IWGO

International Working Group on *Ostrinia* and other maize pests

Regione Veneto
Servizio Fitosanitario

VENETO AGRICOLTURA
Azienda Regionale per i settori Agricolo, Forestale e Agro-Alimentare

Under the aegis of the University of Padova

XXI IWGO CONFERENCE

VIII DIABROTICA SUBGROUP MEETING

PROCEEDINGS BOOK

Legnaro - Padua - Venice - ITALY

October 27 - November 3, 2001

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Proceedings

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Agripolis – S.S. Romea, 16 – 35020 Legnaro (PD)
Tel. 049/8293711 – Fax 049/8293815
e-mail: va@venetoagricoltura.org
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Tel. 049/8293920 – Fax 049/8293919
e-mail : divulgazione.formazione@venetoagricoltura.org

Editing e coordinamento editoriale
Alessandra Tadiotto – Isabella Lavezzo

Preface

The Veneto Region had the honor of hosting the XXI IWGO Conference (International Working Group on *Ostrinia Nubilalis* and Other Maize Pests), the international group dedicated to all maize pests.

This year the meeting was preceded by the VIII Diabrotica Subgroup Meeting, the specific group of study on *Diabrotica Virgifera*, a coleopter that represents one of most serious problems of the corn in USA and present in Europe since 1992. The signaling of a focus of *Diabrotica* in the fields near the Venice airport in 1998 prompts the interest of the Veneto Region that, through the Regional Phytosanitary Service and the cooperation of the University of Padova, has activated a specific program of eradication that has attracted the attention of the IWGO and the other European nations equally involved in the containment of the parasite.

We received more than 100 requests, coming from 24 different Countries, to attend the VIII Diabrotica Subgroup Meeting and the XXI IWGO Conference. A unique opportunity to take stock of the most recent researches on the maize pests.

The meetings were organized by the Regional Phytosanitary Service and Veneto Agricoltura, at the Congress Center of Corte Benedettina at Legnaro (Padova) from 27 October to 3 November 2001.

L'Amministratore Unico
di Veneto Agricoltura
- *Giorgio Carollo* -

THE XXIth IWGO MEETING IN VENICE, ITALY
Oct. 27 – Nov. 4, 2001

Harald K. Berger
IWGO President; Federal Office and Research Center for Agriculture;
Spargelfeldstraße 191, P.O.B. 400; A - 1226 Wien, Austria
Tel.: + 43/1/732 16 – 2002
FAX: + 43/1/732 16 – 2108
e-mail: hberger@bfl.at

The IWGO meeting was the first time within the International Working Group on Ostrinia (IWGO) history that both the "big" IWGO – Meeting and the Meeting of the Diabrotica Subgroup took place at the same time and at the same place. I actually doubt that we will ever find a local organizer who will take on such a workload again ! The response to the first call was larger than we anticipated: more than 120 scientists indicated interest in attending both meetings. At the meeting there were approximately 100 people from 27 countries. At the XIXth IWGO meeting in Guimares, Portugal, we had 30 participants and in Adana, Turkey, at the XXth IWGO meeting there were approximately 50 participants who we thought might be the peak of attendance of IWGO meetings. In Paris and Stuttgart, places of the most recent Diabrotica Subgroup meetings, about 60 people participated.

The reasons for the large amount of participants are two: the appearance of *Diabrotica virgifera virgifera* LeConte in Europe and a joint meeting of both working groups is certainly the most important reason. The second one is - and I do not hesitate to say it - the place of the meeting, Venice is really worthwhile coming to.

Since its establishment in 1968, the "International Working Group on Ostrinia" has been very active. Many meetings took place and several publications were issued:

1968	Founded in Moscow
1969	1st Meeting in Vienna, Austria
1970	2nd Meeting in Zemun, Yugoslavia (Serbia)
1971	3rd Meeting in Bordeaux, France
1972	4th Meeting in Martonvasar, Hungary
1973	5th Meeting in Zagreb, Yugoslavia (Croatia)
1974	6th Meeting in St. Paul; Minnesota; USA
1975	7th Meeting in Leningrad (St. Petersburg); Soviet Union (Russia)
1976	8th Meeting in Madrid , Spain
1977	9th Meeting in Wroczlaw, Poland
1978	10th Meeting in Bergamo, Italy
1980	11th Meeting in Vienna, Austria
1982	12th Meeting in Pistany, CSSR (Slovakia)
1984	13th Meeting in Colmar, France
1986	14th Meeting in Beijing, China
1989	15th Meeting in Varna, Bulgaria
1991	16th Meeting in Martonvasar, Hungary
1993	17th Meeting in Volos Greece
1995	18th Meeting in Turda, Rumania

1997 19th Meeting in Braga, Portugal
1999 20th Meeting in Adana, Turkey
2001 21st Meeting in Venice, Italy

The original idea of IWGO was to exchange inbred lines within the group and test these lines for resistance against the most important maize pest throughout the world, the European Corn Borer (*Ostrinia nubilalis* Hbn.). The most latest results of this breeding program have been presented by Franja BACA within this meeting. But by this time the group had grown considerably and other pests also became of more interest. *Diabrotica* is just one of them and certainly the most important nowadays.

From 1973 – 1980 IWGO published the results of its breeding program and, since 1980, proceedings of all IWGO meetings were issued with the financial help of the Global IOBC.

Since 1980, IWGO publishes biannually the "IWGO - NEWSLETTER". The newsletter contains mainly the summaries of papers presented at previous meetings, but also a large number of original papers have also been published. The newsletter is also a medium to maintain contact within the group and to distribute information about the members. Until now, 21 volumes, consisting mostly of two issues per year have been released. Alltogether 33 issues of more than 30 pages each were published.

PUBLICATIONS OF IWGO 1968 - 2001

1973 **Report of the International Project on Ostrinia nubilalis Phase I; Results 1969 - 1970**;
B. Dolinka (Edit.); Agric. Res. Inst. Hungarian Acad. of Sciences; Martonvasar; 168 p.

1975 **Report of the International Project on Ostrinia nubilalis Phase II**;
B. Dolinka (Edit.); Agric. Res. Inst. Hungarian Acad. of Sciences; Martonvasar; 138 p.

1976 **Report of the International Project on Ostrinia nubilalis Phase III**;
B. Dolinka (Edit.); Agric. Res. Inst. Hungarian Acad. of Sciences; Martonvasar;

1980 **Twelve Years Results (1969 - 1980) of IWGO issue of "Probleme de Protectia Plantelor" 9 (2)**;
D. Mustea (Edit); Agricultural Research Station; Turda, Romania

1984 **Proceedings of the XIIth Symposium of the International Working Group on *Ostrinia***;
J. Janda (Edit.); Proroda, Bratislava pre vyskuminy ustov kukurice v. Trnave; 146 p.

1986 **Proceedings of the XIIIth Symposium of the International working Group on *Ostrinia***;
P. Anglad & C. Cortez (Edit.) Colmar - Bordeaux , France; 116 p.

1986 **Proceedings of the XIVth Symposium of the International Working Group on *Ostrinia***;
Zhou D.(Edit.) Beijing, Peoples Republic of China; 181 p.

1989 **Proceedings of the XV^th^ Symposium of the International Working Group on *Ostrinia*;**
T. Georgiev (Edit.) Technical University "A. Kanchev" ; Rousse, Bulgaria; 172 p.

1993 **Proceedings of the XVI^th^ Symposium of the International Working Group on *Ostrinia*;**
J. Tsitsipis (Edit.) University of Thessalia; Volos, Greece; 72 p.

1995 **Proceedings of the XVII^th^ Symposium of the International Working Group on *Ostrinia*;**
D. Mustea (Edit.) Agricultural Research Station Turda, Romania; 133 p.

1997 **Proceedings of the XVIII^th^ Symposium of the International Working Group on *Ostrinia*;**
Esteves-Silas Pego (Edit.) D. Draed – Numi; Braga, Portugal 341 p.

1998 **Proceedings of the XIX^th^ Symposium of the International Working Group on *Ostrinia*;**
Serpil Kornosor (Edit.) Cukurova University; Adana, Turkey 197 p.

Since 1980: 33 issues of "IWGO NEWSLETTER"
Since 1999: IWGO is also on www: http://www.infoland.at/iwgo/

But times change. While retaining the abbreviation "IWGO", the group changed its name to "International Working Group on Ostrinia and other Maize Pests". The group is now dealing with all aspects concerning corn pests throughout the world. Therefore the number of scientists interested in this Global IOBC working group has increased, primarily because maize is one of the most widespread crops in the world. It is grown in many countries on all continents. And the same main pests in maize occur almost everywhere where maize is grown.

IWGO Diabrotica Subgroup meetings took place in:

Mar. 1995	1st Subgroup Meeting in Graz, Austria
Nov. 1995	2nd Subgroup Meeting in Gödöllő, Hungary
Oct. 1996	3rd Subgroup Meeting in Zagreb, Croatia
Nov. 1997	4th Subgroup Meeting in Gödöllő, Hungary
Oct. 1998	5th Subgroup Meeting in Rogaska Slatina, Slovenia
Nov. 1999	6th Subgroup Meeting in Paris, France
Nov. 2000	7th Subgroup Meeting in Stuttgart, Germany
Nov. 2001	8th Subgroup Meeting in Venice, Italy

The 9th Subgroup Meeting is planned to take place in Belgrade, Yugoslavia, the core of our problem in October 2001.

In order to save program money throughout the world for scientific institutions, universities and even private companies, IWGO decided for the first time to combine the IWGO meeting (which originally took place every year and later every second year) with the Diabrotica Subgroup Meeting.

Previously, the meetings of the FAO Group, the EPPO ad hoc Panel and the EU Research Program Group had joined their meetings, but not together with the

IWGO group. But surely *Diabrotica virgifera virgifera* is the main item in our meeting. Since the 1st Diabrotica subgroup meeting in Graz in 1995, where 25 colleagues from 6 countries were present, the pest has spread over a large area in central Europe. In 1995, only Serbia was infested. In 2001 the pest is also present in Hungary, Croatia, Romania, Bulgaria, Bosnia – Herzegovina, Slovakia, Italy, Switzerland and Ukraine too. Austria, Slovenia and Albania are threatened.

IWGO is the oldest working group within the Global IOBC and is now in its 33rd year of existence . The group was founded in 1968 during the XIIIth International Congress of Entomology in Moscow. But the roots reach back to the US regional project on Ostrinia begun in 1951. The IWGO was established parallel to this US regional project. The founders of the group in 1968 were D. HADZISTEVIC (Yugoslavia), who had the original idea of founding a group of international cooperators, H.C. CHIANG (USA), who brought the ideas of the US projects into the group and was its first president, I.D. SHAPIRO from Russia (USSR), T. PERJU (Romania), C. KANIA (Poland) and B. DOLINKA (Hungary), all well-known researchers or maize breeders and founders and first members of IWGO. The second president after the retirement of Prof. Chiang in 1983, was Pierre ANGLADE from Bordeaux, France. Since 1993 I have been the convenor of the group.

As already mentioned, the main goal of the group was the exchange of inbred lines from different countries, their artificial infestation with corn borer eggs and evaluation of the resistance and tolerance of the lines.

The meeting of IWGO (both, the IWGO meeting and the meeting of the Diabrotica Subgroup) is well known for its high scientific value and for the output of its scientific work. And the meeting in Venice also fulfilled IWGO's high scientific standard.

Last but not least I want to express my thanks: my special thanks go to the local organizers Franco NORIDO, Stefano BARBIERI, Lorenzo FURLAN, Marco VETTORAZZO, Francesca GRANDIS, Silvia CERONI and Vanna FUNES who organized the meeting perfectly. Their challenge was that they had to deal not only with one meeting, but more or less 4 meetings with completely different emphases. In Austria we say "too many cooks spoil the cake". But at the end of the meeting I can say that they managed it perfectly. "The cake tastes wonderful". My warmest thanks goes to all of them.

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VIII DIABROTICA SUBGROUP MEETING

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EGG-LAYING BEHAVIOUR OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE AND POSSIBLE ASSOCIATIONS WITH ITS MORPHOLOGY

Franck J. Muller, Dominique Tauban, Michel Renou, Sylvie Derridj,
INRA, Unité de Phytopharmacie et des Médiateurs Chimiques,
Route de St Cyr, 78 026 Versailles, France

Abstract

The contact of insects with the plant surface is a behavioural step during which the host can be recognised and thus induces acceptance for egg laying. The chemical analyses of water-soluble maize surface leachates show that both growth stages and organs of the maize plant can be discriminated by their primary metabolite composition. We have shown that the maize leaf surface water-soluble leachates stimulate egg laying of *Diabrotica virgifera virgifera* (*D.v.v.)* on artificial substrates.

The question is how the bio-chemicals can influence the egg laying behaviour and where they are perceived.

Morphology of the ovipositor was observed under microscope. Scanning electron microscopy was used to search for sensilla on legs and ovipositor. Laboratory observations were carried out to determine the different steps of *D.v.v.* egg laying behaviour on an artificial substrate. Distribution of the insects in a maize field was investigated in Hungary at different times of the day and at two growth stages.

Before laying eggs on artificial substrate, females extrude their ovipositor, which is used for scanning from back to forth. Preliminary observations show the presence of sensilla on the tarsi and on the upper side of the ovipositor, which are likely to be gustatory. Both were found to be put in contact with the substrate during the "pre-egg laying" event.

Walking and resting on the leaves and then husks were the main behaviours observed in the field. Abundance of females on the maize plants varied with the maize growth stage.

We hypothesise that insect morphology and egg laying behaviour are adapted to the use of bio-chemical information given by the plant surface, which provides the decision making of egg laying at the plant bottom into the soil.

Key words: Egg-laying behaviour, ovipositor, tarsi, gustatory sensillae, maize.

INTRODUCTION

The appearance in Europe ten years ago of *Diabrotica virgifera virgifera*, a new species known as a major corn pest in North America Continent since many years, economically threatens the producers. The first European studies were mainly directed towards population monitoring and distribution in Europe. The pest spread significantly fast all around Yugoslavia where it first appeared in 1992 and is present in Italy and Switzerland since 2000.

Many work researches have been realized since a long time in the U.S.A. However, practically nothing is known about *D.v.v.* egg-laying behaviour. Therefore it was decided to start a study with the aim to understand *D.v.v* egg-laying behaviour to appreciate the feasibility of crop rotations in Europe.

It was shown that corn plant surface washings stimulate or deter *D.v.v* egg-laying according to the corn growth stages (see Derridj, S. *et al.* in the proceedings). The cue contact which stimulates the females must be taken while they are on the corn plant. This prompted us to study in details *D.v.v.* egg-laying behaviour and gustatory sensilla which could be involved.

First we observed the female behaviour on artificial substrate to identify the main organs involved. Then we performed a morphological study of ovipositors and tarsae.

Observations of the localisation of the females in cornfields were carried on to determine on which plant organ the information would be taken to lay eggs.

Female distribution on the corn plant within the day and at two different periods of summer was investigated.

D.v.v. was first found in the south of Hungary in 1995 and has then spread very significantly. The highest population is in the south of the country. Larval damages on corn were already observed in 4 counties including Csongrad County, where we decided to conduct our field test observations.

Material and methods

Behavioural observations on artificial substrate

Behavioural observations in artificial conditions were carried on in a quarantine room of CABI Bioscience Switzerland Centre. *D.v.v.* mass rearing is a non-diapaused US strain. 120 females were taken from the rearing cages and individually put in Petri dishes (100x20 mm) for observations. The cover was lined with 0.5 mm mesh nylon cloth. A disc of nylon with a wrinkle in its middle, used as an artificial egg-laying substrate, was humidified and laid on the bottom of the Petri dish. Macro-photographs of females were taken in the early afternoon before they lay eggs and during the early evening when the females were laying eggs.

Morphological studies

Morphology of the ovipositor was observed after dissections under binocular microscope in order to set informations to orient this organ for scanning electron microscopy observations.

Preliminary studies of ovipositor and leg sensilla were performed by Scanning Electron Microscopy. The ovipositor was studied with a Jeol JSM 840 scanning electron microscope in the entomology laboratory of the National Museum of Natural History, Paris.

The legs were studied with a Phillips 525M scanning electron microscope in INRA Versailles centre. Dorsal and ventral views of extruded ovipositors, fore- middle- and hind-legs were methodically observed. The study was mainly focused towards seeking gustatory-like sensillae on the parts of the body that were formerly showed as implicated in the egg-laying behavioural events.

Field studies of the female distribution on the maize plant throughout the day and the season

Observations were carried on in a 30 ha field situated in Kardoskut (Csongrad County), southern Hungary during July and August 2001. The population of *D.v.v* issued from a 3 years continuous maize field was quite important compared to the nearby maize fields. The corn hybrid was Marista and neither insecticides nor herbicides were applied in the field during the season. There were many weeds in the field such as *Ambrosia artemisiifolia*, *Amaranthus retroflexus* and *Echinochloa crus-galli*.

A 96 m-sided square divided into 64 of 12 m-sided squares was designed in the middle of the 30 ha field. The interest of a little square in the middle of the field was to limit any influence of the female inter-fields migration within the day.

Four consecutive plants were randomly chosen in each plot. 256 plants were observed. Three 2-hour long observation periods spaced by 3-hour intervals were performed on one clay. Females were counted and located on the plant. Walking and resting were noted as well as the organs of the plant, such as upper and under leaf side, stalk, husks and silks, where the females were located when observed.

Results

Behavioural observations on artificial substrate

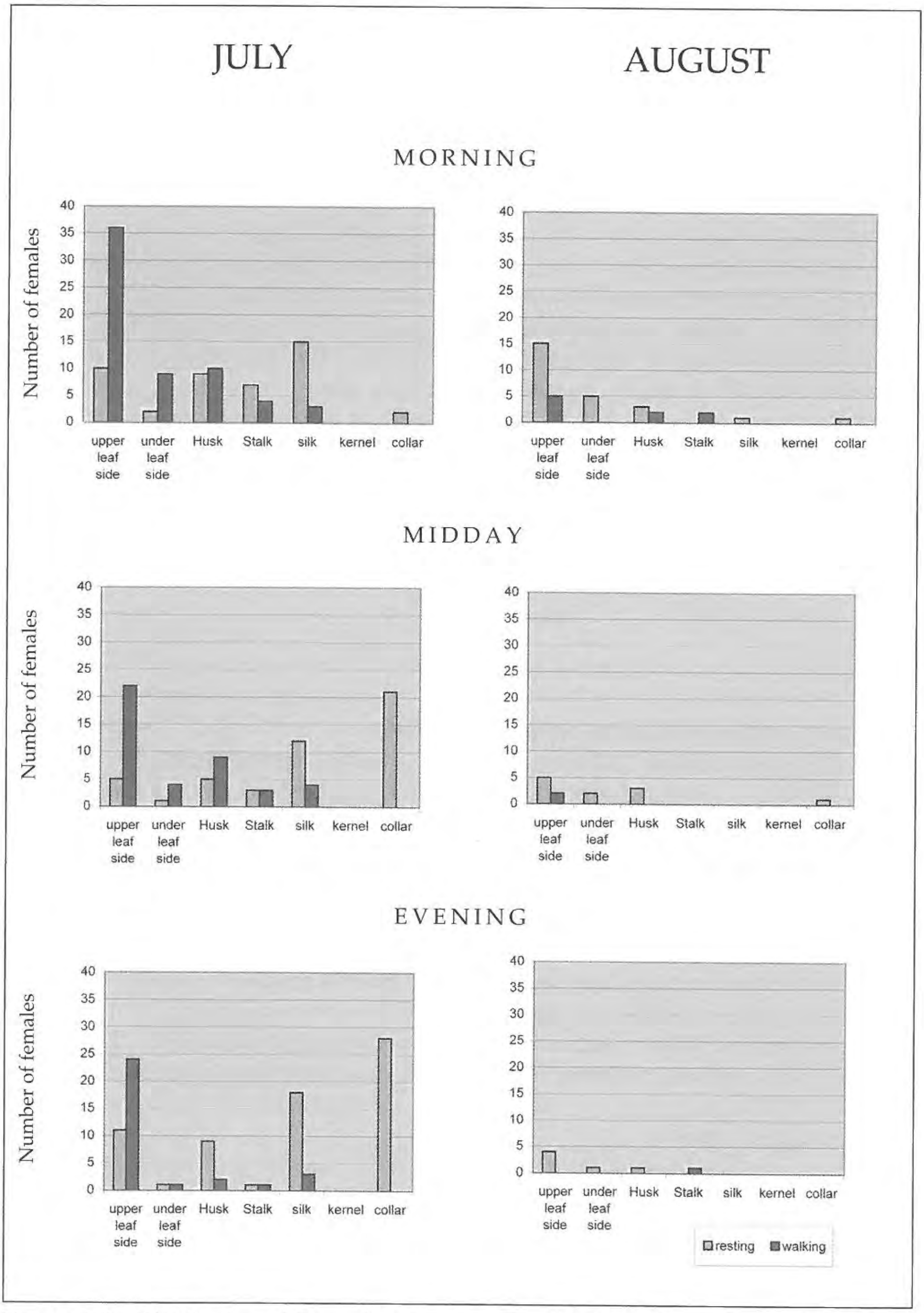
During the observations, different behavioural events were noticed.

Immediately after being introduced in the Petri dishes, the females started to walk everywhere in the boxes. After less then 30 mn, they began to scan the substrate. The ovipositor was completely extruded so that it touched the substrate (*figure 1*). Then it was partially retracted and bent so that the mid-dorsal part was in contact with the substrate (*figures 2* and *3*). Finally females invaginated the whole ovipositor. They repeated this sequence everywhere in the Petri dishes.

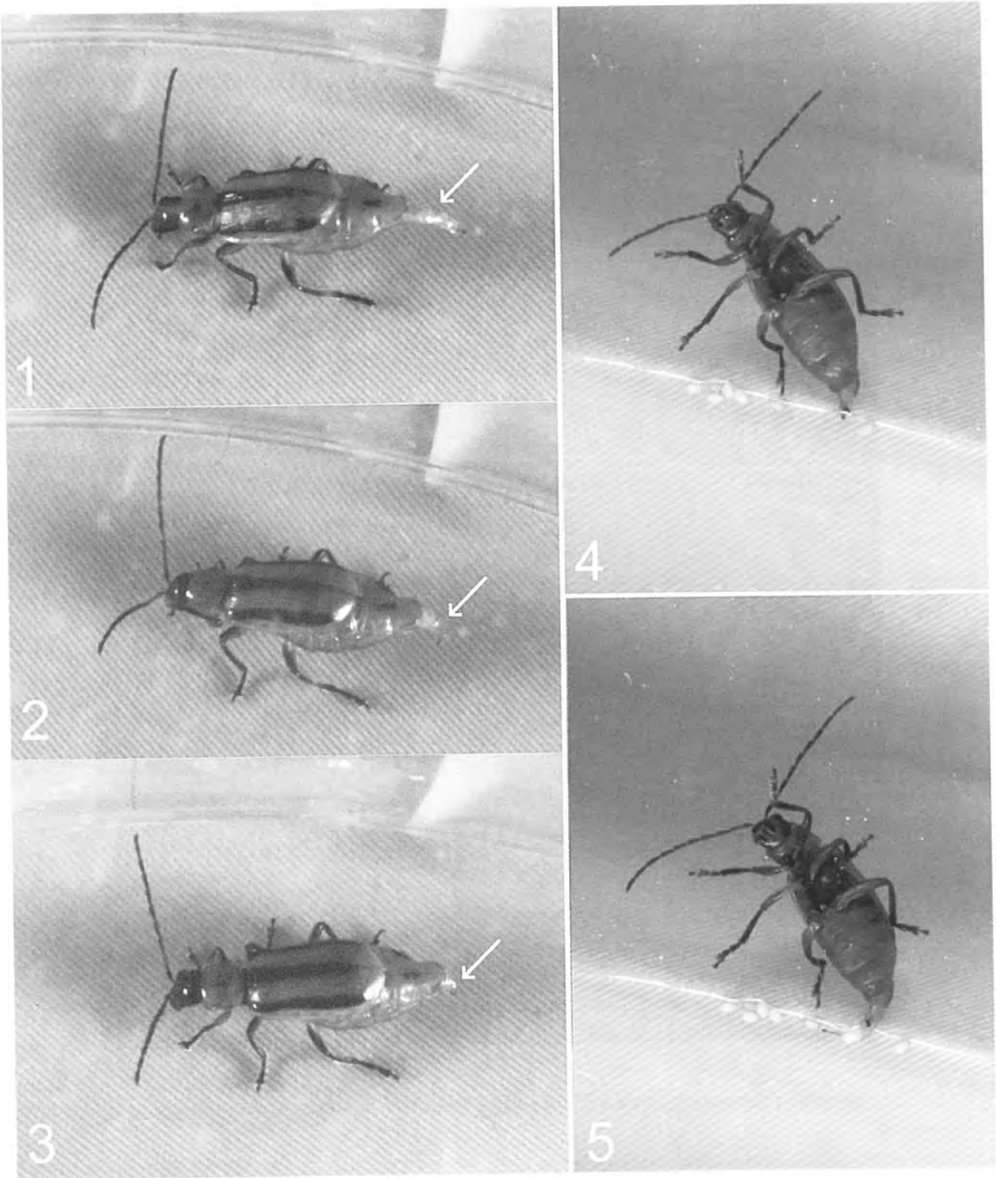
After two hours in the Petri dishes, they stopped scanning the substrate and used their ovipositor in a different way than when they were scanning the nylon. They extruded their ovipositor while introducing it between the humidified nylon disc and the Petri dish bottom where they laid eggs all around the fringe (*figures 4* and *5*).

Morphological studies

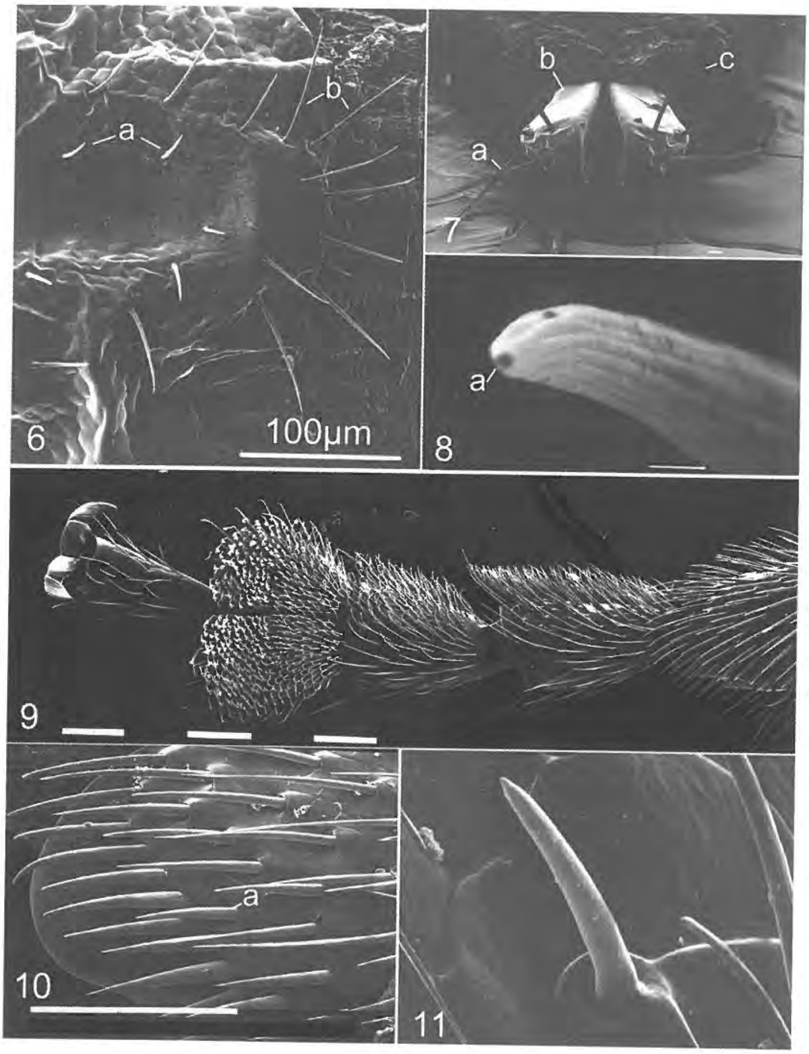
Preliminary microscopic studies of the ovipositor showed sensilla. Between 15 and 20 were found on the mid-dorsal sclerified part of the ovipositor (*figure 6*). This part is in contact with the substrate when the females bend it toward the substrate, and when the females lay their eggs. At least 5 of them appear as gustatory-like sensilla (*figures 6* and *8*). On the terminal sclerified style of the ovipositor 14 to 16 sensilla could be seen (*figure 7*). They all looked like mechanoreceptors of trichodea type. Concerning the legs, gustatory-like sensilla were found on tarsi and tibia (*figures 9*, *10* and *11*).



Figures 1-6 - Distribution of *D.v.v.* females on maize (Marista) plants observed in 2001 in Hungarian field at three periods of the day and two growth stages (July: brown silk, August: maturity)



Egg-laying behavioural events observed in artificial conditions.
Scanning Behaviour:
Fig. 1 - The female is extruding its ovipositor. The extremity of the ovipositor touches the artificial substrate
Fig. 2 - The ovipositor is curved and partially retracted
Fig. 3 - The terminal part of the ovipositor is invaginated. The mid-dorsal sclerified part of the ovipositor is in contact with the substrate. Arrow: mid-dorsal sclerified part of the ovipositor
Egg-laying behaviour:
Fig. 4 and 5 - Ventral view of a female laying eggs under the wrinkled nylon cloth disc



Scanning electron microscopy of *D.v.v.* ovipositor and legs.
Ovipositor:

Fig. 6 - mid-dorsal sclerified part of an extruded ovipositor. a: gustatory like sensillae; b: mechanoreceptors

Fig. 7 - Front view of the terminal part of the ovipositor. a: mechanoreceptors; b: sclerified terminal part (style); c : unsclerified part. Scale bar = 10µm

Fig. 8 - detail of the extremity of a gustatory like sensillum of the mid-dorsal sclerified part (fig. 6). Scale bar = 1µm

Legs:

Fig. 9 - Ventral view of tarsi and extremity of the the tibia (on the right) of a hind-leg. Scale bar = 100µm

Fig. 10 - detailed ventral view of the extremity of a tarsus. a: gustatory-like sensillae. Scale bar = 100µm

Fig. 11 - detail of a gustatory-like sensillum on the tibia

Field studies of the female distribution on the plant during the day and the season
The number of females walking and resting on the plant varies between July (brown silks) and August (maturity). In July the females walk more than they rest on every parts of the plant, except on the collar where they cannot walk but only hide themselves. The number of females observed on the plants was roughly constant throughout the day. It seems that the variation comes almost only from the increasing number of females hiding themselves in collars. In August, less females are present in the field. The proportions of walking and resting females is reversed compared to July. Females walk less and rest more in August. The number of females decreases throughout the day and no females hide in the collars.

CONCLUSIONS

Behavioural events which were observed in artificial conditions are scanning of the substrate which is not always followed by egg-laying, and egg-laying which is always preceded by scanning. The ovipositor is used to scan the substrate. The legs and especially the terminal part of the tibia and the tarsi are always in contact with the substrate. Observed by SEM, many different types of sensilla were found on both organs. A few among them seem to be gustatory sensilla. Further transmission electron microscopy and electrophysiological records are planned. These experiments are needed to confirm that these sensilla are able to detect the corn plant compounds.

Field observations confirmed that the females are very often in contact with the corn plant, even when they are not laying eggs. They walk and rest on leaves, husks and silks, and mainly this added to the fact that *D.v.v.* egg-laying is influenced by leaf surface washings. Washings also support to the hypothesis that chemical cue contacts are rather important in the *D.v.v.* egg-laying behaviour.

Acknowledgements to U. Kuhlman and St. Toepfer from CABI Bioscience Centre Switzerland and to other co-workers of the European project present in Hungary.

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INFLUENCE OF BIOCHEMICALS PRESENT ON MAIZE LEAF SURFACE ON *DIABROTICA VIRGIFERA VIRGIFERA (D.V.V.)* EGG-LAYING

***Sylvie Derridj*, *Ingrid Arnault*, *Franck J. Muller*,**
INRA, Unité de Phytopharmacie et des Médiateurs Chimiques,
Route de St Cyr, 78 026 Versailles, France

Abstract

The ovipositional habits of an insect are critical to the survival of the progeny. During the period of egg-laying, *D.v.v.* females may be present in the maize fields without feeding. Their abundance in fields follows the maize growth stages. The behavioural events leading insect to egg-laying are mediated to a large extent by chemical cues. Maize volatiles emitted by silks, kernel, husk attract *D.v.v.* females. Chemical stimuli, which induce *D.v.v.* acceptability of plant and laying eggs at its bottom, are not known.

Our hypothesis is that plant metabolites present on the maize surface through leaching could stimulate oviposition of *D.v.v.* and explain maize growth stage discrimination.

We collected maize surface water leachates from hybrid Borbala grown in green houses at different growth stages on several organs. Soluble carbohydrates and free amino acids were analysed. Leachates were tested on oviposition in no choice condition on single wild Hungarian females. Oviposition substrate was nylon cloth impregnated with maize leachates and dried in air flux.

Quantities and proportions of primary metabolites on the maize surface discriminate the growth stages, and also the different organs from each other's. Maize leaf surface leachates stimulate *D.v.v* oviposition. The stimulation is correlated with global metabolite composition. Reproductive stages of maize are particularly stimulant and young vegetative stages are deterrent.

Still no firm conclusions can be drawn about the activity of each primary metabolite, and its influence when isolated or associated in a pattern.

Key words: sugars, amino acids, egg-laying, plant surface.

INTRODUCTION

Maize rootworm *Diabrotica virgifera virgifera (D.v.v.)* has been known as the major pest of continuous maize in the mid-west of United States. Maize which is chosen for egg-laying permits development of the progeny. So the traditional management strategy has been crop rotation to break the biological cycle of the insect by avoidance of the larvae to found the good nutrients. However recently population has adapted to the rotation with soybean and females lay their eggs in soybean fields. To estimate the danger for different European crops to be infested by *D.v.v.* and to use the best crop rotation as management, the understanding of egg-laying behaviour and stimuli which influence it, are necessary.

The underlying mechanism of why this insect lays more eggs in maize fields and in older growth stages is unclear. Observations were limited to attraction for females of different part of the maize plants. The host plant selection for insects to lay eggs involves a final step governed by cues from non volatile plant chemicals. The plant acceptance by females can be governed by primary metabolites, which are on plant surface and used as kairomones. These substances were already observed as egg-laying stimulant for *Ostrinia nubilalis* (Derridj *et al.* 1989) and *Cydia pomonella* (Lombarkia and Derridj 2001). *Delia radicum* and *Psila rosae* takes chemical information on plants by running over the leaves before laying eggs in the soil at the base of cruciferous plant for the first, or at the base of carrot, celeriac, parsley for the second (Hurter *et al.* 1999, Degen *et al.* 1999). *D.v.v.* walks, runs, stands stationary over the whole plant throughout the day and mostly on leaf surfaces during the egg-laying season period. We hypothesised that biochemical stimulation leading to plant acceptance and egg-laying is taken by contact on the plant surface and that primary metabolites are the cue contact. We analysed primary metabolites present on the maize plant surface, at several growth stages and on different organs. Then the influence of water-soluble maize leaf washings was tested in laboratory on wild Hungarian *D.v.v.* egg-laying.

MATERIAL

Plants

Maize hybrid Borbala was grown in green houses from July to October 2000. Seeds were first put in small pots with earth and then at the 3-leaf stage were transferred in larger plastic pots of 7 litters. Mineral solutions from Duclos added with «Kanieltra 10 Fe» were used.

Growth stages analysed were 8-9 leaf (V(8)), late whorl (V(12-13)), early tassel (V(14-15)), late tassel (without pollen, VT), beginning maturity fresh silk (R3) and late maturity brown silk (R5).

Insects

Insects were collected in maize fields in Hungary (Kardoskut) from 20 to 27 July 2001 when the maize plants were at the young silk stage. 500 insects were gathered in Bugdorm insect cubic cages of 30 cm side and fed the first three weeks with young maize cobs and nutrient dry diet, then only the last one was used from 20 August at the beginning of the bio-tests. About 10 cages were kept at 24°C, 16:8 photoperiod, 65% H.R.. Dark period began at 2 P.M. 4 fluorescent tubes illuminated 1 m^2 area work plan. Bio-tests were carried on in the same chamber.

Material for bio-tests

Cylindrical transparent plastic boxes of 6 cm diameter and 10 cm high were lined on its sides and bottom with nylon clothe of 0.5 µm mesh. The piece of nylon at the bottom of the box was wrinkled in its middle.

Collect of bio-chemicals on the plant surface

Water-soluble substances were collected by spraying pure water on plant surface by the method described in Fiala *et al.* 1990. The leaves, stalks and husks were washed separately plant by plant. Four plants were sampled and constituted 4 replicates. Leaf washings used to test insect egg-laying were collected on 5 to 8 plants according to the growth stage which represented between 54,000 and 81,000 cm^2 of sprayed leaf surface. This was done at 4 P.M. solar hour and lasted maximum 2 hours and was repeated several consecutive days if necessary. All the leaves of the plants were sprayed on both sides and only the parts of the plants where the insect could access were sprayed. After cutting off the leaves, wounded parts were hidden by paraffin liquid at 40°C and aluminium paper and the stalk are washed. Water solutions were filtered (0.2 µm) and kept at – 80°C until evaporation at 40°C until dryness and then used for chemical analyses.

Chemical analyses

Analyses of trimethylsilyl (TMS) derivatives of sugars and polyols were done by gas chromatography associated to mass spectrometry GC/MS.

Preparation of the TMS derivatives:

Sugars and polyols in dehydrated residues were derivated in TMS derivatives with BSTFA. Standard compound (Phenyl ß-D- glycoside) was added prior to trimethylsilylation.

Separation of TMS derivatives:

The GC/MS system consisted of gas chromatograph 5890 and a quadripole mass spectrometer 5971 (Hewlett Packard). Mass spectrometer was used in scan mode. Gas chromatograph was performed on a 30 m RTx-5Sil MS column with 0.25 µm inner diameter and 0.25 m film thickness (Restek). Injection temperature was 250°C. The temperature program begin at 100°C to 280°C by a 6°C oven temperature ramp and a 5 min isothermal heating at 280°C. The spectral library used was Wiley.

Analysis of *t*-butyldimethylsilyl (TBDMS) derivatives of amino acids by GC/MS:

Chromatographic system is the same as for chemical analyses of TMS sugars and polyols derivatives except that mass spectrometer was used in SIR mode.

Preparation of the TMS derivatives:

Amino acids of dehydrated samples were purified by ion exchange chromatography with an ion exchange resin (AG 50W-X8) and then derivated in TBDMS with MTBSTFA. Norleucine was used as internal standard.

Separation of TBDMS amino acids:

Injection temperature was 100°C to 255°C and to 295°C by a 4°C/min-oven temperature ramp and 10°C/min respectively with a 5-min holds time at 295°C. The injector was heated at 250°C.

Bio-tests

Leaf washings were divided in several aliquots then dried by deep freezing. Each aliquot was used for one day. Nylon clothes were dipped in diluted solutions reproducing the quantities found on the leaf surfaces on nylon clothes, and dried horizontally in a laminaris box in the climatic chamber during half an hour. The nylon clothes put into a box, then a single female was introduced. That was done at 10 A. M. and female left during 24 h after which eggs were counted. Eggs were deposited at the bottom of the box where there was a piece of humidified nylon cloth not impregnated with leaf washings which had no contact with the impregnated nylon cloth. 80 to 100 females were tested on each of the 4 growth stages: 8-9 leaf (V(8)), early tassel (V(14-15)), late tassel (without pollen, VT), and beginning maturity fresh silk (R3).

Results

Primary metabolites present on maize plant surface

All the free amino acids and the three soluble carbohydrates fructose, glucose, and sucrose are present on the plant surface. Sugar alcohols such as quebraquitol, sorbitol, myo-inositol were also detected and malic acid. Their quantities and proportions could characterise plant growth stages and organs.

Proportions on leaves and stalks of the 3 soluble carbohydrates discriminate the early growth vegetative stages from the elder reproductive ones (*figure 1*).

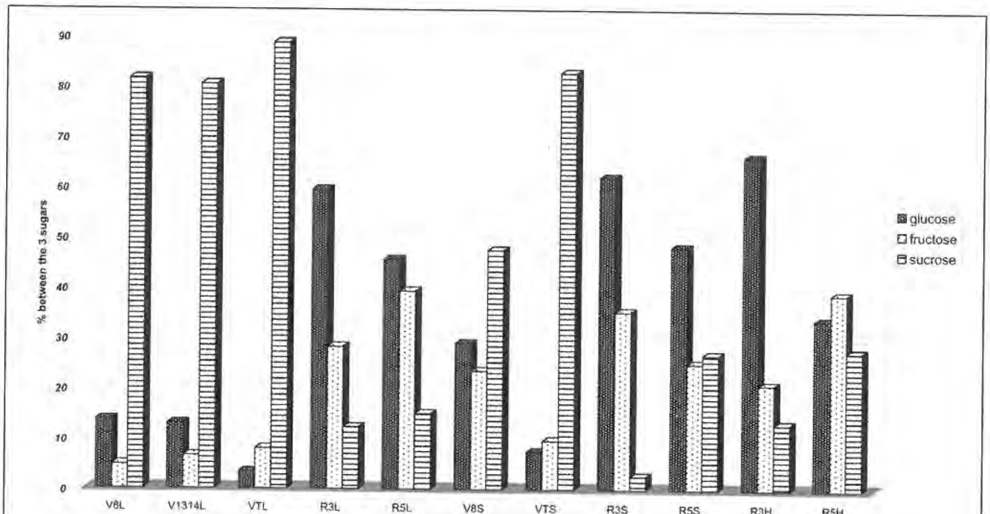

Figure 1 – Proportions of the three soluble carbohydrates on maize (Borbala) organ surface (L: leaf, S: Stalk, H: husk,) at different growth stages

Sucrose is dominant until VT and then glucose during grain maturity. Quantities of soluble carbohydrates and sugar alcohol's vary progressively from

V(8) to R5, quebraquitol diminishes and sucrose and myo-inositol increase. Total of free amino acids present on the leaf surfaces also increases throughout maize growth (*figure* 2). VT is the growth stage which showed the highest quantities of metabolites on leaves and particularly sucrose, quebraquitol, sorbitol, mannitol and amino acids like metionine, threonine, aspartic acid, glutamic acid and arginine.

Leaf surfaces are the poorest part of the plant compared to stalk and husk, which are especially rich in malic acid and sucrose (*figure* 3). Proportions of soluble carbohydrates and free amino acids are rather similar on leaves and stalks.

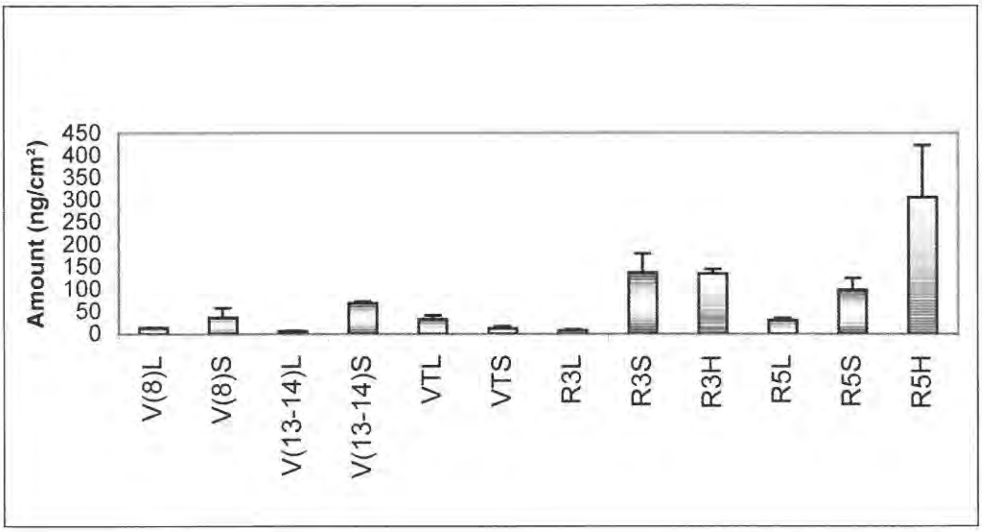

Figure 2 – Total amount of amino acids on Hungarian corn leaf (L), stalk (S) and husk (H) surface at phonological stage V(8), V(13-14), VT, R3 and R5

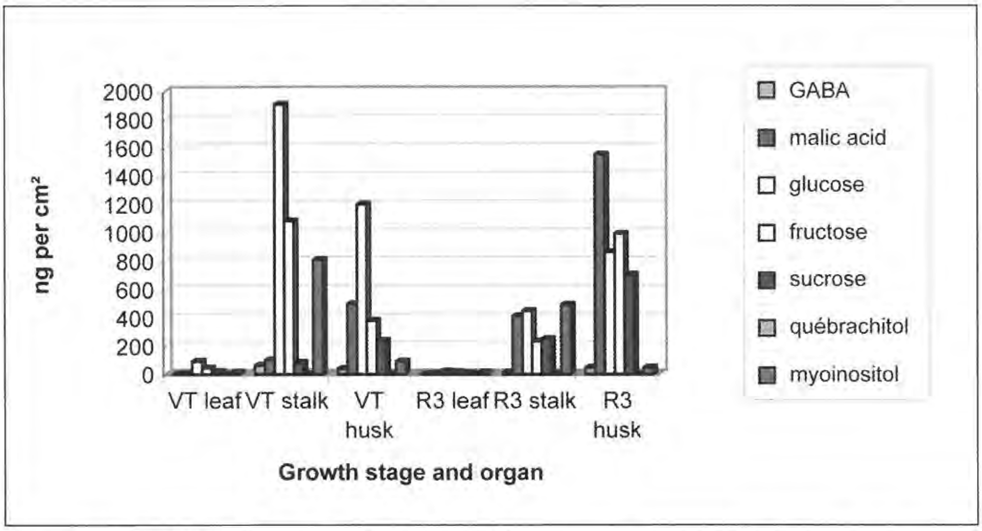

Figure 3 – Quantities of some metabolites on different maize organs at two growth stages

Relationships between leaf washings and egg-laying

Throughout the period of test, egg-laying on control (nylon clothes dipped in pure water) did not change significantly, mean of eggs per female within 24 h was 33.34 (s.e. 27.8). Numbers of females accepting to lay eggs varied from 10 to 60%. Means were calculated on 18 to 29 egg-laying females (except for V(8) on 11 females).

We observed on V(8) washing a reduction of egg-laying female numbers and of eggs laid per female (*figure 4*). On the contrary VT washing is highly stimulant for egg-laying, the number of eggs was multiplied by 6 as compared to V(8) and twice as to control. V(14-15) effect was similar to control 's. R4 was intermediate between VT and V(14-15) which could be very stimulating according to the period of test (*figure 4*).

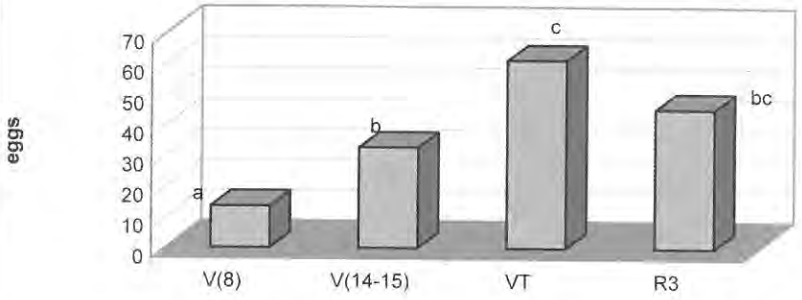

Figure 4 – Number of eggs laid per female within 24 h on nylon cloth impregnated with leaf surface washings of different maize (Borbala) growth stages

Probably several substances are acting together and we are in front of several phenomenon together on females: deterrence at the early stage and positive egg-laying stimulation at the stage VT. In that way the PCA (Principal Component Analysis) is of rather help by considering all the substances together of the different growth stages. The PCA in which variables are quantities of leaf surface amino acid and soluble carbohydrate gives results in the same way as the insects egg-laying response to washings. There is a discrimination between V(8-9) on one side, VT on an other side and V(14-15) and R3 in a third side (*figure 5*). Axe 1 represent 46.5% of the variation and axe 2 : 28.7%. GABA, glutamic acid, lysine, arginine, fructose, glucose, mannitol, sorbitol, and myo-inositol contribute the most to the definition of the axe 1, and alanine, glycine, methionine, serine, phenyl-alanine, ornithine to the axe 2.

Correlation between single substances and egg-laying were less demonstrative and the best ones were obtained with glutamine and sucrose (R^2 = 0.87, and 0.74 respectively).

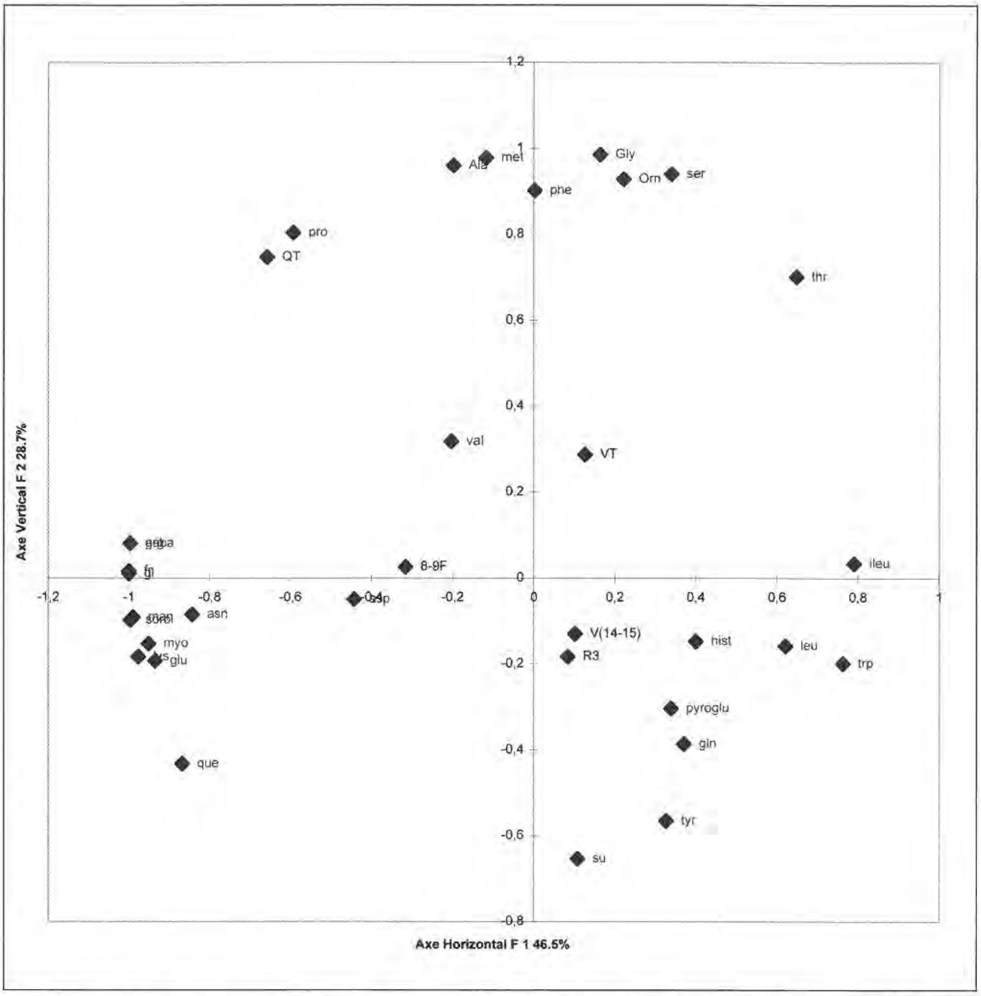

Figure 5 – PCA discrimination of several maize (Borbala) growth stages by quantities of free amino acids and soluble carbohydrates present on leaves

Conclusions

Primary metabolites present on the maize surface discriminate the stages and organs. The insect egg-laying is stimulated by leaf washings including these substances. Young growth stage is deterrent and the VT is specially egg-laying stimulant. The insect localization in maize fields and gustatory like sensilla on tarsae and ovipositor corroborate the hypothesis of chemical plant surface stimulation on egg-laying.

Experiments on artificial substrates with synthetic substances, and electro-physiological recordings will permit to define the active components and their role.

Acknowledgements to Philippe Couzy technician of the INRA laboratory and co-workers of the European project present in Hungary in 2001.

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ALTERNATIVE FOOD RESOURCES FOR ADULT *DIABROTICA VIRGIFERA VIRGIFERA* IN SOUTHERN HUNGARY

Joachim Moeser, Stefan Vidal,
Institute for Plant Pathology and Plant Protection,
Georg-August-University Goettingen, Grisebachstr. 6,
37077 Goettingen, Germany; e-mail: jmoeser@gwdg.de

Abstract

The study presented here gives detailed information on the feeding biology of adult *Diabrotica virgifera virgifera* in Southern Hungary. More than 600 beetles were sampled in two categories of maize fields: fields with a low and a high weed abundance. The maize phenology was recorded in a weekly interval. Furthermore the weed flora was identified and its abundance was estimated by using a transect method. The beetles were treated in a quantitative and qualitative pollen analyses process. The following analyses revealed a feeding biology which is dependent from time and habitat (weedy vs. non-weedy) and additionally shows a possible preference for certain weeds.

Key words: *Diabrotica virgifera virgifera*, feeding biology, host plant interaction, alternative pollen resources, pollen analyses.

INTRODUCTION

Since *Diabrotica virgifera virgifera* LeConte (Galerucinae, Chrysomelidae) was introduced into Europe in the beginning of the 90's, this maize pest is continuously spreading and invading new areas and habitats in Central and Southeast Europe. Because the European agro-ecosystems present a new ecological setting for this beetle, intensive studies are required on possible changes in the ecology and behaviour of the Western Corn Rootworm (WCR) adults which are believed to be specialized maize pollen feeders (Chiang, 1973). The study presented here gives information on the feeding behaviour and use of weeds as alternative pollen resources of adult WCR on the basis of gut content analysis.

MATERIAL AND METHODS

This study was performed in Southern Hungary (Csongrad County) during a 10 week period from the end of June to mid September 2000. Beetles were collected in fields with different degrees of weed abundance to estimate the use of weed-pollen by WCR; 6 fields with a low abundance of weeds and 6 fields with a high abundance of weeds were selected. Beetles were also collected directly from weeds when encountered there. The maize phenology was recorded once a week to obtain information on availability of different maize tissues or organs like pollen or silk. Furthermore the abundance and diversity of flowering weeds was recorded weekly along a 20 m transect which was randomly placed inside each maize field. More than 600 beetles were used for a qualitative and quantitative pollen analysis where the insects were acetolysed so that finally only the pollen remained. The acetolysation process comprises mainly the use of 9:1 mixture of acetic anhydride and sulphuric acid respectively. The insect was dissolved and the pollen residue was concentrated and cleaned by centrifugation and washed once with glacial acetic acid and several times with distilled water. The pollen grains were counted and identified using a previously established reference collection of all weeds encountered inside and along the margins of maize fields.

RESULTS

Adult *D. v. virgifera* were feeding on 19 different plant species (*table 1*), representing 73% of the total weed flora found in maize fields and along their margins in the study area. These 19 weed species were members of 9 different plant families, the Poaceae and Asteraceae being the most prominent by the numbers of species. These results were partly due to observations made in the field and in the laboratory as indicated in *table 1* or were obtained through the pollen analyses.

Table 1 - Plant species and plant organs used by WCR for feeding and source of information (direct observation or pollen analyses)

Family	Host plant species	Observation	Pollen Analyses	Plant organs affected
Poaceae	*Zea mays*	yes	yes	Pollen, kernel, leaves, silk
Amaranthaceae	*Amaranthus sp.*	yes	yes*	Pollen
Chenopodiaceae	*Chenopodium album*	yes	yes*	Pollen
Asteraceae	*Ambrosia artemisifolia*	no	yes	Pollen
	Cirsium arvense	no	yes	Pollen
	Helianthus annuus	yes	yes	Flower Petals, Pollen
	Sonchus asper	yes	yes	Pollen
	Xanthium strumarium	yes	yes	Pollen
Cucurbitaceae	*Cucurbita maxima*	yes	yes	Leaves, Pollen
Fabaceae	*Medicago sativa*	yes	yes	Leaves, Pollen
	unbekannte Fabaceae	no	yes°	Pollen
Malvaceae	*Malva sylvestris*	yes°	yes°	Flower Petals, Pollen
Poaceae	*Echinochloa crus-galli*	no	yes*	Pollen
	Setaria pumila	yes°	yes*	Pollen
	Setaria verticilaria	yes°	yes*	Pollen
	Sorghum halepense	yes°	yes*	Pollen
	Sorghum bicolor	yes°	yes*	Pollen
Scrophulariaceae	*Linaria vulgaris*	no	yes°	Pollen
Solanaceae	*Datura stramonium*	yes°	yes°	Pollen
N families = 9	N species = 19 (73%)			

° = rare event; * = impossible to distinguish within group

The use of the different plant species as pollen resources was not always depending on the abundance of these host plants in the field (*figure 1*). The amount of *Amaranthus* sp./*Chenopodium* sp., *Ambrosia artemisiifolia* or *Helianthus annuus* pollen found in the beetle guts was not proportional to the abundance of these weed species in the maize fields.

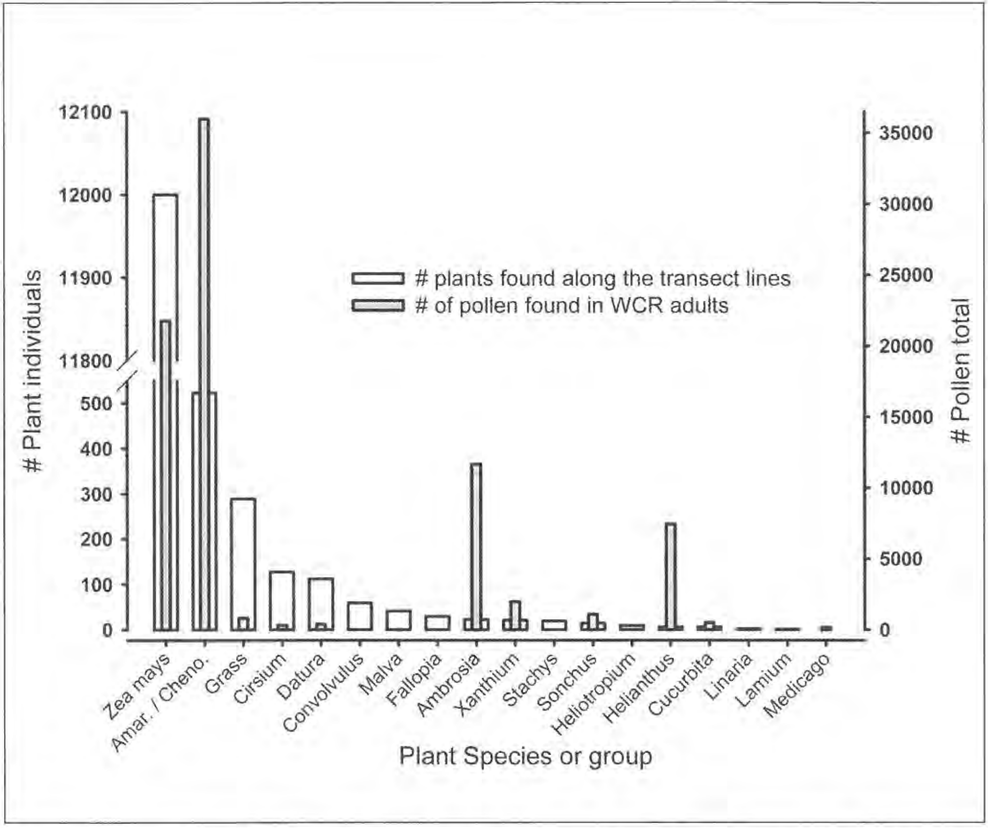

Figure 1 - Number of maize and weed plants found along the transects in the fields in Southern Hungary (left y-axis) and the number of pollen of each plant species found in the guts of the beetle samples in these fields (right y-axis)

The amount of pollen found in the gut of the beetles depended primarily on the availability of pollen, following the changing maize phenology over time. In the same way that the availability of maize pollen decreased, the amount of maize pollen found inside the beetles decreased (*figure 2*).

As the availability of maize pollen decreased the number of *Amaranthus* sp./ *Chenopodium* sp. pollen increased (*figure 3*). In this case, a clear habitat dependency could be observed. The number of *Amaranthus* sp./ *Chenopodium* sp. pollen was significantly higher in beetles from fields with a higher abundance of weeds. *Amaranthus* sp. and *Chenopodium* sp. pollen were treated as a complex, because the pollen could not be clearly distinguished under the light microscope.

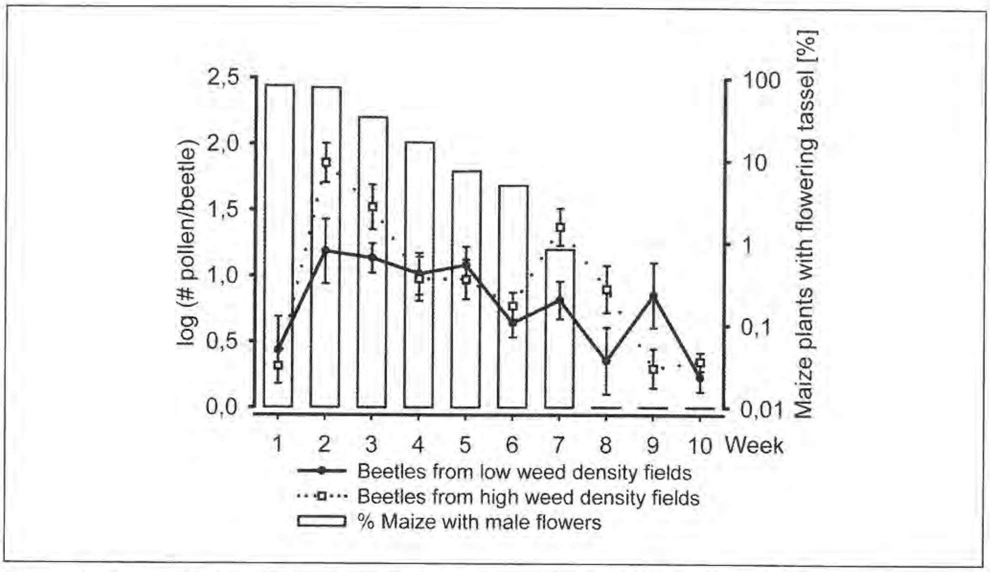

Figure 2 - Log-transformed number of *Zea mays* pollen per WCR adult (left y-axis) compared to the availability of maize pollen (right y-axis; note log-scaling)

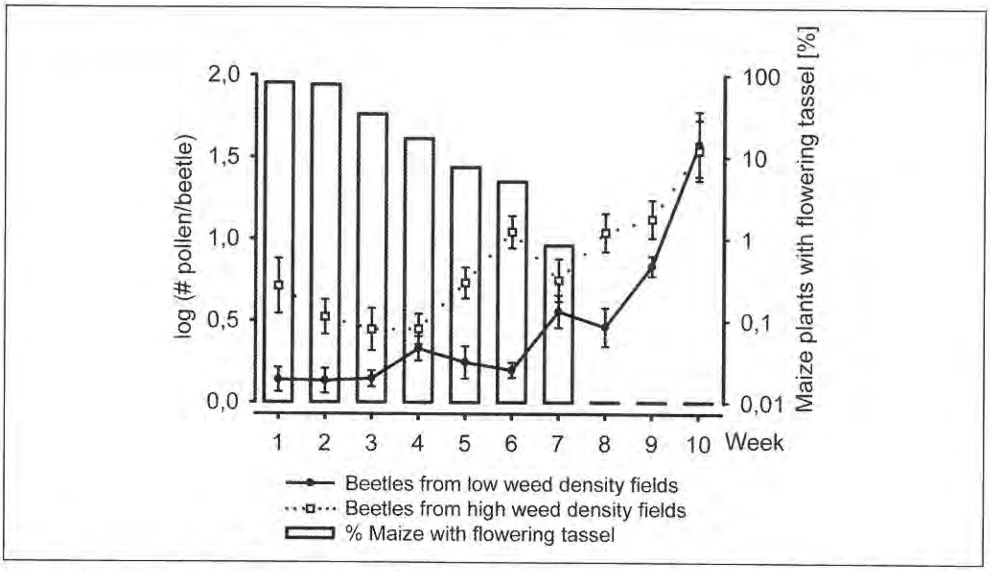

Figure 3 - Log-transformed number of *Amaranthus* sp. / *Chenopodium* sp. pollen per WCR adult (left y-axis) compared with the availability of maize pollen (right y-axis; note log-scaling)

Looking at the use of *Ambrosia artemisiifolia* no differences regarding the number of pollen found inside the gut could be observed (*figure 4*). Beetles from fields where this weed was not at all or only in small numbers present, showed the same number of pollen in their gut, as did the beetles from fields with a high abundance of *A. artemisiifolia*.

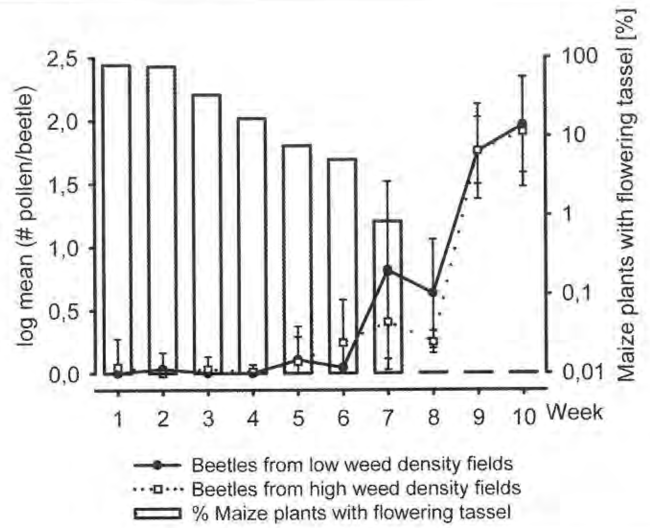

Figure 4 - Log-transformed number of *Ambrosia artemisiifolia* pollen found inside the gut of adult WCR beetles (left y-axis) on the background of the availability of maize pollen (right y-axis; note log-scaling)

Discussion

The feeding biology of *D. v. virgifera* in Southern Hungary is more complex than expected from the literature (Ludwig & Hill, 1975) and depends on various factors:

Time: as maize pollen becomes rare towards the end of the flowering period, other pollen resources are used more frequently. Most flowering plants present in the study area served as alternative pollen resources. Thus the nutritional shortage is compensated by weeds, resulting in another factor affecting feeding biology.

Habitat: our data clearly demonstrate that WCR from fields with a high abundance of weeds uses significantly more pollen of *Amaranthus* and *Chenopodium* than beetles from non-weedy fields. The higher abundance of *Amaranthus* sp./ *Chenopodium* sp. pollen leads to a more frequent use of these weed species, thus the different habitats influence the feeding biology of adult WCR. Our results concerning the use of *Ambrosia* pollen further lead to another factor that may influence feeding behavior.

Host plant specificity: although *Ambrosia* was either absent or only present in very low densities in non-weedy fields, the number of Ambrosia pollen found in WCR from non-weedy fields is not lower compared to beetles from weedy fields, where this plant was more abundant. This may be the result of a preference of this beetle towards this plant species. One possible explanation for this preference is the flowering period of *A. artemisiifolia*. At the end of the study period, this plant is one of the few remaining flowering plants in the study area.

In general the feeding behavior of WCR seems to be influenced by the density of possible host plants to a lesser extend. Maize pollen is not the most common pollen taken into account absolut pollen numbers. The principal pollen source is *Amaranthus* sp./ *Chenopodium* sp. although the density of these weeds was lower than the density of maize plants. This may be due to longer flowering times. While maize pollen was only available in the early summer, pollen of these weeds were present all over the study period. That way the number of these pollen could accumulate to such high quantities. A further example that the feeding biology of WCR is not depending on the host plant density is the *Ambrosia* case. Although *Ambrosia* plants were relatively low in abundance (9^{th} rank), its pollen is ranked three in abundance. Thus, the use of *Ambrosia* as a pollen source was not correlated with its abundance.

Acknowledgements to I. Hatala-Zseller and the staff at the Plant Health and Soil Conservation Station of Csongrad County for technical assistance.

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LIFE TABLE OF *DIABROTICA VIRGIFERA VIRGIFERA*

Stefan Toepfer ***Ulrich Kuhlmann,***
CABI Bioscience Centre Switzerland - Rue des Grillons,1
2800 Delémont Switzerland - stoepfer@cabi-bioscience.ch

Abstract

The western corn rootworm (*Diabrotica virgifera virgifera*) is an exotic invasive species and offers the possibility of classical biological control through the search for effective natural enemies in the area of origin. Life tables will determine mortality factors acting on *D. virgifera* populations in Hungary and will provide information on the most suitable life stages (egg, three larval instars, pupae and adults) to be attacked by exotic biological control agents. Additionally, this three-year study investigates the fecundity of the beetles as well as their reproductive period in the laboratory and in the field (EU Project QLK5-CT-1999-01110). In order to determine mortality factors in the field, several sets of eggs were exposed to winter conditions in two years and recollected during following springs. Population density and mortality of each larval instar and of pupae were obtained by soil-root sampling in three fields (96 samples per week per field). Adult density was measured in emergence cages covering 70 plants per study area (0.4 ha). *Diabrotica* adults were collected in selected fields in Hungary, Croatia and Yugoslavia and kept in cages in order to determine parasitism. The population density of each stage was compared and mortality between each stage was calculated. In a second experiment to determine mortality factors, maize plants were artificially infested by L1 and then L2, L3, pupae and adults were recollected.

Highest mortality was found in diapausing eggs and in the first instar larval stage. There was some mortality observed in prediapausing eggs but lowest mortality was determined during hatching of larvae, during second and third instar larvae and in adults. Additional results are provided on the fecundity, longevity and reproductive period of the adults in the field, as well as on the occurrence of *D. virgifera* natural enemies in Hungary.

In conclusion, the key stages for exotic biological control agents are the late larval instars or the adults of *D. virgifera*. CABI Bioscience is conducting investigations on the search for parasitoids in the area of *Diabrotica*'s origin and will test the suitability of exotic parasitoids in Swiss quarantine facilities.

Work carried out within the framework of EU Diabrotica Project (QLK5-CT-1999-01110).

TWO YEARS OBSERVATIONS ON CORRELATION OF LARVAE DAMAGE OF WESTERN CORN ROOTWORM AND YIELDS

Ibolya Hatala Zsellér,
Plant Health and Soil Conservation Station of Csongrád County,
H-6801 Hódmezővásárhely, P.O. Box 99, Hungary

Endre Széll,
Cereal Research Non-Profit Company,
H-6701 Szeged, P.O. Box 391, Hungary

Abstract
As one of the most infested part in Hungary since 1996, the permanent trapping has been continued in four sites in county Csongrád. Observátions took place at the same fields in Csanádpalota, Hódmezővásárhely, Nagylak and Szeged using pheromone (CSALOMON) and yellow sticky (MULTIGARD and PHEROCON) traps.

Besides trapping of adults determination of larvae development was also carried out by weekly root observations.

On each field 10 emergence cages were set up above one corn plant to measure the number of beetles developed within one root system. In the end of the season the damage on roots were rating by the modified IOWA-scale, the yield of each plant was measured and correlation between adults developed in root system and yield production was measured.

Results of permanent trapping showed significant increase in area of Nagylak and Hódmezővásárhely this year. The population established on the same level in Szeged and less number of beetles was caught by the traps in Csanádpalota. Lodged plants were found only in Szeged experimental field.

Development and seasonal flight of WCR can be compared under rather different weather conditions based on data in 2000 and 2001. The weather in the vegetation season of 2000 can be characterised with extremely high temperature without considerable precipitation for a long period. Season of 2001 was less hot and regular precipitation was measured. In 2000 very early larva hatching was observed: on 25 May both L_1 and L_2 stages were found. This year first L_1 larvae were detected on 30 May. First males occurred 15 June in 2000 and ten days later, 25 June in 2001 in pheromone traps.

The number of emerged adults in cages was low in Csanádpalota and Nagylak observation fields in both years. In Szeged the number of beetles varied between 4 and 53 per plant in 2000. In 2001 number of adults collected in cages increased and varied between 28 and 218. Significant correlation was observed concerning the number of adults, the root damage and the weight of yield of plants.

Work carried out within the framework of EU Diabrotica Project (QLK5-CT-1999-01110).

IS THE WESTERN CORN ROOTWORM ADAPTING TO THE EUROPEAN CROP ROTATION SYSTEM? RESULTS OF A JOINT EUROPEAN TRIAL

József Kiss,
Khosbayar Bayar, Judit Komáromi, Szent István University,
Department of Plant Protection, H-2100 Gödöllő, Hungary

Jasminka Igrc-Barčić, Renata Dobrinčić,
University of Zagreb, Agricultural Faculty,
Svetosimunska 25. Zagreb, Croatia

Ivan Sivcev,
Institute for Plant Protection and Environment,
T. Drajzera 9. P.O.B. 936. 11001Beograd, FR. Yugoslavia

C. Richard Edwards,
Purdue University, Department of Entomology,
1158 Smith Hall, West Lafayette, IN 47907-1158, USA

Ibolya Hatala-Zsellér,
Plant Health and Soil Cons. Station,
H-6801 Hódmezővásárhely, P.O. Box 99, Hungary

Abstract

A three-year crop rotation trial was established in 2000 in Szeged in Southern Hungary (EU-5 project). This trial was included as an activity under the framework of the FAO WCR Network in 2001 and companion studies were initiated in Croatia and Federal Republic of Yugoslavia (FRY). The crops rotated with corn were corn, sunflower, soybean, and a cereal (winter wheat, oat, or spring barley). The presence of WCR was assessed and quantified by using Pherocon AM traps and emergence cages. In Hungary, results for 2000 showed small catches of WCR adults on Pherocon AM traps and no WCR adults in emergence cages in crops other than corn. For 2001, an increased number of WCR adults were caught on Pherocon AM traps in sunflower, soybean, and cereal (latter harvested) plots, in addition to significant numbers caught on traps in the corn plots. Some WCR adults were found in emergence cages in corn after soybean and cereal. In FRY, adult WCR population density was low (both on Pherocon AM traps and in emergence cages). Where WCR catches were recorded, these mostly came from corn. In Croatia, numbers of beetles caught on Pherocon AM traps in corn were high. Additionally, numerous WCR adults were trapped in crops other than corn. WCR adults in emergence cages were trapped almost exclusively from corn. The paper describes the methods and preliminary results.

Key words: Western Corn Rootworm, crop rotation, Europe.

INTRODUCTION

The spread and population build-up of the western corn rootworm (WCR) requires multiple management strategies to be developed for the control of this corn pest. Crop rotation is the primary non-insecticide control strategy used in the USA to control WCR larvae, which feed almost exclusively on corn roots (Branson and Ortman 1970). However, some entomologists in the USA (Edwards, 1996, Levine and Gray, 1996, Gerber *et al.* 1997) and in Serbia in Europe (Sivcev, 1999, pers. comm.) have observed economic WCR larval damage in corn following other crops such as soybean. Barna *et al.* (1998) further substantiated this loss of preference for egg laying in corn fields and noted significant adult activity in soybean fields in Indiana, USA. The egg laying that occurred in these soybean fields resulted in larval damage to the succeeding year's corn crop. Economic thresholds for WCR adults in soybean to predict subsequent damage to corn was established by Gerber *et al.* (2001). There is no relevant data available on how WCR will adapt itself to European crops, to what extent its egg laying preference might change, and which crops in rotation with corn, if any, might eliminate or greatly reduce WCR population build-up.

METHODS

A three-year crop rotation trial was established in 2000 in Szeged in southern Hungary as part of the EU QLK5-CT-1999-01110 project. The crops included in the study were similar to the ones normally grown with corn in many areas of Europe. Preconditions for the trial were as follows.

In order to obtain reliable data allowing proper statistical analysis, the rotation study was established in fields dedicated to this research activity for a period of 3 years. The study areas were noted as having significant WCR population densities, with corn as precrop. Soil conditions (heavy clay soils) favoured egg laying and larval development. Crops in the rotation study were not treated with soil insecticides so as to avoid WCR disturbance. For herbicides, application of post emergence products with less persistence and potential carryover were used. Regular production practices, as applied in the region for the targeted crops, were used so as to mimic those normally carried out by farming community.

Experimental layout

The layout was designed to allow for plot sizes large enough for reliable data to be generated within each plot. Six replications of each treatment were used for the study. The test site was about 3 hectares. The study area was divided into 2 sections (*figure 1*). One half was grown to corn, the other half was planted to strips of the various crops that were in rotation with corn (corn, soybean, sunflower, and winter wheat). Due to starting the study in the spring, oat was planted in the first year as the cereal crop instead of winter wheat. Winter wheat was then planted in the late summer/early fall for the next and subsequent growing cycles. The two sections of the test field will be yearly rotated for the duration of the study. Each plot where the various crops are planted were marked with permanent markers. The field was layed out in the shape of a rectangle. The size of the two sections that made up the study, field sections A and B, was at least 50 m x 217 m each. Distance between A and B was 9 m (thus a roadway to allow for movement and manuvering of machinery, etc.). The uncropped area on the outside perimeter (outer boundary) of A and B was also 9 m. Crops within replications were randomised, but the same crop from each of two replications could not border each other. Also, to reduce the chance of moving WCR eggs from one area of a field to another or from plot to plot, equipment was kept as clean as possible.

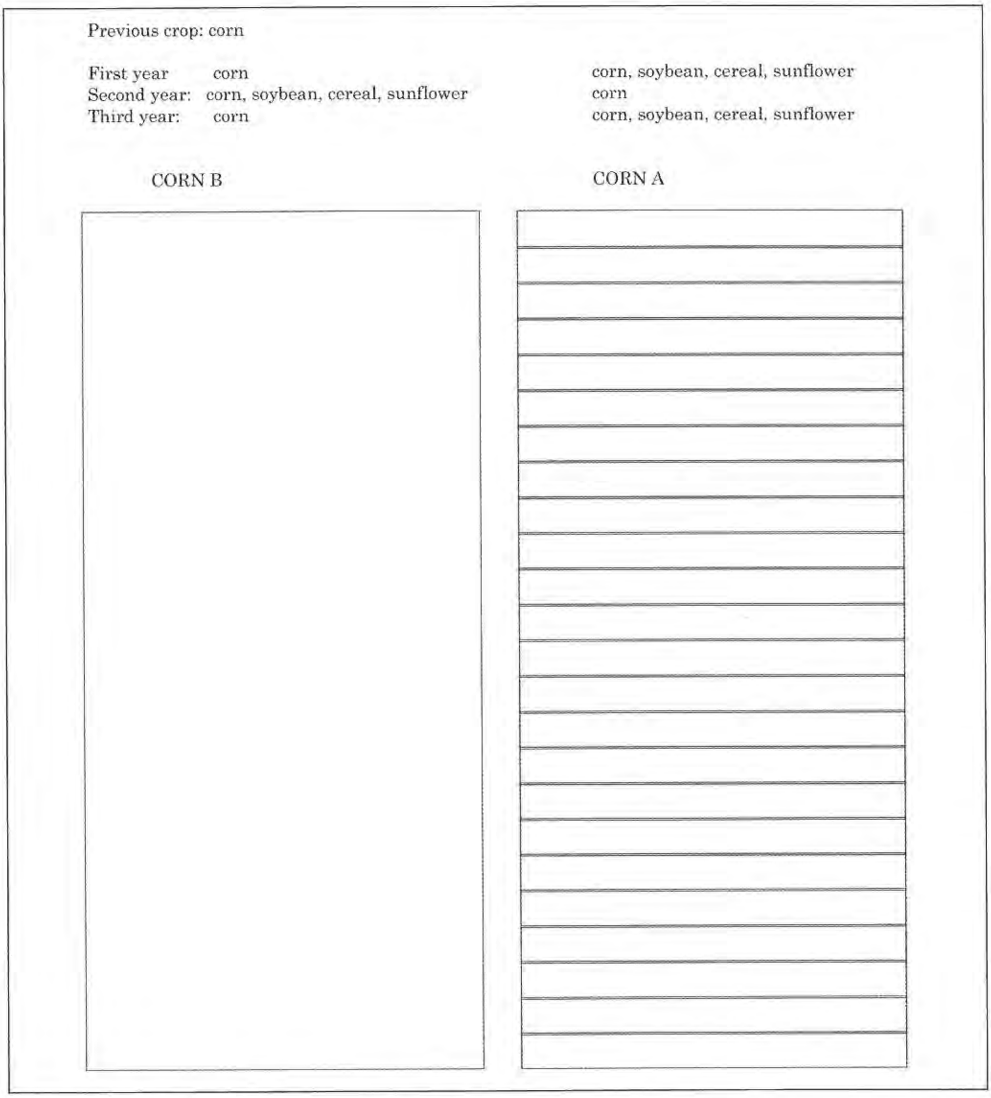

Figure 1 - Field layout for crop rotation trial

Plot layout

In order to properly place 3 traps in each plot, plots were arranged in an elongated, rectangular shape. Each plot was at least 8.6 m x 50 m, thus approximately 12 corn rows in the corn plot, as an example. There was approximately 1m distance between each plot. Three Pherocon AM yellow sticky traps were placed equadistant within each plot in the middle row with the first being about 7 m from the entrance of the plot and the last 7 m from the back of the plot (*figure 2*). Pherocon AM traps were attached to the stems of corn and sunflower and on a stake in the soybean and wheat plots, just above the soil surface. In a row next to the traps, 3 emergence cages were placed within 1-2 m of the Pherocon AM traps. Traps and cages in field B mirrored those in field A. As noted above, the exact location of the plots was marked so as to allow for the proper placement of future plots. The total number of Pherocon AM traps in each field (fields A and B) was 72 (144 total).

These traps were changed weekly and the number of WCR adults on the traps were counted and recorded. The emergence cages were equal in number (72 + 72 = 144) to the Pherocon AM traps. The presence of WCR was assessed and quantified by using the Pherocon AM traps and emergence cages from mid June to mid September.

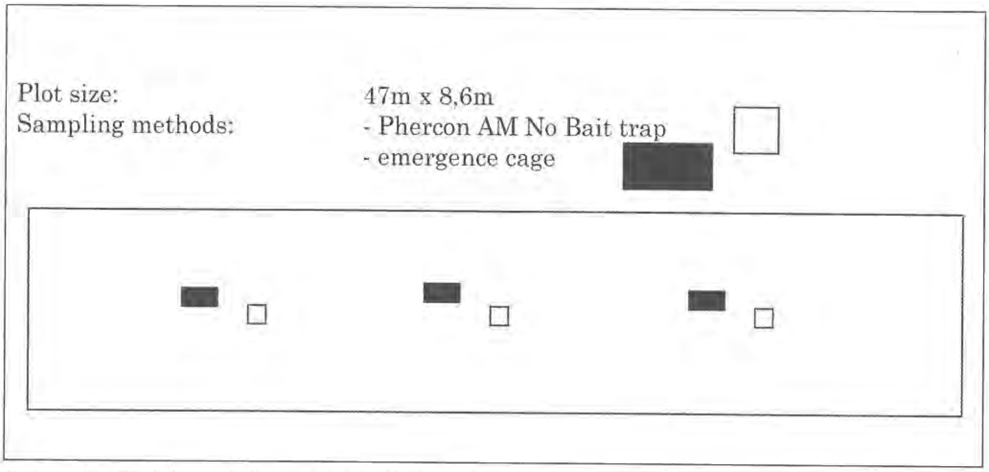

Figure 2 - Plot layout for crop rotation trial

Description of emergence cage

Each cage consisted of two parts - a wooden frame and a screen in the shape of a pyramid. Size of the frame was (inside dimensions): 1.25 m length x 0.4 m width = 0.5 m^2. Height of the screen was about 0.5 m (from soil surface in the middle of the frame to top of the pyramid).

