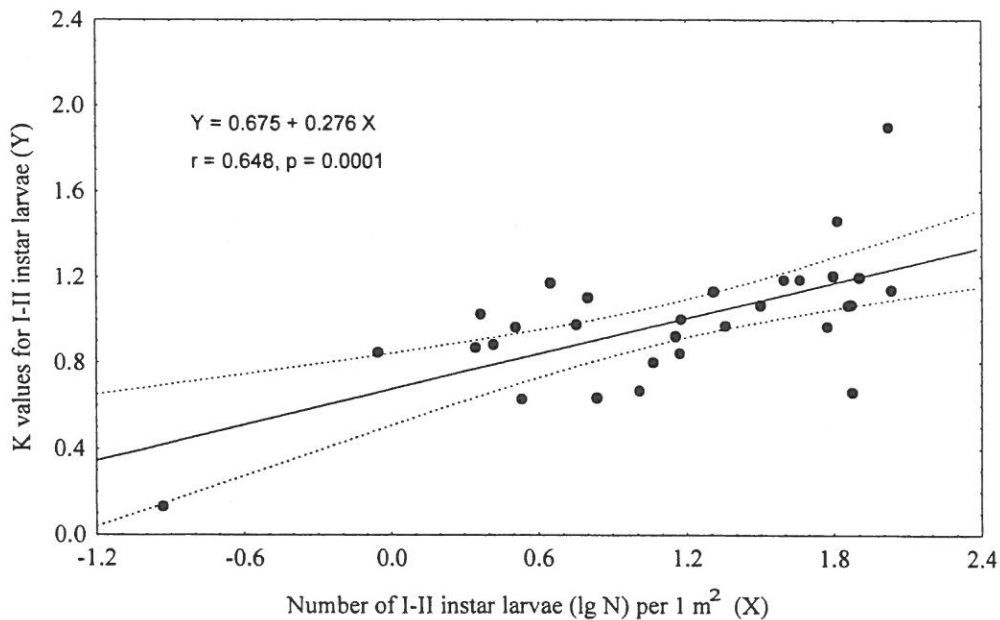


Figure 7. K values in relation with the I-II instar larval densities on maize fields inspected at KES (1994-1998)

In conclusion, the total insect mortality of both the 1st and 2nd generation appeared an evident tendency towards a dependence on initial density of eggs, providing the Krasnodar population with an obvious regulation event (fig. 8).

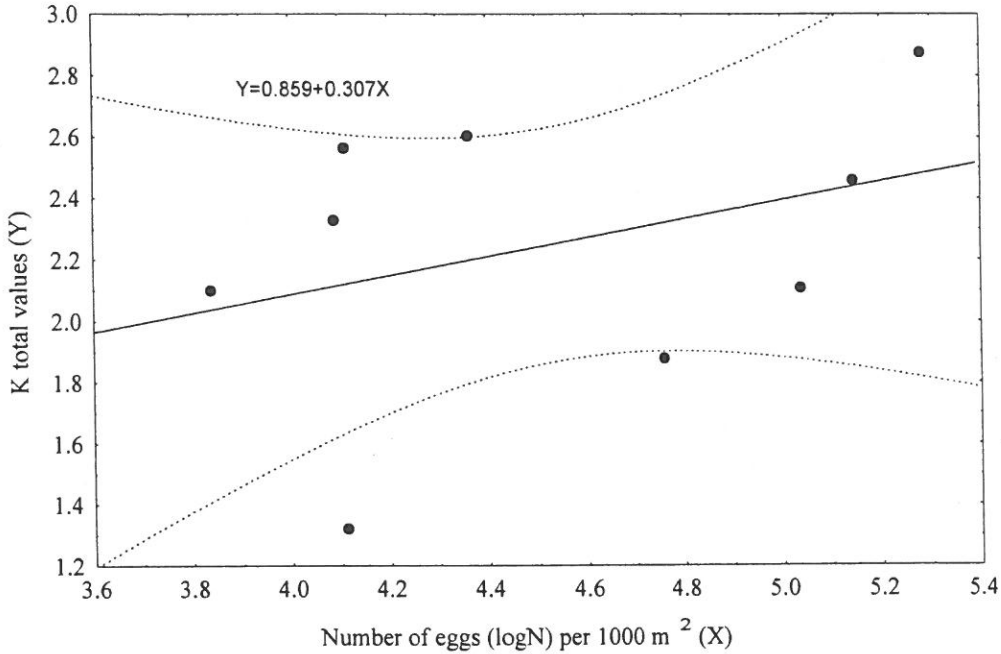


Figure 8. The relationship between total K for a generation and total egg numbers laid (logN) at KES (1994-1998)

The ECB larvae are supposed not to perform a cannibalistic potential. Most probably, the early instar larval density-dependence reflects behavioural reactions of insects to overcrowding; mortality due to predators and/or illness may be another factor promoting density-dependence.

The mortality of adults (normal females) in the Belgorod Province

Life tables involve two main estimates of adult density: 1) the initial number (=normal females) which accounts sex ratio and female fecundity potential before action of different ecological factors, and 2) the final number (=ovipositing females) which is defined by the actual quantity of oviposited eggs. In fact, the difference between expected and actual numbers of eggs in the next generation gives rise to measure drop from initial to final adult numbers during the present generation. Formally, the difference can be considered as adult mortality and/or emigration. However, very many events have happened in adult biology preceding with egg laying. Consequently, a lot of ecological factors may cause population decline during activity of adults. For instance, after eclosion, but before egg laying, adults have to accomplish mating actions and repeated space movements (Showers et al., 1976; Frolov et al., 1996, etc.). So, it seems more correct to speak about a fall in adult capacity to realise its egg production rather than an adult (normal female) mortality. This fall in insect

numbers can come from the effects of different abiotic and biotic factors having influenced both search and choice for mating places; meeting of sexes and copulation; spermatophorous transference; insect movements between aggregation sites and places of oviposition and conversely; searching for appropriate host plants for oviposition, and etc.

The data obtained during 1994-1998 showed that the adult capacity to realise its egg production has tended towards dependence on weather conditions during the ECB flight, i. e. mean air temperature in July ($r = -0.65$) and precipitation in June - July ($r = 0.60$). In spite of quite low r -values, weather effects do not excite any serious doubt (Barlow, 1971; De Rozari et al., 1977; Hudon, LeRoux, 1986, etc.).

On the other hand, the adult capacity to realise its egg production has manifested some tendency towards a density-dependence; the effect seems not statistically proved because of insufficient number of observations (fig. 9). Total egg densities displayed the same tendency; it can be regarded as a result of regulating action of the key factor (fig. 10).

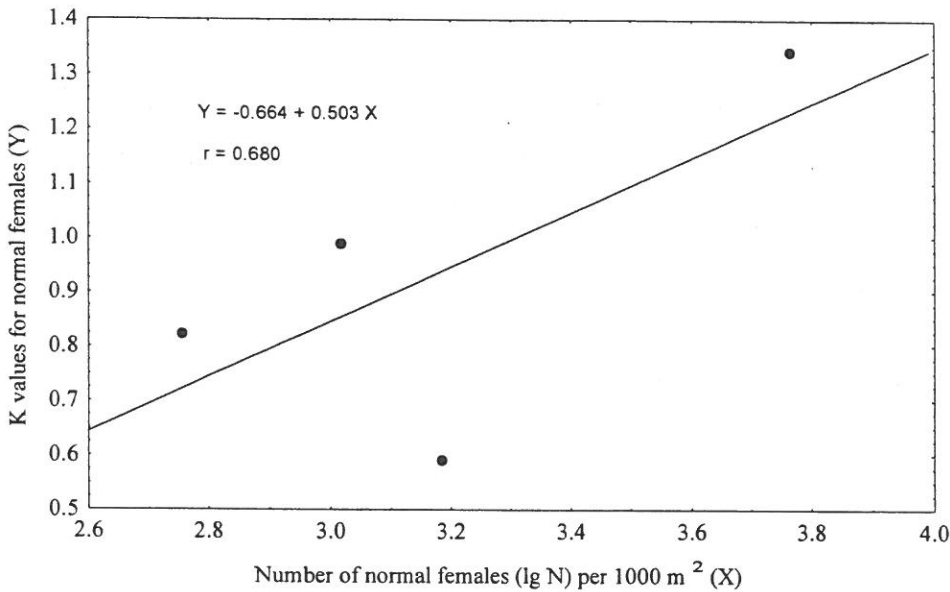


Figure 9. The relationship between a drop in actual egg numbers compared with expected numbers of oviposited eggs at the next generation (K) and the number of ovipositing females of the present generation in BSAA (1994-1997)

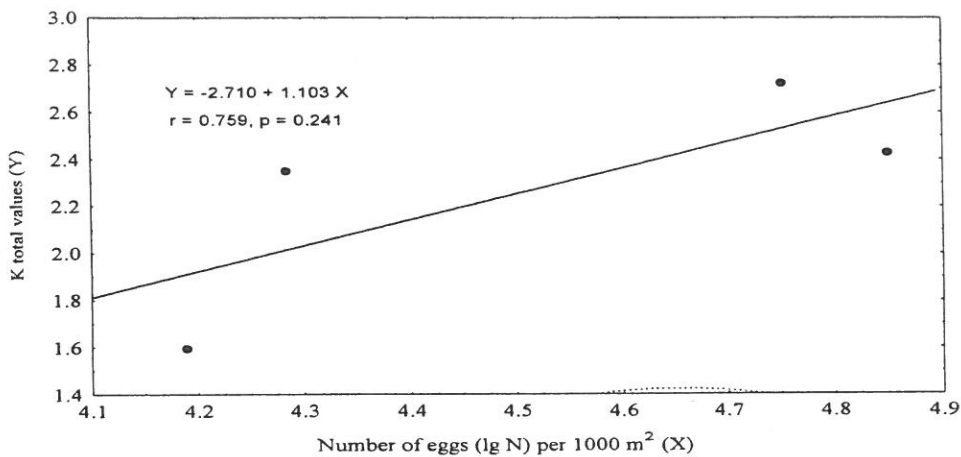


Figure 10. Total K values for a generation in relation with egg numbers laid on maize in BSAA (1994-1997)

An univoltine population dynamics of the ECB was studied in Québec, Canada during 8 years (1957-1965) (Hudon, LeRoux, 1986). The authors made the following conclusions. 1. Within the egg, larval and pupal stages, a population reduction remained relatively low and stable from year to year. 2. A mortality or emigration of gravid females from the study plots ranged from 68 to 98% (mean 95%) and was found the principal key factor affecting Québec population. 3. Weather conditions during egg laying influenced significantly moth mortality. Unfortunately, declared conclusions are not of universal value due to at least two obstacles. First, study plots were located far from commercial maize plantings resulted in a possible enhancement of adult flying away. Second, many common agricultural measures were unused, including those influencing insect numbers dramatically, e. g. tillage cultivation, mowing and threshing at harvest. However, it is of great surprising to find a real similarity in dynamics between the Québec and the Belgorod populations. Consequently, a question arises: either is a density-dependence the characteristic of an adult mortality/emigration in Québec? The answer is easy to obtain through the analysis of life tables being published entirely (Hudon, LeRoux, 1986). Figure 11 shows that the adult capacity to realise its egg production in Québec manifests a density-dependence very significantly. Accordingly, the egg number in Québec tends to be regulated as it does in the Belgorod Province (fig. 12).

Now we may even guess on the mechanism responsible for such a density-dependence in egg realisation by adults. It based most likely on behavioural reactions of adults leaving overpopulated places. Besides, it is not improbable that the growth in insect number can enhance a predator pressure, e. g. birds.

One way or the other identified key factors of the ECB population dynamics characterise with both the density-dependence and density-independence. This combination is ideally suited to the requirements of k-factors typical for herbivorous insects (Hassell et al., 1989). Nonetheless, the density-dependence in the ECB early instar larval mortality when feeding on maize was not detected until the present study, although it was observed when rearing in laboratory (Ramsey, Brown, 1984).

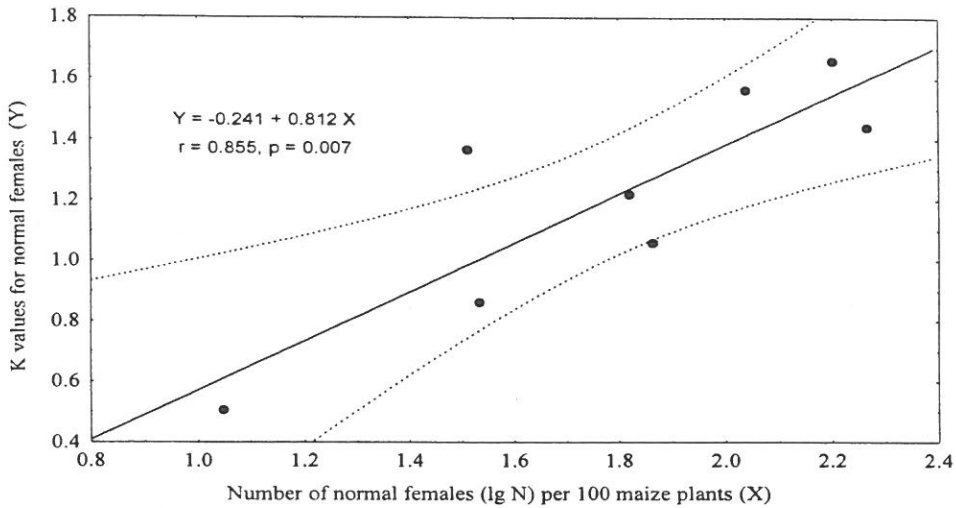


Figure 11. The relationship between a drop in actual egg numbers compared with expected numbers of oviposited eggs at the next generation (K) and the number of ovipositing females of the present generation in Québec (1957-1965) (the handling of data obtained by Hudon & LeRoux (1986))

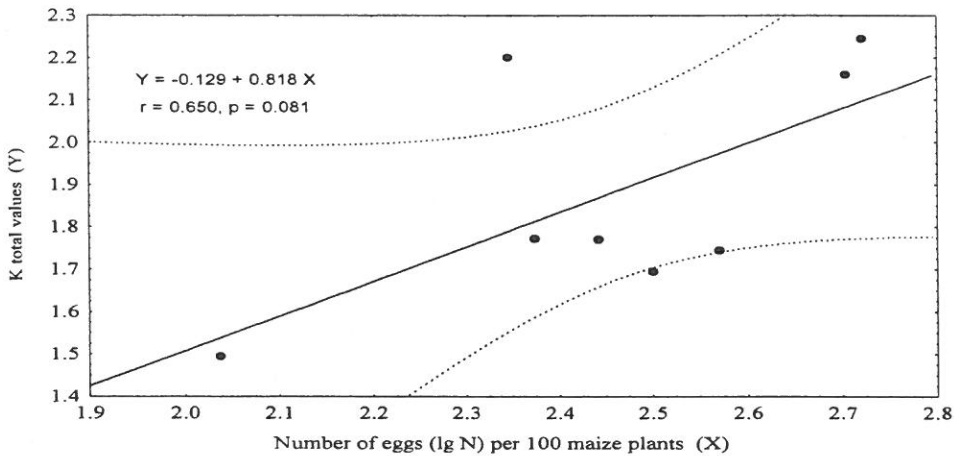


Figure 12. Total K values for a generation in relation with egg numbers laid on maize in Québec (1957-1965) (the handling of data obtained by Hudon & LeRoux (1986))

Plausible causes of the specificity in key factors for two populations tested

It was shown before (tables 1, 2) that the early instar larval mortality and the capacity of adults to realise its egg production made the most significant contribution to the insect death of two populations tested. However, an impact of these mortalities was rather different, and it is not improbable that the only mortality at unique age interval has come into critical importance for dynamics of the population. It means that only one key factor is in operation for the pest at a time.

In any event, the difference between borer populations inhabited the Krasnodar Territory and the Belgorod Province as regards key factors is easy to understand when imagine specificity in insect life cycles and their surroundings.

Feeding of I-II instar larvae is known to be a critical period in the ontogenesis of the ECB. The parts of a plant the insects feed on vary with the stage of growth in maize. Before tasselling, larvae have to feed on whorled leaves, and in such a case their death rate, even on susceptible plant genotypes, can reach 75% and more (e.g. Guthrie et al., 1960). Among the mechanisms of leaf feeding resistance, cyclic hydroxamates such as *2,4-dihydroxy-7-methoxy-1,4-benzoxazin-3-one* (DIMBOA) can be mentioned as fairly well studied (e.g. Klun et al., 1967). At the Krasnodar Territory the great bulk of the 1st generation larvae would have to feed within the leaf whorl. In a consequence of it their mortality is very high almost each year (Frolov, Krapivenko, 1998). In the Belgorod Province a flight of overwintered moths goes on significantly later as compared with the Krasnodar Territory. As a result in average only approximately one-half of I-II instar larvae would be forced to feed on leaf tissues. Consequently, the rate of their death breaks down considerably.

On the other hand, the Belgorod Province and the Krasnodar Territory differ considerably in landscape. In particular, diverse agriculturally unused lands, e.g. ravines overgrown by forest, are rather typical for a forest-steppe in the Belgorod Province. In contrast, almost all the lands in the steppe region of the Krasnodar Territory are used for agricultural production. Besides, maize is the crop of very frequent occurrence at the Krasnodar Territory, as distinct from the Belgorod Province. So, it is quite reasonable to associate higher rate of adult mortality in the Belgorod with the greater efforts exerted by females to search for maize fields which are obviously less common and more hard to reach under more differentiated environment.

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Egg Laying and Survival of the European Corn Borer, *Ostrinia nubilalis*, on Sorghum vs. Maize in Two-Generation Zone of Russia

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Abstract

Although the European corn borer, *Ostrinia nubilalis* (Hbn.) is by far the main pest of maize, it is capable to infest other cereals including sorghum. Less is known about the insect developmental biology on sorghum as well as egg laying preferences to the crop contrary to maize. During 1992-1999 we made observations on natural infestation of sorghum vs. maize as well as conducted artificial infestation experiments at the Kuban Experimental Station, Krasnodar Territory, Russia (two-generation zone of the pest). It is shown that overwintered adults strongly prefer maize for egg laying contrary to sorghum, but during the 1st-generation flight maize is not favoured for egg laying over sorghum. However, larval mortality, especially at early instar is much higher on sorghum opposite maize what was strongly demonstrated from natural infestation observations and an artificial infestation experiment. Some other ecological peculiarities of the insect were demonstrated in respect to its infestation of sorghum.

Key words: European corn borer, *Ostrinia nubilalis*, maize, sorghum, Russia

Introduction

Maize is one of the most important host plants of the European corn borer (ECB), *Ostrinia nubilalis* (Hbn.) all over the world. However, the species is quite polyphagous being capable to attack very many robust herbaceous wild and cultivated plants (Hodgson, 1928, etc.). In particular, it is known to infest cereals besides maize including millet (Dobrodeev, 1921) and sorghum (Painter, Weibel, 1951; Atkins et al., 1983; Wiseman, 1992; Demenichini et al., 1990). The latter, namely broomcorn, seemed to be a carrier promoted the ECB penetration into North America during the I World War from Europe (Smith, 1920).

In North America the ECB is considered as an economically important pest of sorghum (Painter, Weibel, 1951; Dicke et al., 1963). It was shown there that practically all the entries of sorghum are quite resistant to the pest during leaf whorl stage of plant development. Although a level of sorghum resistance usually drops somewhat to flowering, its resistance remains higher than that of maize (Guthrie et al., 1984, 1985; Dharmalingam et al., 1984). Host plant resistance in sorghum is considered to govern by polygenes (Ross et al., 1982); it is a reason to suppose that recurrent selection would be effective (Atkins et al., 1983). Notwithstanding this crop occupying large territories at the North Caucasus and other regions of Russia, host plant relations of the ECB and sorghum have not been adequately studied (Frolov et al., 1995; Andriyash et al., 1995; Dyatlova, Frolov, 1995).

Studying host plant relations of the ECB one must be constantly aware of the fact that the insect is a representative of a complicated taxonomic group which is combined by very

near related forms of the genus *Ostrinia* having trilobed uncus in males (Mutuura, Munroe, 1970).

It was previously proved that the adapted to dicotyledonous host plants populations of *O. nubilalis* form complicated population system together with the naryn borer, *O. narynensis* Mutuura et Munroe, the brush-legged borer, *O. scapularis* (Walker), and the Persian borer, *O. persica* Mutuura et Munroe, at least in the European part of the former USSR. Genetic structure of this population system is appeared to vary according to the range of humidity at territories occupied by borers (Frolov, 1994a, b).

It was proved that a shift of the ECB to maize feeding resulted in a formation of host race at least in the temperate climate of the former USSR. Apart from many distinctions, populations having adapted to maize feeding and those kept their primitive host plant relations with dicotyledonous hosts manifest evident reproductive isolation (Frolov, 1984, 1989, 1994c).

The ECB and the hemp borer, *O. narynensis* × *nubilalis*, inhabit the Krasnodar Territory sympatrically. The ECB develops primarily on maize, whereas dicotyledonous hosts, such as hemp, clotbur, wormwood, and common ragweed are the hosts for the hemp borer (Frolov, 1984). It was observed that larvae of both borers manifested rather strong capacity to feed on their "own" host plants even though they grew adjacent each other (Frolov, unpublished data). A specific morphological feature of *O. narynensis* (the deep invagination on inner surface of male midtibia) is determined by a recessive sex linked gene called *invagination* (*i*) (Frolov, 1984). In populations of the hemp borer frequency of *i* ranges approximately from 0.29 to 0.35; it means that only about 5% of males are expected to perform invaginated midtibiae. The other specific features of *O. narynensis*, namely hair tuft and enlarged curved scales in midtibiae, were possessed also by only few *O. narynensis* × *nubilalis* males. Nonetheless, about 15-20% males of *O. narynensis* × *nubilalis* could be identified morphologically on the basis of mentioned specific characters taken in combination.

So, the first aim of our study is to clear the biotaxonomic nature of insects inhabiting sorghum, and the second one — to investigate insect developmental biology on sorghum compared with maize at the Krasnodar Territory, North Caucasus, Russia.

The first results of this work were reported at the 18th IWGO Conference held in Romania (Dyatlova, Frolov, 1995). The investigation was partly supported by the G. Soros ISF Research Grants # NTH000 and # NTH300, and the RFBR Research Grants # 94-04-11328, # 97-04-48015, and # 99-04-48053.

Materials and Methods

For observation of midtibial morphology in males we used larvae collected in 1985-1998 on sorghum at the Kuban Experimental Station (KES).

An artificial infestation experiment was applied at KES in 1991 to study larval survival depending on origin of the population. Insect stocks were collected during autumn of 1990 at the Krasnodar Territory (KES) on maize and wild dicotyledonous hosts (common clotbur, hemp, and wormwood), in the Kabardino-Balkar (vicinity of Nalchik) on maize and wild dicotyledonous hosts, and at the Ukraine (vicinity of Dnepropetrovsk and Odessa) on wild dicotyledonous hosts. Sorghum variety Kubanskoye krasnoye 1677 was infested with 2 egg masses (=40 eggs) per plant in the middle of whorl stage using 3 replicates and 5 plants per replicate. 20 days later leaf feeding injury was scored (Guthrie et al., 1960) as well as the number of alive larvae per plant dissected was analysed.

To assess the insect – host plant relations we conducted both natural infestation observations on sorghum fields compared with those of maize and an artificial infestation experiment on sorghum. Among plant genotypes used for observations and experiments grain sorghum was represented by the majority of varieties, namely Kubanskoye Krasnoye 1677, Efremovskoye 2, A 83, Yantar' krasnyi 271, and Yantar' Kubanskii. Entries of Sudan grass were few in number: NS, Kubanskaya 183, and Krasnoplenschataya 16/11. Variety Kubanskoye venichnoye was the only representative of broomcorn. Most of maize commercial hybrids were of local breeding (KOS 600, KOS 1089, TOSS 246, KUB 390), and of “Coop de Pau”, France (Nobilis, Alpis, Safaris, Axis, Memphis, Alton). Much more maize inbred lines used for observations were of local origin, namely K 101, K 102, K 111, K 123, K 205, K 395, K 407, K 430, K 347; other inbreds were represented by popular Russian and international genotypes (GK 28, K 611, F 2, F 5, 346, B73). In addition a few parental single crosses (Astra, Lirika, and Lira), one local variety (Urvanskoye beloye) and one popcorn variety (K 550) were used for observations.

In 1992-1993 we inspected the fields occupied by sorghum and maize in the case they grown adjacent each another. Egg masses per plant were counted once (1992) and twice (1993) a peak of oviposition during the 1st and 2nd generation using 5-20 randomised plots (each consisted of 25 plants) per field. 5-10 days after a completion of egg laying the percentage of plant attack was evaluated on the basis of leaf feeding incidence. For this purpose 100 plants (we chose 10 randomised plots each consisted of 10 plants) were observed on each field. 25-30 days later we made estimation of leaf feeding incidence once more. At the same time the number of larvae per plant was counted by dissecting of plants grown on 5-20 randomised plots each consisted of 5 plants.

During 1994-1999 we pursued an assessment of natural infestation of maize and sorghum fields in more detail as follows. Thus, we estimated total numbers of eggs laid both on sorghum and maize, by making a series of sequential counts on spatially fixed plots. Each egg mass found on foliage was marked and a hand lens was used to count the number of eggs as either hatched successfully, or dead due to parasitism, predation, dislodgement, failure to produce embryo, and larval inability to hatch. Each plot was examined for 6-8 times in 5-7 day intervals during a period of moth flight. To estimate the whole numbers of eggs laid as well as the 1st instar larvae hatched we summed up corresponding values found on a plot during successive observations. To raise comparability, both egg and larval densities on a crop were performed on the basis of m^2 because of sorghum plantings populated usually 2-3 times densely than those of maize. Unlike observations made in 1992-1993, we chose for observations fields of both crops even though they were nonadjacent each other.

Overall number of fields inspected was 13 for sorghum and 25 for maize. Periodical surveys of plants to search for eggs were done at 7-23 fixed plots (averaged in size $2.5 m^2$) per field. After a week upon a completion of egg laying we sampled late instar larvae by dissecting plants grown at 10-50 randomised plots sized in average of $1.4 m^2$.

To judge larval survival on sorghum an artificial experiment was carried out in 1992. Each plant (variety Kubanskoye krasnoye 1677) was infested in the middle of whorl stage with 2 egg masses (=40 eggs). 13-20 plants were dissected by 3, 12, 20, and 26 days after infestation and the number of alive larvae per plant was counted as well as places of larval feeding were recorded.

Larval survival according to whether the insect parents fed on maize or sorghum was estimated in 1990 and 1992 by using an artificial infestation experiment. Parental stocks

were collected during autumn of the previous year. Sorghum variety Kubanskoye krasnoye 1677 was infested during the middle whorl stage of plant development. In 1990 we infested plants at the rate of 20 eggs per plant; plant dissections and counts of larvae were made 5 days later. In 1992 plants were infested with 40 eggs per plant and plant were dissected 40 days after infestation. In addition to the number of larvae we assessed score of leaf feeding damage after Guthrie et al. (1960).

Results and Discussion

Both fed on sorghum and maize male adults possessed with midtibiae typical for *O. nubilalis* (in total 125 specimens collected during 1985-1998 were observed). However, significant differences were found when they were compared to males of the hemp borer collected on dicotyledonous hosts.

An artificial infestation experiment evidenced that progenies of collected on maize insects survived on sorghum much better than those of collected on dicotyledonous hosts (table 1). The hemp borer from the North Caucasus as low survived on sorghum as the brush-legged borer or the ECB race kept its original trophic relations to dicotyledonous hosts.

Table 1. Leaf feeding scores and larval densities under artificial infestation of sorghum with egg masses produced by insects collected on maize or dicotyledonous host plants at different localities of the former USSR (KES, 1991)

Borer	Stock origin	Leaf feeding score	Number of larvae per plant
The ECB, <i>O. nubilalis</i>	The Krasnodar Territory, from maize	2.8	0.4
The ECB, <i>O. nubilalis</i>	Kabardino-Balkar, from maize	3.0	0.7
The hemp borer, <i>O. narynensis</i> × <i>nubilalis</i>	The Krasnodar Territory, from dicotyledonous hosts	1.8	0.1
The ECB, <i>O. nubilalis</i>	Kabardino-Balkar, from dicotyledonous hosts	1.8	0.2
The brush-legged borer, <i>O. scapularis</i>	Dnepropetrovsk, from dicotyledonous hosts	1.6	0.1
The ECB, <i>O. nubilalis</i>	Odessa, from dicotyledonous hosts	1.8	0.1

All the mentioned above materials give rise to real suggestion that in the North Caucasus sorghum in couple with maize has infested by just the same population of the ECB specialised to rear on cereals.

Preliminary observations on natural infestation were conducted during 1992-1993 on fields where maize and sorghum grew in close vicinity to each other. On the whole, both overwintered and 1st generation adults attracted actively by sorghum plants to lay eggs on.

Thus, densities of egg masses recorded during an oviposition peak as well as percentages of leaf feeding damage were almost equal in sorghum and maize (table 2, 3).

Table 2. Natural infestation and injury caused by the 1st generation of the ECB to near-by fields of maize and sorghum (KES, 1992-1993)

Genotype	Field acreage, ha	Plants with leaves damaged, %	Number per plant		
			egg masses	holes	larvae
SORGHUM					
1992					
Kubanskoye	8	65.4±0.05	–	–	0.05±0.04
Krasnoye 1677					
Efremovskoye 2	35	71.0±6.40	–	0.05±0.02	0.02±0.01
Efremovskoye 2	20	71.0±4.92	–	0.03±0.02	0.10±0.03
1993					
A 83	0.5	59.0±7.8	0.36±0.09	0.04±0.04	0.12±0.05
Yantar' Kubanskii	0.3	46.0±12.1	0.17±0.04	0.02±0.02	0.08±0.04
Efremovskoye 2	5.0	43.0±9.8	0.20±0.04	0	0
Average		59.2	0.24	0.03	0.06
MAIZE					
1992					
K 101	0.4	34.0±7.50	–	1.75±1.98	0.54±0.78
F 2	9	79.0±4.30	–	2.24±0.32	0.94±0.67
K 550	5	18.8±2.47	–	0.12±0.03	0.08±0.03
1993					
K 101	0.5	73.0±5.0	0.66±0.12	1.92±0.50	0.48±0.17
F 2	0.8	60.0±9.0	0.38±0.06	1.88±0.37	0.48±0.15
B 73	1.5	18.0±3.9	0.14±0.10	0.40±0.13	0.37±0.11
KOS 600, KOS 1089, TOSS 246	45.5	56.0±9.3	0.16±0.04	0.50±0.21	0.28±0.08
KOS 1089	10.0	50.0±7.6	0.40±0.04	0.33±0.13	0.03±0.03
Average		48.6	0.34	1.14	0.40

An artificial infestation experiment was used to estimate the rate of larval mortality on sorghum beginning with leaf whorl stage of plant development. The conclusion on rather high larval mortality on sorghum was confirmed: for the first 3 days after infestation larval death exceeded over 96%. At that time all the larvae were still developing at the 1st instar and fed on whorled leaves; most of infested plants carried one larva at least. Later, the number of larvae as well as percentage of infested plants decreased, most heavily during broom emergence in the middle of July when the insect mortality just exceeded over 50%. During this time larvae passed for the most part into leaf sheaths and central veins and a few penetrated into stalks (fig. 1).

The ECB larval feeding on maize has been studied carefully (e. g. Beck, 1987). Most larvae perish at the 1st instar for three initial days of their feeding, but later insect number gets stabilisation (e. g. Buske, Witkowski, 1985). Larvae at early instar need proteins especially; accordingly their feeding on tassel spikelets is favourable for insect survival. Since the 4th instar larvae are able to bore stalk for tunnelling, and during the 5th instar tunnels represent the place where larvae feed most often until pupation.

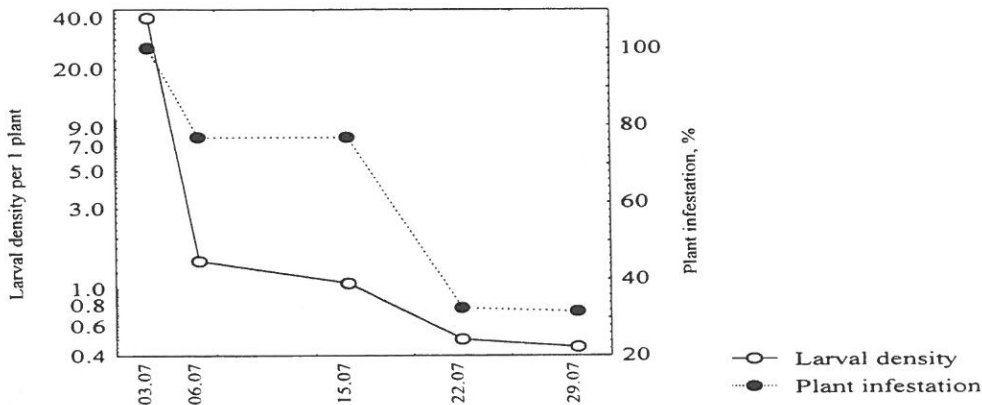


Figure 1. Dynamics of the ECB larval survival and plant infestation under an artificial infestation experiment on sorghum (KES, 1992)

Table 3. Natural infestation caused by the 2nd generation of the ECB to near-by fields of maize and sorghum (KES, 1992-1993).

Genotype	Field acreage, ha	Number of egg masses per plant
SORGHUM		
1992		
Kubanskoye Krasnoye 1677	8	0.80±0.13
Efremovskoye 2	35	0.10±0.02
Efremovskoye 2	20	0.31±0.06
1993		
A 83	0.5	0.09±0.07
Breeding entries	6.0	0.29±0.03
Yantar' Kubanskii	0.3	0.14±0.04
Efremovskoye 2	5.0	0.07±0.03
Efremovskoye 2 ×	18.0	0.11±0.04
Krasnoplentchataya 11/16		
Efremovskoye 2	3.0	0.07±0.04
Krasnoplentchataya 16/11	2.0	0.06±0.02
Average		0.20

MAIZE

	1992	
K 101	0.4	0.43±0.08
F 2	9	0.05±0.01
K 550	5	0.36±0.08
	1993	
K 101	0.5	0.11±0.02
Breeding entries	0.8	0.24±0.09
F 2	0.8	0.02±0.07
B 73	1.5	0.66±0.08
B73 × GK 28	5.0	0.28±0.07
GK 28	5.0	0.22±0.06
KOS 600	3.0	0.16±0.05
KOS 1089	10.0	0.36±0.09
Average		0.26

Thus, dynamics of larval mortality as well as feeding places on sorghum are quite different from those on maize.

Further attention was devoted to quantifying of larval survival on sorghum and maize under natural infestation. Having in mind this purpose, since 1994 we have been estimating total numbers of eggs laid both on sorghum and maize per 1 m² of sowing. Considerable variation in egg and larval numbers was shown between fields, years, and insect generations (table 4). Nevertheless, the averaged data evidenced that the mean density of 1st generation eggs laid on sorghum was 7.2 times less than it was on maize. The mean egg mortality on sorghum was rather less, perhaps, due to their lesser density. In accord, the 1st instar larval density on sorghum was 4.9 times as low as on maize. Since the average larval mortality for 1st-2nd instar was to a marked degree bigger on sorghum (96.6%) than on maize (91.4%), the mean larval density at late instar became 12.3 times greater on the latter host plant. As for the 2nd generation, the average level of egg mortality turned out to be somewhat higher on sorghum (70.1%) as compared to maize (56.6%). Despite rather slight difference in the 1st instar larval densities between sorghum (23.3) and maize (37.0), the late instar larval density on the former host plant (1.14) was 13.4 times less than on the latter (15.32). The remarkable difference was originated from much more great larval mortality for early instar on sorghum (95.1%) opposite maize (58.7%).

Sorghum becomes to attract overwintered moths for egg laying significantly later when compared to maize. It is quite appropriate to explain the low numbers of 1st generation eggs laid on sorghum with two reasons as follows. First, sorghum is sowed usually 15-20 days later than maize, and second, an initial growth of sorghum seedlings is rather slow. However, temporal differences in egg laying on maize and sorghum completely disappear until flying of the 1st generation adults. The results obtained in 1997 are given as an illustration of egg laying dynamics on sorghum and maize (fig. 2).

Table 4. The estimation of the ECB larval survival on sorghum and maize under natural infestation (KES, 1994-1999).

Genotype	Field acreage ha	Plant population m ⁻²	Total insect density, m ⁻²					
			1st generation			2nd generation		
			eggs	larvae		eggs	larvae	
				I instar	III-V instar		I instar	III-V instar
SORGHUM								
1994								
Yantar' krasnyi 271	0.3	10.8	0.46	0.25	0.00	1.28	1.02	0.00
Efremovskoye 2	0.4	15.5	0.52	0.26	0.03	—	—	—
A83 × NS	1	13.9	0.36	0.21	0.01	20.87	5.21	0.00
A83 × NS 1995	1	13.0	1.35	1.28	0.00	—	—	—
Kubanskoye krasnoye 1677	0.1	4.0	—	—	—	10.74	8.86	0.278
Kubanskaya 183	2	16.2	—	—	—	20.40	18.99	0.400
1996								
Kubanskoye venichnoye	4.2	15.3	9.39	9.39	0.33	49.37	34.92	2.68
Kubanskaya 183	2	25.6	3.70	3.70	0.00	—	—	—
Kubanskaya 183	5.5	216.1	16.89	16.89	0.96	—	—	—
1997								
Efremovskoye 2 × Krasnoplachat aya 16/11	15	15.8	51.06	36.07	0.93	280.43	35.93	3.93
1998								
Kubanskoye krasnoye 1677	0.02	6.3	0.91	0.64	0.00	164.60	73.65	1.19
1999								
Efremovskoye 2	0.4	4.6	1.40	1.22	0.18	76.19	7.85	0.65
Efremovskoye 2	4.5	8.5	3.18	1.19	0.00	—	—	—
Average			8.11	6.46	0.22	77.98	23.30	1.14

MAIZE								
1994								
K 407	0.5	5.0	18.33	6.34	0.50	4.55	2.76	0.85
K 611, K 102	0.5	4.6	12.41	4.46	0.30	8.86	4.88	1.52
346	1	4.9	8.75	2.33	0.22	2.91	2.27	2.15
Astra × K 101	18	5.1	24.84	15.63	1.85	1.94	1.31	0.84
TOSS 246	10	5.2	38.35	20.47	1.51	12.75	5.52	2.47
F 5	1	4.7	37.46	20.82	1.33	5.40	2.65	1.28
1995								
K 430	0.5	4.6	—	—	—	8.50	6.24	4.12
K 407	1	2.8	—	—	—	6.21	4.21	2.84
K 611	0.3	4.2	—	—	—	3.39	1.58	1.00
(B73 × GK 28) × GK 28	1	2.9	—	—	—	3.95	3.26	1.54
1996								
(B73 × GK 28) × GK 28	2	4.5	11.02	8.92	2.25	118.99	73.80	28.81
K 430 × K 347	1	5.1	6.23	5.48	0.59	193.45	127.88	62.23
K 430	2	4.4	3.68	2.60	0.85	115.69	82.54	49.02
Lirika × F 5	1	4.2	14.63	8.06	1.54	110.57	68.14	30.83
1997								
K 123, K 395	1	4.3	96.80	53.24	0.19	79.55	39.17	12.00
K 111, K 611	1	4.4	30.56	19.54	0.11	168.72	56.71	14.07
Nobilis, Alpis, Safaris, Axis, Memphis, Alton	4	5.7	82.77	63.10	0.24	123.78	55.88	27.67
1998								
Urvanskoye beyelo	2	5.0	189.41	107.43	7.79	284.25	91.07	20.63
Lira × K 205	20	3.6	89.25	39.44	2.57	190.08	50.23	11.96
K 395	2	7.3	228.64	75.24	16.39	47.58	19.17	11.00
TOSS 246	22	5.1	—	—	—	139.73	46.90	17.04
1999								
Lirika × F 5	18	4.8	13.29	9.13	0.43	132.30	48.26	19.57
KUB 390	3	5.8	18.98	9.62	1.43	—	—	—
B 73 × GK 28	2	5.6	82.26	49.18	5.29	—	—	—
K 611	1	4.9	167.03	113.68	9.29	201.97	57.70	28.80
Average			58.74	31.74	2.73	85.44	37.05	15.32

The specificity in ovipositional terms of overwintered females is not a unique reason for necessity of comparing infestation levels of sorghum and maize on the basis of counting of total egg numbers. It is variation in average size of egg masses laid on different crops that represents another factor of indignation.

It is well actually known that the ECB moths lay their eggs grouping into clusters which vary in size from 1 to 180 and more eggs per one mass (Shchegolev, 1934; Caffrey, Worthley, 1927; Hudon, LeRoux, 1986, and so on). Curiously different estimates of mean

size of egg masses were reported, e. g. 10-25 eggs (Khomyakova, 1962), 12-32 (Andreeva, 1930), 15-20 (Caffrey, Worthley, 1927).

During an examination of plants in 1993-1999 we found totally 30715 egg masses on maize and 1118 on sorghum. The average number of eggs per mass depends to a great extent on a host plant species (more often larger masses were recorded on sorghum) as well as a generation (the 2nd generation masses were much bigger when laid on both maize and sorghum) (fig. 3).

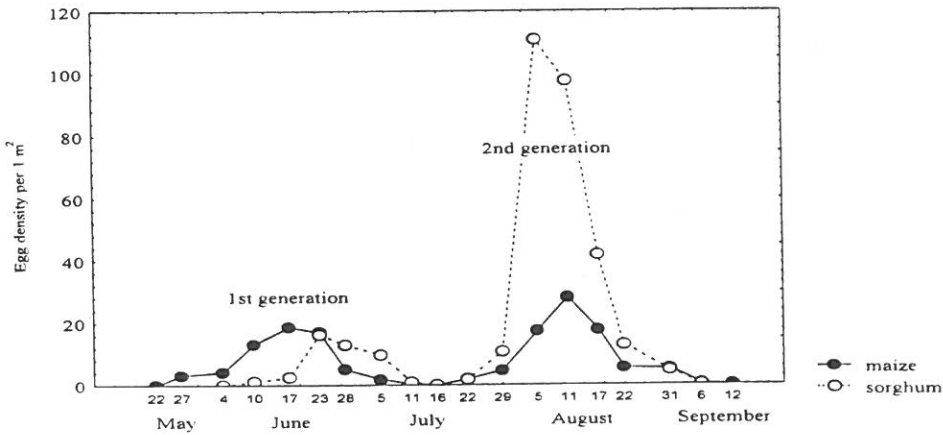


Figure 2. Ovipositional dynamics for the 1st and 2nd generations of the ECB on maize and sorghum (KES, 1997)

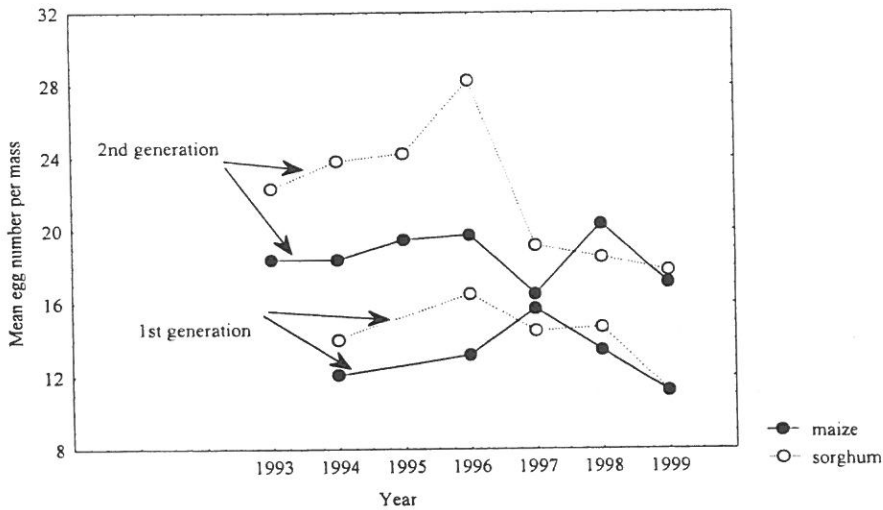


Figure 3. Average numbers of eggs per mass laid on sorghum and maize during the 1st and 2nd generations of the ECB (KES, 1993-1999).

Therefore, the ECB inhabitancy on sorghum is rather specific in many respects. The most curious peculiarity is a conflict combination of low larval survival along with a high

attractiveness to adults for egg laying. It seems to be interesting from an evolutionary point of view. On the one hand, a selection should act against the moths if their offspring is not adapted to sorghum feeding. On the other, if some adaptive features to larval feeding on sorghum occurred, they would catch up due to a selection. One can imagine that such a combination may provoke somewhat like a disruptive selection when the insect has a choice to exploit either sorghum or maize. If so a selection would segregate a population onto specifically adapted to maize and sorghum subpopulations.

Some data obtained after an artificial infestation experiment (table 5) indicated that the formation of host race specialised on sorghum could realise in the ECB under certain conditions. Nevertheless, some obstacles prevent such a process. In particular, larval growth on sorghum is rather slow during the 1st generation as opposed to maize. Actually, during the end of July – the early of August most of insects of the 1st generation fed on sorghum still remain at the larval stage and only a small part of them start pupation. By contrast, at that time most insects fed on maize represent ovipositing adults. As a rule the dates of egg laying on maize and sorghum are coincident during the 2nd generation. So, renewal of sorghum population occurs to a maximum each year from eggs laid by insects reared on maize. Another obstacle, preventing a formation of sorghum specialised race, could be very late happening of developmental stages in sorghum suitable for egg laying by overwintered adults.

Table 5. Survival of the ECB larvae on sorghum under an artificial infestation depending on a host plant of insect ancestors (KES, 1990, 1992).

Insect stock originated from	Leaf feeding score	Larval density per 1 plant
	<u>1990</u>	
Maize	6.1	4.4
Sorghum	6.2	5.9
	<u>1992</u>	
Maize	2.6	0.07
Sorghum	3.7	0.67

Although dicotyledonous plants of the European origin such as hop and mugwort were most probably the initial hosts for the ECB, at present cereals are infested more often and much intensive. Among them common millet, *Panicum miliaceum*, and Italian millet, *Setaria italica*, are certain to be the most ancient cereals which had spread wide over Asia and Europe even in neolite (Lysov, 1968). Sorghum is considered the plant of African origin and it was introduced into the European agriculture much more recently, approximately in 700-400 B. C. (Doggett, 1970). The Europeans had never been familiar to maize until Columbus discovered the Americas as recently as about 500 years ago; besides, the crop occupied considerable acreage towards the 17th century only (Dekapreleevich, 1960). Despite much more extended period of coexistence with sorghum the ECB manifests much less fitness when feeding on this plant contrary to maize.

In fact, the data obtained agree nicely with published materials indicated of worse rearing conditions for larvae and consequently their higher mortality when feeding on sorghum. As a rule, full-grown larvae are sparsely distributed on sorghum, even though very high numbers of eggs laid on the crop. In other words, borer adults can choose sorghum for egg laying very actively, in spite of low feeding quality of the plant for their offspring.

The search, choice, and determination of host availability by an insect for egg laying are very complicated processes specific for different species. From an evolutionary point of view ovipositing behaviour is aimed to make as good as possible compromise between risk spreading (e.g. Root, Kareiva, 1984), making choice for better host for posterity (e.g. Papaj, Rausher, 1987), providing optimal density for offspring over hosts (e.g. Schoonhoven et al., 1990), and realising maternal fecundity to a maximum. More often (Tabashnik et al., 1981; Scriber, Dowell, 1991, etc.), but not necessarily (Dethier, 1959; Courtney, 1981, etc.) females prefer for egg laying to those hosts which are the most appropriate for offspring development. Despite of distinction of criteria used by adults and larvae for host acceptance as exemplified by a specificity of genetic background (Thompson et al., 1990), one could very often determine significant covariation between a choice of host plant for oviposition by adults and its suitability for feeding by larvae (e.g. Via, 1986). Suboptimal and even inappropriate ovipositing on a host plant by an adult resulted in poor feeding by offspring (e.g. Åhman, 1986) can be traced to a number of reasons (Chew, 1977); violating the principle of optimality in host plant preferences by ovipositing adult (Kreslavsky-Smirnov, 1987) is readily apparent from the predicting essence of estimation of host availability for offspring in the future.

As a rule, one determines preferred host plant species for herbivorous insect on the basis of larval attack. However, it is of no less value also to test host plant preferences on the basis of egg laying as it follows from the data presented above.

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Different Possibilities of Using Pheromones for Researches and Control of European Corn Borer, *Ostrinia nubilalis* Hb. in Romania

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Abstract

In Romania at Research Chemistry Institute "Raluca Ripan" - Cluj-Napoca, were developed many pheromones variants for different pest species, but only 25 were registered till now in practice. They are used especially for monitoring of pests, but there are experiments on large scale for using pheromones in control of orchards and vineyards pest. It is presented different ways in which these registered pheromones are used in agricultural practice in Romania. The paper presents results obtained in Romania during 1990-1997 in field trials with pheromone formulations developed for European Corn Borer (*Ostrinia nubilalis* Hb.). Pheromone trap is helpfulness in a future integrated control system of pest by determination of releasing time of males with inherited sterility and oophagous parasites - *Trichogramma* spp. In order to develop an insect pest management, a special attention was paid to the investigations on the ECB dynamics and the estimation of the natural population by means of pheromone traps and markers. It was used synthetic sex pheromone like a monitor pest flight.

Key Words: pheromones, flight monitoring, control

Introduction

In Romania at Research Chemistry Institute "Raluca Ripan" - Cluj-Napoca, were developed many pheromones variants for different pest species, but only 25 were registered till now in practice. They are used especially for monitoring of pests, but there are experiments on large scale for using pheromones in control of orchards and vineyards pest (Hodosan and Oprean, 1979; Ghizdavu, Tomescu and Oprean, 1983; Rosca et al., 1990 A; 1991).

As a consequence of preliminary results recorded during 1982 -1987, from 4 promising pheromone variants were found and from these we choose two, namely: E₅ (Z 11-14OAc + E 11-14OAc at a ratio of 97/3) and I (Z 11- Oac + E 11 - 14 Oac at a ratio of 3/97 + tetradecenyl acetate) and it is very important to establish which is the spreading of these two pherotypes and their proportions (Rosca et al., 1990B; Rosca and Barbulescu, 1997).

The paper presents the results obtained in Romania during 1990-1997 in field trials with pheromone formulation development for European Corn Borer (*Ostrinia nubilalis* Hb.).

Pheromone traps can be used to draw up flight curves of *O. nubilalis* males in order to determine the moment for releasing entomophagous. It is important to determine the level of first and second population of *O. nubilalis* in Romania and pheromone traps could realize this.

A special attention was paid to the investigations on the ECB dynamics and the estimation of the natural population by means of pheromone traps and markers. It is obvious that information on the flight of ECB is highly important and that knowledge of the dispersion and flight range of any insect in all its aspects is meagre indeed and for this reason a great part of our researches were directed toward to establish the distance to which a moth may

fly. Few things were known about the flight distance of ECB. Jermy and Nagy considered that in nature, the adults could migrate on long distances.

It is also very important to know if control of ECB by mass trapping of males or male disorientation could be a tool in integrated control of pest.

Estimate of ECB population is difficult. Tagging large numbers of individual moths and employing a marking/recapture method for estimating the density is a method by which a lot of troubles can be solved. The population density is calculated using the formula of Jenkins.

Material and Methods

Pheromone sticky traps type F-1 (Ghizdavu and Rosca, 1986) were used to draw up flight curves of the two pherotypes (Z and E) of *O. nubilalis* males in the first generation and also in the second one in the years and localities where this appears. The traps were situated from 50 to 50 m. in 4 replicates. Seeking firstly for increased pheromone efficiency, the pheromone lures were changed every 2 weeks, counting of the numbers of ECB captured males/trap was done weekly.

Dye marking method is usually made with an emulsification mixture of Calco oil red N-1700 [150 mg/kg diet (0.015%)] and sunflower oil, mixed homogeneously in the agar of the semisynthetic diet for ECB. The larvae and moths were marked after eating the diet.

Releasing and recapture of marked moths was used in order to study behavior of males, released in field and recapture in pheromone traps situated at 1000, 2000, 3000, 4000 and 5000 m in 1997, far from releasing site in N, S, E and W direction.

Five *Trichogramma* releasing, 200,000 parasites/ha each, like plates of 800 eggs of *Ephesttia kühniella* with parasite, from 7 to 7 m on and between rows.

Results

The pheromones for *Ostrinia nubilalis* Hb., proved to be relatively efficient and specific, obtaining in average for the period 1990-1997, 39% exemplars of target species, 5.5% macrolepidoptera and 55.5% microlepidoptera. Specificity of pheromone formulations are not optimal, since other microlepidoptera like in forested areas *Tortrix viridana* L. is also frequently caught and *Etiella zinkenella* Tr. and *Emelia trabealis* Scop. in the other zones, however these species can be easily separated from the target species.

Biological trials with this pest pursuing capture variation in different localities, years and life cycles, showed that there are significant differences between captures in various years and localities. Broad variations occurred depending on the life cycle, which could be properly established with pheromone traps.

The results from table 1 reveals the relatively high number of *O. nubilalis* males were captured throughout the period under study, this number varying according to year and pheromone variant used. It is to stress that it was undoubtedly that in Romania both the pherotype of this species, CIS (Z) and TRANS (E) exist. The biggest number of males captured/trap [E_5 (Z 11-14Oac + E 11-14Oac at a ratio of 97/3)], 35.25 was registered in 1994.

Table 1. Number of *Ostrinia nubilalis* Hb. males captured/ trap/year – FUNDULEA

Pherotype	YEAR							
	1990	1991	1992	1993	1994	1995	1996	1997
E	1.75	1.25	0.75	2.75	1.5	0.5	0	0
Z	8.25	14.75	23.25	29.5	35.25	17.2	10.5	13.25

Pheromone traps can be used to draw up flight curves of *O. nubilalis* males and to determine importance, appearance time and level of the first and second generation (Table 2). Results underline that the second generation generally which occur in Romania at the end of August or early in September and practically is devoid of significance.

Table 2. Importance of first and second generation of *Ostrinia nubilalis* Hb. determined with synthetic sexual pheromone at Fundulea

YEAR	1990	1991	1992	1993	1994	1995	1996	1997
TOTAL	8.25	14.75	23.25	29.5	35.25	17.25	10.5	13.25
First generation	5.5	10	19.25	21	28	8.75	8.25	11.5
Second generation	2.75	4.75	4	8.5	7.25	8.5	2.25	1.75

Control of ECB by mass trapping of males or even by male disorientation has failed in small corn field surrounded by forest. Using releasing and recapture marked moth technique, it is possible to estimate the population of ECB in that particular area at one period of time.

Experiments from 1996 and 1997 have shown that released tagged ECB moths have been recaptured in pheromone traps at a distance no more than 3000 m from the point of release, and probably this is the distance for which the moths are capable to fly.

Under climate conditions from Fundulea, a number of five *Trichogramma* releasing, 200,000 parasites/ha each, were required to reduce the ECB population with 88.2% in 1993 with 38.1% in 1994 and with only 8.6 in 1995, table 3. Even that population of larvae has decreased after treatment with *Trichogramma* spp., price and effort which has to be paid for releasing entomophagous, seems to be inapplicable for corn in southern part of Romania.

Table 3. Effect of using *Trichogramma* spp. in corn fields at Fundulea

Variant	Attacked stems (%)			No. larvae/ attacked plant			Level of population (%)		
	1993	1994	1995	1993	1994	1995	1993	1994	1995
<i>Trichogramma</i> spp. 5 releases of 200,000 ex/ha	6.0	26.0	86.7	0.72	2.0	2.77	- 88.2	- 33.3	- 8.6
Check	26.5	36.5	91.3	1.38	2.3	2.88	-	-	-

Conclusions

In Romania it were created and registered a large variety of pheromones, part of them are used in field crops, but now the price become restrictive.

The pheromone for *Ostrinia nubilalis* Hb., proved to be less efficient and specific, but they could be used to determine the area of pest, dynamic of pest flight, the structure and size of different generation and of course time evolution of pest in time and space and the moment for releasing of entomophagous

Control of ECB by mass trapping of males or even by male disorientation has failed in small cornfield surrounded by forest.

Using releasing and recapture marked moth technique, seems to be possible to study behavior of pest in field.

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Female-Biased Sex Ratio in *Ostrinia furnacalis* and *O. scapularis*

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Abstract

We have already reported the occurrence of female-biased sex ratio in the Japanese population of the Asian corn borer, *Ostrinia furnacalis*. Here we report that feminization of genetic males caused by bacterial infection is one of the underlying mechanisms for this phenomenon. Three out of 13 females collected at Matsudo, Japan produced offspring with significantly female-biased sex ratio. The female-biased sex ratio was maternally inherited. Treatment of a maternal line that produces only daughters with tetracycline for two generations resulted in production of only male progeny. This indicated that the genetic males were feminized in the maternal line. PCR assay revealed that this feminized maternal line and another line that showed similar maternal inheritance of female-biased sex ratio were infected with *Wolbachia*. *Wolbachia* is a group of bacteria that belongs to the alpha-Proteobacteria, and is known to infect a wide range of invertebrates and manipulate their hosts reproduction in various ways such as cytoplasmic incompatibility, parthenogenesis, feminization and male killing. Feminization of Asian corn borer caused by *Wolbachia* infection found in the present study is the first case in insects.

Key words: feminization, Lepidoptera, *Ostrinia*, sex ratio, *Wolbachia*.

Introduction

Female-biased sex ratios are known to occur in many arthropods, including insects, mites and crustaceans. Many of them are known to be caused by cytoplasmic parasites such as bacteria, viruses or protozoa (Hurst 1993), while female-biased sex ratio due to meiotic drive caused by nuclear factors has been also observed in insects, e.g., several species in *Drosophila* (Mercot et al., 1995; Jaenike, 1996). Major mechanisms for the female-biased sex ratio caused by cytoplasmic parasites can be classified into three categories. 1) Male killing: only male offspring are killed at the immature stage, which is observed in many insects and well studied in ladybird beetles (Hurst et al., 1996). 2) Induction of parthenogenesis (thelytoky): virgin females will produce only daughters, which is observed in several species in Hymenoptera, frequently observed in parasitic wasps and well studied in *Trichogramma* (Stouthamer and Werren, 1993). 3) Feminization: potential males are changed to females, which is observed in crustaceans (Hurst 1993) and well studied in wood louse, *Armadillidium* spp. (Rigaud et al, 1997), and shrimps of *Gammarus* (Dunn, 1993). Since cytoplasmic parasites are transovarially transmitted to subsequent generations of their hosts and not transmitted via sperm, those parasites that distort their host sex ratio toward females will be selectively favored.

These reproductive manipulations are caused by many kinds of microorganisms. *Wolbachia* is a group of bacteria that are known to be concerned with all of the three categories of reproductive alterations. Infection with *Wolbachia* is widespread in arthropods, and many biologists are recently interested in this group of bacteria (reviewed by Werren, 1997).

Female-biased sex ratio in *Ostrinia furnacalis* was observed in several locations over Japan (Miyahara, 1984). However, the underlying mechanism(s), mode(s) of inheritance and causal agent(s) of this trait have remained to be solved in *O. furnacalis*. Recently, we started extensive researches on the female-biased sex ratio in *O. furnacalis* from various aspects. Here, we report the mode of inheritance, mechanism and causal agent of the female-biased sex ratio in *O. furnacalis*. A part of this study has been recently reported in Kageyama et al. (1998).

Materials and Methods

Insects

The Asian corn borer, *Ostrinia furnacalis*, is a major pest of *Zea mays* in the eastern and southeastern Asia. In Japan, six species of the *O. furnacalis* complex (*O. furnacalis*, *O. orientalis*, *O. scapularis*, *O. zealis*, *O. zaguliaevi* and *O. sp.*) occur (Mutuura & Monroe, 1970, Y. Yoshiyasu, personal communication). *O. furnacalis* and *O. scapularis* are common species in Japan.

We collected thirteen female adults of *Ostrinia furnacalis* from Matsudo (Chiba pref.), Japan in 1996. The collected females were allowed to lay eggs individually, and the offspring derived from each female was separately reared on an artificial diet (Silkmate 2S, Nihon Nosan, Yokohama). Species identification was made by morphological characters of male offspring. As to all-female families, female sex pheromone was used for species identification (Ishikawa et al., 1999).

Each cross was conducted in a plastic cup, in which three males and three females were accommodated. After the cross, females were transferred into new cups individually for oviposition.

Antibiotic treatment

Tetracycline was administered to the insects in order to examine whether the causal agent of female-biased sex ratio is a bacterium. Tetracycline hydrochloride was mixed into the artificial diet at a concentration of 0.6 mg/g wet weight and fed to the larvae from the neonate stage. If the female-biased sex ratio was affected with tetracycline treatment, the causal agent is likely to be a bacterium.

DNA extraction and PCR assay

The presence of *Wolbachia* was checked using a diagnostic PCR as described in O'Neill et al. (1992). One of the pair of ovaries in a female adult was ground in 100 µl of STE buffer O'Neill et al., 1992) with 2 µl of proteinase K (20 mg/g), and 10 µl of 10 % SDS. Then the homogenate was incubated for at least 30 min at 37°C. After incubation, the homogenate was successively extracted with phenol-chloroform and chloroform, and the extract was precipitated with ethanol. The DNA pellet was dissolved in 50 µl TE buffer and stored at 4°C. PCR was done in 10 µl reaction volumes (1 µl of DNA sample, 2.5 mM of MgCl₂, 200 µM each of dNTPs, 0.4 µM each of primers, 5 units/ µl of Taq polymerase (Sawady, Tokyo)). A temperature program of 95°C for 1 min, 52°C for 1 min, and 72°C for 1 min was repeated for 30 cycles using PCR thermal cycler MP (Takara). *Wolbachia*-specific primers designed to amplify 16S rDNA were used O'Neill et al., 1992).

Results and Discussion

Thirteen broods were obtained from the females collected at Matsudo. Three of them (M9, M11 and M13) showed female-biased sex ratios (Table 1).

Table 1 Proportion of females in each family produced by *Ostrinia furnacalis* females collected at Matsudo, Japan in June 1996

Family	Proportion male	n
M1	0.49	120
M2	0.49	72
M3	0.48	82
M4	0.42	107
M5	0.37	97
M6	0.47	47
M7	0.42	64
M8	0.47	36
M9†	0.00*	89
M10	0.44	77
M11†	0.00*	29
M12‡	0.44	92
M13	0.21*	28

n, number of pupae.

*Significantly deviated from 0.5 by chi-squared test ($P < 0.01$).

†Infected with *Wolbachia*.

‡Not infected with *Wolbachia*.

The broods from females other than these three thelygenic females were pooled and maintained as the normal strain. Females from the two female-biased families (M9 and M11) were crossed with normal (1:1) sex ratio families (M4, M7, M13). The female-biased sex ratios were maternally inherited to the F1 offspring (Table 2).

Table 2 Proportion of males in the tetracycline untreated and treated families of *Ostrinia furnacalis* for F1 and F2 generations

	Cross	Proportion male	
		F1 generation	F2 generation
Untreated			
	9.7	0.13 (211)	0.00(81)
	9.4	0.24 (29)	0.00(24)
	11.13	0.00 (56)	0.18 (45)
Tetracycline			
Treated	9.7	0.15 (109)	ND
	9.4	0.00 (13)	ND
	11.13	0.00 (111)	1.00 (223)

Family sizes are shown in the paranthesis. ND: Not determined

F1 females were crossed with males from normal strain to produce F2 offspring. Female-biased sex ratios were maternally inherited to the F2 offspring. Therefore, the causal agent of this trait is either cytoplasmic or W-linked factor (in general, sex chromosomes of Lepidoptera are thought to be female heterogametic, i. e. ZW in females, ZZ in males. It is reasonable, at present, to assume that the Asian corn borer also has the ZW-ZZ system of sex chromosomes). Tetracycline treatment over the two generations resulted in production of only male progeny in the M11 strain (Table 2). Since the effect of tetracycline is obvious in M11, the causal agent seems to be a bacterium (-a).

The result that only males were produced by tetracycline treatment is well explained by feminization of the genetic males. If the sex ratio distorter alters a chromosomal male (ZZ) into a functional female, elimination of the sex ratio distorter with an antibiotic will lead a thelygenic females to produce only male progeny. None of the other mechanisms such as male killing, parthenogenesis, meiotic drive or maternal choice of X rather than Y sperm cannot explain the present results (Kageyama et al., 1998).