Frame

Ten centimeter high wood frame was assembled with nails or screws so that any movement of WCR beetles into or out of the cage was avoided. Metal wire, about 45 cm height, was attached to the frame to keep the pyramid-shaped screen above the plant stand (screen not fixed to the plant for cereal, soybean, etc.). The upper end of the frame was circular in corn and sunflower to allow the stem to grow out of the top of the cage. The bottom part of the frame was cut at an angle to improve insertion into the soil to a depth of about 5 centimeters. Soil was pushed up around the outside bottom of the frame to keep WCR beetles from escaping from underneath the frame.

Screen

The screen was carefully fixed in two pieces to the wooden frame with wood strips (to avoid WCR adult escape). The screen had 1-1.5 mm holes to allow air and light into the cage (the holes were small enough to keep beetles from escaping). A zipper was affixed to one side of the screen to allow for entry into the cage. The screen was closed at the top with twine. In the corn and sunflower plots, the twine encircled the corn and sunflower stems. For collecting WCR beetles within emergence cages, Pherocon AM traps were cut into two pieces and one piece was placed in each cage. These were fixed to either the plant stem or onto a stick at about 25-30 cm height. An area encompassing about 3-4 corn plants in a line was covered by each cage. Plants were trimmed back under each cage so as to avoid screen damage, but to allow root development to continue in case larval feeding was taking place.

Emergence cages and the Pherocon AM traps inside the cages were used to catch WCR adults emerging from the soil. WCR emergence proved the presence of eggs in the soil in previous year and development of larvae to adult stage in the crop in the rotation in the present year.

The following data were recorded in the crop rotation trial.

For WCR adult occurrence in corn and in other crops in rotation (on Pherocon AM traps):

- Number of adults;
- Sex ratio for adults (to follow and compare sex ratio by crop over the period of adult activity);
- Number of eggs being carried by WCR females over the trapping period. For WCR larval feeding, development and adult emergence in crop stands (in emergence cage):
- Number of adults emerged from the soil (caught on the Pherocon AM traps inside the cages).

For WCR larval presence:

- Number of larvae and pupae from roots and soil. Took 10 corn plants/plot in the 6 corn replications in the rotated section (the 4-crop section) of the field and the same number (6 x 10) in the field in corn only. Corn plants with about a 20 cm soil cube around the plant base were dug (spade or shovel) and the soil was slowly broken away from the plant root system and the number of larvae and pupae counted;
- Damage to corn roots was evaluated by using the Hills and Peters 1-6 root damage rating scale (10 corn plants/plot between 10-20 July).

For WCR egg laying, presence of eggs in soil in each crop stand:

- Soil samples were taken from late August to late September to determine WCR egg populations at the end of the egg laying period in each plot.

Additional data recorded: Weed species and coverage were recorded during the growing period from early July to late August in 4 sample areas (each 1 m^2) in each plot in the rotation strips and 1 sample (1 m^2) 10 replications for the corn in the large corn field section. Relevant meteorological data (rainfall, daily temperatures, etc.), agronomic data (crop variety, soil preparation, fertilization, seeding rate, sowing time, harvest, yields, and crop phenologies were recorded. Other crops around the trial and their direction from the plots were also recorded.

Results and conclusion

The paper discusses WCR adult occurrence in 2001 in Pherocon AM traps in corn and in crops rotated to corn for the three countries, while adult emergence data in the emergence cages are only available for Hungary in 2001. In Hungary, results for 2000 show low captures of WCR adults on Pherocon AM traps and no WCR adults in emergence cages in crops other than corn (*table 1*). WCR eggs were found only in soil samples taken in 2000 from corn plots. However, egg populations were low in 2000.

Table 1 - Total number of WCR adults caught on traps in crop rotation trial (Szeged, Hungary, June-September, 2000)

Crop	WCR adults in 18 traps	
	Pherocon AM	Emergence cage
Oat	4^a	0^a
Soybean	3^a	0^a
Sunflower	8^a	0^a
Corn-a (strips)	273^b	19^b
Corn-b	262^b	17^b

*values with different letters are significantly different (two sample t-test, p=0.05).

The WCR population increased in the area in 2001. For 2001, an increased number of WCR adults were caught on Pherocon AM traps in sunflower, soybean, and cereal (later harvested) plots, in addition to significant numbers caught on the traps in the corn plots (*table 2*).

Table 2 - Total number of WCR adults caught in traps in crop rotation trial (Szeged, Hungary, June-September, 2001)

Crop	WCR adults in 18 traps	
	Pherocon AM	Emergence cage
Winter wheat	241^a	1^a
Soybean	22^b	0^b
Sunflower	80^c	0^b
Corn-a (strips)	3306^d	207^c
Corn-b	1228^e	98^d

*values with different letters are significantly different (two sample t-test, p=0.05).

The highest WCR adult numbers were trapped in continuous corn (67.6 adult/Pherocon AM trap/week) at the end of July. Significantly fewer WCR adults were found in other crops (0.4 adult/trap/week in soybean, on plots of harvested winter wheat 3.7 adult/trap/week, and 1.8 adult/trap/week in sunflower). Some WCR adults were trapped in emergence cages in corn after soybean and winter wheat, but significantly fewer than in corn after corn. WCR adult numbers on Pherocon AM traps in corn reached economic threshold levels in 2001.

In FRY, the population of WCR in area where the test plot was located reached economic levels in 1993. Damage on corn from WCR larval feeding was recorded during the next 4 years. However, the second-year corn test field selected for the crop rotation trial in 2001 showed extremely low number of adults. There were no damaged plants from WCR larval feeding in the crop rotation trial and populations of larvae were so small that washing the soil to extract rootworm larvae showed no larvae.

During the sampling period (July 14 to September 3) in Field B (corn only), 34 adults were captured. In Field A (rotation strips) a total number of 10 adults were caught in strips with corn and total number of 2 adults in soybean. In sunflower and oat there were no WCR adults caught (*table 3*).

Table 3 - Total number of WCR adults trapped in the crop rotation trial (Dobanovci, FRY, 2001)

Crop	WCR adults in 18 traps	
	Pherocon AM	Emergence cage
Oat	0	0
Soybean	2	0
Sunflower	0	0
Corn-a (strips)	10	0
Corn-b	34	3

* low adult numbers did not allow for statistical analysis.

In 72 emergence cages, only 3 adults were caught in Field B (corn only). In Field A (crop rotation strips), there were no adults caught in the 72 emergence cages. Between July 20 and 25 the corn was checked for a presence of larvae and larval feeding damage. No larvae were found and no damaged roots or goosenecking were observed. Soil samples were taken from each crop stand on September 13 and 14. Presence of eggs in the soil was not detected.

In Croatia, the number of beetles caught on Pherocon AM traps in the corn field were higher compared to FRY plots and were similar to those in 2001 in Hungary. Numerous WCR adults were trapped in corn and crops other than corn (*table 4*).

In corn A (strips) the maximum catch did not exceed 28 adults/Pherocon AM trap/week. This value for corn B was 61 adults/Pherocon AM trap/week. Significantly fewer WCR adults were trapped in other crops (few adults in barley, soybean, and sunflower).

Table 4 - Total number of WCR adults trapped in crop rotation trial (Tovarnik, Croatia 2001)

Crop	WCR adults in 18 traps	
	Pherocon AM	Emergence cage
Spring barley	26[a]	0[a]
Soybean	11[a]	1[b]
Sunflower	47[a]	1[b]
Corn-a (strips)	369[b]	34[c]
Corn-b	801[c]	208[d]

*values with different letters are significantly different (two sample t-test, p=0.05).

WCR adults in emergence cages in corn after corn indicate a high infestation level. Since in Croatia and FRY the trial started in year 2001, egg laying in other crops and larval development in following corn will not be evaluated until 2002.

The first year results of the crop rotation trials indicate the possibility of a slight adaptation of WCR adults (preference for corn, but occurrence in other crop stands) to the European crop rotation system. However, second- and third-year data should give the necessary information for proper conclusions to be drawn.

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COMPARISON OF PERFORMANCE OF DIFFERENT TRAP TYPES FOR MONITORING OF *DIABROTICA VIRGIFERA VIRGIFERA*

Zoltán Imrei, Miklós Tóth,
Plant Protection Institute, Hungarian Academy of Science,
Budapest, Pf. 102, H-1525, Hungary; phone +36-1-3769555;
fax: +36-1-3769729; e-mail: h2371tot@ella.hu

Géza Vörös,
Tolna-megye Station of Extension Plant Protection Service, Szekszárd,
Pf 104., Keselyüsi út 7. H-7101, Hungary

István Szarukán, Tamás Gazdag, Attila Szeredi,
Agricultural University, Debrecen, Pf. 58, H-4001, Hungary

Abstract
The present results come from the first year of a study aiming at the evaluation of the trap types developed at our lab in Hungary for monitoring *Diabrotica v. virgifera*. Results are presented on trap types PAL (pheromone baited), PALs (floral attractant baited) and VARs (baited with both pheromone and floral baits). For comparison, yellow sticky traps were also included. Traps were tested at three sites in Hungary in 2001.

Males were detected earliest in pheromone baited traps PAL and VARs at approximately at the same time. Later on large numbers of males were continously captured.

First female catches were recorded much later than male captures, predominantly in both traps with floral lures - PALs and VARs. The two types captured females during similar periods. There were too few beetles in unbaited traps to draw conclusions about natural sex ratios and sex ratios captured in the different trap types. Next year natural sex ratios are planned to be studied through non-discriminating sampling methods parallel to continuous operation of the trap types with the respective baits.

INTRODUCTION

On earlier meetings, presentations from our team most frequently dealt with the development and optimization trials of several trapping devices for capturing the corn rootworm. In the present study we intended to investigate these trap types in a real-life monitoring situation, comparing their performance in monitoring of the pest.

Main questions we intended to study included:

- are there differences between the monitoring performance of the different trap types according to season?

- are there differences in sex ratio captured and does it change in the course of the season?
- how does the sex ratio captured relate to natural sex ratio of the population?

Materials and methods

The trap types we tested included:

- the CSALOMON® PAL transparent sticky "cloak" trap, baited with the pheromone;
- the CSALOMON® PALs yellow sticky "cloak" traps, baited with the floral bait;
- the recently developed VARs funnel trap, baited with dual bait, consisting of both the pheromone and the floral lure.

For comparison we also tested an unbaited yellow sticky trap, and an unbaited transparent sticky trap (same design as PAL or PALs, but without bait). The yellow colour of the PALs and yellow sticky traps was similar to the human eye to the "Multigard" yellow.

Tests were run by different personnel parallelly at three sites in Hungary, at Bácsbokod (Bács-Kiskun county), Kiszombor (Csongrád county) and Szekszárd (Tolna county). At each test site 4 traps of each trap type were operated. Traps were inspected twice weekly. Sticky sheets of the sticky trap types were replaced by new ones once weekly (first month of the test, when catches were still lower) or twice weekly later on. Baits were replaced at monthly intervals. All captured specimens were sexed, by dissection in case of the Szekszárd captures, or by outside morphology in case of catches from the other two sites.

Results and discusson

Although the present test was not designed to allow for direct comparison of trap performance, as a preliminary evaluation we calculated the mean catches per trap for all the test period (*figure 1*). In males the smallest numbers were caught in the unbaited trap types. Highest catches were observed in the trap types with pheromonal bait (PAL and VARs). At one of the sites the means did not differ, at the two others PAL caught somewhat more than VARs. The floral baited PALs trap also captured a sizeable number of males at two of the sites, although these numbers were significantly lower than catches of the pheromone-baited trap types.

In the females both trap types with floral baits (PALs and VARs) showed good catches; PALs was significantly better at 2 of the sites of the sites. Captures in the unbaited yellkow sticky trap were much smaller than in trap types with floral lure. The pheromone baited PAL and transparent sticky unbaited captured negligible numbers of females.

When looking at the ratio of females captured (means calculated for all the test period), we can leave out from evaluation the type PAL since it practically did not catch females, and VARs, since here, because of the presence of a pheromone bait

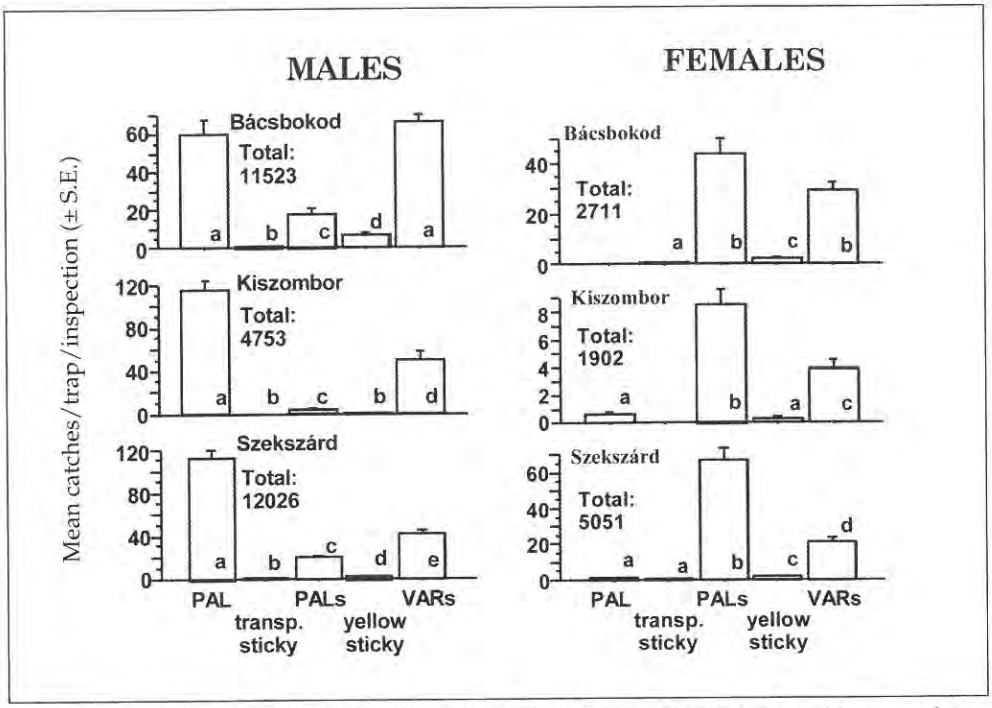

Figure 1 - Mean catches of corn rootworm in different traps. Means followed by same letter within one diagram are not significantly different at P:=5% by ANOVA, Games-Howell

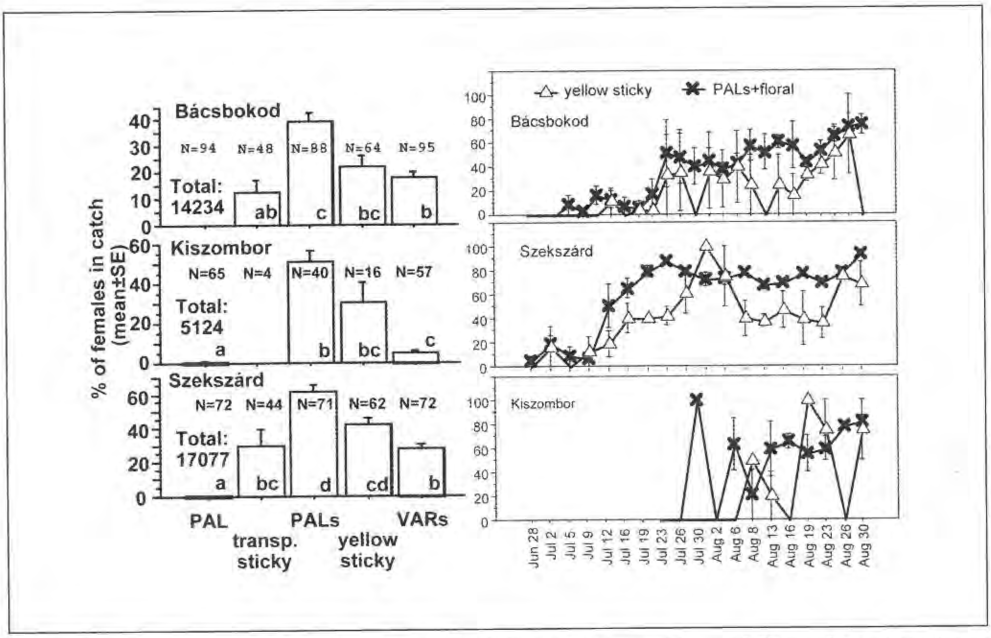

Figure 2 - Percentages of females in catches of different traps. Means followed by same letter within one diagram are not significantly different at P:=5% by ANOVA, Games-Howell

beside the floral bait, the ratio of females captured is distorted. Among the other trap types the same tendency can be observed at all three sites, with the PALs catching most females, followed by yellow sticky and with transparent sticky catching the lowest percentage. This suggests a preference towards females already as an effect of yellow colour, which is further enhanced by the presence of the floral bait. So the female ratio in our PALs or yellow sticky traps did not represent the natural ratio in the population. This finding supports earlier reports in the literature where synthetic floral attractants for *Diabrotica spp.* appeared to be preferentially active for females (Ladd *et al.*, 1985, Andersen and Metcalf, 1986, Lampman *et al.*, 1987, Yaro *et al.*, 1987).

When studying the seasonal changes in female ratio captured, data usually showed quite high variability especialy in case of the yellow sticky type (due to the low numbers captured) (*figure 2*). However, a tendency of female ratios increasing as the season progresses can be observed at all three sites, and in both trap types. We suggest that this perhaps is due to the natural sex ratio changes in the population as males die off faster. Another explanation could be that the phenomenon is due to the competition of natural host plant volatiles from silk or pollen with attractive substances of the artificial floral lure. Silking and pollen production took place in the first month of the tests at each site, and there have been indications reported from North America in the literature on such competition (Andersen and Metcalf, 1986, Lampman *et al.*, 1987, Hesler *et al.*, 1994, Hammack, 1996). Unfortunately the catches of the unbaited transparent sticky traps, which should have shown the natural population's sex ratio, were very low an did not allow an evaluation in favour of the above hypotheses.

In a further approach we looked at the seasonal distribution of catches. We show the Bácsbokod results as an example; results on other sites looked very similar. Males were captured much earlier than females at all sites (*figure 3*).

Comparison of the monitoring performance of the trap types was very difficult since some trap types caught considerably less than others, and it was difficult to compare tendencies of catches. Therefore as a next step we calculated catches normalized against the maximal catch (mean at a given inspection) of each respective trap type, and plotted these data. In *figure 4* we show the Bácsbokod results as an exaple.

In male catches again no clear difference in catch tendencies could be discerned among the trap types. Perhaps the only conspicuous phenomenon is that standard errors of yellow sticky captures are oustandingly high as compared to the other types. This may be mainly due to the low numbers generally caught by this trap type. The same phenomenon can be observed at all the other sites as well, and also in female catches.

Since the seasonal distribution of captures did not show any outstanding differences, we continued our evaluation by establishing some key viewpoints thought to be important for good monitoring performance and tried to compare our trap types according to these viewpoints:

- trap's ability of detection - date of first specimen captured in the season (by any of the 4 replicates);
- trap's ability of signaling continuous presence of population - neighboring dates on which more than 2 of the 4 traps captured a higher number of beetles than the unbaited level (mean catch of transparent sticky unbaited traps);

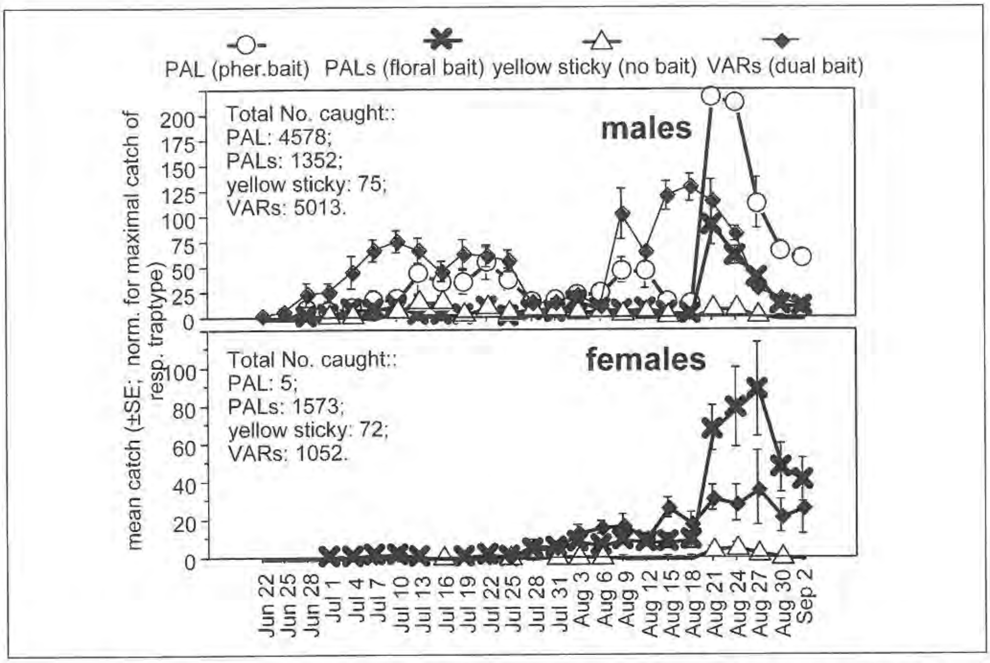

Figure 3 - Catches of corn rootworms in different trap types at one of three sites (Bácsbokod, Hungary, 2001). Each dot represents the mean (±SE) catch of 4 on the given date

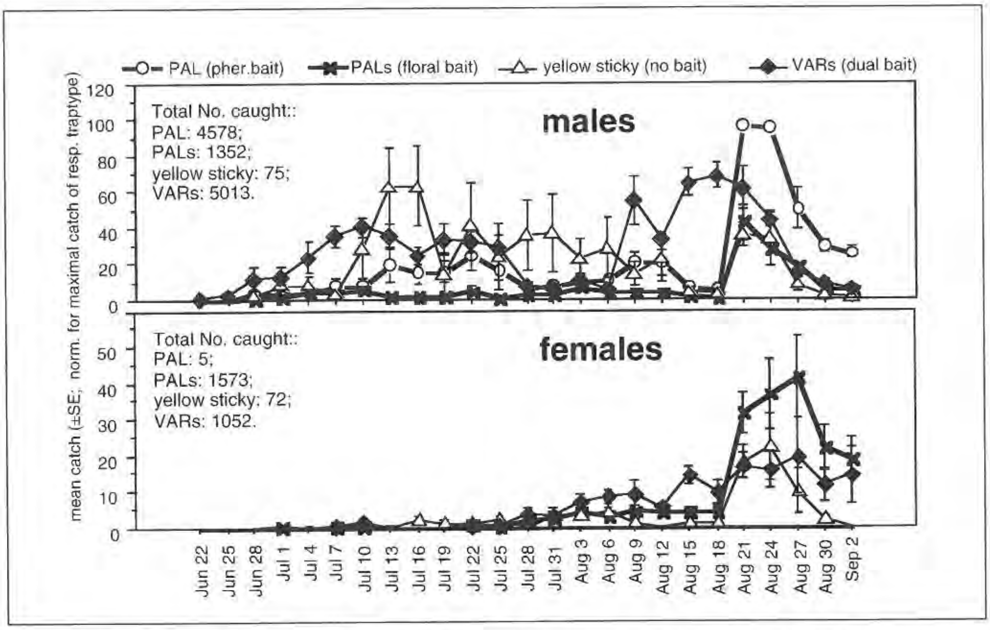

Figure 4 - Catches of corn rootworms in different trap types at one of three sites (Bácsbokod, Hungary, 2001). Each dot represents the mean (±SE) catch of 4 traps (normalized against maximal catch of resp. trap type for ease of comparison) on the given date

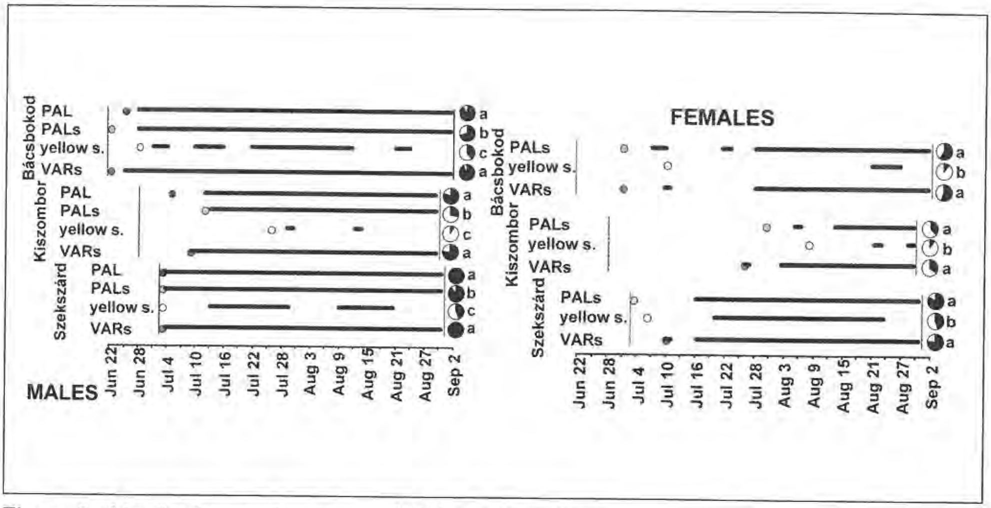

Figure 5 - Monitoring performance of different trap types

- reliability of monitoring - number of inspection dates on which a given trap did capture specimens in larger numbers than the unbaited level (mean catch of transparent sticky unbaited traps).

In males the trap types containing pheromone bait (PAL and VARs) caught the first specimens in most cases at the very beginning of the tests. Type PALs with floral lure was performing similarly, while the yellow sticky unbaited trap lagged behind a little bit at one of the sites.

The PAL, VARs and PALs types were able to continuously signal the presence of males during similarly prolonged and continuous periods, practically almost immediately after the detection date to the end of the test. In this respect yellow sticky traps performed poorly, showing instead of a continuous line several short bouts.

The PAL and VARs traps were showing caches most frequently, followed by PALs which was still pretty good. Again yellow sticky traps were the worst.

In females the two trap types with floral bait were similar, and superior in all aspects over unbaited yellow sticky traps.

Apart from the above "scientific" aspects, we assessed also approximate costs of monitoring for the respective trap types according to our experimental schedule in the present tests. Prices of items were the EXW prices of our CSALOMON trap family.

Table 1 - Monitoring costs (in USD)

Item	PAL	PALs	yellow sticky	VARs
trap+bait (1x)	2.90	4.10	1.80	5.80
replacement bait (2x)	3.90	6.40	not applicable	3.90 6.40
replacement sticky sheets	19x1.20= 22.80	19x1.30= 24.70	15x1.30= 19.50	not applicable
insecticide strip	not applicable	not applicable	not applicable	0.40
Total	29.60	35.20	21.30	16.10

It appeared that in the present situation, when regular exchange of sticky sheets was necessary for reliable monitoring, the costs were lowest in case of the non-sticky VARs trap, which, when comparing trap prices, is most expensive.

Summarising our results we conclude that:

- for catching males the PAL and VARs types are optimal, PALs can also be used, if sensitivity is not a critical consideration.
- for catching females the PALs and VARs types can be recommended.

Usually the sticky types PAL and PALs are better suited to detection situations (when the population density of the pest is low, and no frequent exchange of replacement sticky sheets is necessary). The funnel VARs type seems to be more advantageous in long term monitoring trials.

Throughout the test the unbaited yellow sticky traps performed worst in all aspects. We cannot recommend with a clear conscience this trap type for use in monitoring efforts on *Diabrotica*.

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DIABROTICA VIRGIFERA VIRGIFERA ERADICATION CONTAINMENT TEMPTATIVE IN VENETO REGION: YEAR 2001

Lorenzo Furlan, Andrea Di Bernardo, Vincenzo Girolami,
Department of Agronomy, Entomology,
University of Padova, Agripolis,
via Romea 16, Legnaro PD; phone 0039 049 8272801;
FAX: 0039 049 8272810; e-mail: ento@unipd.it

Marco Vettorazzo, Anna Maria Piccolo,
Phytosanitary Service of Veneto, Venice Office,
via Poerio 34, 30173 Mestre VE;
phone: 0039 041 2795703; FAX: 0039 041 2795702;
e-mail: serv.fitove@mail.regione.veneto.it

Giorgio Santamaria, Lanfranco Donantoni, Vanna Funes,
Freelances

Abstract

The attempt to eradicate or at least to contain the newly arrived population of WCR was implemented in and just around a focus area near the Venice airport in 2001 by using the strategies deployed in 1999 and 2000. Monitoring of WCR population was carried out by using PAL sex pheromone traps placed out in focus and safe area. Insecticide treatments were applied to maize fields in focus area and to those fields in safe area where WCR specimens had been caught. Farmers were required to comply with eradication measures (including the prohibition of maize monoculture in focus areas) by Ministerial Decree and by specific ordinances of the Veneto Region. All the fields in the focus area that had been planted to maize in 2000 were checked to determine what crop was planted in 2001. 0.44 ha of monoculture maize were found and mechanically destroyed from the 2nd to 20th of July. In initial focus area (1200 ha) 6 specimens were captured in traps placed out close to the border of the area, near the monoculture maize fields of the safe area. In the latter, new focus areas were established according to new findings of *Diabrotica* males. In a monoculture maize field 300 m far from the border of the initial focus area, 157 WCR males were captured on a PAL trap in mid July. This was the only trap of the first grid of the safe area that captured specimens. Within few days many other traps were placed out exclusively in monoculture maize fields at increasing distances from the field where the captures were recorded. Totally 67 more specimens were caught in the first and second group of traps placed out in the safe area. Most of the captures were recorded just before the insecticide treatments; a few further captures were observed after 15 days from the last treatment. No specimens were caught on traps placed out in fields planted to crop different from maize

despite the fact these fields were near monoculture maize fields where conspicuous populations had been detected. Several thematic-maps reporting crops, traps and captures positions have been produced.

Key words: *Diabrotica virgifera virgifera*, eradication, containment, Veneto.

INTRODUCTION

The attempt to eradicate or at least to contain the newly arrived population of WCR near the International Airport of Venice was implemented using the strategies deployed in 1999 and 2000 (Furlan *et al.*, 1998, 1999a and b, 2000).

MATERIALS AND METHODS

The eradication program was implemented in and just around a focus area near the Venice Airport. The program was based on:

- *Initial focus area (1200 ha):*
  - Monitoring the WCR population: 159 sex pheromone PAL traps were placed out from 25th to 29 of June in the focus area.
  - Imposing restrictions on the planting of maize in fields where maize was grown the previous year; in a small area (37 ha) around the fields where 77 WCR specimens were captured in 2000 maize cultivation was completely prohibited; in the rest of the focus area was prohibited to plant maize after maize.
  - Applying insecticide treatments to all maize fields to control WCR adults; the insecticide used was Dursban (chlorpyrifos) WG at the rate of 1,1 kg/ha; all maize fields (153 ha) were sprayed between 17th of July and 1st of August; further 20 ha in the part near the safe zone where captures had been recorded were sprayed again between 10th to 23rd of August.
  - Prohibiting the movement of fresh maize or soil in which maize was grown the previous year outside of the focus area.
  - Not allowing maize to be harvested before 1st of October.
- *Safe area (about 35.000 ha):*
  - Monitoring of WCR population:
    in the first phase, 207 PAL traps were deployed from the 9th of July in the safe area according to a 1 km X 1 km grid mainly in monoculture maize fields; later on further 430 PAL traps were added in monoculture maize fields at increasing distances from the focus area. For research purposes, 132 more traps of different types were added in focus and safe area.
  - Applying insecticide treatments to maize fields (and those all around) where WCR specimens are caught: an area of 54 ha of maize was sprayed between the 24th and 28th of July; 25 ha were sprayed on the 3rd of August and further 43.5 ha on the 10th and 11th of August.

Farmers were required to comply with these measures by Ministerial Decree and by specific ordinances of the Veneto Region through specific meetings. The Region also made money (about 78.000 Euro) available to support the eradication programme.

RESULTS

Field checks. After the farmers had been informed of the eradication programme by their organizations, all the fields in the focus area that had been planted to maize in 2000 were checked to determine what crop was planted in 2001.

The field checks were completed in late June and 3 fields totaling 0.44 ha of monoculture maize were found. These fields were mechanically destroyed from the 2nd to 20th of July.

WCR captures (*figure 1*)

- *Initial focus area (1200 ha):*
- 6 specimens were captured: on July 17 (1), July 24 (1), August 18 (2), September 7 (2). All captures were recorded in traps placed out close to the border of the area, near the monoculture maize fields of the safe area.

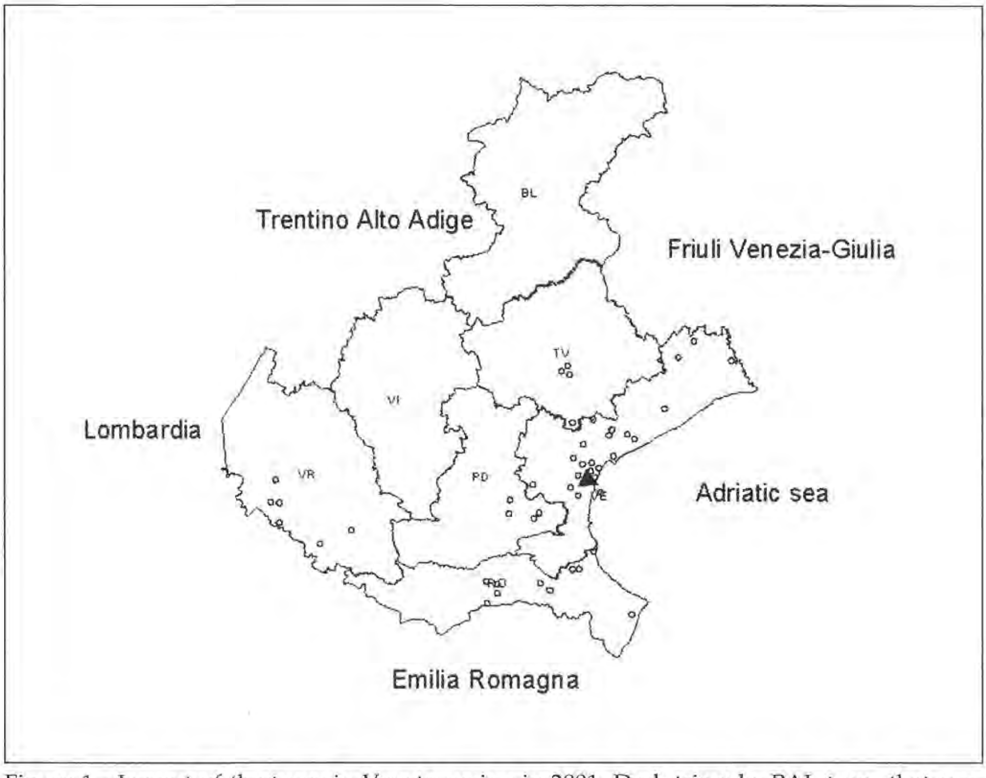

Figure 1 - Layout of the traps in Veneto region in 2001. Dark triangle: PAL traps that captured WCR specimens. Black empty circles: PAL traps that captured no specimens.

- *Safe area and new focus areas*:
  - in a monoculture maize field 300 m far from the border of the initial focus area, 108 WCR males were captured on a PAL trap over a 4 day-period (16th to 19th of July; trap set up on July 16). Further 49 specimens were captured on the same trap just before insecticide treatment (afternoon of the 24th of July) repeated after 7 days. After the treatments, the traps did not catch any more beetles until the 4th of September.

    This was the only trap of the first grid of the safe area that captured specimens. An additional focus area (250 ha) was immediately defined. Within few days (from 22nd of July on) many other traps were placed out exclusively in monoculture maize fields at increasing distances from the field where the captures were recorded. Totally 67 more specimens were caught in the first and second group of traps placed out in the safe area. Most of the captures were recorded just before the insecticide treatments; a few further captures were observed after 15 days from the last treatment. Another small focus area (27 ha) had to be defined about 3 km away from the first one; rescue treatments were made in both the newly defined focus areas. No specimens were caught on traps placed out in fields planted to crop different from maize despite the fact these fields were near monoculture maize fields where conspicuous populations had been detected.

    Several thematic maps reporting crops, traps and captures positions have been produced.

Table 1 - Influence of the field rotation on the level of WCR captures in traps deployed in the safe area by using G test. Two stars mean a significant difference at P=0,01

area with wcr	no.	% in monoculture	% traps (with captures)	G Test
Traps with >5 WCR traps with 1-4 WCR	6 16	100% 56%	within 500 m from a monocolture 57%	0,005 **
area with wcr		% captures	% traps (with captures)	
total traps in monoculture total traps non in monoculture	21 49	71% 14%	within 500 m from a monoculture 57%	0,000 **

CONCLUSIONS

The most important conclusions from this year's work can be summarized as follows:

- The strategies implemented in focus area proved to be very effective in stopping WCR populations; the few specimens captured (6) were collected in the border of the focus area near (within 600 m distance) the small monoculture fields in the safe area where WCR populations were recorded; therefore a re-colonisation of the initial focus area coming from monoculture fields of the safe area (where probably the species had arrived years before but not detected) was observed.

- The key factor of eradication/containment of the species appears to be the interruption of monoculture on all the fields of the focus area. In fact, most of the maize fields where conspicuous captures were recorded were monoculture fields; few specimens were caught in first-year maize near to monoculture fields where high WCR populations had been observed (see *table 1*). Also very small continuous maize fields can allow high WCR reproduction and spread.
- Insecticide treatments directed against the adults are very effective in significantly reducing the populations and stopping their spread.

Acknowledgements to Prof. Rich Edwards (University of Indiana, West Lafayette, USA), for critical review of the manuscript.

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Work carried out within the framework of EU Diabrotica Project (QLK5-CT-1999-01110).

MONITORING OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN LOMBARDY (NORTHERN ITALY) IN 2001

Marco Boriani, Eugenio Gervasini,
Lombardy Region, Phytosanitary Service,
P.zza IV Novembre 5, 20124 Milano, Italy

Abstract

Maize is the most important field crop in Lombardy (Northern Italy) where over 350,000 hectares are cultivated. In 2000, the maize pest western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte (Coleoptera: Chrysomelidae), was detected for the first time near the Milan Malpensa Airport (Varese). Three specimens were collected on two farms using pheromone traps. As a result, two distinct areas of infestation were identified and within these maize cultivation was prohibited in 2001.

An extensive monitoring campaign was carried out in 2001 and over 200 PAL pheromone traps were placed in the 11 provinces of the region. WCR was found at different population levels in 8 provinces covering over 100,000 hectares of maize. In Como and Varese provinces, the highest numbers of WCR adults/trap were observed when compared to the other provinces. However, damage caused by larvae and adults was not noted. Preliminary evidence suggests that the area of Malpensa Airport is not the real origin of WCR spreading and that a different hypothesis on the matter is advanced here.

The results of adult monitoring and population levels observed are also reported.

Key words: Monitoring, WCR, Lombardy, Italy.

INTRODUCTION

Maize is the most important field crop in Lombardy (Northern Italy) where over 350,000 hectares are cultivated. The most important cultivated areas are placed in the Po valley where over 80 percent of the livestock farms are concentrated.

First occurrence of *Diabrotica virgifera virgifera* LeConte was detected in Lombardy in 2000 near Milan Malpensa Airport (Varese). Three specimens were collected on two farms using pheromone traps (Boriani and Gervasini, 2000). As a result, two distinct focus areas were identified and within these maize cultivation was prohibited in 2001.

The monitoring has shown an unexpected spread of WCR. The pest was found at different population levels in 8 provinces covering over 100,000 hectares of maize. In Como and Varese provinces, the highest numbers of WCR adults/trap were observed when compared to the other provinces.

Preliminary evidence suggests that the area of Malpensa Airport is not the real origin of WCR spreading and that a different hypothesis on the matter is advanced here.

The results of adult monitoring and population levels observed are also reported.

Materials and methods

An extensive monitoring campaign was carried out in 2001 and over 200 PAL pheromone traps, produced by the Plant Protection Institute of Budapest, were placed in farms and near potential introduction areas (airports, customs areas, etc.).

For research purposes, traps of different types were added in some fields where high captures had been recorded on PAL traps.

An eradication programme was also implemented in the areas of infestation near the Malpensa Airport. This programme was based on:

- monitoring the WCR population in the focus areas of Somma Lombardo and Vizzola Ticino and in the respectively safe areas;
- imposing absolutely prohibition on the planting of maize in the focus areas;
- application of an insecticide treatment to control WCR adults in the safe areas within five days from the first catch of adults on PAL pheromone traps;
- not allowing corn to be harvested before October 1st.

Results and conclusions

The monitoring has shown an unexpected spread of WCR. The pest was found in areas where the maize have minor importance and that were not monitored in the past, but also where no specimen was detected in 2000 (e.g. near Orio al Serio Airport, Bergamo).

The WCR was found at different population levels. In Como and Varese provinces, the highest numbers of WCR adults/trap were observed when compared to the other provinces. The highest numbers of WCR males/PAL trap/day at the peak of the swarming were recorded, from monocolture maize fields, in some monitoring sites near the border with the Canton of Ticino (Switzerland), with catches levels also above 50 WCR males/trap/day.

Lower population levels were observed in Lecco, Milano and Bergamo provinces and still lower numbers in Sondrio, Lodi and Cremona, where only occasional specimens were caught. In the provinces of Brescia, Pavia and Mantova no specimen was detected.

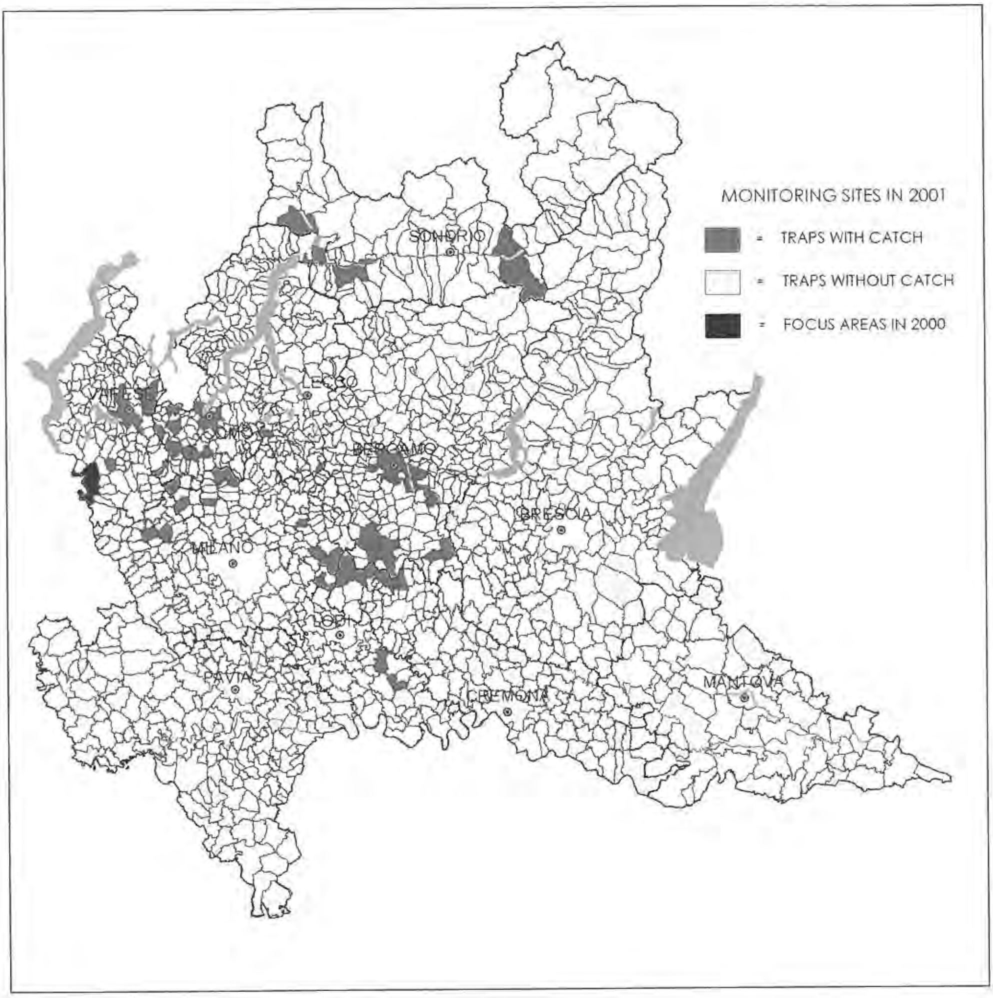
Damage caused by larvae and/or adults was not noted at any of the monitoring sites.

The population levels that were observed in the safe areas near Milan Malpensa Airport (less than 15 WCR males/PAL trap/day at the peak of the swarming) suggest that this area is not the real origin of WCR spreading and that the primary focus should be placed between the province of Como and the Canton of Ticino (Switzerland).

In the focus areas 50 PAL traps were placed beginning from June 15th and no specimen was detected. Within these areas, the maize cultivation was prohibited and only soybean, oil-seed rape and oats were sown. Furthermore, no spread was observed in the south of the infested areas near Malpensa Airport.

Pherocon® AM yellow sticky and Pherocon CWR traps were placed in some fields where high captures had been recorded on PAL traps. Pherocon AM traps did not capture any specimens. Pherocon CWR traps captured only occasional specimens.

The spread of the WCR in Lombardy is shown in the following map.



It is expected that the area of infestation will continue to increase in coming years and that eradication measures will be not applicable.

However, containment measures will be applied through the next management options:

- a good understanding of beetle biology and factors that affect beetle population

dynamics in agroecosystems is needed to effectively use management strategy;
- the rotation on maize and other cultures seems to be the primary strategy advisable for control of WCR. Crop rotation is highly effective in controlling rootworms and has the added benefit of not increasing the selection for insecticide resistance;
- corn hybrids commercially available with source of resistance effective in controlling WCR might reduce damage from larval rootworm feeding, including stalk strength and increased root mass size;
- the decision to use insecticide on the level of rootworms present in individual fields will have to be based on adult scouting and economic thresholds.

Acknowledgements to prof. Rich Edwards (University of Indiana, West Lafayette, USA), for critical review of the manuscript. Furthermore, we are also grateful to the peoples that have worked on monitoring programme and without whom this paper would not have been possible.

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THE PRESENCE OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN ITALY IN 2001: DISTRIBUTION, POPULATION LEVEL AND WHAT HAS TO BE DONE

Lorenzo Furlan, Andrea Di Bernardo,
Department of Agronomy, Entomology,
University of Padova, 35020 Legnaro PD

Marco Vettorazzo,
Veneto Region, Phytosanitary Service,
Venice Office, Via Poerio 34, 30172 Mestre-Venezia.

Marco Boriani, Eugenio Gervasini,
Lombardy Region, Phytosanitary Service,
P.zza IV Novembre 5, 20124 Milano

Giacomo Michelatti,
Piemonte Region, Phytosanitary Service,
Corso Grosseto 71/6 – 10147 Torino

Graziano Caielli,
Agricolture Sector, Novara Province,
Via Dominioni, 28100 Novara

Adeodato Ortez, Carlo Frausin, Giorgio Agazzi,
Friuli-Venezia Giulia Region, OMP,
Via Beato Odorico 13, 33170 Pordenone

Massimo Bariselli,
Emilia Romagna Region, Phytosanitary Service,
via di Corticella 133, 40129 Bologna

Abstract

Diabrotica virgifera virgifera (WCR) monitoring was extended and implemented on the basis of the protocol used in the previous years in Northern Italy (Friuli-Venezia Giulia, Veneto, Emilia-Romagna, Lombardy, Piemonte). Maize fields were selected, in addition to the already defined focus areas, in sites where maize is often grown for multiple years and also at potential introduction areas (such as nearby airports, custom institutions, etc.). PAL sex pheromone traps were set up mainly, or exclusively, in maize after maize fields. In addition, in maize fields where high WCR captures had been recorded the root systems of plants were evaluated following established protocols from the USA. The species was detected in three regions: Veneto (small expansion of original focus area), Lombardy (over 360,000 ha, about 100,000 ha of maize), Piemonte (about 17,800 ha, 3,000 ha of maize).

Maximum population level (over 50 WCR males/ PAL trap/day at the peak population) was observed in Lombardy but no adult feeding damage on leaf tissues or ears was observed. Only in fields planted to maize for at least two subsequent years did PAL traps capture conspicuous numbers of adults. In none of the maize fields with the highest male captures in Veneto and Lombardy did root systems show any visible WCR injury. Pherocon AM yellow sticky traps placed in some fields where high captures had been recorded on Pal traps did not capture any specimens. Therefore no economic populations are present so no insecticide treatments for usual maize cultivation are needed in 2002. Finally, strategies for the future are discussed.

Key words: *Diabrotica v. v.* , PAL pheromone traps, Italy, population level.

INTRODUCTION

Over the past several years, extensive monitoring of *Diabrotica virgifera virgifera* (WCR) was carried out in different regions of Northern Italy, but mainly in Veneto following a common protocol (Furlan *et al.*, 1998, 1999a, 1999b, 2000). In 2001, monitoring was extended and implemented on the basis of the protocol used in the previous year.

MATERIAL AND METHODS

Maize fields were selected, in addition to the already defined focus areas, in sites where maize is often grown for multiple years and also at potential introduction areas (such as nearby airports, custom institutions, etc.). PAL sex pheromone traps were set up mainly, or exclusively, in maize after maize fields of the following regions:

Friuli-Venezia Giulia, 25 traps; Veneto, over 750 traps in safety and focus area, 160 traps in sensitive place 5 to 100 km away from the focus area; Emilia-Romagna, 20 traps; Lombardy, over 200 traps; Piemonte, 12 traps.

In maize fields where high WCR captures had been recorded the root systems of at least 20 plants/field were evaluated following established protocols from the USA.

RESULTS

Presence

In Friuli-Venezia Giulia and Emilia-Romagna no specimens were caught over the last 3 years.

Veneto: in the focus or safe area defined after the first captures recorded in 1998, specimens were captured each year from 1999 to 2001 in monoculture maize fields. The current infested area is about 1,500 ha (in 2001 about 230 ha of maize).

No specimens in the other sites (five to hundred km away from the focus area) were caught despite the fact that hundreds of PAL traps had been deployed in monoculture fields.

Lombardy: three specimens were captured on a few traps placed out near Milan-Malpensa airport (Varese) in 2000 (Boriani and Gervasini, 2000); many traps were placed out in 2001 and numerous specimens were captured in a area of over 360,000 ha (about 100,000 ha of maize) including the provinces of Varese, Como, Lecco, Bergamo, Sondrio, Milano, Lodi and Cremona.

Piemonte: numerous specimens were captured in an area of about 17,800 (3,000 ha of maize) (Novara province) which borders the focus area of Lombardy defined in year 2000; no specimens were captured in fields located near Caselle airport and other provinces (Vercelli, Alessandria).

Population level

Adults: levels over 50 WCR males/ PAL trap/day at the peak of the swarming period were recorded from monoculture maize fields in Lombardy while the peak population was about 25 males/trap/day in Veneto and 15 males/trap/day in Piemonte. Only in fields planted to maize for at least two subsequent years did PAL traps capture conspicuous numbers of adults; most of the infested fields were monoculture maize fields, the others being first-year maize fields located near infested monoculture fields. Pherocon AM yellow sticky traps, which are often used in USA to determine the presence of economic WCR populations within a field, were placed in some fields where high captures had been recorded on PAL traps. These Pherocon traps, however, did not capture any specimens. No adult feeding damage on leaf tissues or ears was observed despite observations made on thousands of plants in the fields where the highest numbers of WCR males were recorded on the sex pheromone traps.

Larvae: in none of the maize fields in Veneto and Lombardy did root systems show any visible WCR injury. Therefore NO economic populations are present so NO insecticide treatments for usual maize cultivation are needed in 2002.

Strategies for the future

Veneto: the eradication/containment programme should be continued following the strategies implemented in the previous years enlarging the area where the cultivation of maize after maize is prohibited (including fields five km from the area where specimens had already been captured).

Lombardy, Piemonte: in order to stop or at least to slow *Diabrotica* spreading a strip some km wide with NO maize after maize fields should be created along the border of the area where WCR specimens have been detected. This measure should be supported by an intensification of the information programme on WCR biology and the importance of crop rotations that is targeted at farmers in the regions.

Friuli-Venezia Giulia, Emilia-Romagna and other regions: monitoring should continue following the instructions utilized over the last several years.

Acknowledgements to Prof. Rich Edwards (University of Indiana, West Lafayette, USA), for critical review of the manuscript.

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Work carried out within the framework of EU Diabrotica Project (QLK5-CT-1999-01110).

SELECTION OF POTENTIAL NON-TARGET COLEOPTERAN HOST SPECIES FOR ASSESSING THE HOST SPECIFICITY OF EXOTIC BIOLOGICAL CONTROL AGENTS

Feng Zhang, Stefan Toepfer, Ulrich Kuhlmann,
CABI Bioscience Centre Switzerland, Rue des Grillons 1,
CH-2800 Delémont, Switzerland

Abstract

The ability of introduced natural enemies to persist in the environment, to reproduce and to spread gives classical biological control a unique advantage as a pest control method. As an exercise in applied ecology, biological control has always been challenged to predict the outcome of complex population processes. Despite many proven benefits, classical biological control has recently come under scrutiny because introduced natural enemies may adversely affect native species, especially rare and endangered species. Concerns should be based on non-target impacts at the population level, rather than attack on individuals but there is debate on which species should be considered when evaluating non-target impacts.

The EU Project DIABROTICA (QLK5-CT-1999-011110) aims to select specific biological control agents of western corn rootworm for potential importation and establishment in Europe. With regard to the safety of biological control the guidelines of the European and Mediterranean Plant Protection Organisation (EPPO) for the importation and release of exotic natural enemies will be followed. Natural enemy surveys carried out in Mexico and Argentina revealed that parasitoids belonging to the genus *Celatoria* (Diptera: Tachinidae) and *Centistes* (Hymenoptera: Braconidae) are found to parasitize adults of *Diabrotica* and *Acalymma* species (Coleoptera: Chrysomelidae).

Prior to the importation of these adult parasitoids, a literature survey has been conducted to determine the host specificity of these potential biological control agents. In addition, the literature has been scanned to determine Coleopteran species present in the European cultivated agricultural habitats (with emphasis on maize, alfalfa, squash, wheat, sunflower, and grassland habitats) which might be at risk to from being parasitized by exotic parasitoids. In Hungary, field surveys using sweep net samples were carried out to identify the Coleopteran species present in selected habitats. Simplifying principles have been developed to select potential non-target Coleopteran host species of greatest relevance for assessing the physiological host range of potential biological control agents in quarantine laboratory.

Work carried out within the framework of EU Diabrotica Project (QLK5-CT-1999-01110).

SIMULATION MODEL SPREADING SCENARIOS FOR WESTERN CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA*) IN CASE OF GERMANY

Peter Baufeld, Siegfried Enzian,
Federal Biological Research Centre for Agriculture and Forestry,
Stahnsdorfer Damm 81, D-14532 Kleinmachnow,
Federal Republic of Germany

Abstract

The dispersal rate of the Western corn rootworm (WCR) in Europe was analysed. The spreading of the WCR ranged from 60 to 100 km per year without containment measures and from 0 to 37 km with (FAO programme TCP/RER/6712). The simulation model used as an average a maximum WCR population-spreading rate of 80 km per year without containment measures and 20 km per year with. The maximum spreading rate is reached by WCR in the succeeding year only if continuous maize is available in the infested area. The concentration of maize in crop rotation is the main factor in the simulation model. Analysis of topography showed that the WCR is not able to fly over altitudes of 900 m, which is factored into the simulation model.

Calculations are carried out using GIS software ArcView/ArcInfo. This simulation model was used for the first time to simulate the spreading rate in Germany over ten years for two cases: 1) introduction via the international airport of Frankfurt am Main with and without containment measures and 2) progression of "natural spread" via Austria with the starting point in Passau with and without containment measures.

As a result of the simulation, an infested area in case of an introduction via Frankfurt am Main of 475,740 ha without containment measures and 23,107 ha with was calculated for a period of ten years. In the second case of "natural spread" from Passau, calculations gave 358,691 ha without containment measures and 118,419 ha with for the same period.

Key words: *Diabrotica virgifera virgifera*, Western corn rootworm, simulation model, spreading scenarios.

INTRODUCTION

Beginning with the discovery of an economic population of Western corn rootworm (WCR) in Serbia at the beginning of the 1990's, WCR has spread continuously and infested many countries of Southeast Europe (Hungary, Croatia, Romania, Bosnia-Herzegovina, Bulgaria, Slovakia, and Ukraine). In 1998 and 2000, there were introductions into Italy and Switzerland, which are now under eradication/containment programme. These cases show that introductions into other maize growing EU member states are highly possible. With the help of a computer model, it is feasible to simulate different spread scenarios. Two cases are presented here to assess the possible introduction and the potential spread of WCR into Germany from infested countries. The simulations allowed for calculating infested areas under different locations and conditions. For the long-term, economic damage and costs for plant protection can be estimated. Additionally, cost/benefit analyses for containment measures can be carried out.

Method

The simulation model for spreading scenarios of WCR is based on the GIS software ArcView/ArcInfo (ESRI). The calculation is based on grid processing of ARC-Macro-Language with a grid extent of 250 X 250 m. The programme checked each grid for kind of crop for further processing.

The following spreading parameters are considered: 1) spreading rates of populations per year, 2) influence of possible containment measures (i.e. phytosanitary measures), 3) concentration of maize (analysis of high-risk areas), and 4) topography (elevation of mountains).

The dispersal rate of WCR in Europe was analysed starting with the discovery of the economic WCR infestation in Serbia at the beginning of the 1990's. Dispersal rates of the WCR differed from year to year. The spreading of the WCR ranged from 60 to 100 km per year without containment measures and from 0 to 37 km with (FAO programme TCP/RER/6712). The simulation model used as an average a maximum WCR population-spreading rate of 80 km per year without containment measures and of 20 km per year with.

The maximum spreading rate is reached by WCR in the succeeding year only if continuous maize is available in the infested area. The concentration of maize in crop rotation is the main factor in the simulation model. In case of low maize concentration, the multiplication factor and spreading pressure are very low. In this case, the spreading rate was reduced by a correction factor **K,** which is defined as follows:
In case of >= 50 % of maize in crop rotation **K = 1** and in case of < 50 % of maize in crop rotation:

$$\mathbf{K} = \frac{\text{concentration of maize in \%} \bullet 2}{100}$$

The following formula was used in the simulation model to calculate the spreading rate of the WCR:

$$\mathbf{AR} = \mathbf{FD} \bullet \mathbf{K}$$

where **AR** = spreading rate of the WCR
FD = distance of flight with containment measures (20 km/year) or without (80 km/year)
K = correction factor (see above)

Furthermore, the topography was analysed in the infested areas of Southeast Europe. Analysis showed that the WCR is not able to fly over altitudes of 900 m, which was considered in the simulation model. The lowest mountain chain in Western Europe is up to 800 m and has valleys (often with maize), which favour progressive dispersal.

All appropriate information is utilised in the WCR spreading simulation model. This simulation model was used for the first time to simulate the spreading rate in Germany over ten years. Two scenarios were chosen: 1) introduction via the international airport of Frankfurt am Main with and without containment measures and 2) progression of "natural spread" via Austria with the starting point in Passau (Southeast Bavaria on the Danube river at the border to Austria) with and without containment measures. With the software ArcView/ArcInfo, different spreading scenarios of WCR were simulated on maps.

The damage of the WCR is expected in the 5[th] to 7[th] year, which is in agreement with findings from the USA and Hungary (C. R. Edwards and J. Kiss, personal communication).

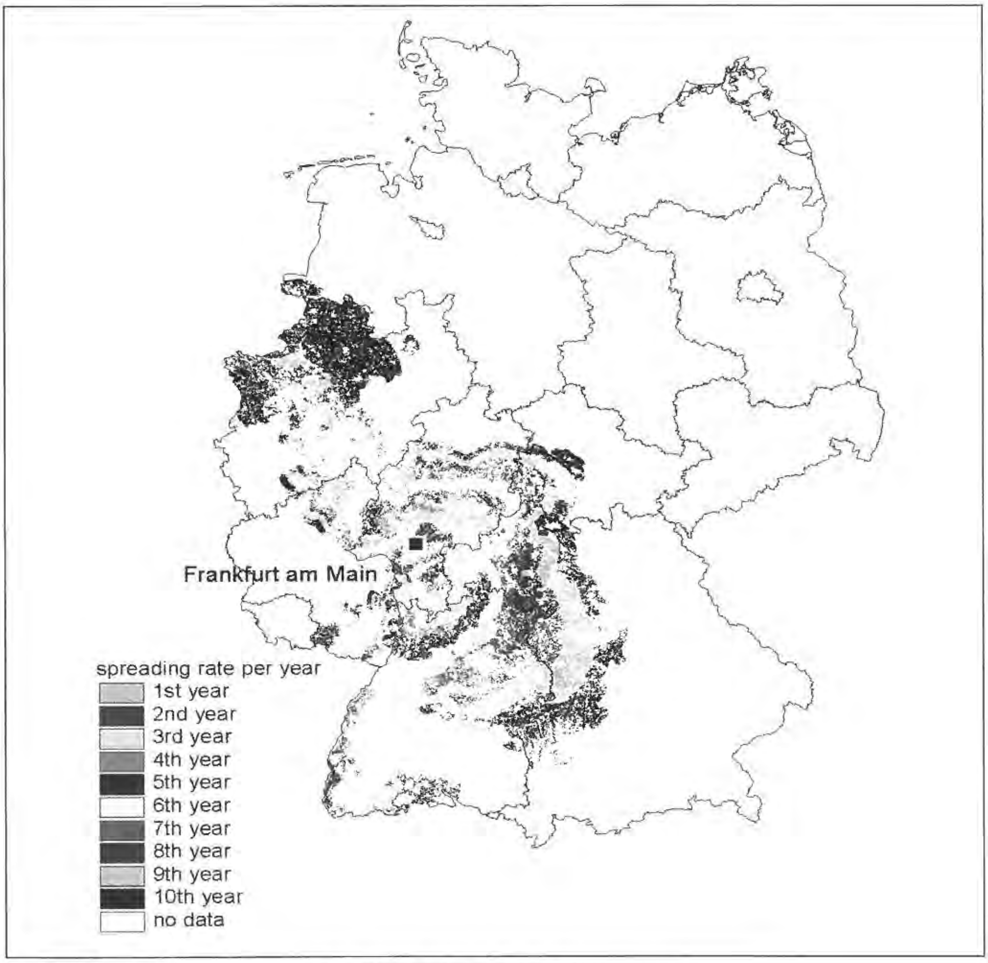

Figure 1 - Simulation of spread without containment measures

Table 1 - Result of simulation of spreading of WCR (infested area) for different scenarios in Germany after 10 years

Year of infestation	Infested area (ha) Frankfurt without containment measures (max. 80 km)	Infested area (ha) Frankfurt with containment measures (max. 20 km)	Infested area (ha) Passau without containment measures (max. 80 km)	Infested area (ha) Passau with containment measures (max. 20 km)
1.	431	376	28,320	293
2.	10,931	1,291	67,152	2,376
3.	10,908	2,097	33,804	6,806
4.	9,698	3,018	43,074	6,175
5.	16,570	4,174	42,431	21,586
6.	19,310	3,659	40,318	30,313
7.	30,343	2,687	21,060	18,444
8.	48,869	2,204	36,129	13,622
9.	62,982	1,686	25,628	10,803
10.	265,698	1,915	20,775	8,001
Sum	**475,740**	**23,107**	**358,691**	**118,419**
Percentage	**100 %**	**4.8 %**	**100 %**	**33 %**

RESULTS

In Germany, of about 1.5 million ha of maize, 348,000 ha are under high risk for WCR infestation assuming that regions with more than 50% of arable land in maize are at high risk. In the four Federal lands of Niedersachsen, Nordrhein-Westfalen, Bayern, and Baden-Württemberg, there are high frequencies of continuous maize. This area amounts to one-third of the maize production area of the above mentioned Federal lands and more than one-fifth of the entire maize production area of Germany (Schaafsma *et al.*, 1999).

These high-risk areas in Germany are the preconditions for high multiplication and maximum spread. Simulation model output in case of an introduction via Frankfurt am Main without containment measures showed that 475,740 ha of maize area (100%) would be infested over a period of ten years (*table 1, figure 1*). With containment measures applied, 23,107 ha would be infested (4.8% of the infested maize area over ten years).

In the case of Passau, the "natural spread" calculation amounted to 358,691 ha of maize (100%) infested over ten years (*table 1*). Containment measures could reduce the infested maize area by one-third (188,419 ha) of the total infested area for the same period. The results of simulation of spreading of the WCR at the two locations are different by year depending on the maize concentration in crop rotation. In the 10^{th} year without containment measures in Frankfurt, a significant ongoing spread of 265,698 ha of maize occurs (*table 1, figure 1*). The reason for this is the high maize concentration in Nordrhein-Westfalen and Niedersachsen, which supports the spreading. In Passau, the conditions for the establishment of WCR are better because the amount of maize makes this a high-risk area. Therefore, the influence of containment measures is not as strong (33%) as in Frankfurt am Main (4.8%) where we have only a low concentration of maize at the beginning.

Under the assumption of damage after the 7th year, the results of the simulation of damaged area in case of an introduction via Frankfurt am Main shows 66,031 ha (100%) without containment measures and 12,589 ha (19%) with after three years. In Passau, calculations gave 425,418 ha (100%) without containment measures and 28,087 ha (6.6%) with for the same period. Potential economic losses would correspond to high-risk areas with more than 50% of maize in crop rotation.

Conclusion

In case of a local introduction, such as Frankfurt am Main, eradication measures should be enacted as soon as possible to avoid WCR establishment (see Italy). In case of unsuccessful eradication or area-wide ongoing of infestation (see case Passau), containment measures should be enacted. The use of containment measures could have an enormous influence on reducing the spread of the WCR and potential damage to maize depending on the local situation. Areas with high concentrations of maize in rotation with other crops have the best conditions for fast ongoing of spread and damage than under slow spreading conditions. The fast spread and the amount of economic loss after seven years of WCR build-up are evident. Therefore, measures that reduce the spread of WCR will reduce the loss observed after seven years.

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ANALYSIS OF A NOVEL FORMULATION OF THE PLANT KAIROMONE MIMIC 4-METHOXYCINNAMALDEHYDE (MCA) USING UV SPECTROMETRY

Wennemann L., Hummel Hans E.,
Institute for Phytopathology and Applied Zoology,
Biological and Biotechnical Plant Protection,
Heinrich-Buff-Ring 26-32, 35392 Giessen, Germany

Ujváry I., Bándiné-Barlai A.,
Hungarian Academy of Sciences, Chemical Research Center,
Institute of Chemistry, P.O. Box 17, 1525 Budapest, Hungary

Sipos P., Szücs-Tóth K.,
Department of Inorganic and Analytical Chemistry,
University of Szeged, P.O. Box 440, 6701 Szeged, Hungary

Abstract
A number of field experiments have already been conducted to evaluate the impact of the plant kairomone mimic MCA (methoxycinnamaldehyde). MCA disrupts orientation of adult western corn rootworm *Diabrotica virgifera virgifera* LeConte (Coleoptera: Chrysomelidae) after application to corn in Hungary and Yugoslavia. Preliminary studies indicated that the application of the non-toxic MCA in corn fields decreased orientation towards MCA - and pheromone-baited traps. Permeation of a corn field with this synthetic substance might mask the female sex pheromone. However, the mechanism of action so far is completely unknown.

For the formulation MCA was dissolved in acetone and blended with corn cob granular material ('grits') using a cement mixer. Grits, available in different sizes, are a by-product after corn is harvested and separated from the cob. In 2000, mixing experiments were conducted with different amounts of acetone, various spraying methods (fog or jet) and in a covered or open mixer to identify the optimal formulation technique. Grit samples were taken immediately after formulation and on regular intervals for 10 days after grits were exposed to the atmosphere. In 2001, grit samples were taken directly after the formulation and over several days after application in a corn field. The residual MCA on the grits was determined by extraction with ethanol and subsequent UV-spectrometric analysis at 320 nm. Aerial MCA was absorbed on a charcoal filter within a glass tube attached to an air pump (so called OLSA, open loop stripping analysis) after field treatment at 80 g/ha in a 12-m^2 sub-plot.

MCA release rates were essentially the same for the different mixing techniques. Grit extraction and analysis showed that over 85% of the applied MCA can be recovered from the formulation. The MCA content of the grit formulations was demonstrated to decrease exponentially with a ca. 2.5 days half life for the experiments in 2000 and 2001. Furthermore, additional experiments were carried out to determine MCA in the atmosphere. The MCA collected in the charcoal filter was below the UV-spectrometric detection limit necessitating the development of a more sensitive analytical method.

Key words: 4-methoxycinnamaldehyde, quantitative UV-spectrometry, formulation, *Diabrotica virgifera virgifera* LeConte, western corn rootworm (WCR), kairomone.

INTRODUCTION

The Institute of Phytopathology and Applied Zoology (Justus-Liebig University, Giessen, Germany) has conducted different field experiments in Hungary and in Yugoslavia in order to study orientation disruption of the western corn rootworm (WCR) (*Diabrotica virgifera virgifera* LeConte, Coleoptera: Chrysomelidae) in corn crops (Wennemann & Hummel 2001a, Wennemann & Hummel 2001b, Wennemann *et al.* 2001, Wennemann & Hummel 2002). One very promising alternative of enhancing the protection of corn from the pest includes the application of substances with species selective chemical signal function during the mating season of the insects. The principle of such treatments is that certain organic substances, mimicking plant kairomonal odours, cause orientation disruption. The mechanisms of disruption by sex pheromones have been investigated repeatedly in other insect species but have not been elucidated for kairomones and for WCR. Thus, in a corn field which is sufficiently permeated, the male insects become disoriented and therefore mating might significantly decrease. Preliminary studies indicated that the application of the non-toxic 4-methoxycinnamaldehyde (MCA) in corn fields significantly decreased orientation towards MCA and pheromone baited traps.

In order to quantitatively characterize the effect of MCA treatment, the determination of the concentration of MCA appeared to be necessary. MCA has a strong UV absorption exhibiting a broad band with a maximum molar extinction coefficient of $\varepsilon_{320} = 28800\ M^{-1}\ cm^{-1}$ at ~320 nm and a weaker shoulder at ~230 nm. The strong UV-adsorption at 320 nm enables the concentration of MCA to be accurately determined in solutions. The objectives of the work performed at the Chemical Research Center, Hungarian Academy of Sciences and the Department of Inorganic and Analytical Chemistry, University of Szeged, included: 1. the optimization of the calibration and the UV spectrophotometric determination of MCA-concentrations in ethanolic substances, 2. the determination of MCA-concentrations in MCA-treated corn grits (carrier for the MCA), to determine the MCA-content immediately after formulation and to qualitatively characterize the rate of MCA-release from corn grits as a function of time and 3. the determination of MCA-concentrations in the atmosphere within the MCA-treated corn fields using direct air sampling.

MATERIALS AND METHOD

Formulation 2000

For the formulation of the MCA, a novel mixing technique was investigated: a portable cement mixer was used to blend MCA dissolved in acetone as a co-solvent with corn grits, a by-product after corn is harvested and separated from the cob. Degrained corn cobs are ground and sieved to standard size. They are used as animal litter, isolation material, industrial absorbent and as a carrier material for a variety of pesticides. They are readily available, cheap, have a large surface area and are therefore a useful tool for MCA application. A 1000 ml plastic hand sprayer, a jar with holes punched in the lid and a modified oil can served to apply 80 g to 120 g MCA/ha on 18 kg corn granules/ha. The acetonic solution of MCA was sprayed directly into the rotating mixer filled with the grits. Experiments with a variety of different mixing procedures were used to identify optimal technique: 1. a volume of 300 or 600 ml of acetone was used to mix with MCA, 2. the mixer was covered with a plastic sheet or left open to investigate influence of an open or closed mixer, 3. the acetonic dilution was applied with a plastic sprayer via jet or atomising technique. Grits were then exposed in plastic saucers to environmental conditions. Samples were taken 10 days from each of the variants to assess MCA release rates via spectrometry. For analysis, MCA-containing corn grits were formulated in the laboratory using a rotary evaporator (atmospheric pressure, open outlet to allow evaporation of acetone).

The samples were stored below –15°C until use. One gram of corn grit was placed in a 30-ml sintered glass filter funnel (G3, 2.5 cm diameter) and solubles were eluted at ambient temperature with analytical grade ethanol (5 x 10 ml during one hour) into a 50-ml volumetric flask. The volume of the eluent was finally adjusted to 50 ml. One ml of this solution was then diluted tenfold for analysis. MCA content was measured by ultraviolet spectroscopy at 320 nm, i.e. the UV maximum of dilute ethanolic solution of MCA (ε_{max} 28,800). During UV analysis eluent from the appropriate control grit sample, not containing MCA, was used as reference (this eliminated any ethanol-extractable UV active "background" material which was nevertheless shown to have absorbance value <0.08 at this dilution). Estimation of the amount of eluted MCA-residue was based on a calibration "curve" obtained by similar analysis of grits freshly coated with known amounts (1-10 mg giving essentially linear concentration dependent absorbance from ~0.02 through 1.73) of MCA in acetone. Residues were air-dried by rotation at ambient temperature for 30 min. In general, analyses were done with at least duplicate grit extracts from four charges (*table 1*).

Calibration 2001

For the UV measurements a Hewlett-Packard 8452A diode array spectrophotometer and a standard quartz cuvette (path length 10 mm) were used. Spectra were recorded over the 200 – 800 nm wavelength range. A 200 mg L^{-1} ethanolic stock solution was prepared from solid MCA and analytical grade ethanol (Reanal, 98%). The stock solution was stored in a refrigerator at 5°C and was used for the preparation of the calibrating solutions. No sign of decomposition of MCA was observed during the experimental period (ca. 60 days). Ethanolic solution series with $[MCA]_T \leq 4$ mg L^{-1} were prepared by accurate dilution for each individual experiment. Aqueous solutions with similar MCA concentrations were also prepared and measured and gave essentially the same results. A blank solution with no MCA was also measured and all the spectra recorded were corrected for the actual background. As expected, the absorbances at 320 nm obtained this way showed linearity with MCA-concentration. The slope of the calibration line was found to be around 0.130 l mg^{-1} and exhibited only insignificant variation in time (ca. ± 0.003 l mg^{-1}, corresponding to ca. ± 2% reproducibility). From these data it can be estimated that under these conditions the detection limit of MCA is ca. 0.2 mg/l (this is the lower threshold value which is necessary to gain analytically significant absorbance values).

Determination of MCA in the atmosphere 2001

Attempts were made to determine MCA-concentrations in the atmosphere of treated fields. For this, the MCA-containing air was sucked through a stainless steel tube, into which a tightly fitting glass tube with charcoal was inserted (CLSA filter, Gränicher, Le Ruisseau de Montbrun, France). Charcoal is known to absorb MCA efficiently. The absorbed MCA can be readily extracted with each μl of a 2:1 mixture of dichlormethan/methanol. If the absorbed MCA is linearly proportional to the MCA concentration in the air, then it can be quantified. However, for such determinations sufficient amounts of MCA have to be accumulated on the charcoal and the MCA concentration in the extract has to exceed the spectrophotometric detection limit. Since the size of the charcoal containing tube and the power of the micro air pump (FürGut, Eichstetten, Germany) could not be changed, the only remaining variable experimental parameter was the pumping time which is directly proportional to the volume of the air flowing through the absorber.

In order to determine in the laboratory if MCA can be detected, a commercial aluminum coffee filter was tightly attached to the inlet of the charcoal tube. A sufficient amount of corn grits (10 g) treated with MCA (0.4 g/100 g) was placed in the coffee filter and air was sucked through it for 1 hour. For reference, corn grits without MCA treatment were also utilized under identical experimental conditions. After the treatment each charcoal charge (2 g) was extracted as described above. The extracts were suspended in a minimum amount of ethanol (5 ml) and their spectra were recorded.

Next, samples were collected from the field. Air pump and absorber were placed in the center of a 12 m^2 sized plot. The inlet of the apparatus was laying on the ground in close proximity of the grits on the ground. The field was treated with the usual charge of MCA treated corn grits (80 g MCA/ha, 20 kg grits/ha). Air pumping was carried out for 1 and 2 hours. Extraction of the MCA was performed as described above.

MCA content of corn grits immediately after formulation 2001

In the formulation, a known amount of MCA was added in the form of acetone solution to 20 kg of corn grits and thoroughly mixed in a cement mixer. Usually 5 or 6 samples were taken from different parts of the mixture. Two charges were prepared for test experiments: 1. Kardoskút (Kardoskút 2 and Kardoskút 3, see *table 1*), prepared on 17 July using 320 g of MCA and 1 August using 160 g MCA respectively) and 2. Ruski Krstur (Ruski Krstur 2 and Ruski Krstur 3, see *table 1*) prepared on 20/7/2001, using 320 g of MCA and prepared on the 8/8/2001, using 160 g of MCA respectively. The MCA content of the MCA-treated corn grits was determined according to the following procedure: The corn grit samples were placed in glass vials with polyethylene lids and were stored and delivered to the analytical laboratories in liquid nitrogen. Usually sample treatments (*i.e.*, weighing and extraction) started immediately after delivery. In some cases, the samples were stored for a few days in a commercial freezer operating at –17°C. Ca. 500 mg of corn grits were accurately weighed on an analytical balance (Sartorius, accuracy: ± 0.1 mg) directly into sealable glass vials. The MCA contents of the grit samples were extracted with 4 or 5 ml of absolute ethanol for at least 48 hours. Longer extraction times were also utilized for some selected samples but were found to cause no significant variation in the obtained analytical data. For the spectrophotometric measurements usually 20 or 25 µl samples were taken from the extracts with an adjustable Eppendorff pipette (maximum capacity 200 µl, accuracy ± 0.1 µl), injected into a 10 ml volumetric flask and set to the mark with 'conductivity water' obtained from a Millipore-MilliQ system. The applied extract volumes were appropriately changed if the observed absorbance at 320 nm was smaller than 0.05 or larger than 0.5.

MCA concentration of field samples 2001

MCA treated corn grit samples (Kardoskút 2 and 3) with a theoretical MCA loading of 16 and 8 mg/g of grits, respectively, were applied on the fields and collected in plastic dishes. After application, samples were collected at regular intervals (*e.g.*, daily) and their MCA content was determined spectrophotometrically.

Results

Formulation 2000

Comparing the different formulation techniques, namely spraying MCA solutions of two different concentrations either as a jet or fog in covered or opened mixing machine, it appears that there is no substantial difference between the various formulations. Preliminary laboratory experiments indicated that 3x10 ml ethanol extracted >85% of MCA from the corn grit. It is notable that parallel 1-gram extractions could give up to 20% differences in estimated MCA content. Using larger amount (3-5 g) of grit samples could presumably improve variance.

The change of MCA-content of the grits over time is similar for all formulations. The attractant is released more or less evenly for all cases. During the first 6-7 days the amount of MCA released from each formulation is the same (virtual first order kinetic) and can be estimated to be in the range of 0.3-0.5 mg per day for each gram of formulated corn grit. After one week the extractable amount of MCA does not change indicating that any residual amount is strongly bound to the formulation. Interestingly, there is a slight increase in the amount of UV-absorbing material for all but one sample taken on 14/9/2000.

When comparing the different formulation methods, it appears that formulation technique No. 4 (300 ml acetone solution applied as jet, covered mixer) results in a product retaining more MCA (i.e., releasing less) and formulations obtained by techniques Number 6 and 7 (MCA applied in 600 ml acetone) release the attractant faster. Nevertheless, these differences are minor and the conclusion is based on the assumption that residual extractable MCA-content correctly reflects the amount dispersed into the air. Finally, we assume throughout the analysis that change of MCA-content in the formulation is solely due to "evaporation" and no chemical transformation into biologically inactive materials takes place under field conditions.

Determination of MCA in the atmosphere 2001

In the laboratory experiment, the reference sample did not exhibit absorption at the critical 320 nm wavelength. However, MCA exposed samples produced well defined MCA-spectra with significant absorption (0.37 after background correction) at 320 nm. In all field samples MCA concentration was below the detection limit. These measurements demonstrated that the experimental setup requires significant improvements to accumulate sufficient amounts of MCA from the atmosphere (*i.e.*, significantly enhanced pumping power or larger air volumes).

MCA content of corn grits immediately after formulation 2001

The results of the analyses are shown in *table 1*. The MCA concentrations found are expressed in terms of percentage MCA found on the grits, where 100 % corresponds to the (maximum) 16 or 8 mg MCA/g grits values, respectively. The data presented in *table 1* indicate that a significant fraction (sometimes up to ~17%) of the MCA is not accounted for, *e.g.*, somehow it has been lost between the addition and the extraction. Since such losses during storage and extraction are unlikely, it seems that the MCA was lost (due to the volatile nature of the substance) during formulation.

Table 1 - MCA contents (expressed in terms of percentage of the theorectical or maximum value) of corn grits determined immediately after formulation for four different charges

	Charge name			
Sample No.	**Kardoskút 2**	**Kardoskút 3**	**Ruski Krstur 2**	**Ruski Krstur 3**
	MCA content (%)			
1	70.5	64.0	86.9	88.0
2	75.0	103.2	102.0	80.8
3	71.5	96.8	98.7	96.0
4	79.0	98.4	102.9	84.0
5	73.0	88.0	89.8	76.0
6	68.0	-	-	-
Average	**73.0**	**88.0**	**96.1**	**84.8**

Conclusions

A realiable and sufficiently accurate UV-spectrophotometric procedure has been developed for the determination of MCA concentrations in ethanolic solutions.

The detection limit for MCA in solutions was found to be ca. 0.2 mg L^{-1}.

It has been concluded that for the determination of MCA in the atmosphere, direct air sampling combined with adsorption of MCA on charcoal accumulates MCA amounts which are insufficient for UV-spectroscopic measurements.

It was found in the two independent studies in 2000 and 2001 that a certain fraction of MCA applied could not be recovered by extraction. This could presumably be due partly to evaporation during the formulation when an open air cement mixer is applied for the coating of corn grits, partly to irreversible adsorption (binding) to the grit matrix and partly to oxidation of the aldehyde to the acid or its polymerization under the influence of oxygen and light.

The MCA content on the corn grits in the field was found to decrease exponentially with ca. 2 days half life.

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USE OF MCA (4-METHOXYCINNAMALDEHYDE) AS AN ORIENTATION DISRUPTION TOOL FOR ADULT WESTERN CORN ROOTWORM *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE

L. Wennemann, Hans E. Hummel,
Institute for Phytopathology and Applied Zoology,
Biological and Biotechnical Plant Protection,
Heinrich-Buff-Ring 26-32, 35392 Giessen, Germany

Abstract

MCA is an organic substance which is investigated for its potential as an orientation disruption tool with a possible effect as a mating disruptant for the Western corn rootworm *Diabrotica virgifera virgifera* LeConte (Coleoptera: Chrysomelidae). For the formulation a cement mixer was used to blend MCA dissolved in acetone and corn cob granules ('grits'). Grits are a by-product after corn is harvested and separated from the cob.

This year's studies included the improvement of the formulation technology by investigating a) 4 corn grit sizes b) 3 loads of the mixer and c) variation in the amounts of liquid mixing media monitored by adding a dye as a marker. Distribution patterns by collecting grits in plastic dishes and counting grits on corn plants were investigated in the field after aerial and high wheel tractor application. Impact of MCA on beetle orientation in experimental fields sites in Kardoskút (Hungary) and Ruski Krstur (Serbia) were evaluated by comparing number of beetles collected in Yellow Sticky traps, VAR traps, PAL traps and plastic cup traps coated with tanglefoot in the MCA treated and control plots. Females were collected in the treated and control plots after different time intervals. The spermathecae were checked for the absence and the presence of sperm to identify mating status.

Results of the formulation experiments showed that the optimal mixer load is 20 kg grits and MCA dissolved in 1 l of acetone. Both airplane and tractor application are valuable for grit distribution in the field. Preliminary analysis of the data showed variable orientation disruption of adult western corn rootworms. The tanglefoot coated plastic cups proved to be a cheap and excellent trapping device for adult beetles. Single female baited traps revealed variable catching results in both the treated and untreated field. The dissection of the spermatheca did not yet show an effect of the MCA application on the mating status.

Key words: orientation disruption, *Diabrotica virgifera virgifera* LeConte, semiochemicals.

INTRODUCTION

The use of semiochemicals to attract insects to trapping devices, to monitor pests, to mass trap insect pests, to modify insect sexual or social behavior and to disrupt insect communication is well documented (Carde and Minks 1997, Witzgall and El

Sayed 1999). The management approach investigated here is to disrupt orientation of adult *Diabrotica virgifera virgifera* LeConte (WCR). Larvae of the western corn rootworm (WCR) cause feeding damage on corn roots. Plants lodge and growth is stunted (Wennemann and Hummel, 2002). Adult beetles feed on leaves, tassels, silk and immature kernels (Hoffman *et al.* 2000). The disruptant investigated is 4-methoxycinnamaldehyde (MCA). MCA is a composite structure derived from about 20 different floral volatiles present in *Cucurbita maxima* Duchesne (Cucurbitaceae). MCA has been discovered and optimised by Metcalf and Lampman (1989). The management approach as described by Wennemann and Hummel (2001 a) is as follows: in MCA saturated corn fields WCR does not locate either MCA or pheromone traps as readily as in untreated plots. This reduced ability to orientate towards the traps might also have an impact on the ability to locate mating partners in the field. If this is true reduced mating frequency and thus reduced pest populations may be expected.

Experiments were conducted to evaluate the impact of the MCA formulation as a potential new management approach to reduce orientation of WCR populations towards artificial MCA and pheromone sources in Hungary and Serbia. Distribution of many grit point sources coated with MCA is of major importance in order to achieve a uniform MCA permeation of the field.

Materials and methods

Formulation technology

The goal was to optimise mixing efficacy of the grits with the acetone/MCA medium. Therefore, preliminary experiments were conducted with different corn grit sizes (RM 6, RM 12, RM 16, RM 20) in conjunction with different amounts of water (300 ml, 500 and 1000 ml). Three different loads of the mixer (20 kg, 30 kg and 40 kg) in conjunction with food dye were used. Grits were put into a portable cement mixer. A 1000 ml plastic hand sprayer served to spray the aqueous/red food dye onto the corn grits. Percentage of grits with no colour (white), with some red (pink) and covered with red more than 80% were calculated to determine most efficient formulation. For the field experiments a similar formulation technique with MCA dissolved in 1 l of acetone was used. Concentrations of 69.5 g to 273 g MCA/ha on 16.6 kg corn grit carrier material/ha in Kardoskút for three aerial applications on July 4th and 28th and August 3rd were used. Applications in Ruski Krstur (Serbia) were conducted on July 10th , 27th and August 11th. 74.4 g to 256 g MCA/ha on 16.6 kg corn grits were applied with a high wheel tractor.

Experimental sites

The test site in Kardoskút consisted of a 15 ha corn field with corn as monoculture since 1997. The experimental site was divided in two 5.9 ha plots each for the MCA treatment and the untreated plot as well as a buffer zone of appr. 3.2 ha. The test site in Ruski Krstur has each 5 ha for the treated and the untreated plot separated by a 5 ha soybean field. Corn has been planted here for two consecutive years. Both fields had sufficiently large WCR populations as evaluated by trapping experiments.

Application technology

Field application of granules mixed with MCA was accomplished using a Dromader aircraft in Kardoskút (Hungary) and a high wheel tractor in Ruski Krstur. The Venturi system of the airplane delivered approximately 18.3 kg grits/ha and the high wheel tractor 16.3 kg grits/ha, respectively. To the horizontal boom of the tractor 6 triangular metal distributors with internal baffles were attached about 2.5 m above ground to facilitate equal spread of the grits while the tractor was moving slowly through the field. Grit distribution was evaluated directly after each application in the field. 9-11 plastic dishes (30-cm) were positioned at regular intervals in each 4 rows in Kardoskút. Additionally, for each sampling date 10 plants were randomly selected within the proximity of the plastic saucers to count grits on leaves and leaf axils. The experimental evaluations in Ruski Krstur were set up the same way but only two rows were used to collect and count grits.

Orientation disruption evalation

The effect of the MCA application was evaluated by the comparison of the number of beetles caught in a variety of traps. For the evaluation the following trap types were used:

- plastic cups coated with tanglefoot and baited with MCA,
- plastic cups coated with tanglefoot and baited with pheromone,
- sticky traps PAL baited with pheromone,
- funnel traps VAR baited with pheromone/floral bait,
- yellow sticky traps.

Cup traps consisted of inverted plastic cups (500 ml, Polarcup, Hungary) with a pheromone or MCA lure put on chromatography paper on top of the cups. PAL and VAR traps were provided by M. Tóth (Hungary), the yellow sticky traps were provided by J. Kiss (St. Istvan University, Gödöllő, Hungary).

The orientation disruption is calculated according to Roelofs and Novak (1981). Beetles in each trap type in the MCA treated and untreated plots are compared and the percent orientation disruption is calculated. The hypothesis is that lower beetle numbers caught in MCA treated plots are due to interference by MCA which as a strong kairomonal mimic reduces the ability of the beetles to locate the traps in comparison to the untreated control plots. Here the beetles can locate point sources of the odour readily without interference by the MCA.

$$\left[\frac{\text{beetles in control plot traps - beetles in treated plot traps}}{\text{beetles in control plot traps}}\right] \times 100 = \%\ \text{orientatiœn disruption}$$

Results

Formulation

Preliminary experiments with red food dye as a marker were useful to identify the best formulation technology for grits (size 12 RM). Only partial results are presented here. With increasing amount of mixing medium the number of grits covered with dye increased. Mixing efficacy differences between the 500 ml and 1000 ml dye/water medium are minor and only a slight difference between uncoloured white and red grits is apparent. Results have important implications on proper formulation of the grits with acetone and MCA for field evaluations of the MCA. The optimal formulation is the use of 20 kg in the mixer and MCA dissolved in 1 l of acetone as shown in *table 1*.

Table 1 - Average number of differently coloured grits after applying various amounts of mixing media (water + dye) to a 20 kg charge of grits in a portable cement mixer (Hodmezövásárhely, Hungary 2001)

grit coloration	amount of mixing medium (water)		
	300 ml	**500 ml**	**1000 ml**
white	26	10	4
pink	59	66	65
red	15	24	31

Application technology

Two application techniques have been used: aircraft and high wheel tractor. Grits collected in plastic dishes as well as the count of the grits on leaves after application demonstrated that grits were distributed over the whole fields in Hungary and Serbia. Aircraft application was more even than high wheel tractor application (*table 2*).

Some inconsistency is apparent between the number of grits counted in plastic dishes and on corn plants and the application rate. Some mechanical problems with the high wheel tractor contributed to the variability. The application had to be stopped several times in order to fix the application device.

Variation in collected grit numbers in the field will be eliminated in the coming field season by increasing further the aircraft height during application. Airplane propeller and wings cause air disturbance and influenced grit distribution patterns which can not be eliminated.

However, due to the bigger size of the treated fields in comparison to 2000, the variability has decreased since the pilot did not have to open and close the valve

Table 2 - Average grit numbers collected in plastic dishes and grits counted on corn plants after aerial application in Kardoskút and high wheel tractor application (Ruski Krstur 2001)

application site and date	Kardoskút 4th July	Kardoskút 19th July	Kardoskút 3rd August	Ruski Krstur 10th July	Ruski Krstur 27th July	Ruski Krstur 10th August
average number grits/dish	13.6 n=44	15.9 n=44	16.68 n= 27	29.06 n=16	30.62 n=16	17.5 n=16
average number grits/plant	4.4 n=160	9.1 n=160	6.1 n=160	4.3 n=160	8.7 n=160	6.6 n=160

releasing the grits as frequently as in 2000. The frequent opening and closing negatively effects precision of the grit deposition in the field. (Wennemann *et al.* 2001b, Wennemann & Hummel 2001c). Proper analysis of the data is on the way.

Orientation disruption

The effect of the MCA treatment was evaluated by monitoring beetle counts in a variety of both pheromone and/or MCA baited traps in each of the MCA treated and untreated plots. Plastic traps coated with tanglefoot and baited with pheromone as well as PAL sticky traps showed excellent results. Traps lured with MCA or with floral baits were less effective. VAR traps were largely inefficient as a trapping tool but, in our research, no pesticides strips were included in the trap body. Plastic cup traps are inexpensive, omni-directional, quick and easy to prepare and attract beetles very sensitively.

Preliminary analysis of the field experiments on both the two test sites in Kardoskút and Ruski Krstur using the particular MCA formulation showed variable orientation disruption. A reduction of variability in orientation disruption in comparison to results from the field season 2000 was not apparent although the size of the treated area, the application rate of the disruptant, the optimising of the mixing technique as well as the improvement of the distribution patterns of the grits has been achieved. As results from the chemical analysis have shown, MCA is released form the grits too readily and therefore the permeation of the corn field is too short lived. Therefore, efforts are increased in order to improve MCA longevity in the field. MCA coated grits release the disruptant within approximately 6 days thus permeating the air space. MCA evaporates from the grits and influences beetle behaviour.

Formulation for research in 2002 has to be improved to allow a prolonged release of MCA from grits. Experiments are on the way to identify suitable additives for the formulation to prolong MCA release at constant rates.

Conclusions

1. Formulation experiments using a hand sprayer, water/dye, grits and the mixer show good coverage of the grits. This technology has been transferred already to apply MCA dissolved in acetone to cover grits. 2. Distribution patterns in the field show some variability but demonstrate good coverage of the field with grits. 3. Orientation disruption effects between 0-100% have been observed. So far no impact of the mating status of female beetles has been found. However, a shift in sex ratio in favour of females has been recorded.

Acknowledgement to M. Meszes, M. Szabo, F. Szeli and A. Tóth for field help. The work was kindly supported by EU project QLK-CT-1999-01110. Thanks to scientists and staff of the Plant Health Station of Csongrad County in Hodmezövásárhely (Hungary) for general support.

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A 2001 UPDATE ON THE WESTERN CORN ROOTWORM, *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE, IN EUROPE

Jozsef Kiss,
Szent István University, Department of Plant Protection,
H-2100 Gödöllő, Hungary

C. Richard Edwards,
Purdue University, Department of Entomology,
1158 Smith Hall, West Lafayette, IN 47907-1158, USA

Manuela Allara,
Global IPM Facility,
Food and Agriculture Organization of the United Nations,
Viale delle Terme di Caracalla, 00100 Rome, Italy

Ivan Sivcev,
Institute for Plant Protection and Environment.
T. Drajzera 9. P.O.B. 936. 11001Beograd, FR. Yugoslavia

Jasminka Igrc-Barčić,
University of Zagreb, Agricultural Faculty.
Svetosimunska 25. Zagreb, Croatia

Husnija Festić,
University of Sarajevo, Faculty of Agriculture,
Sarajevo, Bosnia and Hercegovina

Ivanka Ivanova,
National Service for Plant Protection Quarantine and Agrochemistry,
Sofia, Bulgaria

Gábor Princzinger,
Department of Plant Protection and Agroenvironment,
Ministry of Agriculture and Regional Development, Budapest, Hungary

Peter Sivicek,
The Central Control and Testing Institute of Agriculture,
Bratislava, Slovak Republic

Ioan Rosca,
University of Agricultural Sciences and Veterinary Medicine,
Bucharest, Romania

Abstract

From the first detection of Western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte, on corn in Europe (Surcin, Belgrade Airport, 1992), individuals and scientific communities in Europe and the USA have been working together to address issues related to the problems, or potential problems associated with this pest and to develop sustainable strategies for its management. The cornerstone of this cooperative arrangement is the sharing of knowledge and experiences for the

benefit of all. Infested countries have increased their capacity to deal with WCR through regional cooperation, while endangered countries have used the results, experiences, and expertise of the infested countries to prepare their farmers, governments, institutes, universities, and input supply industry for the likely introduction of WCR into their countries. The infested countries have made available monitoring, spread, research, and economic data to all interested parties. Most of these data were made available through the WCR Network activity supported by the Food and Agriculture Organization of the United Nations (Letter of Agreement No. PR 19713) and through the cooperation of partners not involved officially in the Network, but cooperating with it.

Key words: WCR, spread, economic adult activity, Europe.

Table 1 - Number of WCR monitoring sites in the WCR Network monitoring activity in 2001

	Fed. Rep. Yugoslavia	Romania	Croatia	Hungary	Bosnia - Herzegovina	Bulgaria	Slovakia	Total
Total number of monitoring sites	240	202	150	84	79	41	58	854
No. of permanent monitoring sites	10	49	10	35	12	8	3	127

MONITORING

Monitoring of WCR allowed for the following:
- determinations as to the occurrence of WCR and spread into uninfested regions;
- gauging WCR population increases over years in the permanent monitoring sites.

Hungarian pheromone traps (Csalomon PAL trap for *Diabrotica virgifera virgifera*, attractive only for males, developed and produced by M. TÓTH, Plant Protection Institute, Hungarian Academy of Science, Budapest, Pf 102, H-1525 Hungary) were used for monitoring the population spread of WCR. Though other WCR trap types have been developed by M. Tóth in recent years, the Network has used the PAL trap since it is relatively inexpensive, simple to use, and capture data from previous years can easily be compared. This type of trap is sensitive enough for early detection of WCR males, even at low population levels.

In order to catch WCR females, Pherocon AM yellow sticky traps with no bait (produced by Trécé Incorporated, P.O.Box 6278 Salinas, California, USA) were used. These traps are mostly used in the USA for field-level population surveys and for WCR management decision-making. Pherocon AM traps are more suitable for use at high WCR population densities than are PAL traps.

In 2001, Network partners placed PAL pheromone traps at the 2000 WCR spread line and beyond, while taking into account the natural barriers or pathways that might increase or decrease WCR spread. Pherocon AM traps were suggested to be placed in cornfields within the infested area. If both trap types were placed in the same field or location, a distance of at least 50 m was kept between them to avoid interference of the pheromone on the PAL trap with the Pherocon

AM trap. Traps were monitored at approximately weekly intervals. New traps were placed at each site after approximately 30 days and between the times of trap replacement, WCR's were removed from the sticky surface after counting. The traps in 2001 were monitored from mid to late June through September, or through the beginning of October at some sites. The counts were recorded for each trap, for each sampling period, and for each trap location, and for the date of the counts.

Results of 2001 monitoring

The spread of WCR in Europe continued in 2001. The total area infested increased by approximately 37% from 182,000 km^2 in 2000 to 250,000 km^2 in 2001 (*table 2*). The greatest spread occurred toward northern and eastern areas, where plains (Hungary, Slovakia and Romania) or river valleys (Mures river in Romania) favor beetle spread.

Table 2 - Infested area and area of economic Western Corn Rootworm activity in Europe in 2001 (FAO/J. Kiss and C.R. Edwards based on data from Bertossa, Boriani, Festic, Igrc Barcic, Ivanova, Omelyuta, Princzinger, Rosca, Sivicek, and Sivcev)

	Fed. Rep. Yugoslavia	Romania	Croatia	Hungary	Bosnia-Herzegovina	Bulgaria	Slovakia	Italy	Switzer-land	Ukraine*	Total
Infested area (km^2)	72 250	60 000	15 500	70 000	13 000	7 000	6 300	4 000	728	1	**248 779**
Area of economic adult activity (km^2)	26 500	11 000	4 000	10 000	0	0	0	0	0	0	**51 500**

* estimate for the Ukraine.

In FRY, Croatia, and Bosnia-Herzegovina, the spread of WCR slowed when compared to 2000 numbers. However, the larger the size of the infested area the greater the possibility of a "jumping-spread" movement of WCR beyond the actual spread line (Croatia and northwest Bosnia-Herzegovina in 2001, Lombardy region in northern Italy along with the canton Ticino in southern Switzerland) (*figure 1*). The jumping-spread ability of WCR poses the risk of the establishment of WCR in regions with suitable conditions for the WCR far beyond the established spread line and thus could result in detection delays.

In 2001, the WCR was first observed in the Ukraine (border triangle of Hungary/Ukraine/Romania) and approached Austria and Slovenia. Austria and Slovenia are likely to be infested in 2002.

The permanent monitoring sites within the infested areas enabled the Network partners to measure the population change/increase/decrease over years. The population of WCR at permanent monitoring sites in 2001, in general, increased in Croatia, Hungary, and Romania by 2-3 times compared to year 2000, while it did not change or it decreased in certain regions in Bosnia-Herzegovina and FRY. The decrease is the consequence of unfavorable weather conditions for WCR in those regions (primarily due to too little precipitation and too high temperatures in 2001). However, agronomic practices (growing of continuous corn or crop rotation) may also impact WCR population levels.

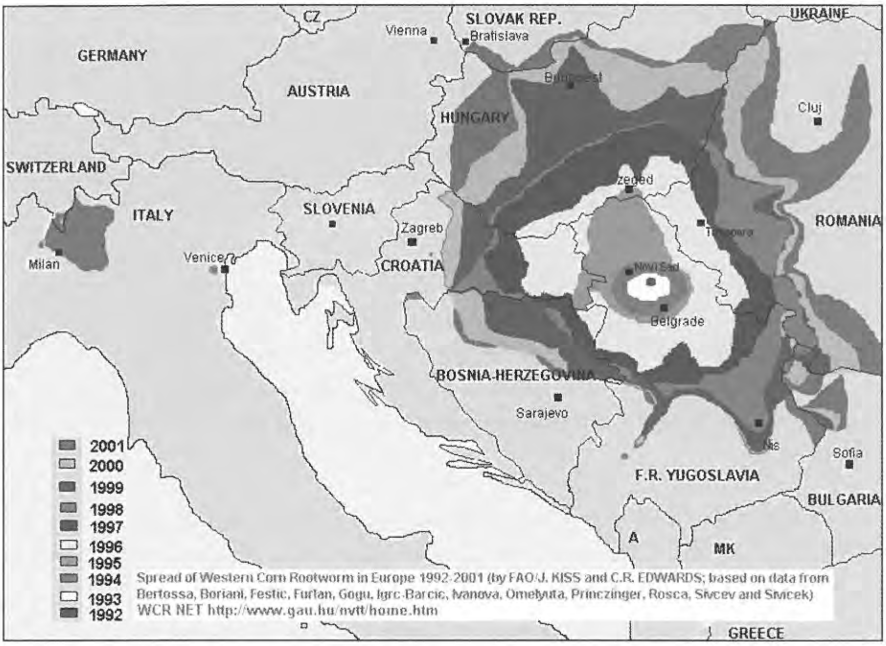

Figure 1 - Spread of Western Corn Rootworm in Europe 1992-2001 (FAO/J. Kiss and C.R. Edwards; based on data from Bertossa, Boriani, Furlan, Gogu, Igrc-Barcic, Ivanova, Omelyuta, Princzinger, Rosca, Sivcev and Sivicek)

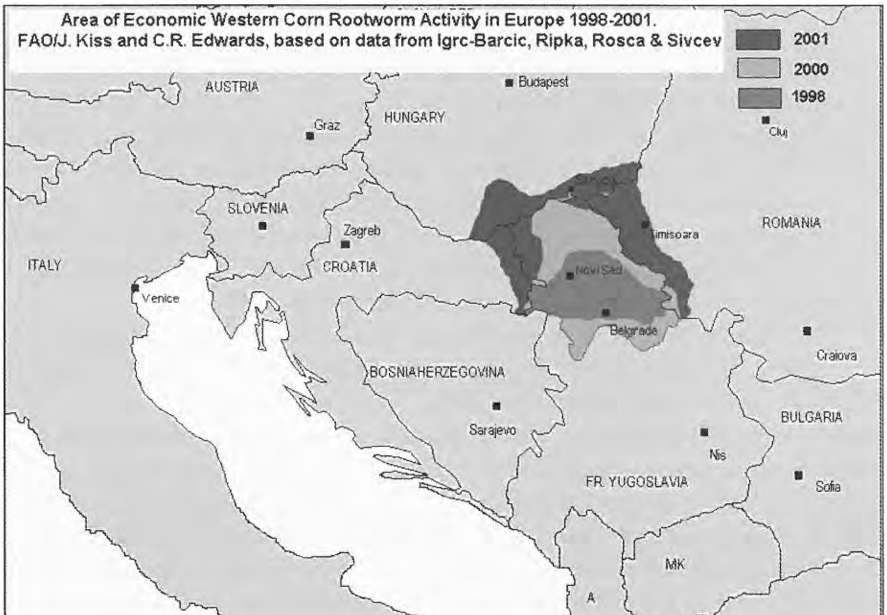

Figure 2 - Area of Economic Western Corn Rootworm Activity in Europe 1998-2001 (FAO/J. Kiss and C.R. Edwards; based on data from Igrc-Barcic, Ripka, Rosca & Sivcev)

The increase of WCR numbers in infested areas is likely to result in economic populations within 5-6 years after the first occurrence of adults. This is demonstrated through the WCR economic activity map in *figure 2*, where the economic area for WCR matches the spread area of 1996. As additional evidence of the presence of economic populations in 2001, larval damage (visible plant lodging) was noted in parts of Croatia, FRY, Hungary, and Romania. The largest area of larval damage was observed in Hungary where root damage ratings in southern counties reached the economic level of 3 (Hills and Peters 1-6 root damage rating scale) on 3,058 ha.

Some conclusions and recommendations

The FAO WCR Network enabled participating countries to continue regional cooperation in Central and Eastern Europe on WCR. This fund is the only external source that is available for all infested countries. Therefore, this support is of crucial importance to the region.

Partners, through the above fund, were able to generate a significant amount of information, which greatly contributed to the body of knowledge on WCR in Europe. This information was useful for the rest of Europe as well.

Due to WCR, the area of corn production has greatly decreased in heavily infested regions of FRY and Romania. This obviously will impact rural communities (both economically and in a social sense) and this has yet to be studied and evaluated. Such studies will be desirable over the next several years.

In the future, further involvement of governments, university communities, young scientists (students), village communities, and farmers is necessary as part of the participatory approach to WCR understanding and management.

Acknowledgements: this update of WCR in 2001 in Europe was made possible through the support of FAO (LoA. No. PR 19713) and with the data received from Lorenzo Furlan (Department of Agronomy, Entomology, University of Padova, Legnaro PD. Italy), Marco Boriani (Lombardy Region, Phytosanitary Service, Milano, Italy), Mario Bertossa (Swiss Federal Research Station for Plant Production, Centre of Cadenazzo, Switzerland) and Victor P. Omelyuta (Institute of Plant Protection, Kiev, Ukraine).

EFFECT OF *THERIDION IMPRESSUM* (ARANEAE: THERIDIIDAE) ON THE SILK CLIPPING OF *DIABROTICA VIRGIFERA VIRGIFERA* ADULTS IN HYBRID SEED CORN IN HUNGARY

***Ferenc Tóth*, Tamás Tuska, József Kiss,**
Szent István University, Department of Plant Protection,
H-2103 Gödöllő, Hungary

Abstract
Hybrid seed corn is more sensitive to biotic and abiotic factors affecting its silk than commercial corn. Silk clipping is the main damage of *Diabrotica virgifera virgifera* adults in corn. Therefore, silking hybrid seed corn can be a sensitive indicator of the actual and potential predatory pressure on *D. v. virgifera* adults. *Theridion impressum* was found to be the most important spider predator of *D. v. virgifera* in Hungary in our earlier studies (Tóth et al. 1998, 1999, 2000).

The present study aimed to determine the ability of *T. impressum* to decrease the silk clipping of *D. v. virgifera* in hybrid seed corn.

The study was conducted in Mezőhegyes, in 2001 (see Tuska *et al.* this volume). 80 corn ears were isolated with ear cages and 48 corn plants with whole-plant cages. The isolated ears were infested with 0, 3, 6 or 9 *D. v. virgifera* adults, and were treated with 0 or 1 *T. impressum* adult female in the latter case (9 beetles/ear) 1-2 days prior to silking (R1). The isolated whole plants were infested with 0 or 9 beetles and were treated as above. Number of replicates was 16 in each case. Silk length was measured daily until the end of silking. Other quantitative and qualitative parameters of the ears and kernels will be measured after harvest in October.

Both the cage types and treatments had a considerable and statistically significant effect on silk length. *Theridion impressum* females could decrease but not terminate silk clipping. Effect on yield parameters and methodological consequences are going to be discussed in details.

This study was partly supported by OTKA F32957 and F. Tóth was a Bolyai Research Grant fellow.

RESEARCHES REGARDING CROP ROTATION, WCR MANAGEMENT AND PEST EVOLUTION IN TIMIS DISTRICT - ROMANIA

Ioan Rosca,
University of Agricultural Sciences and Veterinary Medicine
Bucharest, Bdl. Marasti nr. 59, Bucharest, Romania

Mariana Hincu,
Phytosanitary direction of Timis district

Abstract
During 2001, the Western corn rootworm monitoring in Timis district regarded the symptoms of attacked plant (gooseneck) in field, number of adults/plants, in 81 cornfields, number of adults/pheromone trap and yellow sticky traps in plant protection centers from Jimbolia and Deta. In two cornfields near Jimbolia and Deta, was registered number of WCR larvae/plant and frequency and intensity (1-6 Iowa notes) of root damages. There are other five fields counted for root damages.

Key words: *Diabrotica virgifera*, attack monitorising, crop rotation.

INTRODUCTION

Maize is one of the major crops in Romania as more than 3,000,000 ha/year, covering about 40% of arable fields. Registered for the first time in Romania in 1996 (Vonica, 1996), the pest spread continuously toward east and north (Vonica, 1998 a, b, 2000). The problem of this new pest was analyzed as a possible key pest in Romania (Rosca and Popov, 1999) and strategies for control begin to be elaborated (Rosca, 2000). New zones affected by *Diabrotica virgifera virgifera* in 2001 were 3 new districts in which were registered for the first time, by using pheromone traps, with the pest (Mures, Sibiu, Bistrita-Nasaud) face to 12 districts affected in 2000 (Gogu, 2001). The populations of *Diabrotica virgifera virgifera* have increased strongly, year by year, as indicating by pheromone traps captures reported by Central Laboratory of Phytosanitary Quarantine. For crop rotation and WCR management, relevant conclusions expected until the future years and it is necessary to establish a three-year or four-year crop rotation trial. Sometimes, somewhere the problem of chemical control of fields has arisen, especially due to the necessity of pesticide registration.

FIELD RESEARCH STUDIES ON WCR MANAGEMENT OPTION AND MONITORING OF WCR POPULATION

In 2001, we received Pheromone (60) and Pherocon AM (100) traps after 10 June. Even in these conditions a program based on FAO crop rotation trial started with different cultures (wheat, sunflower, soybean and corn) sowed after corn, in a particular large field near Jimbolia in which the pest was registered in precedent year. Each field has more than 40 hectares. This field will be used in the autumn of this year an in the next year for FAO crop trial rotation in the same way like members of network. After corn harvesting, it will be sowed wheat, like Hungarian plot layout. Three Pherocon AM no bait traps have been placed in the middle of each field and changed weekly. In the same field, separately of Pherocon AM traps no bait, three Pheromone traps and three emergence cages were placed. The results presented in *table 1*, indicate that the most infested fields have been with corn after corn (617.3 adults/pheromone trap, 68 adults/Pherocon no bait trap and one adult/0.5 m^2 emerging from soil).

Table 1 - Captured adults in different field sowed after corn

Culture	Pheromone trap	Pherocone no bait trap	Emergence cage
Corn (67 ha)	617.3	68	1
Wheat (45 ha)	293	1	0
Sunflower (67 ha)	502.3	6.6	1
Soybean (58 ha)	ND	2	0

In this year, there was a selection of cornfield with high WCR population and, in this respect, we choose two fields in the same location in which we made our observations (near Jimbolia and near Voiteg). At the beginning of our activity together with plant protection service staff members, from the end of June until the middle of July, it was searched fields with corn, especially those following corn, taking into account the field size. It were scouted large size fields (more than 10 ha, noted as LS) near Deta (3), Lugoj (2), Periam (2), Varias (2), Sanicolau Mare (1), Ciacova (2), Sanmartinu Sarbesc (1), Lovrin (2). It were scouted 30 small fields (0.5-2 ha) with corn after corn (noted as c/c) and 30 with corn after another culture (noted as c/a), along the main roads Lugoj-Timisoara, Timisoara-Moravita and Timisoara-Jimbolia.

During this scouting activity it was noticed that the pest is present in the district, in some zones, especially in south and south-western part of district, with a density of 0-2.5 adults/plant, during the beginning of July (starting of flight), the adults appearing first in field with corn monoculture There are no high larvae density (< 1/plant) and damaged plants (geese neck) (< 0.1%) (*table 2*).

Exceptions were two fields (near Jimbolia and near Voiteg), in which the next studies were done. Thus in the first field on July 19, 0.73 adults/plant, 0.002% geese neck plants and 0.1 larvae/plant were registered. It is to notice that in a particular zone along the main road on an area of approximately 3 hectares density of larvae, adults and damages produced by pest were 2-10 fold higher on limited 5-20 m^2. In the second field (1.5 ha) on July 26 in average 1.82 adults/plant, 1.2% geese neck plants, and 0.5 larvae/plant were registered.

Table 2 - Presence of WCR in scouted fields

Field	No. of adults/plant	Geese neck plant s (%)	Attacked roots (%)
LS	< 0.01	< 0.001	No evidence
c/c Timisoara-Lugoj	< 0.001	0	No evidence
c/a Timisoara-Lugoj	0	0	No evidence
c/c Timisoara-Deta	< 0.1	< 0.001	No evidence
c/a Timisoara-Deta	0	0	No evidence
c/c Timisoara-Jimbolia	< 0.1	< 0.001	No evidence
c/a Timisoara-Jimbolia	0	0	No evidence

Evolution of captures in Pheromone and Pherocon AM non-bait traps, registered are presented in *figure 1* and 2. From presented data the pest population is higher in Jimbolia zone and adults captured on Pherocon AM non bait traps are higher to the end of the surveying period, level of adult pest population in Jimbolia zone is distributed to the end of observation period and in Deta zone adult flight is earlier but there is no high density, probably due to the fact that field is small and appearing adults migrate towards other fields.

At the middle of September in five fields/districts with corn monoculture it was determined roots attack (there are no evident attacks on Iowa scale, except field from Deta where on Iowa scale note was 2.5/100 plants, but no evident attack in Jimbolia field) and soil samples were taken for determining lying of eggs.

In district Timis, where there is the highest level of population of *Diabrotica virgifera virgifera*, which is maintained in generally at the same level as presented in *table 3*, evolution of WCR was registered during five years and from these data it results that pest population is higher in south and southwestern part of district, but the situation of captures does not reflect the situation of corn plants attack.

Table 3 - Pest evolution in district Timis, registered by number of adults captured/trap

Year	Pheromone traps	Yellow sticky traps
1997	13,167	1,043
1998	7,210	986
1999	23,024	1,086
2000	35,240	910
2001	10,993	854

District Phytosanitary Inspectorate – Timis, by visiting fields with WCR attack, trained farmers and personnel involved in monitoring of Western Corn Rootworm.

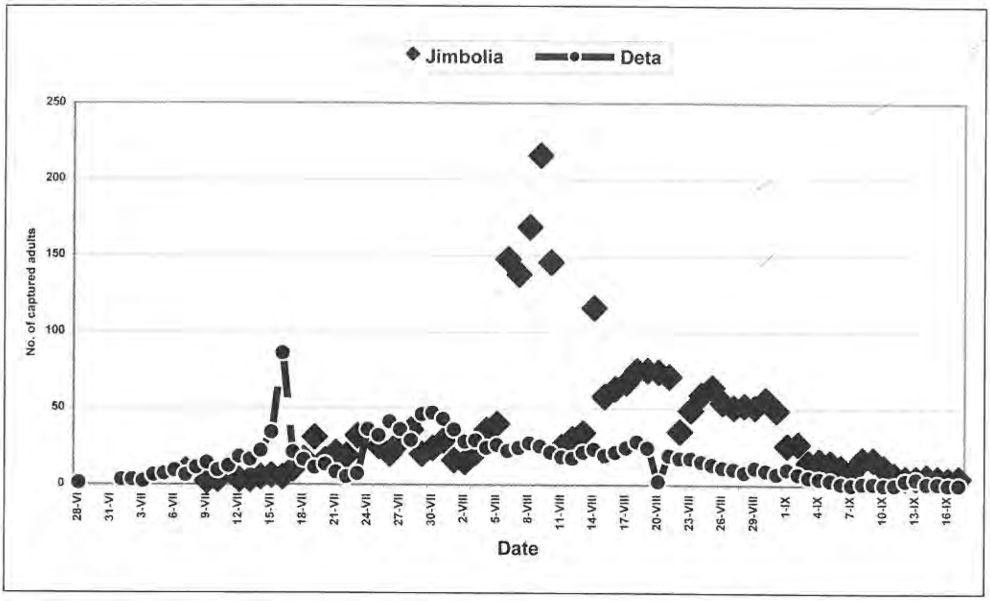

Figure 1 – Dynamic of captures in pheromone traps

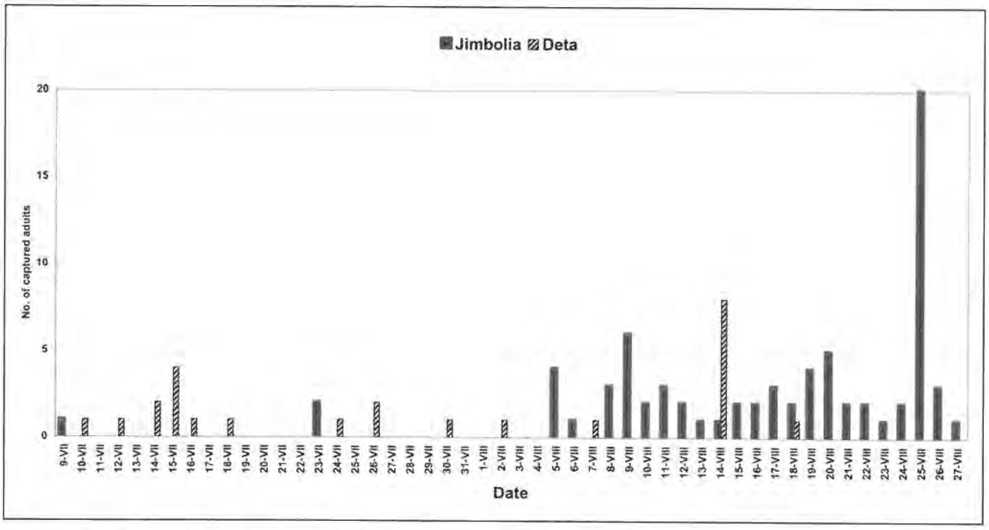

Figure 2 - Dynamic of captures in yellow sticky traps

At the level of General direction for Agriculture Timis, during the several meetings, with responsible with district agriculture, consulting agricultural specialists or directors of agricultural societies, the problem of WCR, its destroying potential for corn fields, was presented each time, not only in this year but from the coming of the pest in Timis district, in 1997, the next year after recording for the first time the pest in Romania. Thus it was possible to reduce the corn area cultivated in monoculture, as it results from *table 4.*

Table 4 - Decreasing of area with corn monoculture in district Timis

Year	Area cultivated corn (ha.)	Area with corn monoculture (ha.)	
		Total	%
1997	126,812	76,902	60.6
1998	130,743	59,946	45.8
1999	127,407	56,569	44.4
2000	141,004	53,731	38.1
2001	189,723	22,549	11.9

Conclusions

For crop rotation and WCR management, relevant conclusions are expected in the future years and it is necessary to establish a three-year or four-year crop rotation trial. In this respect, it has to find financial resources in time. Sometimes, somewhere the problem of chemical control of fields has arisen, especially due to the necessity of pesticide registration.

For WCR monitoring program (spread and population increase) it will be necessary to continue along the eastern infested area of WCR in Romania with Pheromone traps, but in zones were existing WCR is certain, we have to focus on the level of pest populations, their damages, scouting on fields on number of larvae/plant or level of damaged roots on Iowa scale and level of laid eggs in a particular field. (Sometime it is possible to be reported a high level of population, heavy losses, in order to obtain some support or pressing for a pesticide registration).

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ECONOMIC THRESHOLDS FOR WCR ADULTS IN SOYBEAN TO PREDICT SUBSEQUENT DAMAGE TO CORN IN INDIANA, USA

Corey K. Gerber, C. Richard Edwards,
Purdue University, Department of Entomology,
1158 Smith Hall, West Lafayette, IN 47907-1158, USA

Jozsef Kiss,
Szent István University, Department of Plant Protection,
H-2100 Gödöllő, Hungary

Abstract

Adult Western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte, sampling methods were tested in soybean fields, and larval damage was evaluated in first-year cornfields located in northwest Indiana from 1997-2000. The sampling methods included Pherocon® AM unbaited sticky traps, cucurbitacin vial traps, Pherocon® corn rootworm (CRW) non-lure traps, and a 38.1cm diameter sweep net. Calculations for the minimum sample size required for 15% and 25% level of precision were determined for each sampling method using Southwood's formula $n = (s/Em)^2$. Corn root damage was compared with analysis of variance procedures and mean separations using Fisher's protected LSD test. Economic thresholds also were determined by regressing the number of WCR adults collected with Pherocon AM and CRW traps in soybean during 1998 and 1999 with corn root damage in fields the proceeding years, 1999 and 2000. Damage to corn root systems was significantly higher in untreated plots than treated plots in 1997-2000. The Pherocon AM traps and CRW traps, at the 25% precision level, required the least number of traps (4.39 and 5.32, respectively) to determine the estimated adult WCR population in soybean fields. The regression equations developed by adult collections using the CRW traps revealed R^2 values of 0.067 and 0.008 in 1998-1999 and 1999-2000, respectively. Adult collections using Pherocon AM traps during the same time periods revealed R^2 values of 0.579 and 0.360. An R^2 value of 0.460 was determined when the Pherocon AM trap data were combined for both trapping cycles. Although a range of thresholds resulted from this experiment, the economic threshold, as estimated by a root damage rating of 3.5, has been determined as eight WCR adult beetles per Pherocon AM trap per day.

Key words: Western corn rootworm, *Diabrotica virgifera virgifera* LeConte, Pherocon AM trap, CRW trap, economic threshold.

INTRODUCTION

Growing corn in rotation with soybean has been widely adopted by corn producers in the midwestern Corn Belt. One benefit of this rotation is the management of corn rootworm larval populations without the use of insecticides, either soil applied for corn root protection or foliar applied to prevent females from laying eggs. However, an apparent adaptation of the western corn rootworm beetle (WCR), *Diabrotica virgifera virgifera* LeConte, to corn grown in rotation with soybean has occurred in northern Indiana and east-central Illinois, as well as parts of Ohio and Michigan, resulting in economically important numbers of rootworm eggs being laid in soybean. Therefore, growing corn in rotation with soybean is no longer a viable option as the sole management tactic against the corn rootworm for producers in this region.

Sampling plans and economic thresholds are available for corn rootworm eggs, larvae, and adults infesting continuous corn. During the 1990's, in Indiana, no sampling plans or economic thresholds were available for use in soybean to determine if a soil insecticide was needed for corn planted the following year (first-year corn). As a result, an estimated 70% of corn producers in northwest Indiana and east-central Illinois applied a soil insecticide to first-year corn to protect corn roots from rootworm injury and/or other soil insect pests. The objectives of this study were designed to: (1) determine damage to first-year cornfields created by WCR populations in northwest Indiana, (2) develop a sampling program for WCR populations in corn/soybean production systems, and (3) to establish economic thresholds for WCR adults in soybean, which can translate to the amount of subsequent WCR larval injury in first-year corn.

Materials and methods

Determining Damage to First-year Corn

In 1999 and 2000, 16 pairs of corn/soybean fields in six northwest Indiana counties were used in this study. A field pair consisted of a soybean field adjacent to a cornfield. Field size varied from 12-47 hectares. Tillage, planting date, and all crop inputs were not controlled in this experiment, with the exception of the soil insecticide application to corn. The number of treated and untreated rows or strips (continuous multiple rows) in corn depended on (1) the size of the planter used and (2) if the producer wanted to make a yield comparison between treated strips and untreated strips. In fields where check strips were established, producers would either shut off the entire planter or half of the planter while making a pass in the field. Similar passes were made at different points in the field. Check strips were identified by flags. Treated rows received a full rate of soil insecticide (Counter® 20CR, Lorsban® 15G, Force® 3G, and Fortress® 5G) at planting time.

In 10 fields, only one pair of rows was identified as the sampling block. In six fields, where two check strips were established, one pair of rows within each of the check strips was identified as the sampling block. Each block consisted of adjacent treated and untreated rows.

Corn roots were dug from early July to late August. Fifty roots were dug from each treated and untreated row, for a total of 100 roots per field in the fields that contained a single sampling block of one pair of rows. Twenty-five roots were dug from each treated and untreated row, for a total of 100 roots per field in the fields that contained two sampling blocks of one pair of rows. Roots were washed to remove soil and rated using the Hills and Peters (1971, J. Econ. Entomol. 64:3) root damage 1-6 rating scale.

Development of a Sampling Program in Soybean/Corn Production Systems

Adult rootworm populations were estimated weekly, from mid July until early September in 1997, 1998, 1999, and 2000. In 1997 and 1998, four sampling methods at six paired soybean/corn fields in Benton County, IN were used to collect WCR adults. Sampling methods included: 1) the Pherocon® AM unbaited sticky trap (Trécé Incorporated, Salinas, CA 93912); 2) a cucurbitacin vial trap; 3) the Pherocon® CRW trap (Trécé Incorporated); and 4) a 38.1 cm diameter sweep net. In 1999 and 2000, only the Pherocon AM unbaited sticky trap and the CRW trap were used.

In 1997 and 1998, soybean fields were divided into quarters lengthwise, creating three transects running the length of the field. Thus, three trap lines were established and each line consisted of one of the three trap types. Eight traps were placed equidistant in each line. The cucurbitacin vial traps and Pherocon AM traps were attached to 1.2m wood lath that had been driven into the ground. Traps were positioned at the top of the canopy. These traps were adjusted each week to maintain position relative to the soybean canopy. The CRW traps were positioned on 1.8m x 1.27cm PVC pipes. Eight sweep samples were taken within a field. The samples were obtained in the Pherocon AM trap line in each of the soybean fields and consisted of 30 sweeps. Sweeps were made in a continuous pendulum motion in the upper 1/4 to 1/3 of the soybean canopy.

In 1999 and 2000, the soybean fields were divided into thirds lengthwise, creating two transects running the length of the field. Thus, two trap lines were established and each line consisted of either the Pherocon AM unbaited sticky trap or the CRW trap. Eight traps were placed equidistant in each line. Positioning of the two trap types were conducted in a similar fashion as in 1997 and 1998.

The Pherocon AM traps, the contents from the CRW traps, cucurbitacin vial traps and the sweep net samples, for each field, were placed in plastic bags, returned to the laboratory, and refrigerated. The number of WCR beetles was determined for each sampling method.

Development of WCR Economic Thresholds in Soybean

Adult rootworm populations were estimated by using the Pherocon AM unbaited sticky trap in 16 paired soybean/corn fields. WCR beetle populations were sampled weekly from 14 July to 31 August 1998 and from 19 July to 31 August 1999.

In each of the soybean fields, a trap line was established and consisted of either six or eight Pherocon AM unbaited sticky traps that were placed equidistant down the length of each field. Pherocon AM traps were attached to 1.2m wood lath that had been driven into the ground. Traps were positioned at the top of the canopy. These traps were adjusted each week to maintain position relative to the soybean canopy.

The Pherocon AM traps were placed in plastic bags (by field), returned to the laboratory, and refrigerated. The number of WCR beetles on the Pherocon AM traps was recorded. The number of WCR beetles/trap/week was regressed to the root damage ratings in corresponding fields. The root damage ratings used were from the data obtained in "Determining Damage to First-year Corn."

Analyses

Root ratings were analyzed with analysis of variance procedures, and mean separations were made using Fisher's Protected Least Significant Difference (LSD) test.

In developing WCR sampling methods, the minimum sample size required for 15% and 25% level of precision was determined using Southwood's (1978) formula $n = (s/Em)^2$ where s = standard deviation, E = desired level of precision, and m = mean.

A linear regression was conducted between mean number of beetles collected per trap, during the sampling dates of 27 July through 31 August 1998 and 26 July through 31 August 1999, and the mean root rating numbers extracted in 1999 and 2000, respectively. Due to the low number of beetle counts during the first sampling date, we felt that the second, third, fourth, fifth, and sixth sampling dates provided adequate numbers for a better fit of the regression line.

Results and discussion

Determining Damage to First-year Corn

In 1999, only one of 16 (6%) cornfields had a root rating ≥ 3.0. We consider root ratings at or above 3.5 as economically important. Overall, corn treated with a soil insecticide had an average root rating significantly less than those not treated (*table 1*).

Table 1 - Mean root ratings from 16 northwest Indiana cornfields in 1999

Treatment	n	Rating[a]	SE	P
Treated Row	16	1.63 a	0.06	0.0007
Untreated Row	16	2.13 b	0.12	

[a] Iowa and 1-6 scale; 1 = no feeding, 6 = 3 nodes without roots.
LSD = 0.05.

In 2000, 11 of 16 (69%) cornfields had a root rating ≥ 3.0. As noted in 1999, corn treated with a soil insecticide had an average root rating significantly less than those not treated (*table 2*).

Table 2 - Mean root ratings from 16 northwest Indiana cornfields in 2000

Treatment	n	Rating[a]	SE	P
Treated Row	16	2.11 a	0.10	0.0001
Untreated Row	16	3.21 b	0.20	

[a] Hills and Peters 1-6 rating scale; 1 = no feeding, 6 = 3 nodes without roots.
LSD = 0.05.

The low root damage rating in 1999 for untreated rows was expected due to high larval mortality in 1998, which resulted in low adult numbers during July through September of that year. Due to low adult numbers, fewer gravid females were present to lay eggs; therefore, less rootworm larval pressure was present in 1999.

Development of a Sampling Program in Soybean/Corn Production Systems
Pest management decisions require an accurate estimate of a population and use of economic thresholds. The number of Pherocon AM and CRW traps, cucurbitacin vial traps, and sweep net samples needed to make these decisions was determined for soybean (*table 3*). Minimum sample size required for the four rootworm beetle sampling methods varied depending on sampling date and year. Due to the lower numbers of traps required per field to estimate WCR populations, only the Pherocon AM traps and CRW traps were calculated in 1999 and 2000.

Table 3 - Minimum sample size required for four corn rootworm beetle sampling methods in soybean at two levels of precision (1997-2000)

	CRW trap		Vial trap		Sweep net		AM trap	
Year	15%	25%	15%	25%	15%	25%	15%	25%
1997	11	4	9	4	18	7	15	3
1998	19	7	19	7	29	11	47	17
1999	10	4	14	5				
2000	110	4	18	7				
Ave.	13	5	15	6	24	9	31	10

Sample size (n) = $(s/Em)^2$

Pherocon AM or CRW traps can be used effectively as relative sampling methods for WCR adults in soybean based on the fact that each trap type requires low numbers. Therefore, for soybean fields, five Pherocon AM traps or six CRW traps are recommended (at 25% level of precision).

Development of WCR Economic Thresholds in Soybean
Regression analyses were conducted on 1998-1999 and 1999-2000 data. Results show that when trapping methods (in 1998 and 1999) were regressed against root ratings for each of the fields (in 1999 and 2000), it was determined that the Pherocon AM unbaited sticky trap was the best predictor of subsequent corn rootworm root damage. The regression equations revealed R^2 values of 0.579 and 0.360 in 1998-1999 and 1999-2000, respectively (*figure 1* and *2*), for Pherocon AM traps. Adult collections using CRW traps during the same time periods revealed R^2 values of 0.067 and 0.008, respectively (*figure 3* and *4*). The regression analysis in *figure 5* indicates that the threshold value determined in 1998-1999 and 1999-2000 was eight beetles/trap/day with an R^2 value of 0.460.

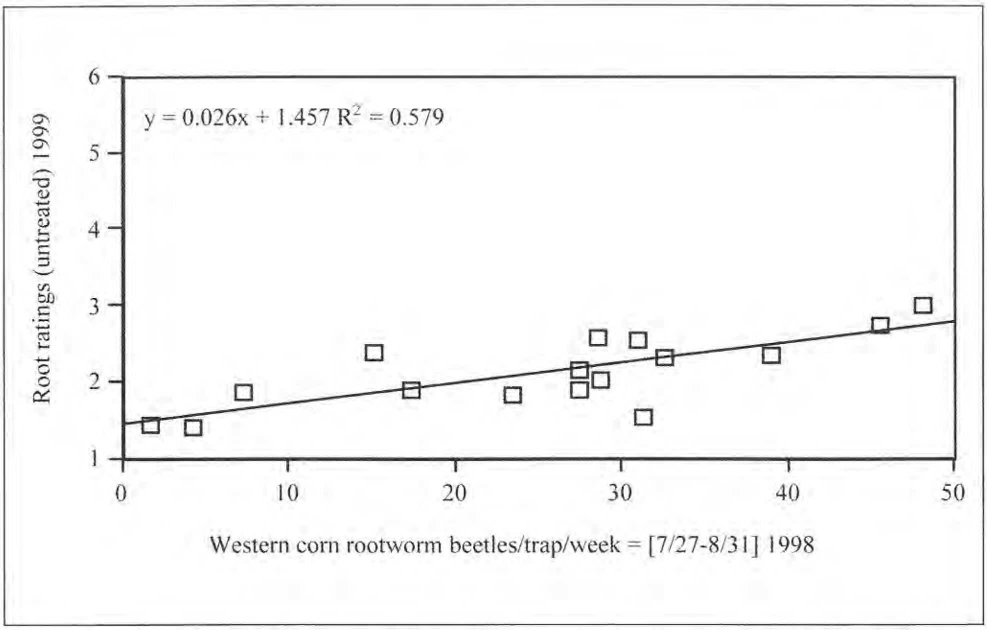

Figure 1 - Regression analysis of mean WCR population estimates/trap/week from 16 Indiana soybean fields using Pherocon AM traps from 7/27-8/31, 1998 versus root ratings taken in these same fields during 1999

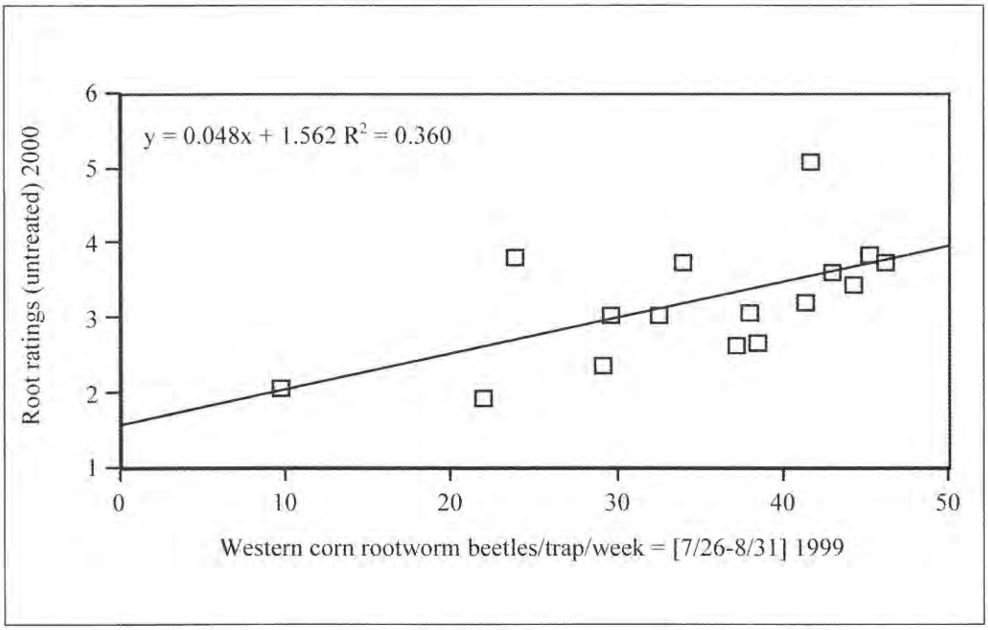

Figure 2 – Regression analysis of mean WCR population estimates/trap/week from 16 Indiana soybean fields using Pherocon AM traps from 7/26-8/31, 1999 versus root ratings taken in these same fields during 2000

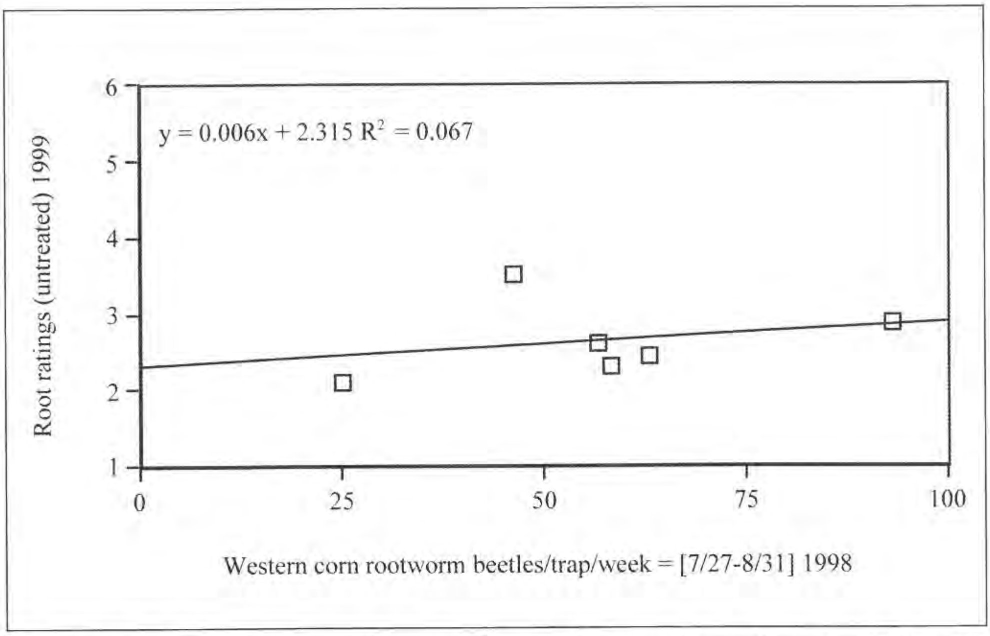

Figure 3 - Regression analysis of mean WCR population estimates/trap/week from six Indiana soybean fields using CRW traps from 7/27-8/31, 1998 versus root ratings taken in these same fields during 1999

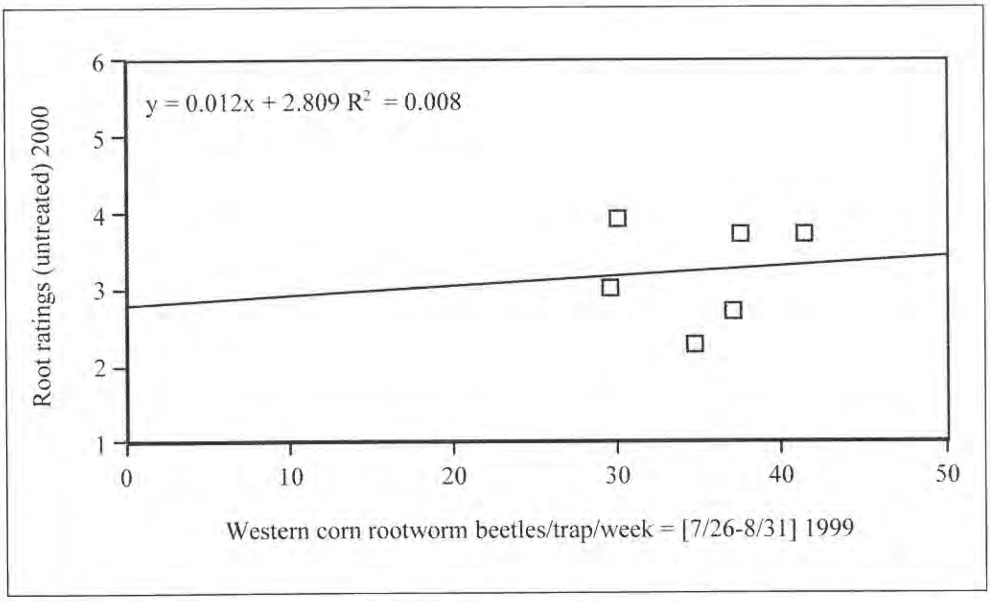

Figure 4 - Regression analysis of mean WCR population estimates/trap/week from six Indiana soybean fields using CRW traps from 7/26-8/31, 1999 versus root ratings taken in these same fields during 2000

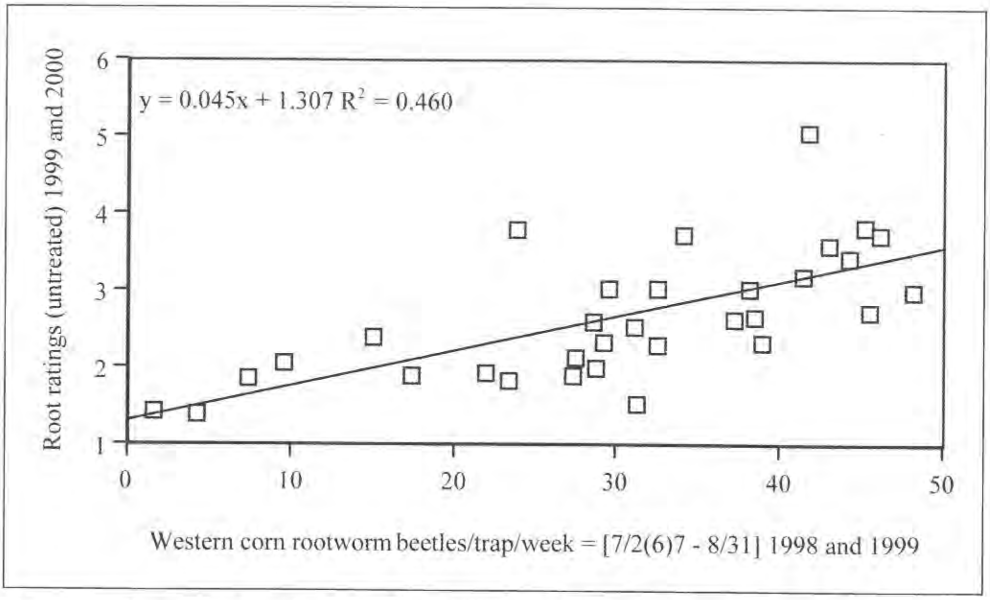

Figure 5 - Regression analysis of mean WCR population estimates/trap/week from 32 Indiana soybean fields using Pherocon AM traps from 7/2(6)7-8/31, 1998 and 1999 versus root ratings taken in these same fields during 1999 and 2000

When selecting a sampling method for corn rootworms in soybean, it is critical to obtain a balance between the number of traps actually needed per field, trap acceptance by producers, commercial availability of traps, and how well the traps actually predict root injury the following season. In 1997-2000, it was determined that by using five Pherocon AM traps or six CRW traps, 75% of the time these traps provide an acceptable estimation of an adult rootworm population in a soybean field. With an increase in trap numbers, the probability that the traps will estimate the actual mean number of beetles increases.

In conclusion, to be effective, Pherocon AM traps should be deployed in soybean fields during the last week of July through the month of August in Indiana. Preliminary reports by Levine and Gray (1996) have suggested that adult WCR peak activity, in the Eastern Midwest, occurs during the first three weeks in August. Five traps placed equidistant, lengthwise, down the middle of a field is the recommended number and placement of traps. Each trap should remain in a field for approximately one week. The data show that an average of eight beetles or more per trap per day can result in economic root damage in corn by WCR larvae the following season. However, due to variability of environmental and soil conditions, we are recommending that five beetles per trap per day be used as the economic threshold. In the research fields where at least five beetles per trap per day were observed, >95% of the cornfields reached economic root damage the following year.

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EFFECT OF SILK FEEDING BY WESTERN CORN ROOTWORM ADULTS ON YIELD AND QUALITY OF SEED CORN

Tamás Tuska, Jozsef Kiss,
Szent István University, Department of Plant Protection,
H-2100 Gödöllő, Hungary

C. Richard Edwards,
Purdue University, Department of Entomology,
1158 Smith Hall, West Lafayette, IN 47907-1158, USA

Zoltán Szabó, Ilona Ondrusz, Péter Miskucza, András Garai,
State Stud-Estate Corporation, Mezőhegyes, H-5820 Hungary

Abstract

The build-up of the WCR population in Europe requires that research be conducted to determine the potential effect of WCR adult silk clipping on seed corn and commercial corn. A silk clipping study was conducted on an early inbred corn line under field production conditions in Mezőhegyes, Hungary, in 2001. Four densities of WCR adults (0, 3, 6, and 9 per ear) were placed into individual, cylindrical nylon bridal veil ear cages at the plant growth stage R1. Two control treatments (0 beetles), one with ear cages and one without were included in the study. Each treatment (beetle density) was replicated 16 times. Presence of WCR adults was observed and silk length was measured daily at the same hour until silks dried. Cages were removed after pollination and the ears were allowed to mature. Ears were harvested, dried, and stored for further testing. Data on silk length, cob length, kernel number, thousand-kernel weight, and kernel fractions were correlated with beetle densities. First-year preliminary results indicate that the economic threshold for WCR adults is between 1-3 adults/ear for the inbred line tested.

Key words: Western corn rootworm, silk feeding, seed corn.

INTRODUCTION

The spread and establishment of Western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte, in Europe has been quite pronounced. Its population build-up in corn production areas will greatly impact farming practices, farmers' income, and corn as a commodity. Corn is a major field crop in Central and Eastern Europe. It is produced for various purposes (sweet corn, popcorn, silage, commercial corn, and seed corn). The continental climate in Central and Eastern Europe favors seed and commercial corn production. Production of seed corn requires higher inputs, but the increased value of this type of corn can results in higher income and profit for farmers. Therefore, the higher risk caused by WCR calls for specific economic threshold values for seed corn based on its higher value. Economic larval damage caused by WCR to corn root systems has been reported from Europe. However, little is known about silk feeding damage caused by WCR adults and its impact on production of corn hybrids in Europe. Silk feeding by WCR adults on inbred lines was tested and economic thresholds established in the USA by Culy *et al.* (1992). The build-up of the WCR population in Europe requires that tests be conducted to determine the potential impact of WCR adult silk clipping on seed and commercial corn production and that management strategies be developed based on these findings. The first-year results for the inbred corn line tested in this trial are presented in this paper. Economic threshold values for a commercial and late inbred corn line will be published later.

The study in seed corn was designed to:

- measure the silk clipping by varying levels of WCR adults;
- measure the yield impact caused by WCR adult feeding;
- evaluate the impact of silk clipping by WCR adults on seed quality;
- estimate the economic threshold value for WCR adult silk clipping.

Material and methods

The study was conducted on an early inbred corn line under field production conditions in Mezőhegyes (State Stud Estate Farm), South Hungary, in 2001.

Seed corn plots were established with male and female plants arranged in a 1:2 and 2:4 planting configuration for early and late lines, respectively. Plants of a female corn row at 30 m from the first row were selected for the test. The first plant used was 10 m from the edge of the field. Four densities of WCR adults (0, 3, 6, and 9 per ear) were placed into individual, cylindrical nylon bridal veil ear cages (40 by 25 cm, with 1 mm openings, fixed by a flat rubber band on the ear and closed by another rubber band on the top, but allowing enough space for silk development and WCR beetle movement) at the plant growth stage R1. WCR beetles were randomly collected from a nearby cornfield 2-3 days prior to beginning the study. Two control treatments (0 beetles), one with ear cages and one without were included in the study to assess the impact of the bridal veil ear cages on pollination. Each treatment (beetle density) for the inbred line was replicated 16 times.

Presence of WCR adults was observed and silk length to millimeter was measured daily at the same hour until the silks dried. Status of WCR adults at the various treatment levels was also checked and beetles were added to meet target levels where needed. WCR population density over the field was determined by counting beetles on 4x25 corn plants, representing whole plant counts, and on 12 Pherocon AM traps.

These values allowed for the estimation of the impact of "natural" WCR adult infestation on the seed corn (uncaged plants) in the field. Cages around the test ears were removed after pollination and the ears were allowed to mature. Ears were harvested, dried and stored for further testing.

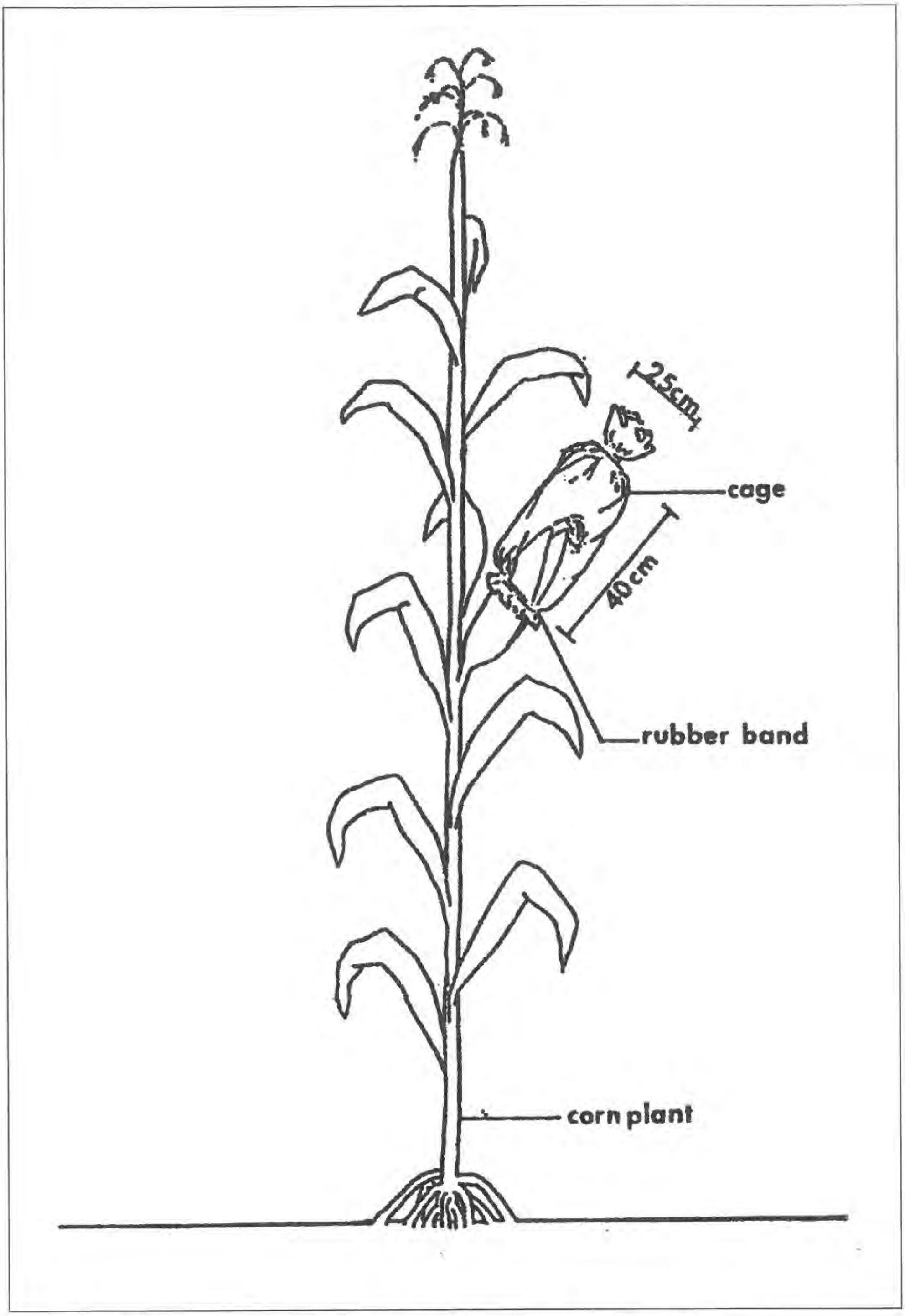

Figure 1 – Schematic drawing of ear cage (after Leva, 1980, in Culy, 1987)

Data on silk length, kernel number, thousand-kernel weight, and kernel fraction (*table 1*) were correlated with beetle densities.

Table 1 - Kernel fractions used for seed kernels

Fraction	Ø round riddle ≠ rift riddle	Size
large flat	Ø	8,5-10 mm
	≠	3,5-5,5 mm
large round	Ø	8,5-10 mm
	≠	5,5-7 mm
small flat	Ø	6,5-8,5 mm
	≠	3,5-5,5 mm
small round	Ø	6,5-8,5 mm
	≠	5,5-7 mm

RESULTS

Silking of the early inbred line started on 9 July 2001. Measuring of silk length started on 10 July and continued until 19 July (10 days). The maximum silk length for uncaged ears reached up to 60 mm. Presence of WCR adults on the ear reduced silk length. Caged ears with 0 WCR adults produced greater silk length compared to the uncaged ones. This was likely a result of shading stimulating the plant to produce longer silks to increase the chance for pollination to take place. The feeding by 9 WCR adults/ear decreased silk length by 50%. Even 3 WCR adults/ear caused a visible silk length reduction (*figure 2*).

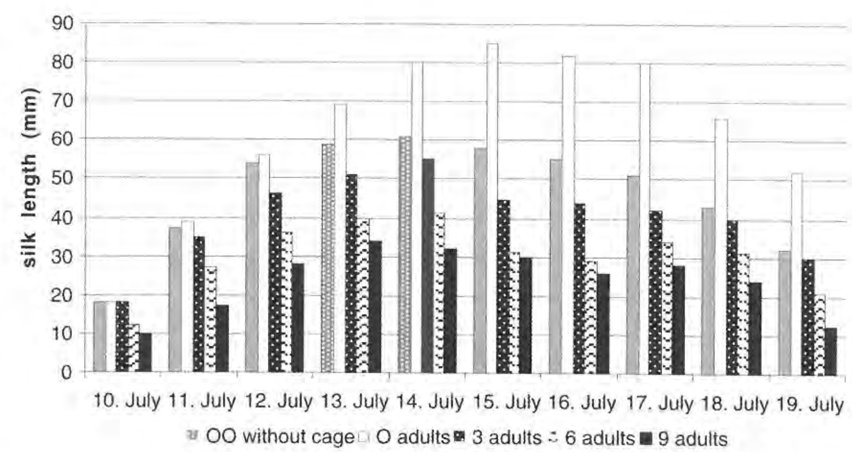

Figure 2 – Silk feeling by WCR on early inbred line (2001, Mezőhegyes, Hungary)

Silk length showed a strong correlation by treatment (number of WCR adults) (R^2=0.9256, *figure 3*). Silk length reduction was expected to lead to a loss in yield. Correlation of silk length to yield (g/kernels/ear) was strong (R^2=0.9478, *figure 4*).

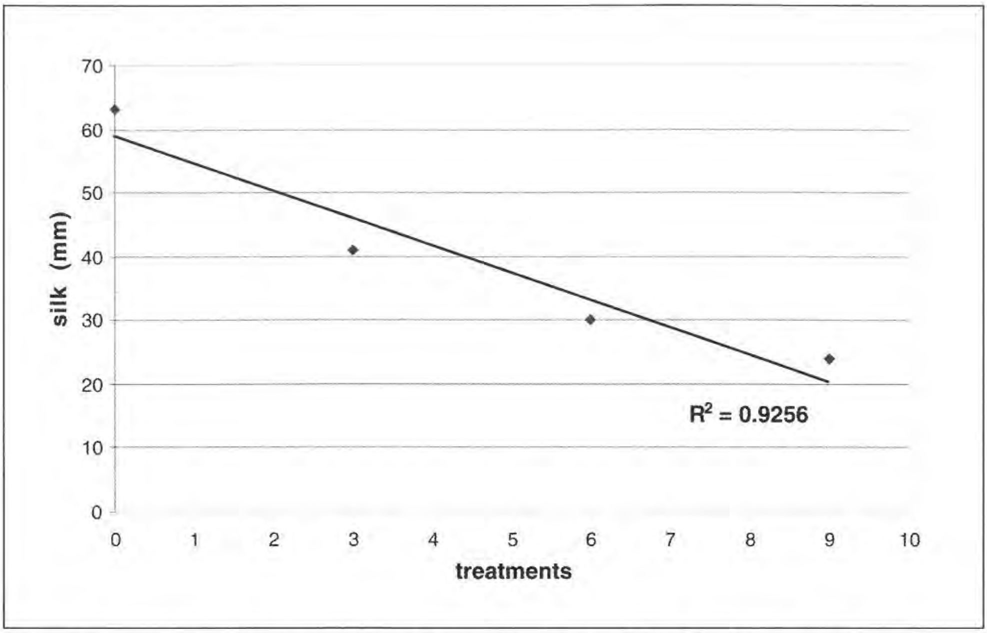

Figure 3 – Correlation of treatment to silk length (early inbred line Mezőhegyes, 2001)

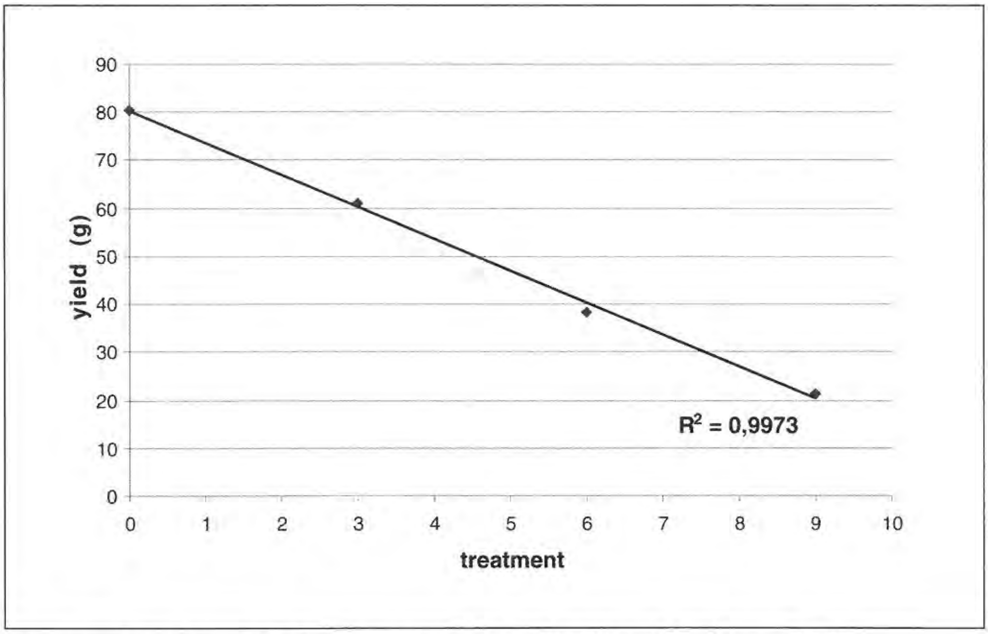

Figure 4 – Correlation of yield to treatment (early inbred line Mezőhegyes, 2001)

Silk feeding by WCR adults significantly reduced yield of seed corn. Correlation of yield to treatment gave an extremely high R^2 value (R^2=0.9973). The lowest treatment level in the trial was 3 WCR adults/ear. This treatment level caused an approximate 25% yield reduction. According to the correlation results, the economic threshold level for the tested inbred line is between 1-3 adults/ear. Adult silk feeding significantly inpacted seed quality (the fractions of the kernels, which is an important parameter for seed production). The percentage of valuable fractions (small flat and large flat) decreased, while those of less valuable fractions (small round and large round) increased (*figure 5*).

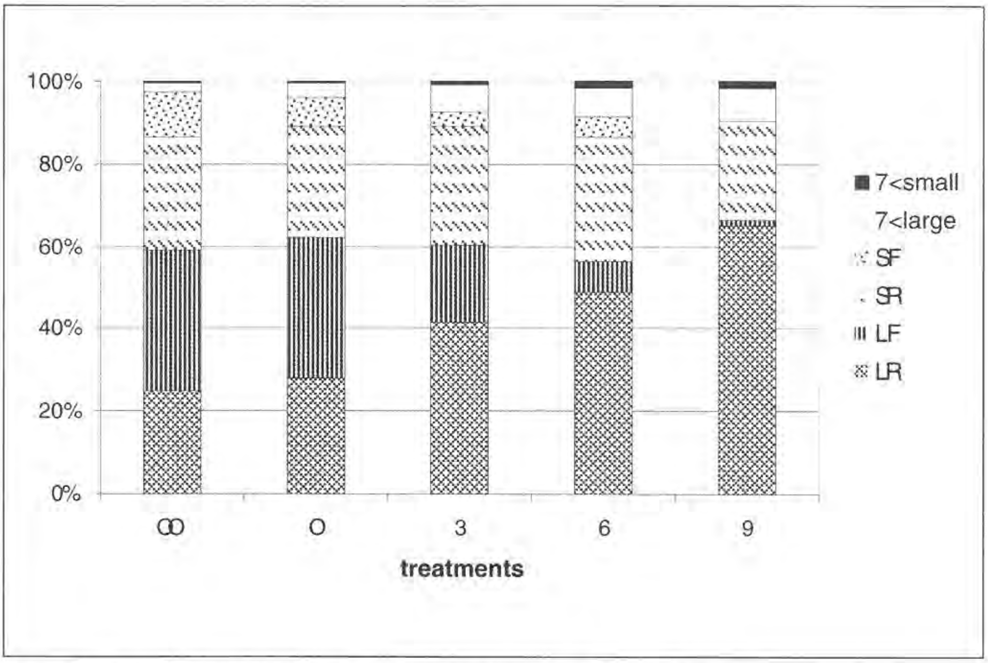

Figure 5 – Seed kernel fractions (early inbred line Mezőhegyes, 2001)

The impact of silk feeding on seed quality shows that the threshold is between 1-3 adults/ear for the inbred tested.

Conclusions

First-year preliminary results for an early inbred line indicates that WCR adult silk clipping can significantly impact yield and seed quality. The economic threshold for silk clipping by WCR adults for the tested inbred line is 1-3 adults/ear.

Acknowledgements to Dr. Mike Culy, Dow AgroSciences, Indianapolis, IN, USA, for information supporting this study.

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MONITORING OF *D. VIRGIFERA VIRGIFERA* LECONTE IN SERBIA IN 2001

Ivan Sivcev, Anicka Galo,
Institute for Plant Protection and Environment, Banatska 33
11080 Zemun - Yugoslavia

Abstract

In 2001 we continued with monitoring of WCR (*D. virgifera virgifera* LeConte) dispersal in Serbia. Sex pheromone traps produced by the Plant Protection Institute, Budapest, were used on 240 sites in order: a) to check presence or absence of WCR imagoes and b) to check population density. Further spread on the south east was registered, along the border with Bulgaria. Economic population and damages were registered sporadically within the area registered during last year. In 2001 there were no newly registered areas with damages from larvae. In general, corn fields were significantly less infested and damaged. Due to abundant rainfalls during the spring damaged corn plants recovered and it was difficult to find heavily damaged fields with logged plants. Yield reduction was insignificant.

Comparing the population density from last year, it is obvious that in 2001 population was reduced significantly on the whole territory of Serbia.

DIABROTICA MONITORING IN AUSTRIA 2001

Harald K. Berger,
Federal Office and Research Centre for Agriculture;
Main Staff Division for Research Integration;
A – 1226 Wien, Spargelfeldstraße 191; Austria

INTRODUCTION

Monitoring of *Diabrotica virgifera virgifera* was carried out by the Federal Office and Research Centre for Agriculture, Main Division Research Integration in Vienna together with co-operators of the relevant Plant Protection Departments in the Chambers of Agriculture of Lower Austria, Upper Austria, Tyrol, Carinthia, Burgenland and Styria and the Pioneer Seed company. In all cases floral baited traps were used. The used traps were received from the Hungarian Academy of Science and were of the type "*Diabrotica v. virgifera* attractant trap" (PALs). With this type of traps both females and males can be caught.

The survey was done because of the rapid spread out of the pest from the place of its first appearance in Surcin, near Belgrade, Serbia to the neighbouring countries such as Croatia, Hungary, Bosnia and Herzegovina, Romania and Bulgaria and due to the spread out of the pest west of lake Balaton, which opens the "door" to the east Austrian maize fields.

In 2000 the most closest appearance of *Diabrotica virgifera virgifera* to Austria from Hungary in was less than approximately 150 km. Danger seems to be serious, especially in Styria where a lot of corn is grown close to the Hungarian border. Additionally farmers in Styria grow also pumpkins for pumpkin-oil production for a rather large amount of hectares, so the pumpkin blossoms are an additional attractant for adult *Diabrotica*. Also the spread out of *Diabrotica* in Slovakia north of the river Danube close to the city Komarom and east of this city seems to threaten the Austrian maize production in the near future.

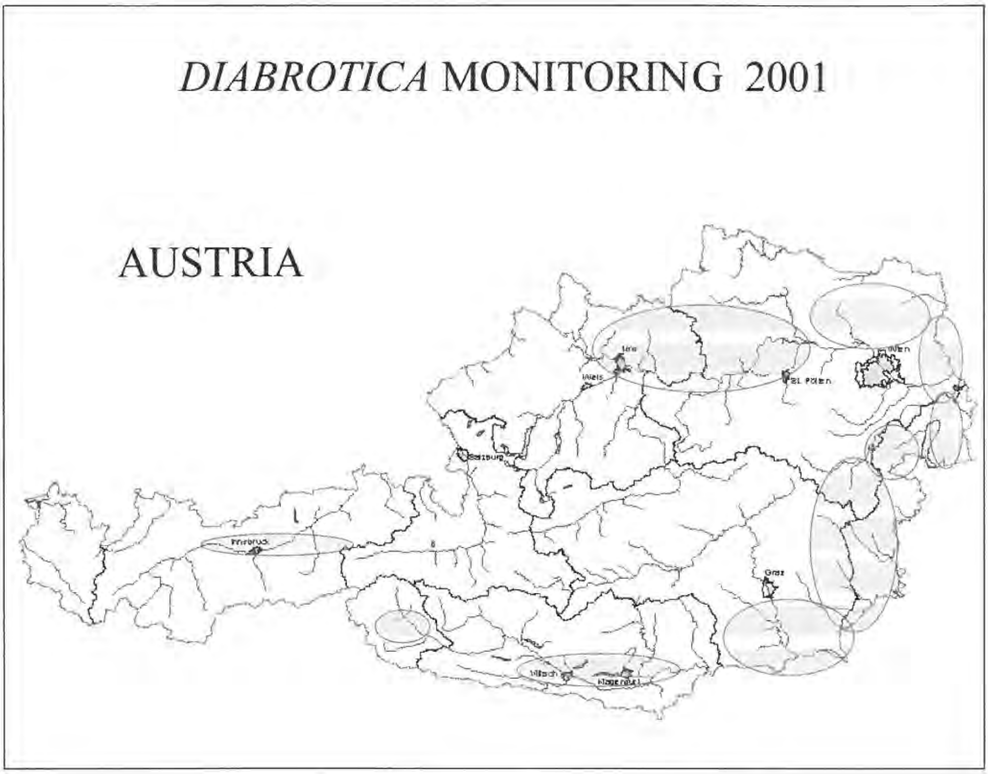
We think that the thread of this pest can be pushed more in the future by a consequent crop rotation which is actually normally done in this region of Austria.

MATERIAL AND METHODS

Monitoring of the adults of the harmful pest *Diabrotica virgifera virgifera* in Austria is characterised by an expected small amount of individuals from population with a very low density. For this purpose floral baited traps from the Hungarian Academy of Science of the type PALs, catching male and female were chosen. The capsules were stored in the fridge until their placement into the maize fields. These traps have been used in all localities. The observations were carried out along the Austrian border to Hungary and Slovakia and also along main roads leading west and southwards from Vienna as well as along the Danube river. Official border crossings to Hungary and Slovakia were preferred as well as the surrounding of the Vienna International Airport ("Schwechat"). While in 1998 60, in 1999 21 and 2000 40 pheromone traps were used the amount of used traps increased up top 300 in 2001.

The selection of the places were made by staff members of the local chambers of agriculture in co-operation with the local farmers and land owners according to crop rotation and share of maize in the crop rotation in the area chosen.