The result of the PCR indicated that the two thelygenic females (M9 and M11) were infected with *Wolbachia*, and that one female that showed 1:1 sex ratio was not infected with *Wolbachia* (Table 1). *Wolbachia* infection seems to be associated with the feminization of genetic males. However, since only three strains were analyzed, it is premature to conclude that the causal agent of feminization is *Wolbachia*. If the causal agent is *Wolbachia*, the present study is the first discovery of feminizing *Wolbachia* in insects.

In conclusion, the female-biased sex ratio distortion in *O. furnacalis* is observed at low frequencies in the field, and an underlying mechanism for this phenomenon is feminization of genetic males. The causal agent of feminization is bacterium, which seems to be *Wolbachia*. The present study is the first discovery of feminization of genetic males in insects.

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Investigations on the Attractiveness of Some Traps Against the European Corn Borer (*Ostrinia nubilalis* Hbn.-Lepidoptera : Pyralidae) in Aegean Region

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Abstract

This studies were carried out in Izmir/Menemen, Torbalı/and Manisa (Salihli) provinces between 1991-1996 investigations on relations among the biology of pest with phenology of plant and observe adult flying progress by using food, light feromone traps, PAA trap substance and there were carried studies to improve the agricultural warning and forecasting program against European Corn Bore (*Ostrinia nubilalis* Hbn.). As a result of this studies usage of food and feromone traps were not provided satisfactory effect however it has been concluded that the light traps as Pennsylvania against European Corn Borer was appropriate. Also delta trap using by PAA trap substanca at dosage of 1.5 ml/trap were found suitable alternative for control of ECB.

Introduction

The most important agricultural production in Turkey is cereals. Corn, with 525.000 ha growing area and 2.225.000 ton production amount is the third crop after wheat and barley. (Anonymous 1994).

European corn borer (*Ostrinia nubilalis*) is the other important pest among some important pests of the product which has large growing area. It has become more important pest. Because, It has more than one generation in Turkey.

The treatment applied in late period of corn plant is very difficult. Therefore the chemical application time is short. All of these reasons, it's a need to determine accurately of the first adult emergence and egg laying of the pest. For this aim the effectiveness of different trap type had been searched for monitoring of first adult emergence of the pest and its generation between 1991 and 1998.

Materials and Methods

The main materials of the study were 2 different bait traps with wine and grape-molasses 8 different pheromones (35:65 and 97:3 combination of 2-11-14-AC/E-11-14-AC, 35:65 and 97:3 combination of 2-12-14-AC/E-12-14-AC, a pheromone from INRA, Trans, Cis, Hybrid formulation produced by Agrisence BCS company), 2 different light trap (Pennsylvania and Robinson type) and phenylacetaldehyde (PAA) and delta type trap used for pheromones. During study, different number of trap were used in trial every year. The trials were designed as randomized block. The traps were hanged with 50 m intervals and changeable heights as parallel product growing. Pheromone capsules and PAA were changed every two weeks while bait traps were changed each week and when sticky surfaces were dirty.

The mixture with vine in bait trap prepared with: 2/3 vine + 1/3 water +20-30g sugar + 2 table spoon vinegar for 1 liter. The mixture of grape-molasses prepared with 1 unite grape-molasses + 5 unite water + 0.1 % carnation oil. These mixtures were hanged in barrel shaped-plastic vessel which has 2.5 liter volume with 18 cm depth and 8 cm diameter. PAA

was tested using only PAA and using the dosage of 1.5 ml/trap with Cis, Trans, Hybrid capsules. PAA was tested as PAA renewing one time a week and one time in two week.

Results and Discussion

The study with bait traps was carried out in two different location (Menemen, Tire) in 1991. The traps have been hanged in trial area between 8.4.1991 and 25.9.1991. After these years the traps weren't tested for they hadn't shown any attractiveness.

The study with light traps had been carried out between 1991 and 1994. The adult numbers caught in traps in those years was given Table 1.

Table 1. The adult numbers of *O. nubilalis* Hbn. caught in light traps during the season between 1991 and 1994

Years	Trap Type	
	Pensylvania	Robinson
1991	61	11
1992	173	42
1993	204	97
1994	124	

According to four year results, Pensylvania trap was shown more suitable as a regard of attractiveness for European corn borer. During the study, it was determined that the pest had given three peak every year (Table 2,3,4,5).

First peak in second week of June, second peak in third week of July and third peak in the last week of August had been generally shown and it was determined that the pest had given three generations. Laying in nature and infections in plants was also parallel with these results. The pheromones used during the study was given.

35:65 and 97:3 combinations of 2-11-14-AC/E-11-14-AC pheromone, 35:65 and 97:3 combinations of 2-12-14-AC/E-12-14-AC pheromone and the pheromone from INRA were tested in 1991. In these 5 different pheromones, totally 21 adult were caught in first and second product of corn plant during all seasons in nature. These amounts weren't enough so these traps weren't tested the other years.

In 1993, Cis, Trans, Hybrid traps produced by Agrisence BCS company was taken for trial as a different character according to MAINI and BURGIO (1990). Cis, Trans, Hybrid and their characters with PAA were tested in trial. As a result of trials, four adults came to Cis character during all season but Trans and Hybrid weren't shown any attractiveness. Of the character of the same traps added PAA, Cis+PAA, trans+PAA and hybrid+PAA caught 22, 25 and 36 adult, respectively. (Of the same added traps with PAA, 22 adult in Cis + PAA, 25 adult in Trans + PAA and 36 adult in Hybrid + PAA was caught).

Hybrid + PAA character which had the most attractiveness in 1993 were taken for test in 1994. Corn cultivation were reduced in 1994 because of the drought, so the study was only carried out on second product and the traps had been in nature between 7.7.1994 and 21.9.1994. During this time, only hybrid caught 1 adult Hybrid+PAA renewed each week and caught 14 adults, Hybrid+PAA renewed every two weeks caught 11 adults, only PAA renewed each week and caught 11 adults and PAA renewed every two weeks caught 21 adults.

As a result, it was seen that none of the pheromones used against European corn borer were effective. However, Palfy (1983) has mentioned that the data obtained from pheromone traps which was special for European corn borer were not compared with that

data obtains from light traps in most of studies in the world. The researcher has also mentioned that the data obtained from light traps was the better.

Table 2. The adult numbers of *O. nubilalis nubilalis* caught in two different light traps in Izmir-Menemen 1991

Date of Control	The adult numbers of <i>O.nubilalis</i>		
	160 W	25 W	Total
08.04	0	0	0
15.04	0	0	0
22.04	0	0	0
29.04	0	0	0
06.05	0	0	0
13.05	0	1	1
19.05	0	0	0
24.05	0	1	1
30.05	2	1	3
06.06	2	3	5
13.06	2	2	4
20.06	0	1	1
27.06	0	2	2
04.07	0	3	3
10.07	0	6	6
18.07	0	5	5
25.07	1	5	6
01.08	2	2	4
08.08	0	4	4
15.08	0	4	4
22.08	0	4	4
30.08	2	9	11
06.09	0	3	3
12.09	0	5	5
18.09	0	0	0
25.09	0	0	0
Total	11	61	72

Table 3. The adult numbers of *O. nubilalis* caught in two different light traps in Izmir Menemen 1992

Date of Control	The adult numbers of <i>O.nubilalis</i>		
	18 W	160W	Total
14.04	0	0	0
29.04	0	0	0
07.05	0	0	0
13.05	0	0	0
21.05	1	0	1
27.05	1	0	1
04.06	5	2	7
10.06	6	0	6
15.06	8	1	9
19.06	4	1	5
22.06	0	0	0
25.06	1	0	1
29.06	0	0	0
02.07	0	0	0
06.07	2	0	2
09.07	0	0	0
13.07	2	0	2
16.07	0	0	0
20.07	2	0	2
23.07	0	2	2
27.07	1	0	1
30.07	0	0	0
03.08	1	0	1
06.08	0	0	0
10.08	2	1	3
12.08	1	0	1
17.08	2	1	3
20.08	14	1	15
24.08	20	9	29
27.08	26	6	32
31.08	44	11	55
03.09	19	3	22
07.09	10	3	13
10.09	0	1	1
14.09	1	0	1
17.09	0	0	0
Total	173	42	215

These results give an opinion that 1.5 ml/trap dosage of PAA in trial renewed one time in two week could be used alternatively to light traps for determining emergence time of European corn borer and its control time. Maini and Burgio (1990) has explained that why the pheromone used against European corn borer were not shown the same effectiveness everywhere was phenomenal polymorphism of the pest. So the researches have announced that obtaining of the purified specific pheromones was difficult and they have attached importance to using PAA.

Table 4. The adult numbers of *O. nubilalis* caught by pensylvania type light trap in Manisa-Salihli in 1993

Date of Control	The adult numbers of <i>O.nubilalis</i>		
	18 W	160 W	Total
21.05	4	0	4
27.05	14	10	24
03.06	51	48	99
10.06	15	15	30
18.06	12	5	17
24.06	0	0	0
01.07	0	0	0
08.07	0	0	0
29.07	14	7	21
05.08	17	2	19
29.07	14	7	21
05.08	17	2	19
12.08	3	0	3
19.08	4	1	5
26.09	12	Broken	-
01.09	12	Broken	-
10.09	14	Broken	-
15.09	1	Broken	-
22.09	0	Broken	-
29.09	0	Broken	-
Total	204	97	

Table 5. The adult numbers of *O.nubilalis* caught by two different light traps in Manisa-Salihli in 1994

Date of Control	The adult numbers of <i>O.nubilalis</i> (The adult number/trap/week)
28.04	2
05.05	1
12.05	7
19.05	17
26.05	Broken trap
02.06	
09.06	8
16.06	1
23.06	0
30.06	0
07.07	0
14.07	1
21.07	5
28.07	13
03.08	18
11.08	14
18.08	5
25.08	2
01.09	Broken trap
08.09	7
15.09	14
21.09	7
29.09	2
06.10	0
Total	124

As a result of the study carried out a long time, it was shown that the main problem of the pest was in second product. The reason of the high damage (sometimes 96%) in second product is there is a good harmony between pest biology and plant phenology. European

corn borer has not preferred egg laying on the plants smaller than 40 cm (Derin 1992). As the reason of this phenomenon was known that the DIMBOA (2.4-dihydroxy-7 mehoxy-2H-1.4 benzoxazin-3-(4H)-one) concentration determined in plant was high (Robinson et al 1983).

As a result, there is not any economical damage in first product of corn planted normal period of the pest, so chemical control is not need.

The pest in late planted first product may sometimes be a problem which need to control. Infections of the second and third generations in second product have caused problems. The situation of *Trichogramma evanescens* West caused 76% parasited in pest eggs in our region must be taken into account well before deciding chemical control (Derin 1992).

It was determined that the most accurate data were obtained by Pennsylvania type light trap in monitoring of adult emergence and its generations and the best way was PAA renewed every two weeks in case the absence light source.

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248 Successive Generations of *Ostrinia nubilalis* Obtained by Mass Rearing on Artificial Diet

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Abstract

Maize resistance to the attack by *Ostrinia nubilalis* is a major target of plant breeders, this imposing approach of investigations on rearing this insect on artificial diet. This report contains a series of data regarding steady rearing of the maize borer on artificial diet for several successive generations.

During 1978-1998 a different number of colonies resulted from the natural population has been set, each in a certain experimental year, with the aim to simultaneous study of different generations reared on artificial diet. The first colony was set up in 1978 and in 1997 the last one. Periodically tests have been performed with these colonies, to estimate the nutritive value by the number of pupae obtained from a rearing dish, duration of larval and pupal instars, and the number of egg-masses laid by one female.

In order to study the behaviour of various *O. nubilalis* generations reared at the same time span, in separate trials fecundity and life-span of adults have been studied. These parameters have been separately recorded for each colony and generation.

It was found that ECB reared on artificial diet reached up to the end of 1998, in agreement with the experimental colony, in the 248-th generation of the colony started in 1979, the 36-th generation of the colony set in 1996, the 36-th generation of the colony set in 1996, and the 22-nd generation of the colony founded in 1997.

The number of pupae per rearing dish, development duration (days) of larvae and pupae, and the number of egg-masses/female were not influenced by the number of generation this belonged to, being reared under continuous flow.

The significant experimental conditions changes have influenced mass-rearing of this insect on artificial diet.

Key words: *Ostrinia nubilalis*, European Corn Borer, mass rearing, artificial diet, infestation, egg-masses.

Introduction

In Romania the European Corn Borer (ECB) continues to be an important pest of maize in areas favourable to this, where yield losses due to its attack can sometimes reach up to 60% (Paulian et al., 1962).

As the use of maize hybrids resistant to this pest is of economic interest, at the Research Institute for Cereals and Industrial Crops-Fundulea a special breeding programme is running since 1975. Tests of biological material are performed under artificial infestation with egg-masses, in view to provide heavy and uniform attacks, able to allow differentiation of reaction of maize forms included in the programme of development of resistant hybrids. As Guthrie et al. (1970) stated, progress in resistance research would be worthless unless artificial plant infestation.

In order to achieve artificial infestation of maize plants with *O. nubilalis* egg-masses, mass rearing of insect on artificial diet is necessary. Following huge investigations performed mainly in the United States of America and elsewhere, this insect can be reared in a continuous flow on artificial diet. In this respect, Rathore and Guthrie (1973) reported laboratory rearing of the European Corn Borer on artificial diet for 87 successive

generations, while Brindley et al.(1975) for 108 successive generations. In our country, Bărbulescu (1982, 1986, 1993, 1996 and 1998) showed that *O. nubilalis* has been reared under continuous flow on artificial diet, and reported the number of generations obtained, as depending on the colony under experiment. Thus, in his latest report (1998) the author mentioned that up to the end of 1995 the insect was reared as follows: 210 generations of the colony set from a natural population in 1979, and 43 generations from the 1992 colony.

The fact that rearing the borer on artificial diet, separately by colonies, starting with the oldest, set in 1978, a bulk of data resulted, which are exposed in the present paper.

Material and Method

The investigations have been carried out at the Research Institute for Cereals and Industrial Crops-Fundulea during 1978-1998, in rearing rooms at temperatures of 24-28°C, air relative humidity 60-90%, continuous ventilation and light, except for room of egg-laying, which was under permanent dark.

The insect was reared on a diet with bean meal as basic ingredient, mixed with wheat bran and a range of other ingredients, named diet A (Bărbulescu, 1982), using the mass-rearing technology previously described (Bărbulescu, 1980).

In view to assess influence in time of nutritive value of diet on the behaviour of insects reared under steady flow, in a different number of successive generations, several colonies resulted from the natural population, have been trialled, one colony from each of the following years: 1978, 1979, 1980, 1985, 1986, 1987, 1990, 1992, 1996 and 1997. These colonies have been tested for various time periods, always including, in the same time span, several colonies with a different number of generations. Periodically, in these colonies the nutritive value of diet was checked, according to: average number of pupae obtained in a rearing dish, average length of larval and pupal instars, average number of egg-masses laid by one female. In separate trials female fecundity and adult life-span have been evaluated. These parameters have been separately recorded, by generation and colony.

Results and Discussion

The results are presented in tables 1-4.

Analysis of data in table 1 reveals that the 10 colonies derived from *O. nubilalis* natural population evaluated according to the experimental period, in a various number of generations. The highest number of generations- 248 - has been recorded in the colony founded in 1979, while the lowest - 22 generations - in the youngest colony, started in 1997.

Data on the number of pupae/rearing dish (tables 1-2) fluctuated from a colony to another, but also within each colony, between very wide limits. When comparing these data referring to colony, it results that there is no link between the longer or shorter period of rearing in continuous flow of these colonies, i.e. between the different number of successive generations and production of pupae/rearing dish. It is to note that high variation, in all colonies, between the values recorded for various generations is mainly explained by different experimental conditions, particularly with fungal infections, inducing moulds. From table 3 it is noted that for the same experimental period, i.e. the same experimental conditions, the number of pupae/rearing dish was very low in all cases for the early generations, as compared to the following ones, thus confirming literature data referring to insect rearings on artificial diets, when the natural populations were brought and reared in laboratory (Boller, 1972).

Referring to development duration of larvae and pupae, it can be seen that the average value for every colony slightly fluctuated from a generation to another, this showing a very low variation, without existence of differences between the respective colonies. Small fluctuations revealed in all colonies, under certain conditions from a generation to another were mainly due to temperature conditions, which were not constant throughout the trial period. Thus, duration of insect development was not influenced by the number of generations reared on artificial diet.

Taking into account data on the egg-masses laid by one female it is to note that the average value of all generations and that calculated for groups of 10 generations, separately by colonies, varied within not too wide limits. Large fluctuations recorded in some instances are usually explained by the various experimental conditions. The fact that the average value of 248 generations of the oldest colony, from 1979, was 3.0 egg-masses/female, while in the youngest the average value of 22 generations was 2.4 egg-masses/female, clearly proves that insect fecundity was not influenced by continuous flow rearing for several successive generations on artificial diet. The same results also detached from analysis of data in table 4.

When comparing data on the number of pupae/rearing dish, duration of development (days) of larval and pupal instars, as well as the number of egg-masses/female in all colonies under experiment, which represent a different number of generations for the same experimental period (table 3), an evolution is obviously noticed, generally similar of insect, irrespective of the number of generations the insect belonged to.

Conclusions

- The European Corn Borer has been reared on artificial diet under continuous flow for a variable number of generations, as depending on the colony generated from the natural population. Until the end of 1998, in the oldest colony, established in 1979, 248 generations have been obtained, and 22 generations from the youngest colony, founded in 1997.

- The number of pupae/rearing dish, development duration (days) of larval and pupal instars, and the number of egg-masses/female were not influenced by the number of generation the insect reared on artificial diet belonged to. Accordingly, the diet used for insect rearing had suitable nutritive value.

- Variations recorded in all colonies, between the values of analysed parameters have been substantially caused by the experimental conditions.

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Table 1. Representative values on the *O. mubilalis* rearing during successive generations on the artificial diet, by colonies.

Colony	Experimental period	Generations obtained	No. of pupae/dish			Duration of larval and pupal development (days)			No. of egg-masses/female		
			Min.	Max.	average	Min.	Max.	average	Min.	Max.	average
1978	1978-1984	80	49	542	252	19	30	23,3	1,5	10,8	4,1
1979	1979-1998	248	27	658	325	19	32	22,0	0,1	8,5	3,0
1980	1980-1992	160	31	589	240	19	30	22,2	0,4	9,9	2,9
1985	1985-1993	113	9	522	230	20	27	21,2	0,4	9,0	2,1
1986	1986-1992	88	7	454	221	19	28	21,5	0,5	6,4	1,9
1987	1987-1992	75	15	525	183	20	27	21,6	0,4	3,6	1,6
1990	1990-1993	48	63	552	258	19	23	20,8	0,5	6,3	2,2
1992	1992-1996	62	51	750	451	19	23	20,1	0,9	7,1	3,1
1996	1996-1998	34	212	533	390	19	23	21,0	1,4	5,4	2,7
1997	1997-1998	22	113	524	346	19	24	21,5	1,7	3,6	2,4

Table 2.-Average data presented by groups of 10 successive generations reared on the artificial diet, as depending on colony of *O. nubilalis*

Generation	Colony										
	1987	1978		1979		1980		1985		1986	
3	1	2	3	1	2	3	1	2	3	1	2
10	210	24	224	21	3,7	239	24	3	90	1,4	24
194	23	1,9	148	22	2,8	203	3,6	22	3,7	262	2,7
11	212	24	3,7	24	4,1	308	22	3,7	161	22	248
21	1,9	207	21	1,5	325	21	3,3	413	21	2,2	2,2
21-30	213	23	244	22	4,5	263	24	4,5	254	21	1,5
160	23	1,7	212	21	1,7	328	2,3	433	20	2,2	2,5
31-40	270	24	3,4	215	24	5,3	348	22	6,0	247	22
138	21	1,4	306	21	1,9	487	2,0	380	21	2,1	2,1
41-50	241	24	4,7	386	23	5,5	333	21	4,0	223	22
160	21	1,3	259	20	2,6	559	1,7	1,7	21	1,3	142
51-60	298	22	5,1	339	22	4,4	189	24	2,0	143	22
170	21	1,1								1,4	272
61-70	333	22	5,0	262	23	2,7	168	23	1,5	233	22
137	21	1,7								1,6	246
71-80	222	23	3,9	201	23	2,1	218	22	1,9	205	21
338	21	2,7								1,6	207
81-90	145	22	1,3	191	21	1,4	201	21	1,2	239	21
91-100	317	22	1,9	221	22	1,6	262	21	2,5		
101-110	273	21	1,9	196	22	1,5	285	20	2,7		

111-120	220	22	1,8	148	22	1,3
121-130	198	22	1,4	159	22	1,6
131-140	157	22	1,6	254	21	1,7
141-150	226	21	1,3	198	21	1,4
151-160	171	21	1,1	176	21	2,6
161-170	229	21	2,2			
171-180	324	20	2,6			
181-190	330	20	2,3			
191-200	396	20	1,8			
201-210	539	20	1,8			
211-220	493	21	3,1			
221-230	464	20	2,7			
231-240	411	20	2,2			
241-248	374	21	2,0			
average number of pupae/dish 2 = average duration of larval and pupal development (days) 3 = average number of egg-masses/female						

Table 3. Behaviour of different generations of *O. nubilalis* reared in the same periods of time

Date of infestation	Colony	Generation	No. of pupae/dish	Development duration: larval and pupal (days)	No. of egg-masses/female
26.04.1985	1979	73	288	24	2.1
	1980	61	219	24	2.2
	1985	1	9	23	2.4
18.03.1987	1979	97	366	23	2.1
	1980	85	182	21	1.9
	1985	24	405	21	1.4
	1986	13	306	21	2.3
	1987	1	55	21	4.8
20.03.1990	1979	136	226	20	1.6
	1980	124	193	21	1.6
	1985	63	296	21	1.5
	1986	52	324	20	3.4
	1987	39	193	20	1.2
	1990	1	63	20	5.8
11.05.1992	1979	164	278	20	2.7
	1985	91	223	21	3.9
	1990	29	223	21	4.0
	1992	3	51	21	3.6
3.08.1993	1979	180	286	19	2.4
	1985	107	288	20	2.7
	1990	45	309	21	2.3
	1992	18	312	19	3.2
17.09.1994	1979	197	521	20	2.5
	1992	35	497	20	2.4
15.03.1996	1979	214	474	21	2.9
	1992	52	472	19	3.0
	1996	1	212	20	3.6
29.08.1996	1979	220	525	20	3.6
	1992	58	475	20	2.8
	1996	7	440	19	2.2
21.03.1997	1979	227	504	20	4.0
	1992	65	510	21	4.6
	1996	14	430	20	3.5
	1997	2	115	20	3.0
7.08.1997	1979	232	343	19	3.0
	1996	19	362	20	2.7
	1997	7	380	19	2.9
21.10.1998	1979	247	406	20	2.4
	1996	34	415	21	2.3
	1997	22	441	20	2.6
23.11.1998	1979	248	403	20	1.4
	1997	23	409	20	1.8

Table 4. Fecundity and life-span of *O. nubilalis* adults reared in a different number of successive generations on artificial diet.

Experimental period	Colony	Generation	No. of egg-masses/female	Adult life-span (days)	
				female	male
1984-August	1979	65	7.5	9.3	10.2
	1984	4	7.3	8.3	9.0
1986-November	1979	91	4.3	9.8	8.8
	1980	78	4.4	7.5	9.0
	1985	19	3.1	7.2	7.5
	1986	8	4.7	7.3	9.7
1987-December	1979	106	5.2	10.2	12.7
	1980	93	4.3	10.2	12.2
	1985	33	4.5	10.6	13.7
	1986	22	4.9	12.9	14.4
	1987	9	4.1	12.5	14.7
1991-May	1979	150	5.3	7.9	8.1
	1985	77	3.7	7.3	9.8
	1990	15	3.5	7.6	8.8
1993-February	1979	173	6.8	10.6	13.3
	1985	100	6.7	10.2	14.7
	1990	38	7.8	12.1	14.8
	1992	11	9.8	12.2	15.3
1996-April	1979	215	8.1	9.7	10.5
	1992	53	9.4	10.9	12.3
	1996	2	12.7	11.7	11.6
1996-November	1979	222	9.3	13.4	14.9
	1992	60	11.3	14.1	17.0
	1996	9	8.1	13.0	15.8
1997-April	1979	227	12.4	12.3	13.8
	1992	65	11.8	13.0	15.0
	1996	14	13.2	12.7	14.4
	1997	2	11.9	9.8	9.9
1998-November	1979	247	5.4	12.2	11.9
	1996	35	5.0	10.7	11.8
	1997	21	4.8	10.5	10.6

The Possibilities of Reducing the Pollution of Environment Utilizing Some Biological Methods in Corn Borer(*Ostrinia nubilalis* Hbn.) Control in Transylvania

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Abstract

In the regions of Transylvania favourable to maize crop, about 8 -13 % of yield is lost due to the damages caused by the European Corn borer (*Ostrinia nubilalis* Hbn.). At the some hybrids the yield loss may reach 21-22%.

These damages produced by *O. nubilalis* on maize crop determined the application one of biological methods of central no polluting the environment and agrofeeding products contributing in equilibrium maintaining in agroecosystem.

Between 1996-1998 at Agricultural Research Station Turda was used as biological methods of this pest control some of parasites species of *Trichogramma* (*T.maidis*, *T. evanescens*, *T. dendrolimi*), biological products (Thuringin 6000, Dipel 2x) and chitine inhibitors (Match 050 EC, Insegar 25 WP) that produce interruption of larvae transformation in pupa and from eggs to larvae.

T. maidis species was proved to be the most adapted at maize crop, followed by *T. evanescens* which reduced the attack frequency of borer very significantly and yield increase about 19%.

The biological products used reduced significantly the attack of this borer and yield increase was about 10%.

Key words: *Ostrinia nubilalis*, *Trichogramma* spp., biological products, attack frequency, yield.

Introduction

It's well known that chemical products represented in the moment of appearance one of the hope of mankind, owing to their quickly effect and often radically in pests control. Beside these positive effects the intensive and long utilisation of these had also the negative effects (the destroying of natural biological equilibrium, the appearance of resistance phenomenons of the pests, of the polluting residues, of the toxicity for the animals and for the people). All of these required the researches reorientation to a new concept of integrated control where the biological methods occupy an important place. In this way, the paper approaches one of reducing ways of environment pollution through utilising of entomophagous *Trichogramma*, of biological products based on *Bacillus thuringiensis* bacteria, and of the less polluting products, from the latest creations, inhibitors of insects increasing with effect on the eggs and larvae in controlling one of the most important pests of maize crop: *O. nubilalis* Hbn.

In Transylvania, in one of favourable maize crops, the corn borer attack recorded the frequencies between 35-95%, and the yield was reduced with 8,0-13,0% (Mustea and colab.,1995). The performed researches sit beside the performed ones in this field by Dulmage (1981); Beegle and colab., 1982; Galani and colab., 1990; Ciochia, 1991; Fabritius and colab., 1992; Amos Navon, 1993; Roman and colab., 1994; Serpil Kornoșov,1995; Uzun and colab., 1995; Florentina Marcu, 1996; Felicia Mureșan and colab., 1996, 1997, etc.

The purpose of this paper is to establish the influence of biological treatments performed between 1996-1998 at Agricultural Research Station Turda on the reduction of corn borer (*O. nubilalis* Hbn.) attack, utilising less polluting products.

Material and Method

The researches were performed in an experience placed in randomised blocks including 8 variants in 3 repetitions. The size of each plot was 7 sqm and maize hybrid Turda 200. The treatments were performed with 3 species of *Trichogramma*: *T.maidis*, *T.evanescens* *T.dendrolimi* in 200.000 individuals/ha, fractionate administrated, depending on eggs depositing of corn borer. The biological products utilized were: Thuringin 6000 - 4,0 kg/ha, and Dipel 2 x - 1,5 kg/ha and among chitinne inhibitors: Insegar 25 WP - 0,3 kg/ha, Match 050 EC - 1,2 l/ha (table 1). The all treatments were performed on the natural and artificial infestation with eggs and larvae at I age. For *Trichogramma* treatments and Insegar 25 WP product the infestations were performed with eggs, and for the treatments with Thuringin 6000, Dipel 2 x, Match 050 EC with larvae at I age, depending on their effect on the eggs or on the larvae. The first artificial infestation and treatments were done at the first borer adults appearance, recorded in the pheromones traps, that corresponds with plants phenofasis about 8-10 leaves, following then another two infestations and treatments at 8 days interval.

At the harvest, were done some observations and notes about the frequency of attacked plants by corn borer, the number of holes and larvae, the length of the tunnels from the stems and the obtained yield.

The results data were calculated utilising the χ^2 (square-chi), test the variance analysis and the square regressions.

Results and Discussions

The evolution of the climatic conditions in the three years of experimentation corresponding to the period of maize vegetation (April-October) influenced in a positive way the treatments applying with *Trichogramma* and biological products. June and July when the treatments were done the warm and the rainfalls were normal (figure 1)*.

For the statistical calculation performed, resulted that the biological treatments considerably reduced the corn borer attack (figure 2). Among *Trichogramma* species the most active was *T. maidis* which in the variants with two or three releases reduced very significantly the attack (with 49,2% and 67,4%). Then, followed *T. evanescens* species that reduced the attack with 43,4% but only in the variant with three releases. At *T. dendrolimi* the attack reducing was only with 15,4% after three releases, that confirms the obtained results by other researchers (Marcu F., 1996) that this species is more active only in pest control in fruit growing. Owing to the way of action of the Thuringin 6000 and Dipel 2x products determined by a specific toxin that actions through an ingestion producing the illness of a digestive apparatus at the larvae at I age, the corn borer attack was reduced significantly (with 11,7%) and very significantly (with 15,3%) comparatively with untreated variant. In corn borer case, Insegar 25 WP product has an effect on the eggs producing a development interruption from egg to larva, the attack reducing very significantly (with 17,0%). The Match 050 EC products, being a chitine inhibitor, the larvae at I age can't pass in the next larval phase, the attack reducing very significantly (with 28,3%) comparatively with untreated variant

The analysis variance (table 2)* shows that the performed treatments influenced very strong the holes number too, the value of F test being very significant (88,70***). An

influence very significant had the climatic conditions in the three years, and the interaction between the factors contributes to reduce very significant this element.

The number of corn borer holes varied between 0,9 - 1,5 holes/plant (figure 3). The most favourable year regarding the reducing of this element was 1996, then 1997 and 1998. Comparatively with untreated variant, the treatments with *T. maidis* and *T. evanescens* (three releases) reduced very significantly and significantly this element. On the other hand at the treatments with *T. dendrolimi* the reducing is no assured statistically, these treatments being more efficient for other pests. From the used products the most efficient was Dipel 2x with a very significant reducing, then Insegar 25 WP and Match 050 EC, which had an effect almost the same, being significant.

On the number of corn borer larvae influenced very strong the performed treatments, the climatic conditions, and the interaction between these factors (table 3), the value of F test being very significant.

In this case too, the year 1996 was the most favourable regarding the reduced number of larvae, followed then by 1997, comparatively with years average (figure 4). After the treatments with the two *Trichogramma* species, the larvae number of corn borer was very significantly reduced comparatively with untreated variant, and among the used products, the most efficient was Match 050 EC, Insegar 25 WP and Dipel 2x with a significant and very significant reducing.

The length of borer tunnels was influenced very significantly by the performed treatments (table 4) and the climatic conditions.

Comparatively with years average, the year 1996 was the most favourable regarding the reducing of tunnels length in the maize plants, (figure 5) followed then by the other two years. This element was also reduced very significantly after treatments with *T. maidis* and *T. evanescens* (from 15,1 cm to 5,7 and 6,1 cm). The treatments with Dipel 2x and Match 050 EC with effect on larvae at 1 age, reduced significantly the tunnels length (from 15,1 cm to 10,2 and 9,8 cm).

Calculating the variance (table 5) it shows the contribution of studied factors (climatic conditions, biological treatments) for realization of beans yield, analysing F test corresponding to factors variations. The biological treatments had an important influence on obtained yield, the F test being very significant. The climatic conditions are also included in yield results. Because the F test is very significant demonstrates that in experimental years existed some differences regarding their favourabilities on the yield, and the interaction between years and treatments had a very significant influence.