Out of a total area of 1,382,000 ha of field crops the share of corn is approximately 19% of is 2/3 grain corn. On most of this area maize is rotated with other crops so the amount of maize monoculture is rather low.



Results and discussion

The observations started between June 20th and July 6th. The traps, pheromones and sticky surfaces were changed twice during the whole observation time. The final checks were made between August 28th and September 13th, 2001. Other checks were made continuously between these days.

Diabrotica virgifera virgifera was not caught in any trap throughout 2001. Therefore, we can say with large likelihood that *Diabrotica virgifera virgifera* is not present in the territory of Austria in the surveyed Federal countries (Lower Austria, Upper Austria, Burgenland, Styria, Carinthia, Tyrol). The possibility of the presence *Diabrotica virgifera virgifera* in these maize growing Federal countries with favourable climate is given. In the remaining Fed. Countries such as Salzburg, Vorarlberg and Vienna the amount of maize in the rotation is rather low (Salzburg, Vorarlberg) or does not pay an important role at all (Vienna).

Nevertheless monitoring will be continued with pheromone traps - such as used in 2001 - also in 2002.

The appearance of *Diabrotica* in Milan and in the Swiss Ticino in 2000 (as already Venice in 1999) showed that there must not be a direct connection between the infested areas. Therefore we will continue our country-wide monitoring in 2002 by help of the Austrian phytosanitary service on the same scale like it has been done in 2001.

Key Words: Diabrotica, Chrysomelidae, monitoring, Austria.

MONITORING THE WCR (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) IN BADEN-WÜRTTEMBERG IN 2001

Hansjörg Imgraben,
Regierungspräsidium Freiburg, Pflanzenschutzdienst, Ref. 33
Bertoldstraße 43, D-79098 Freiburg, Germany

Abstract

Baden-Württemberg is an important area for the production of grain maize, maize for silage and maize seed production in Germany. Since 1997 the Land Baden-Württemberg carries out a monitoring program with the aim to detect Diabrotica if it appears in the country.

On the basis of the current Diabrotica occurrence in Europe, it is considered that the beetle will be imported by aeroplane or other means of transport. The WCR could also arrive along the Danube first to Bavaria and then to Baden-Württemberg.

In 2001 the number of the monitoring locations and the number of the pheromone traps were increased in particular near airports and airfields. As in the previous years traps were set out furthermore along motorways, near railway stations, near warehouses and customs stations. Altogether in 2001 at 67 locations in the country traps were set up.

The monitoring extended to the Rhine Valley, the area from the Bodensee to the Danube, the region around Stuttgart as well as the eastern section of Baden-Württemberg.

As in the previous year only pheromone traps (type PAL, Hungarian origin) were used.

The traps were set out at the beginning of July and controlled regularly.

Diabrotica virgifera beetles were not found in the traps.

THE MONITORING OF WESTERN CORN ROOTWORM IN SLOVENIA - A REPORT FOR YEAR 2001

Aleš Pajmon,
Agricultural Institute of Slovenia,
Hacquetova 17, 1000 Ljubljana, Slovenia

Abstract
In Slovenia, a major part of the farming land is sown with corn. Western corn rootworm (*Diabrotica virgifera virgifera*) population is quickly approaching our border. For this reason, monitoring is more frequently applied in Slovenia. In 2001, baits have been controlled and replaced at control points more often. Furthermore, the number of control points has also been increased to 56 which is more than in previous years. In the vicinity of Italian border we set 10 control points, around the international airport of Ljubljana (Brnik) were another 8 of them. Along the borders with Croatia and Hungary, which is in the eastern Slovenia where the corn production is the most intensive, 38 control points were set. At each point, a pheromonic bait and yellow sticky plate were set. The monitoring was done from the end of June to mid-September while the pheromonic baits and yellow sticky plates were regularly controlled and replaced with new ones. Despite the fact that western corn rootworm was found in Croatia in 2000 only 27 km away from the Slovenian border, it has not yet been detected in Slovenia. Therefore, there is a strong possibility that Western corn rootworm will be found in Slovenia next year.

Key words: monitoring, *Diabrotica virgifera virgifera*, corn, Slovenia.

INTRODUCTION

In Slovenia, a major part of the farming land is sown with corn. Western corn rootworm (*Diabrotica virgifera virgifera*) population is quickly approaching our borders. Unfortunately, climatic conditions in our country are suitable for this pest to spread. Consequently, we have been regularly monitoring the possible existence of the pest in our country since 1996. The monitoring is intensified each year.

METHODS

In 2001, baits have been controlled and replaced at control points more often. Furthermore, the number of control points has also been increased to 56 (*table 1*). As in previous year, we set 10 control points in the vicinity of the Italian border and 8 around the international airport of Ljubljana - Brnik. Along the Hungarian

Table 1 - A review and geographical definition of the control points for the monitoring of Western corn rootworm (*Diabrotica virgifera virgifera*) in Slovenia in 2001

Region	No.	Location	Microlocation of the parcel
Prekmurje	1	Martjanci	100 m ahead of the sign Martjanci - end
	2	Tešanovci	Behind the small bridge
	3	Bogojina	At the green power-point locker
	4	Bukovnica	Near the shooting range
	5	Prosenjakovci	Near the school
	6	Motvarjevci - Kobilje	At the corner, near the paving
	7	Kobilje - state border	Before the border, to the right, at the house
	8	Žitkovci	Near the farm
	9	Mostje - Lenti	Behind the stop sign, to the left
	10	Dolga vas	Before the border crossing, down to the left
	11	Pince	Across the bridge, to the left at the end of the asphalt
	12	Pince - Petišovci	Behind the silos
	13	Petišovci	Grčeva House at the border crossing
	14	Gabrje	At the Y crossroads
	15	Šafarsko - Gibina	Before the sign, to the right of the pumpkins
	16	Razkrižje	Across the Ščavnica, right behind the sign
Dravsko field	1	Moškjanci	Before the hop
	2	Zagojiči - Gorišnica	By the canal
	3	Borovci	Near the football ground, to the right
	4	Zavrč	Near the school, opposite the football ground
	5	Borl	Under the castle, by the nut tree
	6	Zakl	At the petrol station
	7	Mihovci	At the vine grower's
	8	Mihovci - south	At the hunter's observatory
	9	Loperšice	At the bus stop, down to the right
	10	Obrež	At the house No. 76
	11	Središče ob Dravi - east	Between the two border crossings
	12	Središče ob Dravi - north	By the pine trees
	13	Vodranci	Opposite the house with vines
	14	Jastrebci	On the hill, to the left of the small chapel
Krško-breziško field	1	Mali Podlog	By the car park on the turning for Krško
	2	Vihre	Before the small oaks
	3	Velika vas	Between the small bridge and the forest
	4	Obrežje	At the border crossing, to the left
	5	Jesenice	At the road sign "Road company"
	6	Podgračeno	Behind the sign for Cerjak, to the left
	7	Rigonce - Loče	At the turn before Loče, to the left
	8	Gregovce in Bizeljsko	In the village, to the left of the oak tree
Brnik airport	1	Voglje - north	Opposite the hangars
	2	Voglje	After the crossroads
	3	Voglje - east	At the end of the fence
	4	Vodice - Vopovlje	At the end of the field path
	5	Vopovlje - south	At the grove
	6	Vopovlje	Opposite the radar
	7	Spodnji Brnik	Before the forest, to the right of the road
	8	Zgornji Brnik - south	After the crossroads, by the nut tree
Northern Primorska	1	Šempeter	Opposite the Merkator supermarket
	2	Gornja Vrtojba	Opposite the petrol station
	3	Šempeter - Vrtojba	At the purifying plant
	4	Bilje	To the right of the fruit center
	5	Bukovica	Near the graveyard
	6	Vogrsko	Under the motorway viaduct
	7	Vogrsko - Ozeljan	At the farm, to the left of the crossroads
	8	Okroglica at Ozeljan	At the irrigation station
	9	Zemono	At the manor house, to the left towards the field path
	10	Vipava	At the Agromarket shop

Table 2 - A review of the locations, northern latitudes and eastern longitudes of the control points for the monitoring of Western corn rootworm (*Diabrotica virgifera virgifera*) in Slovenia in 2001

Region	Location	Northern latitude	Eastern longitude
Prekmurje	Martjanci	46° 41' 08"	16° 12' 19"
	Tešanovci	46° 40' 59"	16° 14' 22"
	Bogojina	46° 40' 32"	16° 16' 17"
	Bukovnica	46° 40' 58"	16° 17' 41"
	Prosenjakovci	46° 44' 45"	16° 18' 30"
	Motvarjevci - Kobilje	46° 42' 33"	16° 21' 42"
	Kobilje - state border	46° 41' 67"	16° 25' 08"
	Žitkovci	46° 38' 44"	16° 22' 28"
	Mostje - Lenti	46° 35' 70"	16° 26' 96"
	Dolga vas	46° 35' 73"	16° 27' 56"
	Pince	46° 30' 60"	16° 30' 21"
	Pince - Petišovci	46° 31' 17"	16° 28' 16"
	Petišovci	46° 31' 28"	16° 26' 47"
	Gabrje	46° 32' 46"	16° 24' 40"
	Šafarsko - Gibina	46° 31' 12"	16° 17' 76"
	Razkrižje	46° 31' 25"	16° 15' 63"
Dravsko field	Moškanjci	46° 24' 78"	15° 59' 11"
	Zagojiči - Gorišnica	46° 24' 36"	16° 00' 13"
	Borovci	46° 25' 25"	15° 56' 56"
	Zavrč	46° 23' 06"	16° 02' 87"
	Borl	46° 22' 28"	16° 00' 08"
	Zakl	46° 19' 60"	15° 52' 78"
	Mihovci	46° 24' 12"	16° 06' 79"
	Mihovci - south	46° 23' 57"	16° 05' 67"
	Loperšice	46° 23' 90"	16° 12' 28"
	Obrež	46° 23' 70"	16° 14' 54"
	Središče ob Dravi – east	46° 23' 18"	16° 17' 89"
	Središče ob D. – north	46° 24' 35"	16° 16' 66"
	Vodranci	46° 25' 97"	16° 14' 93"
	Jastrebci	46° 26' 29"	16° 15' 46"
Krško-brežiško field	Mali Podlog	45° 54' 76"	15° 28' 06"
	Vihre	45° 54' 60"	15° 31' 22"
	Velika vas	45° 54' 85"	15° 27' 12"
	Obrežje	45° 50' 95"	15° 41' 42"
	Jesenice	45° 51' 54"	15° 40' 98"
	Podgračeno	45° 52' 35"	15° 39' 27"
	Rigonce - Loče	45° 53' 37"	15° 39' 89"
	Gregovce in Bizeljsko	45° 59' 60"	15° 41' 82"
Brnik airport	Voglje - north	46° 13' 12"	14° 27' 57"
	Voglje	46° 13' 04"	14° 27' 98"
	Voglje - east	46° 12' 86"	14° 28' 32"
	Vodice - Vopovlje	46° 12' 85"	14° 29' 06"
	Vopovlje - south	46° 12' 95"	14° 29' 21"
	Vopovlje	46° 13' 35"	14° 29' 22"
	Spodnji Brnik	46° 13' 25"	14° 28' 73"
	Zgornji Brnik - south	46° 13' 88"	14° 28' 25"
Northern Primorska	Šempeter	45° 55' 83"	13° 38' 17"
	Gornja Vrtojba	45° 55' 06"	13° 37' 58"
	Šempeter - Vrtojba	45° 55' 30"	13° 38' 38"
	Bilje	45° 53' 85"	13° 37' 26"
	Bukovica	45° 53' 64"	13° 39' 62"
	Vogrsko	45° 55' 10"	13° 42' 33"
	Vogrsko - Ozeljan	45° 56' 05"	13° 42' 38"
	Okroglica at Ozeljan	45° 56' 31"	13° 43' 02"
	Zemono	45° 51' 48"	13° 56' 52"
	Vipava	45° 50' 46"	13° 57' 54"

and partly Croatian border, which is in the eastern part of Slovenia where the corn production is the greatest in our country, we set 38 control points. 8 of them were placed along the Croatian border, on Krško-brešiško field (at the Sava river) while 16 of them were placed in Prekmurje region - along Hungary and partly the Croatian border. On Dravsko field (at the Drava river) we increased the number of control points to 14. Towards this part of Slovenia, western corn rootworm population is approaching our country very fast. According to the evidence, it was found near Varaždin in Croatia in 2000, which is only 27 km away from the Slovenian border.

At each control point, one pheromonic bait and one yellow sticky plate were set 100 meters away from one another. The monitoring was carried out from the end of June to mid-September. During that time, pheromonic baits and yellow sticky plates were regularly checked, in 7 to 10 day intervals depending on location. The most frequent checks were carried out on Dravsko field, which is because of closeness of Varaždin, the most exposed at the moment. In this area there were 14 control points where pheromonic baits were replaced twice (in 3-4 weeks), while at other points they were replaced once (in 5 weeks). Yellow sticky plates were replaced more frequently at all locations.

Geographically, the control points were defined by GPS 3000 XL apparatus (magellan) which helped us define accurate coordinates of the northern latitudes and eastern longitudes (*table* 2).

The monitoring of western corn rootworm has been re-established by the Slovenian Agricultural Institute while the help with the choice of locations and bait check have been given by Slovenian Inspectorate and Advisory Service for Agriculture.

Results

Considering the fact that western corn rootworm was found in Croatia only 27 km away from Slovenian border in 2000, there was a great possibility it would be found in Slovenia this year. For this reason, especially in that part of Slovenia (Dravsko field), the monitoring has been more intensive than in the past. Nevertheless, the beatle has not been detected in Slovenia this year, therefore there is a strong possibility that WCR corn rootworm will be found in Slovenia next year.

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CURRENT STATUS OF WCR IN CROATIA

Igrc Barčić Jasminka, Dobrinčić Renata, Maceljski Milan,
Faculty of Agriculture, Department of Agricultural Entomology,
Svetošimunska 25, 10000 Zagreb, Croatia;
e-mail: jigrc@agr.hr , tel. 385 1 2393-804

Abstract

The occurence of *Diabrotica virgifera virgifera* LeConte in Croatia was registered in 1995. In 2000, WCR was spread on a 14,000 km^2 area. Of that area, there was about 700,000 ha of arable land on which 200,000 ha of corn was sown.

The Department of Agricultural Zoology, Faculty of Agriculture Zagreb has carried out nation-wide survey of Western Corn Rootworm (*Diabrotica virgifera virgifera* LeConte) in Croatia since 1995.

As in previous years, pheromone and Pherocon AM traps were provided through FAO WCR Network.

The traps were placed out on 145 sites and 75 monitor were involved in the monitoring action. On each site 1 pheromone and 1 Pherocon AM trap were placed 50 m apart. Traps were placed in the field on June 27th. Each 7 days the traps were checked and beetles collected. Fifty five sites (38%) were located at the infested regions, 48 sites (33%) along the line of spread in 2000, and 42 (29%) within uninfested areas. Ten monitoring sites are permanent ones.

The first catch on pheromone trap was registered on June 28th. Before disposing all results on 130 monitoring sites 16,353 beetles were caught. Most of the beetles were caught on pheromone traps. The maximal catch, 62.5% of all caught beetles, was caught between July 10th and August 10th.

The WCR population density varied from locality to locality, we evaluate that the increase factor in the earlier infested area for 2001 is between 1.5 and 2.5. The data from 10 permanent monitoring sites confirm that statement.

Only on one monitoring site located outside the area infested in 2000 one WCR male was caught until now. We can state that in 2001 WCR spread toward west (60 km) to the village Dubrovčak Lijevi. This spread occured along the river Sava. No spread along the river Sava was registered in 2000. No further spread along the river Drava and the Hungarian border was recorded until now.

Three monitoring sites were located around the International Airport Zagreb. WCR has not been found on any of these traps.

In 2001, corn roots in the soil insecticide trial in Tovarnik were dug and evaluated for WCR damage. Plants in a few continuous corn fields in the region Baranja were also evaluated. The average root rating in untreated plots in soil insecticide trial was between 2.5 and 3 (same as in 1999). In some inspected fields in Baranja we found lodged plants. The average root rating in these fields was between 3.5 and

4.0. In one seed corn field a high adult population and the silk damage were registered. The yield loss will be established.

Altogether, in 2001 an area of approximately 15,500 km^2 is infested with WCR, in which area approximately 220,000 ha of corn is grown.

Further spread and an increase in the root damages in 2002 are expected.

THE INVESTIGATION OF THE RELATIONSHIP BETWEEN WCR POPULATION LEVEL AND CORN YIELD CROATIAN EXPERIENCIES

Dobrinčić, Renata , *Igrc Barčić, Jasminka,*
Faculty of Agriculture, Department for Agricultural Zoology,
Svetošimunska 25, 10000 Zagreb, Croatia,
e-mail: rdobrincic@agr.hr, tel. 385 1 2393-969

C.R. Edwards,
Department of Entomology, Purdue University, IN, USA

Abstract

It is well known that larvae of *Diabrotica virgifera virgifera* are oligophagous (Branson and Krysan, 1969, 1970). The only damage effectuated by larvae on cultivated plants was found in corn. Quantitative relatioship between population level and corn yield was investigated by Hills and Peters (1971), Turpin *et al.* (1972), Branson *et al.* (1980, 1982, 1983), Chiang *et al.* (1980), etc. Some investigations were carried out under the conditions of artificial infections and in the presence of high natural WCR population levels. Most authors agree that climatic and edaphic factors can impact yield at the same population level.

The aim of this study was to check how the USA literature data about the correlation between WCR population level and corn yield corresponds to the Croatian conditions of low to moderate population level.

In the region with the highest infestation of WCR, a four year programm started in 1998. In continious corn fields, emergence cages (3 cages per replication) were placed to evaluate number of emerged beetles per plant. To determine the larval population level in the same fields, 20 plants per replication were dug and root damage (using the Iowa 1-6 scale) was rated. To determine the yield, 20 plants per replication were harvested and the yield was measured.

The data from the first 3 years (1998-2000) of the investigation showed that the damage caused by larval feeding on roots in Croatia corresponds to the damage caused in USA. At this level of infestation, the regression coefficient between larval damage and yield could not be established. The literature showed that this coefficient is between 310 and 670 kg/ha of yield loss for each root rating (Iowa 1-6 scale). The full linear correlation (r^2 =0,924) between the number of emerged beetles per plant and the root rating was established ($y = 2.2 + 0.031x$). Thirty-two emerged beetles per plant caused an increase in the root damage rating.

The data obtained in 2001 will be added to and presented at the meeting.

WESTERN CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) SAMPLING AND CONTROL IN THE TERRITORY OF GUNJA, CROATIA

Dražen Džoić,
Ministry of Agriculture and Forestry
10000 Zagreb - Croatia

Marija Ivezić, Emilija Raspudić, Mirjana Brmež,
Univ. of J.J. Strossmayer Fac. of Agriculture in Osijek
31000 Osijek - Croatia

Abstract

A new insect pest was identified in 1995 in Croatia - the western corn rootworm (*Diabrotica virgifera virgifera* LeConte). The first objective of this research was to determine the population density of all stages, except eggs in commercial corn fields. The second objective was to investigate the efficacy of three organophosphate insecticides on larvae. The experiment was conducted in 1999 and 2000 in Gunja, Croatia (44°87'N, 18°94' E). Treatments were corn hybrids (OSSK 444, OSSK 552, Florencia,) and three soil insecticides (terbufos, chlorpyriphos-ethyl, chlormephos) applied at planting. Results showed the highest number of larvae per plant (0.70) in the untreated plot of OSSK 552. In 1999, significant differences in larval numbers occurred between hybrids, but not between the insecticides. In 2000, larval numbers only differed statistically between the insecticide treatments. The highest beetles population counted per plant was 0.55 in 2000. This population level is very close to economic threshold of 0.70 beetles per plant. Significant differences in beetle numbers per plant between hybrids were only detected in 2000. Pheromone traps containing the lure, Csal♀m♂N, caught significantly more beetles than the Multigard® yellow sticky-trap. Terbufos was the only soil insecticide providing a significant yield advantage to the hybrids, but only in 2000. Based on the current value of corn and cost of insecticide, terbufos is the only soil insecticide that would be cost-effective for growers. These studies should be conducted with other insecticides, and growers should avoid planting corn after corn in their fields.

INVESTIGATION OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN BOSNIA AND HERZEGOVINA IN 2001

Festić Husnija, Karić Nedzad,
University of Sarajevo, Faculty of Agriculture

Berberović Husein, Huremović Zijo,
Institute of Agriculture Tuzla

Abstract
In Bosnia-Herzegovina, corn represents a very significant culture. Various hybrids are bread. Spread of hybrids depends on configuration of soil and climatic conditions. Yield of this crop is for the time being very small because Bosnia-Herzegovina has recently been in a war and farmers have not yet managed to improve the necessary agrotechnical measures. In general, this crop is subject to attack by various pests and diseases not only in this country but in neighboring countries as well.

Key words: *Diabrotica virgifera virgifera*, Western Corn Rootworm, occurrence, spread, Bosnia-Herzegovina.

INTRODUCTION

In 2001, monitoring of the spread of Western corn rootworm was continued. Investigation of this pest started in 1997 (Festić *et al.*, 1998). Thanks to the regular monitoring of the pest, since its occurrence until today, we are fully aware of its spread and direction of spreading.

Every year, the area under this crop in Bosnia-Herzegovina occupies 240,000-260,000 ha. In the Federation of Bosnia-Herzegovina, corn covers the area of 60,000-80,000 ha. The largest corn areas are in the plains of north and east of the country. It is estimated that corn in Bosnia-Herzegovina is one-crop at around 35 percent, i.e. 80,000-90,000 ha.

MATERIAL AND METHODS

This year it was suggested to us by the Project Manager that occurrence of corn rootworm might occur earlier. That is the reason why the tests had been conducted a little earlier.

For detection of presence and spread of the pest, Hungarian pheromone-based traps and yellow Traps (Pherocon AM) were used. Traps were placed in the areas of earlier occurrence of the pest, as well as at the places where occurrence was expected.

Investigations were conducted in Posavina Canton, Tuzla Canton, Zenica-Doboj Canton and Una-Sana Canton, and in the Brcko District.

In the Federation of Bosnia-Herzegovina, traps were placed on 79 monitoring spots. One monitoring spot contained one pheromone and one yellow trap each. They were positioned at 50-100 m distance. Traps were inspected once a week. In order to register the date of the first catch, both traps were replaced after 25 days.

During the monitoring, four shifts of traps were made. Number of traps in one municipality was determined on the basis of the area under corn and the degree of danger was estimated on the results from the previous year.

This year, the greatest number of traps was placed between 20 and 30 June. In 2000, the occurrence of imago came a little earlier in relation to previous years (Festić *et al.*, 2000). That is why a certain number of traps were placed on 17 and 18 June in Posavina Canton and in Tuzla Canton.

Traps in the area of Posavina Canton (10+10) were placed at the end of June in municipalities of Orasje, Domaljevac, and Odzak.

In Tuzla Canton, the biggest corn area, there were 39 monitoring spots.

In Zenica-Doboj Canton, traps were placed (12+12) in six municipalities: Tesanj, Doboj-Jug, Usora, Maglaj, Zavidovici, Zepce.

In Una-Sana Canton, 10 monitoring spots were placed in the municipalities of Sanski Most, Kljuc, Bosanska Krupa, Buzim, and Velika Kladusa.

In the area of Brcko District there were six monitoring spots.

During inspection of traps, imago was taken off and put into alcohol. There was a form for registration of the numbers for each monitoring spot.

RESULTS AND DISCUSSION

The first imago was caught on a pheromone trap on the locality of Krtova, municipality of Lukavac, on 27 and 29 June it was caught on a pheromone trap in the Sapna municipality which is in Tuzla Canton. Although the first two imago were registered in June, massive flight of imago was recorded at the end of the second year and in the third decade of July. This, in our opinion, was caused by enormous rainfall in July. We presume that the rainfall in June damaged a great number of larvae and corn rootworm (Maceljski and Igrc-Barčić, 1993; Čamprag *et al.*,1995). The area of Tuzla had 351,5 mm of rain in June. In the period of 17-21 June there was a big flood in this Canton.

In Posavina Canton, rootworm was registered on all localities. This Canton was in the last year under attack by this pest, especially the municipality of Orasje where the number of pest is increasing every year, so we can expect economic damages in the near future. In 2001 in Posavina Canton there were no damages by larvae, and minor damages to beard of corncob in the municipality of Orasje.

In Tuzla Canton, the pest was registered on all 39 monitoring spots. In this area there has been an increased number of caught imago every year. Somewhat greater number of population was in municipalities of Lukavac, Teocak, Tuzla, and

Gradacac. Minor number was registered in the municipalities of Doboj-Istok and Gracanica.

In Zenica-Doboj Canton, the pest was first registered in 1999. In 2000 it was registered in three municipalities, while in 2001 it was registered in six municipalities. Corn rootworm occurred first in municipalities of Maglaj, Zavidovici, and Zepce.

Our expectations from the last year that corn rootworm might occur in the area of Una-Sana Canton came true. The pest occurred in the municipalities of Sanski Most and Velika Kladusa.

Brcko District was attacked by rootworm and the number is increasing every year.

In 2001 there was a spread of corn rootworm into the new areas. Those new areas are in Zenica-Doboj and Una-Sana Cantons, while other areas had an increased number of pest related to the previous year.

The area of municipalities taken by corn rootworm for the first time in 2001 is 1,174 km^2. The total area under the pest in the Federation of Bosnia-Herzegovina is 4,374 km^2 of which 450 km^2 is in Posavina Canton, 2,100 km^2 in Tuzla Canton, 1,304 km^2 in Zenica-Doboj canton, 20 km^2 in Una-Sana Canton, and 500 km^2 in Brcko District.

Conclusion

This year we have noticed the difference in the attack by Western corn rootworm on individual hybrids. We reached the conclusion based on intensity of attack in macro tests of corn hybrids.

Lodging of plants was not noticed.

On areas where corn is one-crop, the catch of imago was considerably higher in relation to traps placed in crop rotation.

As in previous years, dominant catch was by pheromone traps while yellow traps showed smaller number of catch.

In 2001, there was 7,746 imago caught, out of which 7,671, or 99.1% on pheromone, and 75, or 0.96% on yellow traps.

The greatest catch was in August. On most localities, the catch in the first two decades of July was lower than in previous years. The catch considerably falls down in September and becomes merely symbolic in the first decade of October.

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MONITORING OF WESTERN CORN ROOTWORM *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN ROMANIA IN 2001

Gogu Florica,
Central Laboratory for Phytosanytary Quarantine,
Bucharest, Romania

INTRODUCTION

Since 1995 the monitoring of *Diabrotica virgifera virgifera* LeConte has been carried out by the Ministry of Agriculture, Food and Forestry through the Central Laboratory for Phytosanitary Quarantine and the County Phytosanitary Departments. The first find of *D. virgifera* was made in 1996 at Nadlac, near Hungarian border. In the following years, the pest has spread towards the north-east and the population levels have increased, especially in Caras-Severin, Timis, Arad and Mehedinti counties.

METHOD

In 2001 the WCR monitoring system was organized in 22 counties (10 non-infested counties and 12 WCR infested counties), from 25 June till the end of September. We used Pheromone traps (Csalomon®) and Pherocon AM traps in 21 counties and Romanian Pheromone traps in one county (Bistrita Nasaud). The traps were placed in 202 sites: 120 sites in infested counties and 82 sites in non infested counties. The pheromone traps (traps and capsules) were changed every 30 days and Pherocon traps every 15 days. All the traps were checked every 7 days and the results were recorded on the survey sheets.

RESULTS

This year *D. virgifera* was trapped in 15 out of 22 counties, (much more three counties than in 2000). The new infested counties are: Sibiu - 325 captures, Mures - 14 captures and Bistrita Nasaud - 151 captures. The pest is spreading north- east, along the river Mures and in the southern part of the country no further spread was recorded. The farthest point of spread toward north-east is approximately 400 km away from Nadlac (the first site with captures in 1996).

The total number of captured adults was 29,666 (28,129 on the pheromon traps and 1,537 on Multigard traps). The average value of captures in this year, was 146.9 beetles/ installed trap. The highest population density was in Timis county (1,312 beetles/ trap). WCR population have been increased considerable in counties Arad (from 377.7 beetles/trap in 2000 to 909.8 beetles/ trap in this year) and Timis (from 840.1 beetles/trap in 2000 to 1,312 beetles/ trap in this year). At country level most beetles were caught in August: 17,574 beetles (59%) while in July were caught only 7,539 beetles (25%).

The monitoring of larval damages have been performed in the continuous corn fields from 12 counties which were infested in 2000. This year, the larval damages have been observed in counties Arad, Caras-Severin and Timis, but not at economic level.

EVOLUTION OF THE PEST *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN THE TIMIS DISTRICT

Pălăgesiu Ioan, Grozea Ioana,
Banat's University of Agricultural Sciences Veterinary Medicine Timisoara,
Calea Aradului 119, 1900 Timisoara Romania

Hâncu Mariana,
Phytosanitary Direction of the Timis District, Calea Sagului 140 A,
1900, Timisoara, Romania

Abstract

Diabrotica virgifera virgifera LeConte was signaled for the first time in 1996, in Romania in the Arad District. In 1997 it was detected for the first time in the Timis District too, which neighbors on the Arad District and on Yugoslavia and Hungary.

The paper presents the results concerning the pest's evolution from 1997 to 2001 in the Timis District (South- West of Romania).

For its monitoring FAO pheromone traps (Csalomon®), Romanian pheromone traps (Cluj-Napoca), Multigard yellow stiky traps and cucurbitacin traps were utilized from June to October.

In 1997 the *Diabrotica virgifera virgifera* LeConte adults were very frequent along the Yugoslavian border, but they were also registered in the area near to Hungary. In 1998 the pests spread from West to East, within almost the whole Timis District. The Diabrotica population has increased strongly year by year up to 2001. For monitoring the best results with FAO sex pheromone were obtained. In 2000 the first areas with larval attack in a maize field of the Jimbolia zone were found. In this year the attack is stronger than before.

It results that *Diabrotica virgifera virgifera* LeConte is a large disseminated pest in the Timis District and in consequence its control will be necessary.

Key words: *Diabrotica*, monitoring, trap, flight dynamic.

INTRODUCTION

The first paper which signaled a new potential pest for Romania appeared in June 1995, in "Agricultura Banatului" II, (6, 20) the review of the Banat's University of Agricultural Sciences Veterinary Medicine Timisoara (7). Some days after, this piece of information was taken over by the central Romanian press (Romania Libera).

In 1997, for the first time, adults of *Diabrotica virgifera virgifera* LeConte were captured in the Timis District and a scientific paper concerning the monitoring of the pest in Timisoara appeared (8).

Since 1997 many papers about *Diabrotica virgifera virgifera* LeConte in Romania have been published (9), (10), (11), (12), (20), (21), (13). Very important for our investigations, were the scientific informations from IWGO News letter (1), (3), (2), (4), (14), (15), (18), (19), (22) and also from Yugoslavia (6), (15), (16), (5), (17).

The Department of Entomology from the Banat's University of Agricultural Sciences Veterinary Medicine Timisoara, began to study some aspects concerning this new pest for the Timis District and for Romania.

This paper presents the results of the pest's evolution from 1997 to 2001 in this zone, situated in the South-West of Romania.

MATERIAL AND METHODS

For the *Diabrotica virgifera virgifera* LeConte species monitoring, pheromone traps, traps Multigard yellow, sticky traps and cucurbitacin traps were used. The observations were carried out from June to October, in 124 centers in 1997, in 60 in 1998, in 52 in 1999, in 42 in 2000 and 9 in 2001. The observations in all five Plant Protections Centers of the Timis District (Deta, Sannicolaul Mare, Timisoara, Topolovatul Mare, Lugoj) were performed by the center's specialists weekly or daily. The stuff members of the Department of Entomology from the Banat's University of Agricultural Sciences Veterinary Medicine Timisoara were also involved in the investigations concerning this pest.

RESULTS AND DISCUSSIONS

In Romania *Diabrotica virgifera virgifera* LeConte species was signaled for the first time in Nadlac, Arad District in 1996 (*figure 1*). In 1997 it was detected in the Timis District too. This District neighbors on the Arad District and also Yugoslavia and Hungary.

In July 1997 the pest was signaled frequently in the Timis District especially along the Yugoslavian border (*figure 2*). In August 1997 it was signaled along the Hungarian border and it penetrated also into the inner part of the Timis District (*figure 3*). In September, the pest was noticed in 89 centers of the District (*figure 4*).

In 1998 *Diabrotica virgifera virgifera* LeConte species spread out all over the Timis district, from the West to the East, to the Carpathian Mountains zone (*figure 5*). In 1999 the pest was disseminated within almost the whole District.

The Diabrotica populations increased strongly year by year up to 2001, from 15.46 adults/trap in 1997 to 1216.65 adults/trap in 2001 (*table 1*). The population dynamics indicates a great increasing during the last two years, in 2001 especially (*figure 6*).

The adults were captured all over the District, but in the inner part (Topolovatu Mare, Lugoj) the number was less than in the central and western zones (*table 2*). Generally the greatest number of adults was captured in August in all the years.

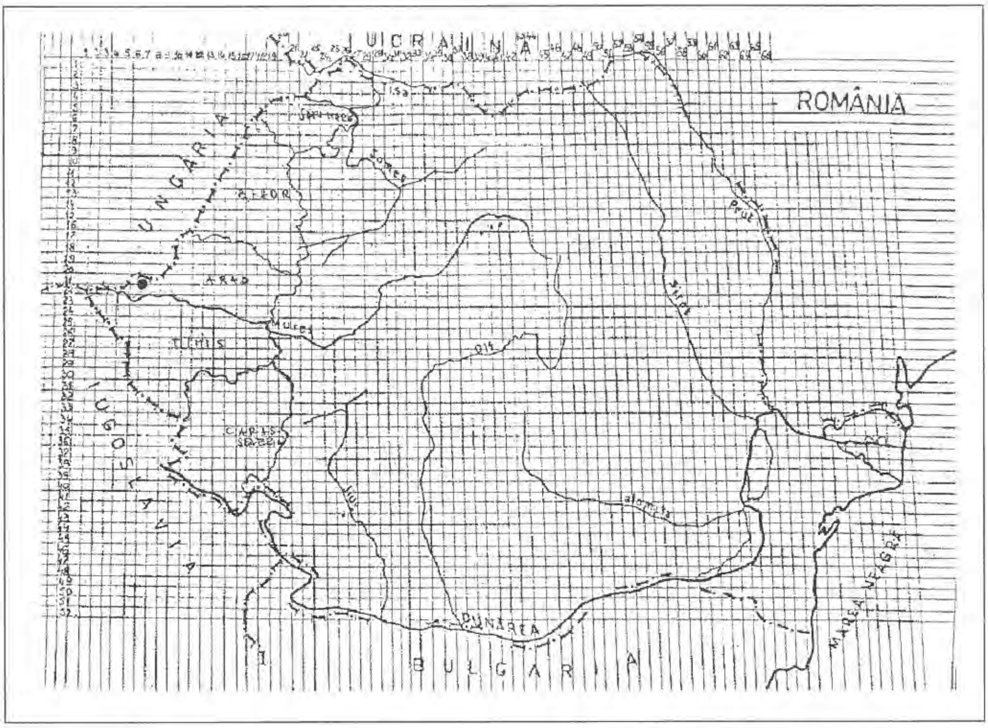

Figure 1 - The distribution of *Diabrotica virgifera virgifera* LeConte in Romania (1996)

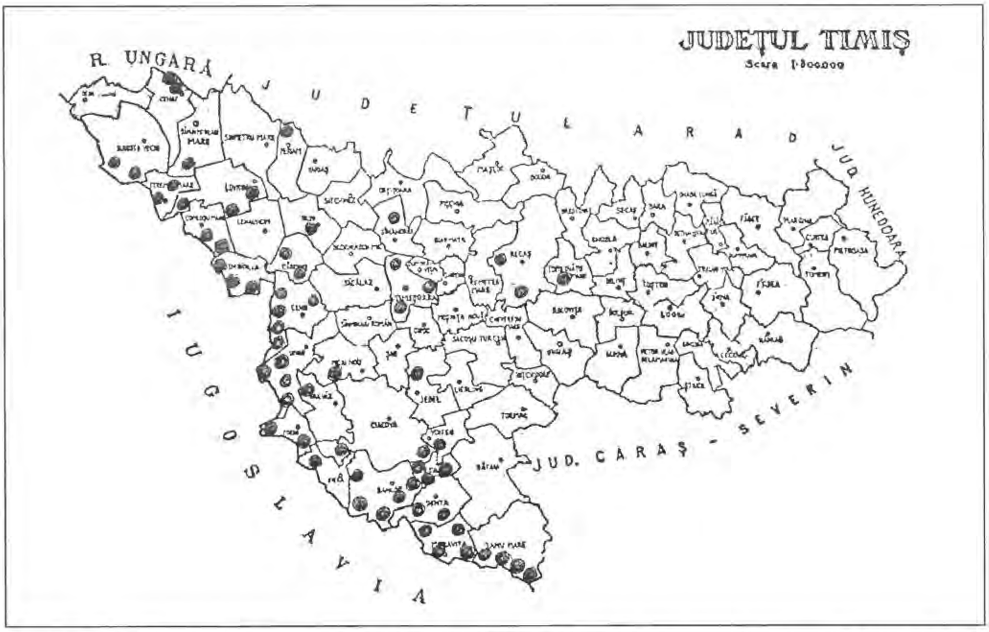

Figure 2 - The distribution of *Diabrotica virgifera virgifera* LeConte in the Timis District - July 1997

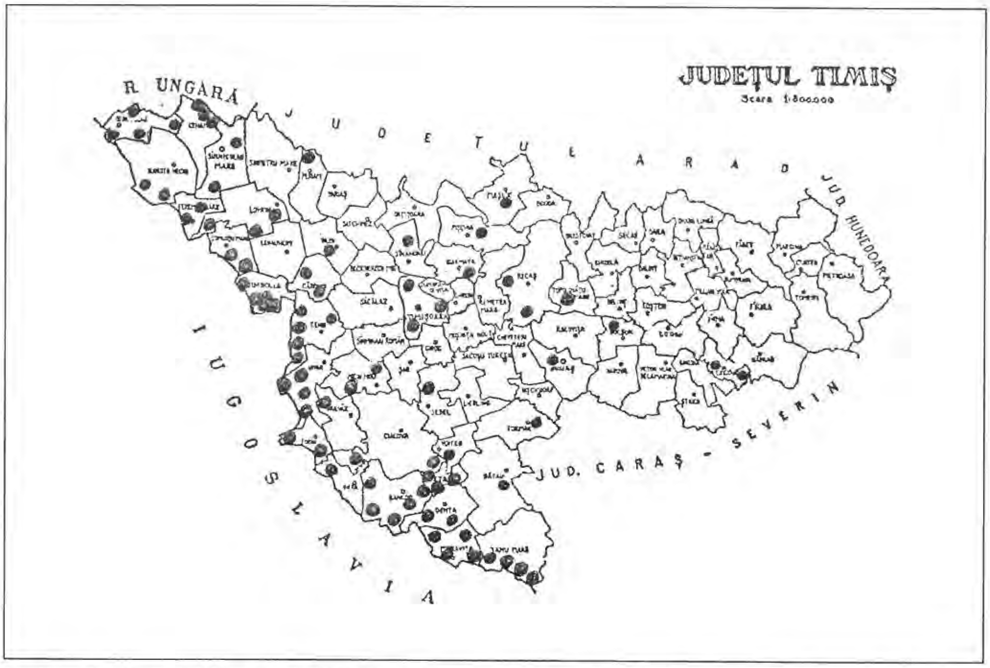

Figure 3 - The distribution of *Diabrotica virgifera virgifera* LeConte in the Timis District - August 1997

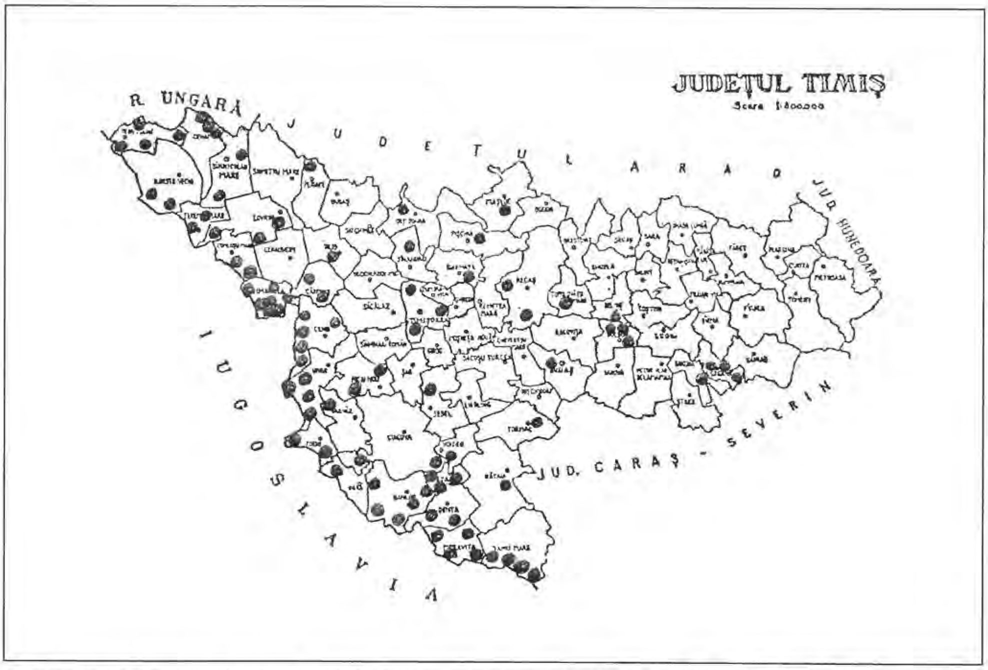

Figure 4 - The distribution of *Diabrotica virgifera virgifera* LeConte in the Timis District - September 1997

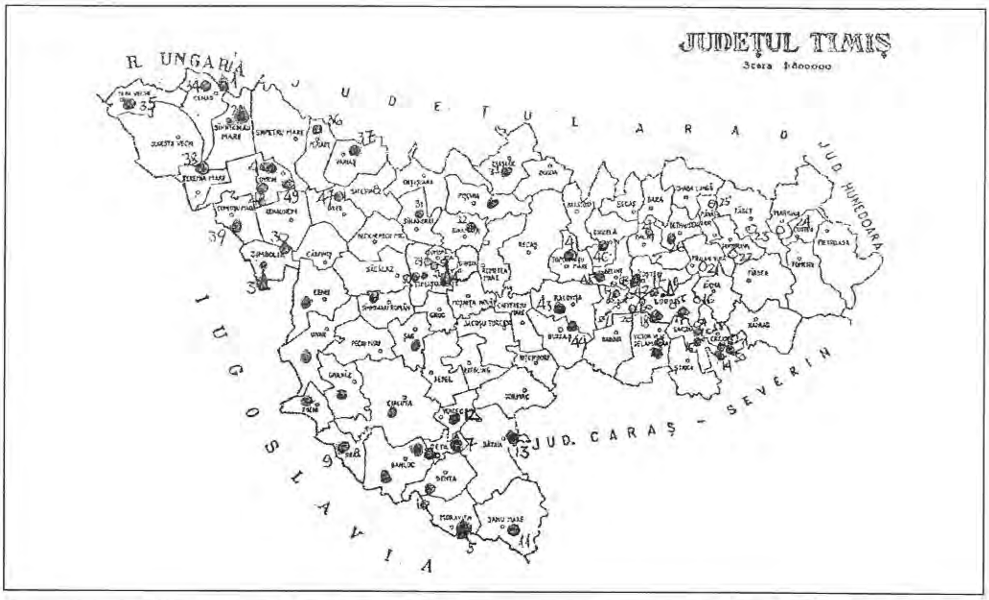

Figure 5 - The distribution of *Diabrotica virgifera virgifera* LeConte in the Timis District in 1998

Table 1 - Number of captured adults/trap in 1997 - 2001 (Timis District)

Year	July	August	September	October	Mean
1997	15.46	71.18	45.34	-	43.39
1998	26.23	53.40	38.55	0.91	29.69
1999	43.90	216.73	30.94	2.29	72.46
2000	388.11	347.33	76.00	-	270.48
2001	432.88	636.44	147.22	-	1216.55

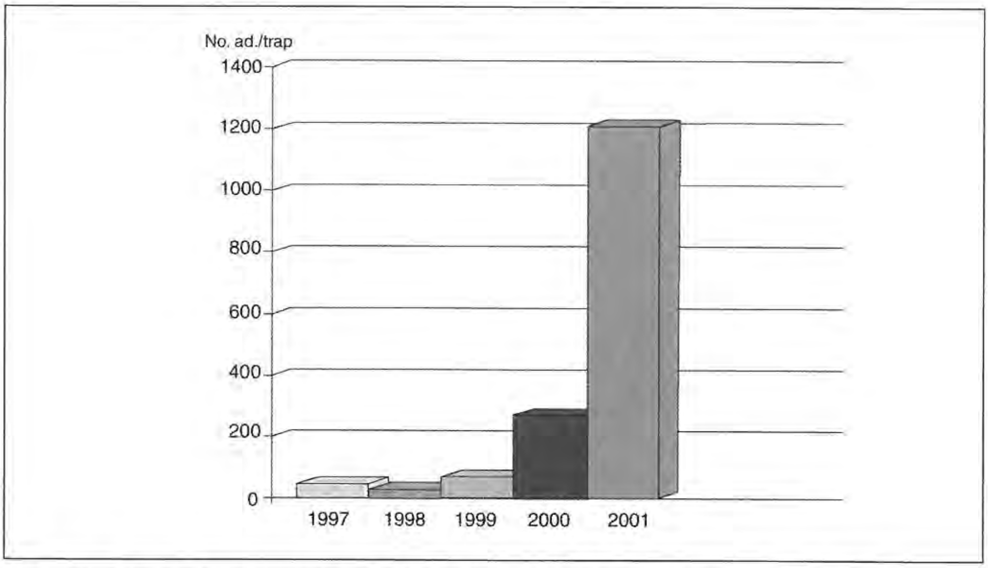

Figure 6 - The *Diabrotica virgifera virgifera* LeConte adults dynamics population

Table 2 - Evolution of the captured adults/trap in the July - October period

Year	Plant Protection Centers	July	August	September	October	Mean
1997	Deta	9.66	53.90	37.40	-	33.65
	Sanicolaul Mare	1.91	6.81	0.89	-	3.20
	Timisoara	3.70	10.06	3.95	-	5.90
	Topolovatul Mare	0.08	.30	0.08	-	0.15
	Lugoj	0.00	0.96	0.35	-	0.40
1998	Deta	13.46	32.98	27.61	0.30	18.59
	Sanicolaul Mare	4.11	14.23	6.55	0.38	6.31
	Timisoara	7.35	4.63	1.36	0.21	3.38
	Topolovatul Mare	0.00	1.30	0.48	0.01	0.40
	Lugoj	1.30	0.25	2.53	0.00	1.02
1999	Deta	15.86	131.61	3.75	-	37.89
	Sanicolaul Mare	6.09	21.61	8.57	-	9.43
	Timisoara	11.05	29.01	12.29	-	13.20
	Topolovatul Mare	9.92	29.61	2.51	-	10.54
	Lugoj	0.90	4.86	3.75	-	2.37
2000	Deta	101.97	133.11	36.25	-	90.47
	Sanicolaul Mare	67.02	123.23	28.11	-	72.78
	Timisoara	120.21	65.97	6.21	-	64.13
	Topolovatul Mare	32.21	10.21	4.28	-	15.56
	Lugoj	66.54	14.07	1.02	-	27.21
2001	Deta	96.66	148.22	10.88	-	85.25
	Sanicolaul Mare	51.22	161.44	7.77	-	73.48
	Timisoara	250.00	323.77	121.77	-	231.84
	Lugoj	35.00	30.77	6.77	-	24.18

The best results for the monitoring were obtained with sex pheromone traps (Csalomon®). Since 1998 the sex pheromone traps Cluj-Napoca and the traps with cucurbitacin have been eliminated because the results were not satisfactory (*table 3*).

In 1998 the daily adult's flight in Timisoara was observed by the staff of the Department of Entomology at Didactic station of Banat's University of Agricultural Sciences Veterinary Medicine.

The flight of adults began in the second decade of July and its maximum was registered in the third decade of July and also in the second decade of August. The last adults were captured in the first decade of October (*figure 7*).

The first adults were captured in 1999 in the third decade of June, the largest number of adults registered in the third decade of July, the third decade of August respectively (*figure 8*).

In 2000 the first adults were captured in the third decade of June and last ones in the third decade of September. The largest number of captured adults was registered in the second decade of July and another increase was noticed in the third decade of July- first decade of August (*figure 9*). In 2001 three increases of the adult flight were registered (*figure 10*).

In 2001 the first areas with larval attack was found in a maize field of the Jimbolia zone. In that year the attack was stronger than before (*figure 11*).

In the Timis District the *Diabrotica virgifera virgifera* LeConte species is now a largely disseminated pest and therefore its control is necessary in the next years.

Table 3 - The efficiency of the different traps for capturing *D.v.v.* adults

Year	Plant Protection Centers	Sex pheromone traps		Multigard yellow	Cucurbitacin
		Csalomon R	Cluj-Napoca		
1997	Deta	82.58	15.00	7.19	0.00
	Sanicolaul Mare	8.86	0.07	0.77	0.00
	Timisoara	13.72	31.39	0.45	0.00
	Topolovatul Mare	0.95	0.00	0.01	0.00
	Lugoj	0.45	0.00	0.00	0.00
1998	Deta	68.25	-	5.78	-
	Sanicolaul Mare	23.03	-	2.25	-
	Timisoara	12.21	-	1.35	-
	Topolovatul Mare	1.80	-	0.00	-
	Lugoj	4.08	-	0.00	-
1999	Deta	151.11	-	8.36	-
	Sanicolaul Mare	36.99	-	0.76	-
	Timisoara	52.40	-	0.50	-
	Topolovatul Mare	41.50	-	0.50	-
	Lugoj	9.51	-	0.00	-
2000	Deta	267.25	-	3.95	-
	Sanicolaul Mare	216.42	-	1.95	-
	Timisoara	180.11	-	12.04	-
	Topolovatul Mare	46.21	-	0.59	-
	Lugoj	80.80	-	0.83	-
2001	Deta	221.22	-	34.55	-
	Sanicolaul Mare	183.00	-	37.44	-
	Timisoara	632.44	-	44.33	-
	Lugoj	72.55	-	-	-

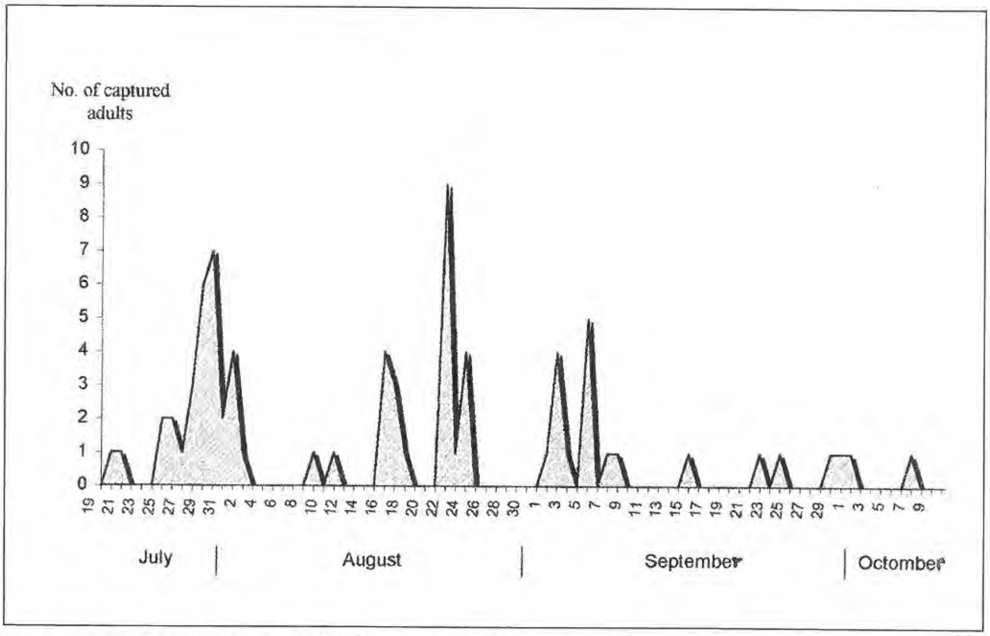

Figure 7 - The daily adults flight dynamics in Timisoara - 1998

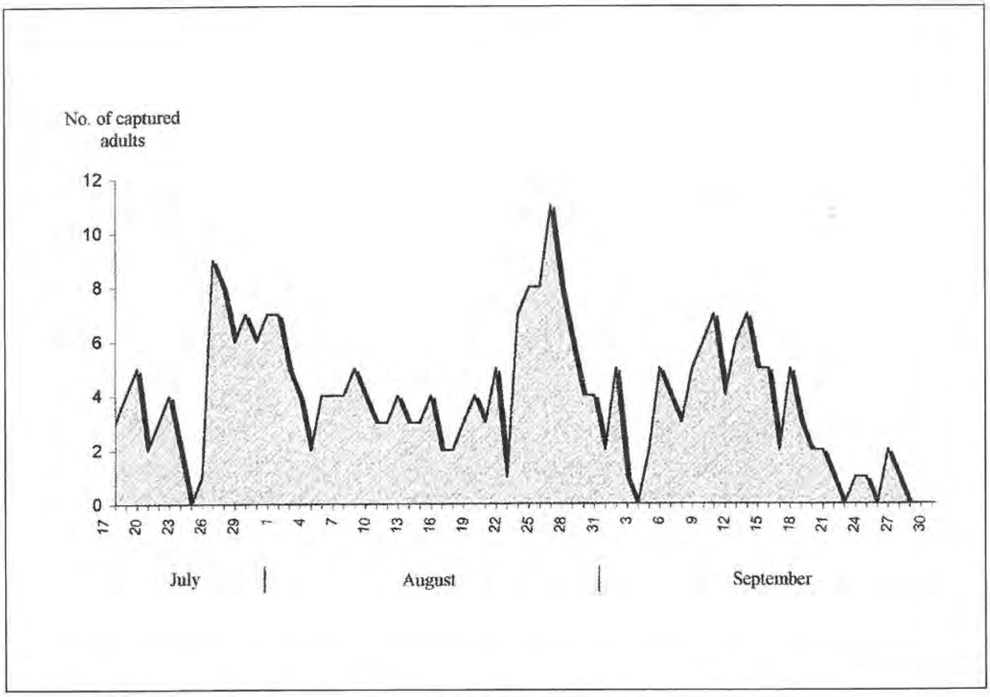

Figure 8 - The daily adults flight dynamics in Timisoara - 1999

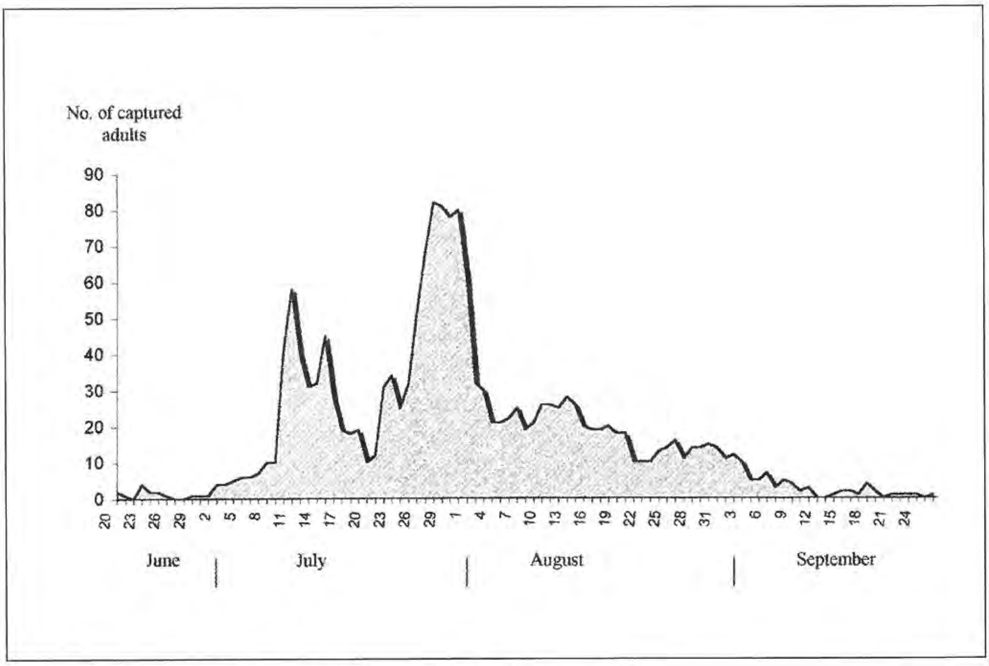

Figure 9 - The daily adults flight dynamics in Timisoara - 2000

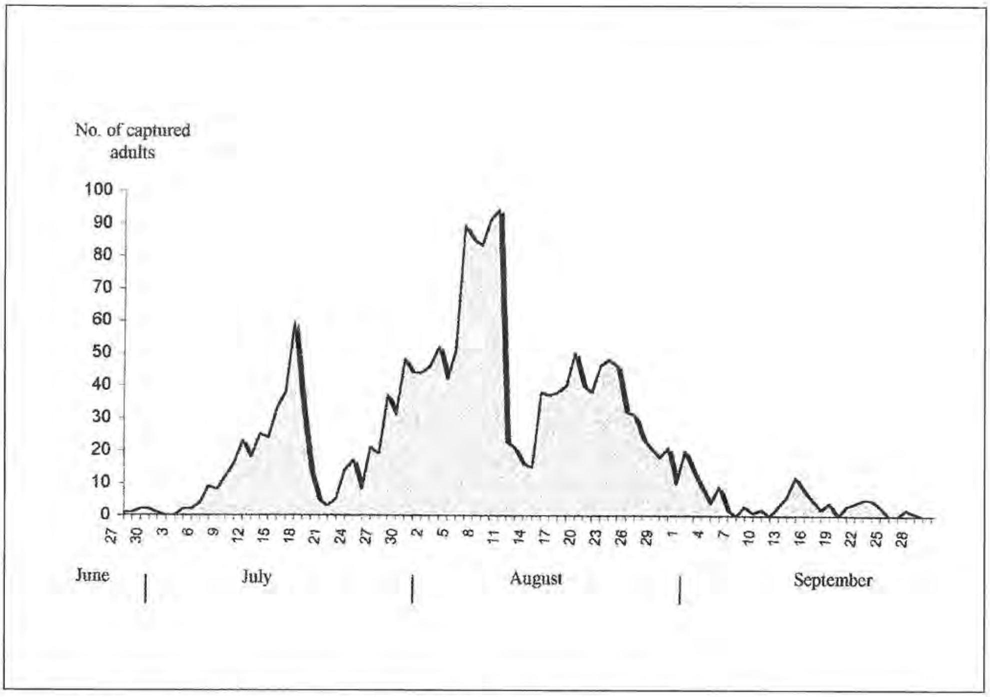

Figure 10 - The daily adults flight dynamics in Timisoara - 2001

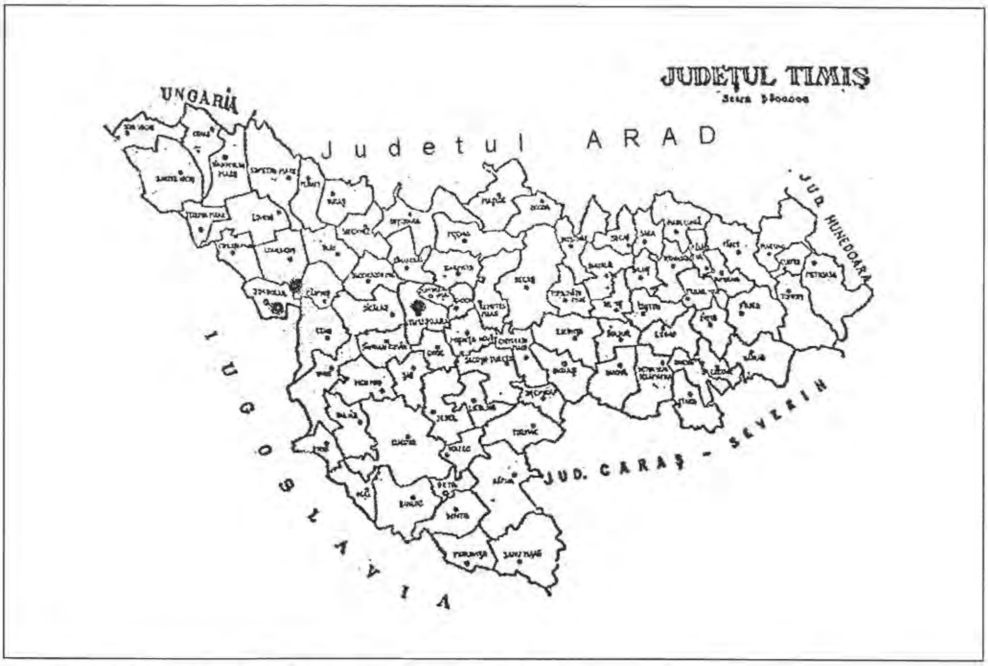

Figure 11 - The areas with *D.v.v.* larval attack in 2001

CONCLUSIONS

The *Diabrotica virgifera virgifera* LeConte species which was detected in the Timis District for the first time in 1997 is in 2001 a largely disseminated pest.

In 1997 it was signaled very frequently along the Yugoslavian and Hungarian border but it penetrated throughout the whole District.

The number of the captured adults increased strongly year by year up to 2001.

The best results for monitoring were obtained with sex pheromone traps. (Csalomon®).

The first signalization of adults was registered between the third decade of June and the third decade of July.

The period of maximum flight was between the third decade of July and the third decade of August. The last adults were captured between the third decade of September and first decade of October.

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EDUCATION AS A FACTOR IN SUPPRESSION OF WCR IN SERBIA

Ivan L. Sivcev, Anicka J. Galo,
Institute for Plant Protection and Environment,
Banatska 33 - 11080 Zemun, Yugoslavia

Abstract
More than 30% of the corn production in Serbia is in repeated sowing or long term continuous corn. In some areas in southern Banat even 80% of arable land was under the corn. Due to serious damages in consecutive 2 or 3 years farmers started with crop rotation which led to decrease of surface under the corn and decrease of WCR population density. In general, there are 10% less cornfields, but in some regions more than 30%. Since farmers are rotating crops without risk assessment it appears that their training is important for justified crop rotation or insecticide application and minimization of damages.

Key words: Corn, *Diabrotica virgifera virgifera*, damages, risk assessment.

INTRODUCTION

Serbia is agricultural country with 3,670,091 ha of arable land. Cornfields are covering 38% of that land. Majority of the corn production is in the northern part of Serbia, predominantly in Vojvodina which has 1,586,994 ha of arable land and percentage of land under corn in that area is - 43%. Farm size is rather small since 77% of the farmers has less than 5 ha of land. Average farm size is 2.8 ha (Anonymous, 2000). Corn production is mainly on private farms.

Ever since corn has been brought to Serbia domestic flora and fauna caused the major problems concerning corn growing. However, the apparent damage has never reached a level which can endanger production seriously. Widespread corn growing in repeated sowing induce population of the certain insects such as *Tanymecus dilaticolis* Gyll to increase (Camprag, 1994).

The occurrence of a new insect species, *Diabrotica virgifera virgifera* LeConte (WCR), which is one of the most important corn pest in the North-American continent, caused worry of corn producers. Fast spread and population density increase in Serbia is related to a significant participation of corn in a repeated sowing, particularly in a long term continuous corn, the favorable climatic and soil conditions as well as a slight regulation effect of natural enemies (Camprag, 1998).

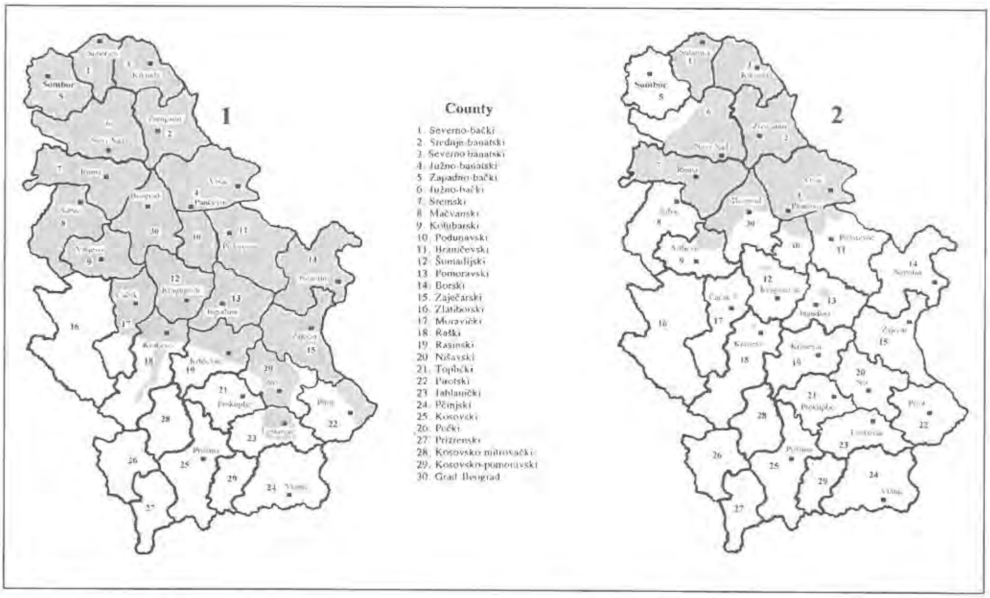
The aim of this paper is to present data on harmful effects of WCR and consequences on corn production in Serbia and to suggest necessity of introducing IPPM.

DAMAGES CAUSED BY WCR

For many years, corn production was managed in repeated sowing and in long term continious corn in 30-40% of land under the corn. On small private farms participation of cornfields on arable land was more than 50% (Gotlin, 1987). Certain areas, neighboring introduction site of WCR, are well known by this practice.

Due to this, population of WCR reached economic level on large area vary fast and damages on corn root from larval feeding occurred. At the beginning, some damages were recorded on the cornfields near airport. Since 1994, majority of damages occurred in southern Banat in villages surrounding city Pancevo. In 2001 an area of approximately 72250 km^2 is infested with WCR (*map 1*). Within that area approximately 26477 km^2 is already registered with economic population level and damages (*map 2*).

Economic population was detected on time, by using pheromone traps, yellow sticky traps and by visual checking of the fields. Farmers were informed about the presence of new corn pest and advised to rotate the crop or to apply soil insecticides.



Map 1 - Infested territories　　Map 2 - Territories were damages occurred

But, many farmers continue growing the continuous corn without applying the insecticides. Similar way of farmer's behavior occurred in other newly infested areas with economic population.

During the period 1992 – 2001 we registered more than 140,000 ha of damaged cornfields (*table 1*). Our estimation on average yield losses is 30% or more than 1 ton per ha. It means than farmers lost about 150,000 tons of corn during that period or equivalent of 22 million US$.

Through extension service, strong propaganda for crop rotation or application of soil insecticides were made. Local media also distributed informations on pest distribution and presence of economic population in many regions of Serbia.

Under present economic situation and with low corn yield during long period of year, farmers did not started to apply soil insecticides. Since it is evident that their needs for corn is significant, they continued with traditional practice of repeated sowing. After having serious damages for consecutive two or three years (and even more years) farmers started to rotate crops. From *table 2* one can see what were the consequences for the corn production in the area of southern Banat where serious damages were recorded during the period of last 6 years. In that area, only in 1996, there were 9,800 ha of damaged cornfields. Out of this, estimated yield loss on 7,600 ha was 20%, on 600 ha up to 30%, on 200 ha up to 40%, 1,220 ha up to 50% and on 200 ha up to 80%. In the following years, reported damaged cornfields were each year lower. It is clear that application of crop rotation resulted in significantly lower surface under the corn. For the whole region there are 21,511 ha or 26% less corn fields than it used to be. In some communities (Opovo) there are decreases of corn fields of 32%.

These represent a second significant point of damages made by WCR.

Table 1 - Corn sown area and damaged corn fields registered in Serbia 1992-2001

Year	Corn sown area in Serbia (000 ha)	Damages from WCR (ha)
1951-1960	1,472	
1961-1970	1,461	
1971-1980	1,411	
1981-1990	1,409	
1991	1,351	
1992	1,510	0,5
1993	1,383	6
1994	1,377	60
1995	1,366	275
1996	1,433	10,787
1997	1,360	5,696
1998	1,351	45,513
1999[1]	1,267	30,543
2000	1,203	50,000
2001	1,219	<1,000

[1] Since 1999 without corn sown surface in Kosovo (95,000 ha)

Table 2 - Corn sown area in southern Banat 1996 - 2001

Region	Arable land (ha)	1996		1997		1998		1999		2000		2001	
		Corn sown area (ha)	% of arable land	Corn sown area (ha)	% of arable land	Corn sown area (ha)	% of arable land	Corn sown area (ha)	% of arable land	Corn sown area (ha)	% of arable land	Corn sown area (ha)	% of arable land
PRIVATE FARMS													
Pancevo	37,291	27,977	75%	25,365	68%	24,123	64%	21,922	59%	22,200	59%	20,862	56%
Kovacica	27,763	20,700	75%	18,820	68%	17,175	64%	13,562	53%	14,359	45%	13,412	51%
Opovo	11,846	9,841	83%	8,070	68%	7,604	59%	7,190	57%	6,633	51%	6,669	51%
Alibunar	32,840	24,116	73%	23,605	72%	22,130	67%	18,844	57%	20,019	60%	20,180	60%
STATE FARMS													
Pancevo	21,469	6,252	29%	6,240	29%	6,200	28%	4,999	23%	5,130	24%	5,799	27%
Kovacica	7,498	2,521	34%	2,270	30%	2,100	29%	2,084	25%	2,108	26%	2,087	26%
Opovo	2,200	390	18%	545	25%	705	45%	622	27%	248	12%	735	35%
Alibunar	11,385	3,002	26%	2,938	26%	2,855	25%	2,489	22%	2,670	24%	2,620	23%

From *table 1* one can see that process of decrease of corn fields is present on the whole territory of Serbia. It is clear that main reason for that is crop rotation and inability of farmers to apply soil insecticides. In 2001 there are about 10% or 100,000 ha less corn fields.

NECESSITY FOR INTEGRATED PRODUCTION AND PEST MANAGEMENT

There is no doubt that traditional practice of long term growing of continuous corn should be reduced. Also, it is our intention to continue recommending crop rotation or insecticide application only when it is necessary but not as a general rule. So far farmers' practice showed that they prefer continuous corn growing up to the point when it starts to be irrational due to damages from WCR. Than they start with crop rotation without estimating real necessity for it.

In 2001 we registered, in some areas, significant decrease of WCR population which can be partly explained by adverse climatic conditions in previous year (2000) but mainly with decrease of surface of continuous corn fields.

Under the program supported by Ministry of Agriculture and FAO, farmers' education and training on risk assessment started in 2001. On two different locations groups of corn producers from private sector were chosen through their Farmers Cooperative Organizations. The program is based on "The Whole–Plant Count Technique" and "The Pherocon AM Sticky Trap Technique" which are widely recommended to US farmers (Ostlie and Noetzel, 1987, Edwards *et al.*, 1997). We noticed that farmers are rotating the crop, as a general rule, without estimating real need for that. In the majority of cases it was possible to repeat corn sowing on the same field.

Having in mind that production of corn is only on 38% of arable land in Serbia, it appears that decrease of surface under the corn is not justified as well as appearance of significant damages from WCR. Also, it can be expected that market needs for corn seed will strongly influence new concept of its production in which risk assessment and IPPM will take its part.

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MONITORING OF WESTERN CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) IN HUNGARY IN 2001

Géza Ripka,
Central Service for Plant Protection and Soil Conservation,
H-1519 Budapest, P.o.b. 340., Hungary

Gábor Princzinger,
Ministry of Agriculture and Regional Development,
H-1860 Budapest, 55. P.o.b. 1., Hungary

INTRODUCTION

The first occurrence of Western corn rootworm (*Diabrotica virgifera virgifera* LeConte, 1868) in Hungary was recorded in the southern part of county Csongrád, close to the Yugoslavian border in 1995. The new maize pest spread considerable rapidly throughout Hungary.

MATERIALS AND METHODS

The Hungarian Plant Protection Organisation has carried out a nation-wide survey of Western corn rootworm in Hungary since 1996. The Plant Protection and Soil Conservation Services in all counties have participated in the study. Similarly to the previous year, in 2001 too, the Western corn rootworm monitoring system was based on two examinations:

1. *Scout trapping*

On 49 locations where western corn rootworm was absent in 2000, Hungarian sex-pheromone traps (Csalomon®) were placed out in maize fields. In the non-infested areas of 8 counties, the traps were monitored from 11 June till the middle of October at 10-day intervals. Within 30 days the traps were replaced by new ones.

2. *Permanent Monitoring Network*

To determine the pest population build-up Hungarian pheromone traps (Csalomon®) and Pherocon® AM yellow sticky traps were placed out in pairs on 35 sites of 16 western corn rootworm-infested counties. The distance between pheromone and Pherocon® AM traps was 50 m. In maize fields, the traps were monitored from 11 June to middle October. From 2 July Multigard® yellow sticky trap and flower volatile baited PALs trap were also used for trapping on 15 and 7 sites, respectively. All traps were changed at intervals of 30 days. Catches were recorded decadely.

RESULTS

Western corn rootworm was trapped in all 19 Hungarian counties.

Out of 49 scout traps, 17 ones had catches over western corn rootworm-infested area. These traps caught a total of 336 beetles. The average number of catches was 6.9 adults/trap.

Out of 35 permanent monitoring pheromone traps, 34 ones had catches. A total of 19,030 beetles were caught. The average catch was 543.7 beetles/trap. It is significantly higher than in 2000. Out of 35 Pherocon® AM traps catches were observed on 25 traps. 3,060 adults were captured. The average catch was 87.4 beetles/trap. Multigard® sticky traps caught more beetles, viz. 4,559, on average 303.9 beetles/trap, than did Pherocon AM traps. The two trapping systems caught a total of 26,985 western corn rootworm adults, much more than in 2000 (16,211 beetles). In addition the 7 PALs traps caught a total of 5,085 adults. The average catch was 726.4 beetles/trap.

Western corn rootworm population considerably increased in counties Fejér, Tolna, Somogy, Baranya, Békés and Hajdú-Bihar. At country level, most beetles were caught in August, 13,672 adults (52.4%), but the catch in July was also high, 12,073 beetles (46.2%). In June 339 adults were trapped only. It is much less than in 2000 (1,945 beetles). But in September and even in October the adults were caught surprisingly in high number. So the flight activity of the pest was long. In southern part of Baranya, Tolna and Bács-Kiskun counties, in long-term (lasting 5-7 years) continuous corn fields, the adult density was very high in the last decade of July and first decade of August. In these counties, several hundreds of hectares of corn fields were sprayed with insecticides using aerial application, in the beetle control programme.

The invasive species was found in counties Vas, Győr-Moson-Sopron and Szabolcs-Szatmár-Bereg where it was absent in the previous year. The farthest points of the insect spread to the northwest and northeast is about 40 km.

In 10 western corn rootworm-infested counties *Diabrotica* larval damages were surveyed in 955 maize fields on 44,895 ha. It is approximately 5% of the total maize fields of these counties. A vast majority of the inspected fields was continuous corn field. The Iowa scale was used for the evaluation of root damage. In counties Baranya, Bács-Kiskun, Békés, Csongrád and Tolna larval damages were observed on 10,311 ha. In counties Baranya, Bács-Kiskun and Tolna the root damages reached the economic level (3<Iowa scale) on 3,058 ha. Lodging plants were found in several corn fields of these counties.

The Hungarian Plant Protection Organisation carried out several trials for the efficacy evaluation of insecticides against Western corn rootworm adults. There are some highly efficacious compounds in the tested products with good knock-down activity and persistence. The organo-phosphates are more persistent than pyrethroids. Endosulfan provides an average persistence.

Table 1 - Results of Western Corn Rootworm scout trapping in Hungary in 2001

County	Location number	WCR detected loc.	June	July	August	September	Total	Average beetles/trap
Zala	3	0	0	0	0	0	0	-
Vas	5	0	0	0	1	0	1	0.2
Veszprém	5	4	0	3	131	2	136	27.2
Győr-M.-S.	8	1	0	3	12	0	15	1.9
Komárom-Esztergom	5	4	0	4	101	16	121	24.2
Heves	5	0	0	0	0	1	1	0.2
Borsod-A.-Z.	4	2	0	9	0	0	9	2.3
Szabolcs-Sz.-B.	15	6	0	35	18	0	53	3.5
Sum total	49	17	0	54	263	19	336	6.9

Table 2 - Results of Western Corn Rootworm permanent trapping in Hungary in 1997-2001

Year	Location number	Caught beetles			Average beetles/trap		
		pheromone	Multigard	Pherocon AM	pheromone	Multigard	Pherocon AM
1997	16	2395	-	-	147.9	-	-
1998	16	1790	84	-	74.3	3.6	-
1999	19	6407	909	-	337.2	47.8	-
2000	27	10014	4586	925	370.9	229.3	54.4
2001	35 (15#)	19030	4559#	3060	543.7	303.9#	87.4

Table 3 - Results of the survey of Western Corn Rootworm larval damage in Hungary in 2000

County	Number of inspected fields (locations)	Inspected area (ha)	Area with WCR root-damage (ha)	Average damage (Hills-Peters scale)
Baranya	80	4215	556	1.22
Bács-Kiskun	62	4203	2133	2.05
Békés	78	2644	310	2.82
Csongrád	119	5158	104	3.52
Tolna	57	2791	0	-
Somogy	26	1789	0	-
Fejér	81	5935	0	-
Pest	92	3287	0	-
Jász-Nagykun-Szolnok	80	3771	0	-
Hajdú-Bihar	294	7564	0	-
Sum total	969	41357	3103	2.4

Table 4 - Results of the survey of Western Corn Rootworm larval damage in Hungary in 2001

County	Number of inspected fields (locations)	Inspected area (ha)	Area with WCR root-damage (ha)	Average damage (Hills-Peters scale)
Baranya	197	11727	7588	2,29
Bács-Kiskun	125	4006	699	1,98
Békés	78	3771	1357	2,39
Csongrád	107	4331	413	1,75
Tolna	52	2828	254	4,36
Somogy	63	3816	-	-
Fejér	90	5028	-	-
Pest	75	2715	-	-
Jász-Nagykun-Szolnok	96	4308	-	-
Hajdú-Bihar	72	2365	-	-
Sum total	955	44895	10311	2,55

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RESULTS OF THE WESTERN CORN ROOTWORM *DIABROTICA VIRGIFERA VIRGIFERA* MONITORING IN THE SLOVAK REPUBLIC IN 2001

Peter Sivicek,
Central Control and Testing Institute of Agriculture,
Hanulova 9/A, 844 29 Bratislava 42, Slovak Republic

Abstract
The phytosanitary inspectors of the Central Control and Testing Institute of Agriculture carried out the WCR monitoring in Slovakia in 2001 as in some years ago. The monitoring was carried out in Slovakia within framework of the FAO project for development of sustainable strategies for monitoring and management of the population WCR in Eastern Europe. Our results confirm the WCR ability to spread to new corn localities.

Key words: Western Corn Rootworm, *Diabrotica virgifera*, monitoring, Slovak Republic.

INTRODUCTION

The corn (mainly for grain and silage but also for seeds) was grown in the Slovak Republic on an area of 220,000 ha in 2001. The corn area in several districts of Slovakia depends on local climatic conditions. The WCR occurred first time in Slovakia in 2001. We continued on WCR monitoring within framework of the FAO project for the development of sustainable strategies for monitoring and management of the population WCR in Eastern Europe.

METHODS AND PROCEDURE

The phytosanitary inspectors of the Central Control and Testing Institute of Agricultural carried out the monitoring in 2001 as in some years ago. The monitoring net was created by 58 monitoring sites. For the monitoring was used 174 pheromone traps Csalomon and 165 Pherocon AM traps from mentioned FAO project. Two types monitoring sites was created. The Csalomon and Pherocon AM traps were located either together or pheromone trap was set separately on monitoring sites. Traps were not located on pair into monitoring sites.

Both types of the traps Csalomon and Pherocon AM have been put along the south state border with Hungary and also into localities where the WCR occurred first time in 2000. The pheromon traps (without Pherocon AM) were put to the margin of the mentioned regions where we predicted the WCR occurrence in this year. The monitoring sites had been located also near the airports in Bratislava and Košice.

The monitoring started on June 18th, 2001 and was finished on September 30th, 2001. The traps had been checked every week. The individuals caught on traps had been taken off from sticky place of the trap and its amount was noted. The pheromone traps were changed every 30 days. It was twice during monitoring season. The Pherocon AM traps were changed every 21 days, i.e. four times during monitoring season.

Results and conclusion

The first WCR beetle was found on July 4th in locality Štúrovo (district Nové Zámky). We can mention for comparison the occurrence of the first beetle in 2000 was noted on August 7th in locality Kosihy nad Ipl'om (district Vel'ký Krtíš). The WCR occurrence was found about 34 days early in 2001 than in 2000.

However we assume these results are not quite sufficient for assessment of the WCR populations development in the Slovak Republic. The WCR occurrence in 2000 was first in Slovakia and we found only basic information on its presence (we caught together 12 adults) in our territory.

The maximal number of the beetles was caught in the monitoring period from July 16th up to August 26th, 2001 (*figure 1*).

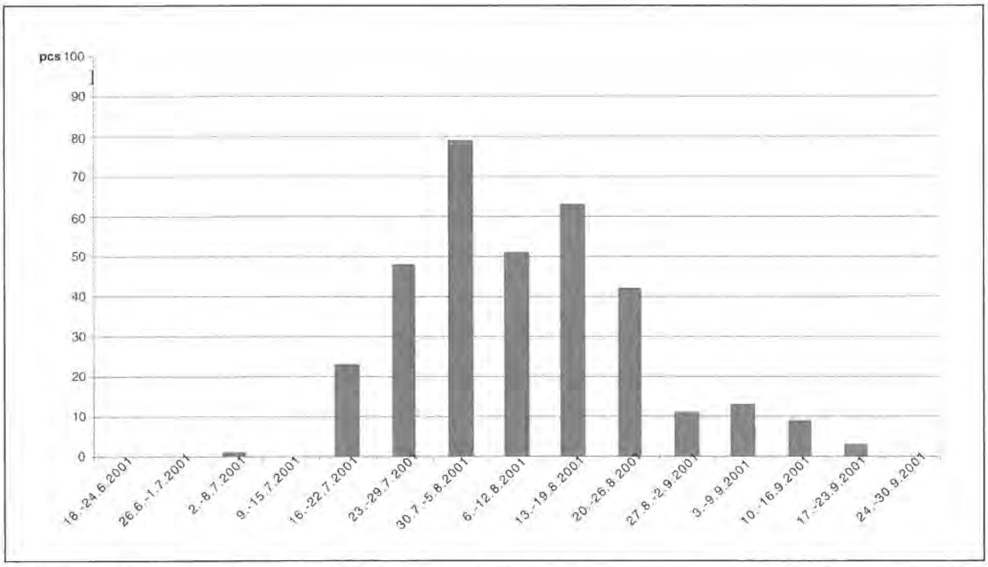

Figure 1 - Number of the WCR beetles cought into pheromone traps in the Slovak Republic in 2001

The WCR infested region with area approximately 6,300 km^2 in 2001. The corn was grown in this region in an area of approximately 68,095 ha. The region with area approximately 500 km^2 was infested in 2000. The corn had been grown on an area of 5,303 ha in that time.

The pest spread mainly to the next south districts of the east Slovakia in 2001 (*figure 2*). The spread to new localities was not so much significant as we predicted in middle and east part of the south Slovakia. The monitoring points were set also in south - east districts of Slovakia however the pest was not caught there.

The WCR is present along river Danube in distance 10 km from Hungary. The pest is present also near river Ipel' that is also bordering with Hungary.

The damages on the corn made by the adults or the larvae were not found out in this year.

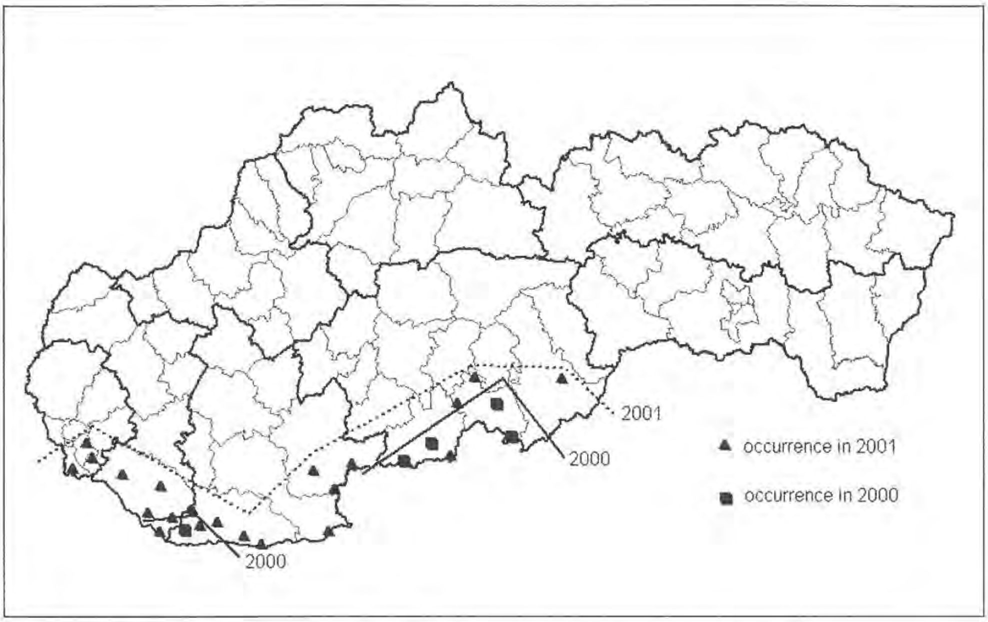

Figure 2 - Spread of the Western Corn Rootworm - *Diabrotica virgifera virgifera* - in the Slovak Republic in 2000 - 2001

MONITORING OF *DIABROTICA VIRGIFERA VIRGIFERA* IN BULGARIA IN 2001

Ivanka Ivanova,
Central laboratory for plant quarantine,
120 Gornobanski Blvd., Sofia 1330, BG

Abstract

The *D. virgifera* has been found for the first time in Bulgaria in 1998. The pest has spread in northwest in Bulgaria near the borders with Serbia and Romania.

In 2001 the monitoring had the following goals:

- permanent observation posts for reporting of the population density;
- limits of the dissemination of the pest;
- eventual damages in the area in which the corn has been groun as monoculture and where in year 2000 were determined adults.

Methods used: For discovering of larvae in the region where in 2000 were determined higher numbers of adults of WCR in May-June soil digging was completed. Soil samplings were taken in Bregovo, Dolna Bijala Retchka, Prevala and in other places in the regions of Vidin and Montana. Soil digging was accomplished from selected places where plants obviously with deviation from the norms of development and also occasionally normal plants were chosen.

The WCR monitoring program for determination of the limits of the spreading and the population density continued with 40 yellow sticky traps and 130 pheromone traps. They were situated on 41 monitoring sides (8 permanent sites). They were situated of Northwest Bulgaria. The monitoring started in beginning of July in the regions of Vidin, Vratza, Sofia.

Results: Until now there were not found larvae and damages by them in the digging activities on places. On 12.07 in Bregovo were captured the first adults -12, in D. B. Retchka – 6. The higher numbers was caught in Gramada –548. Followed by highest numbers in Prevala –265, Bregovo –149. Total numbers of trapped specimens was 1,795.

Conclusion: 1. The distribution of WCR continued forward South and East. 2. For the first time damages were determined in the corn silk and leaves in Prevala. Damages in other area where great density of WCR was considered, were not determined. 3. The pest has been found near town Kneja and also south of Stara planina (near Godetch). 4. From several years in the region of Lom, where the pest was discovered for the first time, adults were not found.

Acknowledgements: FAO support of WCR Network activity , prof. J. Kiss, prof. R.Edwards.

FARM LEVEL MANAGEMENT OF WESTERN CORN ROOTWORM AT THE MEZŐHEGYES STATE STUD-ESTATE CORPORATION, HUNGARY: A CASE STUDY

Zoltán Szabó, Ilona Ondrusz, Péter Miskucza, Tibor Nyári,
State Stud-Estate Corporation, Mezőhegyes, Hungary
Jozsef Kiss, Szent István University, Department of Plant Protection,
H-2100 Gödöllő, Hungary

C. Richard Edwards,
Purdue University, Department of Entomology,
1158 Smith Hall, West Lafayette, IN 47907-1158, USA

Abstract

In 2001, the Western corn rootworm (WCR) population exceeded the economic threshold level in many regions of Hungary. As a result, farmers need WCR management tactics that can be implemented within their typical, multi-crop farming system. This paper looks at the various available management options with special regard to the areawide pest management strategy.

The Mezőhegyes farm encompasses an area of 9,500 ha of which 3,300 ha are in corn (73% seed, 14% silage, 11,7% sweet, and 1,4% commercial corn in 2001). The remaining 6,200 ha are made up of wheat, oats, sunflower, peas, alfalfa, sugar beet, and various vegetables. Of the 9,500 ha, 5,000 ha are under irrigation.

Each year, preparation of a preliminary risk assessment for WCR is required for each field. This is accomplished by estimating the overwintering WCR population in the various fields (pre-crop, pest management applications in pre-crop) and by considering planned insecticide applications (soil application, control of other pests, etc.), while taking into account WCR immigration from the surrounding fields.

For 2001, a preliminary risk assessment was prepared for the total corn growing area of the farm (50% is first-year corn, 45% is second-year corn, and 5% is third-year corn). Thirty-two (32) cornfields were selected for WCR population determinations by surveying with Pherocon AM traps from early July to late August (6 traps on each field, checked weekly).

The WCR infestation level on the farm was highly variable from field to field. The highest number of WCR adults trapped over a seven-day period was 23.1 adults/trap/day in third-year silage corn, 1.7 adults/trap/day in second-year seed corn, 1.1 adults/trap/day first-year seed corn.

Field application tests were made with Cidetrak (Trécé Inc., USA) and Invite (Florida Foods Inc., USA), gustatory stimulant in combination with a small amount of toxicant. Timing of adult control with these compounds and management experiences will be presented in the talk.

UPDATE OF MONITORING DATA OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN SWITZERLAND IN 2001

Bertossa Mario,
Swiss Federal Research Station for Plant Production,
Changins-Cadenazzo, Switzerland

Derron Jacques,
Swiss Federal Research Station for Plant Production,
Changins-Nyon, Switzerland

Colombi Luigi, Brunetti Roberto,
Phytosanitary Service Canton of Ticino,
Bellinzona, Switzerland

Abstract

The detection of four (4) Western corn rootworm adults near Lugano-Agno airport in 2000, led to an increased monitoring in the most important regions of Switzerland where maize is cultivated. A total of 37 strategically important sites were selected. The pheromone traps were set out at the end of June. Adults of WCR were immediately detected in the south border region near Italy, where in four (4) sites a total of 1,715 beetles were trapped over the hole control period of 15 weeks. 40 km northward, three weeks later, other populations of less density for a total of 462 WCR were found in six (6) sites. No WCR was observed northern of the Alps.

The amount of the detections near the border of Italy indicates that a population might be established in that region.

Recapitulation and situation

After the detection of a presence of Western corn rootworm (WCR) in Italy in 1998, authorities decided to place pheromone traps near the main airports in Switzerland in three (3) locations (*figure 1*). In 1999 no infestation could be demonstrated.

After detecting four (4) WCR adults on sex pheromone traps (Csalomon®) near the airport of Lugano-Agno in 2000, measures were taken to eradicate the supposed hazard to maize within the region. It was easy to believe that this infestation occurred at this location, since flights into the Lugano Airport from Serbia took place in the early summer 2000.

The Italian speaking part of Switzerland, the Ticino, where the WCR have been found, accounts a little surface of maize compared to its total surface of 2,812 km^2. However, maize is the most important crop which covers 60% of the arable surface (2,000 ha). The majority of the farmers follow an integrated production management, which forces them to enact a certain crop rotation.

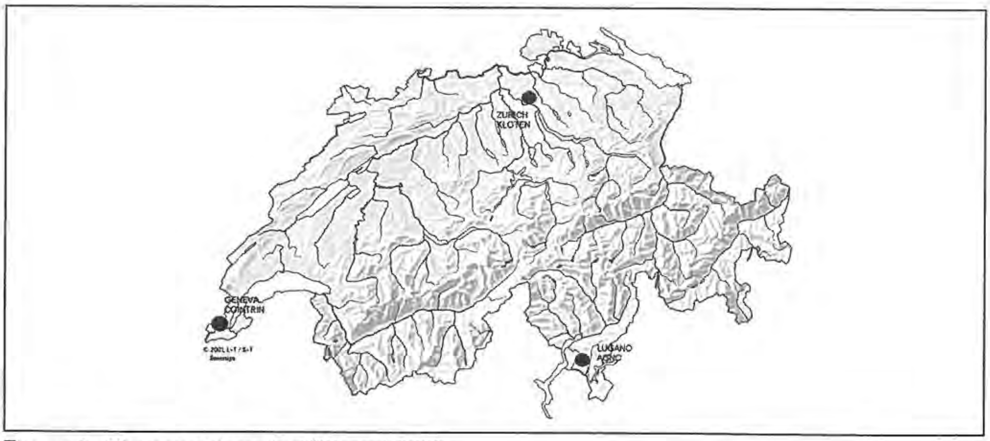

Figure 1 - Trap positions in 1999 and 2000

The measures that have been taken as a result of 2000 WCR catches, among others, was the use of an increased number of traps for 2001.

Methods of monitoring 2001

As the pheromone traps have proved their efficiency in the past and assure comparability to other countries, the same pheromone trap type were used. A total of 74 traps were placed in 37 selected sites of the Swiss territory (*figure 2*): 10 sites in the German speaking part, 13 in the French section, and 14 in the Italian speaking part, including the Lugano Airport site.

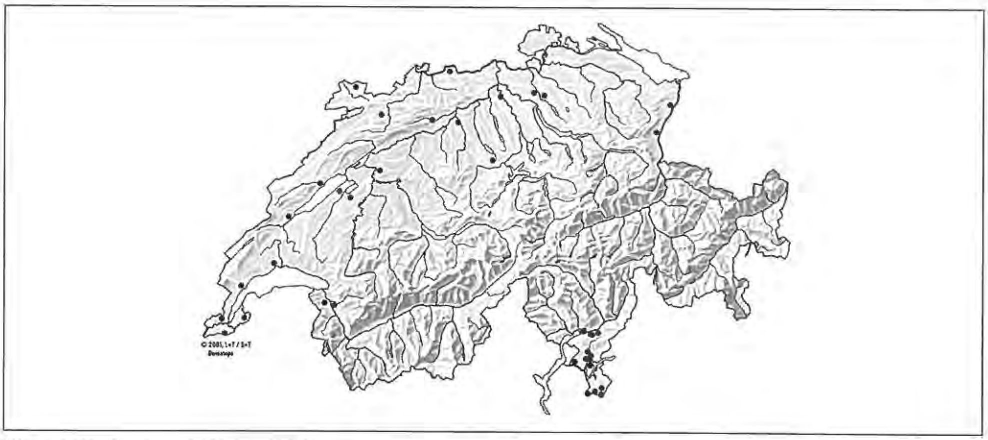

Figure 2 - Trap positions 2001

Installing date of the traps was within the 26th and 27th of June.
The traps were primarily placed along the main transportation routes in the main maize growing regions. All the traps were attached to a wooden stake and placed

approximately at ear height of the maize plant. They were set out in pairs within 20 m distance of each other near the edge of maize fields.

Focus on the Ticino region

The topography shows here three (3) distinguishable maize regions, each separated by mountains and lakes. These include the border region in the south, hereafter called zone A; the Lugano region in the middle, zone B; and the Magadino plain region, zone C. The three zones are located within a distance of 40 km. The traps were monitored once per week from the 5th July until the 10th of October, with the pheromone being changed every 30 days, or 2 times for the sampling period (trap n. 12 and n. 9 with a third exchange).

Results

French and German speaking parts of Switzerland: No beetles were detected in 2001.

Italian speaking part of Switzerland: Ticino

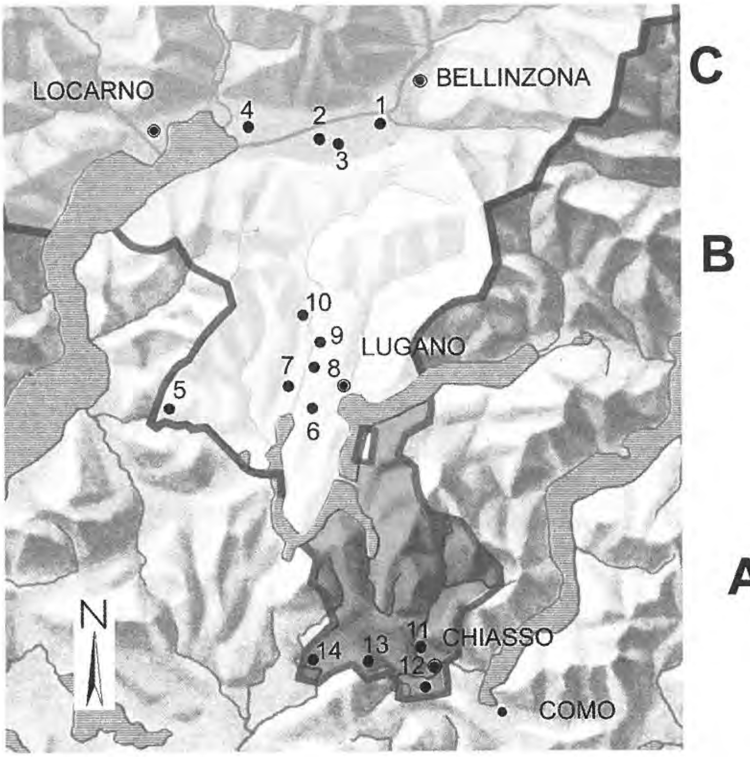

Figure 3 - Trap positions in Ticino 2001

Zone A: In the four trapping sites during the 15-week trapping period, an average of 114 adults per week per site was found.
The first beetles were observed the 5th of July. A total of 1715 insects were trapped with 75 % coming from one location (trap 12). The rest of insects were equally distributed in the remaining locations and a relevant number of adults appeared there only 3 weeks later (*figure 4*).

Zone B: During the same period, an average of 31 adults per week was found. However, catch numbers for July were not significant. The main flight period occurred within the August 2nd and 22nd time period with an average of 108 beetles trapped per week. A total of 462 WCR were captured. The distribution over all sites was almost equal.

Zone C: Only less then 2 WCR beetles per week were trapped over the same monitoring period.

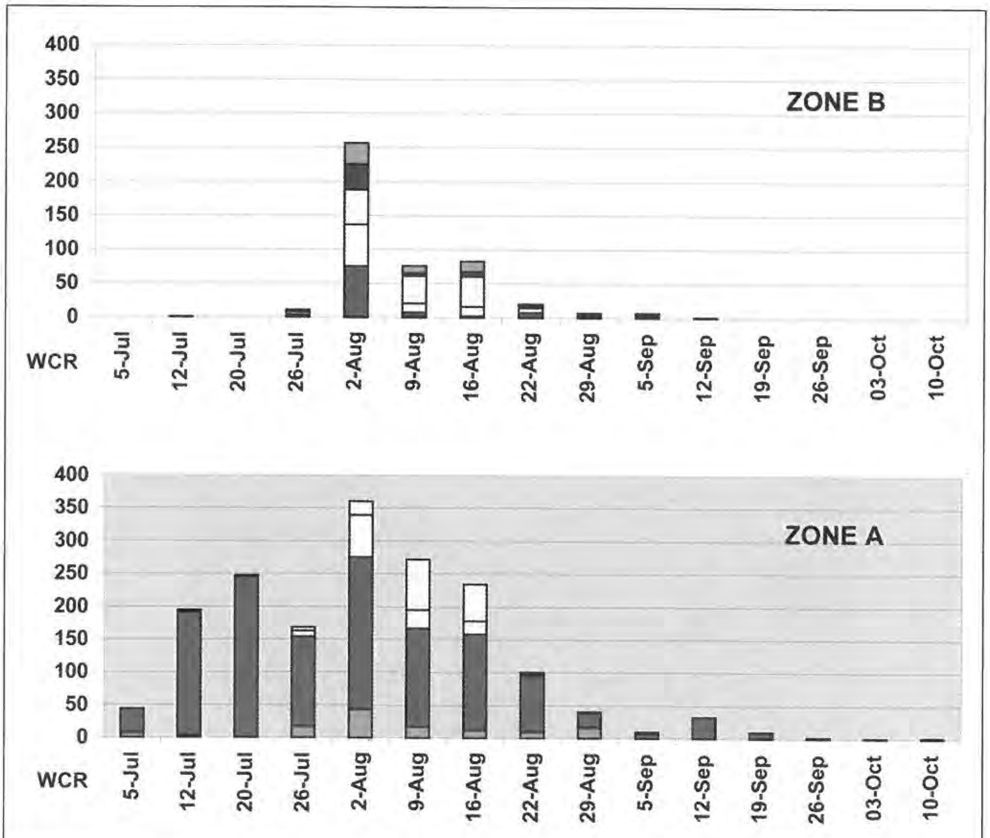

Figure 4 - Amount of catches and time delay in appearance between the zones A and B

It is necessary to say that despite the relative high density (for us) of the population especially in the field of trap site n. 12, no damage to maize (lodging) has been observed.

Conclusions

The number of *Diabrotica* detected in 2001 would indicate that a population of WCR is established near the border zone of Chiasso. In the Lugano zone, where in 2000 the first specimen were found near the airport, it's not clear whether an established, resident population is involved. Time delay in appearance, number of trapped WCR in 2000 and 1999 near Lugano airport, equal distribution between the locations, and crop rotation regulations that have been enacted, indicate a possible migrant population. But there is no certainty that the traps were always located at sites that would prove this.

The four (4) adults captured in 2000 possibly have an origin other than arriving by airplane. They easily could have migrated by themselves from Zone A or from the bordering area of Italy. As the most prolific trap is only few meters away from the border and very near to the main railway station, it is possible that the beetles arrived one or more years before 2000 and became established in the region. In the maize field where beetles were first captured, which has a surface area of one hectare, maize has been grown five times in eight years with 2 interruptions.

Monitoring should not be limited to airports and important transport crossings, it seems more appropriate to install an extended grid over the territory that could be potentially infested.

The ways of migration of the insect are numerous.

For next year, authorities have decided to forbid the growing of maize in the same field in Zones A and B. The transport of silage from Zones A and B to C is also forbidden. An eradication strategy seems inappropriate since *D. virgifera* seems to be well established in Northwestern Italy and would serve as a source for reintroduction of WCR, no matter the steps taken by Switzerland to eradicate it.

Acknowledgments to the colleagues of the Phytosanitary Service, who made the controls, to R. Edwards for his useful advice and J. Derron for his collaboration.

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APPEARANCE OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE ON A BOUNDARY OF UKRAINE

Alla Maksimovna Sadlyak, Adalbert Josifovich Sikura,
Transkarpathian Territorial Department of Plant Quarantine,
Universitetskaya, 21, Uzhgorod, 88017, Ukraine

Petro Ivanovich Yakovets,
Transcarpathian Boundary Inspection of Plant Quarantine, Golovna, 53,
Onokovtsi, Uzhgorodskyy region, 89412, Ukraine

Abstract

The prompt territorial diffusion of *Diabrotica virgifera* in the countries next to Ukraine, in particular with the Zakarpatye region (Hungary, Romania, Slovakia), demands realization of careful monitoring in boundary territories.

For this purpose in the middle of July 2001 lengthways near border with Hungary, Romania and Slovakia in 22 populated localities, and also in villages of the central part of region on sowings of maize pheromone traps and color (yellow) traps were exposed.

The traps were exposed on edges of a maize field on approximately through every 1,000 meters, alternating yellow plastic cylindrical traps, yellow cardboard rectangular traps and pheromone traps. The pheromones of the Hungarian and Moldavian production were used.

At analysis of August 15 of a biological stuff in traps with the Hungarian and Moldavian pheromone first four specimens of *Diabrotica virgifera* were revealed in three frontier populated localities.

Thus, with the help of pheromone traps exposed on sowings of maize along border, it was possible to reveal the appearance of individual specimens on the terrain of the Zakarpatye region, that testifies that the depredator has closely come nearer to Ukraine.

Key words: *Diabrotica virgifera*, monitoring, pheromone traps, Ukraine.

PRESENT SITUATION

Our examinations of *Diabrotica virgifera virgifera* have just begun. From the moment of *Diabrotica virgifera virgifera* appearance in Yugoslavia we have been constantly tracking the reports on its diffusion and harmfulness.

The augmentation of its geographic range in Europe goes by prompt rates and now *Diabrotica virgifera virgifera* is registered in the countries, which adjoins with Ukraine, in particular with the Transcarpathian region - in Hungary, Romania and Slovakia.

Figure 1 – Map of Ukraine and location of Transcarpathian region

Transcarpathian Boundary Inspection of Plant Quarantine, which is heading by Petro Ivanovich Yakovets, carries out the careful examination of all loads and transport units, with which the delivery of this pest is possible. Besides that, the inspection of 3-kilometer zone in items of crossing of frontier is carrying out; the monitoring of *Diabrotica virgifera virgifera* with the help of pheromone traps is annually being carried out.

In 2001 the employees of Transcarpathian Boundary Inspection of Plant Quarantine and Transcarpathian Territorial Department of Plant Quarantine of the Ukrainian Academy of Agrarian Sciences along frontier with Hungary, Romania and Slovakia exposed traps in the 22 populated localities on sowings of maize.

Fields of maize had distances from frontier of 50-100 m. The traps were exposed on edges of field along 1000 m, alternating the yellow sticky plastic cylindrical form, yellow sticky cardboard rectangular and white sticky plastic cylindrical with pheromone on distance 1000 m one from another. In traps we used pheromone of the Hungarian and Moldavian production.

At analysis of a biological stuff in traps with pheromone on August 15 the first 4 specimens of the beetles *Diabrotica virgifera virgifera* in three boundary items were revealed. During the research period from August 15 till September 15 in all were revealed 15 specimens of the beetle *Diabrotica virgifera virgifera* in 7 populated

Figure 2 – Map of places of detection of *Diabrotica virgifera virgifera*

localities along frontier with Hungary and Romania (*figure 2*). Pheromone traps also were exposed on sowings of maize in the central areas of region, Mukachivsky and Irshavsky, and also on 14 items of crossing of frontier. In no one case *Diabrotica virgifera virgifera* was not revealed. The beetle *Diabrotica virgifera virgifera* was not revealed also in color sticky traps. These data allow us to make the conclusion that we succeeded in revealing invasion of western corn beetle on our territory from the very beginning of this process.

WESTERN CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) IN UKRAINE: REALITY AND OUTLOOK

Omelyuta Victor, Filatova Natalia,
Institute of Plant Protection of UAAS
Vasyl'kivs'ka str., 33, 03022, Kyiv, Ukraine
Phone: (044) 263-11-24, Fax: (044) 263-21-85,
e-mail: nikol@mail.kar.net

The analysis of date on geographic - coordinate, zoogeographic (*table 1*) and agro-climatic characteristics of Ukraine (*table 2*) and the range of the western corn root-worm (WCR), which is a native species in America and an invading species in Europe gives the grounds to suppose that WCR is able to inhabit all the territory of Ukraine.

Table 1 - The world range of *Diabrotica virgifera virgifera* LeConte

The range	**Geographic coordinate**	**Zoogeographic provinces and climatic belts**
America - the primary range	30° - 50° north latitude	Eastern-American and Western-American subprovinces of Holoarctic, the subtropical and moderate climatic belts
Europe - the secondary range	43° - 48° north latitude	Circumboreal and Mediterranean subprovinces of Holoarctic, the belt of transitory climate from oceanic to continental (Mediterranean) and the belt of moderate climate
Ukraine - the probable range	45° - 50° north latitude	West of Central-Asiatic subprovince of Holoarctic, the belt of moderate climate
Geographic disposition of Ukraine	44° - 52° north latitude	

Table 2 - The main meteorological indices of Ukraine and countries within the range of the Western corn beetle

Meteorological indices	Ukraine	USA, the corn belt	The countries of Danube basin (Hungary, Croatia, Serbia, Romaine, Bulgaria)
Average temperature of air, grades C:			
January (I)	-2.0	-5.2	-1.6
July (VII)	23.0	23.9	22.0
Season of vegetation (IV-IX)	17.7	18.0	17.7
Average over a year	10.3	9.9	10.7
Sum of precipitation, mm:			
April - June (IV-VI)	122	267	205
July - September (VII-IX)	130	237	163
Season of vegetation (IV-IX)	252	504	368
Average over a year	450	814	644

However, supplying of separate parts of Ukraine with worm and amount of precipitation is very different that stipulates inequivalent conditions for growing of the corn and, consequently, for the run by of normal life cycle of the pest. In the plain part of Ukraine, it is elicited four agroclimatic zones, which are differed by the hydrotermal indices of the periods of vegetation and intensity of growing of the corn in the zones (*table 3*).

In recent years, in Ukraine the corn was sown annually on the area 5.5 million hectares, including 1.2 million hectares in average for grain yield. In so doing, the corn is not growing as monoculture, whereas participation of it in the arable area fluctuates in the range 10-40%. Seldom, in the conditions of irrigation, the participation of the corn reaches 80% and on the same field the corn is rowing 3-4 seasons in consequence. Hence, taking into account food resources for the pest and its demand to climatic factors, one might suppose that it is able to occupy the most part of Ukraine in the agroclimatic zones III-IV, in the subzones Ia and Ib, and partially in the zone II. Therefore, in Ukraine, it is possible to state the following regions with a standpoint of damage by the pest: 1. mass occurrence and high damage, 2. common occurrence and sporadic damage, 3. possible occurrence (*figure 1*).

1. The region of mass occurrence and high damage that spreads on the most part of the area of Ukraine - the Steppe zone, southern part of the Forest-Steppe zone, lowland part of Zakarpattia (Transcarpathian) and Chernivtsi Regions

Table 3 - Agroclimatic zones in Ukraine and intensity of growing of the corn in them

Names of the zones	Indices of hydrotermal coefficient	Indices of sums of air temperatures above 10 °C	Participation of the corn in arable area %	The number of corn fields in consequence of seasons
I - Wet, moderately warm (Transcarpatian region)	2 - 1.3	2400 - 3100	10	1
Ia - Wet, warm with mild winter (lowland part of the region),	1.8 - 1.3	2600 - 3100	40	3
Ib - Wet, warm with moderately cold winter (the low mountain belt);	1.6 - 1.3	2600 - 2900	30	2 - 3
II-Insufficiently wet, warm (Forest - Steppe);	1.3 - 1.0	2500 - 2900	20	1 - 2
III - Droughty, very warm (Steppe);	1.0 - 0.7	2900 - 3000	20 - 30	2 - 3
IV - Very droughty, moderately hot (Dry Steppe)	0.7 - 0.5	3300 - 3400	20 - 40	3 - 4

where corn fields occupy more than 20% of arable area with repeated usage in a shift of crops system.

2. The region of common occurrence and sporadic damage on repeated fields of the corn in the central and western parts of the Forest-Steppe zone where the area occupied by the corn composes 10-20% of the arable area.
3. The region of possible occurrence which spreads over the north part of the Forest-Steppe and Forest zones, where the corn occupies less than 10% of the arable area, and repeated usage of the corn in a shift of crop system is not practiced.

The first beetles (three imagoes on three fields) were found out by the authors by means of pheromone traps in August 2001 in Vynogradovo District of Transcarpathian Region. Moreover, in the same time in a field, it was recorded two clearings within an area up to ten square meters with symptoms characteristic for damage by larvae of WCR, namely lodging.

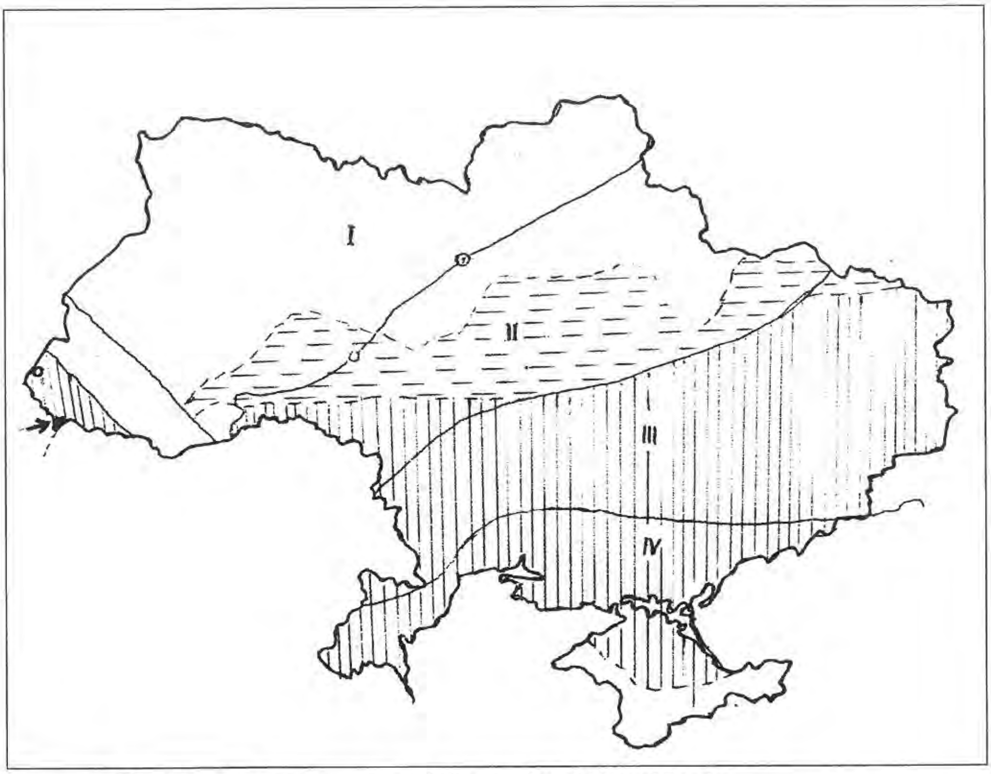

Figure 1 - The regions of occurrence and damage by WCR in Ukraine

The agroclimatic zones:

I. Forest
II. Forest – steppe
III. Steppe
IV. Dry steppe

The regions of occurrence and damage by WCR:

1- Mass occurrence and high damage by WCR

2 - Common occurence and sporadic damage

3 - Possible occurrence

4 - It is found out appearing of WCR in 2001

This fact allows to suppose that primary penetration of the beetles has occurred in previous year - 2000, whereas the presence of WCR has been recorded in the second year as it is known in Yugoslavia.

It is quite probably that in 2002-2005 WCR will spread actively over the lowland part of Transcarpathian Region bearing damage to the corn, which is used in repeated fields in a shift of crop system. The mountain part of Transcarpathian Region where the corn is nearly out of growing will serve as a reliable obstacle for spreading of WCR. The practice of spreading of other quarantine pest species (Hyphantria cunea Drury, Grapholitha molesta Busck.) confirms this supposition.

An appearing of WCR in the rest of Ukrainian territory should be forecasted for the Districts of Odesa Region close to the Donau River moving from Romania when this species would seize the Donau Lowland. It is possible an appearing of WCR in other places in Ukraine in a result of passive transferring of it by aviation and military techniques arrived from dislocation of Ukrainian peace-keeping forces in Yugoslavia. A possibility of such a transportation is confirmed by arising of infestation spots of WCR in Italy and Switzerland.

INFLUENCE OF FERTILIZATION AND HYBRIDS ON WESTERN CORN ROOTWORM DAMAGE ON CONTINUOUS CORN

Tatjana B. Keresi, Pero M. Strbac, Dragana Latkovic,
Faculty of Agriculture, Plant Protection Institute,
Trg D. Obradovica 8, 21000 Novi Sad, Yugoslavia

Radosav R. Sekulic,
Research Institute of Field and Vegetable Crops,
M. Gorkog 30, 21000 Novi Sad, Yugoslavia

Franja Baca,
Maize Research Institute, Slobodana Bajica 1,
11081 Zemun Polje, Belgrade, Yugoslavia

Abstract

Since it has been realized that the chemical control of the Western corn rootworm (*Diabrotica virgifera virgifera* LeConte) is complicated and uncertain, it became important to determine which agrotechnical measures permit continued growing of corn in monoculture.

With this objective in mind, we have studied the effect of fertilization systems and corn hybrids on the harmfulness of the WCR. Experiments were conducted in 2000-2001, in the vicinity of Novi Sad, in a trial with corn monoculture established in 1965. The trial includes three fertilization variants (1. NPK, 2. crop residues + NPK, 3. manure + NPK) and the non-fertilized control and several maize hybrids of different maturity groups (FAO 300, 500, 600, 700). WCR damage was assessed via plant lodging, root damage and yield reduction.

In dry 2000, plant lodging were significantly higher in variants 2 and 3 (52.4 and 61.1%) than in the control (37.6%), while the situation was reverse in wet 2001. In the first year, plant lodging was most intensive in FAO groups 300 and 600 and least intensive in FAO 700. In the second year, the most intensive lodging occurred in FAO group 600 and the least intensive in FAO 300.

In both experiment years, root damage (on the scale 1-6) was largest in the control (4.57 and 4.91) and lowest in variant 2 (3.35 and 4.15). The differences were significant in the first year and non-significant in the second. In 2000, root damage ranged in the hybrids from 4.07 to 4.31, with non-significant differences, while in 2001, FAO groups 300 and 500 experienced significant damages (4.74 and 4.63, respectively) in relation to the two hybrids in FAO group 600 (4.16).

In 2000, the highest yield (2.74 t/ha) was obtained in variant 2, which is significantly higher than in the control and variant 3 (1.18 and 1.25 t/ha). Yields of FAO groups 700 (2.45 t/ha) and 600 (1.96-2.08 t/ha) were significantly higher than those of groups 300 and 500 (1.00 and 1.49 t/ha). The yield in 2001 was the highest in variant 2 and 3 (6.3 and 5.9 t/ha, respectively) and the lowest in control (4.45 t/ha), while differences among hybrids were not big.

Because of the extremely opposite weather conditions (particularly rainfalls) in 2000-2001, relevant conclusions may be expected on the basis of long-term data.

Key words: *Diabrotica virgifera virgifera* LeConte, maize, plant lodging, root damage, yield reduction.

Introduction

Before 1992, when first damage by the Western corn rootworm (*Diabrotica virgifera virgifera* LeConte) was recorded (Baca, 1993), about 40% of corn in Yugoslavia was grown in monoculture (Gotlin, 1987). After that, acreage under continuous maize substantially decreased in the major growing regions.

Since it has been realized that the chemical control of Western corn rootworm (WCR) is complicated, expensive and uncertain, it became important to determine which agrotechnical measures permit continued growing of corn in monoculture, or at least in short-term monoculture, with minimum risk of economic damage from the pest. With this objective in mind, we have studied the effect of fertilization systems and corn hybrids on the harmfulness of the WCR larvae in continuous corn.

Methods

Experiments were conducted in 2000-2001, in the vicinity of Novi Sad, in a stationary trial with corn monoculture established in 1965. The trial includes three fertilization variants (1. NPK; 2. crop residues + NPK; 3. manure + NPK) and the non-fertilized control. Mineral nutrients are introduced using a 15:15:15 formulation at a rate of 60 kg/ha pure N + 60 kg/ha pure P + 60 kg/ha pure K in the autumn and 60 kg/ha N in the spring. Barnyard manure is incorporated every two years at 25 t/ha.

Corn hybrids of different maturity groups (FAO 300, 500, 600, 700) are tested in the trial. Six of them were grown in 2000 and four in 2001, at experimental units of 4 and 6 rows (respectively) long 14.3 m, in 4 replications at randomized block design.

Ever since 1996, when first damages were recorded in the vicinity of Novi Sad (Keresi, 1996) (Keresi *et al.*, 1996), lodging percentage in this trial has been on the increase, hindering the harvest and causing yield losses. In light of this, we carried out a preliminary study of WCR harmfulness in 2000, while in 2001 the study was continued and chemical protection was attempted in two thirds of the trial by applying Furadan 350 F (a.i. carbofuran) at 4 l/ha in bands 28 days after sowing.

Weather conditions during growing season in the two years of study differed greatly, especially in the terms of precipitation, with regard to which the two years were diametrically opposite. The 2000 growing season was characterized by catastrophic drought (60% less precipitation than the long-term average) and very high air temperatures (about 2°C above average), while the 2001 growing season had twice more precipitation than usually (particularly in April, June and September) and average air temperatures.

Western corn rootworm harmfulness was assessed at two unprotected rows of each hybrid and expressed via the percentage of plant lodging (a), extent of root damage (b) and yield reduction (c).

RESULTS AND DISCUSSION

a) In extremely dry and warm 2000, maize emergence was not uniform, the plants were considerably shorter than average and many of them were barren. The WCR appeared significantly earlier than usually, so the damage caused by the larvae was recorded and assessed earlier too (on June 26). The percentages of lodging (*table 1*) were significantly higher in treatments with barnyard manure + NPK (61.1%) and ploughing down crop residues + NPK (52.4%) than in the control (37.6%). Plant lodging was most intensive in FAO groups 300 and 600 (53.8 and 51.6%, respectively), and least intensive in FAO 700 (38.2%).

In extremely wet and average warm 2001, maize emergence was uniform, the plants were taller and stronger than usually and very well stand attack of WCR larvae. Generally speaking, there was no lodging in the Vojvodina province at large. However, in our trial with 35-year continuous corn, we recorded a similar average percentage of lodged plants as in previous year (48%), but lodging was the greatest in the non-fertilized control (61.8%), whereas in the different fertilization treatments it was about the same (41.5-45.8%). The most intensive lodging occurred in FAO group 600 (about 52.0%) and the least intensive in FAO 300 (37.8%).

The obtained results from first year are in accordance with the results obtained by Spike and Tollefson (1988) and Baca *et al.* (1998), which indicated that increase of mineral and organic fertilizer rates led to the increase of plant lodging. In second year our results are the opposite from the results of mentioned authors. Regarding plant lodging in different hybrids, our results in first year are partly and in second year completely similar to those of Baca *et al.* (1998), who found plant lodging in FAO group 400 about 24.2% and in FAO group 500-600 about 46.0%.

In evaluating the efficacy of chemical protection in two thirds of the trial, only the lodging percentage has been established, while the yields are yet to be calculated. The results show that the effects of the treatment are very significant, since the lodging percentage in the protected part of trial was on average only 5.7%, or 88% less than in the unprotected, where the lodging was 48%. Differences among the various fertilization systems and hybrids were non-significant (*figure 1*).

Table 1 - Damage by WCR larvae on continuous corn depending on fertilization and hybrids (Novi Sad, 2000-2001)

Fertilization system	Lodged plants (in %)			Root damage (1-6 scale)			Yield (t/ha)		
	2000	**2001**	**Ø**	**2000**	**2001**	**Ø**	**2000**	**2001**	**Ø**
NPK	40.33	41.46	40.90	4.37*	4.39	4.38	1.92	5.48	3.70
Crop residues + NPK	52.43*	45.81	49.12	3.35	4.15	3.75	2.74**	6.30	4.52
Manure + NPK	61.11**	41.82	51.46	4.47*	4.25	4.36	1.25	5.90	3.58
Control (no fertilizing)	37.62	61.77	49.70	4.57*	4.91	4.74	1.18	4.45	2.82
Average	47.87	47.72	47.80	4.19	4.42	4.30	1.77	5.53	3.65
$LSD_{0.05}$	14.26	34.89		0.88	0.97		1.00		
$LSD_{0.01}$	20.48	50.13		1.26	1.39		1.44		
Hybrids									
NS – 300	53.79*	37.76	45.78	4.07	4.74**	4.40	1.00	5.42	3.21
NS – 542	-	49.05	-	-	4.63**	-	1.49	5.92	3.70
NS – 606	-	51.43	-	-	4.16	-	2.08**	5.12	3.60
NS – 640	51.62	52.61	52.12	4.18	4.17	4.18	1.96**	5.30	3.63
Balkan	38.20	-	-	4.31	-	-	2.45**	-	-
Average	47.87	47.71	47.79	4.19	4.42	4.30	1.80	5.44	3.62
$LSD_{0.05}$	14.63	22.33		0.77	0.34		0.53		
$LSD_{0.01}$	19.83	29.92		1.04	0.46		0.70		

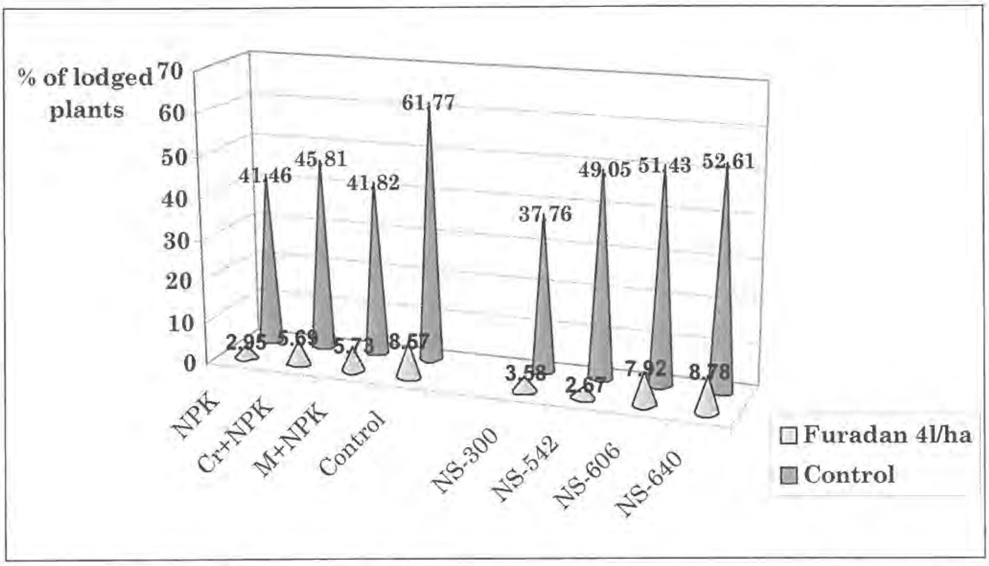

Figure 1 - Efficacy of chemical protection against WCR larvae via percentage of plant lodging at different fertilization systems and hybrids (Novi Sad, 2001)

b) In both experiment years, the level of root damage (on the scale 1-6) was largest in the control (4.57 and 4.91) and lowest in the treatment with ploughing down crop residues + NPK (3.35 and 4.15). The differences were significant in the first year and non-significant in the second.

In year 2000, the level of root damage ranged in the hybrids from 4.07 to 4.31, with non-significant differences. In 2001, FAO groups 300 and 500 were damaged significantly more (4.74 and 4.63, respectively) than the two hybrids from FAO group 600 (4.16-4.17).

c) Due to extremely unfavorable weather conditions in 2000, the average yield of maize was disastrously low. The highest maize yield of 2.74 t/ha was obtained in variant 2, with ploughing down crop residues + NPK, which is significantly higher than in the control and variant manure + NPK (1.18 and 1.25 t/ha, respectively). Yields of FAO groups 700 (2.45 t/ha) and 600 (1.96-2.08 t/ha) were significantly higher than those of groups 300 and 500 (1.00 and 1.49 t/ha).

The yield in 2001 (assessed on October 24) was much higher, due to favorable weather conditions, which enabled rapid and full recovering of damaged corn roots. Obtained yield was the highest in treatments with crop residues + NPK (6.3 t/ha) and manure + NPK (5.9 t/ha) and the lowest in non-fertilized control (4.45 t/ha), while among hybrids differences were inconsiderable (5.12-5.92 t/ha).

The average results showed that in both years percentage of lodged plants was identically high (around 48%), as well as level of root injury (4.3). However, while the average yield in the first year was disastrously low (1.77 t/ha), obtained yield in the second year was entirely satisfactory (5.53 t/ha).

According to the average results, the treatment with ploughing down crop residues + NPK, in spite of high plant lodging caused by WCR larvae, had least root-

injury and the highest yield. Explanation for this giving us Baca and Veskovic (1999), which found that organic matter balance, as well as better soil aeration increased by crop residue ploughing down. Under such conditions, egg and larvae mortality is probably greater, and/or soil conditions are less favorable for oviposition.
Differences among hybrids in average were small, regarding all three parameters.

CONCLUSIONS

The present study has shown that Western corn rootworm populations are strongly influenced by cultural practice methods which can partially eliminate the risks associated with maize production by repeated sowing and even monoculture. In both years the treatment with ploughing down crop residues + NPK, in spite of high plant lodging caused by WCR larvae, had least root-injury and the highest yield.

Since the studied parameters were intensively influenced by climatic factors, particularly the rainfall, which went into the opposite extremes in the two experiment years, relevant conclusions may be expected after 2-3 more years and than can be recommended to the corn growers *(table 1)*.

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METHODS FOR EVALUATING TOLERANCE OF CORN TO CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) LARVAL INJURY

Jon J. Tollefson,
Iowa State University, Department of Entomology,
Ames, Iowa, USA 50011-3140

Abstract

The mechanisms of host resistance to insects have been classified as non-preference, antibiosis, and tolerance. Non-preference describes plants that have lower populations of a pest because the pest prefers other plants. Antibiosis refers to the harmful effects of a host on a pest, resulting in suppression of pest densities. Tolerance is the ability of a host to produce higher yields while sustaining similar densities of a pest. Because resistance due to non-preference and antibiosis results in the presence of fewer pests, pest density is commonly used to evaluate these forms of resistance. With tolerance, however, pest densities do not differ measurably and comparisons must be made using differences in host growth and yield.

In the USA, an extensive search has been made for corn germplasm that is resistant to western corn rootworm (*Diabrotica virgifera virgifera* LeConte (WCR)) larval feeding. The WCR larvae are subterranean, making estimates of pest density difficult to obtain. Consequently, plant indices of larval density have been used to screen for non-preference and antibiosis. The most common index has been the root-injury rating. Plant characteristics that have been used to compare corn germplasm for tolerance to CRW larval feeding include root size, root regrowth, plant lodging, and the amount of vertical force required to pull plants from the soil.

Attempts have been made to contrast root size using volumetric displacement of water and visual ratings. Water displacement provides a quantitative measurement of size, but the measurement error has been too large for it to be useful. Although visual ratings are subjective, statistical differences in the ratings have been found and the ratings have been useful in ranking tolerance.

Root regrowth is the measure of a plant's ability to recover from injury caused by WCR larvae. Quantitative measurements of regrowth have been made by weighing root masses during and after larval feeding and calculating the difference. Visual ratings of regrowth have also been used to evaluate for tolerance by using a 1–6 scale of fibrous root growth. While root weights are more precise than the subjective visual ratings, they are more difficult to obtain because the plots must be evaluated twice and the root masses dissected, dried, and weighed.

Plant lodging and the force required to pull plants from the soil are general estimates of root mass and health. Because lodging results only under certain environmental conditions, it has not been a reliable indicator of corn tolerance to WCR feeding. The force required to pull plants from the soil is less subject to environmental variability and has been more useful. The measurements of pulling force are quite variable, however, and require sample sizes that are larger than visual estimates of root size and regrowth.

Key words: western corn rootworm, *Diabrotica virgifera virgifera*, tolerance, root size, root regrowth.

INTRODUCTION

Soon after the Western corn rootworm (*Diabrotica virgifera virgifera* LeConte) was discovered damaging U.S. corn, the search for varieties that were resistant to the pest began. In the 1920s, a difference in reaction to larval feeding was noticed (Bigger *et al.*, 1938). It was subsequently determined that certain lines had superior resistance to rootworms (Bigger *et al.*, 1941). As the western corn rootworm became resistant to insecticides and rapidly spread eastward across the U.S. Corn Belt during the 1960s, a systematic search for corn lines that were resistant to the pest was conducted. Plant materials that were evaluated included: inbred lines (Eiben and Peters 1965, Fitzgerald and Ortman 1965), plant introductions (Wilson and Peters 1973), various exotic corns (Melhus *et al.*, 1954, Fitzgerald and Ortman 1964), synthetic varieties (Owens *et al.*, 1974), and commercial hybrids (Rogers *et al.*, 1975).

The mechanisms of host resistance to insects have been classified as non-preference, antibiosis, and tolerance (Painter 1951). Non-preference describes plants that have lower populations of a pest because the pest prefers other plants. Antibiosis refers to the harmful effects of a host on a pest, resulting in suppression of pest densities. Because resistance due to non-preference and antibiosis results in the presence of fewer pests, pest density is commonly used to evaluate these forms of resistance. The western corn rootworm larvae are subterranean, however, making density estimates difficult to obtain. Consequently plant indices of larval density have been used to screen for non-preference and antibiosis. The most common index has been the root-injury rating. Tolerance is the ability of a host to produce higher yields while sustaining similar densities of a pest. With tolerance, pest densities do not differ measurably and comparisons must be made using differences in host-plant growth and yield. Plant characteristics that have been used to compare corn germplasm for tolerance to corn rootworm larval feeding include: root size, root regrowth, plant lodging, and the amount of vertical force required to pull plants from the soil.

ROOT-INJURY RATING

Classifying the amount of root injury corn has suffered is a direct estimate of the number of corn rootworm larvae that fed on the root system (antibiosis or non-preference). The most common scale that had been used was a 1 to 6 scale where 1 = little or no damage and 6 = severe larval feeding (three nodes of roots destroyed) (Hills and Peters 1971). Recently a node-injury scale has been proposed that ranges from 0 (no feeding) to 3 (three nodes of roots destroyed) (Oleson and Tollefson, unpublished). The node-injury scale is easier to learn, linear, and more precise.The heritabilities of root-injury ratings have been low; which means that they have little use in selecting for resistance (Owens *et al.* 1974). Until recently, little resistance against corn rootworms due to antibiosis has been found.

Lodging

Researchers have used counts of the number of plants that are lodged more than 30° or 45° from the vertical to classify rootworm resistance. Lodging resistance can be a function of reduced larval populations (antibiosis or non-preference) or more vigorous root systems (tolerance); the two cannot be separated. Farmers desire lodging resistance because standing corn is easier to harvest and suffers less mechanical and physiological yield loss. Unfortunately lodging is not consistent. Not only is it a function of corn rootworm resistance, but also it is dependent on storms (wind and rain) at or shortly after maximum root injury. Because of the strong environmental influence on and unpredictability of lodging, lodging alone has not been used to select for resistance. However, strong correlation of lodging with root size and secondary root development have been shown and these indices have been used together to select for resistance (Owens *et al.*, 1974).

Plant-pull resistance

Another estimate of root injury (antibiosis or non-preference) and root vigor (tolerance) is the amount of force required to pull the plant from the soil. The technique uses a scale and lever to lift plants vertically until they break loose from the soil. Like lodging, the technique measures overall root health it does not depend on environmental conditions such as windstorms. The amount of force required to pull plants from the soil differs depending on soil type, soil moisture, and plant size. The relative resistance of corn lines will remain the same, however, if all lines are included at each location (replicated across soil types), and if a whole location or replicates within a location, are evaluated at the same time (similar soil moisture and plant size).

Root volume

In attempts to quantify corn rootworm resistance directly, root volume has been measured. The most common method is to immerse the root system in water, then measure the amount of liquid that is displaced. The technique is not suitable for evaluating corn lines because of phenotypic variation in root growth and the lack of uniformity in digging the plants from the soil.

Root size

Size ratings are relative ratings of all the roots at a location. When size is rated, the evaluator attempts to not be influenced by the amount of larval feeding, but to consider only the relative size of a root system (tolerance and not antibiosis or nonpreference).The original root-size rating was described by Eiben (1967) and is a 1-6 scale similar to the Iowa State University root-injury scale. Eiben rated size by classifying the smallest root in an experiment as a 1 and the largest root system as a 6. Then representatives of the four classifications (2 – 5) that fall between these two extremes are selected. The six representative roots from that location are laid side-by-side and the remaining roots are compared to them to assign each root a size rating. In the root-injury rating scale, 1 is the best rating (least injured), but in Eiben's root-size scale 1 is the worst (least vigorous root). Because of this, some researchers have reversed the root-size scale and assigned the largest root a 1 and the smallest a 6 so that the two scales are parallel.

Secondary root development

The secondary root development scale described by Eiben (1967) is very similar to the root-size scale. It also is a relative rating of tolerance and not antibiosis or nonpreference, that varies from 1 to 6. The root within an experimental location that has the least secondary root development (branching of nodal roots) is assigned a rating of 1 and the root system with the most branching is assigned a rating of 6. Representatives of the intermediate ratings are chosen and the branching of the rest of the roots is classified according to the six representative roots. Some researchers reverse this rating scale so that 1 is the best rating (greatest secondary root growth) and 6 is the worst (least regrowth) to bring it into agreement with the root-injury and reversed root-size scales.

Attempts have been made to measure secondary root development using a quantitative scale. The most common approach has been to cut the nodal roots from the stalk, dry them, and weigh the dried roots. To obtain a measure of the amount of regrowth a corn line is capable of the weights must be taken twice. The nodal roots are weighed the first timed at approximately pollen shed. At this time the roots have reached their maximum volume and the most of the larval feeding has been completed. Approximately two weeks later, the roots are dried and weighed again after the injured nodal roots have had a chance to branch and produce secondary roots. The potential to produce secondary roots is calculated by subtracting the first root weight from the second. The secondary root-development measurement is correlated with the visual secondary root development ratings and does not substantially improve the selection for tolerance. Because weighing roots to calculate secondary root development requires that plants be dug twice, it is less efficient then the visual rating and is not used as often in resistance screening.

YIELD

The corn characteristic of ultimate interest to the farmer is yield, which also has been used to measure tolerance of varieties to corn rootworm larval feeding. Tolerance to yield loss was used by Rodgers *et al.* (1975) to compare commercial corn varieties. Yield tolerance is measured by planting the experimental corn lines in paired-row plots and treating one row with a soil insecticide, but not treating the other row. Yields are then compared in the two rows, one with a rootworm infestation and the other where the roots were protected from significant larval feeding; the smaller the difference between the yields in the treated and untreated rows, the greater the tolerance of rootworm injury.

CONCLUSIONS

Ratings of root injury by corn rootworms measure the level of toxicity of corn (antibiosis) or the ability of the host to avoid injury (non-preference). High levels of these forms of resistance to rootworms have not been found in corn.

The resistance that has been found has been due to tolerance, larger root systems, and greater secondary root development (Owens *et al.*, 1974, Rogers *et al.*, 1975). The most efficient measurements of these characteristics are visual ratings of root size and secondary root development (Owens *et al.*, 1974). Plant-pull resistance is more variable than the size and regrowth ratings and takes twice the number of replications to produce equal precision. Root volume measurements have too much error to be of use. Lodging resistance and yield tolerance are the characteristics of most interest to farmers. While they can be used to demonstrate corn rootworm tolerance, they are influenced by environmental conditions too much to be of use in a corn breeding program. The best improvements in corn tolerance to corn rootworm larval injury can be made visual ratings of root size and secondary root development.

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OCCURRENCE OF WESTERN CORN ROOTWORM ADULTS ON DIFFERENT TRAPS AND HYBRIDS

Pero Strbac, Tatjana Keresi,
Faculty of Agriculture, Novi Sad, Yugoslavia

Vera Pecarski,
Agricultural Productive Institute "Sokolac", Novi Becej, Yu

Abstract

Since its first occurrence, until now, Western corn rootworm (*Diabrotica virgifera virgifera* LeConte) has spread quickly, first in the main maize growing area of Yugoslavia, and then in other parts of country. This pest has tendency of further spreading, occupying first of all neighboring countries, followed by some European ones, whose ecological conditions favor to its reproduction and development.

With the aim to prevent economical losses, different sampling techniques and programs have been developed for all stages of corn rootworm. Imagoes sampling, i.e. plant inspection for damage prediction represents a standard method.

Trap use has advantages, for population assumption is done in longer time period, whereas during plant inspection it is performed in definite terms only, with the risk of being subjective. By trap use need for well-educated specialists for inspection of whole plants is eliminated. However, opinions on efficiency and credibility of different trap use are divided.

From the given reasons, during 2001, in the region of mid Banat (Yu), in hybrid fields of different ripening groups (NS 680, NS 644, NS 599 and NS 420), USA yellow sticky traps ("Pherocon AM" and "Multigard") and Hungarian pheromone trap were tested. Traps were set up on July 18, and the last reading was on September 5. Trap assessment and visual assessment of plants was performed weekly.

The greatest occurrence of WCR adults was during August in all hybrids, after which population kept reducing. The greatest total imagoes number was caught in the field of the hybrid NS 599. Hybrids NS 680 and NS 640 had similar imagoes number, whereas the smallest population was in the field of the hybrid NS 420. Among different traps have been determined significant differences in catch. The highest catch was on pheromone, similarly lower on yellow "Pherocon AM" and "Multigard" trap, with somewhat lower catch by visual assessment. These differences among applied traps and methods are evident in all studied hybrids.

The results should be considered preliminary ones. In the following year studies will continue and probably be completed by data on larvae density and harmfulness, as well as by the number of adults in emergence cages.

STUDY OF THE SEQUENTIAL SAMPLING FOR DETERMINING THE ADULT POPULATION OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE AT FIELD LEVEL IN HUNGARY

Krisztina Mucsi,
Csongrád County Plant Health and Soil Protection Service,
6800 Hódmezővásárhely - P.O. Box 99 - Hungary
e-mail: kmucsi-csnta@fki.gov.hu

Katalin Pálfi, Cecilia Vaszi-Kovács, Ede Petró,
Centre Service for Plant Health and Soil Conservation, 1519
Budapest, P.O. Box 340 - Hungary

Abstract

In 2000, the western corn rootworm (WCR) was present all over Hungary, except for three counties (Győr-Moson-Sopron, Zala and Szabolcs-Szatmár-Bereg). This year, the traps have caught individuals in all regions of the country.

Supported by the forecasting specialists of the Hungarian Plant Protection Organisation, a sequential sampling was adapted according to the American literature.

Investigations were made between 23 July and 7 September 2001.

The survey procedure is simple and rapid, easy to learn for everybody and no special equipments and tools are required.

During the sampling process, the growing in monoculture and in one-year field is studied in a separeted way. Investigations are made on minimum 20 and maximum 50 plants/pair of plants.

Putting the obtained data in a table of infestation threshold, a rapid and simple decision can be made on whether treatments are needed or not on the particular field.

This technique does not only provide a simple decision model for specialists, but it can be easily learnt and safely applied by farmers.

DEMECOLOGICAL STUDY OF THE ADULT POPULATION OF *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE NEAR SZEGED IN 1999-2001

To the 80th anniversary of Dr. Nagy Barnabás

Krisztina Mucsi
Csongrád County Plant Health and Soil Protection Service,
6800 Hódmezővásárhely, P.O. Box 99., Hungary
e-mail: kmucsi-csnta@fki.gov.hu

Ede Petró
Centre Service for Plant Health and Soil Conservation,
1519 Budapest, P.O. Box 340., Hungary

Abstract
Results of the first-year structure analysis were reported in 2000. The investigations have been continued and the results were compared with those of the previous year.

This year again six surveys have been made in the region of Szeged and the results were analysed on the basis of formal and functional structure elements.

The obtained results were analysed and compared with data of the American literature.

Changes of the distribution, in time, of the population density were examined in 2000, in dry and rainless weather, while in 2001, in a relatively rainy weather.

Made in the frame of the SKK/SooC project.

BIOTECH APPROACH TO CORN ROOTWORM CONTROL: DEVELOPMENT STATUS OF MONSANTO'S CORN ROOTWORM RESISTANT MAIZE

Jay C. Pershing,
Monsanto Co., 700 Chesterfield Pkwy North,
Chesterfield, MO 63198

Abstract

Maize is the largest crop in the United States in terms of acreage planted and net crop value. In 2000, the U.S. maize crop covered 79 million acres. The western corn rootworm (CRW), *Diabrotica virgifera virgifera* LeConte, (Coleoptera: Chrysomelidae) is one of the most serious pests of maize in the United States. Larvae damage maize through feeding on the roots, which reduces the ability of the plant to take up water and nutrients from the soil and causes harvesting difficulties due to lodging. There are currently two common options for controlling CRW larval feeding, including synthetic insecticides and crop rotation. The later tactic can be ineffective as a result of behavioral modifications. The most common insecticide regime is an at planting time application, where the most widely used insecticides have been the organophosphate and synthetic pyrethroid insecticides. In 2000, these insecticides for CRW control were applied on over 18% of the maize acreage. This accounted for the largest insecticide usage in any one crop, totaling approximately 12 million pounds of active ingredient targeting CRW.

Recently, a third option has been developed using molecular biological techniques to produce transgenic maize expressing a *Bacillus thuringiensis (B.t.)* Cry3Bb protein. The Cry3Bb protein has been successfully transferred to corn to produce maize resistant to CRW feeding.

This new transgenic management tool has demonstrated superior larval feeding protection over current insecticides and has the potential to greatly reduce the amount of insecticides applied in maize cropping systems, thus benefiting the environment and potential adverse effects throughout the manufacturing, distribution and use chain.

This presentation describes the development status of Monsanto's Cry3Bb expressing maize, including the results of efficacy and safety testing.

EVALUATION OF CROATIAN CORN HYBRIDS FOR TOLERANCE TO CORN ROOTWORM (*DIABROTICA VIRGIFERA VIRGIFERA* LECONTE) LARVAL FEEDING

Marija Ivezić, Emilija Raspudić,
Univ.of J.J. Strossmayer in Osijek, Faculty of Agriculture,
Trg sv. Trojstva 3, 31000 Osijek, Croatia

Jon J. Tollefson,
Iowa State University, Department of Entomology,
17 Insectary Bldg., Ames , Iowa 50011-3140

Bruce E. Hibbard,
USDA-ARS, Plant Genetics Research Unit,
and Department of Entomology,
Univ. of Missouri, Columbia, MO 65211

Ivan Brkić,
Institute of Agriculture,
Department of Maize Breeding and Genetics,
31000 Osijek, Croatia

Abstract

The Western corn rootworm (*Diabrotica virgifera virgifera* LeConte (WCR)) is a major pest of corn in the USA. It was first recorded in Europe from Yugoslavia in 1992. The pest arrived in Croatia in 1995, and today over 200,000 ha are infested. In the USA insecticides are regularly used to control WCR, but the cost is high, can pose environmental risks, and may become ineffective due to resistance. Growing corn that is resistant to corn rootworms would be a valuable alternative to insecticides.

Ten Croatian (Institute of Agriculture, Osijek) and two Pioneer Hi-Bred Int. Inc. (Johnston, Iowa, USA) commercial corn hybrids were evaluated for WCR resistance at four locations using a randomized complete block design with four replications. Two locations were in east Croatia (Gunja, Osijek) and two were in the USA (Iowa, Missouri). At the Missouri location the plots were artificially infested with WCR eggs; the others contained natural rootworm infestations. At all locations, root injury was rated using the Iowa State University Node-Injury Scale (0-3) and the hybrids' tolerance to larval feeding was compared by evaluating root size and root regrowth. In the USA locations, tolerance was also measured by weighing (g) the root regrowth. In addition the Iowa experiment contained side-by-side insecticide treated and untreated rows to measure yield tolerance.

In an analysis of variance combined across locations, root injury, size and regrowth were significantly different among hybrids and there was a significant location by environment interaction. Root injury ratings averaged 1.52 and 1.67 at

Iowa and Missouri, respectively, and 0.07 and 0.36 at Osijek and Gunja, respectively. Analysis by location showed the hybrids differed significantly in root injury and root size at Croatia and Iowa, but not Missouri. Root regrowth was significantly different among hybrids at all four locations.

Key words: Western corn rootworm, root node injury, root size, root regrowth, commercial Croatian hybrids.

INTRODUCTION

The Western corn rootworm (*Diabrotica virgifera virgifera* LeConte (WCR) is a major corn pest in the USA. Corn rootworms cost US growers an estimated $ 1 billion annually in crop losses and control costs (Metcalf, 1986).

More then 50% of fields under continuous corn production are treated with insecticides for control of corn rootworm. However, many fields are unnecessarily treated each year (Gray *et al.*, 1993). As an alternative to insecticides, investigations on resistance to corn rootworm started 60 years ago and continue today. Tolerance, conferred by larger root size and greater root regrowth, has been identified as the primary mechanism. A tolerant plant sustains as much feeding damage as a susceptible plant, but is able to develop and produce high grain yield regardless of the injury (Riedell and Evenson 1993). Studies in Missouri in 1997 and 1998 identified several crosses that had significantly less damage caused by corn rootworm larvae (Hibbard *et al.*, 1999). Identifying commercial hybrids with tolerance to corn rootworm would reduce the amount of insecticides that needed to be applied.

In Europe the first record of WCR was from Yugoslavia in 1992 (Baca, 1994). The pest was identified in Croatia in 1995, and today it infests approximately 200.000 ha (Igrc-Barcic *et al.*, 2000). During 2000 near the border to Yugoslavia, some cornfields were found to be infested with 0.37 to 0.51 beetles per plant (Ivezic *et al.*, 2001). There was no visible lodging. This pest has been discovered in several other European countries and is spreading very fast. European scientists in the infested countries are trying to discover the best control measures for their production systems.

The objectives of this research were to evaluate 10 commercial Croatian hybrids under natural and artificially infestations to obtain information about their tolerance of tolerance to corn rootworms larval feeding different environmental conditions.

MATERIALS AND METHODS

Field experiments were conducted in 2001 at four locations. Two of them were in USA (Sutherland, Iowa and Columbia, Missouri), and two were in Croatia (Gunja and Osijek). The Missouri location was artificially infested with 1000 eggs/ft. The other locations used natural rootworm infestations. The experimental design at each location was a randomized complete block with four replications. In Croatia hybrids were planted in plots 6 m long with 2 seeds per hill, 25 cm between hills, and

0.76 m row spacing. Plots were thinned to one plant per hill. In the USA, the plots were machine planted with a 0.76 m row spacing with plants spaced approximately 23 cm apart. In Iowa the plots were two rows wide and 25 m long. One of the paired rows was treated with tefluthrin insecticide applied in the seed furrow at planting and the other was not treated with insecticide. The Missouri plots were only single rows 1.5 m long because they were artificially infested with WCR eggs. Because they are the most popular hybrids grown by Croatian farmers, the following ten Croatian hybrids were selected: Os 499, Os 84246-2, OsSK 444, OsSK 552, OsSK 596 R, OsSK 602, OsSK 617, OsSK 644, OsSK 654, and OsSK 659 (Institute of Agriculture, Dept. for Maize Breeding and Genetics, Osijek, Croatia). Two commercial hybrids from Pioneer, 3563 with known tolerance to WCR and 3573 known to be susceptible (Pioneer Hi-Bred Int. Inc. Johnston, Iowa, USA), were chosen as standards. Five competitive plants in each plot were chosen, tagged for identification, and dug from the soil. The root systems were cleaned with pressurized water and rated for rootworm larvae feeding injury. The root ratings were conducted on July 30 at Osijek, July 31 at Gunja, July 31 at Missouri, and July 19 and August 3 at Iowa. At the Iowa location, roots were cut off and dried so regrowth could be measured quantitatively.

Root injury was rated on the Iowa State University 0-3 Node-Injury Scale. The Iowa State University Node-Injury Scale is as follows: 0.00 – no feeding damage; 1.00 – one node, or the equivalent of an entire node, eaten back to within approximately 5 cm of the stalk; 2.00 – two nodes eaten; 3.00 – three or more nodes eaten. Damage between complete nodes eaten is noted as the percentage of the node missing, e.g., 1.50= one and half nodes eaten and 0.25 = one quarter of node eaten (Oleson and Tollefson 2000; Nowatzki et al. 2000).

The Eiben 1-6 Scale, reversed, (Rogers 1975) was used to rate root size and regrowth. On the reversed scales a rating of 1 indicated a large root system or well developed secondary roots, and a rating of 6 indicated a small root system or poorly developed secondary roots (*figure1*).

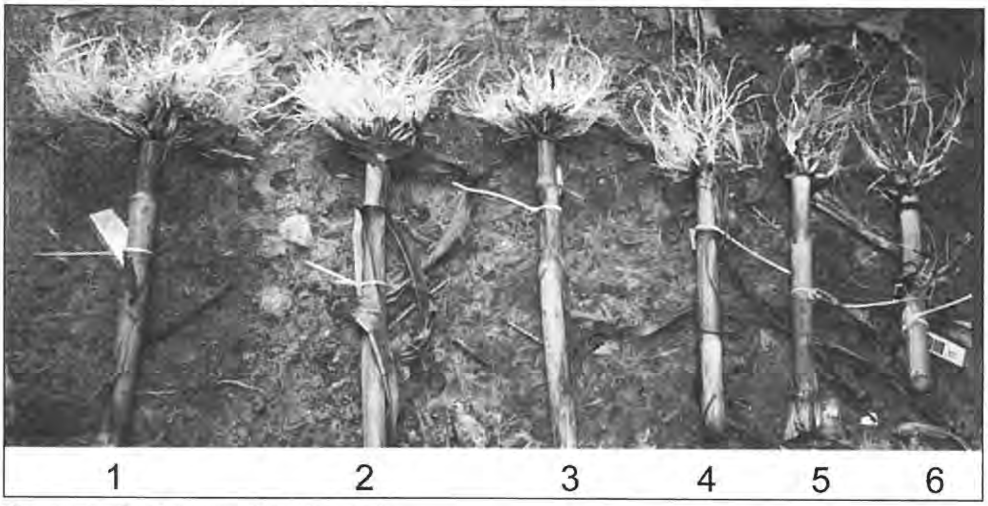

Figure 1 - Root regrowth rating scale

STATISTICAL ANALYSIS

Data for root injury, root size, and secondary root regrowth rating and weight in g were analyzed by location and hybrids using analysis of variance (SAS Institute 1999). Response variables were analyzed as least squares means, and means were separated using least significant differences (LSD) of the least squares means. The standard errors for means of hybrids and location were calculated. At the Iowa location, linear regression was used and coefficients of determination (R^2) calculated for relationship between root size, secondary root regrowth rating, and weight (g).

RESULTS AND DISCUSSION

In an analysis of variance combined across locations, root injury and root size were not significantly different, but regrowth differed significantly among hybrids. There was a significant location by environment interaction for all response variables (*table 1*). Root injury ratings averaged 1.52 and 1.67 at Iowa and Missouri, respectively, and 0.07 and 0.36 at Osijek and Gunja, respectively.

Table 1 - Combined Analysis; F-values

SOV	df	Root Injury	Root Size	Root Regrowth
Environment	3	37,59**	22,40**	12,00 **
Rep (Env.)	12	20,21**	7,21**	3,59**
Hybrid	11	0,90 ns	1,51 ns	9,95**
Env x Hybrid	33	4,05**	3,22**	3,42**

Separate analyses by environments showed that the hybrids differed significantly in root injury and root size at Croatia and Iowa, but not Missouri. Root regrowth was significantly different among hybrids at all four locations (*tables 2-5*).

There were significant differences (P = 0.01) among hybrids for root injury, root size, and secondary root development at Osijek. OsSK 644 had the least injury. The hybrids with the greatest regrowth were OsSK 617, OsSK 659, OsSK 596 R, P 3563, OsSK 602, and OsSK 644.

At Gunja there were significant differences (P = 0.01) among hybrids for root injury and secondary root development, but not for root size. The hybrids with the least injury were P3563, Os 499, and OsSK 617, and those with the highest regrowth were P 3563, OsSK 602, OsSK 659, and OsSK 617.

In Missouri there were no significant differences among hybrids for root injury and root size, but significant differences (P = 0.01) for secondary root development. OsSK 644, OsSK 617, and OsSK 602 had the greatest regrowth.

At the Iowa location, the root injury, root size, and secondary root regrowth in the insecticide treated and untreated plots was significantly different (P = 0.01). In untreated plots, root injury, root size, and secondary root growth were significantly different (P = 0.01) among hybrids. Hybrid OsSK 444 had the least injury. There was less plant lodging in the Croatian varieties OsSK 444 (10%), OsSK 644

(17.50%), OsSK 617 (25%), OsSK 654 (27.50 %), and OsSK 602 (30%), then the Pioneer hybrid 3563 (30%) known to be tolerant to WCR.

Secondary root rating were well correlated with regrowth measured in (g) (r=0.62) (*figure* 2), and with root size (r=0.57). The highest regrowth was at OsSK 617, OsSK 602 and OsSK 644.

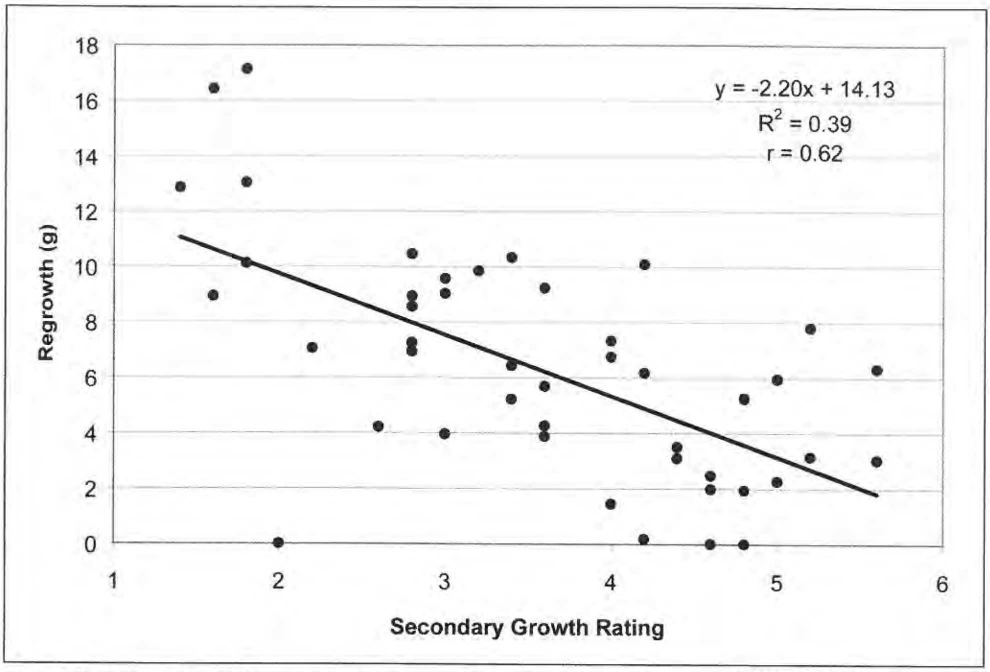

Figure 2 - Correlation between secondary growth scale rating and regrowth (g) at the Iowa location

Table 2 - Root injury by location (Node – Injury Scale 0-3)

Hybrid	Iowa	Missouri	Gunja, Croatia	Osijek, Croatia
Os 499	1.93 a	1.63 bcd	0.22 e	0.10 ab
Os84246-2	1.58 abcd	1.53 cd	0.80 a	0.10 ab
OsSK 444	0.90 e	1.47 cd	0.45 b	0.08 abc
OsSK 552	1.48 bcd	1.91 ab	0.30 bcde	0.07 bcd
OsSK 596 R	1.71 abcd	1.66 bcd	0.29 cde	0.07 abc
OsSK 602	1.36 d	1.63 bcd	0.40 bcd	0.08 abc
OsSK 617	1.56 abcd	1.97 ab	0.24 e	0.06 bcd
OsSK 644	1.30 de	1.47 cd	0.45 bc	0.02 e
OsSK 654	1.42 cd	1.70 bc	0.34 bcde	0.11 a
OsSK 659	1.90 ab	2.09 a	0.28 ed	0.05 cde
P 3563	1.31 de	1.66 bcd	0.20 e	0.07 bcd
P 3573	1.80 abc	1.34 d	0.34 bcde	0.03 de
Mean	**1.52**	**1.67**	**0.36**	**0.07**
LSD 0.05	0.42	0.36	0.16	0.04

Means followed by same letter not significantly different

Table 3 - Root size by location (Scale 1-6)

Hybrid	Iowa	Missouri	Gunja, Croatia	Osijek, Croatia
Os 499	2.25 e	2.69 d	1.95 abcde	1.35 de
Os84246-2	3.80 bc	3.00 bcd	1.65 bcde	1.40 cde
OsSK 444	2.25 de	3.38 abcd	2.25 ab	1.05 e
OsSK 552	2.95 de	3.63 ab	1.55 de	1.05 e
OsSK 596 R	2.95 de	3.50 abcd	2.50 a	1.50 bcde
OsSK 602	2.80 de	2.75 cd	1.70 bcde	2.00 a
OsSK 617	2.70 de	3.38 abcd	2.20 abc	1.95 ab
OsSK 644	3.15 cd	2.69 d	1.95 abcde	1.85 abc
OsSK 654	3.15 cd	4.00 a	1.60 cde	1.80 abcd
OsSK 659	3.90 ab	3.69 ab	2.10 abcd	1.55 abcd
P 3563	3.25 bcd	3.56 abc	1.45 e	1.40 cde
P 3573	4.55 a	3.56 abc	2.15 abcd	1.95 ab
Mean	**3.17**	**3.32**	**1.92**	**1.57**
LSD 0.05	0.72	0.85	0.61	0.48

Means followed by same letter not significantly different

Table 4 - Root regrowth by location (Scale 1-6)

Hybrid	Iowa	Missouri	Gunja, Croatia	Osijek, Croatia
Os 499	3.35 cde	3.06 cdef	2.95 bc	4.35 a
Os84246-2	4.15 b	3.69 bc	2.50 bc	2.70 c
OsSK 444	4.95 a	4.19 ab	4.40 a	4.60 a
OsSK 552	3.20 cdef	3.44 bcd	3.05 b	2.00 cde
OsSK 596 R	3.10 def	3.06 cdef	2.50 bc	1.70 e
OsSK 602	2.60 fg	2.56 def	1.45 d	2.00 cde
OsSK 617	2.05 g	2.50 ef	1.60 d	1.55 e
OsSK 644	2.85 ef	2.25 f	2.10 cd	2.45 cd
OsSK 654	4.90 a	4.63 a	3.35 b	4.25 a
OsSK 659	3.50 cd	3.50 bc	1.55 d	1.55 e
P 3563	3.80 bc	3.38 bcde	1.25 d	1.85 de
P 3573	4.90 a	3.75 abc	3.00 b	3.45 b
Mean	**3.61**	**3.33**	**2.48**	**2.70**
LSD 0.05	0.63	0.89	0.90	0.74

Means followed by same letter not significantly different

CONCLUSIONS

Analyses by location showed the hybrids differed significantly in root regrowth at all four locations. Secondary root ratings (visual scale) correlated well with regrowth weight (r = 0.62), and with root size (r = 0.57). It confirms that the visual scale for root regrowth could be used instead of measuring the weight of the regrowth. Root size, root regrowth, and lodging differed among hybrids and identified OsSK 617, OsSK 602, OsSK 644, and OsSK 444 as Croatian varieties that would stand reasonably well under severe rootworm feeding. These commercial varieties had tolerance that was comparable or better than the commercial Pioneer hybrid with known tolerance. Growing hybrids more tolerant to WCR does not eliminate corn rootworm populations, but it could reduce grower reliance on soil insecticides.

Table 5 - Root regrowth (g), Iowa

Hybrid	Root weight 1	Root weight 2	Regrowth
Os 499	7.72 cd	15.71 bcd	7.99 ab
Os84246-2	6.69 d	13.17 d	6.47 abc
OsSK 444	10.40 abc	12.16 d	1.76 c
OsSK 552	8.68 bcd	14.65 cd	5.97 abc
OsSK 596 R	9.53 abcd	14.74 cd	5.21 abc
OsSK 602	10.60 abc	20.27 ab	9.66 a
OsSK 617	12.84 a	21.18 a	8.34 ab
OsSK 644	11.73 ab	18.35 abc	6.62 abc
OsSK 654	8.54 bcd	13.22 d	4.68 abc
OsSK 659	8.58 bcd	14.28 cd	5.70 abc
P 3563	9.02 bcd	15.54 bcd	6.52 abc
P 3573	7.06 cd	10.99 d	3.93 bc
Mean	**9.28**	**15.35**	**6.07**
LSD 0.05	3.66	5.04	5.40

Means followed by same letter not significantly different

Acknowledgments to Tim Nowatzki for help with statistical analysis, Jim Oleson for helping with rating root injury, Stephanie Miller, student of Luther College, Decorah, Iowa, Mirjana Brmez and Drazen Dzoic, for their assistance in Croatia, to Institute of Agriculture in Osijek, Croatia and Pioneer Hi-bred Int. for providing the seedcorn, and to the Fulbright Program – USDS Council for International Exchange of Scholars for providing financial support for the senior author's visit to the USA.

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CORN ROOTWORM DISPERSAL IN THE SOUTH DAKOTA AREAWIDE MANAGEMENT SITE

Brian Wade French,
Northern Grain Insects Research Laboratory,
USDA–ARS, 2923 Medary Ave., Brookings, SD, USA, 57006

Laurence D. Chandler,
Red River Valley Agricultural Research Center, USDA-ARS,
1307 North 18th Street, Fargo, ND, USA, 58105

Abstract

The use of transgenic maize may soon be a reliable substitute for insecticides as a control tactic for corn rootworms (CRW). However, resistance among CRW could quickly evolve thereby diminishing the durability of the transgenic maize. To counteract the spread of resistance, refugia (non-transgenic maize) probably will be planted to sustain non-resistant CRW numbers. To keep resistant numbers low, gene flow among resistant and non-resistant CRW populations must occur. Dispersal is a means by which gene flow occurs among populations. Our goal is to examine the dispersal and reproductive biology of the western and northern CRW under field conditions. With respect to CRW movement, we wish to estimate the dispersal potential of western and northern CRW across the landscape in relation to sex, size, and reproductive status. Our study location is in the South Dakota CRW areawide management site. Using sticky traps, we captured 2460 northern CRW and 1116 western CRW dispersing between fields of continuous maize, first year maize, and soybean. Peak dispersal occurred in August for western CRW and August through September for northern CRW. Most CRW were captured between continuous and first year maize fields and at canopy height. Also using sticky traps attached to the top of conduit poles to detect vertical flight, we captured 805 northern CRW and 45 western CRW. More northern CRW were captured emigrating from maize fields than immigrating into maize fields. We found no differences in the number of western CRW immigrating and emigrating from maize fields. The number of western CRW was extremely low and may not be indicative of their migratory behavior into and out of maize fields.

EFFECTS OF PRECIPITATION AND TEMPERATURES ON THE LEVEL OF *DIABROTICA VIRGIFERA VIRGIFERA* POPULATION IN THE REPUBLIC OF SRPSKA IN 2000 AND 2001

Franja Baca, Dragisa Lopandic,
MRI Zemun Polje, 11085 Belgrade-Zemun, Yugoslavia

Jovo Stojcic, Vojislav Trkulja,
Institute of Agriculture, Banja Luka Slavko Radanovic,

Dragica Zivanovic,
Agricultural Station, Bijeljina

Dusko Paravac,
Agricultural Station, Doboj, Republic of Srpska

Abstract
At least 200 mm of precipitation during the maize growing season are necessary for maize growth under rainfed conditions. Two-fold higher precipitation sum is considered satisfactory, while 500-600 mm are considered ideal. Effects of drought and high temperatures in the eastern regions of RS fill a role of stress in 2000. Temperatures induced earlier larval hatching in region of Semberia. The more precipitation sum approached the minimum value of 200 mm, the more unfavourable effects on maize and WCR were. On the other hand, precipitation and temperatures in 2001 were not a limiting factor in all regions of RS.

Monitoring of incidence and spreading of WCR imagoes over pheromone traps were done as in previous years. The obtained results were analysed by comparison of the number of WCR imagoes over years and sums of precipitation and temperatures in Bijeljina, Doboj and Banja Luka. The favourable year of 1999 supported the development of the WCR population in 2000. Moreover, very warm April in 2000 with about 150°C HU accelerated larval hatching in maize continuous cropping in the area of Bijeljina; it also slowed down and reduced maize root development. Lack of nutrients and faster silk maturing resulted in significant WCR reduction in 2000 in comparison to 1999.