Comparatively with years average, the greatest yield was realised in 1998 when the increase was very significant (figure 6), and in 1997 the difference of yield was significant negative. 1996 was close to years average. The effect of biological treatments is reflected in the increases of obtained yield. Thus, after the treatments with *T. maidis* and *T. evanescens* the yield increases recorded, were about 12,9% and 10,3% very significant, with Dipel 2x and Insegar 25 WP about 2.8% and 3.4% significant and at Match 050 EC the increase was very significant 5,8%.

From years interaction and treatments (figure 7) even the climatic conditions influenced the obtained yield, the greatest increases were realised in the all years after the treatments with *T. maidis* (12,2-18,4%), *T. evanescens* (10,2-13,1%) with Match 050 EC (9,9-10,1%), then Insegar 25 WP (5,2-7,0%) and Dipel 2x (4,7 - 7,6%).

Graphics picture of the effect of reducing the corn borer populations applying the biological treatments, presents the image of square regressions calculated between of holes number, of larvae, of tunnels length/plant and obtained yield/plant. The performed

treatments assured a reducing of holes number on the maize plants (figure 8) recording also a saved yield/plant significantly comparatively with untreated variant, depending on the *Trichogramma* species or used product. Thus, the treatments with *T. maidis*, *T. evanescens*, Match 050 EC assured a reducing very significantly of the holes number/plant (from 2,7 to 0,6; 0,8; 1,0), recording a saved yield/plant significantly comparatively with untreated variant (from 135 g/plant to 160 g/plant). The relation between the obtained yield/plant and the reduction of holes number is very tight, the value of correlation rapport η is very significant (0,919***).

The mentioned treatments assured the highest level of yield/plant reducing also very significantly the larvae number/plant and also the tunnels length in the maize plants, comparatively with untreated variant (from 1,3 to 0,2; 0,4; 0,6 larvae/plant, and from 15,0 to 5,0; 6,0; 7,0 cm the tunnels length/plant)(figure 9). In this case too the value of correlation rapport η is significant that proves a tight relation between the obtained yield/plant and reducing of larvae number and of tunnels length/plant.

Conclusions

1. In the years 1996-1998 the conditions, the biological treatments with *Trichogramma* spp. reduced the frequency of the attack plants by *O. nubilalis* Hbn. from significantly to very significantly (from 13,4 - 67,4%) depending on parasites species and on the number of releases.

2. From the used products, Thuringin 6000, Dipel 2x, Match 050 EC with effect on larvae, reduced the attack of corn borer with 11,7%, 15,3% and 28,3% and Insegar 25 WP product with effect on eggs reduced the attack with 17,0%, comparatively with untreated variant.

3. As a result of the attack reducing of *O. nubilalis* Hbn., after the application of treatments with *Trichogramma* spp. and the mentioned products, the saved yield recorded was significantly and very significantly (with 2,8 - 12,9% comparatively with untreated variant).

4. The utilisation of these biological methods in reducing the corn borer populations form maize crop, contributes at reducing the environment pollution - which is the most dangerous flagel in present, at the maintaining of the used fauna existent, protecting the crop till harvesting against different pests.

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*Tables and figures were not sent by the author.

Biological Control of *Ostrinia nubilalis* Hübner (Lepidoptera; Pyralidae) by *Trichogramma evanescens* Westwood (Hymenoptera; Trichogrammatidae) and Its Natural Parasitization Rate on Maize in Çukurova Region of Turkey

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Abstract

This study was conducted to determine the biological control possibilities of the main pest of maize, *Ostrinia nubilalis* Hübner with inundative releases of *Trichogramma evanescens* Westwood, beside this, natural parasitization rate of *T. evanescens* Westw. was studied on the maize field in Çukurova region of Turkey during the maize vegetation period in 1995.

O. nubilalis, *Ephestia kuehniella* Zeller (Lepidoptera, Pyralidae) and *T. evanescens* were reared in a climatic room under constant temperature ($25\pm 1^{\circ}\text{C}$), relative humidity ($65\pm 10\%$) and appropriate light regime for three species.

T. evanescens was applied in two releases with 10 days interval, at the beginning of the oviposition period of third generation of *O. nubilalis* in second crop maize. A total of 1.500.000 parasitoids per 100 da ($750.000 + 750.000$ parasitoids / 100da) were released. Egg parasitism was 89.19 % and the reduction of infested plant with *O. nubilalis* was 63.63 % in this experiment.

Natural parasitization rate of the egg parasitoid in Çukurova region which were assigned to intensity of pesticide use and ease of transportation were studied in three subregions. The highest natural parasitism rate found in the first subregion (Ceyhan, Kozan, Kadirli, Osmaniye) than the third (Tarsus, Mersin) and the second (Yüreğir, Karataş) subregion. Even though the parasitism rate of the first and third subregion were 45.93% and 38.26%, second subregion had a lower parasitism rate by 1.45%.

In conclusion, natural parasitism by *T. evanescens* was 38.05% in Çukurova region in 1995. Natural parasitism was high in the first and third subregion, where pesticide use was minimal; and was low in the second subregion where pesticide use was intensive.

Key words: Maize, *Ostrinia nubilalis*, *Trichogramma evanescens* , inundative releases, natural parasitization

Introduction

The European corn borer, *O. nubilalis*, continues to be one of the most destructive pest of maize in Turkey. Control of this pest often requires multiple insecticide applications. Growers often apply at least 2-3 insecticide treatments to each planting to control only *O. nubilalis*, at an estimated annual statewide cost of about 6000 \$ in 1995. This heavy use of insecticides is expensive and poses undesirable effects on the environment, wildlife and man. Besides, chemical applications sometimes has given unsatisfactory control if an application was carried out after the insect had bored into the stalk. That's why, alternative tactics for control are needed. Among natural enemies of the corn borer, *T. evanescens*, is the most widely used and efficient biocontrol agent against this pest in corn fields in

Turkey. Because they kill their hosts during the egg stage before larval feeding causes damage. This is important in controlling corn borer. Also, *Trichogramma* spp. is easy to rear and release in large numbers.

In the world, *Trichogramma* spp. attacks more than 400 pest species, mostly lepidopterous and some pests on agriculture and forestry. It is estimated that over 32x10⁶ ha have been treated with *Trichogramma* spp. annually in more than 30 countries. During the last 30 years, *Trichogramma* spp. have been used on maize, sugarcane, cotton, vegetables and fruit trees (Li Li, 1994). The utilization of *T. evanescens* in controlling the European corn borer is reported by several authors (Zilberg 1972, Krehbiel and Wittwer 1979, Chiang et al. 1980, Hassan and Heil 1980, Berger 1984, Hassan 1981a, Hassan 1981b, Voronin and Grinberg 1981, Ivanov 1982, Hassan 1983, Hassan et al. 1986).

The objective of these studies was to determine the efficiency of *T. evanescens* on egg masses of the European corn borer in field conditions in 1995. This allows an evaluation of the possibility of using *T. evanescens* to control European corn borer in a field Integrated Pest Management (IPM) program.

Material and Methods

This study was conducted as laboratory and field experiments.

Laboratory experiments

O. nubilalis, *Ephestia kuehniella* Zeller (Lepidoptera, Pyralidae) and *T. evanescens* were reared in a climatic room under constant temperature (25±1°C), relative humidity (65±10%) and appropriate light regime for three species (Kayapınar 1991). Also, sometimes *T. evanescens* was reared on the eggs of *O. nubilalis* in order to prevent the parasite from quality deterioration.

Field experiments

Inundative Release of *T. evanescens* against *O. nubilalis*

In 1995, a total of 10 ha have been treated by releasing the parasitoids in second crop maize field in the Faculty of Agriculture in Adana. The distance between the treated and untreated control plots was 500 m. The adult population of the pest was observed by means of Robinson light trap which was located near the experimental plots to determine the releasing time.

The first *Trichogramma* release was performed when the first adult of *O. nubilalis* was captured (01.08.1995) in the light trap. But, before releasing, previous counting of egg masses was done at 20 different points by controlling five plants totally in 100 plants.

T. evanescens was released two times 75.000 parasitoids/ha with 10 days interval (01.08.1995 and 11.08.1995) at the beginning of the oviposition period of the third generation of *O. nubilalis* in second crop maize (Hassan et al. 1986; Stein 1987). The distance between two releasing points was 14 m (Hassan 1981; Hassan 1984).

Different development stages of parasitoids from 50 points were released with hanging the releasing bag to the leaf of the maize plant. Each releasing bag included approximately 1500 *E. kuehniella* eggs parasitized by *T. evanescens* (Hassan and Heil 1980).

The effectiveness of the parasitoids has been studied by sampling pest eggs before and after releasing the parasitoids in treated plots till harvesting time. The number of egg masses examined two times in a week. Every two plants in four edges of the plant on which the releasing bag was hung were examined for counting the pest eggs totally in 100 plants. If the egg masses are parasitized, it is recorded. If they are not parasitized the leaf and the egg masses were marked and then observed until the eggs either turned black or

larvae of *O.nubilalis* emerged from them. The untreated plot was also monitored at the same time for possible parasitism.

The evaluation of the effect of release applications was determined by assesing percentage of parasitism throughout the growing season (Bigler 1986). Moreover, the efficiency of the parasitoids was evaluated at harvesting time by counting the number of infested plants and 1000 grain weight in treated and untreated plots. Among 100 plants per plot taken at random were cut and examined. The number of larvae, pupae and entrance holes was recorded for every sample plant. Infestation rate was calculated by means of Abbott formula.

Natural Parasitization Rate of *O. nubilalis* eggs by *T. evanescens*.

Three subregions were selected in Çukurova region to determine the natural parasitizm of *T. evanescens* in the East Mediterranean Region (Table 1).

Table 1. Three subregions according to the using insecticide application density in Çukurova.

1 st Subregion	Ceyhan, Kozan, Kadirli, Osmaniye
2 nd Subregion	Yüreğir, Karataş and their villages
3 rd Subregion	Tarsus, Mersin and their villages

These subregion were surveyed once a week during the vegetation period in 1995 and 100 plants from every field were checked according to the plant phenology and the densities of the different stages of *O. nubilalis*. European corn borer mainly lays its eggs under the leaf, rarely on the leaf. This part of plants was carefully examined. Egg masses of *O. nubilalis* were collected by cutting the leaf to bring back to the laboratory. In the laboratory, the number of egg masses as well as the number of eggs were counted in order to determine parasitism percentage and then placed into glass vials separately. Vials were kept at 25±1°C, relative humidity 70±5 RH and 16:8 L:D. The vials were controlled daily if the eggs were hatched or a parasitoid emerged until they died. Parasitization rates of egg masses and the eggs per egg masses were calculated for each surveyed area

Result and Discussion

Inundative Release of *T. evanescens* against *O. nubilalis*

The average parasitism of corn borer eggs masses by *T. evanescens* is shown in Table 2.

Table 2. The parasitization rate of *T. evanescens* on the eggs of *O. nubilalis* in treated and untreated plots in 1995.

	The Num. of Eggs	The Num. of Par. Eggs	Parasitization Rate (%)
Treated plot	731	652	89.19
Untreated plot	416	34	8.17

The first unparasitized *O. nubilalis* egg mass was found before releasing parasitoids on 1 August 1995. The same day, totally 75.000 parasitoids/ha were released. During the first week of release when very low numbers of pest eggs were present in the release field, parasitism averaged 59.52%. After 10 days (11.08.1995), the second release was applied. The high egg parasitism peak reaching 100% was observed after second releasing in mid-

August when the *O. nubilalis* egg densities increased (Lopez and Morrison 1985). This high parasitism lasted till the end of the season.

In the treated plot, totally 731 eggs were counted and 652 eggs were found to be parasitized. Average parasitism rate was 89.19% (Table 2). It is known that *Trichogramma* spp. has short life cycle and a high reproductive potential (Pak and Oatman 1982ab, Brower 1988) that combine to produce a rapid population increase. That's why, parasitism increased at the end of the season.

In untreated plot, totally 416 eggs were counted and only one egg mass including 34 eggs were found to be parasitized (8.17%) (Table 2). Because of having a high passive spread of the egg parasitoids by wind effect, a low parasitism was observed in untreated plot (Yu et al. 1984, Keller and Lewis 1985) (Figure 1).

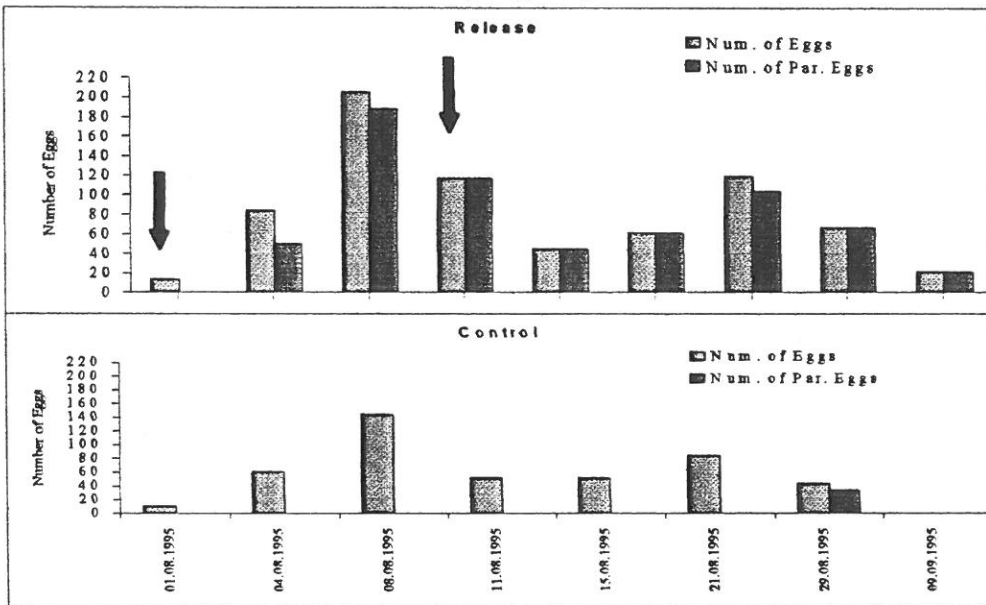


Figure 1. The parasitization rate of *O. nubilalis* by *T. evanescens* in release and control plot in 1995.(01.08.1995= Previous counting) (→: Release).

The release studies of other countries have similar results (Hassan 1981b, Hassan et al. 1986, Bigler 1986 and Stein 1987). For example, Hassan et al. (1978) made four weekly release of *T. evanescens* at rates of 45.000 parasites/ha/release to control European corn borer on corn resulted in parasitism rates from 63.0 to 84.3%, damage was reduced 75.5 and 94.8% compared to the control in Germany. Bigler (1983) reported that releases of *T. evanescens* at weekly intervals (150.000 parasites/ha/yr) to control European corn borer on corn in Switzerland resulted in parasitism rates of 75.2, 18.8, 83.3 and 82.7% in 1978, 1980, 1981 and 1982 respectively. Neuffer (1982) reported that 2-4 releases of *T. evanescens* at a rate of 240.000-300.000 parasites/ha gave effective results between 80.5 - 96.8% in reduction of infestation. In Germany, two treatments 75.000 per ha are being carried out starting at the beginning of the *O. nubilalis* adult flight, as indicated by light traps and the reduction in the number of *O. nubilalis* larvae was obtained by the releases compared to untreated plots, varied between 70 and 93% in this experiment (Hassan et al. 1986, Stein and Hassan 1988).

As a result, in this study, percentages of parasitism in the egg parasitoid-treated plot and the untreated plot were 89.19% and 8.17% respectively and the reduction of the number of infested plants in released plot in comparison to the control was 63.63% in 1995 (Table 3).

Suter and Babler (1976) made three weekly augmentative release of *T. evanescens* in the experimental corn fields in Switzerland and reported that rates of parasitism of European corn borer eggs were 91.8 and 89.9% as compared to 8.3% in untreated area. Infested plants in two untreated fields totaled 68.3-95.2% compared to 4.0 and 5.6% in the release fields. Research by Suter and Babler (1976), Hassan et al. (1978) and Bigler (1983) showed that parasitism rates 80% or greater were effective in reducing larval population of the European corn borer. These results were supported our studies.

At harvest, the average grain was increased by 772.8 kg/da in treated plot and 631.1 kg/da in the untreated plot. And also, the average 1000 grain weights were 326.0 gr and 275.9 gr in the treated and untreated plot respectively (Table 3). The number of larvae and pupae in the egg parasitoid treated plot was lower than that in the untreated plot. This clarified that the density of European corn borer was decreased by the release of the parasitoids and at least two applications of chemical insecticides were completely avoided. In addition, Hassan et al. (1978) reported that the reduction in the level of infestation by *O. nubilalis* at harvest was 77.6-89.8%.

Table 3. Production level in treated and untreated plot at harvest in 1995.

Data at harvest	1995	
	Treated plot	Untreated plot
Yield (kg/da)	772.8	631.1
1000 grain weight (gr)	326.0	275.9
The reduction of the number of infested plant (%)	63.63	-
The reduction of the number of larvae (%)	71.44	-

According to all these results showed that efficiency of *T. evanescens* in controlling eggs of *O. nubilalis*, in reducing the damage rate of European corn borer was satisfied and therefore resulted in good production and quality of maize. And also, it is clear that, *T. evanescens* is a better biological control agent for *O. nubilalis* eggs and must be used in the Integrated Pest Management (IPM) in Turkey.

Natural Parasitization Rate of *O. nubilalis* eggs by *T. evanescens*.

In the second crop maize, *O. nubilalis* gave two generations in Çukurova region of Turkey. The beginning of the first generations of *O. nubilalis* occurred on 20 July 1995 and lasted till the first week of August. The beginning of the second generations of *O. nubilalis* occurred on 24 August 1995 and continued till mid-September. The periods of adult activity for *O. nubilalis* and *T. evanescens* are similar throughout the vegetation period.

In the first subregion, the first parasitized egg mass was found on 20 July 1995. From this date, parasitism continued till the end of the season and gave 100% parasitism on the 24 August 1995. During the vegetation period, totally 4143 eggs were collected and 1903 eggs were found to be parasitized. Average parasitism rate was 45.93%.

In the second subregion, the first parasitized egg masses was found on 31 August 1995. During the vegetation period, totally 896 eggs were collected and only one egg mass was found to be parasitized because of using heavy insecticide applications. Average parasitism rate was 1.45%. In this area, maize plantation covered nearly 60% of the total. Farmers

were accustomed to control stem borers by applying insecticides at least three times in 10 days interval. Because of these, naturally occurred parasitization rate by *T. evanescens* was very low in this second subregion.

In the third subregion, the first parasitized egg mass was found on 13 September 1995. A total of 656 eggs were collected and 251 eggs were found to be parasitized. Average parasitism rate was 38.26% (Figure 2).

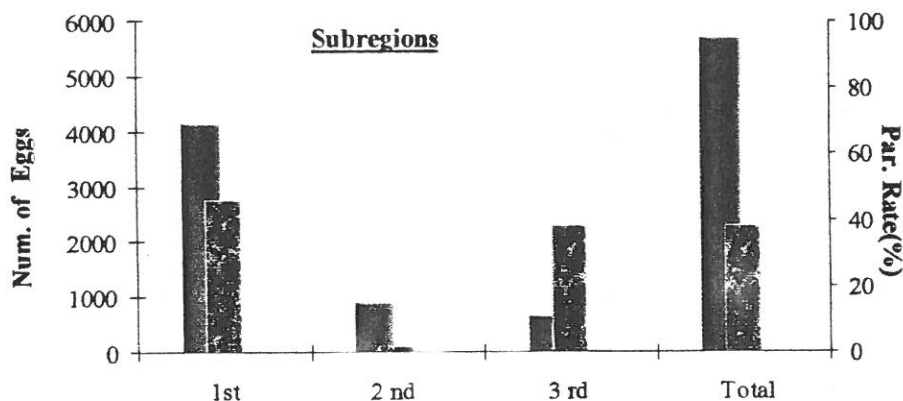


Figure 2. The natural parasitization rate of *O. nubilalis* eggs by *T. evanescens* in three subregion of Çukurova.

When we compared to the three subregion, the highest parasitization rate was occurred in the first subregion (45.93%). The third (38.26%) and the second (1.45%) subregion followed this respectively (Table 4).

Table 4. The natural parasitization rate of *O. nubilalis* eggs by *T. evanescens* in three subregion of Çukurova.

Sampling places	1995	
1 st Subregion	The number of egg	4143
	The number of parasitized egg	1903
	Parasitization rate (%)	45.93
2 nd Subregion	The number of egg	896
	The number of parasitized egg	13
	Parasitization rate (%)	1.45
3 rd Subregion	The number of egg	656
	The number of parasitized egg	251
	Parasitization rate (%)	38.26
Total	The number of egg	5695
	The number of parasitized egg	2167
	Parasitization rate (%)	38.05

As a result, natural parasitization rates in three subregion were 45.93%, 1.45% and 38.26% in the first, second and third subregion respectively. The average of natural parasitism was 38.05% in 1995 (Table 4). The results indicated that natural parasitism was

high the first subregion where pesticide use was minimal and low in the second subregion where pesticide use was intensive.

Similar results were obtained in the previous studies about natural parasitism of egg parasitoids in the different parts of Turkey. Kayapınar and Kornoşor (1992) reported that *T. evanescens* was the most effective parasitoid of *O. nubilalis* in Çukurova region with an average natural parasitization rate of 2.30%, 27.17% and 51.06% in 1988, 1989 and 1990 respectively. During a three-years study, parasitization rate have been found to be 52.21%, 1.30% and 5.01% in the first, second and third subregion respectively. Coşkuntuncel (1995) found that natural parasitism by *T. evanescens* was 52.66% and 16.16% in Çukurova region in 1993 and 1994. In Black Sea Region, Özdemir (1981) recorded that *T. evanescens* was the most effective parasitoid of *O. nubilalis*. In this region, the rate of parasitization was reach to 96%. Uzun (1994) indicated that natural efficiency of this parasitoid was 2.69-100% on eggs and 6.5-100% on eggs masses of the second generation of *O. nubilalis* during 1991-1992 in the Aegean Region. Melan and Kedici (1993) found that parasitization rate of eggs was 87.24-98.61% and 97.03% in Bolu and Sakarya during 1991-1992. Besides, parasitism on eggs of overwintered adult of *O. nubilalis* was 20.79%, 31.58%; 58.53% and 90.17% on eggs of first generation in Bartın and Zonguldak in 1993.

If we compare the natural parasitization rate in some European countries, parasitization rates of the eggs were 22.7% and 24.6% in Romania (Ciudarescu 1982, Muresan 1987) and 3.2-7.0% in Yugoslavia (Manojlovic 1984). Heyde (1990) reported that *T. evanescens* is the most important egg parasitoid and has been used extensively in the world with an effectiveness up to 90% in the corn fields.

In conclusion, *T. evanescens*, Turkish race, is an effective egg parasitoid and approximately 1.45-53 % natural parasitization if supported with inundative releases with this parasitoid, biological control of European corn borer will be successful in Turkey.

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**Natural Impact of Egg Parasitoid, *Trichogramma evanescens*
(Westwood) (Hym.: Trichogrammatidae) of *Ostrinia nubilalis*
(Hbn.) (Lep.: Pyralidae) in West Black Sea Region**

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Abstract

Studies related to the determination of the egg parasitoids of European corn borer (*Ostrinia nubilalis* Hbn.) have been carried out in 1991 –1993 in Bolu, Sakarya, Bartın and Zonguldak provinces.

During the surveys, first and second generation egg batches of the pest were collected. Egg batches were collected from randomly selected maize plants. At least 10 egg batches were collected from each field. All samples were cultured in the laboratory until the adult emergence of the parasitoids completed and so parasitization rate was determined.

Eggs parasitization rates of the pest were determined as 94.74 % in Bolu province. Parasitization rates of the eggs of ECB were determined as 97.03 % in Sakarya province. Parasitization rates of the first generation eggs of ECB 20.79 % in Bartın and 31.58 % in Zonguldak provinces. These rates were 58.53 % in Bartın and 90.17 % in Zonguldak in the second generation eggs .

It was concluded that natural impact of egg parasitoids was high in Bolu, Sakarya, Bartın and Zonguldak provinces.

Key words : *Ostrinia nubilalis*, egg parasitoid, parasitization, *Trichogramma evanescens*

Introduction

Maize is the third major crop after wheat and barley with 550.000 ha. growing area and 2.500.000 ton production in Turkey (Anonymous 1995). It is consumed as human food as well as industrial crop.

European corn borer (ECB) is one of the most important pests of maize in Turkey . It has become more serious with the second crop maize production. Since the larvae of the pest feed in the stems, it is impossible to obtain sufficient control through chemical treatments. Because of the low effect of chemical control and undesirable effects of the insecticides on the environment, biological control method is also used for the control of the pest and this method is the only effective way for the control of the pest in many countries.

The first biological control application was the introduction of 2.993.605 parasitoids species from different genera from Europe to USA in 1920-1938. From these species *Eriborus tenebrans* Graw, *Phaeogenes nigridens* Wesm, *Chelonus annulipes* Wesm, *Macrocentrus grandis* Goid, *Eulophus viridulus* (*Sympiesis viridula* Tohms) and *Lydella thompsoni* established in the region (Clausen, 1978). In successive years studies are focussed on the most promising parasitoid species, *Trichogramma* spp. During the last three decades the use of egg parasitoids, *Trichogramma* has become very common. These parasitoid species have been largely used against many pests on sugar cane, fruits and vegetables in more than 30 countries.

Detailed studies have been carried out on the bio-ecology and chemical control of ECB in Turkey. Besides, natural enemies of ECB were determined and mass rearing and release of egg parasitoids were investigated (Coşkuntuncel and Kornoşor, 1996).

This study was carried out in order to determine the egg parasitoids of ECB and their natural impact on the pest in Bolu, Sakarya, Zonguldak and Bartın Provinces.

Material and Method

Material of this study were ECB eggs, *Trichogramma evanescens*, Laboratory host of the parasitoid, flour moth (*Ephestia kuehniells* Zell.) and light traps.

Studies related to the determination of egg parasitoid of ECB have been carried out in Bolu, Sakarya, Zonguldak and Bartın provinces in 1991-1993.

Survey Studies

Egg masses of the first and second generation of ECB were collected from randomly selected maize plants. At least 10 egg masses were tried to collect from each field. These egg masses were put into tubes individually, labeled and kept in icebox. In the laboratory these eggs were examined as normal eggs and parasitized (black) eggs and incubated in a room maintained at 25 ± 2 °C and 65 ± 5 % RH for adult emergence. After the completion of parasitoid emergence the eggs were counted again as parasitized-normal, hatched – nonhatched eggs. Natural parasitization rate was found by dividing the number of parasitized eggs to the number of total eggs. These parasitoids were reared on flour moth eggs in the laboratory using the methods given by Bulut (1985) and Bulut and Kılınçer (1987) to obtain parasitoid cultures for identification.

Results

Second generation ECB egg masses collected from Bolu province and parasitization rates were given in Table 1. Parasitization rates were determined as 98.61 % and 87.24 % in Çilimli (Topçular) and Cumaovası counties respectively. Average parasitization rate was found as 94.74 % in Bolu province.

Parasitization rate of the first generation ECB eggs was found as 98.00 % in Sakarya province (Table 2). This rate was 97.03 % averagely in the second generation ECB eggs in Sakarya . In the second generation the maximum parasitization rate was 100 % in Büyükesence and the minimum was 94.28 % in Akyazı counties (Table 2).

Data related to the parasitization rates of the first and second generation ECB eggs were given in Table 3 and Table 4'in Bartın and Zonguldak provinces.

Average parasitization rates were found as 20.79 % and 58.53 % in the first and second generation ECB eggs respectively in Bartın (Table 3).

In Zonguldak these rates were 31.58 % and 89.10 % in the first and second generation ECB eggs (Table 4).

All parasitoid samples were identified as *T. evanescens* West.

Table 1. Egg parasitization rates of the second generation ECB in Bolu province in 13-14 / 8 / 1991

Place	Total egg masses	Egg number	Number of parasitized eggs	Number of emerged parasitoid adult	Egg Par. rate (%)
Cumaovası Merkez-Kışla mahallesi	10	149	130	137	87.24
Çilimli Topçular	18	288	284	290	98.61
Total and average	28	437	414	327	94.74

Table 2. Egg parasitization rates of the ECB in Sakarya province

	Place	Locality	Total egg masses	Egg number	Number of par. eggs	Egg par. Rate (%)
6-8.7.1992 first generation	Adapazarı	Küçükesence	1	9	9	100.00
	Akyazı	Kuzuluk	1	12	12	100.00
	Ferizli	Merkez	4	55	55	100.00
		Kavakdüzü	6	72	72	100.00
	Hendek	Merkez	4	36	36	100.00
	Söğütlü	Akgöl	2	40	40	100.00
	Adapazarı	Büyükesence	13	195	195	100.00
		Küçükesence	31	408	398	97.55
		Sarıcalar	21	248	241	97.18
	Total and average		65	851	834	98.00
19-20.8.1992 second generation	Akyazı	Kuzuluk	16	227	226	99.56
		Yahyalar	24	297	280	94.28
		Total and average	40	514	506	98.44
	Ferizli	Kavakdüzü	22	326	312	95.71
	Hendek	Merkez	18	187	177	94.65
	Söğütlü	Akgöl	38	573	559	97.56
		Total and average	183	2461	2388	97.03

Table 3. Egg parasitization rates of the ECB in Bartın province

	Place	Locality	Number of egg masses	Number of egg	Number of parasitized eggs	Egg Par. rate (%)
8.7.1993 first generation	Merkez	Aygırlar	2	21	21	100.00
	Merkez	Kazpınarı	2	39	0	0.00
	Merkez	Terkehaliller	3	41	0	0.00
	Total and average			7	101	21
26-27.8.993 second generation	Merkez	Aygırlar	1	11	3	27.27
		Esenyurt	12	135	128	94.80
		Kazpınar	5	62	12	19.35
		Terkehaliller	4	50	8	16.00
	Total and average			22	258	151

Table 4. Egg parasitization rates of the ECB in Zonguldak province

	Place	Locality	Dates	No. of egg masses	No. of egg	No. of par. eggs	Egg par. rate (%)
first generation	Merkez	Türkali	17.6.1993	1	13	0	0
	Alaplı	Kılçak	6.7.1993	2	17	0	0
			26.7.1993	2	23	10	43.48
	Çaycuma	A.Kayıkcılar	7.7.1993	1	7	5	71.43
		Kayabaşı	8.7.1993	2	23	13	56.52
	Ereğli	Aydınlar	6.7.1993	3	34	0	0
		Yazıcılar	17.6.1993	1	14	0	0
			6.7.1993	2	21	20	95.24
	Total and average				14	152	48
second generation	Merkez	Türkali	24 -26. 8.93	5	74	74	100.00
	Çaycuma	A.Kayıkcılar		15	256	224	87.50
		Kayabaşı		4	52	52	100.00
	Top. ve ort.			19	308	276	89.61
	Devrek	Albuzlar		26	309	278	89.98
		Kaypaklar		15	214	188	87.85
	Total and average				41	523	466
TOTAL AND AVERAGE				65	905	816	90.17

Discussion

At the end of survey studies 94.74 % egg parasitism was determined in the second generation ECB eggs in Bolu province in 1991. This rate was 100 % in the first generation ECB eggs. High egg parasitism was also observed in the first and second generation ECB eggs in Sakarya province in 1992. In a study carried out in Blacksea region 96 % egg parasitism was determined (Özdemir, 1981).

In Bartın and Zonguldak provinces parasitization rates were 20.79 % and 31.58 % in the first generation ECB eggs respectively in 1993. In these provinces there is no irrigation and the sowing dates were very different and so the maize vegetation. Because of this there was not a synchronism between the plant phenology and insect biology and the population density of the pest was very low, especially in coastal area. During the second generation plant phenology was suitable for the pest and the egg parasitism was higher when compared with the first generation. Egg parasitization rates were 58.53 % and 90.17 % in Bartın and Zonguldak provinces respectively during the second generation.