In 2001, the population increased by about 50% in relation to 2000 and equalled the abundance in 1999. The total number of imagoes per population amounted to 3,532, 1,972 and 3,722 in 1999, 2000 and 2001, respectively.

The data obtained by imago monitoring in RS during 2000 and 2001 point out to a fact that a significant reduction of WCR population can occur in years with extreme weather conditions such as 2000 was. The population abundance increased westward.

Table 1 - Fluctuation of population abundance 2000:1999 and 2001:2000

Distribution	Years			Index	
East-West	1999	2000	2001	2000:1999	2001:2000
< 50 km	202.4	133.4	256.6	66	192
51-100 km	43.0	58.5	94.8	136	162
101-150 km	43.5	68.6	41.1	158	60
> 150 km	0.2	0.4	5.8	200	1450

Key words: *Diabrotica v. virgifera*, maize, pheromone traps, population fluctuation.

EVALUATION OF POTENTIAL DAMAGE INCURRED BY CORN WEBWORM IN RUSSIA

Sergei S. Izhevsky, Vladimir N. Zhimerikin,
All-Russia Research Institute for Plant Quarantine.
Pogranichnaya str., 32, Bykovo,
Moscow region, 141050, Russia

Abstract

Western corn rootworm (WCR) is not yet registered on the territory of Russia. It is included in the group of unregistered species in The National List of Quarantine Objects. The most probable way of establishment of the beetle in Russia is via Ukraine. Its distribution will be active (by summer flight migration), and passive (by means of transport). The period between the moment the WCR is registered in Ukraine and its close approach to the Russian borders can vary from 8-9 years (invasion through Transcarpatia) to 3-4 years (invasion through the south of Moldavia). Thus, the most probable years for the appearance of the corn webworm in Russia are 2005-2010.

The northern border of the constant establishment zone of the WCR in Russia will be January isotherm -8°C. It goes through Lugansk, Kamensk-Shakhtersky, Astrakhan. The beetle will be able to form temporal local populations to the north of the described border, within the zone of maize cultivation. The probability of their existence will be limited by winter temperatures in the soil at the depth of egg deposition. The zone of constant will be the territory of about 360 thousand hectares, covering maize cultivation areas of Rostov region, Krasnodar and Stavropol Territories. With 20% losses (the average loss in the USA on condition of chemical protection), the total yearly damage incurred by the WCR in the described zone will be able to reach 40 million USD. At present, along the whole length of the western border of Russia that embraces the zone of probable acclimatization of the WCR, a complex of quarantine measures is applied, including pheromone monitoring.

INVITE* EC, AN ARRESTANT / FEEDING STIMULANT FOR CORN ROOTWORM CONTROL

Dr. Robert F. W. Schroder,
Director of Entomology Research

Tom Brown,
Vice President for Marketing Florida Food Products, Inc, P.O. Box 1300, Eustis, Florida USA, 32727-1300

Abstract

The corn rootworm is one of the most serious pests on corn in the United States. Growers routinely apply soil insecticides to 50-60% of the 30 million acres of corn annually to control the pest. Insecticides are routinely applied prophylactically and are frequently unnecessary, and may cause imposed health risks to growers, livestock and wildlife, in addition to possible ground water and environmental contamination. Florida Food Products, Inc, in cooperation with USDA, identified, extracted and developed a feeding stimulant/arrestant specific to the corn rootworm, known as Invite*. Invite*EC is an inert, emulsifiable concentrate that is mixed with low levels of insecticides (less than 10%) to control adult corn rootworm beetles. Invite* contains a naturally occurring, EPA approved arrestant/feeding stimulant upon which the corn rootworm compulsively feeds. We have clearly demonstrated in our field trials and aerial applications this summer that Invite* applied with low levels of organophosphates, carbamates, pyrethroids and other inert products dramatically enhanced insecticide activity, resulting in accelerated knockdown and sustained performance. The impact of Invite* mixed with reduced rates of pesticides on non-target insects including lady beetles, honey bees, lacewings, and other parasites and predators has been minimal. Invite* was applied with reduced rates of pesticides to over 50,000 acres of corn in 2000 and several 100,000 acres in 2001 with excellent results. We have provide the farmer with an alternative, safe, economical and effective product for control of the corn rootworm pest complex. We are planning to market this product in Europe and South America.

THE PROBLEM OF CORN ROOTWORM *DIABROTICA VIRGIFERA VIRGIFERA* LECONTE IN UCRAINE

Movchan O.M.,
The Main State Inspection Service of Plant Quarantine in Ukraine

Melnyk P.O.,
Ukrainian Scientific-Research Station of Plant Quarantine

Konstantynova N.A.,
Ukrainian Central Scientific-Research Laboratory of Quarantine

Abstract
Corn is one of the most important cultures in Ukraine, where much attention is paid to its growing. Early this culture occupies the square equal to 1,7 mln. ha.

Taking into account a considerable potential of corn in the frames of general corn balance, all technological processes directed at providing its stable production, are under special attention of specialists and scientists.

Control over the development of harmful organisms, prognosis and control in advance aimed at avoiding damages of yield, gained special weight today.

As the adventine species of *Diabrotica virgifera virgifera* appeared in Ukraine, the first years were dedicated, first of all, to propagandising the knowledge about this harmful species not only among specialists, but also among common public.

As a result, in these years a multitude of conferences, TV and radio programmes have been organized, methodical recommendations, posters and postcards were edited, containing detailed data (according to American and European authors) on biology, morphology, and other data concerning the development and harmfulness of *Diabrotica virgifera virgifera*.

Ukraine gave its specialist the opportunity to take part in all international conferences, dedicated to this problem. Specialists and scientists were informed about the materials, provided by researchers from abroad.

During the recent years Ukrainian National Service of Phytosanitary has been monitoring *Diabrotica virgifera virgifera*. Active and goal-oriented work of specialists gave its results. It helped to discover the pest in August 2001, in Zakarpattya, on the bordering line of Ukraine with Hungary and Romania. They were caught on the pheromone traps made in Moldova. Revealing in time provides the effective control over distribution of *Diabrotica virgifera virgifera* in Ukraine in the future.

On the whole, in 2001 the specialists of National Service monitored the territory of 500 thousand ha in 25 geographical regions and 106 administrative units in Ukraine. In general, pheromone traps of Moldavian, partly Hungarian and Ukrainian syntheses were used. The total amount of 1,200 traps were made use of.

Pheromone traps were installed in the second half of July. Every ten days the inspectors of Phytosanitary Service made calculations and took specimen form pheromone traps, which were sent afterwards for analyses to the Laboratory of Quarantine. Pheromone capsule in traps was changed every 30 days.

During investigations more attention was drawn to Zakarpattya, a bit later – also in Dyakovo. On August 22, 2001 in Beregiv region of Zakarpattya the imagos of Western Corn Rootworm were noticed in Kosyno and Chetovo, later – in Astej (*table 1*). These dwelled places are situated 1-7 km from Hungarian and Romanian borders. Visual inspections of corn plantations found no larvae of Western Corn Rootworm. The results of all conducted tests were recorded in a special journal.

Western Corn Rootworm was detected in the Laboratory of Quarantine in Zakarpattya zone. The identification of *Diabrotica virgifera virgifera* LeConte was confirmed by Central Scientific Laboratory of Quarantine.

Scientific institutions continue their researches. So, Ukrainian National Service Station of Plant Quarantine synthesized 8 analogs of natural genitor liquids. Some of them were installed along the perimeter with Romanian border in 3 regions of Ukraine. At the same time, thanks to friendly assistance from partners from Hungary, these liquids were approbated in the region with stable development of *Diabrotica virgifera virgifera*. The signs of effectiveness of different liquids, as well as their modifications, were defined and necessary results were attained. It will be the basis of further improvement of their effectiveness and fitting to modern requirements.

At this point we would like to underline that such cooperation helps to work out the system of control over development and distribution of adventive species on European territory more effectively.

Besides, scientific searching for the signs of corn plants resistance to *Diabrotica virgifera virgifera* also took place. According to this, Ukrainian National Service Station of Plant Quarantine modified the device (published before by Dr. F. Bocza from Yugoslavia) for the identification of the maturity stage of corn root system in the first half of vegetation. By means of this device, we received data about resistance of root system in different hybrids. This can be used in further approaches to selection and seedage of hybrids with strongly developed root system.

So, in Ukraine the main directions of control over *Diabrotica virgifera virgifera* have been already identified.

Table 1 – The review of *Diabrotica virgifera virgifera* LeConte occurrence in Ukraine in Zakarpattya region in 2001.

District	Place of occurrence	Date of occurrence	Number of caught adults
Vynogradiv	Holmovets	15.08.2001 27.08.2001 13.09.2001	1 1 1
	Yulivtsyi	15.08.2001 27.08.2001 13.09.2001	2 1 1
	Dyakovo	27.08.2001	4
	Bobove	15.08.2001 27.08.2001	1 1
Beregove	Kosyno	22.08.2001	1
	Chetovo	22.08.2001 12.09.2001	1 1
	Asteii	12.09.2001	1

SPATIAL VARIABILITY OF NORTHERN CORN ROOTWORM DISTRIBUTION IN RELATION TO LANDSCAPE POSITION

M. M. Ellsbury,
USDA-ARS, Northern Grain Insects Research Laboratory,
Brookings, SD, USA, 57006
e-mail: mellsbur@ngirl.ars.usda.gov

D. D. Malo, S. A. Clay, D. E. Clay, C. G. Carlson,
Plant Science Department, South Dakota State University,
Brookings, SD, USA, 57007
e-mail: david_clay@sdstate.edu

Abstract

The soil environment plays a significant role in determining changes of corn rootworm distribution in the soil from the time eggs are deposited until the damaging larval populations occur. Because of the influence of the soil environment on the survival of the immature stages of corn rootworm, we have hypothesized that measurable soil properties, such as soil electrical conductivity, may be used to predict where corn rootworms are most likely to survive and cause economic loss. The distribution of the immature stages of corn rootworms in relation to landscape position and soil electrical conductivity is discussed with reference to prospects for site-specific management of corn rootworms.

Key words: northern corn rootworm, site-specific management, spatial variability, landscape position, soil ecology.

INTRODUCTION

Corn rootworms, *Diabrotica* spp. (Coleoptera: Chrysomelidae), are major pests of corn in North America. The western corn rootworm, *Diabrotica virgifera virgifera* LeConte, has appeared in Eastern Europe and its range of distribution in Europe is presently expanding. A second species, the northern corn rootworm, *D. barberi* Smith and Lawrence, is not found outside North America. Northern corn rootworms, unlike western corn rootworms, are capable of surviving over two winters as an egg stage in the soil, and thus may produce economic loss in corn rotated with soybean. Survival of corn rootworm immature stages is highly influenced by properties of a variable soil environment that mediate spatial variability in field distributions of the emerging adults (Ellsbury *et al.*, 1998). Corn rootworm larvae are dependent on a narrow range of host plants, primarily corn, and are incapable of moving from oviposition sites more than about one meter through the soil to locate host plant resources (Short and Luedtke 1970, Gustin and Schumacher 1989).

Because of spatial variability in their field distributions and the relative immobility of the soil-dwelling stages, corn rootworms are potential candidates for site-specific management strategies. Moreover, the influence of the soil environment on survival of the immature stages of corn rootworms suggests that measurable soil properties, such as soil electrical conductivity, may be used to predict where corn rootworms are most likely to survive and cause economic loss. Our objectives were to compare spatially explicit data for northern corn rootworm egg and adult population distributions, root injury by the larval stage, and electromagnetic induction (EMI) properties of the soil from different landscape positions in two rotated cornfields.

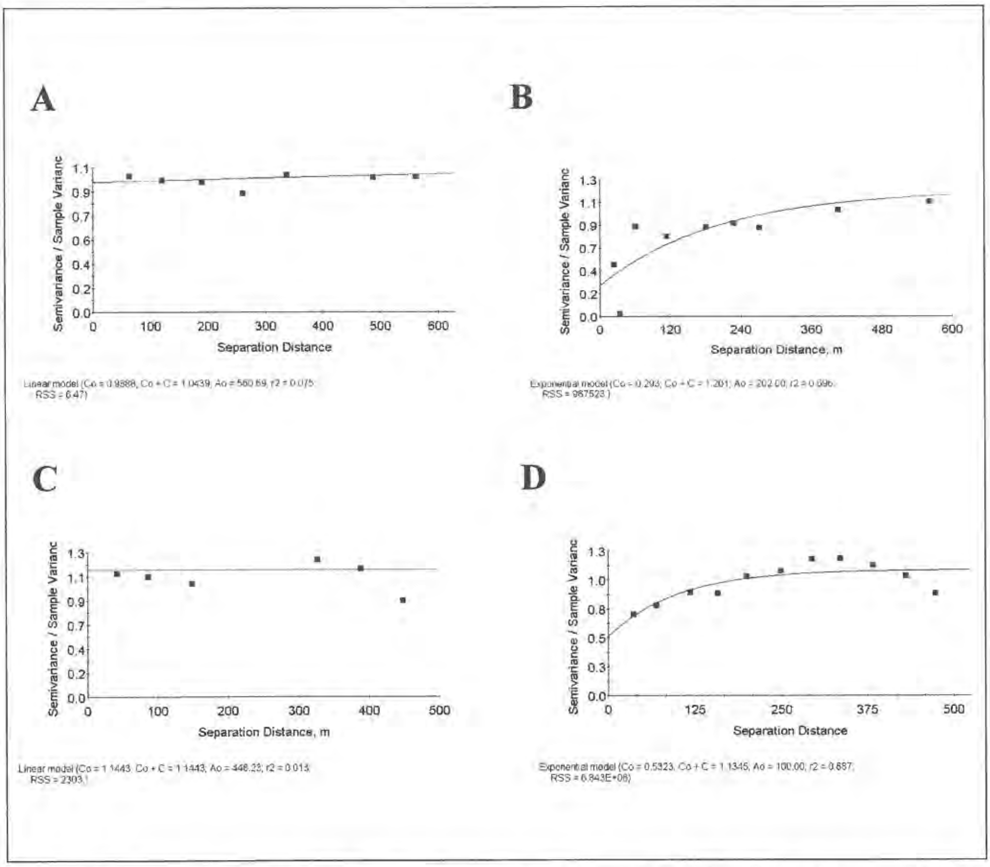

Figure 1 - Semivariograms for egg density per liter of soil in the Brookings County (A) and Moody County (C) study sites, and for adult emergence distributions in the Brookings (B) and Moody County (D) study sites in South Dakota, USA

Specifically we were interested in determining whether lower rootworm populations and reduced injury to corn occurred consistently in poorly-drained footslope/toeslope areas that are characterized by higher soil electrical conductivity values.

Methods

Egg and adult stages of northern corn rootworms, *Diabrotica barberi* Smith and Lawrence, were grid-sampled at study sites, in Brookings and Moody Counties, South Dakota, both farmed in rotation with soybean. The Moody Co. site was located in the SW1/4, S8, T108N, R48W of the 5th principal meridian, Moody Co., South Dakota or latitude 44° 10′ 15″ N and longitude 96° 37′ 25″ W. The study site was 64.8 ha (160 a) in size with 16.5 m (54 ft) of relief. The average elevation at the site was 523 m (1715 ft) above sea level. The Moody site had an undulating topography with water seepage and poor drainage in toeslope/footslope areas, particularly in the northeast quadrant. The field also was known to have been fenced and divided along a line running from north to south about 300 m from the eastern margin.

Soils at the Moody Co. site formed in late Wisconsin glacial till and loess. Soils at the summit and shoulder hillslope positions are fine-silty and fine-loamy, mixed, superactive, frigid Calcic Hapludolls (Kranzburg, Venagro, and Vienna series). Soils at the backslope positions are fine-silty, mixed, superactive, frigid Pachic Hapludolls (Waubay series). At the footslope and toeslope positions the soils are fine, smectitic, frigid Typic Argiaquolls (Badger series); fine-silty, mixed, superactive, frigid Aeric Calciaquolls (Cubden and McIntosh series); and fine-silty, mixed, superactive, calcareous, frigid Cumulic Endoaquolls (Lamoure series).

The Brookings Co. study site was located in the SE1/4, S24, T109N, R49W of the 5th principal meridian, Brookings Co., South Dakota or latitude 44° 13′ 41″ N and longitude 96° 39′ 04″ W. The study site was 64.8 ha (160 a) in size with 14.4 m (47 ft) of relief. The average elevation at the study area was 509 m (1670 ft) above sea level. Topography of the Brookings Co. site was relatively flat, except for a high point in the northeast quadrant and a low-lying waterway running from northeast to southwest and underlaid by drainage tile.

Soils at the Brookings Co. site formed primarily in late Wisconsin glacial till. Soils at the summit and shoulder hillslope positions are fine-loamy and coarse-loamy, mixed, superactive, frigid Calcic Hapludolls (Barnes, Egeland, and Vienna series). Soils at the backslope and footslope positions are fine-silty, mixed, superactive, frigid Aquic Hapludolls (Brookings series) and fine-silty, mixed, superactive, frigid Aeric Calciaquolls (McIntosh series).

Georeferenced sample sites were established at each study site in transects about 65-75 m apart. Within transects, sites also were 65-75 m apart and were offset about 33 m from those in adjacent transects. Additional sites were established within selected transects to provide data at lag distances of less than 65 m between sites. Egg populations were estimated from four one-liter soil core samples taken at each study site during mid-April, bulked, and subsampled to one liter. Eggs were washed from the one-liter subsamples using the methods of Shaw *et al.* (1976). Adult northern corn rootworm emergence density was monitored weekly at each grid node by use of emergence enclosures subtending 0.5 m^2 of ground surface. Adult emergence was expressed as total numbers of beetles emerging over the growing season prior to the first killing frost. Three whole-plant samples also were taken at each sample site to characterize larval injury to roots. Root systems were washed free of soil and root injury was visually rated using the 1 to 9 scale of Welch (1977), where 1= no root injury and 9 = three nodes of roots destroyed. Root injury level at each site was expressed as the average rating for the three samples. Soil electromagnetic induction (EMI) was measured using an EM38 meter (Geonics Limited, Mississauga, Ontario, www.geonics.com) fitted to a wooden trailer and pulled about 30 cm above the soil surface at 11 km/hr behind a GPS-equipped vehicle (Fritz *et al.*, 1999). Georeferenced EMI measurements were taken over grid transects at 15 m spacing.

Data for rootworm egg and adult populations, root ratings, and EM-38 values were assigned to landscape positions defined by arbitrary elevation limits depending on topography of each study site using Arcview. Zones of landscape position in the Moody Co. field were established as: footslope/toeslope, < 525 m (below 1723 ft); backslope, 525 - 530 m (1723-1740 ft); and shoulder/summit, > 530 m (above 1740 ft). Zones of landscape position in the Brookings County field were established as: footslope/toeslope, < 510 m (below 1723 ft); backslope, 510 - 514 m (1723-1740 ft); and shoulder/summit, > 514 m (above 1740 ft). Values for rootworm population parameters, EM-38 readings, and crop injury ratings within each landscape position zone were subjected to analysis of variance.

Results and discussion

Semivariograms for density of northern corn rootworm eggs, shown in *figures 1A* and *c* for both study sites, showed little spatial dependence in the distribution of rootworm eggs at the time of sampling, just before the start of the growing season. Egg densities were higher in the Moody Co. field than in the Brookings Co. field (*figure 2*). Mean egg densities in the Moody Co. field did not vary with landscape position (*table 1*) even though areas of higher density were evident in a contour map of egg data (*figure 2A*).

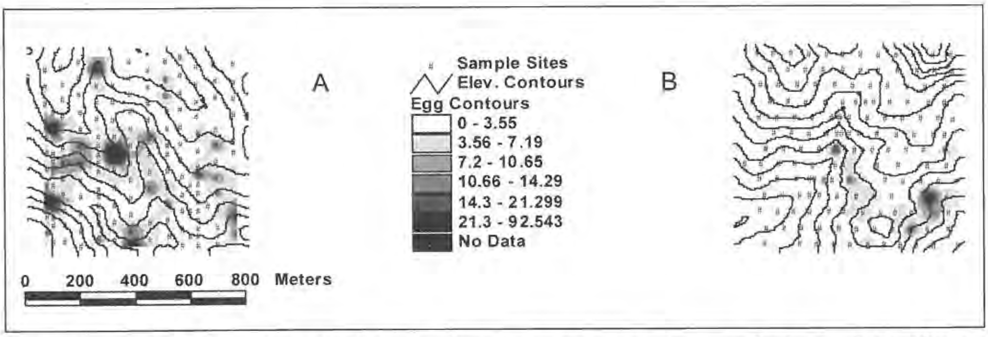

Figure 2 - Egg density per liter of soil for northern corn rootworms from study sites in fields of rotated maize during 1999 in Moody Co. (A) and 2000 in Brookings Co. (B), South Dakota. Egg numbers are shown as grayscale overlaid on contour lines for elevation

Conversely, contour mapping (*figure 2B*) of egg data from the Brookings field showed lower overall densities and no obvious areas of higher numbers, yet mean egg densities were significantly higher at the lower (toeslope) landscape positions than at the backslope and summit/shoulder positions of the Brookings site (*table 2*). The data suggest that lag distances of 65-75 m were too large to detect spatial dependence in egg densities and that oviposition occurred randomly over the fields.

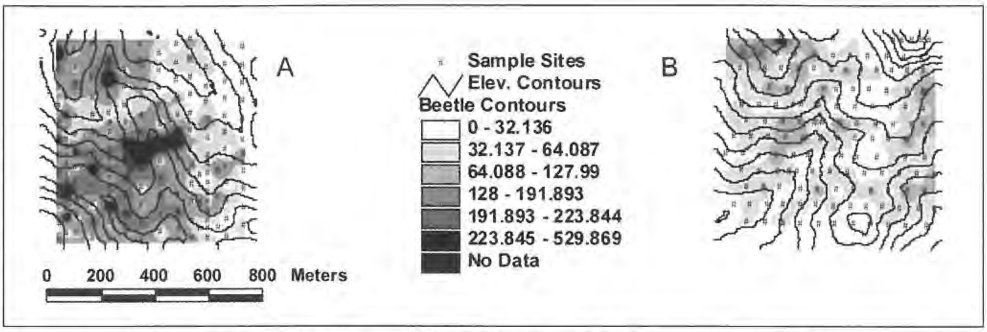

Figure 3 - Adult emergence density per 0.5 m² of soil surface for northern corn rootworms from study sites in fields of rotated maize at locations during 1999 in Moody Co. (A) and during 2000 in Brookings Co. (B), South Dakota. Total numbers of adults emerged are shown as grayscale contours overlaid on contour lines for elevation

Semivariograms for adult emergence densities (*figures 1B* and *D*) at both study sites showed a distinct sill and suggested spatially dependent variation that was not evident in the distribution of the egg stage. Adult emergence at the Moody Co. site was lowest at footslope positions (*figure 3A*) where seepage and higher salinity was evident. Conversely, emergence of adult rootworms and root injury ratings were highest along ridges and better-drained backslope areas (*figure 3A*) of the Moody Co. field. Adult population levels generally were lower in the Brookings Co. field (*figure 3B*) than in the Moody Co. field. The highest adult corn rootworm emergence densities were found in the southwest quadrant of the Brookings Co. site. This quadrant was the lowest (footslope/toeslope) portion of the field, but did not have the seepage and poor drainage characteristics, that were found at lower elevations of the Moody Co. field. In both fields, the highest EMI readings were associated with soil at the footslope/toeslope landscape positions (*tables 1* and *2*). In the Moody Co. field, adult emergence appeared negatively correlated with the higher EMI readings (*table 1*) at poorly-drained footslope and toeslope positions. In contrast, adult emergence in the Brookings Co. field was highest yet root injury ratings were lowest at the footslope/toeslope positions, where drainage was facilitated by a tile system and EMI readings also were highest. The reasons for these apparently diametrically opposed and inconsistent results from the two fields are unknown, but may be related to differences in the interactions of the soil-dwelling stages of corn rootworms with the distinctive soil types and drainage characteristics of the two fields.

Table 1 - Landscape position in relation to northern corn rootworm egg density, total adult emergence, root injury ratings, and soil electrical conductivity (EMI) for the Moody Co., SD study site during 1999. Numbers in parentheses are standard errors; means within a column followed by the same letter were not significantly different, Fisher=s protected LSD ($p<0.05$). Root injury was visually estimated on the 1 to 9 scale of Welch (1977)

Landscape Position	Elevation interval, m	EMI mS m^{-1}	Eggs per liter of soil	Mean total adults per 0.5 m^2	Root injury Ratings
Footslope/ toeslope	< 525	41.06 a (0.18)	5.1a (1.8)	66.8 a (9.4)	3.9 a (0.2)
Backslope	525 - 530	35.9 b (0.17)	5.0 a (1.2)	107.0 b (13.2)	4.0 a (0.2)
Shoulder/ summit	> 530	31.02 c (0.07)	4.8 a (1.4)	115.5 b (18.1)	4.5 b (0.3)

Table 2 - Landscape position in relation to northern corn rootworm egg density, total adult emergence, root injury ratings, and soil electrical conductivity (EMI) for the Brookings Co., SD study site during 2000. Numbers in parentheses are standard errors; means within a column followed by the same letter were not significantly different, ($p<0.05$). Root injury was visually estimated on the 1 to 9 scale of Welch (1977)

Landscape Position	Elevation interval, m	EMI mS m^{-1}	Eggs per liter of soil	Mean total adults per 0.5 m^2	Root injury Ratings
Footslope/ toeslope	< 510	33.27 a (0.43)	2.6a (0.6)	44.6 a (3.5)	3.6 a (0.1)
Backslope	525 - 530	30.75 b (0.24)	1.4 b (0.2)	46.1 a (5.1)	3.8 a (0.2)
Shoulder/ summit	> 530	31.54 b (0.48)	1.4 b (0.4)	30.6 a (11.5)	4.2 a (0.6)

In an earlier publication we concluded that soil characteristics, such as EMI measurements or pH, may be valuable as indicators of infestation potential for corn rootworms (Ellsbury *et al.*, 1999). Comparison of results from the two present study sites suggest that it will be difficult to develop generalized guidelines for site-specific management of corn rootworms based on any single sampling protocol or indicator variable. In the examples from our Moody Co. and Brookings Co., South Dakota study sites, rootworm populations appeared to behave quite differently in each field with respect to field topography, root injury, and soil EMI properties. These results still suggest that sufficient spatial variability exists in rootworm populations to justify consideration of site-specific management, but also indicate that this will require an approach that incorporates multivariate information on individual field history, soil characteristics, and a better knowledge of the complex interactions of corn rootworm populations with the soil environment.

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A COMPARISON OF THE TRECE AND CSALOMON (EUROPEAN) CRW TRAP FOR MONITORING POPULATIONS OF *DIABROTICA* SP. IN KANSAS

Jasdeep Singh, Gerald Wilde,
Department of Entomology.
Kansas State University. Manhattan, KS. USA

Two plant allelochemic based traps were compared for their efficiency in monitoring populations of western, northern and southern corn rootworm beetle populations near Abilene, Kansas in 2001.

Materials and Methods

Eight traps of each type were placed alternately on the perimeter (outside row) and internally in six different cornfields. Traps were placed at least 30 m apart and examined every 7 days. Traps were initially placed in the field when corn was in the green silk stage and examined over a six-week period. Trap catches were placed in resealable plastic bags, brought back to the laboratory, and the number of beetles of each species counted.

Results

Combined data from the six fields for each of the last three sample periods are summarized in *tables 1-3*. There were significantly more western (*D. virgifera virgifera*) southern (*D. undecimpunetata howardi*) and northern (*D. longicornis*) com rootworm beetles caught in the Trécé than in the Csalomon traps on most dates. For the western corn rootworm, the Trécé trap caught from 2-5 times the number of beetles caught in the Csalomon trap on most dates. Differences were greater in traps placed in the field than those on the perimeter of the field. Similar results were recorded for the southern and northern species. In general, Trécé traps placed in the field caught more beetles than Trécé traps placed on the field perimeter. On the other hand, the Csalomon trap caught approximately equal numbers of beetles in the inner field and perimeter traps. We were able to examine and emptv the Trécé traps more rapidly than the Csalomon traps.

Summary

Both the Trécé and Csalomon com rootworm traps were effective in monitoring populations of the *Diabrotica sp.* occurring in Kansas. Since the Trécé trap was more efficient to examine and caught more beetles, it would be the trap of choice where beetle populations are extremely low.

Table 1 – Comparison of Western Corn Rootworm Catch by the Csalamon and the Trécé Western Corn Rootworm Traps, Abilene, KS. 2001. G. Wilde, Kansas State University

Date	Location[1]	Species	Trap	Mean[2]± SEM
8/6/01	In	WCR	European Trécé	6.4 ± 1.5 a 33.3 ± 8.5b
	Out	WCR	European Trécé	9.3 ± 6.9 a 10.6 ± 2.7 a
8/15/01	In	WCR	European Trécé	6.5 ± 1.7 a 20.0 ± 5.4 b
	Out	WCR	European Trécé	5.3 ± 2.3 a 8.5 ± 2.5b
8/21/01	In	WCR	European Trécé	1.6 ± 0.4 a 5.7± 1.7a
	Out	WCR	European Trécé	0.6 ± 0.3 a 3.3 ± 1.1 a

[1] Location: In = traps placed at least 100 paces into the field; Out = traps placed around the edge of the field

[2] Means within a date and location followed by the same letter are not significantly different (P>0.05)

Table 2 - Comparison of Southern Corn Rootworm Catch by the Csalomon and the Trécé Western Corn Rootworm Traps, Abilene, KS. 2001. G. Wilde, Kansas State University

Date	Location[1]	Species	Trap	Mean[2]± SEM
8/6/01	In	SCR	European Trécé	6.7 ± 1.6 a 27.3 ± 3.9 b
	Out	SCR	European Trécé	6.5 ± 2.7 a 19.4 ± 4.3 b
8/15/01	In	SCR	European Trécé	5.6 ± 1.3 a 14.3 ± 2.6 b
	Out	SCR	European Trécé	5.9 ± 1.9 a 10.7 ± 1.9 b
8/21/01	In	SCR	European Trécé	0.6 ± 0.2 a 1.8± 0.3 a
	Out	SCR	European Trécé	0.8 ± 0.3 a 2.7 ± 0.5 a

[1] Location: In = traps placed at least 100 paces into the field; Out = traps placed around the edge of the field

[2] Means within a date and location followed by the same letter are not significantly different (P>0.05)

Table 3 - Comparison of Northern Corn Rootworm Catch by the Csalomon and the Trécé Western Corn Rootworm Traps, Abilene, KS. 2001. G. Wilde, Kansas State University

Date	Location[1]	Species	Trap	Mean[2]± SEM
8/6/01	In	NCR	European Trécé	0.7 ± 0.2 a 3.2 ± 0.6 b
	Out	NCR	European Trécé	1.7 ± 1.0 a 5.3 ± 1.5 b
8/15/01	In	NCR	European Trécé	0.6 ± 0.2 a 1.2 ± 0.3 a
	Out	NCR	European Trécé	1.7 ± 0.7 a 1.1 ± 0.3 a
8/21/01	In	NCR	European Trécé	0.03 ± 0.03 a 0.5 ± 0.1 a
	Out	NCR	European Trécé	0.3 ± 0.2 a 0.6 ± 0.2 a

[1] Location: In = traps placed at least 100 paces into the field; Out = traps placed around the edge of the field

[2] Means within a date and location followed by the same letter are not significantly different (P>0.05)

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Proceedings

FINAL RESULTS ON ESTIMATION OF IWGO INBREDS RESISTANCE TO BORERS *OSTRINIA NUBILALIS* HBN. AND *O. FURNICALIS*

F. Bača,
Maize Research Institute Zemun Polje,
11080 Belgrade-Zemun, Yugoslavia

H. Berger,
BFL Inst. F. Phytomedicine A-1126 Wien, Austria

P. Anglade,
INRA Station de Zoologie, 33883 Bordeaux, France

and *IWGO cooperators**

Abstract

The fundamental intention of establishing the IWGO programme in 1968 was to standardise methodology on resistance testing of maize inbred lines to both European (ECB - *Ostrinia nubilalis* Hbn.) and Asian Corn Borers (ACB - *Ostrinia furnacalis*). In order to achieve set goals, the work of maize breeders and entomologists was united and performed through several stages.

The 14-year results obtained in 14 countries of Europe, Asia and North America are discussed in this paper. As the testing stage is finished, data are unified and some questions important for maize selection can be answered.

Resistance tests were performed by plant artificial infestation with ECB/ACB egg masses. In some countries, wild population egg masses were used, while in others, egg masses of populations artificially fed were used. Depending on a country, resistance estimations encompassed leaf feeding scores, general damage rating, grain yield, yield response and data on stalk dissection. Inbreds, selected as checks, were evaluated under different agroecological conditions over years. According to results of resistant and susceptible standards, complexity of ECB/ACB dependence on agroecological conditions and inbred responses to larvae causing damage to them are the best observable.

The overall estimation can be determined on the basis of resistance parameters and their correspondence to yield response to damages caused by ECB larvae obtained in Zemun Polje. Significant differences were observed in all registered resistance parameters over investigation years. Inbred responses to larval damages

* W. Faber† (Austria), T. Georgiev and L.Gerginov (Bulgaria), M. Hudon and M.S. Chiang (Canada), D. Zhou (China), P. Anglade (France), B. Dolinka and B. Nagy (Hungary), G. Mariani and F. Coppolino (Italy), C. Kania (Poland), P. Silas (Portugal), D. Mustea (Romania), A. Monteagudo† (Spain), D.I. Shapiro†, D.S. Pereverzev and A.N. Frolov (Russia), V. Masler†, A. Piovarci†, M. Nešticki, J. Longauerova and B. Ryšava (Slovakia), H.C. Chiang, W.D. Guthria and L.C. Lewis (USA), D. Hadžistević and Z. Kaitović, Yugoslavia

and pest responses to agroecological conditions were also determined. Results obtained in different countries varied not only over years, but also over inbreds, signifying that the final inbred estimation should been done with caution.

Data on over 360 inbreds were catalogued according to their breeding value and shall be presented in the IWGO Newsletters.

Key words: European corn borer (ECB – *Ostrinia nubilalis)* Asian corn borer (ACB - *Ostrinia furnacalis*), inbred lines, resistance, productivity.

THE BEHAVIOUR OF A MONSANTO MAIZE HYBRID DEKALB 512 Bt TO THE ATTACK BY THE EUROPEAN CORN BORER *(OSTRINIA NUBILALIS)* IN ROMANIA

Alexandru Bărbulescu, Constantin Popov, Ion Sabau
Research Institute for Cereals and Industrial Crops,
8264 Fundulea-Călărasi, Romania
e-mail: fundulea@ns.ricic.ro;
Phone/fax: 401-311.07.22

Abstract

The European Corn Borer is a major pest in many areas of Romania, and by its attack during the late verticil phase and especially in the panicle, stem and peduncle of the cob, causes significant grain production losses. The basic method to prevent its attack is growing resistant hybrids, and several of such hybrids were promoted, especially in the USA; lately there is a tolerant experimental hybrid in Romania, too. To promote genetically modified hybrids in Romania, Monsanto made available for testing the hybrid Dekalb 512 Bt. This paper presents the results.

The experiment was performed under artificial infestation of plants, using 20 egg-mases per plant for at least 10 plants for each variant. For the infestation to take place under different vegetation phases and weather conditions, including the usual egg-laying period for moths in natural conditions, sowing and infestation were performed at several times. Together with the hybrid Dekalb 512 Bt, two widely-grown indigenous hybrids were used, caracterized by a lower attack of the borer, namely Fundulea 376 and Olt. Separately, the hybrid Dekalb 512 Bt was compared to a tolerant indigenous hybrid, HSF 1071-96 and a pest-susceptible indigenous hybrid, HSF 1147-96. The attack was assessed using a longitudinal cross-section of the plants during corn harvesting period and recording the attack cavity lengths and the count of live and dead larvae/plant.

The results showed that, although the attack of the corn borer during the experiment period was rather low, due to unfavourable weather conditions for the pest, attack values gave the opportunity to note a significant differentiation in the reaction of the test hybrids. The hybrid Dekalb 512 Bt showed in all cases a significant resistance to the attack of *Ostrinia nubilalis*, the attack values showing only sporadic traces of attack by the pest. The resistance of the hybrid Dekalb 512 Bt was satisfactory for the vegetation phase when the infestation was performed, including after the panicle had appeared, which corresponds to the period of evolution of the only generation that has economic significance in Romania. Although the indigenous hybrid in the experiment HSF 1071-96 had a low level of attack, the genetically hybrid Dekalb 512 Bt was practically not attacked by the corn borer.

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

INTRODUCTION

It is a well-known fact that the European Corn Borer (ECB) is one of the major corn pests in Romania, especially in the central and western region of the country, in hill areas and along the main rivers, in the Danube floodplains and under irrigations. Its attack, in the later verticil phase of the leaves and especially in the panicle, stem and peduncle of the cob, can cause significant damage, reaching 60% of the grain production under particularly favourable conditions (Paulian *et al.*, 1962).

The fact that, in the USA, *Ostrinia nubilalis* has two generations that are dangerous to corn crops, has caused, as early as the first half of the last century, that complex and multiple studies be performed on this pest, especially on how to control it. Following the many years of research, it was proven that plant resistance is the most effective way to protect corn crops against both the first and the second generation (Hudson *et al.*, 1989). Many hybrids that were created tolerant or resistant to the attack of the pest were made available for production and quickly became widespread and dominant, especially in the Corn Belt areas.

The high economic significance of the corn borer in the USA made it necessary that, once major breakthroughs were achieved in genetic engineering, a series of studies be started for the use of genetically modified corn hybrids and, within a short period, corn growers were given corn hybrids with the Bt gene, According to Edwards (1999), of the total area of 31,417,004 ha cultivated with corn in the USA in 1998, genetically modified hybrids occupied an area of 4,858,300 ha.

In Romania the various studies performed especially during the last decades of the past century, regarding the resistance of corn against *Ostrinia nubilalis* attacks, lead to creation, by conventional means, of inbred lines with a certain degree of resistance that at the same time had high combinative value and superior agronomic features (Barbulescu and Cosmin, 1997). By crossing some of these lines tolerant corn hybrids were obtained during the past few years (Barbulescu *et al.*, 1999).

In order to promote genetically modified corn hybrids in our country as well, Monsanto made available the hybrid Dekalb 512 Bt for testing under the conditions found in Romania, and the results that were obtained are mentioned in this paper.

MATERIALS AND METHODS

The experiments were conducted during the period between 1998 and 2000, under artificial infestation conditions. The hybrid Dekalb 512 Bt was used together with widely grown indigenous hybrids, Fundulea 376 and Olt, characterized by a certain attack reduction degree. In a separate experiment, the Dekalb 512 Bt was compared with an experimental hybrid HSF 1147-96, susceptible to the attack of

ECB and with a tolerant hybrid HSF 1071-96, created from inbred lines with a certain degree of resistance to this pest.

In order to have the plants infested in various vegetation phases and under various climatic conditions that would usually include the egg-laying period under natural conditions, sowing was performed at two different dates, and the plants were infested at several dates, starting with a few weeks before the appearance of the panicle and, in some cases, until after the panicle had appeared. Twenty egg-mases per plant were used for infestation, and for at least 10 plants of each variant, the egg-mases were in the black head phase, and were inserted into the leaf verticil or attached to the leaves with pins where the panicle had appeared. Infestation with a different number of egg-mases, 5, 10, 15, 20 per plant was also experimented, with the plants in different vegetation phases.

The attack was assessed in the fall, during the corn harvesting period, by cutting a longitudinal cross-section of the plants and recording the total length in cm of attack cavities for each plant, and the number of live and dead larvae/plant.

RESULTS AND DISCUSSIONS

For the overall degree of attack produced by the corn borer during the experiment period (*tables 1* and 2), it was observed that, due to weather conditions that were generally unfavourable for the pest, a low degree of attack was recorded, compared with a year that was favourable to the evolution of the pest, such as 1988, when, in a susceptible corn line, the average length of cavities was 63 cm/plant, with the maximum value of 135 cm/plant, and the average larvae count was 7.3 per plant, up to 13.7 per plant (Bărbulescu, 1989). Nevertheless, the attack was more visible during the first year of the experiment and much reduced, almost sporadic, during the last year, when temperature during infestation and immediately thereafter was very high, frequently exceeding 35°C, and drought was also very severe.

Reviewing the attack data for the two indigenous hybrids by sowing date and especially by infestation date, it was observed that the attack degree was usually not influenced by the period between sowing and infestation.

However there was an exception in 1998, for the second sowing period, June 3, to the first infestation period, June 8, only 35 days after sowing the attack was lower, due to a less advanced vegetation phase of the plants. Comparing the data with data from the same year during the first sowing period (May 7) and the infestation period of July 6, in similar environment conditions, it was observed that higher attack values were recorded in the latter case, the difference in the attacks being caused by the different vegetation phases at the moment of infestation.

Considering the data on the infestation with a different number of egg - mases per plant (*table* 2) it occured that the attack was not influenced by the infestation level, the attack values being similar regardless of the number of egg-mases used during the same infestation period. The differentiation of data in some cases by infestation date (vegetation phase) was caused by oscillating environment conditions from one infestation period to another.

As for experimental hybrids, it is worth mentioning that, for the hybrid Dekalb 512 Bt, in all cases, regardless of the year conditions, sowing and infestation period - i.e. vegetation phase of the plants, of the number of egg-mases used for infestation, no attack values were found in the length of cavities or larvae count, while for the hybrids F 376 and Olt the attack was obvious, with high values, greatly differentiated from that of Dekalb 512 Bt.

The same results showed the *figures 1 - 4*, which represent the mean data on the reaction of maize hybrids to the attack of ECB as depending on the number of days from sowing to infestation or the number of egg-masses/plant.

Analyzing the attack data on the behaviour of corn hybrids in the experiment depending on the vegetation phase of the plants, the same good behaviour of Dekalb 512 Bt was noted to the corn borer attack, even if pest attacked after the panicle had appeared, a phase which corresponds to the evolution of the second generation in the conditions of the USA (Guthrie, 1971) and, at a much smaller scale, to some extent, with the evolution of the partial second generation in southern Romania (Bărbulescu *et al.*, 1997).

The data obtain concerning the reaction of the two indigenous hybrids in the experiment and of the Dekalb 512 Bt to the attack of the corn borer showed very clearly that, although the indigenous hybrid that is considered tolerant HSF 1071-76 showed a lower attack degree compared with the susceptible indigenous hybrid HSF 1147-96, the genetically modified hybrid, Dekalb 512 Bt with the Bt gene included showed only sporadic traces of attack produced by *Ostrinia nubilalis* (*table 3*).

Table 1 - Reaction of corn hybrids to the ECB attack as depending on the sowing date and infestation date

Year	Date		Days	Cavity length cm/plant			Larvae count per plant		
	Sowed	Infested		Dekalb 512 Bt	F 376	Olt	Dekalb 512 Bt	F 376	Olt
1998	15.07	06.20	44	0	16.7	14.8	0	2.6	2.1
		06.26	50	0	12.8	13.5	0	1.6	1.5
		07.06	56	0	11.7	17.9	0	1.9	2.3
		07.16	66	0	14.5	17.2	0	1.5	1.3
	06.03	07.08	35	0.1	8.8	6.9	0	1.5	1.4
		07.20	47	0.1	13.6	15.0	0	2.2	2.1
1999	05.03	06.25	53	0	4.1	8.2	0	1.0	1.6
		07.05	63	0.1	8.8	9.5	0	1.8	2.1
		07.15	73	0	9.8	10.7	0	1.7	2.3
	06.01	07.19	48	0	9.9	13.3	0	3.0	2.7
		07.23	58	0	7.5	8.2	0	1.4	2.0

Table 2 - Reaction of corn hybrids to the attack of the ECB as depending on infestation date and number of egg-mases per plant

Date			Hybrid	Cavity lenght, cm/plant				Larva count per plant			
sowing	infestation	days S-I		Egg-mases per plant				Egg-mases per plant			
				5	**10**	**15**	**20**	**5**	**10**	**15**	**20**
05.07.1998	06.30	54	Dekalb 512 Bt	0	0	0	0.3	0	0	0	0.1
			F 376	6.1	5.6	4.7	5.9	1.0	1.6	0.5	1.2
			Olt	5.1	4.9	6.2	9.3	2.1	1.0	2.3	1.8
	07.07	61	Dekalb 512 Bt	0	0	0	0	0	0	0	0
			F 376	8.9	6.2	8.6	8.3	1.5	1.9	2.8	1.6
			Olt	10.2	10.9	7.5	9.3	3.1	3.4	1.9	1.8
	07.14	68	Dekalb 512 Bt	0.2	0	0	0	0	0	0	0
			F 376	17.2	16.9	16.1	22.2	1.9	5.1	5.4	6.4
			Olt	14.1	17.1	16.3	19.9	4.3	6.6	5.1	5.8
	07.21	75	Dekalb 512 Bt*	0	0	0	0	0	0	0	0
			F 376*	9.9	11.9	7.0	14.4	1.8	4.4	2.0	4.0
			Olt*	6.0	11.7	7.0	10.3	3.2	4.7	3.8	4.5
	07.28	82	Dekalb 512 Bt*	0	0	0	0	0	0	0	0
			F 376*	5.0	7.0	7.0	2.8	0.5	0.9	1.4	1.7
			Olt*	7.0	9.0	4.9	7.4	1.0	1.6	2.7	3.9
05.03.1999	06.24	52	Dekalb 512 Bt	0	0	0	0.1	0	0	0	0
			F 376	1.7	2.8	0.8	1.1	0.4	0.3	0.1	0.3
			Olt	3.5	2.6	2.0	2.0	0.7	0.5	0.5	0.2
06.01.1999	07.15	44	Dekalb 512 Bt	0	0	0	0.8	0	0	0	0.8
			F 376	14.4	13.6	9.2	16.0	2.4	2.2	1.8	2.5
			Olt	10.0	11.8	13.5	11.0	2.6	2.8	1.9	1.8

Infestation with pins, as the panicle had appeared

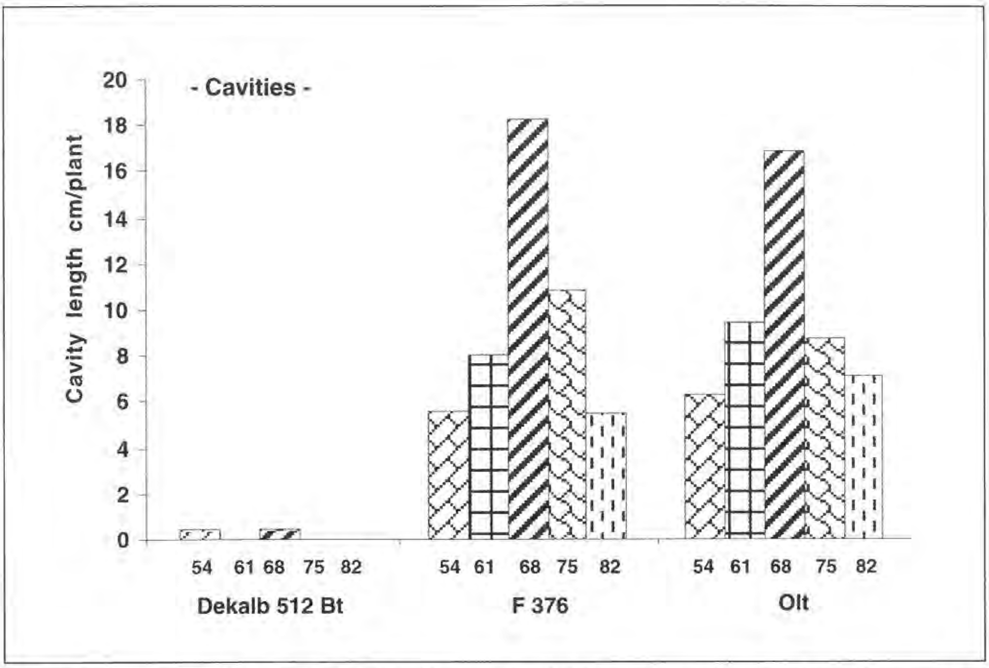

Figure 1 - Reaction of maize hybrids to the attack of ECB as depending on days from sowing to infestation (cavities)

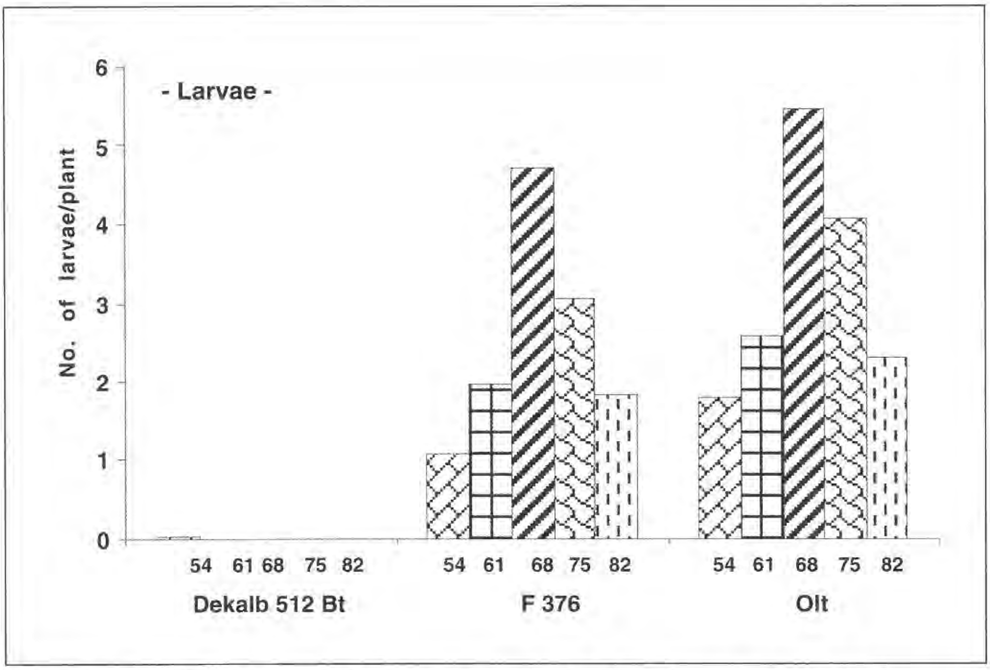

Figure 2 - Reaction of maize hybrids to the attack of ECB as depending on days from sowing to infestation (larvae)

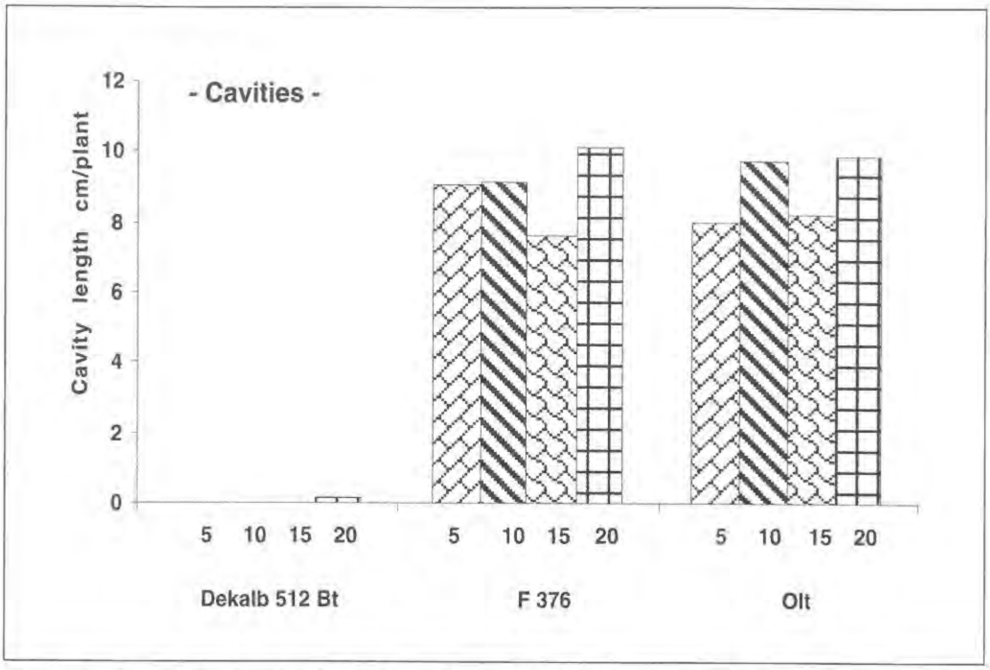

Figure 3 - Reaction of maize hybrids to the attack of ECB as depending on the number of egg-masses/plant (cavities)

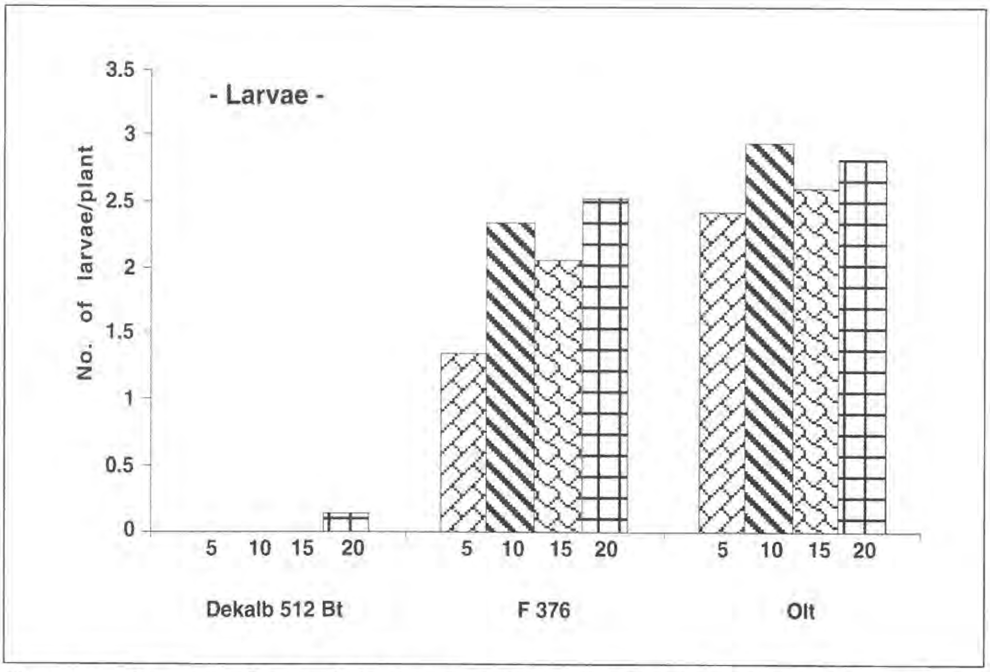

Figure 4 - Reaction of maize hybrids to the attack of ECB as depending on the number of egg-masses/plant (larvae)

Table 3 - Reaction of the hybrid Dekalb 512 Bt to the attack of the ECB, compared with a resistant hybrid and a susceptible one

Hybrid	Experiment	Cavity length, cm/plant	Larvae count per plant
Dekalb 512 Bt	1	0	0
	2	0	0
	3	0.2	0.4
	4	0.7	0.1
HSF 1071	1	0.7	0.2
	2	2.2	0.2
	3	1.0	0.2
	4	1.3	0.2
HSF 1147	1	9.0	2.3
	2	16.9	3.9
	3	20.2	3.9
	4	15.4	3.2

Conclusions

Although the attack of the corn borer during the experiment period was low, due to weather conditions that were unfavourable for the pest, the attack values recorded gave the opportunity to make a definite differentiation on the reaction of the test hybrids.

The hybrid Dekalb 512 Bt showed in all cases a significant resistance to the attack of *Ostrinia nubilalis,* the attack values showing only sporadic traces of attack by the pest.

The resistance of the hybrid Dekalb 512 Bt was satisfactory for the vegetation phase when the infestation was performed, including after the panicle had appeared, which corresponds to the period of evolution of the only generation that has economic significance in Romania.

Although the indigenous hybrid in the experiment HSF 1071-96 had a low level of attack, the genetically modified hybrid Dekalb 512 Bt was practically not attacked by the corn borer.

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MONITORING OF CORN BORERS RESISTANCE TO Bt-MAIZE IN SPAIN: FORECAST OF RESISTANCE

Pedro Castañera, Marta De la Poza, Gema P. Farinós, Pedro Hernández-Crespo, Félix Ortego, CSIC, CIB - Dpto Biología de Plantas, Velázquez 144, 28006 - Madrid -Spain

Abstract

Maize is the main summer cereal crop in Spain with a cultivated area of about 450,000 ha. The Mediterranean corn borer (MCB), *Sesamia nonagrioides* Léfèbvre, and the European corn borer (ECB), *Ostrinia nubilalis* (Hübner), are key pests in most of the Spanish maize growing regions.

The commercialisation of transgenic maize expressing the Cry1Ab toxin from *Bacillus thuringiensis* (Bt-maize) provides a new tool for an effective control of these two major maize pests. However, the development of resistance in target pests to Bt plants has been considered the main risk for the success of this control strategy. So far, field resistance to Bt-maize has not been documented, but it is expected that large-scale planting could result in rapid selection for resistance in field populations of corn borers.

A surface ranging from 20,000 to 25,000 ha of Bt-maize (event 176, cv. Compa CB, Syngenta) has been grown annually in Spain since 1998. Accordingly, a monitoring research project was established (funded by the Spanish Ministry of the Environment) to detect changes in susceptibility of corn borers through regular monitoring on Bt-maize fields.

Baseline susceptibility to the Cry1Ab toxin was determined for Spanish populations of MCB and ECB from larvae collected on non-transgenic maize in the most important growing areas (Galicia, Ebro, Madrid, Andalucía, Badajoz and Albacete). Annual monitoring of field populations of both species collected on Bt-maize in the same geographical areas has not revealed changes in susceptibility after three years of Bt-maize cultivation in Spain.

The expression of the toxin in Compa CB tissues is not maintained all season and, therefore, laboratory selection to induce resistance might be more relevant than in other cultivars where high expression of Bt toxin is maintained throughout the maize cycle. Laboratory selection for eight generations yielded ECB and MCB strains 5.0- and 2.2-fold less susceptible to Cry1Ab than unselected populations, respectively.

IMPACT OF Bt- MAIZE ON NON-TARGET ARTHROPODS IN CENTRAL SPAIN

Gema P. Farinós, Marta De la Poza, Pedro Hernández-Crespo,
Félix Ortego and Pedro Castañera,
CSIC, CIB, Dpto Biología de Plantas, Velázquez 144,
28006, Madrid, Spain

Abstract

The greater specificity of Cry1Ab toxin with respect to conventional insecticides to control corn borers, and the likely reduction of the latter under a transgenic crop system should result in a more favourable environment for the non target-arthropods in the crop. However, it has been reported from laboratory experiments that Bt-maize may have a negative impact on some natural enemies by a combined effect of Bt exposure and nutritional deficiency caused by a reduction in the quantity and quality of their food supply. Laboratory experiments are very useful to point out possible deleterious effects of Bt-maize, but they must be verified with field experiments.

In 2000 we have started a three years field-study to assess, in a commercial field (about 5 ha) near Madrid (Spain), the potential impact of Bt-maize on the abundance and diversity of non-target arthropods. Three treatments with three replicates (≈ 0.6 ha/plot) were arranged in a completely randomised block design: Bt-maize (cv. Compa CB) was compared with the isogenic cv. Dracma under conventional farm practices, with or without Imidacloprid treatment. Two sampling techniques (visual counts and pitfall traps) were used to compare their effects on non-target arthropods that comprise maize-based food webs from late June to the end of September.

Visual surveys reveal that *Orius* spp. and spiders were the most abundant predators on the maize plants. A common pattern of the frequency of *Orius* on plants was found in both years, with few specimens at the beginning of the summer and a big increase in September. Conversely, spiders were present in the plant all through the summer.

Ground beetles and spiders were the most abundant poliphagous predators found in pitfall traps in all treatments. Five carabid species accounted for 98% of the total beetles collected. Similarly, six species of spiders accounted for 97% of their total number.

No detrimental effects associated with the transgenic cultivars have been detected so far, although no firm conclusions can be drawn until these studies are completed.

THE PROJECT TO IMPLEMENT IPM STRATEGIES AGAINST *AGRIOTES* SPECIES IN EUROPE: WHAT HAS BEEN DONE AND WHAT IS STILL TO BE DONE

Lorenzo Furlan,
Department of Agronomy, Entomology, University of Padova,
via Romea 16, Agripolis, Legnaro PD;
phone: 0039 049 8272801; fax: 0039 049 8272810;
e-mail: ento@agripolis.unipd.it

Miklós Tóth,
Plant Protection Institute, Hungarian Academy of Sciences,
Budapest, Pf 102, H-1525 Hungary

Venyamin G.Yatsynin,
Krasnodar Lukyanenko Research Institute of Agriculture,
Krasnodar 12, 350012, Russia

István Ujváry,
Central Chemistry Institute, Hungary, Academy of Sciences,
Budapest, Pusztaszeri ut 59/67, H-1025, Hungary

Abstract

A specific project to develop effective *Agriotes* sex pheromone traps was begun in 1995. It has been developed through the following phases: 1) Collection of available information on: a) the most important *Agriotes* species that damage maize in Europe; b) the pheromones of these species; 2) Rearing of the species for which no or not enough information was available (*Agriotes brevis*, *Agriotes sordidus*, *Agriotes lineatus*) in order to obtain sufficient numbers of experimental insects to study; 3) Extraction of the pheromones from female's gland; gas chromatography and mass spectrometry analyses to characterise them; 4) Synthesis of the components identified; 5) Field optimisation (components ratio, dosage, dispenser type, etc.) of baits prepared from synthetic pheromone components; 6) Development of a trap model suitable for catching the different species; 7) Evaluation of the effectiveness of the traps in different areas with different populations. Phases 5 to 7 were sometimes carried out concurrently. To ensure rapid development of new materials, protocols were often modified during the season in the light of results obtained. After seven years work, a non-saturable trap and different lures suitable for monitoring all the most important European *Agriotes* species (*A. brevis* Candeze, *A. lineatus* L., *A. litigiosus* Rossi, *A. obscurus* L., *A. rufipalpis* Brullé, *A. sordidus* Illiger, *A. sputator* L., *A. ustulatus* Schäller) have been made available for all the European countries. A description of the main features of the new materials is given.

The cost to realize the phases one to six was over 300,000 euro, not including overheads and the amortization of the facilities used. Finally what has to be done to complete the work is discussed.

Key words: *Agriotes*, sex pheromone traps, Europe.

Introduction

In Europe many farmers apply soil insecticides to maize at planting without any evaluation of the actual presence of an economically damaging wireworm population. Effective, easy and inexpensive tools are needed to identify the fields that have high wireworm populations in order to ensure that treatments are only applied where necessary or to enable non-infested fields to be avoided. Pheromone traps might represent a solution as they monitor the only stage that lives outside the soil: the adults (Furlan *et al.*, 1996; Furlan *et al.*, 1997).

A specific project to develop effective *Agriotes* sex pheromone traps was begun in 1995. It has been developed through the following phases.

1) Collection of available information on:

a) the most important Agriotes species that damage maize in Europe

Little information is available about the *Agriotes* species really damaging crops; for several countries no information was found about larvae damaging crops; sometimes no information about adult presence was discovered. Studies for all Europe concerning larval presence and dangerousness are not available, since most of the few papers dealing with this issue generically refer to wireworms. Anyway, the publications collected allowed us to identify which species were likely to be the key species from an agricultural point of view; for example *A. brevis* Candeze can be considered the main pest to maize and other crops in Italy (Furlan, 1999; Furlan *et al.*, 2000, Rusek, 1972a, 1972b) but also in the Czech Republic and other European countries it must be considered a key species (Rusek, 1972a). According to observation of adult presence *A. brevis* is widespread in central and southern Europe (Platia, 1994).

Agriotes sordidus is certainly important too. It appears to be a very important species in Italy (Furlan, 1999; Furlan *et al.*, 2000, Rusek, 1972a) but also in Spain and France where larvae collected in heavily damaged fields belonged mainly to this species (Furlan, unpublished observation). The species should be also important in Portugal where it was first described (Platia, 1994). No doubt about the importance of *Agriotes ustulatus*, *Agriotes litigiosus*, *Agriotes lineatus*, *A. obscurus* and *A. sputator* in several European countries (Dolin, 1964; Furlan, 1999; Furlan *et al.*, 2000; Kosmacejsky, 1955; Rambousek, 1929; Platia, 1994), which are also reported as damaging to plants in many entomological textbooks). *A. rufipalpis* should be relevant in Eastern European countries (Dolin, 1964). Finally the following list about the most important *Agriotes* species on which research efforts had to be concentrated was put together: *A. brevis* Candeze, *A. lineatus* L., *A. liti-*

giosus Rossi, *A. obscurus* L., *A. rufipalpis* Brullé, *A. sordidus* Illiger, *A. sputator* L., and *A. ustulatus* Schäller.

b) the pheromones of these species

Information about the main components of the sex pheromones was already available in the following spp.:

Agriotes ustulatus: main component E,E - farnesyl acetate (Yatsynin, 1984, Kudryavtsev *et al.*, 1993);

Agriotes litigiosus: geranyl isovalerate (Yatsynin *et al.*, 1980);

Agriotes sputator: geranyl butanoate (Kudryatsev *et al.*, 1993; Yatsynin *et al.*, 1986);

Agriotes obscurus: geranyl hexanoate + geranyl octanoate (Borg-Karlson *et al.*, 1988);

Agriotes lineatus: geranyl octanoate + geranyl butyrate (Yatsynin *et. al.*, 1980, 1991, 1996).

2) Rearing of the species for which no or not enough information was available (*Agriotes brevis, Agriotes sordidus, Agriotes lineatus*) in order to obtain sufficient numbers of experimental insects to study:

Adult *A. brevis, A. sordidus* and *A. lineatus* specimens were collected from the rearings maintained at the San Donà laboratory according to the methods described (Furlan, 1998). The rearing in cages and vials started from females or larvae collected in Piemonte (north-western Italy), Veneto (north-eastern Italy), Lazio (central Italy) and Molise (southern Italy). The females obtained in overwintering cells were kept isolated in single vials with humid soil and some rye grass leaves until pheromone extraction was performed at the time when the males' swarming was observed in the field.

3) Extraction of the pheromones from females' glands; gas chromatographic and mass spectrometric analyses to characterise them:

Alive insects were taken to the Budapest laboratory. Female sex pheromone gland extracts were prepared as described by Russian authors (Ivashchenko and Adamenko, 1980; Oleschenko *et al.*, 1976), by carefully piercing the pheromone gland by a fine glass capillary and collecting the liquid inside into the capillary. The liquid samples obtained were dissolved in hexane (a.g., MERCK KGaA, Darmstadt, Germany) to make up stock solutions. Extracts prepared were analysed by capillary gas chromatography and coupled gas chromatography/mass spectrometry using several column types, i.e. SP 2340 fused silica (Supelco Inc., Bellefonte, USA); HP Ultra 1 crosslinked methyl silicone gum phase (Hewlett-Packard, Palo Alto, USA); FS capillary column coated with FFAP (Macherey & Nagel, Düren, Germany).

Results

The following components were identified for the species studied:

A. sordidus: geranyl hexanoate (the same ompound can attract *A. rufipalpis* males in the field (Tóth *et al.*, 2001);

Agriotes lineatus: geranyl octanoate + geranyl butyrate

Agriotes brevis: geranyl butyrate + *E,E* farnesyl butyrate (Tóth *et al.*, 2002).

Table 1 - *Agriotes* species captured using different compounds in different European countries in 1998 - 2001 (in some regions monitoring was done only for 2-3 years). Species indicated with bold letters: the bait proved to be clearly effective in capturing the species (at least 50 males captured per trap per year). Smaller letters mean that the bait showed some activity but the captures were not conspicuous.

	geranyl-isovalerate	E,E farnesyl acetate	geranyl-hexanoate	geranyl butanoate	geranyl octanoate + geranyl butyrate	geranyl butyrate + E,E farnesyl butyrate	geranyl hexanoate + geranyl octanoate
AUSTRIA	A. litigiosus	A ustulatus	A. rufipalpis	***A.sputator***	***A. lineatus***	***A. brevis***	NOTHING
BULGARIA	NOTHING	A.ustulatus	Cidnopus pilosus	A.sputator	***A. lineatus***	***A. brevis***	NOTHING
CROATIA	A. ustulatus	***A. ustulatus***	***A. rufipalpis***	***A. sputator***	***A. lineatus***	A. sputator	***A. obscurus***
GERMANY	NOTHING	***A. ustulatus***	NOTHING	A.sputator	***A. lineatus***	A. brevis	A. obscurus
GREECE	***A. litigiosus***	NOTHING	***A. rufipalpis***	NOTHING	A. lineatus	NOTHING	***A. rufipalpis***
HUNGARY Central	NOTHING	***A. ustulatus***	***A. rufipalpis***	***A. sputator***	***A. lineatus***	A. sputator	NOTHING
HUNGARY East	NOTHING	***A. ustulatus***	***A. rufipalpis***	***A. sputator***	***A. lineatus***	A sputator	not tested
ITALY North-east	***A. litigiosus***	***A. ustulatus***	***A. sordidus***	NOTHING	NOTHING	***A. brevis***	***A.sordidus***
ITALY North-west	***A. litigiosus***	NOTHING	***A. sordidus***	NOTHING	***A. lineatus***	A. acuminatus	***A.sordidus***
ITALY central	***A. litigiosus***	NOTHING	***A.sordidus***	NOTHING	A. lineatus	***A. brevis***	***A.sordidus***
ITALY South	***A. litigiosus***	NOTHING	***A. sordidus***	A. acuminatus	***A. lineatus***	***A. brevis***	***A.sordidus***
PORTUGAL	NOTHING	NOTHING	NOTHING	***A. proximus***	A. proximus	***A. proximus***	not tested
ROMANIA	NOTHING	A. ustulatus	A. rufipalpis	A. sputator	***A. lineatus***	A. sputator	***A. obscurus***
SLOVENIA	NOTHING	***A. ustulatus***	NOTHING	***A. sputator***	***A. lineatus***	***A. brevis***	***A. obscurus***
SWITZERLAND Ticino	***A. litigiosus***	NOTHING	NOTHING	***A. sputator***	A. lineatus	***A. brevis***	***A. obscurus***
SWITZERLAND Zurich	NOTHING	A. ustulatus	***A. gallicus***	***A. sputator***	***A. lineatus***	not tested	***A. obscurus***
UNITED KINGDOM	NOTHING	NOTHING	NOTHING	***A. sputator***	***A. lineatus***	not tested	***A. obscurus***
YUGOSLAVIA	not tested	***A. ustulatus***	not tested	A. sputator	not tested	not tested	not tested

4) Synthesis of components identified

All the synthesis were carried out at the Budapest laboratories. Syntheses usually involved esterification of geraniol and farnesol by standard methods.

5) Field optimisation (components ratio, dosage, dispenser type, etc.) of baits prepared from synthetic pheromone components

For all the species trials were carried out in fields where the presence of conspicuous larval populations had been ascertained by using bait traps (Chabert and Blot, 1992) and/or soil sampling in the previous year (species overwintering as adults) or in the same spring (species non overwintering as adults) following a common protocol. Traps were set up in blocks. Each block was a group placed out in a circle and comprised of one of each treatment (different ratio, dispensers, dosages and respective combinations). The distance of traps within a block was 5-10 m. The distance between blocks ranged between 25 - 100 m. Traps were moved one position forward within a block at each occasion when the traps were inspected; at the same time, captured beetles were recorded and removed. The inspections were made at least once per week but usually 2-3 times per week from the beginning of the adult emergence for 2-3 months.

Results

The most effective dispensers were 0.7 ml polyethylene vials with lid (No. 730, Kartell Co., Italy). After the first trials all the subsequent ones were made using these vials.

For all the species the biological active compounds, the most effective ratio between them and dosage were established.

6) Development of a trap model suitable for catching the different species
Different traps were devoloped during these studies (Tóth *et al.*, 1997, 1998). **Bottle** traps were funnel traps home-made from 2 litre transparent plastic bottles, by cutting the bottle below the neck (ca 10 cm), and placing the cut part upside down into the remaining bottom of the bottle. The resulting funnel entrance diameter was ca 8 cm; funnel hole diameter was ca 2 cm. A transparent plastic sheet (10 x 10 cm) was placed in a vertical position immediately above the opening of the funnel. The bait dispenser was attached to the vertical plastic sheet so that the attractant-containing part was hanging into the large opening of the funnel. **VARb** funnel traps were the plastic CSALOMON® VAR funnel traps produced by the Plant Protection Institute, Budapest (Hungary), except that a transparent plastic sheet (23x10 cm) was placed in a vertical position immediately above the opening of the funnel. A bait dispenser was attached to the vertical plastic sheet so that the attractant-containing part was hanging into the opening of the funnel. Funnel entrance was 13 cm (i.d.), hole of funnel was 3 cm (i.d.), height of funnel was 10 cm. Beetles falling down through the funnel were caught in a ca 1 litre plastic container. The **TAL** traps were modified pitfall traps. They consisted of a brick shaped plastic box, a roof made of a folded plastic sheet in which the hanging bait was fixed and two other sheets on both sides of the plastic box which provided smooth surface for beetles entering the trap and leading them into the plastic box. At the bottom of the box a 10x16 cm sticky sheet was placed to make the escape harder from the trap. Dimensions of the plastic box were: length 17 cm, width 11 cm, and height 6 cm.

The **YATLOR** traps were made of plastic at the Italian laboratory and were similar in shape and size to the "Estron" trap described earlier (Oleschenko *et al.*, 1987, Kudryavtsev *et al.*, 1993). They were modified to prevent adults from escaping. **YATLORfunnel** traps were made of a bottom part like YATLOR and an upper part resembling the Bottle trap. With TAL, YATLOR designs insects could get in without flying ("crawl-in" traps), into BOTTLE, VARb traps only by flying ("fly-in" traps). The latter proved to be completely unsuitable for catching some species (*A. brevis*, *A. obscurus*) while the "crawl-in" traps proved to be much less effective in catching the flying species. YATLORfunnel traps proved to be effective in monitoring all the species along all the season.

7) Evaluation of the effectiveness of the traps in different areas with different populations
This was done in several European countries. First data were presented at XX IWGO Conference in Adana (Furlan *et al.*, 1999) and other specific presentations have just been given at XXI IWGO Conference in Italy. An overview of the potential of the new baits in Europe is given in Table 1 where data collected over last four years are summarized. The suitability of the new traps for monitoring the populations of the most important *Agriotes* species cited above is clear; some compounds proved to be suitable for monitoring other species. Their importance from an agricultural point of view is under evaluation.

Table 2 - The project to implement ipm strategies against *Agriotes* costs (euro) (assumptions: 1 h of manpower=15 Euros; average car cost: 0.4 E/km)

Rearings			
	materials		7,500
	manpower (400 h/y x 5 years)		30,000
Laboratory extractions and analyses			
	material, chemical analyses		18,000
	trips and accomodation (Italy – Hungary;Russia - Hungary – Russia - Italy)		10,000
Materials for cap preparation			8,000
Field tests for optimizing component ratio and dosages			
	materials		10,000
	manpower	Italy	48,500
		Hungary	15,000
	car movements		
	(12,000 km/y x 7 y x 0.4 E/km)		34,000
	(8,000 km/y x 7 y x 0.4 E/km)		22,000
Traps development			
	silicone templates		5,000
	iron templates		34,000
	manpower		35,000
	trap preparation		9,000
	car movements (8,000 km/y x 5y)		16,000
		TOTAL	**302,000**
SPONSORS			
Copagri - Venice Province			25,000
Centro Quadrifoglio			15,000
Consorzio Regionale Molisano di Difesa			4,000
University of Padua			20,000
Enea			2.000
Institute of Agronomic Research of Zurig			5,000
RO-SA Micromeccanica			38,000
Piemonte Region			10,000
Plant Protection Institute of Budapest			46,000
Furlan's salary and savings			75,000
Furlan's work			62,000
		TOTAL	**302,000**

Conclusions

After seven years of work, a "high capacity" trap and different lures suitable for monitoring all the most important European *Agriotes* species (*A. brevis, A. lineatus, A. litigiosus, A. obscurus, A. rufipalpis, A. sordidus, A. sputator, A. ustulatus*) have been made available for all the European countries.

The cost to realize the phases one to six was over 300,000 euro not including overheads and amortization of the facilities used (*table 2*).

What has to be done to complete the work:

- to establish the biological significance of the pheromone trap catches; to determine the actual range of attractiveness, and to study the relationship between males captured and level of the female population.
- to establish a reliable correlation between adult trap catches and subsequent larval populations for all the species and varieties in different climatic and agronomic (mainly rotation) conditions.

For both these aspects, studies have already started and the first encouraging data have been obtained. Replications in many different conditions are needed to meet the practical requirements of the effective implementation of an IPM strategy.

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FIRST RESULTS OF CLICK BEETLE TRAPPING WITH PHEROMONE TRAPS IN HUNGARY 1998 - 2000

Miklós Tóth, Zoltán Imrei,
Plant Protection Institute, Hungarian Academy of Sciences,
Budapest, Pf. 102, H-1525, Hungary;
phone: +36-1-3918639; fax: +36-1-3918555;
e-mail: h2371tot@ella.hu

István Szarukán, Róbert Körösi,
Agricultural University, Debrecen, Pf. 58, H-4001, Hungary

Lorenzo Furlan,
Department of Agronomy, Entomology, University of Padova,
Via Romea 16, Agripolis, Legnaro, I-35020 Italy

Abstract

Pheromone traps for *Agriotes* click beetle pests were operated at several sites in Hungary in 1998 - 2000 with the principal aim of determining the key pest species and to obtain information on their seasonal occurrence.

During the tests at all sites regular catches of *A. sputator, A. lineatus, A. rufipalpis* and *A. ustulatus* were observed in traps baited with the respective baits. Catches in these traps were highly selective; only in traps baited with the *A. lineatus* bait were sporadic catches of *A. sputator* observed. In the course of our trappings seasonal occurrence of all four species captured in greater numbers could be readily followed by the pheromone traps.

No catches were observed in traps baited with *A. litigosus* or *A. obscurus* baits. In traps baited with *A. brevis* baits all specimens captured proved to be *A. sputator*, evidently coming to the one component of the *brevis* bait.

The results suggest that the most abundant and economically important click beetle species in Hungary include *A. sputator, A. lineatus, A. rufipalpis* and *A. ustulatus*. From these the importance of *A. sputator, A. lineatus* and *A. ustulatus* has been realized in earlier plant protection literature. *A. rufipalpis*, although known to be present in Hungary, so far has not been thought to be an important pest. Based on our results its pest status should be reconsidered.

The conspicuous absence of *A. obscurus* from the present trappings is highly interesting as this species was listed as one of the most abundant pest elaterid in earlier literature. The cause of this discrepancy has to be the subject of further studies.

In soil samplings performed at Debrecen in 1999 and 2000 at the sites of the trappings no or negligible numbers of click beetle larvae were found, suggesting that numbers trapped in the present study still do not represent an economically important population level.

Introduction

Pheromone traps for *Agriotes* click beetle pests were operated at several sites in Hungary in 1998 - 2000 with the principal aim of determining the key pest species and to obtain information on their seasonal occurrence.

Materials and methods

Trials were carried out from 1998 to 2000; in the first 2 years YATLOR and VARb traps were used while in 2000 a trap design suitable for both flying and crawling species (YATLORfunnel) was also used. The traps were baited with the lures for *A. lineatus, A. obscurus, A. sputator, A. sordidus, A. brevis, A.litigiosus* and *A.ustulatus*. The areas of the regions where the experimentation was carried out have the following characteristics:

Agárd, Fejér county; weedy edge of fields; latitude: 18°42', 48°51'; ph: 7.3, kind of soil: brown chernozyom, yearly average temperature: 11.1°C (1998); 11.2°C (1999); 12.2°C (2000), total rainfall: 697 mm (1998); 795 mm (1999); 403 mm (2000), common cultures: maize, sunflower, wheat, rape, sugarbeet, alfalfa.

Tedej-Hajdúnánás, Hajdú-Bihar county; alfalfa fields; latitude: 21°30', 48°29'; ph 7.3, kind of soil: brown chernozyom; yearly average temperature 9.8°C; average rainfall 556 mm, common cultures: wheat, maize, sugarbeet, alfalfa.

Only in 1999 a test was run also at Martonvásár (ca 20 km distance to the Agárd site, Fejér county, 18°52', 48°23', ph: 7.3, kind of soil: brown chernozyom, yearly average temperature: 11.1°C (1998); 11.2°C (1999); 12.2°C (2000), total rainfall: 697 mm (1998); 795 mm (1999); 403 mm (2000), common cultures: maize, sunflower, wheat, rape, sugarbeet, alfalfa.

At each site two traps of each bait type were operated.

Results and discussion

In Hungary at all the sites 4 *Agriotes* spp. were captured in large numbers in the course of the tests with pheromone baited traps, each by its respective pheromone composition. These species were *A. lineatus, A. rufipalpis, A. sputator* and *A. ustulatus*.

Traps baited with the pheromone composition for *A. litigiosus* did not capture any species. Traps baited with the pheromone composition for *A. brevis* caugth only lower numbers of *A. sputator*, evidently being attracted to one component (geranyl butyrate) of the *brevis* bait. Traps baited with the pheromone composition of *A. obscurus* did not catch any *A. obscurus* in the above tests.

Sporadic catches of both *A. lineatus* and *A. rufipalpis* were observed, in both cases due to the presence of the respective main pheromone component of these two spp. (geranyl octanoate for *A. lineatus*, and geranyl hexanoate for *A. rufipalpis*, resp.).

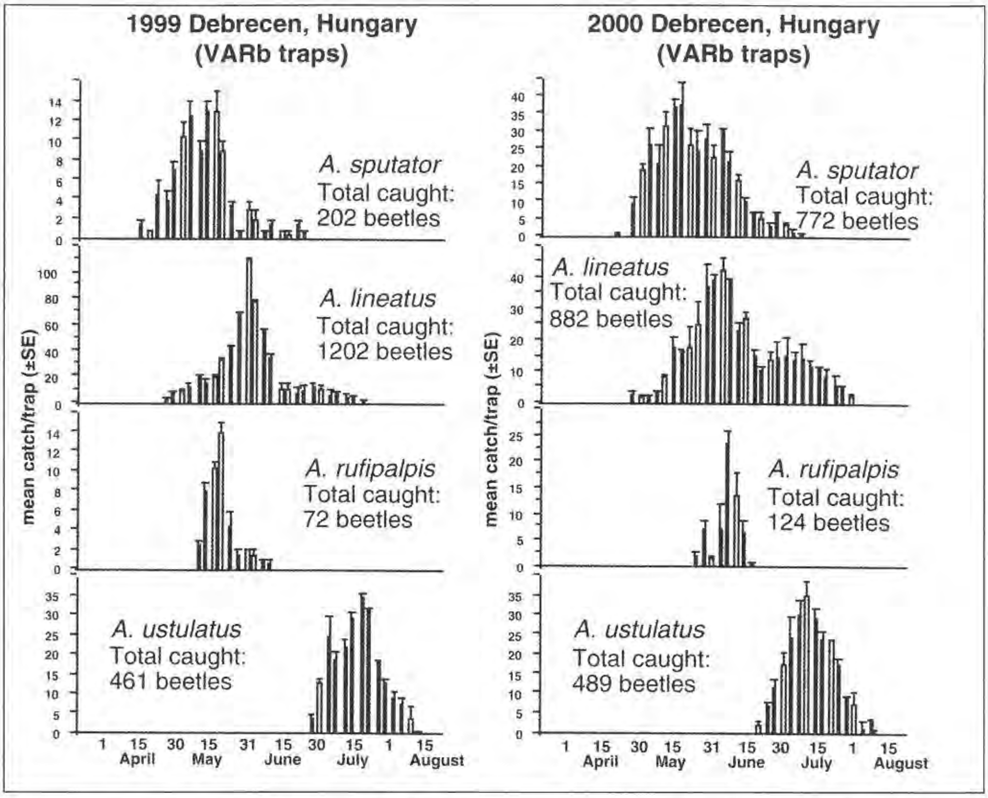

Figure 1 - Flight dynamics of four Agriotes spp. captured in pheromone traps in Hungary at Debrecen, 1999 and 2000. Columns show mean(±SE) catches of two traps

It has to be mentioned that in previous preliminary tests at other sites in central Hungary including baits with geranyl octanoate and hexanoate, some specimens of *A. obscurus* were recorded by us; suggesting that the species is present, but not in very large numbers.

The absence of *A. obscurus* from trap captures was quite surprising, as in previous literature this species is listed among the most abundant and economically most important click beetle pests in Hungary (Tóth, 1990).

On the other hand *A. rufipalpis*, although mentioned to be present in the fauna, was not thought to be an important pest in Hungary. Based on its regular appearance and relatively large numbers caught in the present trappings, its pest status may have to be reevaluated.

The remaining two spp., *A. lineatus* and *A. ustulatus* are listed as important click beetle pests also in previous literature; our trapping data support this notion.

Conspicuous differences in flight dynamics of the four spp. could be observed by pheromone trap captures (*figure 1*).

In each year the very first species to appear was *A. sputator*. First catches were recorded already at the end of March, early days of April. The flight was going on at a high intensity during all April and May, prolonging at lower intensity to the middle of summer.

A. lineatus was recorded almost as early as the previous species, and its flight was also very long, reaching over several months. Usually the peak flight is somewhat later than that of *A. sputator*.

A. rufipalpis started to fly only at the beginning of May. Its flight was never so prolonged as in the previous two spp., usually it stopped after ca 5 - 6 weeks.

The flight of *A. ustulatus* took place in the summer, starting in late June/early July. Usually the flight is not long; sizeable catches can be recorded for a period of not more than 4 - 5 weeks.

Pheromone traps proved to be excellent when studying differences of flight patterns between sites (*figure 2*).

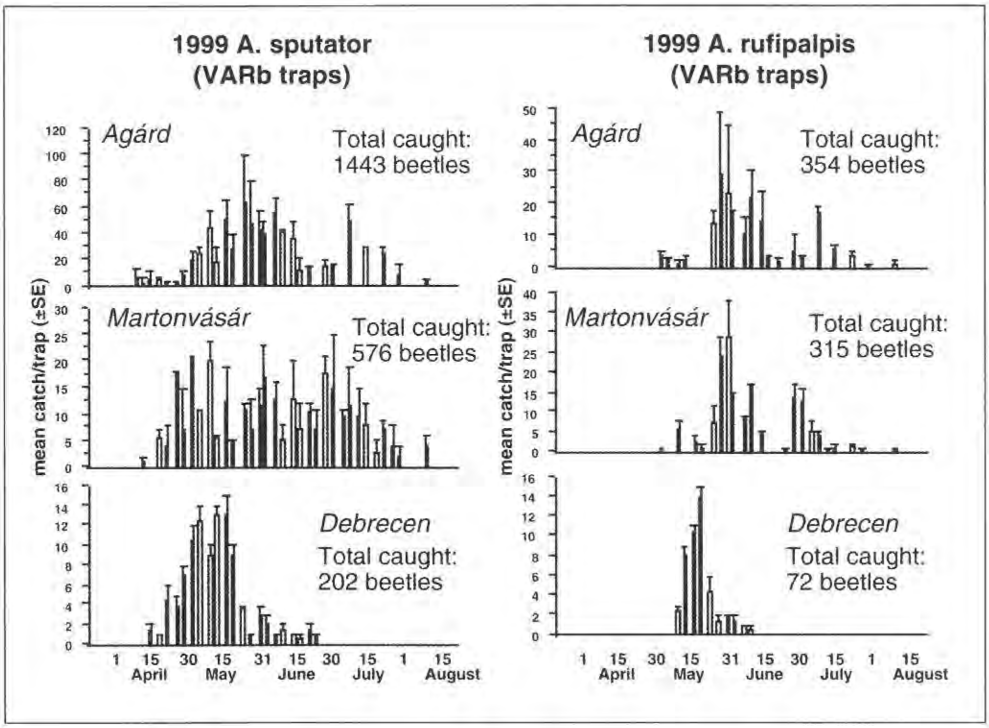

Figure 2 - Flight dynamics of Agriotes sputator and A. rufipalpis captured in pheromone traps at three sites in Hungary in 1999. Columns show mean(±SE) catches of two traps

As an example, in 1999 *A. rufipalpis* had a somewhat earlier flight peak at Debrecen (where weather was hotter and drier in 1999) than at the two central sites (*figure 2*). Similar tendency is shown in *A. sputator*, although here the difference is not so clearcut (*figure 2*). In this species however, the flight terminated much more abruptly at Debrecen, probably due to the draught in that locality.

Yearly differences between flight pattern can also be clearly demonstrated by pheromone trap captures (*figure 3*). *A. rufipalpis* started to fly much earlier in 1999 and 2001 at Debrecen, than in 2000 (*figure 3*). Also, populations increased dramatically by 2001 at this site.

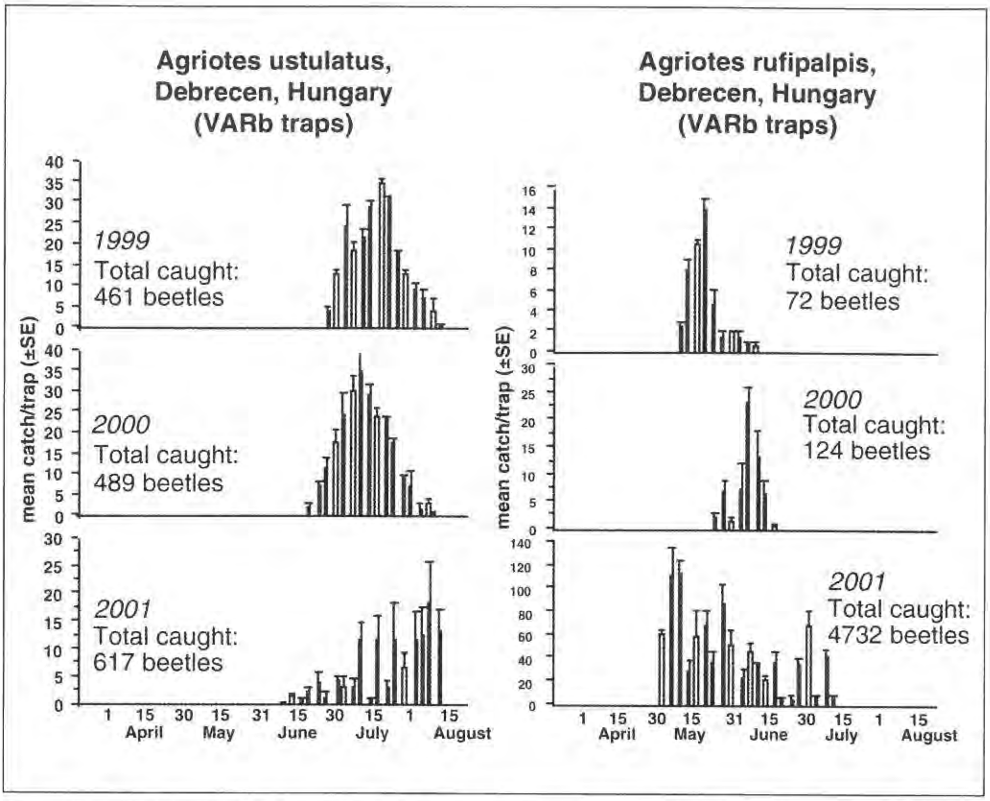

Figure 3 - Yearly flight dynamics of Agriotes ustulatus captured in pheromone traps at Debrecen between 1999 - 2001. Columns show mean(±SE) catches of two traps

In *A. ustulatus*, the appearance of the first beetles was recorded earlier in each year between 1999 - 2001 (*figure 3*). In 2001, the usually clear and sharp flight peak was depressed probably due to sudden cold spells in late June - early July.

We conclude that click beetle pheromone traps can be used for all the conventional, qualitative aims of insect population monitoring. Trials are underway to study their performance in quantitative monitoring objectives.

In preliminary soil samplings performed at Debrecen in 1999 and 2000 at the sites of the trappings no or negligible numbers of click beetle larvae were found, suggesting that numbers trapped in the present study still did not represent an economically important population level at the sites where the tests were performed.

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SEASONAL FLUCTUATION OF ADULT AND LARVAE OF *AGRIOTES SPP.* (COLEOPTERA: ELATERIDAE) IN CENTRAL GREECE

K. Karabatsas, V. Tsakiris, K. Zarpas, J. A. Tsitsipis,
Laboratory of Entomology and Agricultural Zoology,
University of Thessaly, Fytokou Str. 38446, Nea Ionia, Magnesia, Greece

L. Furlan,
Department of Agronomy, Entomology, University of Padova,
via Romea 16, 35020 Legnaro PD, Italy

M. Tóth,
Hungarian Academy of Sciences,
Budapest, Pf. 102, H-1525, Hungary

Abstract

The seasonal abundance of adults and larvae of seven wireworm species (Coleoptera: Elateridae) was examined in Velestino, Magnesia, Central Greece (latitude 39°22′, yearly average rainfall 450 mm, yearly average temperature 14.37° C) during years 1998-2001. Sex pheromone (YATLOR (1999, 2000), YATLORfunnel (2001) and VARb (1998, 1999, 2000)) and soil bait traps were used and soil samplings were made. Different combinations of the various types of traps were installed each year in the experimental field. Sex pheromone traps operated from late March to September. Soil bait traps were installed for 15 days in April (covered soil), October and November (bare soil), respectively. Soil samplings were performed in April and October-November every year at a depth of 30 cm. The species *Agriotes rufipalpis* Brullè and *Agriotes litigiosus* Rossi were examined in all four years, while *Agriotes lineatus* L., in 1998 and 1999, *Agriotes sputator* L. and *Agriotes ustulatus* Schaller in 1998, *Agriotes brevis* Càndeze in 1999 and *Agriotes obscurus* L. in 2000.

The swarming period of *A. rufipalpis* and *A. litigiosus* lasted from late March to mid September and from early May to early July with two peaks in mid April and early June and one peak in late May, respectively in all four years. In the first year 1998, 284.5 and 278.5 adults per pheromone trap of *A. rufipalpis* and *A. litigiosus* were captured, respectively. Adults of *A. sputator* and *A. ustulatus* were not caught and only five adults of *A. lineatus* were captured. In 1999, 660.5 and 256.5 adults per pheromone trap of *A. rufipalpis* and *A. litigiosus* were captured, respectively. Adults of *A. brevis* were not caught and only three adults of *A. lineatus* were captured. During 2000, 2443.8 and 248.7 adults per trap of *A. rufipalpis* and *A. litigiosus* were captured, respectively. No *A. obscurus* insects were caught in the pheromone traps. In 2001, 6721.8 adults of *A. rufipalpis* and 218.8 adults of *A. litigiosus* were captured. While in 1998 one larvae of *Melanotus* sp. was caught out of 30 traps/month in May. In 1999, no larvae were found in both soil bait traps and soil sampling. In 2000, larvae of *A. rufipalpis* only were captured in soil bait traps, three, six and eight all out of 33 traps/month, in April, October, and November, respectively. In March 2001, 12 larvae *A. rufipalpis* and one *Melanotus sp.* were found out of 44 soil bait traps. In soil samples, six larvae *A. rufipalpis* were found, two in May 1998, two in November 2000 and two in April 2001 out of 30, 33 and 44 samples respectively.

Key words: Fluctuation, *Agriotes*, Central Greece, captures.

Introduction

Larvae of *Agriotes* species are pests of several herbaceous crops: mainly corn, small grains, potato, sugarbeets, tobacco, cotton, strawberry, sunflower (Stamopoulos, 1995). The larvae feed on germinating seeds, roots and hypocotyls. Sometimes wireworm infestations can cause a substantial loss and resowing of the crop can be required (Furlan, 1989, 1990).

The present knowledge on *Agriotes* species in Greece is very limited and there is data available on key species, their seasonal occurrence and economic importance in few bibliographic resources (Pelekasis, 1962; Dalianis, 1977; Tzanakakis, 1980; Tolis, 1986; Stamopoulos, 1995; Tsitsipis, 1999).

The present study aimed at: (1) evaluating the seasonal abundance of adults and larvae of *Agriotes* species in Central Greece, (2) identifying the key species in this region and (3) assessing the effectiveness of new sex pheromone traps. The whole study has a particular interest since it is the first one on the seasonal fluctuation of this genus in Greece.

To meet these objectives, a common protocol with other European countries was adopted.

Materials and methods

The experimental site was located in Velestino, Magnesia, Central Greece (latitude 39°22', yearly average rainfall 450 mm, yearly average temperature 14.37°C). In the first two years 1998 and 1999 it consisted of one 1-ha field. In 2000 there were two fields (2.5 and 1.1 ha) and in 2001 four (1.1, 1.7, 0.9 and 1.6 ha). The first year, part of the field was cultivated with maize and sugarbeets while the main crop in the area was wheat. In the following years the experimental fields were cultivated exclusively with wheat. In all fields no soil insecticides, fertilizers or herbicides were applied.

The Plant Protection Institute of Budapest supplied pheromone caps for the following species containing the following chemical compounds:
Agriotes rufipalpis Brullè: geranyl-hexanoate,
Agriotes sputator L.: geranyl-butanoate,
Agriotes litigiosus Rossi: geranyl isovalerate,

Agriotes obscurus L.: geranyl-hexanoate + geranyl octanoate,
Agriotes ustulatus Schaller: E,E-farnesyl acetate,
Agriotes brevis Càndeze: geranyl-butyrate + E,E-farnesyl butyrate,
Agriotes lineatus L.: geranyl octanoate + geranyl butyrate.

The species *A. rufipalpis* and *A. litigiosus* were examined in all four years, while *A. lineatus* in 1998 and 1999, *A. sputator* and *A. ustulatus* in 1998, *A. brevis* in 1999 and *A. obscurus* in 2000.

In the first year only VARb traps were used, 10 for five species, in 1999 six VARb and two YATLOR (soil level) traps, for four species, 2000 six VARb and four YATLOR traps, for three species and in the last year 2001 eight YATLORfunnel traps, for two species. The pheromone dispensers were replaced every 30 days and the traps were inspected twice a week. All the specimens were removed at every inspection. The monitoring period lasted from late March to late October in all four years.

Two methods were used for estimating the larval population: soil bait traps and soil samplings. Laboratory studies have suggested that wireworms are attracted to germinating seeds by respired carbon dioxide (Doane *et al.*, 1975). Baiting is a relative sampling method that reflects both wireworm density and behavior (Williams *et al.*, 1992). Soil samplings provide estimates per unit surface area of the field. Although this information is useful, it is often very expensive and not practical for farmers or consultants to use (Simmons *et al.*, 1998).

The soil bait traps were installed for 15 days in three trapping periods. In April with covered soil, in October and in November with bare soil, respectively. A number of 30, 30, 33 and 45 bait traps per trapping period were installed in the years from 1998 to 2001. All traps were made just as Chambert and Blot (1992) have described. They were manually observed and the larvae found were counted.

The soil samples were taken near the soil bait traps (1-2m) at a depth of 30 cm in two periods, in April and in October-November. The number of samples per sampling period in these four years was identical to the number of bait traps: 30, 30, 33, 45. The samples were a soil cylinder 30 cm(H) x11cm (D) taken using a manual soil sampler and were put to dry into funnels for at least 30 days. The larvae were collected in vials placed in the bottom of the funnels and counted.

All the adults and larvae specimens collected by using the methods described above were sent to the Institute of Agricultural Entomology of Padova for identification.

Results

In the first graph (*figure 1*) it is clearly shown that the captures of adult males of *A. rufipalpis*, in these four years presented only little differences referring to the first capture and great differences in the mean number of adult males captured. The swarming period lasted from late March (probably earlier since the traps started capturing high numbers just after they had been placed out) to mid September and presented two peaks, one in mid April and another one in early June.

For *A. litigiosus*, the captures of adult males during 1998-2001 (*figure* 2) were more similar presenting only one peak in late May in all four years. The swarming period lasted from early May to early July and the mean number of adult males captured was almost constant.

The traps for *A. lineatus* installed only in the first two years 1998 and 1999, captured only five and three adult males respectively in total from late April to mid July.

We had no captures of the species *A. ustulatus* and *A. sputator* in 1998 and *A. brevis* in 1999.

The mean number of adult males of all species captured per trap in these four years is represented in *figure 3*.

We can clearly distinguish that in year 2000 the number of adult males of *A. rufipalis* captured in pheromone traps was higher than in 1998 and 1999, while in 2001 we had the highest number of captures. This is probably due to the new type of trap.

In the first year, 1998, only one larva of *Melanotus sp.* was found after the first trapping period. In the other two trapping periods no larvae were found like in 1999. In year 2000 after three trapping periods three, six and eight larvae of *A. rufipalpis* respectively were captured. Finally, in the first trapping period of 2001 twelve larvae of *A. rufipalpis* and one of *Melanotus sp.* were found.

The number of larvae collected by soil sampling was very low. Only *A. rufipalpis* larvae were collected. In 1998 two larvae (7.02 larvae/m^2) in the first sampling period were collected while in the subsequent year no larvae were found. The first period of year 2000 did not give any results, but in the second period two larvae (6.38 larvae/m^2) were found. The same number of larvae (4.68 larvae/m^2) was found in the first period of 2001. Expressed as larvae/m^2 the above numbers become 7.02, 6.38 and 4.68 in 1998, 2000 and 2001 respectively.

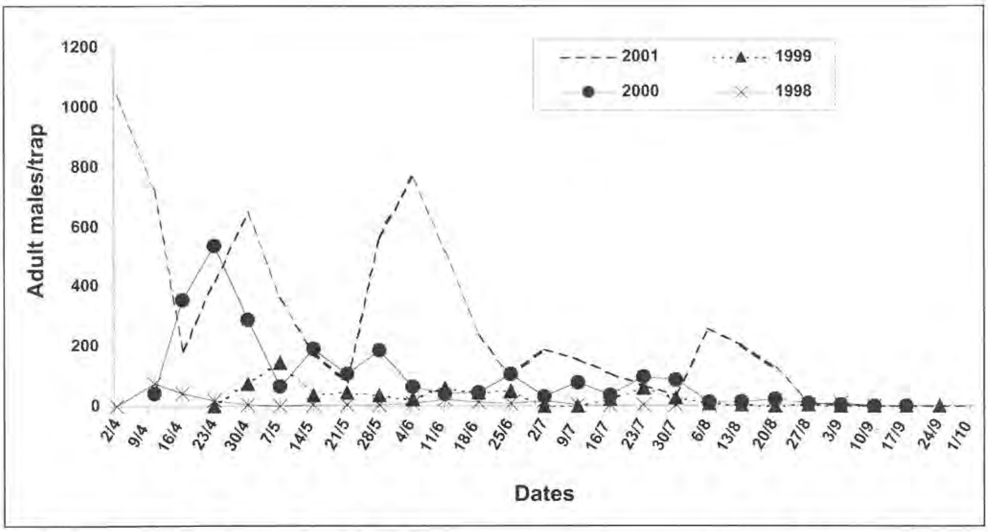

Figure 1 - Adult males of *A. rufipalpis* captured in pheromone traps in Velestino, Greece during 1998-2001

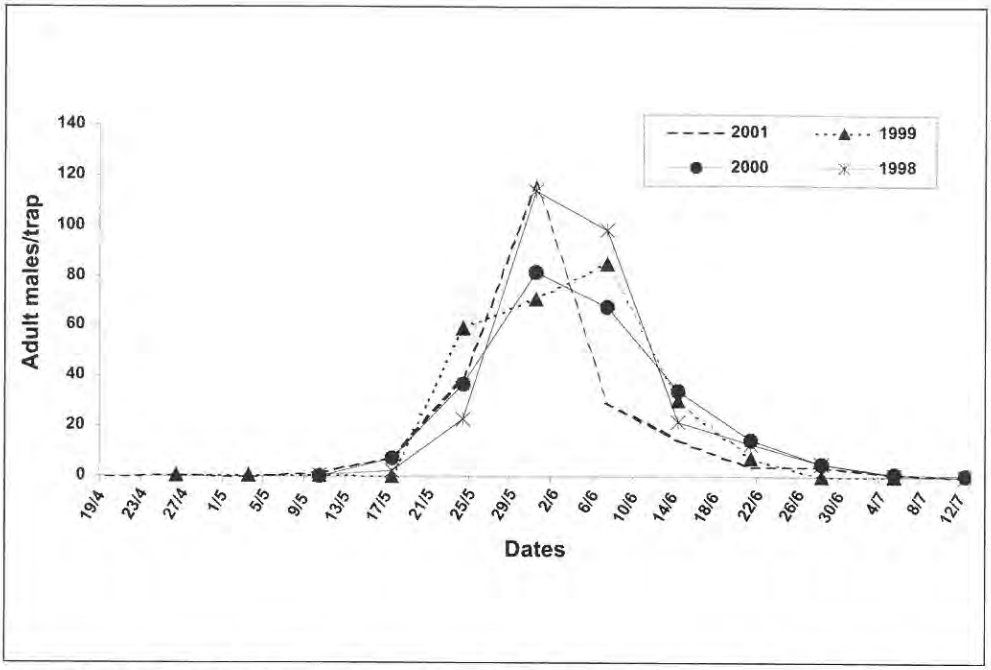

Figure 2 - Adult males of *A. litigiosus* captured in pheromone traps in Velestino, Greece during 1998-2001

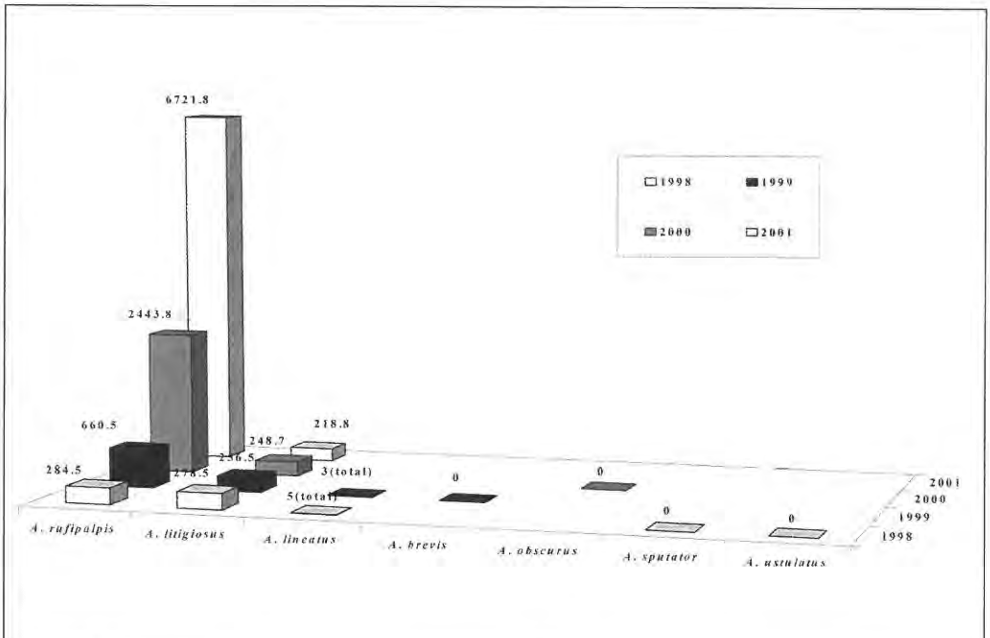

Figure 3 - Mean number of adult males of *A. rufipalpis, A. litigiosus, A. lineatus, A. brevis, A. obscurus, A. sputator and A. ustulatus* captured/trap in Velestino, Greece during 1998-2001

Conclusions

After four years work *A. rufipalpis* and *A. litigiosus* proved to be the key species in the area tested. The numbers of *A. rufipalpis* captured every year is probably the result of the different types of traps used.

The pheromone traps captured only the species *A. rufipalpis, A. lineatus* and *A. litigiosus. A. obscurus, A. brevis, A sputator* and *A. ustulatus* were absent or their populations consistence was very low. Geranyl-hexanoate + geranyl octanoate, used as pheromone for *A. obscurus,* captured only adults of *A. rufipalpis* but in lower numbers than only the geranyl-hexanoate used as pheromone.

Using soil bait traps and the soil sampling, few larvae were collected belonging mainly to *A. rufipalpis*. Some *Melanotus sp.* larvae were found too.

The number of larvae found in bait traps is higher than the number of larvae found in soil cores and is in concordance with the result obtained by Parker (Parker, 1996).

As to swarming patterns *A. rufipalpis* appeared from late March-early April to mid September, while *A. litigiosus* appeared from early May to early July.

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FIRST PRACTICAL RESULTS OF CLICK BEETLE TRAPPING WITH PHEROMONE TRAPS IN ITALY

Lorenzo Furlan, Andrea Di Bernardo,
Department of Agronomy, Entomology, University of Padova

Roberto Ferrari, Luca Boriani,
Centre for Agriculture and Environment "G. Nicoli", Crevalcore (BO)

Stefano Maini,
DiSTA, Entomology, University of Bologna

Paola Nobili,
Biotechnology and Agricolture Division
Enea, Casaccia Research Centre, Roma

Giancarlo Bourlot,
Piemonte Region, Phytosanitary Sector

Annalisa Turchi,
Province of Torino, Agricolture Service

Vincenzo Vacante, Carmelo Bonsignore, Gianni Gilioli,
Department of Agrochimics and Agrobiology
"Mediterranean" University of Reggio Calabria

Miklós Tóth,
Plant Protection Institute
of the Hungarian Academy of Sciences; Budapest

Abstract

From 1999 to 2001 trials were done in six different Italian regions: Veneto, Piemonte, Emilia-Romagna, Lazio, Calabria, Sicilia. The areas studied have variable climatic and agronomic conditions. Latitude ranged from 37°- 39° in the southern regions (Sicilia and Calabria) to 45°- 46° in the northern regions (Veneto and Piemonte). All trials were done using a common protocol. In the first two years, YATLOR and VARb traps were used while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was used. Individual traps were baited with the synthetic sex pheromones for one of the following species: *Agriotes lineatus* L., *A. obscurus* L., *A. sputator* L., *A. sordidus* Illiger (the same pheromone compound can attract *A. rufipalpis* Brullè males), *A. brevis* Candeze, *A. litigiosus* Rossi and *A. ustulatus* Schäller. Larval populations were estimated using bait traps and soil sampling. The traps proved to be effective in detecting the key species in different regions. In eastern areas, only larvae of *A. ustulatus*, *A. sordidus* and *A. brevis* were found damaging the main herbaceous crops; larval populations were correlated with pheromone trap data. Only adult males of the three species were caught in high numbers. The same correlation between larval populations and males caught in pheromone traps was observed in Veneto, Piemonte and Emilia Romagna. Here, the main species trapped were *A.litigiosus*, *A. sordidus* and *A. brevis*. Pheromones attracted two types of *A. litigiosus*: the dark form (var. *laichartingi*) in Veneto, Piemonte and the light brown form in Emilia Romagna. The main substance active to *A. litigiosus*, geranyl-isovalerate, proved to be sufficient to monitor efficiently the different varieties. Other compounds present in female glands added to geranyl-isovalerate had no synergistic effects, as demonstrated by a specific experiment done in Emilia–Romagna. *A. lineatus* larvae were also found in bait traps in Piemonte, but this was the only northern region where *A. lineatus* beetles were found in pheromone traps. Only in Piemonte traps for *A. obscurus* caught a few males, while only one larva of this species was found in bait traps. No reliable information on the *Agriotes* larvae damaging crops in central and southern Italy (Lazio, Calabria, Sicilia) has been available so far. The pheromone traps indicated that, with the exception of *A. ustulatus*, the same species present in northern Italy are also the key soil pests in these regions. All the other species are present everywhere; conspicuous *A. sordidus* populations were recorded at almost all the sites studied. *A. sputator* and *A. rufipalpis* were not found either in sex pheromone traps or larval samplings. Males of minor species were found in some pheromone traps (*Agriotes acuminatus* males were captured by *A. sputator* and *A. brevis* traps). The sex pheromone traps proved to be much more sensitive than the tools used to monitor larval populations. Click beetles numbers appeared to be correlated with larval populations estimated by soil sampling and bait trapping both in northern and southern regions.

Key words: *Agriotes*, sex pheromone traps, Italy.

Introduction

The potential suitability of sex pheromone traps for implementing IPM strategies against *Agriotes* populations was first suggested with experimentation in Hungary and Italy (Furlan *et al.*, 1996) and at the XIX IWGO Conference in Braga, Portugal (Furlan *et al.*, 1997). Subsequently, the efficacy of the new *Agriotes* sex pheromone traps in detecting different species and populations was reported at the XX IWGO Conference in Adana, Turkey (Furlan *et al.*, 1999). The first practical implications of the use of the new traps in Italy are described.

Materials and methods

From 1999 to 2001 trials were done in six different Italian regions: Northern Italy

– Piemonte, Veneto, Emilia-Romagna (1999 - 2000), Central Italy - Lazio; Southern Italy - Calabria, Sicilia (2000 - 2001). These areas have variable climatic and agronomic conditions. Latitude ranged from 37°- 39° in the southern regions (Sicilia and Calabria) to 45°- 46° in the northern regions (Veneto and Piemonte). Acid soils are present in Piemonte (north) and southern regions, while basic soils are prevalent in the other areas. All trials were done using the same protocol described in Furlan *et al.*, 2001. All trials were done using a common protocol. In the first two years, YATLOR and VARb traps were used while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was used.

In addition, a specific trial to evaluate the possible synergistic effects of minor compounds found in female glands of the red variety of *A. litigiosus* was done in Emilia-Romagna. Four pheromone component mixtures (treatments) were compared: geranyl isovalerate (0-0); geranyl isovalerate plus E,E farnesyl isovalerate (1% of geranyl isovalerate) (1-1); geranyl isovalerate plus E,E farnesyl isovalerate (1%) plus E8 hidroxygeranyl diisovalerate (1%) (1+1); geranyl isovalerate plus E,E farnesyl isovalerate (3%) (3-3). Bottle traps were set up in three blocks. The distance between traps within a block was 5-10 m. The distance between blocks ranged between 20 and 30 m. Traps were moved one position forward within a block at each occasion when the traps were inspected (from June 26 to July 7, i.e. nine times). At the same time, captured beetles were counted and removed. Capture data were $(x+0.5)^{1/2}$ transformed and analysed by ANOVA. Treatment means were separated by Duncan's New Multiple Range Test. In case one of the treatments was catching nil, the difference of the catch of other treatments from zero catch was analysed by one-sample t test.

Results

Detection of the key species in different regions (*table 1*): in eastern areas of Veneto only larvae of *A. ustulatus, A. sordidus* and *A. brevis* were found damaging the main herbaceous crops (Furlan *et al.*, 2000).

Only adult males of the three species were caught in high numbers and the data on larvae, collected both in bait traps and soil sampling, confirmed the pheromone trap data. The same correlation between larval populations and males caught in pheromone traps was observed in Veneto, Piemonte and Emilia-Romagna. Here, the main species trapped were *A.litigiosus, A. sordidus* and *A. brevis.* Pheromone baits for *A. litigiosus* attracted two varietes of *A. litigiosus:* the dark form (var. *laichartingi)* in Veneto and Piemonte and the light brown form in Emilia Romagna.

The main substance active to *A. litigiosus,* geranyl-isovalerate, proved to be sufficient to monitor efficiently the different varieties. Other compounds present in female glands added to geranyl-isovalerate had no synergistic effects, as demonstrated by a specific experiment done in Emilia-Romagna (*table 2*).

Table 1 - Adult population level estimated by using the different *Agriotes* traps in different Italian regions in 2000 - 2001 period (1999-2000 for Piemonte)

REGION	SOIL	Agriotes litigiosus	Agriotes ustulatus	Agriotes sordidus	Agriotes sputator	Agriotes brevis	Agriotes lineatus	Agriotes obscurus
VENETO east	basic	Low	Medium - High	Medium - High	No	Medium - High	No	No
VENETO west	basic	Medium - High	No	Medium - High	No	Medium - High	No	No
EMILIA ROMAGNA	basic	Medium - High	No	Medium - High	No	Medium - High	No	No
PIEMONTE	acid	Medium - High	No	Medium	No	Medium - High	Medium	Low
LAZIO	basic	Medium - High	No	Medium	No	Medium	Low	No
CALABRIA	acid	Low	No	No	No	Low	Medium	No
SICILIA	basic	No	No	High	No	No	No	No

High= more than 500 adults/trap/season
Medium= between 50 and 500 adults/trap/season
Low=less than 50 adults/trap/season
Data average of at least 2 traps in each year

Table 2 - Effect of the addition of minor compounds to geranyl isovalerate. No compound increased the captures of traps placed out in Emilia-Romagna from June 26 to July 7, 2000. The differences were not significant at P=0.05

Treatment	Count	Sum	Minimum	Maximum	Average (Mean)	Standard error	Standard deviation
0-0	27	182	0	46	6.74	1.92	9.95
1-1	27	241	0	30	8.93	1.47	7.63
1+1	27	185	0	45	6.85	1.89	9.82
3-3	27	157	0	33	5.81	1.37	7.13
Total	108	765	-	-	7.08		8.68

Larvae of *A. lineatus* were found in bait traps in Piemonte and this was the only northern region where *A. lineatus* beetles were found in pheromone traps. Only in Piemonte traps for *A. obscurus* caught a few males, while only one larva of this species was found in bait traps. No reliable information on the *Agriotes* larvae damaging crops in central and southern Italy (Lazio, Calabria, Sicilia) has been available so far. The pheromone traps indicated that, with the exception of *A. ustulatus,* the same species present in northern Italy are also the key soil pests in these regions. All the other species are present everywhere; conspicuous *A. sordidus* populations were recorded at almost all the sites studied. *A. sputator* and *A. rufipalpis* were not found either in sex pheromone traps or larval samplings. Males of minor species were found in some pheromone traps (*Agriotes acuminatus* Stephens males were captured by *A. sputator* and *A. brevis* traps).

Traps efficacy and relationship between pheromone trap catches and wireworm population levels: the sex pheromone traps proved to be much more sensitive than the tools used to monitor larval populations. Click beetle numbers appeared to be correlated with larval populations estimated by soil sampling and bait trapping both in northern and southern regions. For example in Calabria conspicuous numbers of *A. lineatus* males were caught in fields where only larvae of this species had been found in bait traps and on damaged plants. In eastern Veneto, sex pheromone traps caught few *A. litigiosus* adults despite the fact no larvae of this species had been collected over the last ten years (*table 3*).

Regional differences in adult emergence patterns were also clearly detected. For example *A. litigiosus* var. *laichartingi* males started emerging from the soil about one month earlier than the light brown form of the same species in Emilia-Romagna.

Table 3 - *Agriotes lineatus, A. brevis* and *A. litigiosus* males captured by using YATLOR and YATLORfunnel traps at different localities of Italy with different mature larvae population levels in 2000-2001

LOCALITY	Veneto east	Veneto west	Piemonte	Emilia Romagna	Lazio	Calabria	Sicilia	Veneto east	Veneto west	Piemonte	Emilia Romagna	Lazio	Calabria	Sicilia	Veneto east	Veneto west	Piemonte	Emilia Romagna	Lazio	Calabria	Sicilia
SPECIES	*A. lineatus*	*A. lineatus*	*A. lineatus*	*A. lineatus*	*A. lineatus*	*A. lineatus*	*A. lineatus*	*A. brevis*	*A. brevis*	*A. brevis*	*A. brevis*	*A. brevis*	*A. brevis*	*A. brevis*	*A. litigiosus*	*A. litigiosus*	*A. litigiosus*	*A. litigiosus*	*A. litigiosus*	*A. litigiosus*	*A. litigiosus*
Larval Population level	L	L	H	L	H	H	L	H	H	H	H	L	L	L	L	H	H	H	H	L	L
Males captured	NO	NO	M	NO	L	M	L	M-H	M-H	M-H	M-H	M	L	NO	L	M-H	M-H	M-H	H	L	L

L= no larvae of the species found by using attractive traps and soil sampling in the fields surrounding the sex pheromone traps;

H= larvae of the species easily found by using attractive traps and soil sampling in the fields surrounding the sex pheromone traps.

H= more than 500 adults/trap/season; M= between 50 and 500 adults/trap/season; L=less than 50 adults/trap/season; NO=no adults found.

Data average of at least 2 traps in each year.

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TWO YEARS OF MONITORING CLICK BEETLES AND WIREWORMS IN SLOVENIA

Stanislav Gomboc, Lea Milevoj, Polona Bitenc, Aleksander Bobnar, Franci Celar,
Biotechnical Faculty, Department of Agronomy, Institute of Phytomedicine, Jamnikarjeva 101, Ljubljana, Slovenia

Lorenzo Furlan,
University of Padova, Department of Agronomy, Via Romea 16, I-35020 Legnaro PD, Italy

Miklos Tóth,
Hungarian Academy of Sciences, Plant Protection Institute, Pf 102, H-1525 Budapest, Hungary

Abstract

Ten species belonging to the genus *Agriotes* have been found to exist in Slovenia to date: *A. atterimus* L., *A. acuminatus* Steph., *A. ustulatus* Schäll., *A. litigiosus* Rossi, *A. pilosellus* Schönh., *A. lineatus* L., *A. obscurus* L., *A. sputator* L., *A. medvedevi* Dolin, *A. brevis* Cand. Wireworms are known to be responsible for damage to small plant crops – maize, sugar beet, vegetables and potato. In order to find a simple tool to sample wireworm populations, the Biotechnical Faculty of Slovenia decided to participate in an international programme, "The effectiveness of *Agriotes* sex pheromone traps in implementing IPM strategies" from 2000. In the first year YATLOR and VARb traps were used, while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was employed. Individual traps were baited with lures for one of the following species: *A. lineatus*, *A. obscurus*, *A. sputator*, *A. rufipalpis* (the same pheromone compound can capture *A. sordidus* males), *A. brevis*, *A.litigiosus*, *A.ustulatus*. In both years the monitoring period was from March (from May for *A. ustulatus* in *A. litigiosus*) to the end of August. Larval population was estimated by using bait traps. Bait traps were laid in April and September, before and after the swarming period. In 2000 monitoring was carried out in two locations. In 2001 the number of locations was increased to five, in Central and Eastern Slovenia. At all sites soils are acidic. The traps appeared to be selective enough to distinguish *A. sputator* and *A. brevis* despite these two species being systematically very close. *A. litigiosus*, *A. sordidus* and *A. rufipalpis* were not found in any of the locations studied. The dominant species were: *A. lineatus* in Ljubljana (1743 specimens/trap/season in 2000 and 474 in 2001); *A. obscurus* and *A. sputator* in Lendava (247/18 specimens *A. obscurus*, 263/470 specimens *A. sputator* in 2000/2001); *A. ustulatus* in Komenda (1117 specimens in 2001); *A. sputator* in Zagorje (286 specimens in 2001) and *A. sputator* in Žalec (261 specimens in 2001). The efficacy of the pheromone traps in detecting the regional variations of swarming patterns of the species is also discussed.

Key words: *Elateridae*, *Agriotes*, click beetles, wireworms, pheromone traps, integrated pest management.

INTRODUCTION

95,500 hectares in Slovenia are planted with sensitive crops to wireworms attack (maize, sugar beet and potato), representing 56% of arable land. 21,000 hectares of these crops are regularly treated with soil insecticides. Assuming a mean quantity of 0.3 l/ha of imidacloprid, currently the most applied soil insecticide (as seed dressing), this means that a total of about 6,600 litres of insecticide are yearly applied. Most of the treatments are applied to maize and other crops at planting without any evaluation of the actual presence of an economically – damaging wireworm population. In order to ensure that treatments are only applied where necessary our knowledge of the biology and ecology of click beetles in Slovenia must be improved.

At present precise data concerning click beetle species in Slovenia are not available, as the family has been poorly studied. Our own estimate is that there are at least 150 species living in Slovenia. In the Czech Republic and Slovakia a total of 169 species have been discovered (Laibner, 2000). The genus *Agriotes* should be the predominant in Slovenia. Ten species belonging to this genus have been discovered (Brelih pers. Comm.; Vrabl, 1992; Matis and Vrabl, 1997; Čamprang, 1997; Milevoj, 1985, Urek, 1985). From the agronomic point of view the most important species in arable land should be *A. sputator* L., *A. obscurus* L., *A. lineatus* L., *A ustulatus* Schäll., *A. litigiosus* Rossi and *A. brevis* Candeze while in forest areas *A. atterimus* L., *A. acuminatus* Steph., *A. medvedevi* Dolin, *A. pilosellus* Schoenh should be the key species.

Due to the shortage of reliable and inexepensive prognostic methods and increasing pressure to reduce reliance on soil insecticides, over last years new methods for forecasting the critical number of wireworms in the soil began to be evaluated: those based on using bait for larvae (Chabert, Blot, 1992; Parker, 1996) as those using pheromone traps for beetles (Siirde *et al.*, 1993, Merivee and Erm, 1993; Yatsynin *et al.*, 1996; Furlan, 1999). In order to achieve the best comparable results a voluntary international research programme entitled The Effectiveness of *Agiotes* Sex Pheromone Traps in Implementing IPM Strategies, led by Dr. Lorenzo Furlan from Italy, has been running in Europe since 1997. In 2000 Slovenia joined this programme.

MATERIALS AND METHODS

In 2000 traps were placed out in two locations; in 2001 the number of locations increased to five, as shown in *figure 1*.

Location characteristics (meteorological data average of a 30 years period):

- Ljubljana, laboratory field: traps placed in grass-clover mixture fields, elevation

299 m a.s.l., latitude 46° 03′, longitude 14° 29′ 14″, mean temperature 9.8° C, total amount of precipitation 1393 mm, soil acid, loamy.

- Petišovci near Lendava: traps placed in maize field – crop rotation: sugar beet-maize-wheat, elevation 159 m a.s.l., latitude 46° 31′ 05″, longitude 16° 28′ 42″, mean temperature 10.0° C, total amount of precipitation 805 mm, soil acid, lomy-clay.
- Lendava: traps placed in maize field – crop rotation: sugar beet-maize-wheat, elevation 170 m a.s.l., latitude 46° 33′ 36″, longitude 16° 27′ 27″, mean temperature 10.0° C, total amount of precipitation 805 mm, soil acid, lomy-clay.
- Komenda: traps placed on field with sugar beet and maize – crop rotation: sugar beet-maize-wheat, elevation 338 m a.s.l., latitude 46° 12′ 09″, longitude 14° 31′ 47″, mean temperature 9.8° C, total amount of precipitation 1393 mm, soil acid, lomy-sandy.
- Poljče near Žalec: traps placed in the border of meadow fields, elevation 288 m a.s.l., latitude 46° 16′ 14″, longitude 15° 03′ 23″, mean temperature 9.1° C, total amount of precipitation 1145 mm, soil acid, lomy-sandy.
- Zagorje – Mali Kum: traps placed in the border of meadow fields, elevation 512 m a.s.l., latitude 46° 04′ 49″, longitude 15° 04′ 33″, mean temperature 9.0° C, total amount of precipitation 1145 mm, soil acid, lomy.

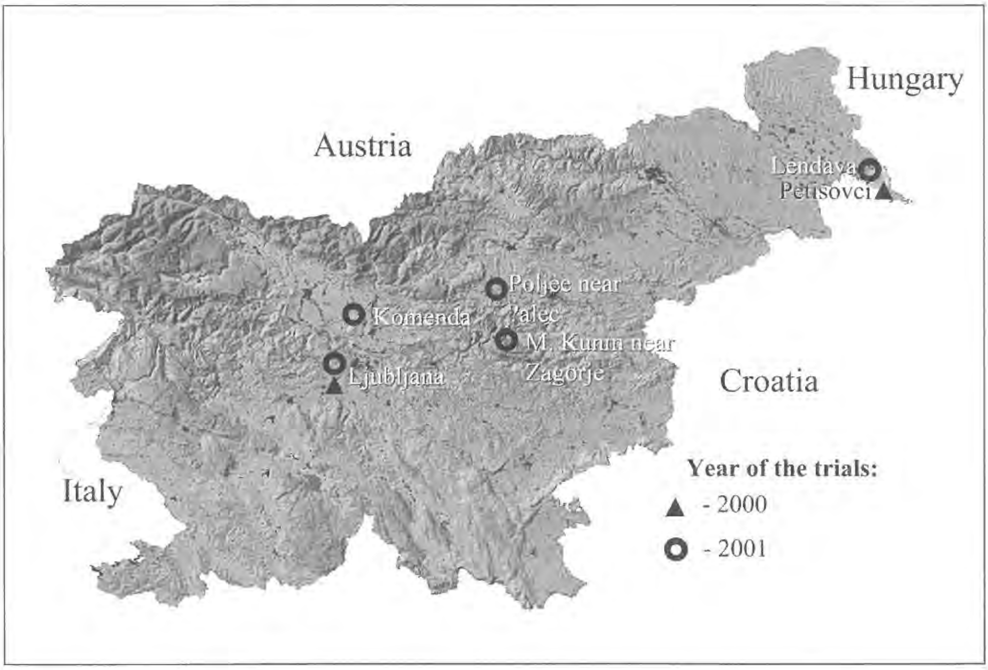

Figure 1 - Locations of pheromone traps

In the first year YATLOR (for the crawling species *A. lineatus, A. obscurus, A. brevis*) and VARb (for the flying species *A. litigiosus, A. ustulatus, A. sputator, A. sordidus/rufipalpis*) traps for adults were used while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was used. The same common protocol described in Furlan *et al.*, 2001 was followed with the exception that larval populations were evaluated by using the bait traps only (no soil sampling was done).

RESULTS

Five of the eight species monitored were collected in the pheromone traps. These were *A. brevis, A. lineatus, A. obscurus, A sputator* and *A. ustulatus*. They were found in all locations, although in different numbers (*table 1*). The predominant species were *A. lineatus* in Ljubljana – high population level (1743 specimens in 2000 and 474 in 2001); *A. obscurus* and *A. sputator* in Lendava – medium population level (*A. obscurus* 247 specimens in 2000 and 18 in 2001, *A. sputator* 263 specimens in 2000 and 470 in 2001); *A. ustulatus* in Komenda – high population level (1117 specimens in 2001); *A. sputator* in Zagorje – high population level (286 specimens in 2001) and *A. sputator* in Žalec – medium population level (261 specimens in 2001). The criteria to classify population density were uniform within Europe (Furlan *et al.*, 2001): high population level means more than 500 adults/trap/season; medium population level 50 to 500 adults/trap/season and low population level less than 50 adults/trap/season.

Table 1 - Number of specimens captured per trap per season at different locations in 2000 and 2001

Location and year of trials	*Agriotes brevis*	*Agriotes lineatus*	*Agriotes obscurus*	*Agriotes sputator*	*Agriotes ustulatus*
Ljubljana 2000	95	**1743**	228	88	127
Ljubljana 2001	240	**474**	125	84	44
Petišovci near Lendava 2000	4	104	**247**	**263**	113
Lendava 2001	52	194	16	**470**	13
Komenda 2001	23	251	43	27	**1117**
Poljče 2001	2	60	4	**261**	7
Zagorje - Mali Kum 2001	5	5	14	**286**	33

The traps appeared to be sufficiently selective to distinguish between *A. sputator* and *A. brevis*, although these two species are very close. *A. litigiosus, A. sordidus* and *A. rufipalpis* were not found at any of the locations. Untargeted species trapped were less than 1%. Species found in traps for other species were: *A. sputator* in *A. brevis* trap, *A. lineatus* in *A. sputator* trap, *A. acuminatus* in *A. sputator* trap, *A brevis* in *A. sputator* trap and *A. ustulatus* in *A. sputator* trap. Data clearly demonstrated that numbers varied significantly from year to year at the same location (Ljubljana) but greater variances were found between different populations. In damp and clay soils the species *A. lineatus* was predominant, whereas in medium damp soils *A. sputator, A. ustulatus* and *A. obscurus* were predominant.

Swarming pattern of the species appeared quite specific (*figures* 2 to *8*). First the species *A. brevis* appeared – end of March to end of April. This was followed by the species *A. sputator* with peak in the middle of May but quite steady from the middle of April until the middle of August. *A. lineatus* had an extended period of activity – from the middle of April to the middle of July, peaking in May. *A. obscurus* and *A. ustulatus* had a shorter period of activity, especially *A. ustulatus*. *A. obscurus* appeared from May to the middle of July, peaking in May. *A. ustulatus* appeared from June to August, peaking in June.

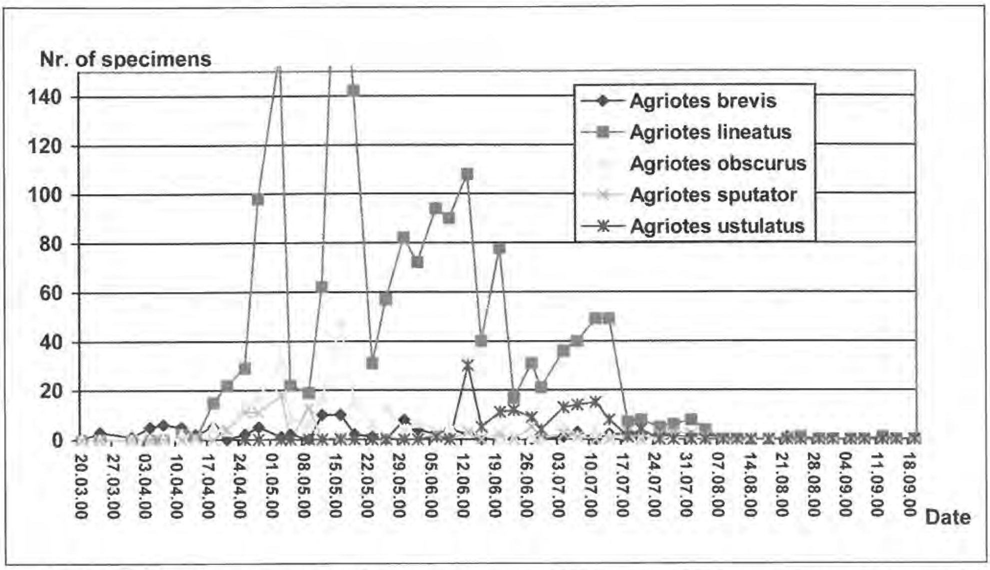

Figure 2 - Click beetles captured in pheromone traps in Ljubljana in 2000 (grass-clover mixture field)

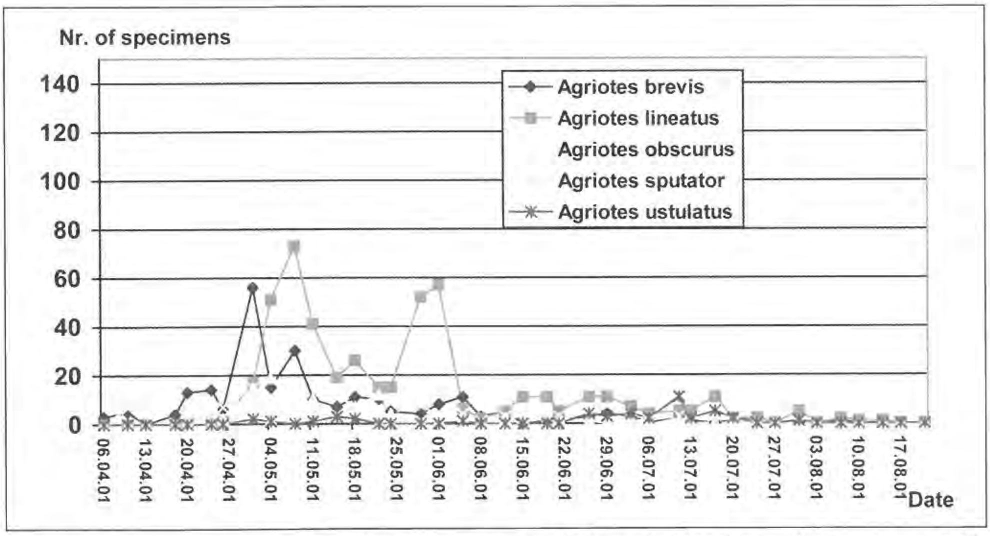

Figure 3 - Click beetles captured in pheromone traps in Ljubljana in 2001 (grass-clover mixture field)

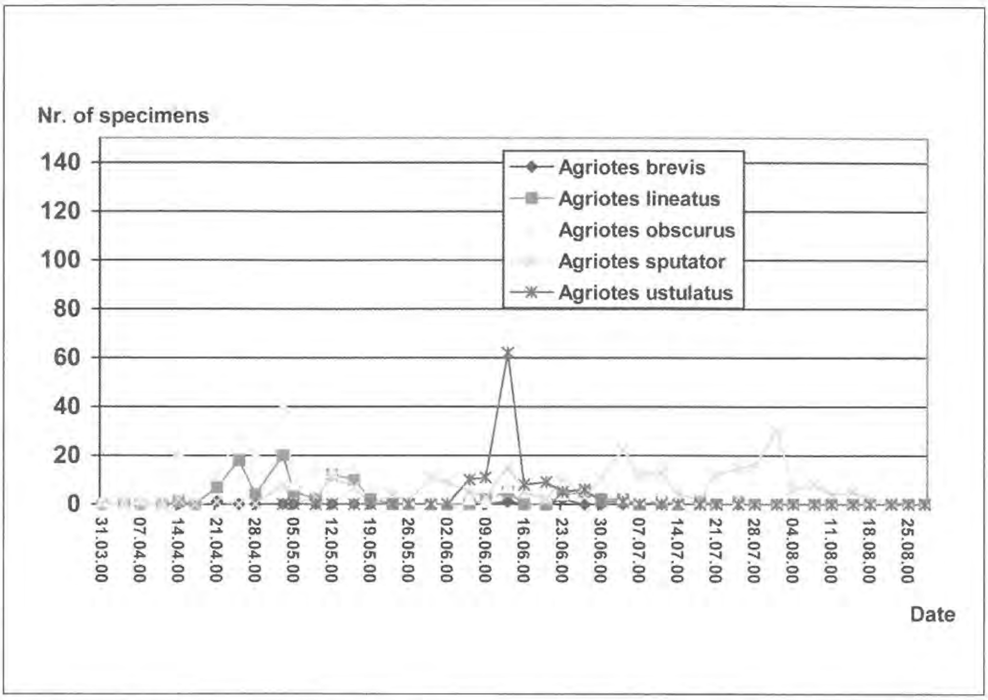

Figure 4 - Click beetles captured in pheromone traps in Peti?ovci near Lendava in 2000 (maize field)

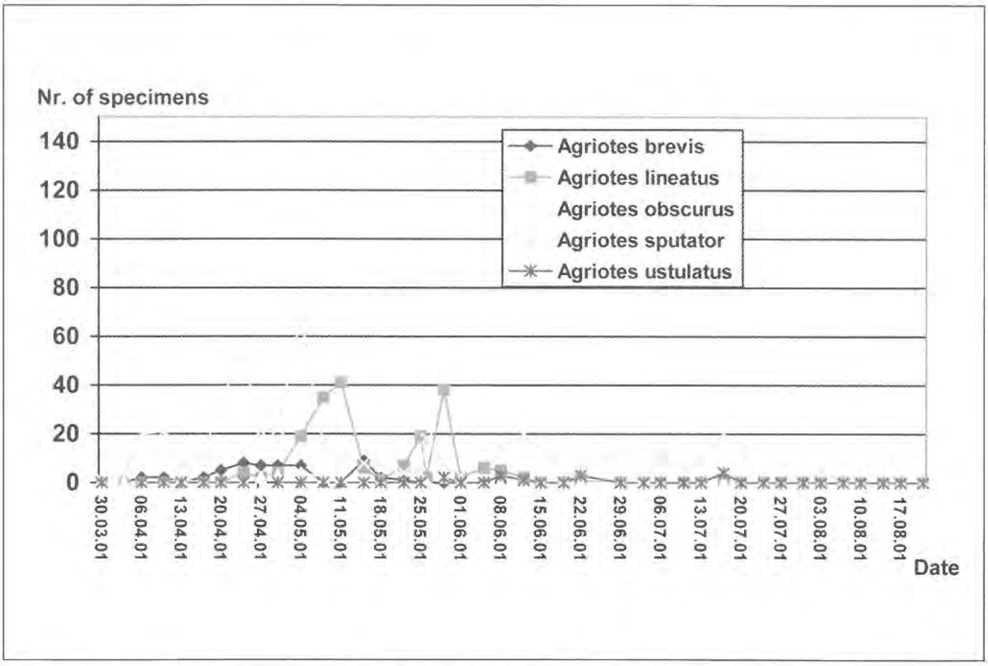

Figure 5 - Click beetles captured in pheromone traps in Lendava in 2001 (maize field)

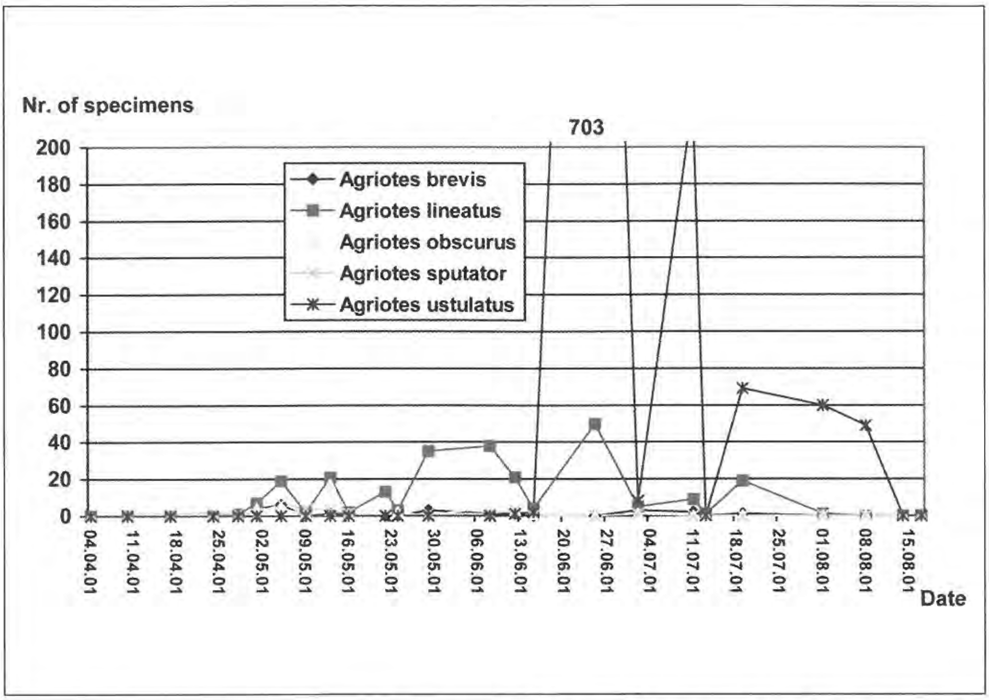

Figure 6 - Click beetles captured in pheromone traps in Komenda near Kamnik in 2001 (maize and sugar beet field)

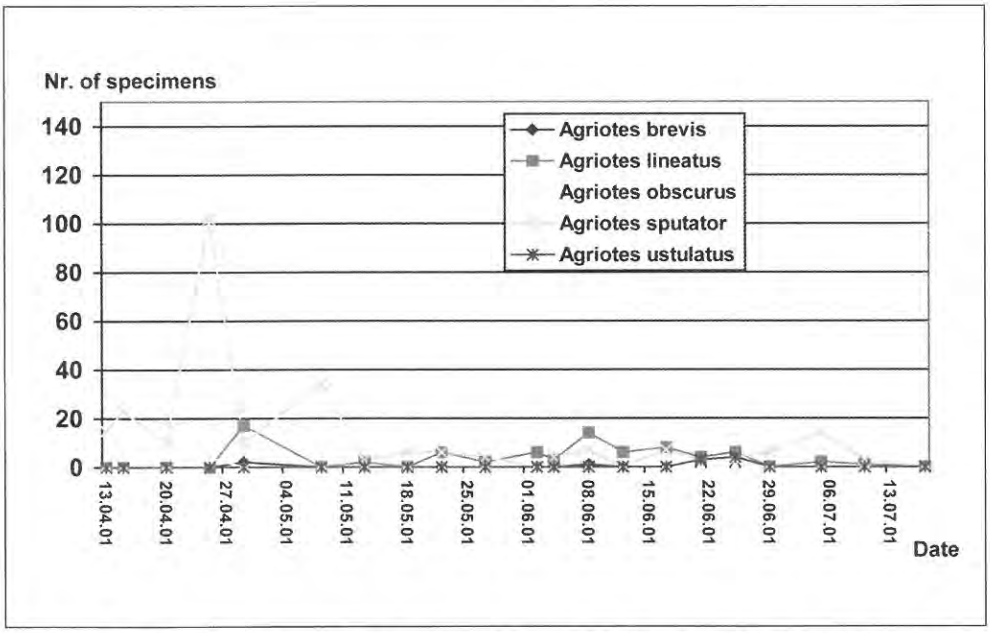

Figure 7 - Click beetles captured in pheromone traps in Polj?e near ?alec in 2001 (border of a meadow field)

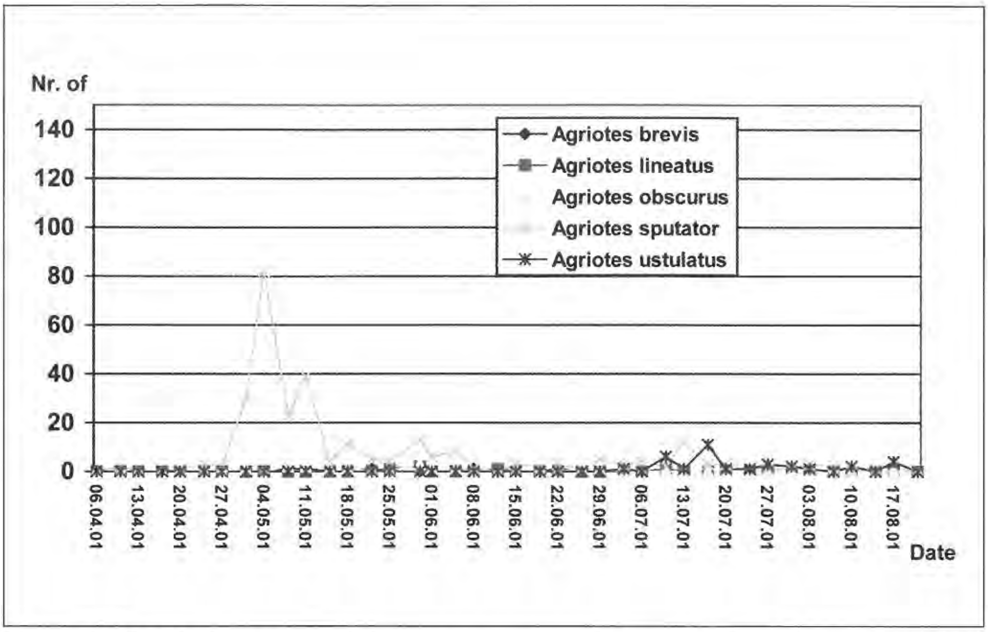

Figure 8 - Click beetles captured in pheromone traps in Zagorje – Mali Kum in year 2001 (border of a meadow field)

Larval populations

Table 2 shows the species composition estimated by using 40 bait traps/ field. It appears that everywhere the average numbers of wireworms were below the critical number, according to the threshold of Vrabl (1992). The higher catches of wireworms were expected in the grass-clover fields of Ljubljana, but here too the number was low. This was likely caused by the fact that the soil was not bare when the traps were placed out. In Dolnji Lakoš the number of wireworms was also low. Most of the wireworms were caught in meadows fields, confirming that high populations of wireworms live in soils constantly covered with plants.

In 2001 only a small number of wireworms were caught in Ljubljana and in the fields around Lendava, where only 3 of the 8 traps caught specimens. As the threshold was not exceeded in these fields, owners of the fields did not treat seeds with insecticide. In Ljubljana in spite of a high number of *A. lineatus* beetles caught, no larvae of any species were caught in the bait traps. This was probably due to the presence of the grass which does not permit bait traps to perform correctly since the larvae are attracted by the roots of plants. In reviewing the composition of larvae, there was an apparent difference between locations and sampling times. In Ljubljana the wireworm *A. obscurus* was not caught by bait traps and neither was *H. hirtus*, that usually appears in natural meadows. These results confirm the hypothesis that the composition of wireworm species differs according to region and therefore there may be differences in crop damage with respect to the ecological characteristics of species.

Table 2 - Number of larvae captured per 40 bait traps

YEAR 2000 Ljubljana:	Lendava - Dolnji Lakoš:
19 APRIL 2000: (grass-clover mixture) - *Agriotes ustulatus* (8) 21 September 2000: (same field) - *Agriotes ustulatus* (16) - *Agriotes sputator* (3)	14 April 2000: (before maize planting) - *Agriotes sputator* (3) 30 September 2000: (sugar beet) - *Agriotes obscurus* (4) - *Hemicrepidius hirtus* Herbst (5) 30 September 2000: (meadow) - *Agriotes obscurus* (12) - *Agriotes sputator* (2)
YEAR 2001 Ljubljana:	Lendava: (bait traps placed on 8 fields)
25 APRIL 2000: (grass-clover mixture) - *Agriotes ustulatus* (5)	23 April 2000: (before maize planting) field Pince: - *Hemicrepidius hirtus* (2) field Benica: - *Hemicrepidius hirtus* (2) field Pince – near forest: - *Athous* sp. (6)

Conclusions

Pheromone traps are very effective, selective and easy to use (much more than the bait traps for larvae).

Data reported are the first concerning presence and swarming period of different *Agriotes* species in Slovenia.

The most common species in Slovenia (depending on the region and soil type) are *A. lineatus*, *A. obscurus*, *A. sputator* and *A. ustulatus*.

The results also show the differences in population structures between the species and regions.

The IPM strategy against *Agriotes* species based on sex pheromone trap use seems to be very promising; anyway more data in different regions with different rotations are necessary to study the correlation between click beetle captures and subsequent larval populations.

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THE EFFICACY OF THE NEW AGRIOTES SEX PHEROMONE TRAPS IN DETECTING WIREWORM POPULATION LEVELS IN DIFFERENT EUROPEAN COUNTRIES

Lorenzo Furlan,
Department of Agronomy, Entomology,University of Padova, Italy

Miklós Tóth,
Plant Protection Institute, Budapest, Pf 102, H-1525 Hungary

William E. Parker,
ADAS, Woodthorne, Wolverhampton, WV6 8TQ, UK

Maria Ivezić, Stjepan Pancic, Mirjana Brmez,
J.J.Strossmayer University, Osijek, Croatia

Renata Dobrinčić, Jasminka Igrc Barčić,
Faculty of Agriculture, University of Zagreb, Croatia

Felicia Mureşan,
Agricultural Research Station Turda, Romania

Mitko Subchev, Teodora Toshova,
Institute of Zoology, Sofia, Bulgaria

Zoltan Molnar, I.C.P.C., Brasov, Romania

Barbara Ditsch, Dagmar Voigt,
Botanical Garden of Dresden, Germany

Abstract

New *Agriotes* sex pheromone traps were tested in different European countries enabling the suitability of the traps in monitoring different populations and the relationship between pheromone trap catches and wireworm population levels to be studied under a range of climatic and agronomic conditions. Trials were done in the United Kingdom, Eastern Croatia, Central Croatia, Central Romania, Central Eastern Romania, Germany, Bulgaria from 1999 to 2001. The areas of the regions where the experimentation was done have very variable climatic and agronomic characteristics (latitude ranged from 42° to 56°). All trials were done using a common protocol. In the first 2 years YATLOR and VARb traps were used while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was used. Individual traps were baited with the lures for one of the following species: *A. lineatus* L., *A. obscurus* L., *A. sputator* L., *A. sordidus*

Illiger, *A. rufipalpis* Brullé, *A. brevis* Candeze, *A.litigiosus* Rossi, *A.ustulatus* Schäller. Larval population was estimated by using bait traps and soil sampling.

In all regions conducting studies the pheromone traps were confirmed to be a sensitive tool for detecting the key wireworm species present. At all sites it was possible to detect the dominant species. Furthermore the traps appeared to be selective enough to distinguish *A. sputator* and *A. brevis* despite these two species being systematically very close. Males of minor species (also not *Agriotes*) were found in some traps (for example *Cydnopus pilosus* Leske in Bulgaria). The response of the different species to the different monitoring tools was very variable. Generally the sex pheromone traps proved to be a much more sensitive tool than soil sampling and bait traps for larvae. For all species traps were able to detect wireworm populations below those that can be reliably detected using soil sampling and bait trapping. In addition pheromone traps proved to be effective in detecting the regional variations of swarming patterns of the same species.

Key words: *Agriotes*, wireworms, sex pheromone traps, Europe.

Introduction

First data on the effectiveness of the new *Agriotes* sex pheromone traps in different European countries with different species and populations were presented at XX IWGO Conference in Adana, Turkey (Furlan *et al.*, 1999). In subsequent years, pheromone traps were tested in several new European countries under a range of different climatic and agronomic conditions. The objectives of the work were 1) to assess the effectiveness of the traps, and 2) to investigate the relationship between pheromone trap catches and wireworm population levels in the soil. The results from this work are summarised in this paper.

Materials and methods

Site locations

In 1999, 2000 and 2001 trials were done in the United Kingdom (five different localities), Eastern Croatia, Central Romania, and Bulgaria (2000 and 2001 only). In 2001 only, additional trials were done in Central Eastern Romania, Germany and Central Croatia. The regions where the experimentation was done have very variable climatic and agronomic characteristics. Latitude ranged from 42° in South eastern regions to 56° in the most northern regions of the UK. In most cases the soils were acid. All experiments were done using a common protocol.

Trap and pheromone materials

In the first two years, YATLOR and VARb traps were used, while in 2001 a trap design suitable for both flying and crawling species (YATLORfunnel) was used (Furlan *et al.*, 2001). Individual traps were baited with the synthetic sex pheromones (Tóth *et al.*, 1997; Tóth *et al.* 1998) for one of the *Agriotes* species given in *table 1*. Kartel 730 vials were used as lure dispensers for all pheromones.

Table 1 - Pheromone composition for *Agriotes* species

Agriotes species	Pheromone composition	Source reference
A. ustulatus	E,E - farnesyl acetate	Kudryatsev *et al.*, 1993
A. litigiosus	geranyl-isovalerate	Yatsynin *et al.*, 1980
A. sputator	Geranyl-butanoate	Yatsinin *et al.*, 1986
A. obscurus	geranyl hexanoate + geranyl octanoate	Borg-Karlson *et al.*, 1988
A. rufipalpis/sordidus	geranyl-hexanoate*	Toth *et al.*, 2001
A. lineatus	geranyl octanoate + geranyl butyrate	Yatsinin *et. al.*,1980, 1996
A. brevis	Geranyl butyrate +*E,E* farnesyl butyrate	Toth *et al.*, 2002

* The same pheromone compound can attract A. rufipalpis males.

Field experiment procedure

1. *Site selection:* fields chosen for the work were in a typical rotation for the area. Where possible, the field had a known history of wireworm attack (or wireworm attacks confirmed in the locality), and/or grass fields in the vicinity. No soil insecticides were used in the year prior to the start of the work, or were used in the spring of the year in which the work was done. Where possible, fields that had remained bare until April were chosen in order to allow evaluation of larval populations using bait traps as well as soil sampling, If this was not possible, bare (uncropped) areas in the middle of the field were obtained by cutting down small areas of the crop. In some cases some pheromone traps were placed out in fields planted with crops not usually treated for wireworms (soybean, winter cereals, meadows).
2. *Trap position in the field*: see *figure 1.*
3. *Trap separation distance*: the distance between traps for the same species or different species was at least 30 m.
4. *Period of monitoring*: 1 March to 15 September for *A. brevis, A. obscurus, A. lineatus, A. sputator, A. rufipalpis.* 15 April to 31 August for *A. litigiosus* and *A. ustulatus.*
5. *Trap inspections:* pheromone traps were inspected once or twice a week. All beetle specimens were removed from the traps at each observation and retained.
6. *Replacement of pheromone caps:* every 30 days.
7. *Replication:* work was done in at least two fields.
8. *Soil sampling:* a 10 x 5 grid (20 m x 30 m, figure 1) of 50 soil samples was taken covering the area where the pheromone traps were set up. Each soil sample was 12 cm in diameter and 30 to 60 cm deep according to the season (30 cm deep

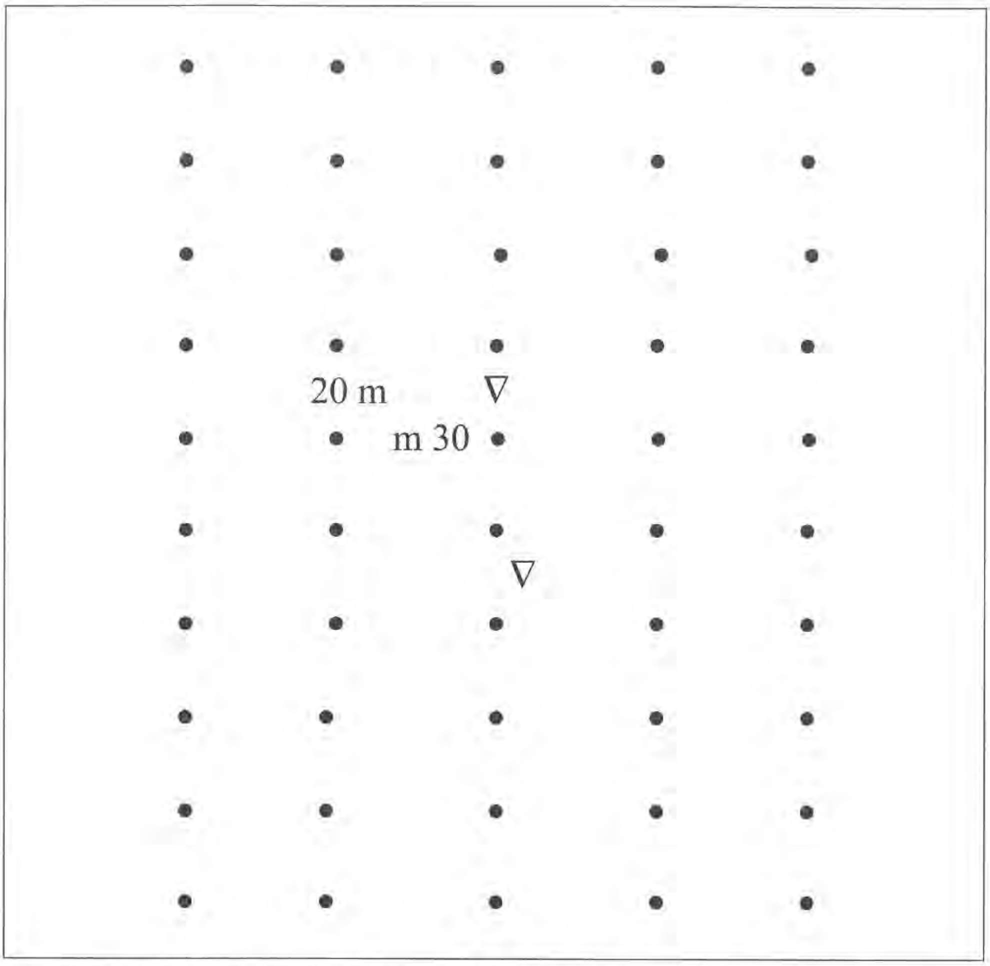

Figure 1 - Layout for soil sampling/bait trapping points & pheromone trap location

∇ = sex pheromone traps

• = bait trap and soil sampling (1-2 m from the bait trap)

cores are adequate in the spring when temperatures are above 10°C and soil moisture is high). After collection, soil samples were processed by putting soil cores into Tullgren funnels, 26 cm in diameter, provided with a 0.5 cm mesh at the bottom. The collecting vial under each funnel was filled with moistened soil to keep the larvae alive. The soil was allowed to dry for at least 30 days in a sheltered place. Larvae that fell into the collecting vials were counted and identified.

9. *Bait traps*: bait traps were placed 1 to 2 m away from each point where the soil cores had been taken provided the soil was bare. Each trap was made and used according to the description given by Chabert and Blot (1992). These comprise a plastic pot 11cm in diameter provided with holes in the bottom; the pots are filled with

vermiculite, 30 ml of wheat seeds and 30 ml of corn (maize) seeds. The pots were wetted before being placed into the soil just below the surface and covered with an 18 cm diameter plastic lid placed a few cm above the rim of the pot. Traps were checked by hand-sorting the contents after 10 to 15 days. After hand-sorting, bait traps contents were processed by putting it into Tullgren funnels as per soil samples (see above).

RESULTS

In all regions conducting studies the pheromone traps were confirmed to be a sensitive tool for detecting the key wireworm species present (*table* 2). At all sites in the United Kingdom only three species (*A. lineatus, A. obscurus, A. sputator*) were captured. *A. obscurus* was the dominant species. In acid fields of the other nations a different species range was found. In Bulgaria *A. brevis* and *A. lineatus* appeared the dominant species while *A. ustulatus* was dominant in Germany and Eastern Croatia. The traps appeared to be selective enough to distinguish *A. sputator* and *A. brevis* despite these two species being systematically very close. Agriotes litigiosus and Agriotes sordidus were not found in any of the nations studied while males of minor species (also not *Agriotes*) were found in some traps (for example *Cidnopus pilosus* in Bulgaria).

Table 2 - Adult population level estimated by using the different *Agriotes* traps in different European nations over 2000-2001 period

European nations	*Agriotes litigiosus*	*Agriotes ustulatus*	*Agriotes sordidus/A. rufipalpis*	*Agriotes sputator*	*Agriotes brevis*	*Agriotes lineatus*	*Agriotes obscurus*
BULGARIA	No	Low	No	Low	Medium	High	No
CROATIA - central	-	Medium	No	Medium	Medium	High	Medium
CROATIA east	No	Medium	Low-ruf	Medium	Low	Medium	Medium
ENGLAND - south west	No	No	No	Medium	-	Medium	High
ENGLAND - west	No	No	No	Low	-	Low	Medium
ENGLAND - north	-	-	-	Low	-	Low	Medium
GERMANY	No	Medium	No	Low	No	Low	Low
ROMANIA central	-	Medium	Low-ruf	No	Medium	Medium	Low
ROMANIA central eastern	No	No	-	Medium	Low	High	High
SCOTLAND - central	-	-	-	No	-	No	Low
WALES - mid	-	-	-	High	-	High	High

High = more than 500 adults/trap/season; Medium = between 50 and 500 adults/trap/season; Low = less than 50 adults/trap/season; NO = no specimens; - = trap not placed out
Data average of at least 2 traps in one or 2 years.

Taking into consideration also data obtained in previous years (Furlan *et al.*, 1999) it is now possible to prepare reliable maps on the distribution of *Agriotes* species in Europe (*Figures. 2-8*). *A. ustulatus* is clearly a species distributed only in the eastern part of Europe above about 42° latitude, while *A. litigiosus* is found in the southern-eastern regions of Europe. In contrast, *A. lineatus* proved to be a very adaptable species, being present from southern Italy (39° latitude) to the most northern locality of UK (56° latitude). In Portugal, *A. lineatus* seems to have been displaced by *Agriotes proximus*. *A. obscurus* and *A. sputator* are also widespread, but they are clearly not important in the southern regions. The active ingredient of the *A. sordidus* pheromone (geranyl hexanoate) was effective in capturing other species, specifically *A. rufipalpis* in southeastern regions, *A. gallicus* in Switzerland, and some specimens *of Agriotes flavobasalis* in Portugal.

The response of the different species to the different monitoring tools was very variable. For example, very high numbers of *A. lineatus* males were captured by the sex pheromone traps at sites (e.g. Croatia, Romania) where bait traps for larvae and soil sampling recovered few or no larvae specimens. In contrast traps for *A. ustulatus* caught 2-3 times less adults than the *A. lineatus* traps cited above in fields with conspicuous *A. ustulatus* larval populations. Generally the sex pheromone traps proved to be a much more sensitive tool than soil sampling and bait traps for larvae. For all species traps were able to detect wireworm populations below those that can be reliably detected using soil sampling and bait trapping. In addition pheromone traps proved to be effective in detecting the regional variations of swarming patterns of the same species. The peak of the swarming could be clearly detected for all the species (e.g. *figure 9*). Despite the fact that very different regions were studied the start of male emergence and their peaks of activity did not vary greatly between sites for individual species. However, large differences were observed between different species.