It was concluded that natural egg parasitism by *Trichogramma evanescens* has great importance to suppress the ECB population in Bolu, Sakarya, Bartın and Zonguldak provinces. Depend on the results of the studies carried out in many countries natural enemies, especially egg parasitoids have important role to suppress the pest (Özdemir, 1981; Manojlovic, 1985 ; Sheng et al., 1986 ; Bigler, 1986; Zhili, 1986) It can be said that conservation and manipulation of present natural enemies have great importance for this region. This is very important for natural biological control. ECB problem can be prevented by preserving the present status of the region. So chemical control should be avoided in corn fields. Besides in the places where pest density is high but the parasitization is low augmentative releases should be implemented to support the natural egg parasitization.

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Performance of Pioneer Bt-Hybrids to Control ECB and Pink Stalk Borer (PSB)

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Abstract

A management study was conducted to determine the performance of Pioneer corn hybrids containing the MaisGard* gene under different levels of European corn borer (*Ostrinia nubilalis* Hbn.) or Pink stalk borer (*Sesamia nonagrioides* Lef.).

1. Pioneer conducted studies in 1997 and 1998 to evaluate its hybrids under different levels of corn borer infestation in USA.
2. Pioneer conducted also studies in 1998 to evaluate its hybrids under different levels of stalk borer infestation in Turkey and France

Results:

Results from 39 locations showed that Pioneer hybrids with the MaisGard* gene:

- for ECB

- Had significantly less feeding damage under low, medium and high corn borer infestations
- Provided a yield advantage of 8.8% over similar genetics without the MaisGard* gene for every corn borer cavity present in the untreated checks.

Results from 4 locations showed that Pioneer Bt-hybrids:

- for PSB

- Had significantly less feeding damage (length of tunneling, number of tunnels) under low, medium and high stalk borer infestations.
- Reduced significantly the average number of larvae per plant.
- Provided a yield advantage from 12.7% to 109% over similar genetics without the MaisGard* gene.

Conclusion: Pioneer hybrids with the MaisGard* gene have proven value, even at low levels of corn borer or stalk borer infestation.

Key Words: *Ostrinia nubilalis*; European Corn Borer, *Sesamia nonagrioides*; Pink Stalk Borer, Bt, Maize.

Introduction

The introduction of maize in different continents was followed by the attack of native insect species that became pests on maize fields. The pyralid *Ostrinia nubilalis* (Hbn.) is the main pest of maize in France, Italy, Central Europe and also in North America, where it has been introduced accidentally (DICKE and GUTHRIE, 1988). Now the European Corn Borer (ECB) is the number one pest problem in the United States. ECB typically produce two damaging generations per year in Corn Belt States.

In some zones, several species of noctuids of the *Sesamia* genus have been reported as the main problems for the cultivation of maize. Among them, *Sesamia nonagrioides* Lef. causes important losses in Morocco (LESPEL and JOURDAN, 1940) and Southern Europe (ALFARO, 1955; ANGLADE, 1961a; PROTA, 1965; TSITSIPIS and al., 1984). In Greece, late corn is heavily infested, and losses of 80-100% are not unusual (TSITSIPIS and al.,

1987). In Turkey Pink Stalk Borer cause important damage at second crop maize (Symposium on Corn borers and control measures, 1988).

Pink Stalk Borer generally produce two to four generations per year depending on regions and climatic conditions.

Designing effective means of control appears to be more and more important as these pests damaging more and more maize crops in diverse regions worldwide with severe yield losses. In spite of insecticide control, good control is very difficult to succeed due to application timing. The discovery of insecticidal properties of *Bacillus thuriengensis* since 1901, then the work done by researchers on identifying the gene which coded for the protein, isolating it, and using tools of biotechnology, inserting it into the genome of plants allow now farmers to use Bt maize hybrids to manage both ECB and PSB.

ECB and PSB pressures vary widely due to geography and weather. Because of this variation, Pioneer has conducted studies to evaluate its hybrids under different levels of ECB and PSB. This presentation will report on the results of these studies, and the performance they demonstrate for Pioneer hybrids with the MaisGard* gene.

Objectives of Corn Borer and Stalk borer Management Studies

These Management studies were conducted to determine the value of Pioneer maize hybrids containing the MaisGard* gene under different levels of ECB or PSB pressure.

Specifically, the trials addressed the following questions:

- How effective are Pioneer maize hybrids containing the MaisGard* gene in controlling damage from ECB or PSB?
- What is the yield advantage of resistant Pioneer maize hybrids under various levels of corn borer infestation?

Experimental Procedures

Pioneer hybrids with the MaisGard* gene and genetically similar Pioneer hybrids not containing the gene were planted:

For ECB -In plots 6m long by 4 rows wide at 39 locations across the Corn Belt in USA.

For PSB -In plots 5m long by 4 rows wide, 3 replications, and 2 planting dates at 1 location in Turkey.

-In plots 12m long by 4 rows wide, 3 replications, and 2 planting dates at 3 locations in France.

IN ECB experimentation different hybrids were used:

- Use of non-resistant hybrids not treated (3223, 3394, 3489, 3563, 3730, 3752, 3751, 37M81, 38P05 and 3893).
- Use of Pioneer brand maize hybrids containing the MaisGard* gene (31B13, 33A14, 33V08, 34T14, 33Y09, 34R06, 34R07, 34A03, 35M02, 35N05, 36G32, 37R71, 38P06, 36K27, 36F30, 38B22, and 38W36).

Entries varied at each location to fit areas of adaptation

IN PSB experimentation different hybrids were used:

- Use of non-resistant hybrids not treated (3394, 3489, 3751, 3563)
- Use of Pioneer brand maize hybrids containing the MaisGard* gene (33V08, 34R07, 36F30, 35N05)

Evaluations

Plots were evaluated for corn borer damage by examining 10 consecutive plants (15 in PSB experimentation) in each plot. Since stalk tunneling is the best single indicator of yield loss potential, stalks were split on these 10 or 15 plants and the number and total length of tunnels were recorded.

The 39 ECB locations were grouped into low, medium and high levels of infestation, defined as follows:

- Low – less than 2.5 cm of total stalk tunneling/plant
- Medium – 2.5 to 5 cm of total stalk tunneling/plant
- High – greater than 5 cm of total stalk tunneling/plant

The 4 locations x 2 planting dates showed up different PSB levels of infestation in terms of average number of larvae per plant Figure1, allowing to observe the comportment of Pioneer maize hybrids under different PSB pressures. We did not get any result from the first planting in France (no PSB infestation).

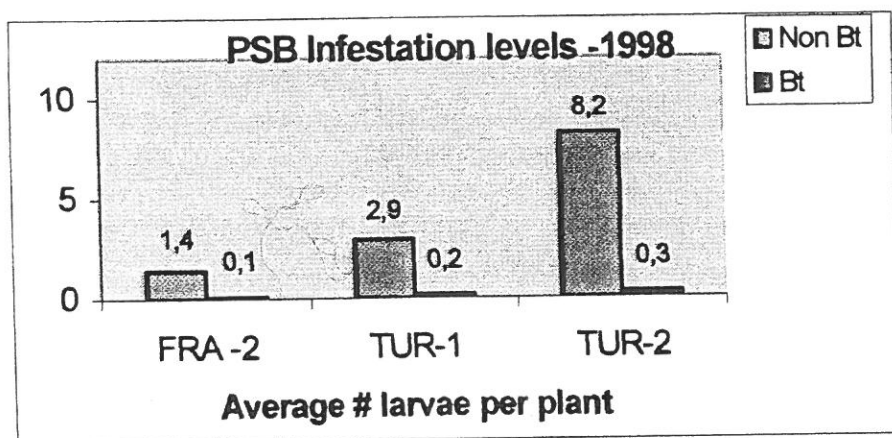


Figure 1. PSB infestation levels based on average number of larvae per plant

Experiment Results

Yield results are shown in Figure 2. And 3. Pioneer maize hybrids containing the MaisGard* gene had significantly higher yields than untreated checks, under low, medium and high levels of infestation of ECB or PSB.

In ECB experiments, Pioneer hybrids produced significantly higher yields with a 2.5 qu/ha yield advantage over the check in low infestation environment. Under medium and high European corn borer pressure, Pioneer hybrids with MaisGard* produced 12.6 and 17.6 qu/ha yield advantages over the untreated check, respectively.

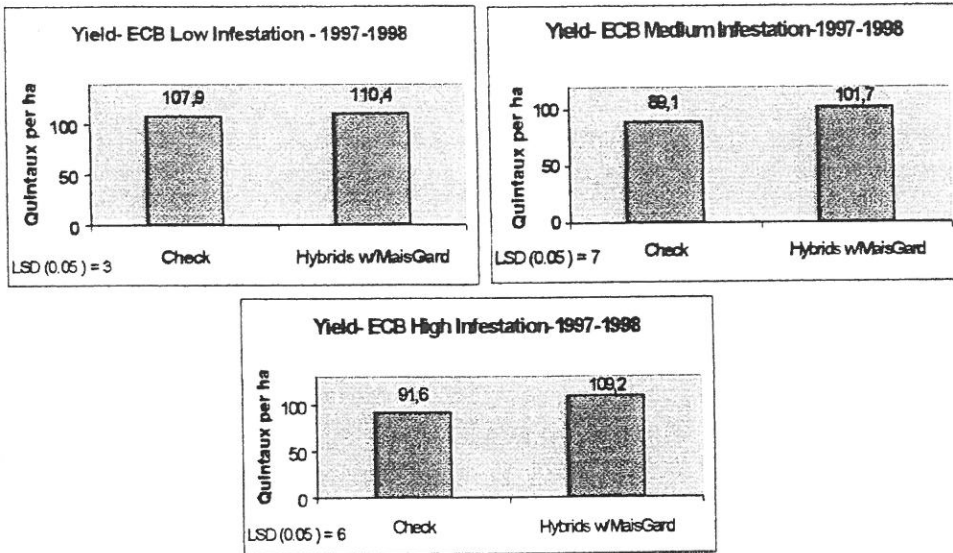


Figure 2. Yield results under low, medium and high infestation levels of European Control Borer

In PSB experiments, as we observed with ECB experiment results Pioneer hybrids with MaisGard* produced significantly a 11.6 qu/ha yield advantage over the check in low infestation environment (1.4 larvae per plant). Under medium and high PSB pressure, respectively 2.9 and 8.2 larvae per plant, Pioneer hybrids with MaisGard* produced 17.5 qu/ha and 47.2 qu/ha yield advantages over the untreated checks, respectively. Due to low pressure at first planting date in France we didn't get any results.

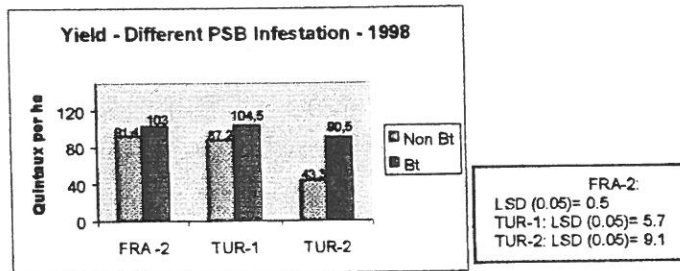


Figure 3. Yield results under low, medium and high infestation levels of Pink Stalk Borer

The relationship between number of cavities and yield advantage of Pioneer hybrids with the MaisGard* gene is depicted in Figure 4. These data indicate that over 39 locations and 2 years, Pioneer hybrids with the MaisGard* gene gave a yield advantage of 8.8% for every corn borer cavity. This is higher than the "classic" corn borer loss estimates of 5% to 6% per borer, which have been used extensively in economic threshold formulas to determine whether to treat for corn borer.

Number of ECB Tunnels vs Yield Advantage for Hybrids with the MaisGard Gene

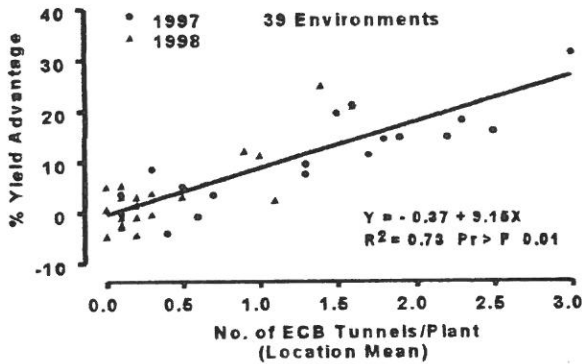


Figure 4. % yield advantage by number of tunnels for resistant vs non-resistant hybrids.

Despite of few locations with PSB experimentation, we observed the same pattern Figure 5., which means more tunneling is observed in non-resistant versus resistant hybrids more yield advantage you gain with Pioneer hybrids with the MaisGard gene

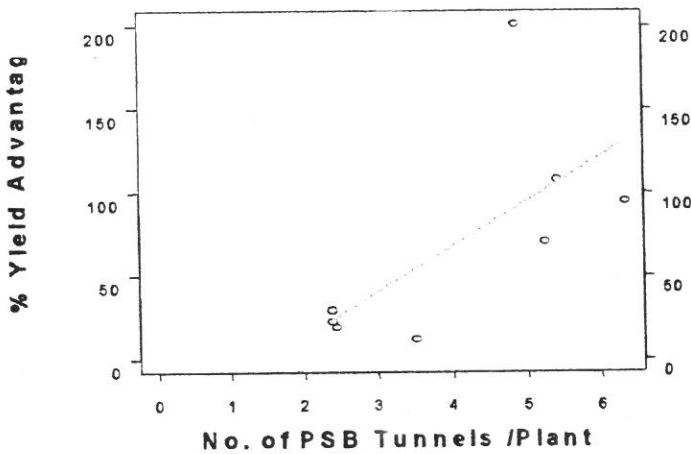


Figure 5. % yield advantage by number of tunnels for resistant vs non-resistant hybrids

Value of Pioneer Resistant Hybrids

Pioneer hybrids with the MaisGard* gene have proven value, even at low levels of corn borer infestation. Nevertheless, it is critically important to preserve the long-term effectiveness of this exciting new technology for corn insect management. Pioneer is committed to resistance management and has implemented a strategy for managing insect resistance that includes:

- Discovery of additional resistance genes
- Monitoring of ECB and PSB populations in the field
- Promoting refuges for ECB and PSB populations
- Developing education programs
- Responding quickly to unexpected damage complaints.

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Determination of *Bacillus thuringiensis* Varieties Pathogenic on Maize Stem Borers and their Insecticidal Activity

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Abstract

This study aimed to isolate *Bacillus thuringiensis* varieties from Maize stem borers; to determine these varieties; to select virulent ones; to produce them and to test their insecticidal activity against *Ostrinia nubilalis*.

Seventeen isolates obtained from 55 dead/diseased *O. nubilalis* larvae and 2 *Sesemia nonogroides* isolates were identified as *B. thuringiensis* var. *kurstaki* on the basis of biochemical and serological tests.

In the pathogenicity tests performed with 6 isolates selected from *O. nubilalis* isolates at the end of fourth day 9 of 10 alive larvae were exist in negative control, but larvae number varied from 0 to 0.6 in other characters, showing high insecticidal effect on the pest.

Spore-crystal complexes of 2 *O. nubilalis* isolates (Bt/5 and Bt/26) which were formulated by HD-1 technique and produced by deep fermentation method controlled *O. nubilalis* larvae at the rate of 67.8 and 92.8 % at 3000 µg/ml respectively while *Lobesia botrana* isolate (Bt/21) was 92.8 % effective at 1000 µg/ml dosage on *O. nubilalis* larvae. *L. botrana* isolate was also found to be superior to the others by having high spore productivity of 1.6×10^{12} spore/gram.

It would be useful to continue the studies by optimizing fermentation condition, by finding new very well adapted varieties to the environment as well as by solving pilot production pit falls.

Introduction

Microbial insecticides are widely used along with the conventional insecticides and with the other control measures and take place in IPM programs for insect management in agriculture, forestry, and for common fly controlling.

Bacillus thuringiensis has the 90 % part of the total microbial insecticides used in the world. This organism has been registered to be utilized against 92 insect species. The studies, beginning with the first commercially available preparations in 1958, have been concentrated on improvement and field applications up to 1970's at the time widely usage have been built up (Burgess, 1981). Recently, the studies have mostly been centered upon finding the new varieties, determination of host range and searching out the application conditions.

B. thuringiensis isolates are classified on the basis of morphology and biochemical characters in 1958 (Heimpel and Angus, 1958). Serological classification is done by means of antigenic features of the flagellum proteins (Barjac and Bonnefoi, 1962) and definitions on the variety level were achieved by electrophoretic partition of esterases in the vegetative cells (Norris, 1964). On the other hand, like crystal serology (Krywienczyk and Fost, 1980), plasmid type electrophoresis (Selander et al., 1986) some modern techniques are being utilized for determination.

Although there are many definition methods, serological classification is considered to be the simplest and the most reliable one and 27 antigenic groups and 34 serovarieties in the 7 subgroups are identified (Barjac and Frachon 1990). Recently, 58 serovar have been identified which fall into 45 antigenic groups.

Materials and Methods

For the isolation of *B. thuringiensis* strains, diseased/dead larvae collected from maize production areas were first washed in sterile water and one or two then transferred in to the vials of sterile water and pasteurized in water bath at 65 °C for 10 min, so that the larvae were cleaned from vegetative cells on their surfaces. Larvae were extracted in 1-2 ml of sterile water or saline, from extracted 0.2 ml was taken and put into the 5 ml Nutrieth Broth (Difco) tubes and incubated at 32 ± 1 °C for 48 hours. Inoculations were made by loops into the Petri dishes which contains Nutrient Agar (NA-Difco) from the tubes showed turbidity. Pure isolates were maintained on NA slants at 4 °C.

Pathogenicity tests: Pathogenicity tests were performed on the first instars of *O. nubilalis*. From the 24 h old NA cultures of 6 selected isolates was taken by a loopfull bacteria and transferred into the 5 ml of NB and incubated at 30 °C for 48 hours. 4 % of these liquid cultures was then transferred into the 200 ml NB medium and incubated into the shaker (280 rpm) at 30 ± 2 °C for 72 hours. The cultures were centrifuged at 200 g for 20 min in order to partition into spores and crystals (Dulmage, 1970). From the spore-crystal pellets, 10^8 - 10^9 spore/ml suspensions were prepared in sterile water and sprayed by a hand sprayer onto the larvae feeding media composed of maize leaves and stems which were placed in to Petri dishes in which 10 *O. nubilalis* first instars were placed when the food got dried. For each isolate 30 larvae were employed for 3 Petri dishes which were incubated at 25 ± 1 °C for 4 days in growth chambers. At the end of this period, alive larvae were counted and the effectiveness of the isolates was determined. For negative control dishes only sterile water, for positive control dishes *B. t.* var. *kurstaki*, var. *ostrinia* and var. *thuringiensis* reference strains which were supplied from Pasteur Institute (France) were used. Re-isolations were performed by utilizing the same technique.

Isolate identification: For identification serological tests were used along with the some morphological, physiological and biochemical ones. The presence of endospore (staining by malahite green) and colony pattern were the morphological feature to be checked. The physiological and biochemical tests were selected from Barjac and Bonnefoi (1968) and Norris (1964).

Serological identification was based on H-antigens. A loop full culture was taken from the 24-hour old NA cultures of both 19 isolates originated from maize stem borers and *B. t.* var. *kurstaki*, var. *ostrinia* and var. *thuringiensis* reference ones and inoculated into 5 ml NB tubes and incubated at 30 °C at 280 rpm by rapidly shaking. Inoculation was made at the beginning of logarithmic growth (6-7 hours after inoculation) into the 0.3 % of soft NA in the Craige tubes from which another inoculation was made 24 hour later into the new Craige tubes. At the end of the incubation at 30 °C for 24 hours, inoculation was completed into the 100 ml of sterile NB which were shaken at 30 ± 1 °C for 10 hours. 0.5 % formalin was added to the cultures and maintained at 4 °C as stock suspension. Stocked cell suspensions were then diluted to 10^6 cell/ml (at Spectronic 20 with 72 % permeability at 490nm) by adding 0.95 % NaCl solution. First of all, the titers of antisera were determined. For this 0.1 ml of dilution series of antisera was put into the Kahn tubes on which 0.9 ml of reference culture suspensions was added and incubated at 37 °C for 2 hours in dark. At the end of the incubation on the bottom of the tubes floccular sediment, which was easily dispersed by shaking, was considered to be positive (Barjac, 1981). The isolates originated from insects were also determined by same serological procedures. In addition, by using saturation technique (Barjac, 1981) it was checked whether the antigenic features of the isolates were the same of the identified *B. thuringiensis* varieties.

Producing and preparation of some *B. thuringiensis* isolates: *O. nubilalis* isolates marked Bt/5 and Bt/26 and *L. botrana* Bt/21 isolate were prepared for experimental use by deep fermentation technique.

NB liquid cultures prepared according to Dulmage (1970), were added at 3 % into the erlenmayer flasks of 2 liter sterile Tryptone medium (Tryptone 10.0g, dextrose 5.0g, corn starch 5.0g, yeast extract 2.0g, K_2HPO_4 1.0g, distilled water 1000ml). Cultures were incubated at 32 °C at 280 rpm by shaking until the vegetative cells broke down (85hrs). In order to get spore-crystal complexes HD-1 steps were followed (Dulmage et al., 1970). For this, pH of fermentation output was checked and adjusted to pH 7.0 with HCl, then centrifuged for 10 minutes at 500 g, supernatant was removed and 0.1 volume 5 % lactose solution was added to the remainder and stirred for 30 minutes on the magnetic stirrer. 4 volume acetone was slowly added during the stirring and stirred another 30 minutes. After letting stand for 10 minutes filtered through 1 No. Whatman, and the filtrate was removed. 5 ml acetone was added to the remnant on the filter paper and refiltered. Washing and filtering was repeated one more time. Remnant was room temperature dried and ground to homogenous dust.

Determination of spore number: 1000 mg of the homogenous dust was taken twice and diluted with sterile water (10^{-1} 10^{-10}). Suspensions were pasteurized at 65 °C for 10 minutes and 0.5ml of dilution series was added to 10ml NA tubes at 47 °C and poured to the Petri dishes. For each dilution serial 3 dishes were inoculated and kept at $32 \pm 1^\circ C$ for 48 hours. Colonies were counted and active spore numbers in 1gram dust were determined at the end of the incubation period.

Bioassay: Tests were conducted on the 0-24 hour old *O. nubilalis* larvae. Dosage series of 0-10-30-100-300-1000-3000 $\mu g/ml$ were made from the dust preparation and sprayed onto the feeding medium composed of maize leaves and stems in culture jars by hand sprayer. 10 larvae were put into each for and kept at $25 \pm 1^\circ C$ and $60 \pm 10\%$ RH for 5 days in a growth chamber, mortality ratio was defined at the end of the incubation period. For negative control, water was sprayed onto the feeding media. Tests were carried out with three replications.

Results and Discussion

Totally 19 *B. thuringiensis* isolates were obtained from 55 dead/diseased larvae samples of *O. nubilalis* and 2 *S. nonogroides* collected from maize production areas. On the pathogenicity tests performed with 6 isolates selected from *O. nubilalis* strains at the end of the fourth day, 9 alive larvae were exist in the negative control, while alive instar number was varied from 0 to 0.6 in the treated characters showing high insecticidal effect of isolates.

Colonies were shallow, dull and irregular on the NA medium and endospores were produced. Biochemical and physiological features of these isolates were given on the Table 1. As shown on Table 1 the isolates exhibited almost the same features.

The data obtained from serological tests were shown on the Table 2. Totally 19 strains originated from maize pests and the reference strain gave reactions to the var. *kurstaki* antiserum up to 1/10240 dilution. But these isolates showed no reaction to the antiserum prepared for the var. *ostrinia* while var. *ostrinia* reference strain displayed reaction up to its 1/10240 dilution.

The biochemical and physiological properties of 19 isolates obtained from the 57 dead/diseased maize pests showed similarities between those of var. *kurstaki* as described by Barjac (1990)'s identification table. These isolates also gave positive reaction against the

var. *kurstaki* antiserum. It was proven by field and laboratory tests that *B. t.* var. *kurstaki*, var. *ostrinia*, var. *thuringiensis* and var. *alesti* had entomopathogenic effect on the *O. nubilalis* larvae (Krieg and Langenbruch, 1981). Otherwise, it was declared that var. *kurstaki* had pathogenic effects on *S. inferens* (Krieg and Langebnbruch, 1981). Nowadays, transgenic maize varieties which held toxin gene of *B. t.* var. *kurstaki* had place in the commercial production for the control of stem borers.

Data connected with the insecticidal activities of the Bt/5, Bt/26 and Bt/21 preparations which were formulated by HD-1 technique and produced by deep fermentation method to the *O. nubilalis* larvae were given on Table 3. As shown on Table 3, Bt/5 preparation caused the highest mortality ratio at 3000 µg/ml dosage. Bt/21 *L. botrana* strain gave rise to 100 % mortality at 3000 µg/ml, whereas Bt/26 strain 93.33 % at the same dosage. Spontaneous death ratio was 6.66 % in this trial. Bt/21 isolate was found to be superior to the others by having high spore productivity of 1.6×10^{12} spore/gram. It was found out that for determination of the efficacy of the *B. thuringiensis* formulations it was not sufficient to detect the spore numbers, fermentation circumstances or serotype, but it was compulsory to test the insecticidal activities by bioassays. On the other hand, endotoxin quantity had the inclination to vary depending on the whether the fermentation conditions or the varieties even with the different strains (Dulmage, 1970).

It would be useful to continue the studies on the preparation of formulation by eliminating or reducing negative impacts on the applications, by optimizing fermentation conditions, by finding new very well adapted strains to the environment as well as by solving pilot production pitfalls.

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Table 1. Some biochemical and physiological properties of *Bacillus thuringiensis* isolates obtained from maize stem borers.

Strains	Origin	AMC	ADH	TW- esterase	Urease	Amylase	Aesculin	Proteol.	Mannose	Sucrose
Bt/1	On	+	+	+	+	+	+	+	-	-
Bt/2	On	+	+	+	+	+	+	+	-	-
Bt/3	On	+	+	-	+	+	+	+	-	-
Bt/4	On	+	+	-	+	+	+	+	-	-
Bt/5	On	+	+(s)	+	+	+	+	+	-	-
Bt/7	On	+	+	+	+	+	+	+	-	-
Bt/9	On	+	+	+	+	+	+	+	-	-
Bt/10	On	+	+(s)	+	+	+	+	+	-	-
Bt/11	On	+	+(s)	+	+	+	+	+	-	-
Bt/12	Sn	+	+	-	+	+	+	+	-	-
Bt/13	Sn	+	+	-	+	+	+	+	-	-
Bt/18	On	+	+	+	+	+	+	+	-	-
Bt/19	On	+	+	-	+	+	+	+	-	-
Bt/20	On	+	+	-	+	+	+	+	-	-
Bt/26	On	+	+	-	+	+	+	+	-	-
Bt/28	On	+	+	+	+	+	+	+	-	-
Bt/31	On	+	+(s)	-	+	+	+	+	-	-
Bt/32	On	+	+	+	+	+	+	+	-	-
Bt/33	On	+	+(s)	+(s)	+	+	+	+	-	-

On: *Ostrinia nubilalis*, Sn: *Sesemia nonogroides*

AMC: acetyl methyl carbinol, ADH: arginin dihydrolase, TW-esterase: Tween esterase
s: slight reaction

Table 2. Serological reactions of *Bacillus thuringiensis* isolates obtained from dead/diseased larvae of maize pests against tested antisera

Host	Isolate number	Antiserum			H-Antigen	Serovar
		kurstaki	thuringiensis	ostrinia		
<i>O. nubilalis</i> <i>S. nonogroides</i> Reference strains	17	+	-	-	3a, 3b, 3c	Kurstaki
	2	+	-	-	3a, 3b, 3c	Kurstaki
	var. kurstaki	+			3a, 3b, 3c	Kurstaki
	var. thuringiensis		+		1	Thuringiensis
	var. ostrinia			+	8a, 8c	Ostrinia

Table 3. Effectiveness of some *Bacillus thuringiensis* strains isolated from dead/diseased insect larvae against 0-24 hours old *Ostrinia nubilalis* instars

Treatments	Dosage	Dead larvae number			Mortality (%)	Effect (%)
		I	II	III		
Bt/5 1.3x10 ⁹ spore/gr	10 µg/ml	4	6		50.00	46.4
	30 µg/ml	5	6		55.00	51.8
	100 µg/ml	6	6	6	60.00	57.1
	300 µg/ml	6	8	6	66.66	64.3
	1000 µg/ml	6	7	7	66.66	64.3
	3000 µg/ml	7	8	6	70.00	67.8
Bt/26 1.6x10 ¹² spore/gr	10 µg/ml	4	4		40.00	35.7
	30 µg/ml	4	5		45.00	41.1
	100 µg/ml	4	6	5	50.00	46.4
	300 µg/ml	5	5	6	53.33	50.0
	1000 µg/ml	9	7	7	76.66	75.0
	3000 µg/ml	9	9	10	93.33	92.8
Bt/21 2.2x10 ¹² spore/gr	10 µg/ml	5	8		40.00	35.7
	30 µg/ml	6	5		55.00	51.8
	100 µg/ml	8	6	6	66.66	64.3
	300 µg/ml	8	10	8	86.66	85.7
	1000 µg/ml	9	9	10	93.33	92.8
	3000 µg/ml	10	10	10	100.00	100.0
Sterile water		1	0	1	6.60	

Effect of Some *Bacillus thuringiensis* Formulations, Neem Extract and Juvenil Hormone Mimic on Corn Infestation By Large Sugar-Cane Worm, *Sesamia cretica* Led. and Their Associated Predators

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Abstract

The effect of the *Bacillus thuringiensis* formulations (SAN 415 I, Dipel, Thuricide HP, Ecotech, Delfen, Protecto), the Neem extract (Nemazal) and the Juvenil hormone mimic (Admeral) besides the mixtures of SAN 415 I and Nemazal or Admeral on corn infestation by large sugar-cane worm, *Sesamia cretica* Led., and the associated predators were evaluated. Data revealed that the B.t-formulation Ecotech and the mixtures of SAN 415 I and Nemazal or Admeral were the most toxic compounds against the pest, since they exhibited infestation reduction of 82.99, 83.19 and 80.80%, respectively. On the other hand, the least effective compounds were Dipel and Thuricide HP with infestation reduction of 54.7% and 56.50%, respectively.

Data also indicated that the plants in control plots harbored significantly more number of the pest than the treated plots. This led to a significant reduction in the yield. The highest yield amount were found in the plots treated with the B.t-formulation Ecotech and the mixtures of the B.t-formulation SAN 415 I and Nemazal or Admeral and the maize yield amounts were 34.75, 33.25 and 31.50 kg/plot (1/100 feddan) with increase rates of 98.57, 90.00 and 77.14%, respectively.

The tested B.t-formulations were less harmful to the associated predators whereas the Neem extract (Nemazal) and the Juvenil hormone mimic (Admeral) were the most harmful compounds.

Introduction

Maize (*Zea mays* L.) is the main food for some 100 million people. The crop is especially important to many developing countries in Africa, Asia and Latin America where it serves as a human subsistence crop. In Egypt, it is the most dominant summer cereal crop and the area under cultivation amounted to be 1.95 million feddan. many varieties of insects attack maize plant's roots, stem, ears and foliage. The large sugar-cane worm, *Sesamia cretica* Led. is one of the major insect pests of maize in africa and in Assiut (El-Naggar, 1967). The number of egg-masses of *S. cretica* laid on maize plants and intensity of their infestation were faried greatly according to the planting date (Willcocks, 1925; El-Sherif, 1965; El-Sadany, 1965 and El-Naggar, 1967).

In the last few years some authors, recorded an increase in tolerance of many insects in field against organophosphate, carbamate and pyrethroid insecticides. Under such conditions, other compounds of other groups (such as Juvenil hormon mimics and neem extract) that have biological importance, have been introduced to play a basic role in pest management.

Integrated pest control, by minimizing the use of chemical pesticides and using biological agents, promises to reduce pest losses to maize on a continuing basis and at the lowest cost. Some of these agents such as *B. thuringiensis* have shown much potential and offer an excellent alternative to chemical pesticides for IPC program. *B. thuringiensis* as a

bacterial pathogen infecting a wide range of insect pests, is the most common microbial insecticide in use today (Walker, 1979).

The present study was conducted to evaluate the:

Effect of certain chemical and microbial insecticides on large sugar-cane worm, the associated predators and the maize yield.

Materials and Methods

This work was carried out at the farm of experimental station of Faculty of Agriculture, University of Assiut. An area of about $\frac{3}{4}$ feddan was divided into 75 plots of 42 m² each in 1998 season. The plots were planted with maize cultivar "diploid hybrid 215" on April 20, 1998.

The maize plants were sprayed with the tested compounds two times. The first spraying was 3 weeks after planting and the second 2 weeks after the first (Table 1). The tested compounds were applied using a knapsack sprayer. Split plot design was adopted in the experiment and each treatment was replicated four times.

The numbers of infested maize plants with the large sugar-cane worm and the numbers of the associated predators on 10 plants in each plot were recorded. The counts were made after 7 and 14 days from each spraying. The mean reduction of *Sesamia* spp. infestation, as a result of the treatments, were calculated according to Hinderson and Tilton equation, 1955. Analysis of variance was used, and the averages were compared using LSD (0.05). The percentage of the pest infestation reduction and the number of associated predator were used as criterion to evaluate the effectiveness of the tested compounds.

Results and Discussion

The effect of the *Bacillus thuringiensis* formulations (SAN 415 I, Dipel, Thuricide HP, Ecotech, Delfen, Protecto), the Neem extract (Nemazal) and the Juvenil hormone mimic (Admeral) besides the mixtures of SAN 415 I and Nemazal or Admeral on corn infestation by large sugar-cane worm, *Sesamia cretica* Led., and the associated predators were evaluated.

Data summarized in table 2 and illustrated in Fig. 1. The insect pest of major importance and dominance after 3 weeks from the planting of maize was *Sesamia cretica* Led. The associated predators were *Coccinella undecimpunctata* L., *Orius* spp. (*Orius laevigatus* and *Orius albidipennis* Reut.), *Scymnus syriacus* Mars. and true spiders.

Data revealed that the B.t-formulation Ecotech and the mixtures of SAN 415 I and Nemazal or Admeral were the most toxic compound against the large sugar-cane worm, since it exhibited infestation reduction of 82.99, 83.19 and 80.80%, respectively. In agreement with results obtained by Navon (1989), who found that *B. thuringiensis* HD-1-S, 80 was active compound against *Sesamia nonagrioides* larvae. The least effective compounds were Dipel and Thuricide HP and they exhibited reduction of 54.78 and 56.50%, respectively.

The neem extract (Nemazal) and the Juvenil hormone mimic (Admeral) were effective against *S. cretica* and with these treatments the level of the pest infestation has been reduced.

Data also indicated that the plants in control plots harbored significantly more number of the pest than the treated plots. This inevitably led to a significant reduction in the yield. The highest yield amount were found in the plots treated with the B.t-formulation Ecotech and the mixtures of the B.t-formulation SAN 415 I and Nemazal or Admeral and the maize yield amounts were 34.75, 33.25 and 31.50 kg/plot (1/100 feddan) with increase rates of

98.57, 90.00 and 77.14%, respectively. The lowest ones were recorded in untreated plots (17.5 kg/plot).

The maize yield increased moderately by using the other tested insecticides (Nemazal, SAN 415 I, Admeral, Protecto, Dipel and Thuricide HP). The tested B.t-formulations were the safest tested compounds against the beneficial natural enemies (*Coccinella* spp., *Orius* spp., *Scymnus* spp. and true spider) (12.81-19.90% reduction), while Nemazal and Admeral were the most harmful compounds, since they exhibited reduction of 72.08 and 54.00%, respectively.

The combination of B.t-formulation SAN 415 I with Nemazal or Admeral at the field recommended rate of SAN 415 I with 25% of the field recommended rates of the other chemical compounds resulted significant increase in yield and effectiveness against *S. cretica*, while it lead to increase the harmful effect of the B.t-formulation against the natural predators. The combination effects may involve a many and different mechanisms such as simple compatability of the microorganism and the pesticide and this lead to B.t. spores activation.

The statistical analysis of yield data, the infestation of the insect pest and the associated predators on maize plants revealed that B.t-formulations exhibited a higher yield and reduced *S. cretica* infestation with least harmful effect on its associated natural enemies and this leads to quite effect against the pest when combined with resident natural enemies (Falcon, 1971).

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Table 1. Microbial and chemical insecticides tested against the large sugar-cane worm and their rates of application.

Trade name	Chemical or scientific name	Rate of application (%)
SAN 415 I W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Dipel W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Ecotech W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Delfin W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Protecto W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Thuricide HP W.P.	<i>B. thuringiensis</i> var. <i>Kurstaki</i>	0.10
Admeral EC 10 %	4-phenoxy phenyl (RS)-2-(2-pyridyloxy) propyl ether	0.04
Nemazal EC 10%	Neem extract formulation	0.20
SAN 415 I + Admeral	<i>B. thuringiensis</i> var. <i>Kurstaki</i> + 4-phenoxy phenyl (RS)-2-(2-pyridyloxy) propyl ether	0.1 + 0.01
SAN 415 I + Nemazal	<i>B. thuringiensis</i> var. <i>Kurstaki</i> + Neem extract formulation	0.1 + 0.05

Table 2.Effect of some microbial and chemical insecticides on large sugar-cane worm infestation, the associated predators and yield of maize.

Treatments	% Infestation of <i>Sesamia cretica</i>						% Infestation* reduction	Av. no. of predators**/ 100 plant	Yield Kg/plot
	Pre count		After first spraying		After second spraying				
	7 days	15 days	7 days	15 days	7 days	15 days			
SAN 415 I	0.87	10.50	5.25	10.50	11.25	12.50	72.95 c	87.50 b	26.50 b
Dipel	0.97	10.67	6.79	10.67	18.44	23.30	54.78 f	91.25 b	22.25 c
Thuricide HP	1.06	13.82	7.44	13.82	21.27	24.46	56.56 f	87.50 b	21.25 cd
Ecotech	0.83	5.25	4.16	5.25	6.33	7.50	82.99 a	93.00 ab	34.75 a
Delfin	0.99	12.75	1.25	12.75	15.25	15.50	70.52 cd	95.25 ab	26.00 b
Protecto	0.81	12.63	6.31	12.63	15.78	15.78	63.32 a	89.25 b	22.50 c
Nemazal	0.83	6.25	2.50	6.25	7.50	9.75	77.88 b	30.50 e	28.25 b
Admeral	1.15	16.45	8.86	16.45	18.98	18.98	68.93 d	50.25 d	24.25 c
SAN + Nemazal	0.85	6.32	2.53	6.32	6.32	7.59	83.19	67.00 c	33.25 a
SAN + Admeral	0.81	5.81	2.81	5.81	7.04	8.26	80.80 a	71.25 c	31.50 ab
Control	1.14	36.61	16.90	36.61	50.70	60.56		109.25 a	17.50 e

*Passed on the % pest infestation after 15 days of the second spraying and spraying.

** Average number of the predators (*Coccinella* spp., *Scymnus* spp., *Orius* spp. and True spider) of the four after treatment observations/100 plant.

Study for Comparing the Pesticide Applications Based on the Dosage per Decar and Concentration per 100 Lt. of Water Against the Maize Pests

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Abstract

This investigation was carried out for comparing the applied by motorized mistblower and knapsack sprayer with the dosage per decare and the concentration in the 100 lt. water used against the maize pests in 1995 and 1996.

As a result of the study carried out in 1995, chemical control applications with motorized mistblower were not found successful. For that reason in 1996, the same study was carried out using only knapsack sprayer.

In 1995, in the applications of knapsack sprayer which were the dosage rate recommended to the per acre and the concentration of 100 lt. water were obtained the average effects %87,41 and %92,10 according to contamination ratio and also %90.86, %93.81 according to the number of alive larva respectively. They were classified in the same effect group in statistic analysis.

In the same application with knapsack sprayer in 1996 was obtained the average effects % 84 and % 89 respectively. They were classified in the same effect group in statistic analysis.

As a result of two years work was noticed the applications in maize using knapsack sprayer were more suitable than motorized mistblower. Both applications in the dosage rate recommended the acre and the concentration ratio of 100 lt. water were sufficiently effective.

However, quantity of pesticide applied in 3 th treatments in the application of the concentration ratio of 100 lt. water was less than that of the dosage rate recommended to the per acre. This is a very important advantage on economy and environmental pollution. Therefore it is suggested that the application of the concentration ratio of 100 lt. water is more suitable.

Materials and Methods

Materials are maize, insects of maize (*Ostrinia nubilalis* Hbn. - *Sesamia nonagrioides* Lef and *Heliothis* spp) triazophos formulation was used hostathion 40 EC (420 g/lt. a.i., AgrEvo Inc.), the knapsack sprayer, the motorized mistblower and the water sensitive paper.

In 1995, applications had 5 characters (2 applications + 2 apparatus + control) and 4 repetitions and in 1996 the applications had 3 characters (dosage + concentration + control) and 5 repetitions.

In 1995 description of two machines table 1. We have used the knapsack sprayer, in 1996.

In this project we have used the physical droplet particulermnt method (Deligönül 1984). Sampling is based on taking leaves from three-levels (bottom, middle and top) which is interior for the pest activity.

The water sensitive papers have been used in monitoring spray distribution (Oztürk and Yalcın 1991).

After 3 th treatment was down 15 days later maize were counted for a biological effect. 100 maize plants were randomly chosen from in the middle of parcel and their corn and stalk were check for damage rate. At the end of the process, statistical analyze was applied.

Table 1. Diagram of the production sprayer.

Sprayer	Knapsack sprayer	Motorized mistblower
Producer	Volkan	Taral
Type-Model	Mod-3	Taral Port 512
Name of desc.	M-1	M-2
Spray tank capacity	14 liters	12 Liters
Agitation	-	Pneumatic
Pump	Double piston	-
Pressure gauges	2...4 Atm	-
Nozzles	Hydraulic	Pneumatic
Wide- angle	80°	-
Nozzle	1 peace	1 peace
Capacity of nozzle	(1/min)	(1/min)
1 No or adj.	0.560	0.8
2 " " "	-	1.2
3 " " "	-	1.9
4 " " "	-	2.2
Airflow-volume	-	640 (m ³ /h)
Airflow-speed	-	95-100 (m/sec.) at outlet

Results and Discussions

Droplet deposits (cover rate), dosage and the indexes of leaf area in 1995 is shown in table 2, the values 1996 are shown in table 3, 4, 5, 6, 7 and 8.

Table 2. In the application for maize in Salihli in 1995 cover rate, dosage and maize canopy.

WSP	Knapsack sprayer 100 Lt.			Knapsack sprayer da			Motorized mistblower 100 Lt.			Motorized mistblower da		
	Treatment			Treatment			Treatment			Treatment		
	1	2	3	1	2	3	1	2	3	1	2	3
TUP												
TUN												
MUP												
MUN												
BUP												
BUN												
ST												
App. rate l/da	83	138	120	83	79	69	25	35	5	25	15	5
Leaf surf. Ind.	0.5	1.7	3.3	0.5	1.7	3.3	0.5	1.7	3.3	0.5	1.7	3.3

- Cover rate < % 1
 ▲ Cover rate % 1...25
 ▽ Cover rate % 26...50
 X Cover rate % 51...75
 ● Cover rate > % 75
- TUP: Top Upper Leaf Surface
 TUN: Top Under Leaf Surface,
 MUP: Middle Upper Leaf Surface,
 MUN: Middle Under Leaf Surface,
 BUP: Bottom Upper Leaf Surface,
 BUN: Bottom Under Leaf Surface
 ST: Soil Surface

Table 3. Cover rate at various applications within the maize canopy and details of the application (1996).

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	84		9		10		17	39
Cover rate (%)	100	3	1		10		36	10
Cover rate (%)	72		25		17			19
Cover rate (%)	39		33		33		10	19
Cover rate (%)	100		69		42		39	44
Cover rate (%)	79	3	27		22		26	26

Heights: 1.40 m Leaf surface index: 2.313

Table 4. Cover rates at the first application of concentration in 100 liters for maize (1996).

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	97		11		42		19	97
Cover rate (%)	83		44	5	25		31	85
Cover rate (%)	86		9		31		32	32
Cover rate (%)	69		38		27	25	14	46
Cover rate (%)	94		52		43		28	41
Cover rate (%)	86		31	18	34	25	25	60

Table 5. Cover rates at the second application of concentration in 100 liters for maize (1996).

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	53	1	34		39			47
Cover rate (%)	27	39	94		61		100	11
Cover rate (%)	25	33	50		64		42	42
Cover rate (%)	14		42	33	39		81	83
Cover rate (%)	24		100		61	9	44	58
Cover rate (%)	29	24	64	33	53	9	67	48

Heights: 2 m Leaf surface index: 3.94

Table 6. Cover rates at the second application of dosage in da for maize (1996).

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	5		64	20	38		31	76
Cover rate (%)	8	11	53	8	36		27	64
Cover rate (%)	27		31		25		29	16
Cover rate (%)	62			35	46		11	87
Cover rate (%)	18	4	45	4	52		27	54
Cover rate (%)	24	8	48	17	39		25	59

Table 7. Cover rates at the 3 th application of concentration in 100 liters for maize (1996).

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	23	42	33		4		6	42
Cover rate (%)	49	100	53		36		28	62
Cover rate (%)	18		44	54	29		46	59
Cover rate (%)	21	100	57		14	13	27	44
Cover rate (%)	14	31	25		11		19	16
Cover rate (%)	25	68	42	54	19	13	25	45

Heights: 2.6 m Leaf surface index: 4.94

Table 8. Cover rates at the 3 th application of dosage in da for maize (1996)

Repetition	TUP	TUN	MUP	MUN	BUP	BUN	ST1	ST2
Cover rate (%)	8		17				13	0.8
Cover rate (%)	9	11	20		7	10	11	13
Cover rate (%)	4		0.19		11		47	4
Cover rate (%)	3	26	8		2		9	31
Cover rate (%)	63		78		8	3	14	9
Cover rate (%)	17	19	25		7	7	19	12

Applications details are given in table 9.

Table 9. Details of the application in Manisa (Salihli-Tekelioğlu), 1996.

	The application of dosage for da		The application for 100 lt. concentration	
	Application rate (l/da)	a.i. (ml/da)	Application rate (l/da)	a.i. (ml/da)
1. application	54	200	51.5	128.5
2. application	104	200	74	185
3. application	62	200	68	170

In 1995,1996 the results of biological effect of spray treatments for maize are shown in tables 10 and 11.

Table 10. Effect of spray treatments for maize in Manisa -Salihli Çökelek 1995.

Characters	Repetitions	Damage rate (%)	The effect of spray treatments Accord. damage rate (%)	Living larva 100 plant	The effect of spray treatments to living larva (%)
Knapsack sprayer the application of a dosage for da	1	12	75.0	12	86.36
	2	4	90.9	4	93.75
	3	4	90.0	4	92.30
	4	4	93.75	4	96.0
Average			87.41 b	6	92.10 b
Knapsack sprayer the application of concentration for a 100 lt. water	1	4	91.66	4	95.45
	2	8	81.81	8	87.50
	3	4	90.0	4	92.30
	4	0	100.0	0	100.0
Average		4	90.86 b	4	93.81 b
Motorized mistblower the application of a dosage for da	1	0	100.0	0	100.0
	2	4	90.0	4	93.75
	3	24	40.0	32	38.46
	4	28	56.25	40	60.0
Average		14	71.78	19	73.05 ab
Motorized mistblower the application of concentration for a 100 lt. water	1	24	50.0	28	68.18
	2	24	45.45	32	50.0
	3	36	10.0	36	30.76
	4	20	68.75	28	72.0
Average		26	43.55 a	31	55.23 a
Control	1	48		88	
	2	44		64	
	3	40		52	
	4	64		100	

Table 11. Effect of spray treatments for maize in Manisa - Tekeliođlu (1996).

Characters	Repetitions	Damage rate (%)	The effect of sprayer treatments according to damage rate (%)
Knapsack-sprayer the application of a dosage for da	1	12	75.0
	2	6	83.3
	3	2	95.2
	4	8	85.4
	5	8	81.1
Average		7.2	84.0
Knapsack-sprayer The application of concentration for a 100 lt. water	1	8	83.3
	2	2	94.4
	3	5	88.1
	4	6	89.1
	5	4	90.1
Average		5	89.0
Control	1	48	
	2	36	
	3	42	
	4	55	
	5	44	
Average		45	

The results of both years were in the same level. There is only different is that the concentration ratio of 100 lt. water was 117 ml less than of the dosage rate per decar.

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Population Development of Egg Parasitoid *Platytenomus busseolae* (Gahan) (Hym.:Scelionidae) on the Eggs of *Sesamia nonagrioides* Lef. (Lep.:Noctuidae) in Adana/TURKEY

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Abstract

Population development and natural parasitization rate of *Platytenomus busseolae* (Gahan) on the eggs of *Sesamia nonagrioides* Lef. on second crop maize were determined in Adana/Turkey during 1994 and 1995. 63.3 and 58.0% of all eggs collected throughout the season were found to be parasitized with *P. busseolae* in 1994 and 1995 respectively. In both years parasitism was found to be low in August followed by an increase in September and October. *P. busseolae* were determined very promising biological control agent for the control of *S. nonagrioides*.

Introduction

The corn stalkborer, *Sesamia nonagrioides* is the most destructive corn pest in Mediterranean countries (Tsitsipis et al., 1987; Alexandri and Tsitsipis, 1990). The population density of *S. nonagrioides* start to increase during August, and causes economic damage in September and October on second crop maize in the Southeast Mediterranean region of Turkey (Sertkaya, 1993). A similar pattern of population development was observed in Greece (Tsitsipis et al., 1984). It has been reported that at least 2-3 spray applications are necessary to achieved reasonable protection (Tsitsipis, 1988).

Many parasites were detected as natural enemies of *S. nonagrioides*, including the egg parasitoid *Platytenomus busseolae*. This has been reported as the most effective natural enemy of *S. nonagrioides* in this region (Sertkaya, 1999). The efficiency of *P. busseolae* has been studied since 1990, and it is a very promising agent for biological control of *S. nonagrioides* (Kornoşor et al., 1992; 1994).

The aim of this study was to investigate the population development and natural parasitization rate of *P. busseolae* on eggs of *S. nonagrioides*.

Key words: *Sesamia nonagrioides*, *Platytenomus busseolae*, natural parasitism

Materials and Method

This study was conducted during 1994 and 1995 at the University of Çukurova, Faculty of Agriculture, in experimental fields of the Plant Protection Department in Adana. Second crop maize (approximately 5 da) was sown. at the beginning of June and July. Maize plants were sampled once a week. Fifty plants were examined at random in a transect across the field for egg parasitoids on each sampling date. Collected egg masses of *S. nonagrioides* were brought into the laboratory and each egg mass was counted and put into a separate glass vial. These were kept at 25±1 °C, 65±10 % relative humidity and 16 hours light daily. All egg masses were checked daily and parasitized and non -parasitized eggs were recorded. Emerged parasitoids were then distinguished according to their sexes.

Result and Discussion

The first parasitized eggs were seen on 15th July and parasitism continued until the beginning of November. The highest parasitism rate (92.8%) was recorded on 13th September in 1994 (Table 1). In 1995, the first parasitized eggs were seen on 18th July and parasitism continued until mid-October. The highest parasitism rate (79.1%) was recorded on 14th September (Table 2).

Table 1. Natural parasitization rate of *Platytenomus busseolae* on the eggs of *Sesamia nonagrioides* in Adana, 1994.

Date	Number of Eggs	Number of Parasitized Eggs	Parasitism (%)
01.07.1994	0	0	0.00
08.07.1994	122	0	0.00
15.07.1994	152	113	74.34
21.07.1994	161	143	88.81
09.08.1994	198	153	77.27
15.08.1994	262	28	10.68
19.08.1994	226	99	43.80
23.08.1994	512	358	69.92
30.08.1994	274	194	70.80
06.09.1994	463	411	88.76
13.09.1994	418	388	92.82
19.09.1994	896	672	75.00
27.09.1994	2761	960	34.77
04.10.1994	4564	2693	59.00
11.10.1994	2483	1944	78.29
19.10.1994	1224	959	78.34
26.10.1994	1113	841	75.56
02.11.1994	266	197	74.06
09.11.1994	270	218	80.74
Total	16365	10371	63.37

The data showed that population densities of *S. nonagrioides* and *P. busseolae* increased at the end of August, and 63.3 and 58.0 % of all eggs collected throughout the season were found to be parasitized with *P. busseolae* in 1994 and 1995 respectively. In both years, parasitism was found to be low in August followed by an increase in September and October (Figure 1).

Alexandri & Tsitsipis (1990) *S. nonagrioides* eggs parasitized by *P. busseolae* for the first time in Europe, and reported that populations of *P. busseolae* on the eggs of *S. nonagrioides* increased at the end of August and during September. It is concluded that low parasitism in August could be due to high temperatures at that time. Chabiolaye et al. (1997), reported that *T. busseolae* could not parasitize *S. calamistis* at 34 °C under laboratory conditions. The parasitism efficiency of *S. nonagrioides* by *P. busseolae* found in the present study confirms data reported by other researchers (Moutia and Courtois, 1952, Hafez et al., 1979, Scheibeireiter, 1980, Baniabbasi, 1981).

Table2. Natural parasitization rate of *Platytenomus busseolae* on the eggs of *Sesamia nonagrioides* in Adana, 1995

Date	Number of Eggs	Number of Parasitized Eggs	Parasitism (%)
05.07.1995	0	0	0.00
12.07.1995	66	0	0.00
18.07.1995	75	35	46.66
25.07.1995	142	54	38.02
01.08.1995	156	67	42.94
08.08.1995	184	57	30.97
13.08.1995	312	97	31.08
18.08.1995	391	147	37.59
25.08.1995	682	259	37.97
31.08.1995	1256	538	42.83
08.09.1995	870	646	74.25
14.09.1995	2240	1772	79.10
22.09.1995	1397	671	48.03
30.09.1995	2237	1348	60.25
07.10.1995	1469	956	65.07
13.10.1995	254	159	62.59
Total	11731	6806	58.01

The sex ratio ($\sigma^{\circ}:\text{♀}$) of *P. busseolae* observed in this study were 1:1.2 and 1:1.3 in 1994 and 1995 respectively. A sex ratio for *P. busseolae* of 1:1.5 has been previously reported by Hafez et al. 1977; and Alexandri & Tsitsipis 1990. Our results confirm these data.

During this study, egg parasitism of *S. nonagrioides* by *Trichogramma evanescens* was also detected but at very low levels. In conclusion, the population densities of *P. busseolea* increased at the end of season with the increase of its host *S. nonagrioides* and gave high rates of natural egg parasitism. This indicates a very promising biological control agent for the control of *S. nonagrioides*.

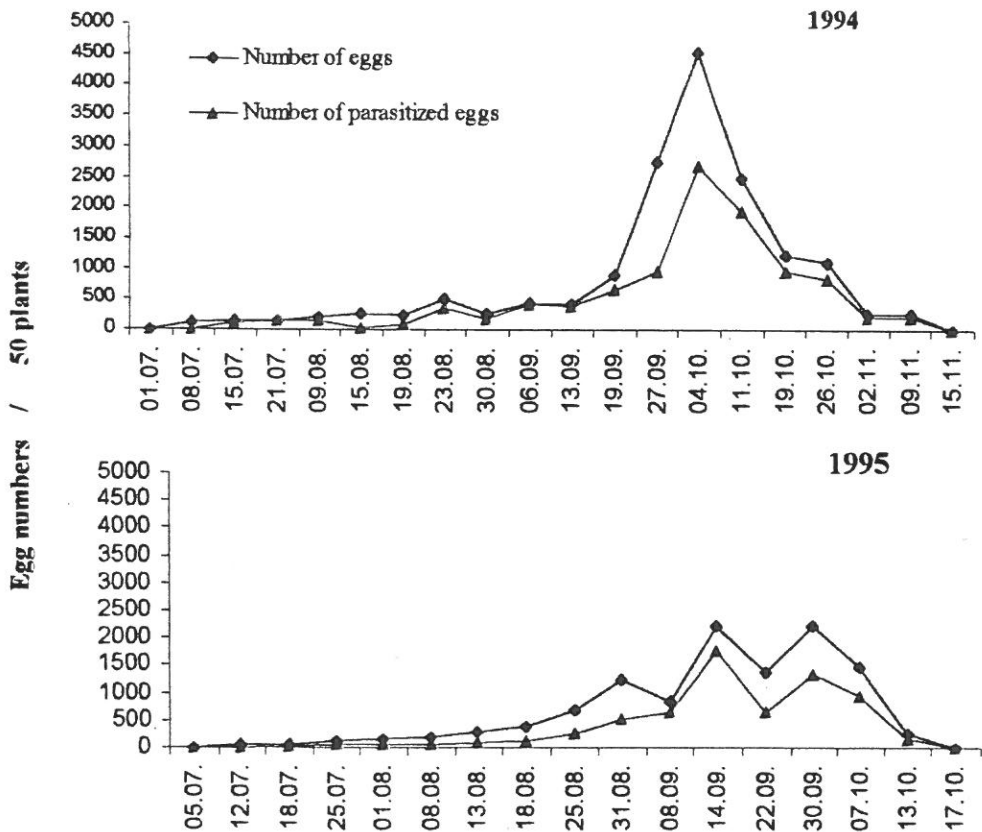


Figure 1. Population changes of *Platytelenomus busseolae* on the eggs of *Sesamia nonagrioides* in Adana in 1994 and 1995.

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Biological Features of *Trichogramma evanescens* Westwood (Hymenoptera, Trichogrammatidae) on the Eggs of *Sesamia nonagrioides* Lefebvre (Lepidoptera, Noctuidae)

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Abstract

Sesamia nonagrioides and *Ostrinia nubilalis* are the most destructive pests threatening the agricultural production of second crop maize in Çukurova Region. *Trichogramma evanescens* was found to be most effective natural enemy of *O. nubilalis* and also was able to parasitize *S. nonagrioides* eggs at low rate in the nature. The aim of present study was to evaluate effectiveness and biological features of *T. evanescens* on *S. nonagrioides* eggs when it was used in inundative releases to control *O. nubilalis*.

Effect of temperature on biological properties of the egg parasitoid *T. evanescens* such as longevity, number of parasitized eggs and life time fecundity, emergence rate, sex ratio, number of emerged parasitoid per parasitized egg on the eggs of *S. nonagrioides* were determined in climatic chambers at respective constant temperatures $20\pm 1^{\circ}\text{C}$, $25\pm 1^{\circ}\text{C}$ and $30\pm 1^{\circ}\text{C}$, $60\pm 10\%$ relative humidity and 16 h photoperiod.

Developmental time and adult longevity of the parasitoid were negatively correlated with increasing of temperature. Number of parasitized eggs and total fecundity were found to be greatest at 25°C .

Much efforts must be devoted to control of *S. nonagrioides* by inundative releases of *T. evanescens* in the nature.

Key words: *Sesamia nonagrioides*, *T. evanescens*, egg parasitoid Introduction

Introduction

The Corn stalk borer, *Sesamia nonagrioides*, is one of most significant pest in Mediterranean countries. Eggs are deposited in leaf sheaths and after hatching the larva into stalks causing considerable economic losses mainly on second crop maize and damaging population levels are reached between August and October in south-east Mediterranean region of Turkey (Sertkaya, 1999) and in Greece (Tsitipis, 1988; Tsitipis and Alexandri, 1989). Also, recent studies have shown that the pest has two important egg parasitoids, *Platytenomus busseolea* and *Trichogramma evanescens*, in the region and Greece (Kornoşor et. al., 1994; Alexandri, 1993; Sertkaya, 1999) though the former has not been reared on artificial hosts which would be needed to keep the pest below economical injury threshold.

The objective of this study was to investigate the effect of temperatures on biological features of *T. evanescens* indigenous to southeastern Mediterranean region of Turkey to obtain basic information for the biological control of *S. nonagrioides*.

Materials and Methods

The egg parasitoid, *T. evanescens* was obtained from parasitized *Sesamia nonagrioides* eggs on second crop maize fields from Research and Implementation Fields of Plant Protection Department on the Faculty of Agriculture Farm of Çukurova University in 1997. The parasitoid colony was maintained on eggs of the corn borer eggs at 25 ± 1 °C, 16:8 L:D, %60-70 RH. *T. evanescens* was reared in glass tubes (2x20 cm) plugged with cotton where about 500-600 one day old eggs of *S. nonagrioides* were glued to 1.5x10 cm paper cards on both sides of which one honey drop scattered.

S. nonagrioides used in the study was also collected from the same place and reared on fresh maize stalks and ears at 25 ± 1 °C, 16:8 L:D regime, 60-70 % RH. New hatched adults were placed into plastic mating cages with 15-20 cm tall maize plants to lay their eggs on. Laid fresh *S. nonagrioides* eggs were removed from the plants by careful dissection and then the plants were replaced with new ones twice a day.

The effect of temperature on the biological parameters of *T. evanescens* was studied at 20, 25 and 30 ± 1 °C. Experiments were conducted in 2x20 cm glass tubes in which 75 of 0-24 h old *S. nonagrioides* eggs glued to cards (1x5 cm). These were exposed daily to less than 1-day old mated female parasitoid that reared on *S. nonagrioides* eggs. Less than 1 day old eggs were provided every 24 h. Number of alive *T. evanescens* females were recorded until death by checking 3 times a day. Eggs provided to the parasitoids were labelled and held at the tested temperature until parasitoid emergence.

Parasitized eggs were inspected at the same time each day by a stereomicroscope in order to indicate parasitism, parasite emergence, sex of emerged parasites. The number of parasites developing per host egg was also recorded. Parasitism was confirmed by blackening of the vitelline membrane of the host egg as described by Flanders (1937). After counting and distinguishing of sexes of emerged offsprings of *T. evanescens*, total parasitized eggs were calculated and dissected in order to investigate daily and total emergence rate. Each temperature had 20 replicates. One way analysis of variance was conducted on all data and LSD' test was used to separate the means. Percentages were obtained from emergence and sex ratio transformed to $\arcsin \sqrt{x}$ before analysis.

Results and Discussions

No significant differences was recorded in longevities of the parasitoid between 25 °C and 30 °C that was shorter than the longevity was recorded at 20 °C. Significant difference in developmental times was observed among temperatures and with average values ranging from 18.50 ± 0.05 days at 20 °C, to 7.99 ± 0.02 at 30 °C. Longevity of oviposition period of *T. evanescens* was highest at 20 °C while there were no significant differences in longevities of oviposition period of parasitoid between at 25 and 30 °C (Table 1).

The number of parasitized eggs and offsprings were greatest at 25 °C and fewest at 30 °C; the significant differences were recorded only temperatures of 25 °C and 30°C (Table 1).

Oviposition time at 20 °C was longer than those were recorded at 25 °C and 30°C. These results are in contrast with those reported by Özpınar (1994) who found shorter longevity of ovipositing *T. evanescens* at 25 °C on natural host *O. nubilalis* and factitious host *E. kuehniella* eggs.

Table 1. Biological properties of *Trichogramma evanescens* on *Sesamia nonagrioides*

Biological Features	Temperatures (°C)		
	20	25	30
Longevity (day)	15.45 ± 1.27 A	10.45 ± 0.63 B	8.90 ± 0.48 B
Developmental Time (day)	18.50 ± 0.05 A	10.34 ± 0.02 B	7.99 ± 0.02 C
Oviposition (day)	12.65 ± 1.04 A	8.80 ± 0.73 B	7.40 ± 0.46 B
Number of parasitized eggs	67.50 ± 9.48 AB	85.80 ± 6.48 A	49.50 ± 5.39 B
Number of offsprings	98.00 ± 14.90 AB	126.35 ± 8.66 A	74.75 ± 6.94 B
Emergence (%)	83.7 ± 1.01 B	86.2 ± 0.43 A	84.8 ± 1.06 AB
Sex ratio (%)	77.4 ± 1.25 AB	81.7 ± 0.55 A	73.8 ± 2.37 B
No. Parasites emerged/host egg	1.62 ± 0.10 A	1.58 ± 0.06 A	1.70 ± 0.06 A

Means within a row followed by the same letter are not significantly different at $p \leq 0.05$ (LSD test)

Results of previous studies on parasitism capacity of *Trichogramma* spp. vary widely. Reasons for this may include differences in species, hosts, rearing conditions and techniques, strains, keeping duration of the parasitoid under laboratory conditions, number of generation rearing on factitious host and other factors (Noldus, 1989).

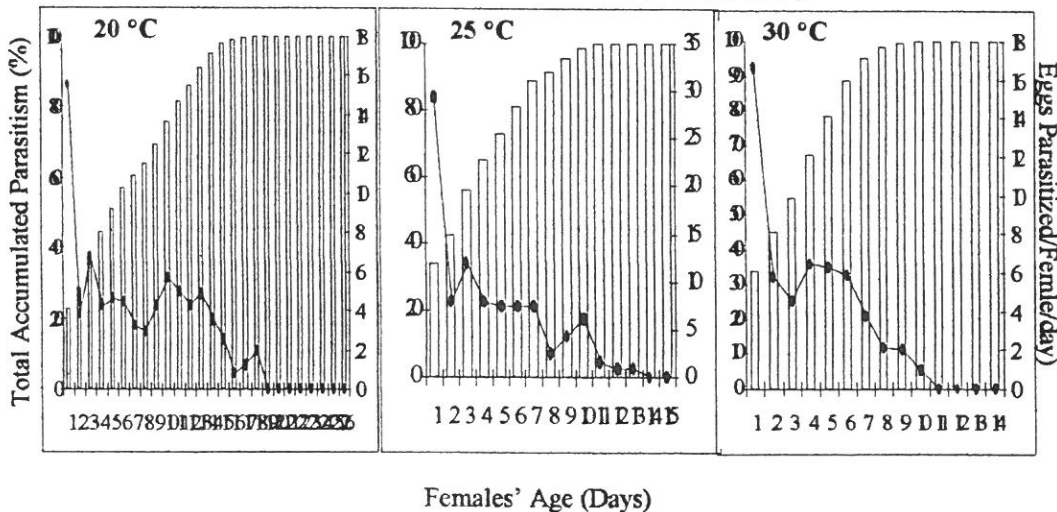


Figure 1. Accumulated parasitism (percentage) and average daily parasitism (eggs parasitized per female) of *T. evanescens* on the eggs of *S. nonagrioides* at different temperatures

Parasitism rates were highest on the first day after emergence, ranging from 25.7 % at 20 °C, to 42.2 % at 30°C. On the second day, the mean number of parasitized eggs per female declined greatly at all tested temperatures. In the following days, the number of parasitized eggs tend to decrease to zero until all females died.

The parasitism percentages achieved in the first week were 60.2 % (20 °C), 89.3 % (25 °C) and 95.1 % (30 °C) (Figure 1). The results in trends of parasitism were similar to those obtained by Kayapınar and Kornoşor (1993) for the same parasitoid on the eggs of *E. kuehniella*.

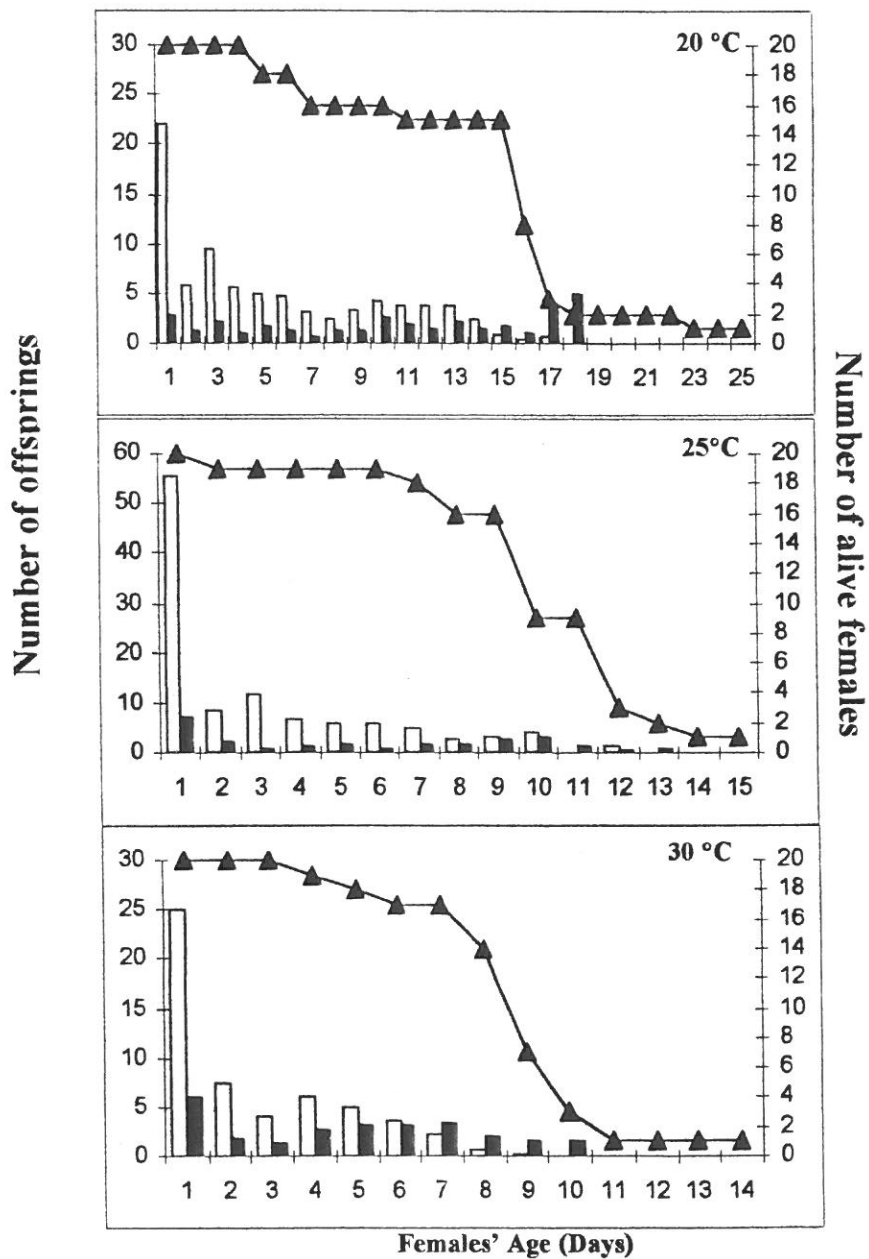


Figure 2. Mean daily production of progeny produced by *T. evanescens* and number of alive females at different temperatures

Parasitoids began laying eggs immediately after emerging. Percentages of offspring produced in the first day compare to total production in their whole life were 25.6 % at 20 °C, 60.1 % at 25 °C and 41.6 % at 30 °C and reached to 96.3 % (20°C), 90.1 % (25°C) and 97.8 % (30 °C) at the end of first week of their life (Fig. 2.). Conversely, percentages of female progeny produced was higher during first day, from 29.9 % (20°C), 53.4% (25°C) and 47.7 % (30°C), than on any successive day of their life. Number of female offsprings was decreased with age ovipositing females (Fig. 2.).

Emergence rates of *T. evanescens* at 25 °C and 30 °C were the same but considerably higher than emergence rate recorded at 20 °C (Table 1.). Various results on the emergence rate of different *Trichogramma* spp. obtained by other authors, [e.g. Kayapınar and Kornoşor (1993)] reported that emergence of the same species on *E. kuehniella* eggs was about 88 % at 25 °C. Whereas, Babi (1994) found emergence rate of both *T. cacoeciae* and *T. daumalae* on laboratory host decreased as temperatures increases. However, some authors reported that emergence rates of parasitized eggs by *Trichogramma* spp. were higher at high temperatures.

The ratio of female progeny was highest at 25 °C and lowest at 30 °C; the significant differences were recorded only temperatures of 25 and 30 °C. Means of emerged parasites per parasitized *S. nonagrioides* eggs did not differ at the three temperatures (Table 1.).

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Evaluation of The Effectiveness of the New *Agriotes* Sex Pheromone Traps in Different European Countries

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Abstract

New sex pheromone traps for monitoring the most important European *Agriotes* species (*Agriotes litigiosus*, *Agriotes ustulatus*, *Agriotes lineatus*, *Agriotes obscurus*, *Agriotes brevis*, *Agriotes sordidus*, *Agriotes rufipalpis*) have been discovered and set up in studies carried out mainly in Hungary and Italy in last four years. At present in order to implement an IPM against *Agriotes* species in Europe using the new traps we need:

- to ascertain if the traps are really effective in monitoring different *Agriotes* populations of the same species in different european countries; (we know that the sex pheromone composition of female glands may vary according to the population);
- to establish if there are conspicuous differences in the life cycle between populations of the same species;
- to estimate the correlation between the amount of adults captured by the sex pheromone traps and the subsequent larval population in different areas in different years.

In order to meet these goals an informal specific European working group was established in 1998. It involves the following researchers, who are co-authors of the work, and Institutes: S. Arnone and P. Nobili, Enea Casaccia, Italy; P. Bassetti, Stazione Federale di Ricerche Agronomiche, Cadenazzo, Switzerland; H.K. Berger, P. Cate, H. Klapal, Federal Office and Research Center of Agriculture, Wien, Austria; G. Bourlot and A. Turchi, Plant Protection Service of Piemonte, Torino, Italy; R. Ferrari and L. Boriani, Centro Agricoltura Ambiente, Crevalcore, Italy; L. Furlan, Intitute of Agricultural Entomology, Padova, Italy; M. Ivezic and M. Mlinarevic, University of J.J. Strossmayer, Faculty of Agriculture, Osijek, Croatia; W. Jossi, Station Federale de Recherches en Agroecologie et Agriculture, Zurich, Switzerland; T. Keresi, Faculty of Agriculture, Institute for Plant Protection, Novi Sad, Yugoslavia; F. Muresan, Agricultural Research Station Turda, Turda, Romania; M. Rostàs, Freie Universitat Berlin, Institute fur Zoologie, Berlin, Germany; I. Szarukan, University of Debrecen, Hungary; M. Toth, Plant Protection Institute of Budapest, Hungary; J.A. Tsitsipis, University of Thessaly, Volos, Greece; A. Xavier, Ministerio da Agricultura, Direccao regional de Entre Douro e Minho, Porto, Portugal.

All the participants have followed a common protocol including soil sampling carried out by using attractive traps for larvae and the collection of soil cores in the area where the traps have been placed. Lures and traps produced by the Plant Protection Institute of Budapest have been used. All the specimens captured have been identified by Dr Furlan, Dr Giuseppe Platia and Dr P. Cate.

Very promising preliminary results have been obtained in the first two years: several traps proved to be effective in different countries.

Key words: sex pheromones, *Agriotes*, Europe, I.P.M.

Introduction

Soil insecticides are a very important group of pesticides, often making up the majority of insecticides applied in several cultivated areas (Furlan, 1989). However, extensive studies carried out in north-eastern Italy have demonstrated that less than 5% of the fields planted with corn need to be treated with soil insecticides (Furlan, 1989, Furlan *et al.*, 1992). Despite this fact at present most of farmers use soil insecticides when planting mainly with the aim of controlling wireworms. Therefore effective and inexpensive methods are needed to identify the soils that have high wireworm populations in order to treat these only, or to avoid cultivating maize in the same soils. *Agriotes* sex pheromone traps might effectively help us to attain this goal since they can easily give us useful information without making expensive and laborious surveys of the soil layers when searching for larvae. In fact they can monitor the only stage which lives outside the soil: the adult. Experimentation carried out mainly in Italy have shown that sex pheromone traps can indeed be useful for implementing Integrated Pest Management (Furlan *et al.*, 1996a, Furlan *et al.*, 1996b, Furlan *et al.*, 1997). At the moment we need to know if this can apply to the different European countries. In order to evaluate if it is possible to implement easy and low cost IPM strategies against *Agriotes* larvae all over Europe and therefore to reduce the use of soil insecticides in Europe we need:

- to ascertain whether the traps are really effective in monitoring different *Agriotes* populations of the same species in different European countries;
- to identify the key species in the different areas;
- to ascertain if there are conspicuous differences in the life cycle between populations of the same species;
- to estimate the correlation between the number of adults captured by the sex pheromone traps and the subsequent larval population in different crops in different areas in different years.

In order to meet these aims during and just after the XIX IWGO Conference a group of researchers, who must be considered co-authors of this paper, from different European countries in 1997 agreed to start working according a common protocol:

L. Furlan, Institute of Agricultural Entomology, Padova University,

M. Toth, Plant Protection Institute, Hungarian Academy of Sciences, Budapest

S. Arnone and P. Nobili, Enea Casaccia, Italy;

P. Bassetti, Stazione Federale di Ricerche Agronomiche, Cadenazzo, Switzerland;

H.K. Berger, P. Cate, Klupal, Federal Office and Research Center of Agriculture, Wien, Austria;

G. Bourlot and A. Turchi, Plant Protection Service of Piemonte, Torino, Italy;

R. Ferrari and L. Boriani, Centro Agricoltura Ambiente, Crevalcore, Italy;

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W. Jossi, Station Federale de Recherches en Agroecologie et Agriculture, Zurich, Switzerland; for Plant Protection, Novi Sad, Yugoslavia; F. Muresan, Agricultural Research Station Turda, Turda, Romania;

M. Rost...s, Freie Universitat Berlin, Institute fur Zoologie, Berlin, Germany; I. Szarukan, University of Debrecen, Hungary; M. Toth, Plant Protection Institute of Budapest, Hungary; J.A. Tsitsipis, University of Thessaly, Volos, Greece;

A. Xavier, Ministerio da Agricultura, Direccao regional de Entre Douro e Minho, Porto, Portugal.

Materials and Methods

PHEROMONE CAPS set up after specific studies by the Plant Protection Institute of Budapest and the Institute of Agricultural Entomology of Padova for the following species:

<i>Agriotes brevis</i> Candeze	geranyl-butyrate and E,E-farnesyl butyrate;
<i>Agriotes sordidus</i> Illiger	geranyl-hexanoate
<i>Agriotes sputator</i> L.	geranyl-butyrate
<i>Agriotes rufipalpis</i> Brullè	geranyl-hexanoate
<i>Agriotes lineatus</i> L.	geranyl octanoate + geranyl butyrate
<i>Agriotes litigiosus</i> Rossi	geranyl-isovalerate
<i>Agriotes ustulatus</i> Schaller	E,E - farnesyl acetate

Traps: VARb: funnel trap; on top of it larger funnel made from plastic sheet .

BOTTLE: "funnel" traps made from plastic water bottles, diameter 8 cm (effectiveness like VARb).

SOIL-LEVEL: model suitable for species which prefer crawling

Sex Pheromone Trap Management: at least 2 traps per species per each locality were set up according the following indications:

HEIGHT of the traps: 40 - 60 cm above the ground;

POSITION in the field: in the middle of the field area sampled for larvae;

DISTANCE BETWEEN THE TRAPS: at least 50 meters

PERIOD OF MONITORING: May - September for *A. ustulatus* and *A. litigiosus*, March - August for the other species.

REPLACEMENT OF THE CAPS: every 30 days.

INSPECTIONS: twice per week. All the specimens were removed at every inspection and identified.

IDENTIFICATION: species and sex identification was carried out by:

Dr Lorenzo Furlan, Institute of Agricultural Entomology, Padova, ITALY

Dr Giuseppe Platia, Italian expert, ITALY

Dr Peter Cate, Federal Office and Research Institute for Agriculture, Vienna, AUSTRIA

Larval Population Evaluation

1) Soil sampling: 20-30 soil samples (diameter 12 cm and 25 to 60 cm deep according to the season) were collected per field (0,3 to 1 hectare) at random, using a manual soil sampler. The soil cores were then put into funnels, 26 cm in diameter, provided with a 0,5 cm mesh and a vial at the bottom according to the Tullgren method. The soil was allowed to dry for at least 30 days in a sheltered place and the larvae which fell into the vials placed at the bottom of the funnels were counted and identified.

2) Attractive traps: attractive traps were placed 3 m from each point where the soil cores had been taken. Each trap was made and used according to the description given by CHABERT and BLOT (1992). They were manually observed after 15 days and the larvae found were counted and identified.

First Results

Sex pheromone traps appear a sensitive tool for the identification of the key species and for evaluating population levels in the different European countries as shown in Table 1 which refers to 1998 and 1999 for all countries with the exception of Croatia and Yugoslavia for which only data collected in 1998 are available at the moment. Geranyl isovalerate proved to be extremely effective in capturing the different varieties of the species *A. litigiosus* Rossi. In fact high numbers, sometimes more than one thousand per trap in May — August, were captured both where the red variety (fen. typ.) is prevalent and where the dark variety (laichartingi) is predominant. In the regions where no specimens were caught no larvae of this species had been captured. The same considerations apply to E,E farnesyl acetate which proved to be extremely effective in catching both the dark and the brown forms of *Agriotes ustulatus* Schaller (in this case however the 2 forms of the males are present together in the same locality). On the other hand, the other compounds were seen to have a good effect on more than one species. Geranyl hexanoate is effective in capturing males belonging to 3 species: *Agriotes sordidus* Illiger in Italy, *Agriotes gallicus* Lacordaire in Northern Switzerland, *Agriotes rufipalpis* Brullè in the other countries. The compound did not capture any *Agriotes* specimens in Ticino but no larvae of the above cited species had been found by using soil sampling and bait traps in the last four years. Geranyl butanoate appears to be the suitable sex pheromone for *Agriotes sputator* L. but in Portugal it is effective in monitoring the populations of *Agriotes proximus* L.. Trials carried out in Switzerland and Italy in recent years (Toth et al., 1998; Toth et al., 1999) have demonstrated that a reliable monitoring of *Agriotes lineatus* L. can be carried out by exploiting the synergistic effects of geranyl butyrate and geranyl octanoate. Two compounds are also necessary to set up effective traps for *Agriotes brevis* Candeze (geranyl butyrate and E,E farnesyl butyrate). Anyway these association of compounds is effective for capturing *Agriotes proximus* Schwarz too and in a few regions can also capture *Agriotes sputator* L. and *Agriotes acuminatus* Stephens.

Provided that the traps can be considered effective in the different European countries data about the practical implications of their use are not enough. As far as this aspect is concerned this data must be considered preliminary. A considerable amount of data obtained in different years and in different agronomic conditions would allow us to interpret the records of adults from a practical point of view.

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Recent Developments in Wireworm Management in the U.K.

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Abstract

The importance of wireworms (*Agriotes* spp.) in U.K. agriculture and horticulture has varied considerably over the last 50 years. Their importance was at a peak during the Second World War but declined from the mid-1950s as the use of persistent organochlorine insecticides became more widespread. However, the last five years has seen a resurgence in the importance of wireworms. A particular feature of these recent wireworm problems has been an apparent increase in infestations in all-arable (no grass) rotations. This may in part be due to long-term (five-year) set-aside providing a suitable habitat for wireworm survival. Recent research work has concentrated on improving risk assessment methodologies. This has resulted in the development of a bait trapping system that is now in widespread commercial use. Work has also been done to develop and evaluate more sophisticated multi-variate models based on field characteristics that could be used to predict the presence and level of wireworm infestation in individual grass fields. A limited amount of work has also been done on evaluating insecticides for wireworm control and evaluating the susceptibility of different potato cultivars to wireworm damage. Current work is concentrating on improving risk assessment methodologies by investigating adult monitoring systems, and investigating the interaction between variety, insecticide use and harvest date on wireworm control in potato. Work is also required on risk assessment and control thresholds in cereals. With increasing interest in organic production in the U.K., acceptable non-chemical means of wireworm management also need to be developed.

Key words: Wireworm, *Agriotes*, management, control.

Introduction

In the UK, much of the current knowledge on wireworms is derived from work done during the 1940s. This was prompted by the conversion of large areas of grassland to arable production during the Second World War. The work involved extensive surveys of wireworm incidence and research on sampling, biology and control (Miles, 1942, Anon., 1948). The nationwide effort towards wireworm control in the 1940s resulted in control strategies based on protective seed treatment (e.g. γ -HCH on cereals) and broadcast applications of persistent insecticides on other crops, coupled with limited risk assessment by soil sampling. These basic wireworm control strategies remained largely unchanged from the mid-1950s to the late 1980s, although work on organophosphorous (OP), carbamate and pyrethroid insecticides for wireworm control continued sporadically through the 1960s and 1970s (Caldicoff & Isherwood, 1967; Griffiths, 1977; Griffiths & Bardner, 1964, Oriffiths et al., 1967). However, the increasing trend towards all-arable rotations reduced the status of wireworms to a sporadic, but locally important problem, principally in the mixed arable and livestock farming areas in western and northern England and Wales (Strickland et al., 1962). In the late 1980s, the impending withdrawal of aldrin for wireworm control on potato resulted in a renewed interest in wireworm risk assessment and control. During the 1990s, the upward trend in the importance of wireworms has continued, principally for potato growers, but very recently (1999) the withdrawal of γ -HCH seed

treatments on cereals (wheat and barley) has highlighted potential shortcomings in wireworm management for other crops. The upsurge in interest in organic farming also raises new questions about wireworm management techniques.

This paper briefly reviews the developments in wireworm research and practice in the U.K. in the last 10 years, and suggests priorities for future work.

Wireworm species

One of the features of the increased wireworm problems in the U.K. in the last five years has been the occurrence of unexpectedly high infestations in all-arable rotations (i.e. in fields with no recent history of long-term grass in the rotation). Initial investigations have concentrated on identifying the species involved, and characterising the situations in which these so-called 'arable wireworm' problems occur.

On the limited data available so far, it is likely that the main species involved are those normally associated with crop damage in the U.K., principally *Agriotes lineatus*, *A. obscurus* and *A. sputator*. There is no evidence of an increase in the importance of other wireworm genera (e.g. *Athous*, *Corymbites*, *Adrastus*, *Brachylacon*, *Cryptohypnus*, *Dotopius*, *Hypnoidus*, *Selatosomus*, *Prosternon*,. (Erichsen Jones, 1944; Cooper, 1945, 1947; Van Emden, 1945). These are sometimes found in both grass and arable fields, often in mixed populations with *Agriotes* species, though rarely exceeding 10% of the total population.

The situations in which 'arable wireworm' occurs are very varied. However, a small survey conducted in 1998 (Parker, unpublished data) suggested that fields with a history of at least two years of set-aside (unmanaged fallow) were at particular risk from this type of infestation. Experimental work in the U.K. has demonstrated that set-aside can support wireworm populations (Hancock et al., 1992), and advisory information from North America has also indicated that grain crops grown after set-aside are considered to be at greater risk from wireworms. Clearly, the undisturbed conditions afforded by set-aside could favour the build-up of wireworm populations. Further work is required to substantiate this.

Chemical control

Recent work on the chemical control of wireworms has focused on the potato crop, principally to identify alternatives to the persistent organochlorine insecticide aldrin. The use of aldrin was approved for wireworm control on potato until 1989 and was regarded as the first choice product, even though phorate was also Approved for this use. Hancock et al. (1986) found that although some products, including organophosphates (phorate, chlorpyrifos, ethoprophos, fonofos, carbofuran) and carbamates (bendiocarb, befuracarb and carbofuran), significantly reduced the incidence of wireworm damage on potatoes, the level of control achieved was inconsistent and overall rarely as effective as aldrin. Further work compared aldicarb, carbofuran, ethoprophos, fonofos and thiofanox with aldrin. The results suggested that ethoprophos and, to a lesser extent, carbofuran were potential replacements for aldrin, although again the level of control achieved with all products (including aldrin) was variable (Parker et al., 1990).

On cereals, wireworms have not generally been a major problem, and where control was required, soil applications and seed treatments of lindane (γ -HCH) provided effective control. However, very recently (1999) the use of lindane seed treatments has been banned and the use of spray treatments curtailed. Alternative insecticidal seed treatments containing imidacloprid or tefluthrin are available and are known to give some control of

wireworms, although the range of wireworm population densities over which these products will provide cost-effective control is not known.

On forage maize, bendiocarb seed treatment or lindane applied pre-drilling are the only recommended treatments.

Risk Assessment Techniques

The majority of recent research work on wireworms in the U.K. has concentrated on improving methodologies for identifying the incidence and level of wireworm infestation in individual fields. For most of the last 50 years, risk assessment has been done using labour-intensive soil sampling techniques developed during the 1940s, coupled with laboratory extraction of invertebrates using flotation (Salt and Hollick, 1944; Cockbill et al., 1945). However, in the early 1990s, it was recognised that improved methods were required which ideally required less labour and a greater chance of detecting low wireworm populations. Work was therefore done to evaluate bait trapping systems, based on the extensive work done in North America (e.g. Ward & Keaster, 1977; Doane, 1981; Kirfman et al., 1986; Bynum & Archer, 1987) and to a lesser extent in Europe (Chabert & Blot, 1992). Initial work identified that cereal baits were the most effective, were potentially less labour-intensive and more sensitive than soil sampling, and could be incorporated into a simple trapping system (Parker, 1994). This system was further refined and tested (Parker, 1996), and with industry support was extensively field tested throughout the U.K. (Parker et al., 1994). A modified version of the original trapping system is now commercially available and is widely used, particularly by the potato industry.

Despite the success of the bait trapping system, it does have some limitations. Current indications (e.g. Parker, 1996) are that the traps are best used as presence/absence indicators only, and cannot be used to estimate the likely damage level, particularly to high-value crops such as potatoes. In addition, a negative trap result does not necessarily mean that a field is wireworm-free. Additional information is therefore required to make a full assessment of wireworm risk. Efforts have been made to use multiple site characteristics to predict the incidence and level of wireworm infestation in individual fields (Parker & Seeney, 1997), but these have only been partially successful. Current work is now concentrating on the possible role of monitoring adult populations in aiding the risk assessment process, particularly for those fields whose infestation status is uncertain.

Current work

The largest current research programme on wireworms in the U.K. is being funded by the potato industry, and is therefore biased towards the needs of that particular commodity. Work includes assessment of potato varietal susceptibility to wireworm damage, assessing the interactions between insecticide use, harvest and variety, and assessing the role of adult monitoring systems in field risk assessment. The latter area clearly has application to wireworm risk assessment for other crops. It is hoped that this will have a spin-off into the cereal sector, where it is possible that work may be funded soon re-examining long-established thresholds for insecticide treatment, as well as assessing the efficacy of newly-introduced seed treatments.

Future priorities

Improvement of risk assessment methodologies remains the most pressing research need as this applies to all crop commodities susceptible to wireworm attack. Work on adult

monitoring in particular should be of great value. Risk assessment will become increasingly important if the interest in organic production continues to grow. An accurate assessment of the likely risk of wireworm incidence in organic systems in general, and on individual crops in particular, will be critical to the effective organic management of wireworms. A better understanding of the biology and dispersal of adult beetles will also help to assess the possible role of more novel approaches to managing wireworm populations at a number of different points in the crop rotation. In the U.K., changes in agronomic practice that may be affecting wireworm populations, particularly the impact of set-aside, also require further investigation.

Acknowledgements

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Monitoring Wireworm Populations in Greece by Sex Pheromone and Soil Bait Traps

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The seasonal abundance of wireworm species (Coleoptera: Elateridae), important pests of many crops, was examined in Central Greece. Sex pheromone traps, soil bait traps and soil sampling were used. The experimental field was located in Velestino, Magnesia (Central Greece) and monitoring covered the period from late March until late October. Ten pheromone traps were set up in an area of about 1-hectare (200x50m). The pheromone dispensers were replaced every month. The traps were placed inside wheat, maize and sugar beet crops. Pheromones of the following *Agriotes* species were used: *A. lineatus* L., *A. rufipalpis* Brullé, *A. litigosus* Rossi, *A. sputator* L. and *A. ustulatus* Schaller. Two traps were set up per pheromone species distributed randomly. Soil sampling was performed in April and May and soil bait traps were placed in the ground to monitor the larval stages. All captured individuals were males. The traps for *A. ustulatus* did not capture any individuals of this species, but they caught two individuals belonging to *A. rufipalpis* and *Hemicrepidius hirtus* Hbst. The traps corresponding to *A. sputator* captured ten individuals for the whole monitoring period: one *Cardiophorus crassicollis* Hbst., one *A. paludum* K., two *A. rufipalpis* and six *H. hirtus*. The traps for *A. lineatus* captured 12 individuals from late April to mid July, showing no particular population peaks: five *A. lineatus*, four *A. paludum*, two *A. rufipalpis* and one *H. hirtus*. The traps for *A. litigosus* captured a total of 566 males from mid May to mid July, 556 of which were *A. litigosus*. One *A. rufipalpis* was also caught along with nine *H. hirtus*. Finally, the *A. rufipalpis* pheromones attracted 569 *A. rufipalpis* males along with four *A. paludum*. Three larvae were found with the soil sampling: two *A. rufipalpis* and one *Melanotus* sp. Also, one *Melanotus* sp. larva was caught by the soil bait traps.

Potential of the Western Corn Rootworm (*Diabrotica virgifera virgifera* Laconte) to be Included Like a Key Pest of Corn in Romania

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Abstract

In analysis it has chosen five temperature criteria [four are referring to soil temperature 10 cm deep (1) it should never be lower than -10°C , (2) the April mean should be above 12.8°C , (3) the May and June mean should be above 17°C and (4) the October mean should be lower than 12.8°C and the fifth is related to air temperature, the June-August means should be above 17°C], needed for an optimal development of WCR and compared them with climatic data for representative localities in Romania. From these five criteria, soil temperature seems to be the most important limitation criteria for spreading of WCR in Romania.

Analysis shows that in main corn cultivated area from Romania, in most favorable zone, first favorable zone and second favorable zone, were is produced over 80% of corn yield in Romania, all criteria are fulfilled in almost all of the years.

It was done a comparison between WCR and the most important corn pest, till now in Romania, *Tanymecus dilaticollis* Gyl.. The actual IPM strategies which should be adopted in Romania for attempt to control WCR pest is presented.

Key Words: *Diabrotica virgifera*, soil temperature limiting factor

Introduction

The western corn rootworm (*Diabrotica virgifera virgifera* La Conte), [WCR], which was for the first time in Europe, in Yugoslavia in 1992, has spread rapidly into other east European countries. This most serious insect pest on maize in the USA and Canada has great economic importance for European maize growing countries, too. The EPPO has included this pest on its quarantine list as A2 pest, and it is assumed to become a quarantine pest for the EC in 2000. The establishment potential is a main part of risk assessment.

Romania has the largest maize-growing area in the Europe. The pest was recorded for the first time in Romania in 1996, and has established during 1997-98 in the southwestern part of the country. As for other countries, the establishing of *Diabrotica virgifera virgifera* in Romania would have a serious impact on maize yield and corn pests' management.

Therefore, the evaluation of its establishing potential as a part of pest risk assessment is absolutely necessary. In 1997-98, a countrywide system was worked out for records of presence of the pest in Romania. This system is based on the Integrated Forecasting System. The application of pheromone traps proved a large spreading of WCR. However, no any notable success in stopping the spreading of the pest in Romania was registered and no damage was registered in Romania, even at places with highest number of trapped beetles.

The WCR ability to adapt itself to the new conditions and absence of its natural enemies makes especially a large effort for the new studies on the ecology and ethology of this pest in these new habitats.

Establishment possibilities for the WCR in Croatia, Germany, Italy and France were reported by many authors (Csamprag et al., 1994; Baufeld and Enzian, 1997).

Materials and Methods

The emergence of the different developmental stages of *D. virgifera virgifera* was calculated for 6 main corn growing area in Romania by means of temperature sums based on the lower development threshold of 11^o C for larvae and of 9^o C for the other stages (Fischer, 1989; Jackson and Eliot, 1988; Levine et al., 1992; Schaafsma, 1991). These multiannual data were from National Institute for Meteorology and Hydrology.

In our analysis we have chosen the same five temperature criteria [four are referring to soil temperature 10 cm deep (1) it should never be lower than -10^oC, (2) the April mean should be above 12.8^oC, (3) the May and June mean should be above 17^oC and (4) the October mean should be lower than 12.8^oC and the fifth is related to air temperature, the June-August means should be above 17^oC] as Barcic-Igrc and Maceljiski (1998), needed for an optimal development of WCR and compared them with climatic data for representative localities in Romania.

Results and Discussion

Maize is cultivated now in Romania over than 3 million of hectares situated, upon the ecological map of corn in Romania (Fig.1), country is divided in 5 zones (most favorable, first favorable, second favorable, less favorable and unfavorable), in general depending on thermic zones (five from six), taking into account average sum temperature over than 10^oC (Fig. 2) (Bilteanu and all., 1983). It is assumed that districts, which are included in most favorable zone, first favorable zone and second favorable zone, produced over 80% of corn yield in Romania.

This area amounted larger than to two third of the maize production area of Romania it suppose to be needed to be treated with insecticides in the next 5-10 years. Till now the most important pest for corn is in Romania *Tanymecus dilaticollis* Gyll. is controlled by seed treatment with pesticides based on carbofuran on more than 700,000 ha in each year. Even than the experience of insecticide control of WCR in USA and Canada, conduct to assumption that Romania has a great potential for up than 1 million of hectares treated with soil insecticides, especially with those based on terbufos, but till now COUNTER 5 G is registered in Romania for other pest. In the next future presence of WCR all over the country, will presume a new corn technology including: machine system for applying soil insecticide in a band (applying of insecticide in-furrow at planting, seems to be inadequate, due to the fact that that sowing time is at the beginning of April and larvae emergence is at the middle of June, when the killing larvae effect has passed), proper calibration, placement, incorporation, planting date, physical and chemical properties of the insecticide, taking into consideration soil factors and weather.

Romania has the largest maize-growing area in the Europe and the Western Corn Rootworm would find maize in all regions of Romania, but the most endangered areas are those with the highest maize concentration.

Under our climatic conditions temperature, like anywhere in the world, is the main factor for the survival of WCR. Egg mortality depends on winter temperature and could reach 50% at -10^oC for four weeks and 100% at -15^oC for one week (Chiang, 1973).

Analysis of 16 zones in which is cultivated corn, our five chosen temperature criteria, four referring to soil temperature, at 10 cm deep were unusual lower than -10^oC, the April temperature were often above 12.8^oC, the May and June mean always were above 17^oC and the October mean in general were lower than 12.8^oC and the fifth is related to air temperature, the June-August means always were above 17^oC (Fig. 3), needed for an optimal development of WCR shows that in main corn cultivated area from Romania, in

most favorable zone, first favorable zone and second favorable zone, were is produced over 80% of corn yield in Romania, all five criteria are fulfilled in almost all of the years and temperatures are favorable for a good development of WCR.. From these five criteria, soil temperature seems to be the most important limitation criteria for spreading of WCR in Romania.

From figure 4, January long-term mean temperature range from -2.9°C to $+1.6^{\circ}\text{C}$ at 5-cm depth and from -2.4°C to $+1^{\circ}\text{C}$ at 10-cm depth. From the last 10 years data registered at Fundulea shows that 10 years average of soil temperature at 5, 10 and 20 cm deep, is favorable for overwintering eggs (table 1) and winter mortality rate is expected to be low. This idea is sustained by the fact that in general the lower temperatures are registered only for a short period of time smaller than one week, thus as example, at Fundulea minimum temperatures were: in December at 5 cm deep, $-10.8/1997$ and $-9.8/1989$, at 10 cm deep $-6.8/1989$, at 20 cm deep, $-3.9/1997$ and $-3.1/1989$; in January at 5 cm deep, $-10.3/1993$ and $-6.2/1996$, at 10 cm deep, $-9/1993$ and $-5.2/1992$, at 20 cm deep, $-6.7/1993$ and $-3.2/1992$; in February at 5 cm deep, $-6.8/1993$ and $-6.2/1996$, at 10 cm deep, $-8.7/1997$ and $-5.8/1996$, at 20 cm deep, $-5.8/1996$ and $-3.6/1991$; in March at 5 cm deep, $-0.7/1997$ and $-0.5/1995$, at 10 cm deep, $-2.3/1996$ and $-0.2/1993$, at 20 cm deep, $-0.6/1993$ and 1996 .

Table 1. Soil temperature (Fundulea 1989-1999)

Month/Deep	5 cm.		10 cm		20 cm.	
	Average	minimum	Average	minimum	Average	minimum
December	0.26	-4.85	0.71	-3.3	1.66	-1.37
January	-0.44	-4.29	0.17	-3.28	0.3	-1.99
February	0.99	-3.7	1.04	-2.84	1.12	-1.77
March	5.88	-3.7	5.68	0.78	5.48	2

For emergence of the different developmental stages of *D. virgifera virgifera* was calculated for main corn growing area in Romania by means of temperature sums basses on the lower development threshold of 11°C for larvae and of 9°C for the other stages. These multiannual data were from National Institute for Meteorology and Hydrology and shows that on 16 analyzed zones it is possible to develop a large population of WCR.

It is necessary that more bioclimatic data should be included in further analysis. Based of actual bioclimatic data it is considered that temperatures in almost all maize growing regions are of Romania are favorable for a good development of WCR.

Conclusions

From five criteria, soil temperature in January at 10 cm deep, seems to be the most important limitation criteria for spreading of WCR in Romania.

Based of actual bioclimatic data our analysis shows that in main corn cultivated area from Romania, in most favorable zone, first favorable zone and second favorable zone, were is produced over 80% of corn yield in Romania, all criteria are fulfilled in almost all of the years and temperatures are favorable for a good development of WCR.

Due to the fact that it was impossible to establish a quarantine barrier, the actual IPM strategies, which should be adopted in Romania for, attempt to control WCR pest has to include crop rotation, a new machine system for applying insecticides in the sowing time and after this and registration of a large variety of effective insecticides.

Fig.1 Ecological map of corn in Romania

Fig. 2 Thermic zones of corn in Romania, taking into account average sum temperature over than +10⁰C

Fig. 3 Isotherms of July in Romania (multiannual average)

Fig. 4 Isotherms of January in Romania (multiannual average) and decade's temperature and average at 5 and 10 cm deep from 16 localities situated in different zones of cultivated corn

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The Western Corn Rootworm (*Diabrotica virgifera virgifera*) – Potential Loss and Insecticides Use in Germany

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Abstract

Diabrotica virgifera virgifera is the most serious insect pest on corn in the USA and Canada. The non-native leaf beetle was introduced into South-East Europe at the beginning of the 90ies and has continuously spread since then. Therefore, it is expected that the pest will reach further European corn-producing areas like Germany, too.

The Western corn rootworm would find corn in all regions of Germany, but the most endangered areas are those with the highest corn concentration. It is assumed that regions with more than 50 % of arable land in corn have significant areas in continuous corn. These areas have been defined as “corn areas with high risk”. The production of continuous corn is the precondition for a high establishment potential and a further rapid development of corn rootworm populations resulting in crop losses.

In Germany, in the four federal lands of Niedersachsen, Nordrhein-Westfalen, Bayern and Baden-Württemberg there is a concentration of continuous corn. About 348,000 ha of corn (130,000 ha grain corn, 218,000 ha silage corn) in Germany are under high risk of Western corn rootworm infestation. This area amounted to one third of the corn production area of the four above mentioned federal lands and more than one fifth of the whole corn production area of Germany in 1995. The long-term experience of insecticide use in Canada (Ontario), supports the assumption that Germany has a potential for up to 111,000 ha treated with rootworm insecticide annually. That would evoke an added insecticide (Counter SG, terbufos) cost of 25 million DM per annum. Without any insecticide use, rootworm infestations could cost German corn producers 49 million DM in crop losses each year. Experience from Canada supports the changing of crop rotation away from continuous corn to reduce the potential for the establishment of Western corn rootworm, the potential for economic loss, and increased application of insecticides.

Introduction

The Western corn rootworm *Diabrotica virgifera virgifera* LeConte is a serious pest of corn in North America and causes considerable crop losses. The cost of pest control and crop losses can approach 1,000 million US \$ in the USA annually (KRYSAN and MILLER, 1986). The larvae cause the main damage by feeding on roots of corn plants. This leads to a reduction in growth and in the case of severe root damage, corn lodging and loss of ears during harvest (GAVLOSKI *et al.*, 1992, GROENEWEGEN, 1992). Adults cause secondary damage by feeding on silks which results in insufficient pollination and thus problems for corn seed production (CULY *et al.*, 1992). Because of the economic importance for the EU of the *Diabrotica*-group, 5 species/subspecies including the Western corn rootworm are regulated as quarantine pests in Council Directive 77/93/EC (Annex I).

In Europe, *D. virgifera virgifera* was introduced to Yugoslavia in 1992 (BACA, 1994) and has spread to other European countries like Croatia, Hungary, Bosnia-Herzegowina, Romania and Bulgaria. Since 1997 there has been a second outbreak in Italy which is still under eradication. The corn pest may arrive in Germany in the near future in case of introduction or in the distant future in case of ongoing infestation in Southeast Europe.

Host availability and suitable climatic conditions are the main reasons of the establishment of a non-native pest. The analysis of the climatic conditions leads to the result that *D. virgifera virgifera* would find suitable conditions in Germany (BAUFELD *et al.*, 1996, BAUFELD and ENZIAN, 1997). Furthermore, the assessment of the impact of cropping practices on rootworm in Canada allowed to evaluate the trophic conditions and establishment potential for the Western corn rootworm in Germany. The Canadian data allowed to predict the potential for rootworm damage in Germany and the relative adoption of crop rotation as a primary control strategy for the Western corn rootworm (SCHAAFSMA *et al.*, 1999).

Materials and Methods

Data for the land area under corn production for 215 counties of 12 corn-producing federal lands in Germany in 1995 were put at our disposal by the Federal Statistical Office. However, the average percentage of the county area in corn does not reflect the real area in corn of the individual corn-growing farms and is in nearly all farms below 50 %. Precise data on the basis of farms were not available (protection of data privacy). The Erzeugergemeinschaft Mitteldeutscher Körnermaisbauern w. V. had made random surveys of corn-growing farms. On this basis it provided data on crop rotations and calculated the average percentage of corn within the crop rotations for 213 counties (ACKERMANN, personal). It was assumed that areas including farms with > 50 % of arable land in corn were at high risk for rootworm establishment and damage (above the economic threshold), because these areas are likely to have a high frequency of continuous corn.

In the U.S. fields (Iowa) where corn is cultivated continuously *Diabrotica* adults exceeded the economic threshold of 1 beetle/plant in 95 % of fields surveyed over 3 years, with an overall mean density of 4.2 beetles/plant (FOSTER and TOLLEFSON, 1996). Corn rootworm infestation has caused corn yield decreases of 10-13 % (APPLE *et al.*, 1971; PETTY *et al.*, 1968). A 10 % yield loss was assumed in untreated fields for the calculation for Germany. The expected loss was estimated on a basis of an average yield of 8.05 t/ha for grain corn and 47 t/ha for silage corn. Therefore returns were expected to make up 2310.35 DM/ha and 876 DM/ha for grain and silage corn, respectively (GRAF *et al.*, 1996).

In Canada (Ontario) long-term data have shown a mean of more than 50 % continuous corn with an area of about 32 % treated with rootworm insecticide (SCHAAFSMA *et al.*, 1999). The same potential rate of use for rootworm insecticides was assumed for Germany. Insecticide costs were calculated for Counter SG (terbufos), the only registered insecticide for control of corn pests (wire worm). Calculations started from the recommended rate of 1.25 g/m (ANONYMOUS, 1996), 0.75 m row spacing (16.7 kg/ha) and a market price of 13.40 DM/kg. Application costs have not been considered because insecticides are usually applied with the planting equipment at seeding time.

Results and Discussion

About 1.5 million ha of corn were grown in Germany in 1995, which represented nearly one third of the EU production and the second biggest corn producer of the EU behind France. The concentration of corn production was inhomogeneous in the different federal lands. In some regions corn cultivation reached relatively high concentrations (Fig.). Beside favorable climatic conditions the production of continuous corn is the precondition for rapid development, and the probability of establishment of Western corn rootworm populations is very high. It is assumed that regions containing more than 50 % of arable land in corn have significant areas in continuous corn. These areas have been defined as "corn areas with high risk". The total high risk land area in Niedersachsen, Nordrhein-Westfalen, Bayern, and Baden-Württemberg regions was 133,509 ha, 114,113 ha, 86,992 ha, and 13,006 ha, respectively (Table). With 50 % Nordrhein-Westfalen had the highest percentage of total high risk area, followed by Niedersachsen with 44 %, Bayern with 22 % and Baden-Württemberg with 11%.

The total corn area at high risk amounted to 347,620 ha, one third (33 %) of the corn production area of these four federal lands (1,043,743 ha) and about 23 % of the entire corn production area of Germany. The area with the highest percentage of corn had 217,867 ha in silage corn, compared to 129,753 ha in grain corn (Table). However, grain corn contributed proportionally more high risk area than silage corn. For example, an estimated 56 % of the entire grain corn area (230,812 ha) was estimated to be at high risk, whereas for silage corn only 17 % of the entire silage area (1,251,788 ha) were at high risk.

Corn is produced in three of four years continuously (with a maximum of 75 % corn in a crop rotation) in the counties Ammerland (7,374 ha), Grafschaft Bentheim (18,866 ha) in Niedersachsen and in the counties Borken (34,558 ha), Steinfurt (35,974 ha) in Nordrhein-Westfalen. Furthermore, some farms in Baden-Württemberg in the Oberrhein region and in the south of Bayern (bordering Austria) are known for mono-culture of corn.

Corn silage production dominated in both northwestern federal lands Niedersachsen (87,755 ha silage corn high risk area) and Nordrhein-Westfalen (68,755 ha) and in the southeastern federal land Bayern (59,585 ha) (Table). In contrast, the southwestern federal land, Baden-Württemberg, has a favorable micro-climate in the Rhine valley for grain corn production (11,234 ha grain corn high risk area) and many corn producers are well established there.

Conclusion

In Canada, where crop rotation has changed in the last ten years, crop rotation has clearly been an effective tool for the control of corn rootworms in Ontario (SCHAAFSMA et al., 1999). Crop rotation is one of the most effective long-term control measures for *D. virgifera virgifera*. This strategy would reduce the potential for the establishment of the Western corn rootworm and the potential for economic loss.

The Western corn rootworm would find suitable climatic and trophic conditions for the establishment in Germany. The climatic conditions in winter and in the period of its development are not a limiting factor for this non-native pest in Germany (BAUFELD et al., 1996, BAUFELD and ENZIAN, 1997). *D. virgifera virgifera* would find its host throughout Germany, but the development conditions and the potential for damage are different in each federal land depending on the concentration of corn for each region. Germany still has a high concentration

of continuous corn in four of the main corn-producing federal lands. The federal lands of Niedersachsen, Nordrhein-Westfalen, Bayern and Baden-Württemberg have a large percentage of corn area under high risk for the establishment of the Western corn rootworm. This pest would find ideal development conditions in these regions and quick establishment is to be expected resulting in damage above the economic threshold in some years. The consequence will be economic loss or an insecticide treatment will be necessary.

Baden-Württemberg, with its special micro-climate for corn seed production, presents an potential area for higher economic losses by corn rootworm. Seed corn inbreds are more susceptible to rootworm damage because of lower plant vigour and smaller plant habit, by comparison with corn hybrids. One to three adults per ear reduce kernels per ha and kernels per kg (CULY *et al.*, 1992). A more integrated approach might be necessary in this specialized region to reduce potential seed crop losses. On the basis of the Canadian long-term data of about 32% fields treated with rootworm insecticide and a high risk area of corn of up to 347,620 ha in Germany, the potential treatment area in Germany was calculated at 111,238 ha annually. Until now only one insecticide is registered to control pests in corn in Germany, Counter SG. Starting on the recommended rate of 16.7 kg/ha for Counter SG and a treated area of 32 % of the high risk corn area, an additional 1857.7 t of insecticide would be applied annually at an added cost of 25 million DM per year for insecticides. If the Western corn rootworm was left untreated and a 10 % yield loss was assumed, a loss of 231 DM/ha of grain and 87.60 DM/ha of silage corn could be expected. Applying these losses to the high risk area would result in a total loss of 30 million DM for grain corn and 19 million DM for silage corn. A plant protection cost of 25 million DM would mitigate a crop loss of 49 million DM.

It is assumed that the spread of this pest will continue over all corn growing areas in Europe but will reach Germany in some years. The prevention of introduction is supported by the regulation as quarantine pest in Council Directive 77/93/EC. The infestation in Italy in 1998 shows how important it is to avoid an introduction and to eradicate an infestation from the beginning. However, up to now the pathway is not clear and the spread will go on. Therefore it is necessary to elaborate a package of measures for Germany to prevent introduction of the pest and its establishment, and to contain the "natural" spread rate. The changing of crop rotation away from continuous corn in Germany is one possibility to reduce the potential for establishment of the Western corn rootworm, potential economic loss, and reliance on insecticides.

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Table 1. Estimate of the areas in Germany at high risk for corn rootworm infestation based on the proportion of arable area under corn production and the potential for corn production in two or more successive years. 1995.

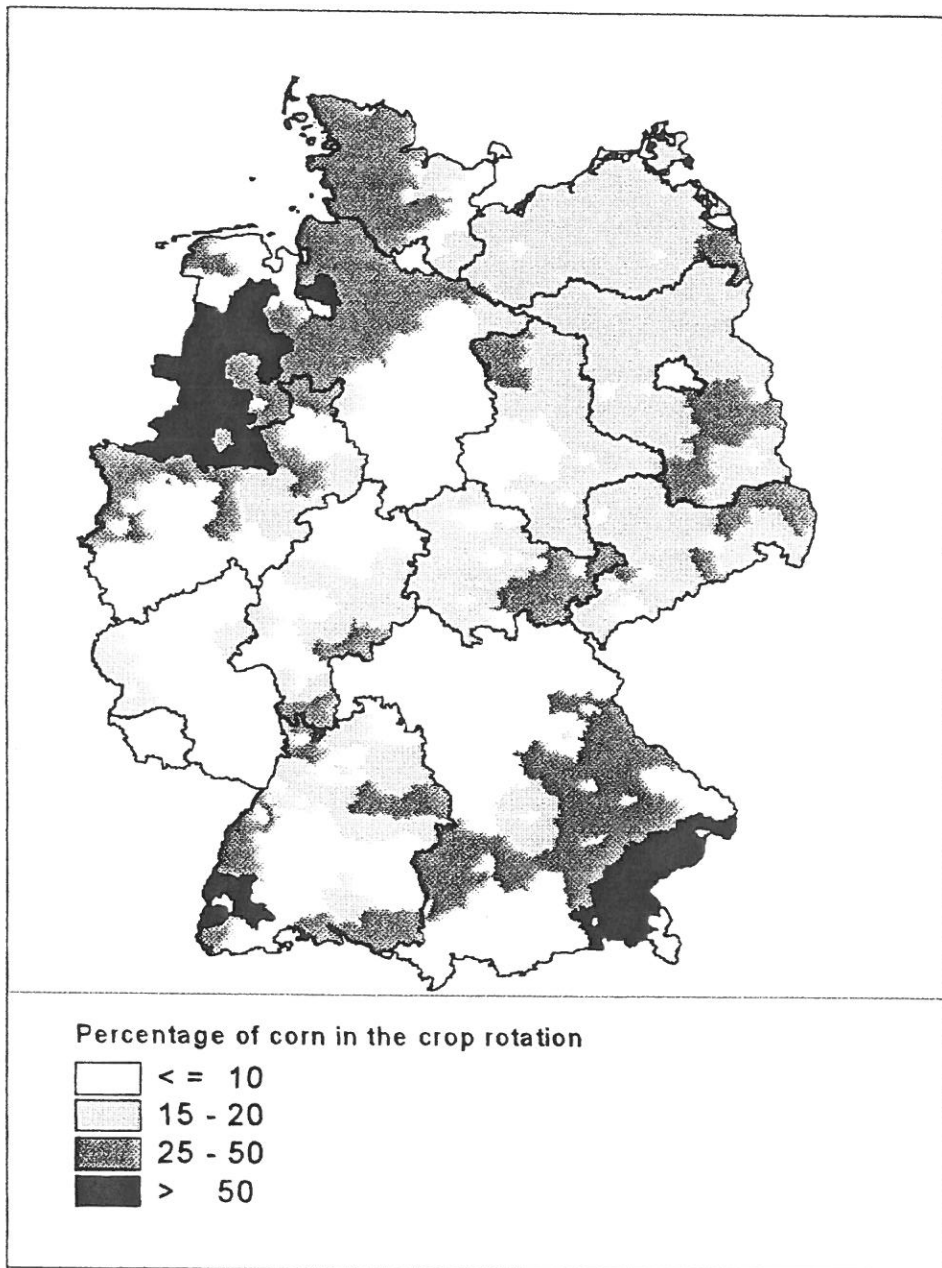
Federal Land	Area of high risk corn in Germany (ha)*			Percentage of total corn area with high risk
	Grain	Silage	Total	
Niedersachsen	45,754	87,755	133,509	44
Nordrhein-Westfalen	45,358	68,755	114,113	50
Bayern	27,407	59,585	86,992	22
Baden-Württemberg	11,234	1,772	13,006	11
Total	129,753	217,867	347,620	33

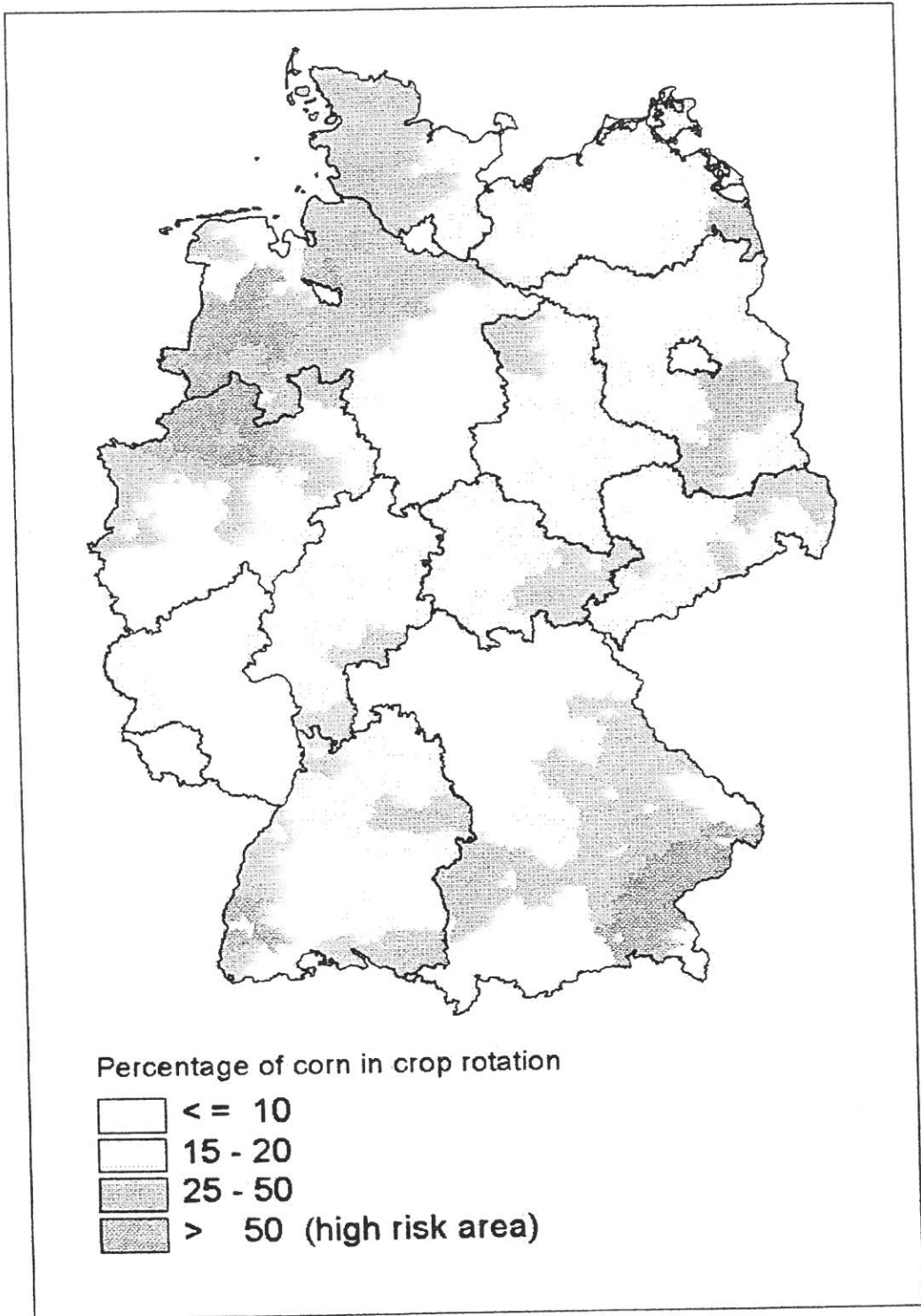
*area sums for counties within german federal lands that had > 50 % of arable land in corn production in 1995

Figure Captions

Fig. (as black and white or color figure):

The percentage of corn in the crop rotation in the counties across Germany in 1995.





Incidence of Corn Leaf Blights in Çukurova Region and Important Corn Diseases in Turkey

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Abstract

It was found many different diseases on corn in Turkey. Two of those diseases, Maydis and Turcicum Leaf Blights are performed in this survey study in Çukurova Region of Turkey. According to the results of surveys, Maydis Leaf Blight is more important than Turcicum Leaf Blight for Çukurova Region. Both of them are found more prevalent on second crop corn as regards first crop corn.

Introduction

Corn is grown for a long time, in Turkey. Because of it hasn't any marketing problem and its high yield potential, especially after the 80's, hybrid corn is easily entered in Turkey and corn growing areas increased. Important corn growing areas of Turkey are Black Sea, Mediterranean, Marmara and Aegean Regions with total area of 485.000 ha and production of 1.850.000 tons. Çukurova Region is a part of Mediterranean Region and has 10.2 % of growing areas and 19.22 % of production in Turkey (Anonymous, 1996).

Most important diseases of corn are *Turcicum* and *Maydis* leaf blights, *Fusarium* stalk rot, smut, rust, *Curvularia* leaf blight, *Cephalosporium maydis*, *Rhizoctonia solani* (Ataç, 1984; Hatat and Maden, 1988; Saydam et al., 1992; Tunçdemir et al., 1996). In addition to those diseases, also some fungal pathogens including *Fusarium* spp., *Alternaria* spp., *Cladosporium* spp., *Epicoccum* sp., *Penicillium* spp., *Aspergillus* spp., *Rhizopus* spp., *Mucor* spp., *Termonyces lanuginosus*, *Artrobotrys* spp., *Neurospora oryzae*, *Stemphyllum* spp., *Trichoderma* spp. and *Phoma* spp., were determined on corn seed (Hatat and Maden, 1988; Soran and Asan, 1989; Aktaş and Tunalı, 1990). As regards of our observations, there is an increase for *Pythium* stalk rot.

Turcicum and Maydis leaf blights caused economically loses in 1993 and 1994, and this study carried out to determine prevalence, incidence and severity of the Turcicum and Maydis leaf blights on corn in Çukurova Region, in 1994, 1995 and 1996.

Material and Methods

The trials were set on first and second crop corn, to determine Turcicum and Maydis leaf blights caused by *Exserohilum turcicum* and *Helminthosporium maydis* respectively, at sampling units that 1:10.000 of growing areas and 8. and 9. growing stages of corn (Anonymous, 1966). The observations performed at one point if the field chosen as a sampling unit is 0,1-1 ha, and at three separate points for 1-5 ha, five for 5-10 ha, seven for 10-20 ha and 10 for larger than 20 ha. Observations realized at side by side 25 plants on a row. Elliot and Jenkins (1946)' s scale and Townsend- Hauberger formula are used to obtain the diseases severity.

Results and Discussion

Result of the study is shown at Table 1. As it is shown at Table 1, for the first crop, Turcicum leaf blight didn't observe in 1996. In 1994 and 1995 prevalences were 32.3 % and 36.7 %, and incidences were 0.67 % and 0.64 %, severities were 0.07 and 0.07, respectively. On the other hand, in 1994, 1995 and 1996, for Maydis leaf blight, prevalences were 54.8, 83.3 and 45.5 %, and incidences were 5.73, 13.47 and 3.40 %, and severities were 0.65, 2.63 and 0.46, respectively.

Table 1. Prevalence, incidence and severity of Turcicum and Maydis Leaf Blights on Corn in Çukurova Region

Disease	First or Second Crop Corn	1994			1995			1996		
		Prevalence (%)	Incidence (%)	Severity (%)	Prevalence (%)	Incidence (%)	Severity (%)	Prevalence (%)	Incidence (%)	Severity (%)
TLB	F.C.	32,30	0,67	0,07	36,70	0,64	0,07	0,00	0,00	0,00
	S.C.	50,00	25,62	8,23	44,40	3,72	0,78	29,40	2,87	0,39
MLB	F.C.	54,80	5,73	0,65	83,30	13,47	2,63	45,50	3,40	0,46
	S.C.	70,80	30,00	7,66	72,20	13,24	2,44	88,20	51,77	14,11

T.L.B.= Turcicum Leaf Blight, M.L.B. = Maydis Leaf Blight, F.C.= First Crop, S.C.= Second Crop

Turcicum leaf blight prevalences were 50.0, 44.4 and 29.4 %, and incidences were 25.62, 3.72 and 2.87 %, and severities were 8.23, 0.78 and 0.39 %, for Maydis leaf blight prevalences were 70.8, 72.2 and 88.2 %, and incidences were 30.00, 13.24 and 51.77 %, and severities were 7.66, 2.44 and 14.11 %, respectively on the second crop corn.

As it is shown at Table 1, all records for second crop is higher than first crop except 1995 for Maydis Leaf Blight. Because after the wintering inoculum increases on the first crop corn and second crop corn presents at the same time with first crop corn which has not harvested yet, very abundant conidia that increase on the lesions on first crops cause more severe diseases on the second crop corn. Besides, many researchers reported that increased relative humidity causes an increase of disease level (Ayaydın, 1975; Khatri, 1993; Krausz et al., 1993) According to Ayaydın (1975) because of some rivers on the plain, some swamp area and trees around the fields diseases are widespread at Çarşamba and Terme district. In parallel to this report, there are similar conditions with Seyhan and Ceyhan rivers, irrigation and drainage canals and intensive irrigation, high relative humidity in Çukurova. Also, according to Tunçdemir et al. (1992) Turcicum Leaf Blight is more prevalent in temperate and damp conditions and late sowing and especially second crop corn.

There are differences between the years. This difference may be due to climatic condition or different cultivars or hybrid seeds sown in fields. It is known that there are many different studies to obtain resistant varieties (Ataç, 1995; Tunçdemir et al, 1996; Pataky et al., 1998). According to investigation carried out by Ataç (1984), the highest severity level of Turcicum Leaf Blight was 60.00 % on the local variety, but 16.00 % on P-3360 in Adana province. At the surveys in 1996, incidence of Turcicum Leaf Blight was found 84.00 %, and severity was

21.20 % on a local popcorn variety. So, changes of the cultivars year by year would make problems depending on their susceptibility.

According to the results of the survey, Maydis Leaf Blight is more important for Çukurova Region on both first and second crop corn compare to Turcicum Leaf Blight. During the survey, prevalence of diseases was 100 % and incidence 63.67 % in some fields. It means that, if the susceptible varieties is sown, problem will increase. On the other hand, in the Blacksea region which is another important corn growing region, the most important disease is Turcicum Leaf Blight (Ayaydin, 1975; Hatat and Maden,1988) Difference of between Mediterranean and Blacksea Regions is suitable to climatic requests of pathogens. In spite of both diseases can be found in most corn growing areas, Turcicum Leaf Blight prefers relatively cool conditions as 18 – 22 °C, but Maydis Leaf Blight as 20 – 32 °C (Anonymous, 1980). Average temperatures in Çukurova are always higher than Blacksea Region. Besides, according to investigations on dynamics of the 2 species on maize performed by Jacqua (1989) show that an antagonism in favour of *Helminthosporium maydis* occurs between it and *Exserohilum turcicum*. Due to its extremely short incubation period and its rapid development, this species can impair the expression of *E. turcicum* in mixed inoculations on already unfolded maize leaves. Braun and Howard (1994) have found that conidia of *Helminthosporium maydis* keeps to leaves strongly in a short time like 50 – 60 minutes after germination. However, lesions of *Helminthosporium maydis* rapidly grow and appear in 1 –2 days after inoculation. But lesions of *Exserohilum turcicum* appear after 7 –12 days. This situation explains why Maydis Leaf Blight is widespread in Çukurova Region. Because, climatic conditions of Çukurova are more suitable for *Helminthosporium maydis* if there are adequate inoculum for both pathogens.

At the same time, the difference between Mediterranean and Blacksea Regions is parallel with their names. Because some researchers called Turcicum Leaf Blight as Northern Leaf Blight and Maydis Leaf Blight as Southern Leaf Blight.

Finally, it is clearly understood that Maydis Leaf Blight is more important in Çukurova Region. But, if the susceptible varieties sown to Turcicum Leaf Blight, it would make damage.

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