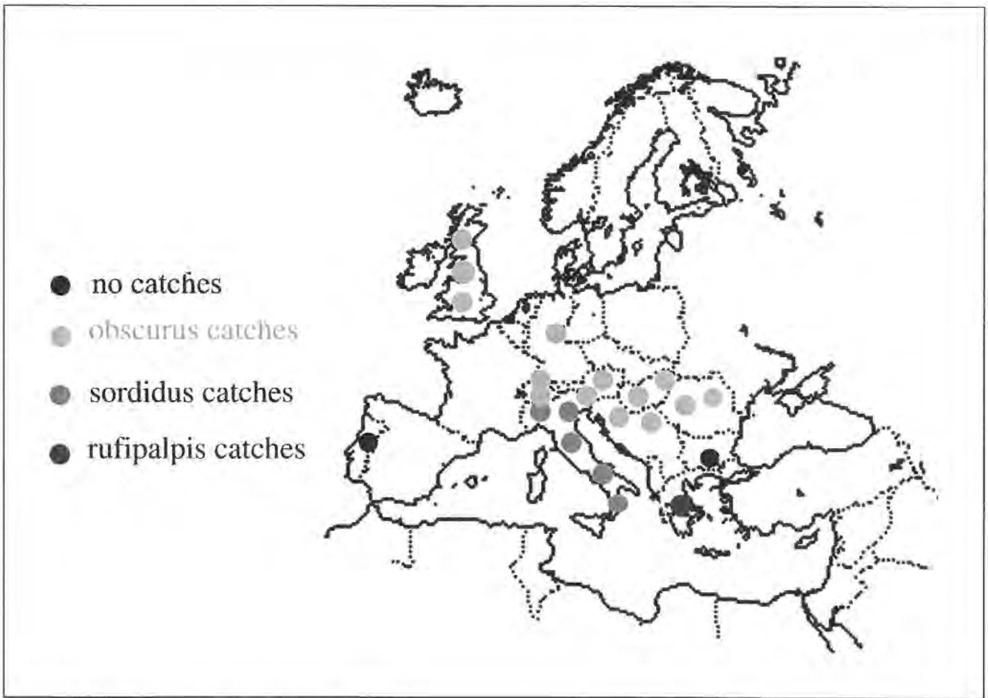

Figure 2 - *A. obscurus* bait 1998 - 2001

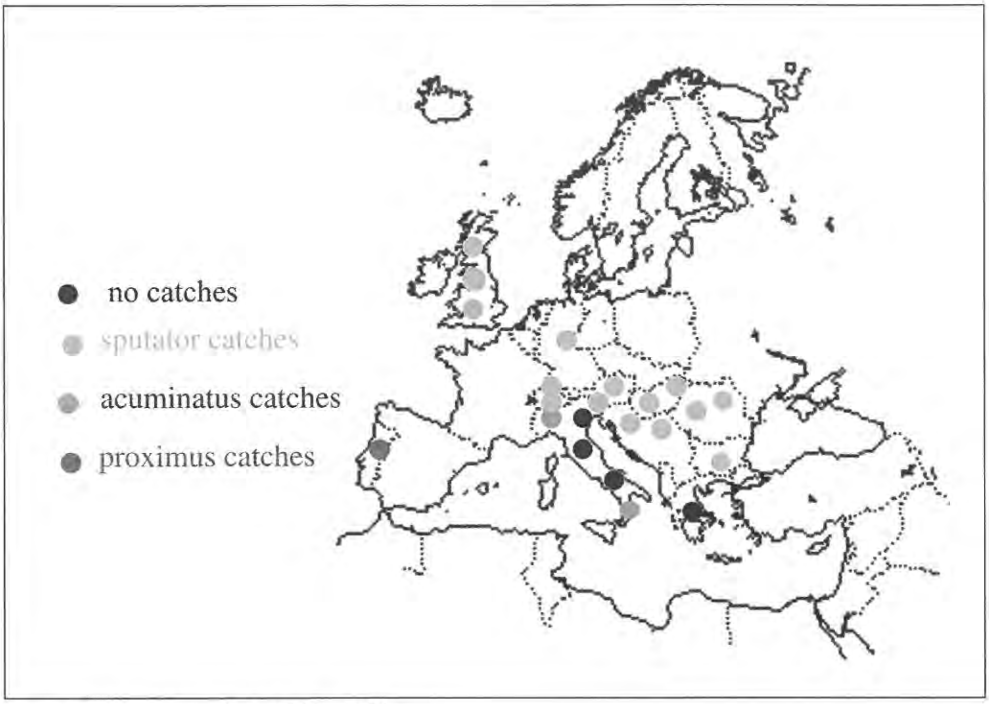

Figure 3 - *A. sputator* bait 1998 - 2001

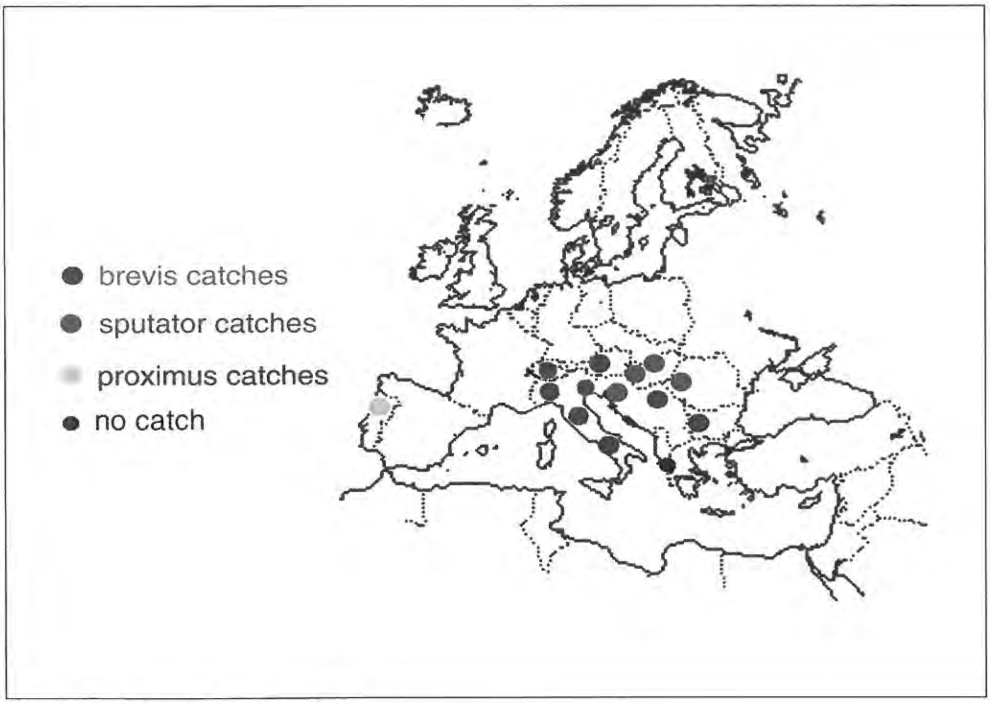

Figure 4 - *A. brevis* bait 1998 - 2001

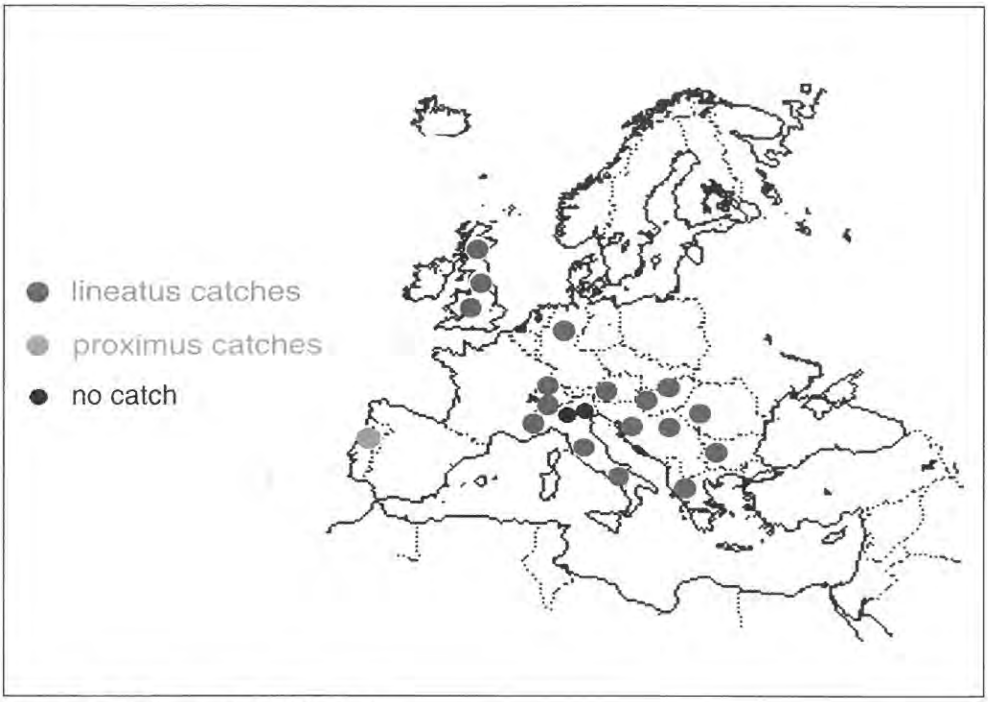

Figure 5 - *A. lineatus* bait 1998 - 2001

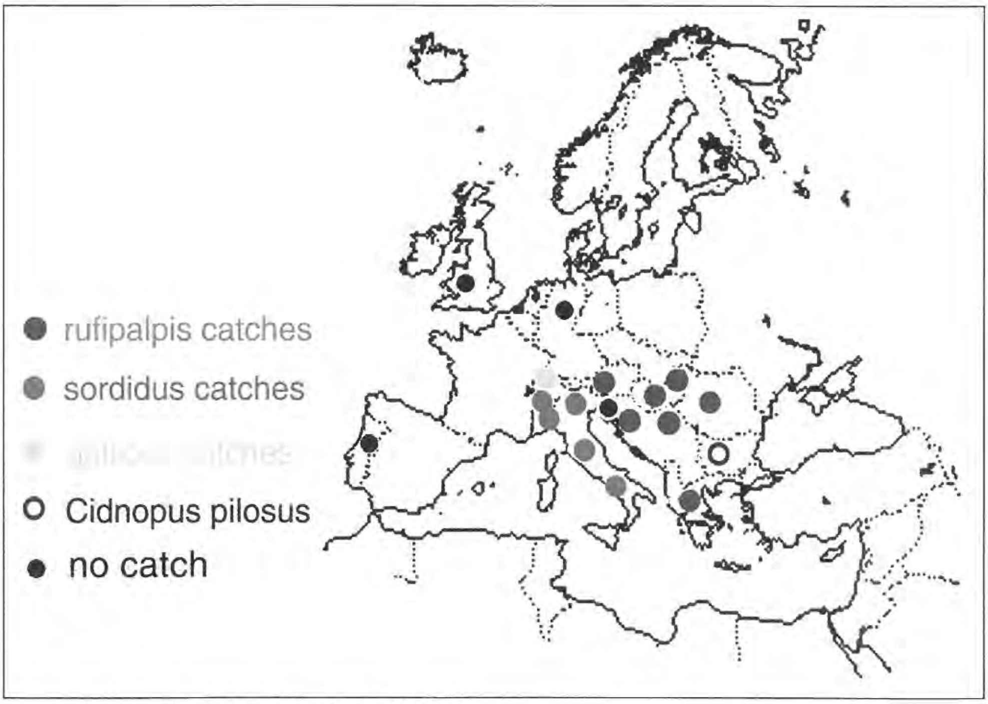

Figure 6 - *A. rufipalpis/sordidus* bait 1998-2001

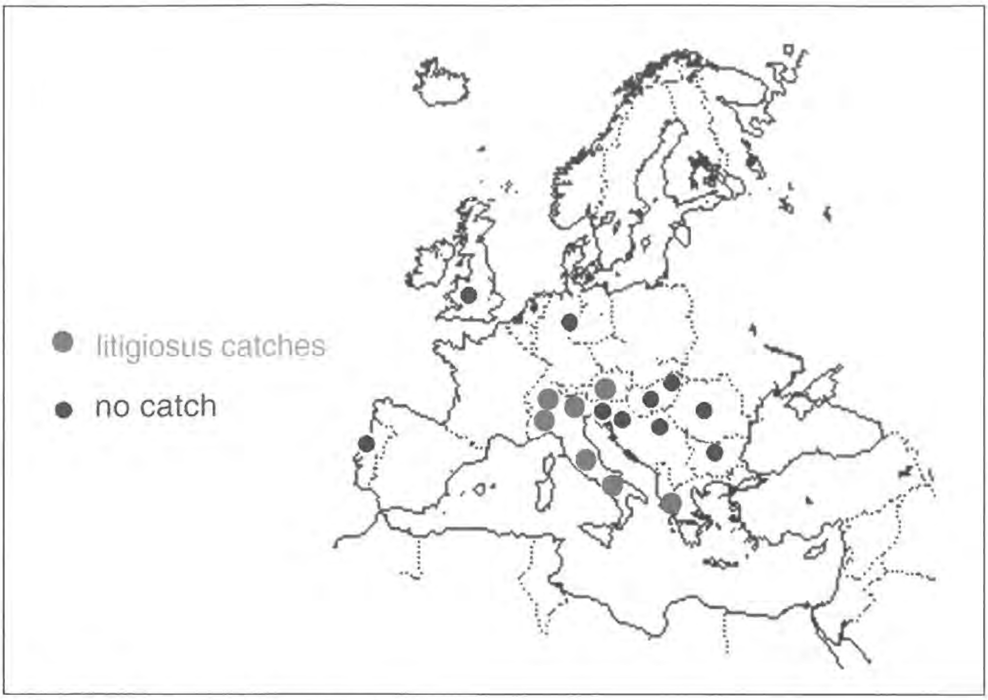

Figure 7 - *A. litigiosus* bait 1998-2001

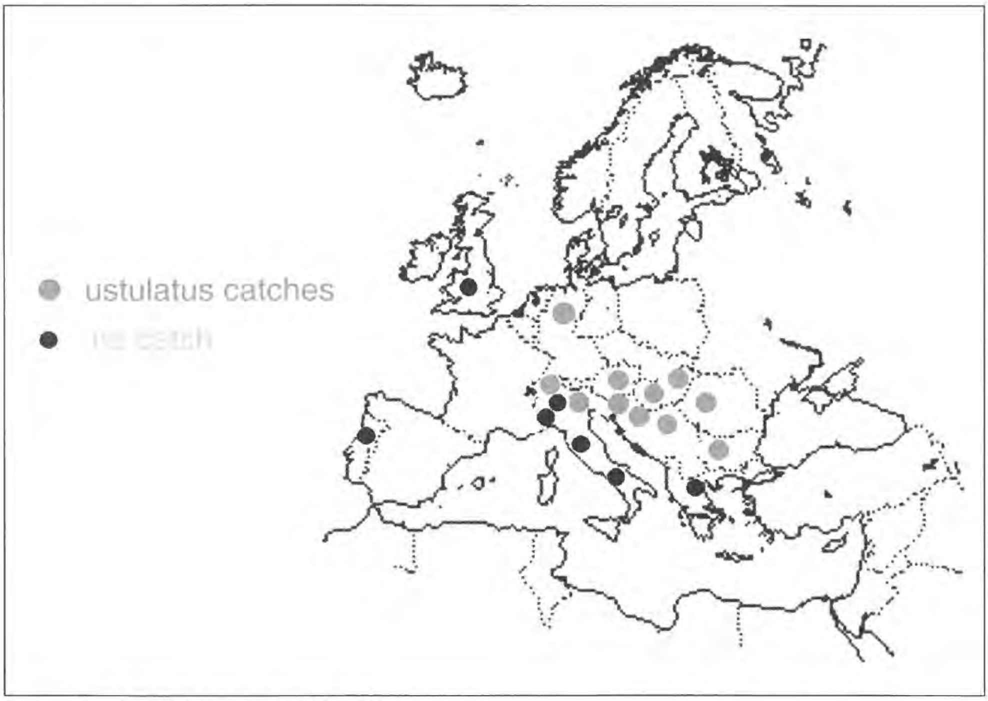

Figure 8 - *A. ustulatus* bait 1998-2001

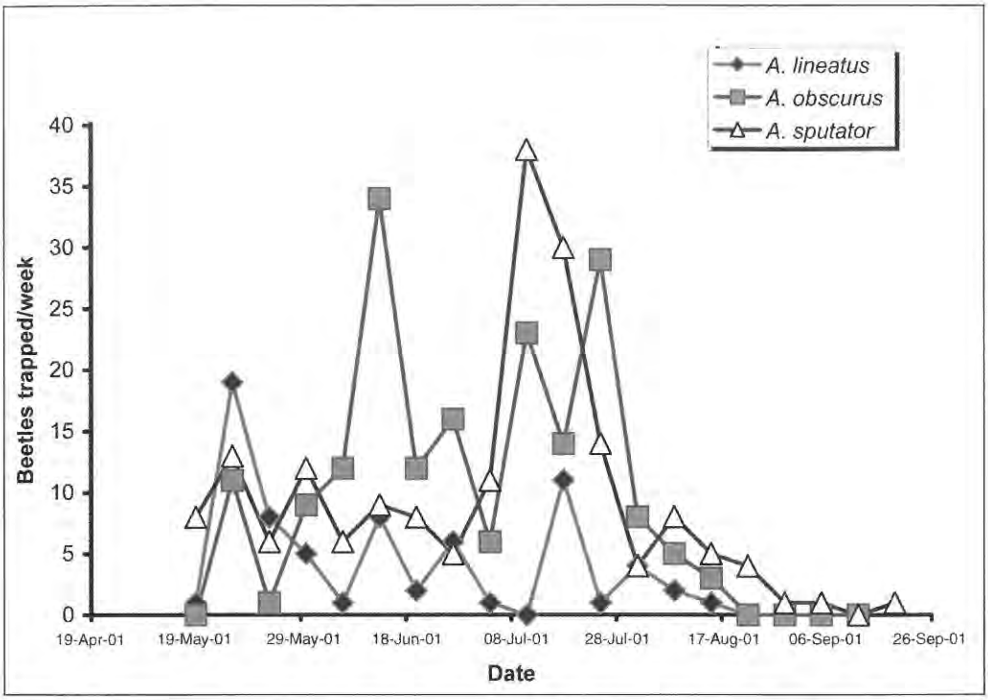

Figure 9 - Swarming patterns of 3 *Agriotes* species at Buckfastheigh, UK, in 2001

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PREVIOUS RESEARCH OF MONITORING OF *AGRIOTES* SPP. WITH SEX PHEROMONES

A.Ester, K. van Rozen,
Applied Plant Research (PPO),
P.O. Box 430, 8200 AK, Lelystad, The Netherlands

F.C. Griepink,
Plant Research International (PRI),
P.O. Box 16, 6700 AA, Wageningen, The Netherlands

Abstract
Wireworms (*Agriotes* spp.) is the main soil pest in many arable crops. Current chemical protection is primarily based on soil treatment with chlorpyrifos and ethoprophos. This soil protection depends on presence or absence of wireworms by baiting. PPO is developing a new strategy to control wireworms by monitoring the adult click beetles. The click beetles were trapped by pitfall traps. The main components of species specific sex pheromones were used to monitor three *Agriotes* spp. in different types of vegetation. *A. lineatus*, *A. obscurus* and *A. sputator* were abundant. Most *A. sputator* were caught in the verge only. *A. lineatus* and *A. obscurus* were common in grass seed, barley and the verge. A mixture of 95% geranyl octanoate and 5% geranyl butanoate attracted nearly 100% *A. lineatus*. The mixtures used to attract *A. obscurus* (50% geranyl octanoate and 50 % geranyl hexanoate) and *A. sputator* (80% geranyl butanoate and 20% farnesyl hexanoate) were rather species specific.

Key words: Sex pheromones, *Agriotes* spp., arable crops.

INTRODUCTION

Wireworms (*Agriotes* spp.) are larvae of click beetles which live in the soil for 3 to 5 years. They are known as an economic pest mainly in arable crops. Potatoes can be affected severely by wireworms. A soil sampling method was developed in the past (Salt & Hollick, 1944), but this is a laborious and not very effective method. Although baiting offers a more efficient method of assessing wireworm populations than taking soil samples, it can only be used as a presence/absence indicator of wireworm infestation (Parker, 1996). An adequate threshold for use in a supervised control system is not available.

A new approach to predict wireworm infestation may be monitoring the adult click beetles with sex pheromones. Pheromone compounds have already been identified for several *Agriotes* species (Yatsynin *et al.*, 1996).

The aim of this previous study was to determine the presence of certain *Agriotes* spp. and the specificity of sex pheromone components to these species. Flights of certain click beetles were monitored and peaks determined. The research focused on four types of vegetation related to the abundance of *Agriotes* spp., namely grass seed (*Festura rubra* L.), summer barley (*Hordeum vulgare* L.), potatoes (*Solanum tuberosum* L.) and the verge of a road.

MATERIALS AND METHODS

Pheromones

In 1998 and 1999 different sex pheromone mixtures were formulated and applied to catch three different species of click beetles (*table 1*). The pheromone components of *A. lineatus*, *A. obscurus* and *A. sputator* are primary or secondary components of the complete sex pheromone (Yatsynin *et al.*, 1996). In 1998 and 1999 each dispenser contained 20 mg of pheromone. In 1999 the ratio of the *A. sputator* pheromone was changed (*table 1*).

Table 1 - The pheromone mixtures used for specific click beetles, 1998 and 1999

Agriotes spp.	Pheromone components
A. lineatus	(A) = 100 % geranyl octanoate
A. lineatus	(B) = 95 % geranyl octanoate and 5 % geranyl butanoate
A. obscurus	(C) = 50 % geranyl octanoate and 50 % geranyl hexanoate
A. sputator (1998)	(D) = 50 % geranyl butanoate and 50 % farnesyl hexanoate
A. sputator (1999)	(E) = 80 % geranyl butanoate and 20 % farnesyl hexanoate

Traps

As click beetles move mainly on the soil surface, 3 litre pitfall buckets (Ø 18 cm) were used, to monitor their activity. Each trap was filled with 0.5 l water and 3 ml surfactant to reduce the surface tension. Traps were covered by white lids 5 cm above soil surface. Dispensers were placed under the middle of the lid. The lids each had three-iron feet, to press and secure them into the soil.

Experimental conditions

The experiments were done at the experimental fields of PPO in Lelystad. The experimental lay out is given in *table 2*. The pheromone traps were randomised within three replicates, with a distance of 20 m between the traps and 40 m between the replicates. *Agriotes* spp. were monitored in four types of vegetation, namely grass seed, barley, potatoes and the verge. The numbers of captured click beetles were counted and the species were identified. Observations were mainly done every three or four days, or once a week in periods when click beetles catches were low. The numbers of click beetles were corrected into three-days intervals (*figures 1 – 4*) to exclude peaks caused by the variations in observation intervals.

Table 2 - Characteristics of conditions and design of field trials 1998

	Year	Year
	1998	1999
Crop Sowing time Previous crop	Grass seed; variety: Mocassin 24th October 1996 Winter wheat	Grass seed; variety: Mocassin 23rd October 1997 Winter wheat
Crop Planting time Previous crop		Potatoes; variety: Turbo 3rd/4th May 1999 Sugar beet
Crop Sowing time Previous crop		Summer barley; variety: Reggae 1st April 1999 Potatoes
Period of monitoring	7th July – 13rd August	21st May – 17th August

Statistical analysis

Data were analysed using analysis of variance (ANOVA) in Genstat 5. From the ANOVA means, least significant differences (LSD) and F-probabilities were obtained.

RESULTS

The numbers of *A. lineatus, A. obscurus* and *A. sputator* caught are shown in *figure 1*. The pheromone mixture of 95 % geranyl octanoate and 5 % geranyl butanoate (B) was significantly ($P < 0.001$) more attractive to *A. lineatus* then geranyl octanoate (A) alone. These pheromone mixtures A and B caught exclusively *A. lineatus*, whereas pheromone mixtures C and D attracted some *A. lineatus* as well (data unpublished).

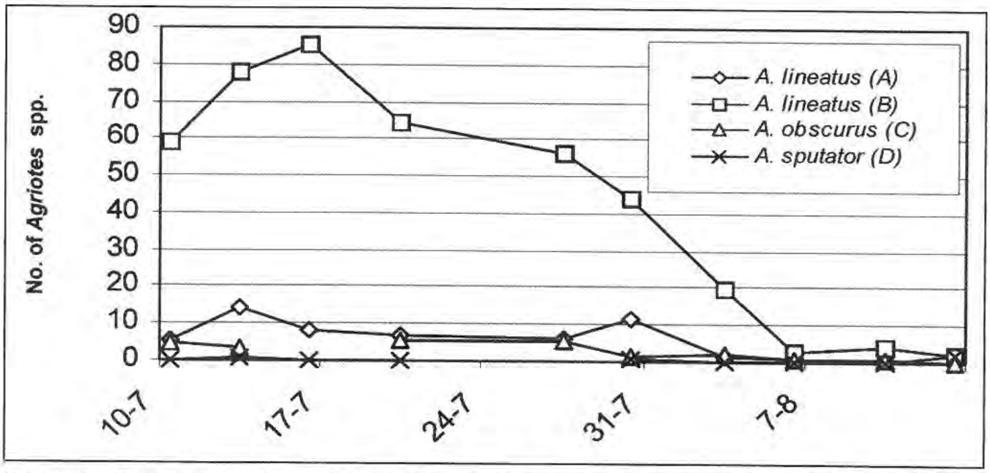

Figure 1 - Average numbers of *A. lineatus, A. obscurus* and *A. sputator* caught in pitfall traps with their specific pheromones (in brackets) in 1998

The lowest numbers of click beetles were caught in potatoes (*table 3*). *A. lineatus* and *A. obscurus* were both common in the other vegetation. The high abundance of *A. sputator* in the verge was striking. The mixture of 95 % geranyl octanoate and 5 % geranyl butanoate (B) gave nearly 100 % catches of *A. lineatus*. The catches with pheromone C and E were less specific.

Table 3 - Total numbers of *Agriotes* spp. caught by their own specific pheromone and the specificity (%) of the specific species (total catch per trap minus the catch of other species), 1999

Vegetation	1999					
	A. lineatus		*A. obscurus*		*A. sputator*	
	No.	%	No.	%	No.	%
Potatoes	53	100.0	23	67.6	3	75.0
Barley	522	99.4	369	84.4	12	52.2
Grass seed	498	100.0	122	76.7	3	75.0
Verge	407	99.8	345	93.2	205	91.9

The catches for 1999 are given in *figures 2, 3* and *4*. All three species were caught mainly at the end of May and the beginning of June. Thereafter, small peaks occurred around the 15th of June (*A. lineatus*) in grass seed and around the 18th and 2nd of June (*A. lineatus* and *A. obscurus*) in the verge. *A. lineatus* was caught in barley and grass seed crops and the verge, but less were caught in the potato crop in June (*figure 2*). *A. obscurus* were caught mainly in barley and in the verge at the end of May and the first week of June (*figure 3*). At the same time, beetles of *A. sputator* were caught in the verge only (*figure 4*).

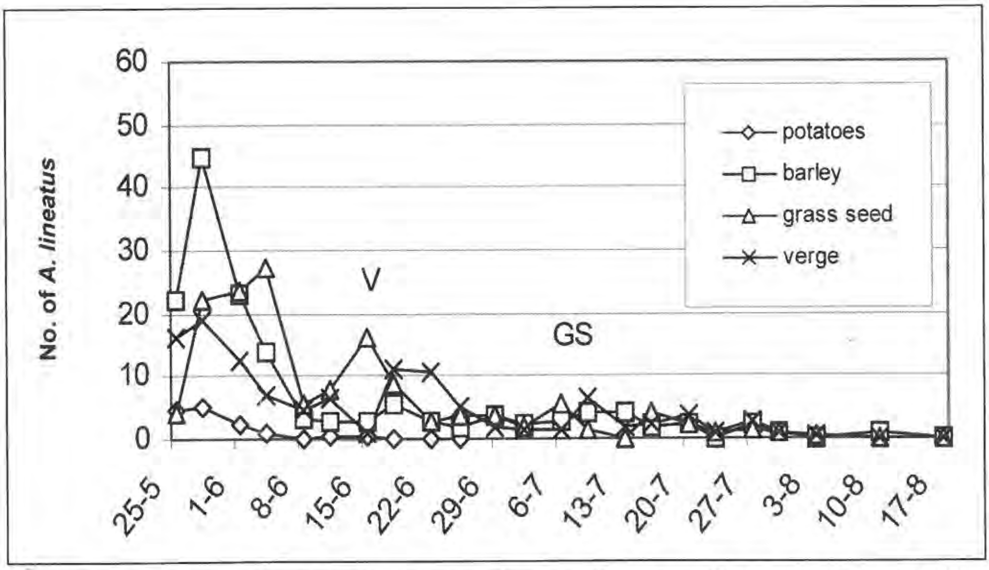

Figure 2 - Average numbers of *A. lineatus* caught with pheromone B per pitfall trap in 1999
* V and GS is the moment of mowing of respectively the verge and grass seed crop

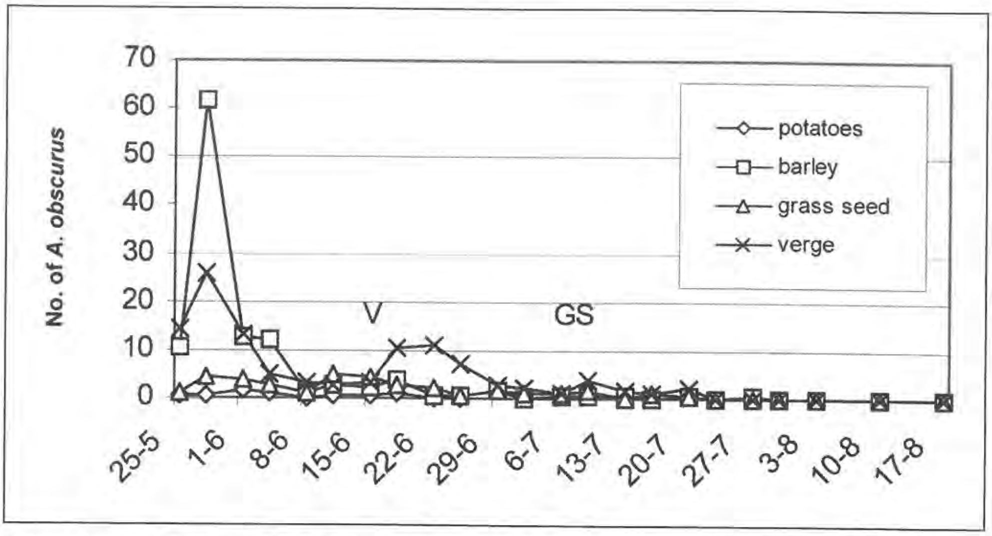

Figure 3 - Average numbers of *A. obscurus* caught with pheromone C per pitfall trap in 1999
* V and GS is the moment of mowing of respectively the verge and grass seed crop

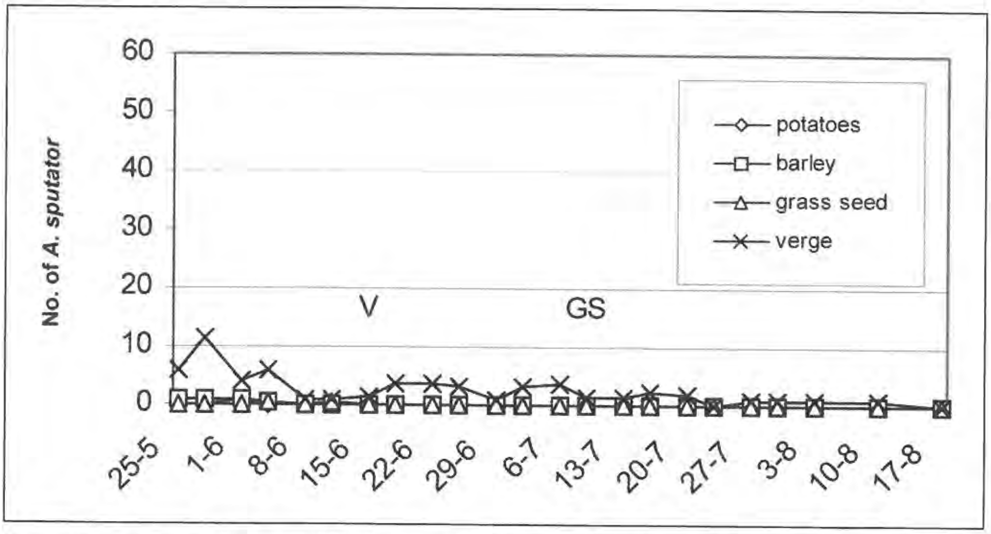

Figure 4 - Average numbers of *A. sputator* caught with pheromone G per pitfall trap in 1999
* V and GS is the moment of mowing of respectively the verge and grass seed crop

Discussion

A. lineatus, *A. obscurus* and *A. sputator* were abundant in the verge and in field crops. Kudryavtsev *et al.* (1993) predicted the distribution of *A. lineatus* and *A. obscurus* in northern Europe and *A. sputator* in Central Europe.

Monitoring *Agriotes* spp. with the specific sex pheromone mixtures gave a good indication of presence and peaks of these populations. The specificity of the pheromone mixtures used is acceptable for these different *Agriotes* spp. In 1998 and 1999 the main peaks were immediately after installation of the pitfall traps and pheromones. This could have been be caused by the relative late start of monitoring. Peaks catches of *A. lineatus* and *A. obscurus* in the verge were related to mowing of the vegetation. Perhaps the strong increase in 'green leaf volatile' concentration during mowing had a synergistic effect on the attractiveness of the pheromones. *A. sputator* was only common in the verge and may be less important as harmful species in arable crops. In contrast, *A. lineatus* and *A. obscurus* showed no clear preference for the grass seed crops, barley or the verge. This may be due to the previous crops in the different fields. Parker & Seeney (1997) mentioned, that a high abundance of *Agriotes* spp. was associated to a lower bulk density of the soil and longer grass duration. As the verge was more sandy than the other sites with a long duration of a mixed vegetation, this could indicate that *Agriotes* spp. abundance in certain habitats is rather species specific. Although potatoes were not expected to be a host for adult *Agriotes* spp., small numbers were found early in the season. Wireworms maybe develop into adult click beetles in the summer, hibernate in the soil and start flying early the next year. This research showed that sex pheromones can be used to predict the presence of different *Agriotes* spp. in arable crops.

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OBJECTIVES OF THE RESEARCH ON CLICK BEETLE SPECIES IN THE KUBAN REGION

Venyamin G. Yatsynin, *Elena V. Rubanova*,
Krasnodar Lukyanenko Research Institute of Agriculture,
Krasnodar 12, 350012, Russia

Abstract

Wireworm species have been regarded as a danger for the crops in Kuban region (Southern Russia) for many years. Over the past years wireworm outbreaks were observed mainly in fields where high amounts of soil insecticides had been applied to crops in the previous years. The main click beetle species detected in Kuban region are: *Agriotes tauricus Heyd.*, *A. gurgistanus Fald.* and, usually less harmful, *A. sputator L.*, *A. lineatus L.*, *A. ponticus Step.*, *A. obscurus L.*, and *Melanotus fusciceps Gyll.*, whose pheromone has not been completely studied yet.

The current main objectives of the research on click beetles in the Kuban region are the following:

- identification and synthesis of click beetle female pheromones attracting males;
- designing a new pheromone trap, not catching beneficial insects, for monitoring and mass trapping for integrated pest management;
- development of repellent to wireworms among substances derived from tartaric acid to be used as seed dressing;
- creating pheromone analogues for click beetles. For example thiogeranyl butyrate, *trans* - geranyl crotonoate or L - a - alanine geranyl ester, reconstructed forms of a single pheromone substance of *A. gurgistanus Fald.* might be added to synthetic pheromones to obtain synergistic effects and therefore a more effective tool for mass trapping or mating disruption.

Key words: *Agriotes*, pheromone, identification, synthesis, control.

INTRODUCTION

In the Kuban region wireworms caused conspicuous yield losses over the past years. Recently wireworm populations further increased. In fact in 2001 the Regional Plant Protection Station ascertained that 111,000 ha out of 226,100 surveyed revealed the presence of wireworm populations. Where soil sampling was properly done, wireworm populations between 10 and 30 specimens per square meter were estimated. These reported data are not complete since only the one-fifth part of the fields yearly planted with maize, sugar beet, sunflower was actually sampled. So far the chemical structure of the most harmful *Agriotes* species has been identified both in the Kuban region (Yatsynin *et al.*, 1996) and many other European countries (Furlan and Tóth, 1999). Anyway knowledge on the pheromones of important species as *Melanotus fusciceps Gyll.*, *Selatosomus latus F.* and *Selatosomus aeneus L.* still remains insufficient. This research should be addressed to optimizing pheromone components for pest control not only for monitoring. In order to meet this aim a specific research on analogues of females pheromones containing sulphur, nitrogen, fluorine, selenium and other heteroatoms is being conducted. It has already been demonstrated that antennal receptors may respond to analogues of insect sex pheromones (Bengtsson *et al.*, 1990; Subchev *et al.*, 1989, 1994) or be inhibited by them (Berger *et al.*, 1987; Camps *et al.*, 1990). Obviously to implement a mass trapping strategy effective compounds should be available.

The strategy proposed to control wireworm species damaging maize crops is based on two basic actions:

1) protection of young plants by using repellent compounds as seed dressing;
2) prevention of the reproduction of click beetle species as a result of mating disruption implemented by placing out effective sex pheromone traps.

Materials and Methods

The study was conducted both in laboratory and field conditions.

Laboratory experiments

Pheromones, their structural analogues and repellents were synthesized by the authors in the Krasnodar laboratory according to established methods of organic synthesis and subsequent purification. The chemical structure of the synthesized substances was in addition checked by GC – MS analysis and IR – spectroscopy.

Traps were made of plastic materials at laboratory level.

Field experiments

Trials were carried out in fields with wireworm populations of at least 12 – 15 larvae/m^2. Distance between the traps: at least 30 meters.

The pheromone dispensers were pieces of medical rubber hose 10 mm long and 8 mm in diameter. The dosage was at least 5 mg of pheromone compound per trap.

Period of monitoring: June – August for *A. tauricus* and *A. gurgistanus*.

Seed dressing with repellent compounds: the substances were dissolved in ethyl acetate and mixed with talk before dressing maize seeds. 30 seeds per each treatment were sown in May in a field (Armavir) where more than 20 *A. tauricus* larvae/m^2 had been detected.

Results and Discussion

The studies based on bait traps and soil sampling have shown that in several areas of Kuban region a gradual increase of *A. tauricus* and *A. gurgistanus* populations occurred. This might be due to the following basic reasons:

- constant increase of pesticide applications (mainly pyrethroid);
- prevalence of crops like cereals and sunflowers (63.1%);
- changes of agronomic practices especially with regard to tillage of soil surface;
- increase of the number of fields irrigated in the period of egg laying of the pest;
- absence of click beetle control during the reproduction period.

However the main reason should be the alteration to fauna equilibrium caused by the increase of soil insecticide applications where the rules for application of pesticides are usually not properly followed. As a consequence of this in these sites, where potatoes are often planted, the number of wireworms has sharply increased to 18 – 20 larvae/m^2 and more in some cases (*table 1*). Also in other farms where high amounts of soil insecticides were applied it is seldom to find predators of soil insects, which may feed on elaterid eggs and larvae.

Table 1 - *A. tauricus* larval infestation levels in different areas of Kuban region, Russia between 1990 and 2001

Area	Soil insecticide application	Number of larvae/ m^2	Total number of insects caught in a season	
			Carabids	*A. tauricus* males
Coast of the Kuban river:				
large farms	constant, following instructions	5 –6	120 – 160	4200 – 5500
private farms	constant, not following instructions	8 – 10	60 - 80	6900 – 8400
East area:				
rich farms	constant and repeated	14 – 20	10 - 40	12200 – 18100
Western area:				
poor farms	sometimes, low dosages	0,4 – 0,6	470 – 830	340 – 520
private farms, main crop potato	constant, not following instructions	12 - 18	30 - 60	10400 – 15700

As to seed dressing it would be preferable to use repellent compounds instead of chemicals dangerous to nature. Unfortunately few of the repellent compounds patented against mosquitoes, flies, ants and other pests have a potential to be used to keep away wireworms. During the research it was observed that isovaleric acid effectively kept away *A. tauricus* larvae from seeds. However in the pure state this acid is phytotoxic and decreases seed germination. Therefore esters of isovaleric acid were synthesized and tested in field conditions. The best results were given by diisovalerate tartaric diisoamyl ester (*table 2*).

The possibility of implementing successfully mass trapping against *A. tauricus* and other species depends on the features of trap design. The traps made in Estonia, such as "Estron" proved to be effective in field conditions with the exception of "Estron – 1" but had two important shortcomings:
- catching of useful species like carabids;
- their design does not allow easy entrance of click beetles.

Table 2 - Estimation of repellent action of different isovaleric acid esters used as seed dressing on *A. tauricus* larvae (Kuban region, 1997)

Substances	10 days after sowing		
	Total number of plants emerged	Number of plants damaged	Number of wireworms near the plants
Untreated	25	13	8
Insecticide ("Semaphor", FMS, US)	22	5	1
Isoamyl isovalerate	26	6	9
Tartaric diisovalerate diisoamyl ester	29	4	0
Thymol isovalerate	12	9	15
Hydroquinone diisovalerate	24	7	12
sec – Phenethyl isovalerate	4	4	18

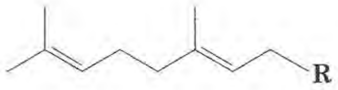
The trap 'Kniphan – 1" has no obstacles for click beetles attracted so that an increase of male captures was observed, however, this trap still does not prevent *Carabus* species from entering the traps. The new trap "Kniphan - 2" does not have a conic form, allows high adult captures without catching carabids (*table 3*). Such traps were used in 2001 to carry out *A. tauricus* mass trapping trial in a 100 ha field. No carabids were caught despite the fact that over one million of male click beetles were captured.

Table 3 - Captured of *A. tauricus* males in five different traps (pheromone dosage 10 mg, n=5, Krasnodar region, 2001)

Type of traps	Total number of insect caught		Average number of males/trap
	A. tauricus	Carabids	
Conic traps: Estron – 1	144	228	29
Estron – 2	2584	471	517
Estron – 3	2061	303	412
Kniphan - 1	4171	119	834
Pail traps: Kniphan - 2	5667	0	1133

The conventional theory that even insignificant modifications of chemical structure of insect sex pheromones inevitably leads to sharp reduction of biological activity or complete inactivation is not always true. Our research demonstrated that chemical modifications of female produced pheromones of *A. gurgistanus* by replacement of one chemical element does not always result in loss of biological activity. For example, thiogeranyl butyrate (*table 4*) only slightly decreased the attractiveness of the natural pheromone but increased evaporation.

Table 4 - Field attraction of A. gurgistanus males to traps baited with pheromone analogues with heteroatom (Krasnodar region, Bruchovetskaj area, 2001); n=5, p<0,005



Substances (5,0 mg), R	FW	Mean number of males captured per trap
$OCOCH_2CH_3$ (geranyl butyrate – control)	224	685±18
$SCOCH_2CH_2CH_3$	240	554±21
$SCSCH_2CH_2CH_3$	256	59±6
$SeCOCH_2CH_2CH_3$	287	1137±72
$NOCOCH_2CH_2CH_3$	237	85±14
$OCOCH(NH_2)CH_3$	225	943±49
$OCOCH_2CH_2NH_2$	225	171±12
$NHCOCH_2CH_2CH_3$	225	26
$OCOCH=^{E}CHCH_3$	222	796±27

The introduction of selenium in a pheromone molecule caused the increase of hydrophobic property of the substance, the increase of molecular weight but nevertheless geranylselenol butyrate attracted more males than natural geranyl butyrate. Geranyl crotonate is not less effective than the natural compound in capturing males. The inclusion of nitrogen in the molecule of the natural component interferes with reception of biologically active connections; L – alanine geranyl ester appeared especially active. Since the latter has a lower evaporation rate than geranyl butyrate, this substance may allow a prolongation of activity of blends of different biologically active substances.

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FUTURE EUROPEAN PRIORITIES FOR WIREWORM RESEARCH

William E. Parker,
ADAS, Woodthorne, Wolverhampton, WV6 8TQ,
United Kingdom

Lorenzo Furlan,
University of Padova, Agripolis
Via Romea 16-35020 Legnaro PD, Italy

Miklos Tóth,
Hungarian Academy of Science, Budapest,
Pf 102, H-1525, Hungary

Abstract
Wireworms (*Agriotes* spp.) are important pests of agricultural crops throughout Europe. Several countries are now working on a range of aspects of wireworm biology, risk assessment and control but those doing research on wireworms in Europe are not fully aware of the efforts of their colleagues in other countries. This means that there has been some duplication of effort, lack of standardisation or comparison of experimental techniques, and no overt prioritisation and co-ordination of research goals. This paper reviews current research priorities and suggests possible ways of developing European Commission (EC)-funded scientific collaboration between wireworm researchers in Europe.

Key words: wireworms, research priorities, European Commission.

INTRODUCTION

Wireworms are the soil-dwelling larvae (grubs) of click beetles (family Elateridae). Over 40 species from at least 12 genera are recognised worldwide as important agricultural pests Radcliffe *et al.*, 1991, Jansson & Seal, 1994). The type of crops attacked are very varied, and include maize and other grain crops (wheat, barley, sorghum) and vegetable root crops (sugar beet, potato, carrot), oil crops (sunflower) and grassland.

The wireworms of economic importance found in Europe are principally species in the genus *Agriotes.* This is a different species range from that found in North America (dominated by the genera *Limonius*, *Melanotus* and *Conoderus*), the main geographical focus of wireworm research during the 1970's and 1980's. Although there are similarities in biology and management between species, the substantially different cropping regimes, climate and available control measures in North America mean that a European dimension to wireworm research is essential.

Indeed, during the 1990's the focus of wireworm research in the world has shifted to Europe (see Parker & Howard, 2001). Wireworms are also relatively unusual amongst European pests in having a pan-European distribution, spanning both northern and southern European countries from the United Kingdom to Greece. They are also recognised pests in many eastern European countries such as Romania, Hungary, Poland and Russia.

Wireworms have long life cycles. Depending on species and geographic location, it can take two to four years for them to complete their development from egg to adult on the basis of the information available so far. Identifying whether wireworms are present in a particular field can be difficult and control measures are rarely fully effective. As a result, farmers often perceive that repeated application of broad-spectrum soil insecticides (principally organophopshorous and carbamate compounds) is required to prevent wireworm damage. In Europe, a sound scientific basis for this intensive use of insecticides for the control of wireworms is lacking. It is very likely that further research into novel methods of risk assessment and control could lead to significant reductions in insecticide use, with associated environmental and economic benefits.

In recent years, a number of European countries have initiated research programmes on wireworms covering topics such as biology, novel risk assessment techniques, thresholds and chemical and non-chemical control. However, active wireworm researchers in Europe are not fully aware of the efforts of scientists in other countries. This means that there has been some duplication of effort, lack of standardisation or comparison of experimental techniques, and no overt prioritisation and co-ordination of research goals. Some 'unofficial' co-ordination of work has been done under the auspices of the International Working Group on *Ostrinia* and other maize pests (IWGO), but this needs to be extended into all areas of European wireworm research. This paper reviews current research priorities and suggests possible ways of developing European Commission (EC)-funded scientific collaboration between wireworm researchers in Europe.

RESEARCH PRIORITIES

As a starting point for formulating a bid to the EC (assuming a suitable funding route can be identified, see below), we propose that the following research themes are those which have the highest priority in terms of preventing over-use of insecticides for wireworm control:

Theme 1: Biology of wireworms of agricultural importance

Research work should be done under controlled experimental conditions to determine important aspects of the biology of the species present in each country. The most critical of these is the duration of the larval (wireworm) phase of the life cycle. Comparative work done on the same species in widely separated geographical locations will help determine the extent to which life cycle duration is innate or governed by climatic factors. Methodologies for doing this type of research have already been developed and shown to be effective (Furlan, 1996, 1998). This theme may also include work on the local dispersal of adult click beetles. This work will clarify which species are important, and help determine the selection and timing of appropriate control strategies in participating countries.

Theme 2: Development and validation of novel risk assessment techniques

Further research is required to continue the development and validation of novel methods of identifying the level of wireworm infestation in individual fields. This will specifically cover the evaluation of pheromone traps for adult click beetles (Furlan *et al.*, 1996, 1997), and the relationship between catches of click beetles and the level of wireworm infestation in the soil. Some work has already been done on this latter topic (see papers in these Proceedings) but there is scope to do considerably more under a range of climatic and agronomic conditions. This work should allow infested fields to be identified well in advance of growing crops susceptible to wireworm damage, and allowing management strategies for specific crops to be modified accordingly.

Theme 3: Development of sustainable control techniques

Work should be done to determine the level of damage caused by different population of wireworms to crops grown in participating countries. This work will improve the scientific basis of predicting damage caused by wireworms, and will enable those fields unlikely to benefit from insecticide treatment to be identified reliably. This will reduce the need for soil insecticide use. Work on the response of wireworms to treated soil may also help to improve the placement of soil-applied insecticides.

Extension priorities

It is essential that a planned effort is made within the Action to ensure that the research is publicised at conferences and published in a co-ordinated way in the scientific literature. Specific efforts must also be made to transfer the information gathered to farmers in all participating countries. This will be done by writing a booklet on best practice in wireworm management that can be translated into the languages of participating countries, and used as a resource by national extension agencies from which to produce local promotional material. The booklet (text and illustrations) will also be posted on an open web-site that will also hold summaries of the detailed scientific information on which the booklet is based. The existence of the booklet and the web-site can be advertised through the national media in participating countries.

Possible funding routes within the European Union

There are two possible approaches to developing a proposal for EC-funded pan-European research on wireworms. Probably most of these approaches would allow the participation of some eastern European countries currently outside the European Union, as many of these are now pre-accession states and hence are eligible to participate in most EC research schemes.

To undertake a one or two year 'concerted action' (under either the EC Framework V or VI programmes), which provides funds for researchers to meet, exchange information and plan a full research proposal. A similar approach could be taken via a COST action – COST is not an EC programme, but is administered by the EC.

Both concerted actions and COST actions have the advantage of being relatively simple to apply for, but provide no money for actually doing research. However, this does rely on individual researchers having sufficient funds to continue their research until such time as a full research proposal is submitted and (although there is no guarantee of this!) accepted.

To proceed to the submission of a full research project proposal at the earliest opportunity. There are currently three possible routes for such a proposal.

To make a bid under the final round of the EC Framework V programme. Although the currently published deadline for the last submissions under this programme (18 October 2001) has finished, it is possible that there may be a further call early in 2002, with a submission deadline three months later.

To await the opening of the EC Framework VI programme. The content and scope of this is still under discussion, and is likely to have a substantially different set of objectives and priorities to Framework V. The interim time between this conference and the opening of Framework VI could be used to develop the basis of a proposal.

To submit a bid under the LIFE-Environment programme. This would have to be a demonstration project rather than a research project. Demonstration projects must set out to test an innovative solution to an environmental problem and lead to concrete, practical results of the project. They must be implemented at a scale that allows evaluation of the technical and economic viability of the large-scale introduction of this solution. On the basis of the currently published criteria, it is uncertain whether crop protection related issues are appropriate for funding under LIFE-Environment.

Most EC research schemes only provide part funding (up to 50% of eligible costs for research projects in Framework V, 30-50% of eligible costs in LIFE-Environment). Finding additional sponsorship to meet the remaining costs of the project is therefore a major issue for EC-funded research projects.

Conclusions

There is a clearly defined need for further European-wide research work on wireworms, and a well-defined community of researchers willing to participate. This is a very good starting point for developing an EC-funded research network or research project.

However, the difficulties of obtaining EC-funds for research, due both to the competitive nature of the bidding process and the administrative complexity involved requires considerable commitment from all concerned to maximise the chances of success.

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GENETICS OF RESISTANCE TO PINK STEM BORER ATTACK IN FLINT MAIZE POPULATIONS

Ana Butrón, Pilar Soengas, Pedro Revilla,
Amando Ordás, and Rosa A. Malvar,
Misión Biológica de Galicia (CSIC),
Apdo. 28, 36080 Pontevedra, Spain

Abstract

The most important maize pest in northwestern Spain is the pink stem borer (*Sesamia nonagrioides* Lef.). Evaluations under infestation conditions have revealed that some maize inbreds and populations could bring resistance genes to pink stem borer attack. More studies, however, should be addressed to look for new sources of resistance and to know how to manage those materials in a selection program to develop varieties resistant to *Sesamia nonagrioides*. The objective of this work was to know the genetic effects involved in the resistance to *Sesamia nonagrioides* in a diallel among diverse flint maize populations. That knowledge will allow us to design the most suitable selection program to develop resistant varieties. Ten flint maize populations, seven of them from diverse areas of Spain, two from Argentina, and one from United States, were crossed following a diallel design. The diallel was analyzed following Gardner and Eberhart's design. The 45 crosses, plus ten populations, plus a check hybrid were evaluated under infestation with *Sesamia nonagrioides* at Pontevedra (northwestern Spain) in 1998 and 1999. Tunnel length and general appearance of the ear were recorded as estimates of damage produced by the pink stem borer larvae on stems and ears, respectively. There were significant differences among genotypes and non significant genotype x environment interactions for tunnel length and general appearance of the ear. Variety effects were the only significant effects for tunnel length. Variety and average heterosis effects were significant for general appearance of the ear. Therefore, the additive gene action was the most important in the inheritance of the resistance to *Sesamia nonagrioides*. The population Basto/Enano levantino (Ba/El) showed the best variety effect for tunnel length and was among the populations with the most favorable variety effects for general appearance of the ear. Ba/El could be the base material in a recurrent selection program to develop flint maize resistant varieties.

Key words: *Sesamia nonagrioides*, pink stem borer, resistance, genetics, heterosis.

INTRODUCTION

The pink stem borer is the main pest of maize in northwestern Spain. Most of the larvae development takes place inside the maize plant and can provoke stem lodging, ear drop, and direct yield losses (Anglade 1961). Maize yield losses averaged on a set of 45 hybrids was the 15% of the yield of the insect-attacked free crop (Butrón *et al.*, 1999b). Larue (1984) reported that yield losses could reach up to 30% of yield. The use of maize resistant cultivars appears as one of the most promising methods of control within an integrated pest control strategy (Ortega *et al.*, 1980; Pathak, 1991). As a previous step to develop resistant cultivars, it is necessary to find sources of resistance. Many efforts have been devoted in that direction, but just a small part of the maize germplasm susceptible of being used in the areas affected by the pink stem borer attack has been tested (Malvar *et al.* 1993; Cartea *et al.*, 1994, Butrón *et al.* 1998; 1999a; b; Velasco *et al.*, 1999a; b). In Europe, most maize hybrids come from the cross between European flint maize and American dent maize inbreds (Moreno-González, 1988; Ordás, 1991). Flint maize material supplies adaptation to European conditions, while American dent maize brings high yielding potential. Some Spanish flint maize populations along with other populations were evaluated for resistance to *S. nonagrioides* (Malvar *et al.*, 1993). In that study, the flint maize population "Amarillo temprano de Aragón" stand up for being the most resistant to stem attack by *S. nonagrioides*. However, resistance to pink stem borer of flint maize populations has not been fully prospected.

Local flint maize varieties supply characteristics, such as early vigor, earliness, resistance to stem lodging, and relative resistance to drought stress. In addition, flint kernels possess a better ability than other kernels for developing high quality flours. Once sources of resistance have been found, studies should de addressed to determine the genetics involved in the inheritance of the resistance. That knowledge will allow us to design the most suitable selection program to develop resistant cultivars from those materials where resistance factors were detected. The Gardner and Eberhart (1966) analysis II applies to a group of populations and all possible crosses among them and divides genetic effects in variety and heterosis effects, which can be partitioned into mean, variety, and specific heterosis effects. The goal was to test if variability for resistance to *S. nonagrioides* was present among flint maize materials, and then, to determine the genetics involved in the resistance to pink stem borer attack.

MATERIAL AND METHODS

Ten flint maize populations from different origins were used (*table 1*).The varieties were crossed in a complete diallel without reciprocals in 1997, using paired rows with 15 plants per row. Five sets of paired rows were used for each cross, using each plant only once, as male or female. Also, each variety was multiplied to obtain seed in the same environmental conditions.

The 10 parental varieties and their 45 crosses plus the commercial check D.M.B. 15-70 were evaluated under infestation with *Sesamia nonagrioides* in the Northwest of Spain. Trials were planted in Pontevedra (42°24'N, 8°38'W, 20 m above sea level) in 1998 and 1999. Randomized complete block designs with three replications were employed to evaluate the treatments. In 1998, each experimental plot consisted of two rows with 17 two-plant hills, while in 1999 plots consisted of two rows with 15 two-plant hills. In both years, rows were spaced 0.80 m apart and hills were spaced 0.21 m apart. Plots were overplanted and thinned, obtaining a final density of approximately 60 000 plants ha^{-1}.

Table 1 - Flint maize populations studied

Population	Origin	Abbreviation†
Gallego‡	Northern Spain	Gallego
Gallego × Hembrilla Norteño‡	Northern Spain	Ga/Hn
Norteño‡	Northern Spain	Norteño
Norteño largo‡	Northern Spain	Nl
Fino‡	Center of Spain	Fino
Basto × Enano levantino‡	Eastern Spain	Ba/El
Tremesino‡	Southern Spain	Tremesino
Longfellow	United States	Longfellow
Amarillo precoz de Simone	Argentina	Simone
Relámpago ocho hileras	Argentina	Relámpago

† Abbreviations will be use to name the populations

‡ Sánchez-Monge (1962)

At silking, all plants from a row in each plot were infested with a mass of about 40 eggs of *S. nonagrioides* per plant. Eggs were placed between the main ear and the stem. Eggs were obtained in the lab following Eizaguirre's method (1989). At harvest, stems of infested plants from each plot were dissected, and tunnel length in centimeters was recorded. General appearance of the ear, rated on a 9-point scale (from 1, wholly damaged ear, to 9, ear without injury) was used as an estimate of damage produced by the pink stem borer larvae on ears.

Combined analyses of variance over years were made for each trait using the PROC GLM (SAS, 1989). The source of variation due to the diallel was divided according to the Analysis II suggested by Gardner and Eberhart (1966), that fits the population and population cross means to the linear model:

$Y_{ij} = m_v + 1/2\,(v_i + v_j) + h_{ij}$

where:

Y_{ij} : observed mean of the cross of varieties *i* and *j*.

m_v : mean of all varieties.

v_i, v_j : variety effects. Variety effects are the difference between a particular variety mean and the mean of all varieties

h_{ij}= observed heterosis of the cross of varieties *i* and *j*. Heterosis is measured as the difference between the mean of the two parental varieties and its cross. It is due to differences in gene frequencies among the varieties *i* and *j*, and to dominance. Besides, heterosis is subdivided in the following manner:

$h_{ij} = d\,(h + h_i + h_j + s_{ij})$

where:

$d = 0$, when $i = j$ and $d = 1$ when $i \neq j$.

h = average heterosis of mean of crosses respect to the mean of all varieties.

h_i, h_j : variety heterosis. The contribution of each variety to the expression of heterosis.

s_{ij} : the specific heterosis that occurs when variety i is mated to variety j .

Variety effects and mean, variety, and specific heterosis effects were obtained according to Gardner and Eberhart (1966), as well as the least significance difference to compare those effects.

RESULTS AND DISCUSSION

There were significant differences among years and populations for tunnel length and general appearance of the ear (*table* 2). The only subdivision of the variability among populations that was significant for tunnel length was the one due to variety effects. Hence, additive effects were the only significant gene effects in the inheritance of the stem damage by the pink stem borer. That agrees with some works previously published (Anglade and Bertin, 1968; Butrón *et al.*, 1999b). Cartea *et al.* (1999), however, found that the gene model involved in the inheritance of stem damage by the pink stem borer could vary from an exclusively additive to an additive-dominant model, depending on the germplasm considered.

Table 2 - Analysis of variance of the diallel crosses among 10 flint maize populations evaluated under infestation with S. nonagrioides in 1998 and 1999

Source of Variation	Degrees of freedom	Mean square	
		Tunnel length	General appearance of the ear
Year	1	1615.74*	70.2440*
Population	54	422.01*	2.1425*
Variety	9	1279.81*	6.9066*
Heterosis	45	250.45	1.1936
Average	1	85.55	5.8064*
Variety	9	232.28	0.4725
Specific	35	259.83	1.2472
Genotype × Year	54	204.82	0.9423
Error	215	221.64	0.9426

* Significant at the 0.05 probability level

On the other hand, two components of the variability among populations for general appearance of the ear were significant, the variety and mean heterosis components. Crosses would receive a better value than populations *"per se"* because crosses, generally, improve ear size and other yield components. Therefore, the inheritance of ear damage by pink stem borer larvae appears to be mainly additive as other authors suggested (Butrón *et al.*, 1998; Velasco *et al.*, in press). Cartea *et al.* (2001), however, found that dominance gene effects could be as important as additive gene effects.

The interaction year x population was not significant either for stem damage or ear damage. Therefore, mean and effect comparisons were performed with the data combined over years. Norteño x Longfellow, Gallego x Norteño, Ba/El x Tremesino, Ba/El x Relámpago, Tremesino x Relámpago, Ba/El x Ga/Hn, Tremesino x Longfellow, Ba/El x Longfellow and Ba/El showed the least stem damage (*table 3*). The population Ba/El had the most favourable variety effects for tunnel length (*table 3*). Ba/El might represent the remnant maize from the first introductions brought to Spain from the Caribbean area and Central America, while the remaining populations used in this study could be more related to North American maize (Revilla *et al.*, 1998). The population Tremesino and its crosses to other populations, excepting Gallego x Tremesino and Norteño x Tremesino, were among the genotypes least damaged on the ear (*table 4*). Tremesino, Fino, Relámpago, Longfellow, Simone, and Ba/El showed the largest variety effects for general appearance of the ear (*table 4*).

Table 3 - Means for tunnel length (cm) of 45 crosses (above diagonal) and 10 parental varieties (on the diagonal), and variety effects for those parental varieties (bold characters within brackets)

	Gallego	Ga/Hn	Norteño	NL	Fino	Ba/El	Tremesino	Longfellow	Relampago	Simone
Gallego	50.1(**-5.7**)	53.2	47.5	54.6	68.6	59.6	49.8	62.2	56.8	62.7
Ga/Hn		50.1(**-5.6**)	69.7	66.5	57.4	43.7	54.9	60.1	60.6	67.3
Norteño			57.2(**1.5**)	63.5	65.9	62.3	55.3	48.5	65.9	61.1
NL				57.9(**2.2**)	54.7	50.6	57.8	61.3	54.9	53.1
Fino					66.2(**10.5**)	56.6	51.4	52.5	57.0	65.3
Ba/El						32.1(**-23.6**)	47.1	40.1	45.4	58.4
Tremesino							49.5(**-6.3**)	41.4	44.7	60.3
Longfellow								56.2(**0.5**)	64.3	69.3
Relampago									65.7(**10.0**)	64.2
Simone										72.4(**16.6**)

LSD (0.05) = 16.6 for genotype means
LSD (0.05) = 16.5 for variety effects

However, mean and variety effect differences among populations for general appearance of the ear were not as large as those presented for tunnel length. Butron *et al.* (1998) and Velasco *et al.* (1999a; b) already detected that differences among populations and inbreds for ear damage by the pink stem borer are not too large.

As conclusions, additive effects were the most important effects in the inheritance of stem and ear damage produced by the pink stem borer. Since the population Basto/Enano levantino (Ba/El) showed the best variety effect for tunnel length and was among the populations with the most favorable variety effects for general appearance of the ear, Ba/El could be the base material in a recurrent selection program to develop flint maize resistant varieties.

Table 4 - Means for general appearance (from 1= bad to 9 = excelent) of 45 crosses (above diagonal) and 10 parental varieties (on the diagonal), and variety effects for those parental varieties (bold characters within brackets)

	Gallego	Ga/Hn	Norteño	NL	Fino	Ba/El	Tremesino	Longfellow	Relampago	Simone
Gallego	5.30 (**-0.95**)	5.85	5.45	6.75	6.75		6.59	6.51	6.22	5.78
Ga/Hn		5.92(**-0.33**)	6.43	6.93	7.43	6.25	6.99	6.57	6.43	6.56
Norteño			5.75(**-0.50**)	6.22	6.67	6.33	6.35	6.09	6.29	6.63
NL				5.87(**-0.50**)	7.37	5.99	7.11	6.22	6.50	5.81
Fino					7.00(**0.75**)	6.61	7.73	7.27	7.55	6.81
Ba/El						6.12(**-0.12**)	6.80	7.30	7.18	7.45
Tremesino							7.21(**0.97**)	6.64	7.49	7.41
Longfellow						5.91		6.40(**0.15**)	5.82	5.28
Relampago									6.67(**0.42**)	6.15
Simone										6.22(**-0.03**)

LSD (0.05) = 1.13 for genotype means

LSD (0.05) = 1.12 for variety effects

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YIELD LOSSES CAUSED BY LEPIDOPTEROUS PESTS ON SECOND CROP MAIZE IN ÇUKUROVA REGION OF TURKEY

M. Güllü,
Plant Protection Research Institute,
POBox 21, 01321 Köprüköy- Adana/Turkey
e-mail: mgullu83@hotmail.com

S. Kornoşor,
Çukurova University,
Faculty of Agriculture, Department of Plant Protection,
01330 Balcali-Adana/Turkey
e-mail: kornosor@mail.cu.edu.tr

Abstract

Lepidopterous pests, *Spodoptera exigua* Hbn. *Sesamia nonagrioides* Lef., *Ostrinia nubilalis* Hbn., *Pseudaletia unipuncta* Haw., *Heliothis armigera* Hbn. and *Spodoptera littoralis* (Boisd.) cause yield losses in economically significant level in second crop maize in Çukurova Region of Turkey. The yield losses caused by the lepidopterous pests on the most common hybrids, Pioneer P-3394', DeKalb DK-698', Funks G-4207', Cargill C-7993', Limagrain LG-60' and Sandoz PX-9540', were studied in Do?ankent Agricultural Research Institute between 1995 and 1996.

Experiments were set up in a randomized complete block design within a split-plot arrangement with 5 replications. Hybrid varieties were main plots and insecticide application (applied or non applied) was the sub plots.

Interactions among varieties, insecticide applications and variety by insecticide applications were found statistically significant at 0.01 and 0.05 levels in both years. Yield was higher at pesticide-applied plots comparing to non-pesticide applied plots. In 1995, yield increase due to pesticide application was over 30% for P-3394, DK-698, PX 9540, and LG-60; but about 20% for two other varieties. However, yield increase did not exceed 20% in 1996 for any varieties, even it was as less as 10% for P-3394. These data showed that yield losses due to the lepidopterous pests could reach 40% in varieties and years.

Key words: Yield losses, maize varieties, lepidopterous pests, second maize crop.

THE EFFECTS OF DIFFERENT IRRIGATION SYSTEMS ON INUNDATIVE RELEASE OF *TRICHOGRAMMA EVANESCENS* WESTWOOD (HYMENOPTERA:TRICHOGRAMMATIDAE) AGAINST *OSTRINIA NUBILALIS* HÜBNER (LEPIDOPTERA: PYRALIDAE) IN THE MEDITERRANEAN REGION OF TURKEY

S. Öztemiz,
Plant Protection Research Institute,
01321, Adana/Turkey

S. Kornoşor,
University of Çukurova,
Faculty of Agriculture, Department of Plant Protection,
01330 Adana/Turkey

Abstract

This study was conducted to determine the effects of different irrigation systems on inundative releasing the egg parasitoid, *T. evanescens* against *O. nubilalis* in maize in Çukurova Region of Turkey in the years of 1999-2000.

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.

T. evanescens was applied two times at a rate of 75,000 parasitoid/ha with 10 days interval at the beginning of the oviposition period of third generation of *O. nubilalis* in the second crop maize. It was found that irrigation was an important factor for effectiveness of *T. evanescens* and significant differences in two irrigation systems were obtained. The efficiency of *T. evanescens* was higher in the flooding irrigation than the sprinkle irrigation. Egg parasitism was 81.06% and 84.31% in the flooding irrigation; 66.37% and 69.26% in the sprinkle irrigation in 1999 and 2000 respectively. The reduction of infested plant with *O. nubilalis* was 80.00% and 88.31% in the flooding irrigation, 60.71% and 68.85% in the sprinkle irrigation respectively. Also, the yield was increased approximately 150-200 kg/per da in the flooding irrigation system than the other one.

In conclusion, maize is a very important and widely planted agriculture product in the Çukurova Region. Especially, *O. nubilalis* is a main pest on the second crop. So far, it was controlled at least three application in 10 days interval by chemicals. After this research, for releasing *T. evanescens* to *O. nubilalis* showed that the better control than the insecticides. Because of these, in near future to conduct the biological control against *O. nubilalis* with egg parasitoids will be effective in this region of Turkey.

METHODS FOR ASSESMENT THE EFFICIENCY OF CHEMICAL AND BIOLOGICAL TREATMENTS IN THE REDUCTION OF ECB (*OSTRINIA NUBILALIS* Hbn.) IN TRANSYLVANIA

Felicia Mureşan, D. Mustea,
Agricultural Research Station Turda Str. Agriculturii, 27,
3350, Turda, jud. Cluj România

Abstract

In this paper some methods for estimating of yield losses caused by European Corn Borer (*Ostrinia nubilalis* Hbn.) to maize crops are presented. All the methods are based on a large number of weighing and notations during 1998-2000. The 4 methods presented are: frequency of damages plants (K=0.219), mean number larvae of plants damaged (K=0.180 for one larva), degree of damaged plants in according to rating scale (1-5): (2=0.15; 3=0.30; 4=0.50; 5=0.80). In maize crops from Transylvania, the yield losses depending on attack frequency, attack degree and the number of larvae were between 15.4-23.7%, at 10 maize hybrids created at Agricultural Research Station Turda.

These damages caused by *Ostrinia nubilalis* Hbn. at maize crops, determinated the application of some biological and chemical methods unpolluting the medium and the feeding products. Such as biological methods in this pest control, a few species of *Trichogramma,* the biological products, the chitin inhibitory and some chemical selective products were used. The efficiency of chemical products was between 25-71%, of biological products was between 40-65% and of *Trichogramma* species between 41-75%, regarding the reduction attack of *Ostrinia nubilalis* Hbn.

Key words: Maize, *Ostrinia nubilalis* Hbn., the yield losses, the treatments.

INTRODUCTION

The consequence of application of some incomplete or incorrect technologies, practising monocultures, unrespecting the measures of cultural hygiene as maize stems remained in the field after harvesting, permitted the maintaining of one of the most pest-corn borer (*Ostrinia nubilalis* Hbn.). The attack of this pest in many ecological areas in Transylvania is about 30-95%(Mureşan *et al.*,1995; Mustea *et al.*,1995; Mureşan *et al.*, 1999), and the damages produced are considerable, till 40% from beans yield (Mustea *et al.*, 1975).

In reducing the population of this pest a special attention was paid to chemical and biological methods that form a technological part with an efficiency more than satisfactory in realizing the mentioned objective. Thus in the countries from UE through applying the biological and chemical treatments, the corn borer attack was reduced with 40-60% and the realized yield increase was about 2-6 q/ha (Baicu, 1992; Grinberg *et al.*, 1993). The researches in this stream were performed by Mustea

(1971); Mureşan (1997); Mureşan *et al.*, (1999); Mureşan (2000); Tancik *et al.*, (1999); Oztemiz *et al.*, (2000); Mark *et al.*, (2000).

The aspects pointed in this paper refer to assessment of chemical (insecticides) and biological (products and entomophagues) treatments efficiency depending on: attack frequency, the number of holes/plant, the number of larvae/plant, the number of cavities/stem, the cavities length, obtained yield.

MATERIALS AND METHODS

Between 1998-2000 studies were performed for 10 maize hybrids created and cultivated at the Agricultural Research Station Turda and in the influenced area of this, regarding the natural attack of corn borer (*Ostrinia nubilalis* Hbn.).

The studied aspects were referred to:

a) the frequency and degree of attack;
b) the number of larvae/plant;
c) the yield losses regarding these aspects.

In an experience with 11 variants, three repetitions, located after randomizated blocks method, where the size of one lot was 7 m^2, it was in view the efficiency of biological and chemical treatments in reducing corn borer population, through different methods of assessment:

a) number of holes/plant;
b) number of larvae/plant;
c) number and length of cavities/stem;
d) the yield increased.

The chemical treatments were done using: Regent 200 SC-0.1 l/ha, Bulldock 025 EC-0.3 l/ha, Conquest 25 WG-0.1 l/ha, the biological ones were done using: Thuringin 6000-4.0 kg/ha, Dipel 2x-1.5 kg/ha, Match 050 EC-1.2 l/ha, Insegar 25 WP-0,3 kg/ha and those with entomophagues using: *T.maidis*, *T.evanescens*, *T.dendrolimi*-200,000 individuals/ha. The chemical treatments and the biological ones were applied from the appearance of the first adults of corn borer at the sexual pheromons traps (plant phenophasis of 6-10 leaves) and at the maximum flight of the pest. The notes referring to all the studied aspects mentioned were done at the harvesting.

The utilized calculation methods were: the variant analysis, the simple regressions, the Abbot formula for efficiency, and for calculation of crops losses depending on the frequency and the degree of attack, the number of larvae, were utilized the following formulas: $P\,\% = F\,\% \times Kf\ (0.219)$; $P\,\% = F\,\% \times K\,l\ (0.18) \times$ no. larvae/plant and $P\,\% = [\,F\% \times K\,i_2\ (0.15)] + [\,F\,\% \times K\,i_3 (0.30)] + [\,F\% \times K\,i_4(0.50)] + [\,F\,\% \times K\,i_5\ (0.80)]$, where Kf, Kl and $K\,i_{2\text{-}5}$ are values for each element.

RESULTS AND DISCUSSION

The observation in the mentioned period (1998-2000) demonstrated that the frequency of natural attack of corn borer (*Ostrinia nubilalis* Hbn.) maize hybrids cultivated at A.R.S. Turda and in the influenced area, was about 35.7% and 69.3%, this pest appearing in the first decade of June, mantaining in this culture till August (*table 1* and *figure 1*).

Table 1 - The natural attack of European corn borer (*Ostrinia nubilalis* Hbn.) at maize hybrids cultivated in the counties near A.R.S. Turda (1998-2000)

Crt.nr	Counties	Genotype	Attack frecquency (%)	Nr.larvae/plant
1	Poiana	Turda 200	46,7	1,1
2	T.Nouă	Turda 200	65,0	2,0
3	M.Viteazul	Elan	49,7	1,1
4	Moldovenești	Turda 200	55,7	1,8
5	C.Turzii	Betuflor	43,0	1,0
6	Luna	Elan	53,3	1,7
7	Luncani	Turda 200	56,7	1,8
8	Boldut	Turda 200	49,7	1,2
9	Petrești	Elan	43,0	1,1
10	A.R.S. Turda	Turda super	35,7	0,9
		Sweet maize	58,8	2,0
		Popcorn	60,0	2,1
		>Average	61,6	1,8

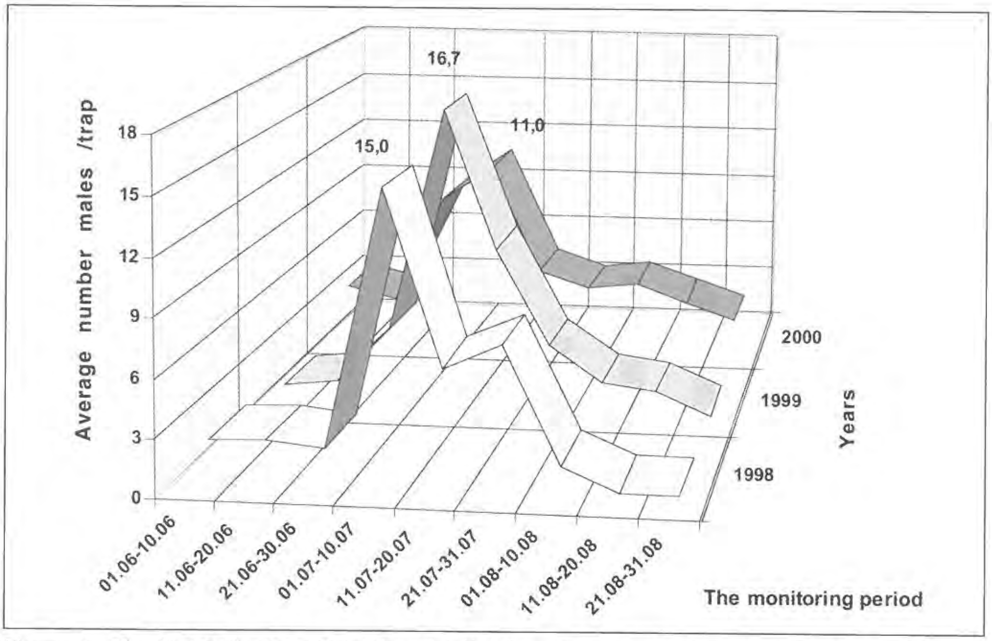

Figure 1 - The flight of adults *Ostrinia nubilalis* Hbn. movement obtained at the traps with synthetic sexual pheromone CIS and TRANS 11 - tetradecenylacetat (Turda, 1998-2000)

Also, these observations showed the existance of a relation between the yield losses and the attack frequency of number of larvae/plant, the strongest is the attack, the greatest are the yield losses (*table 2*).

Table 2 - Comparative estimation of yield losses depending on frequency and degree of *Ostrinia nubilalis* Hbn. attack and mean number of larvae/plant (Turda, 1998-2000)

Genotype	Attack frequency		Attack degree (Rating 1-5)	Larvae/plant		
	Kf = 0,219		Ki = 0,15; 0,30; 0,50; 0,80			Mean
	Freqv. at. (%)	Losses (%)	Losses (%)	Nr. larvae/pl.	Losses (%)	yield losses (%)
Betuflor	69,3	15,2	28,5	2,2	27,4	23,7
Betulisa	69,0	15,1	28,0	2,1	26,1	23,1
Elan	58,5	12,8	22,6	1,8	19,0	18,1
Turda 200	61,4	13,4	25,3	1,8	19,8	19,5
Turda 215	52,0	11,4	19,8	1,6	14,9	15,4
HT 167	63,2	13,8	25,9	2,0	22,7	20,8
Turda 160	50,9	11,1	20,8	1,6	14,6	15,4
Doina	55,0	12,1	21,6	1,7	16,8	16,8
Turda 200 plus	52,1	11,4	21,0	1,6	15,0	15,8
Turda super	44,5	9,7	18,1	1,3	10,4	12,7

The assessment of efficiency of biological and chemical treatments in reducing the corn borer population were done regarding:

a) number of holes/plant;
b) the number of larvae/plant;
c) the number and length of cavities/stem;
d) obtained yield.

At maize hybrids cultivated in the influenced area of A.R.S. Turda the yield losses depending on frequency of ECB attack was between 11.4% – 15.2% (*figure 2*).

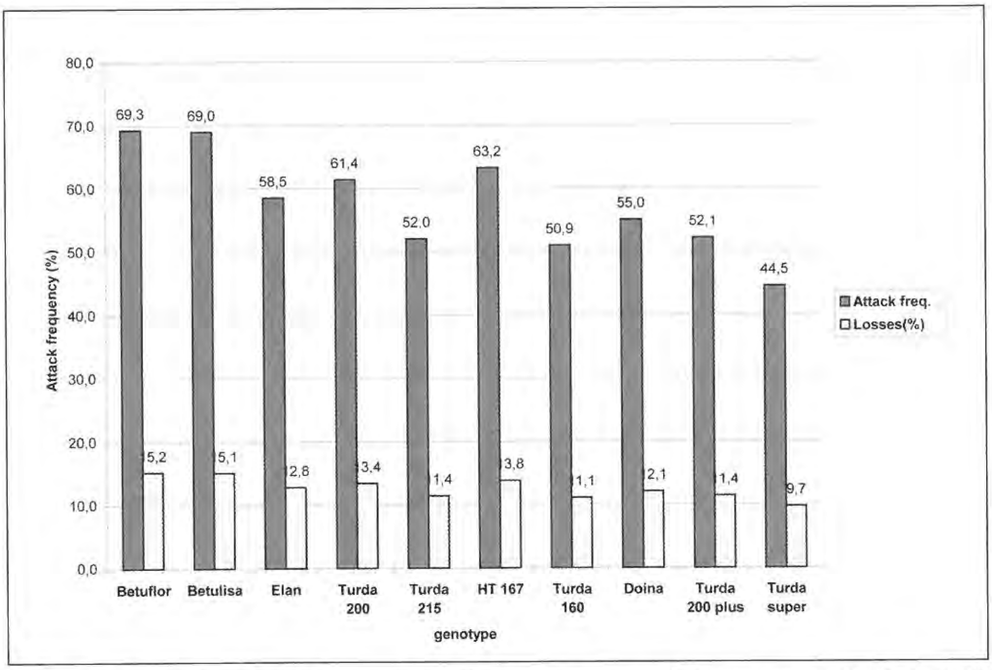

Figure 2 - Estimation of yield losses depending on frequency of *Ostrinia nubilalis* Hbn. attack (Turda, 1998-2000)

From the statistical calculation performed, it resulted that at hybrids with very strong attack, the yield losses were significantly. The value of correlation coefficient between the attack frequency and the yield losses at maize hybrids mentioned is significant (0.987^{***}) (*figure 3).*

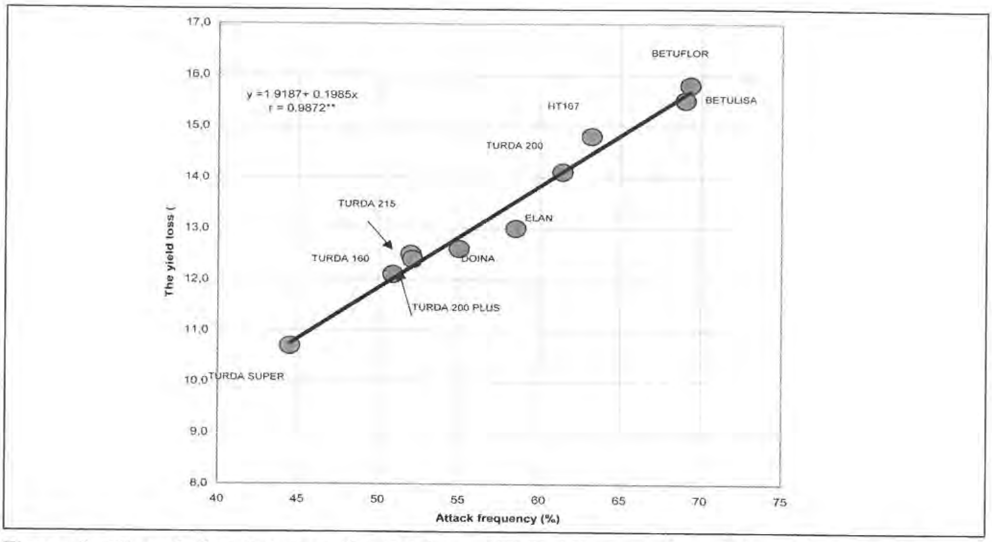

Figure 3 - The relation between the *Ostrinia nubilalis* Hbn. attack and the yield losses at some maize hybrids (Turda 1998-2000)

Also, the relation between the average number larvae / plant and the yield losses is high, the value of correlation coefficient is very significant (0.993^{***}) (*figure 4*).

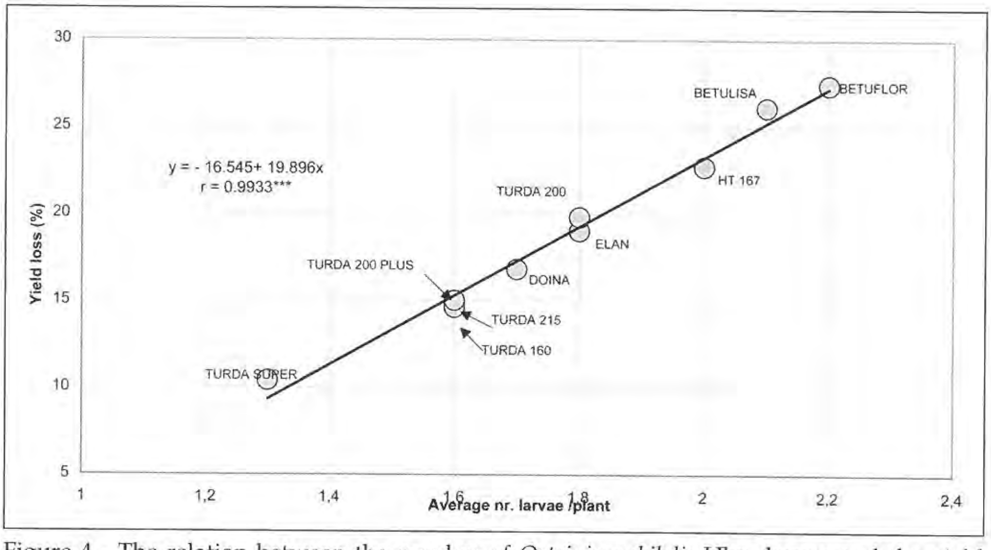

Figure 4 - The relation between the number of *Ostrinia nubilalis* Hbn. larvae and the yield losses at some maize hybrids (Turda 1998-2000)

The chemical and biological treatments assured a very significant reduction of number of cavities/plant, of number holes/plant, of number larvae/plant (*figure* 5) and of length of cavities from the maize plants (*figure* 6) comparatively with untreated variant.

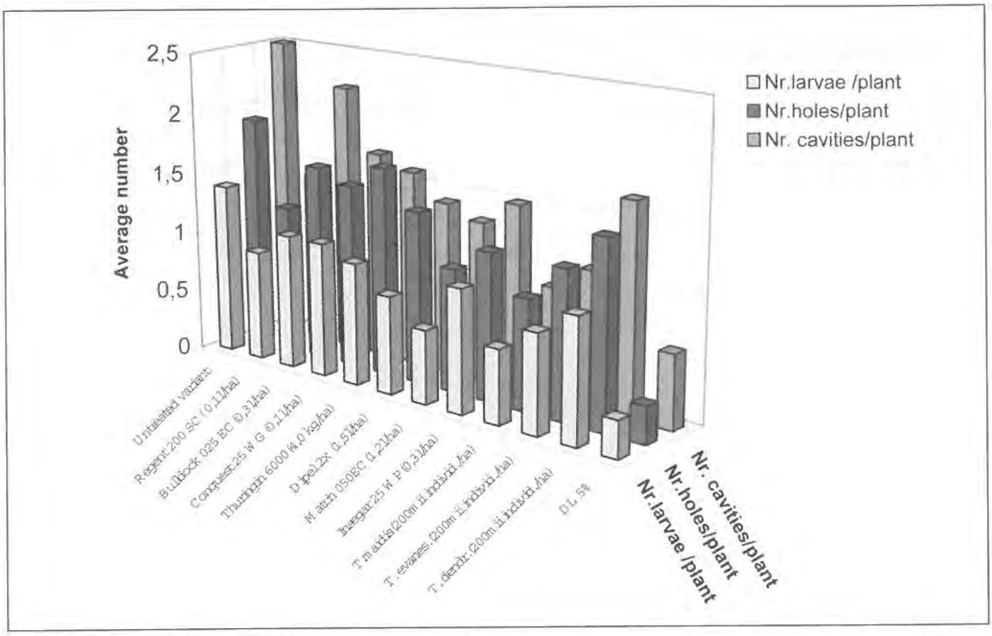

Figure 5 - The effect of chemical and biological treatments on some elements of *Ostrinia nubilalis* Hbn. attack (Turda, 1998-2000)

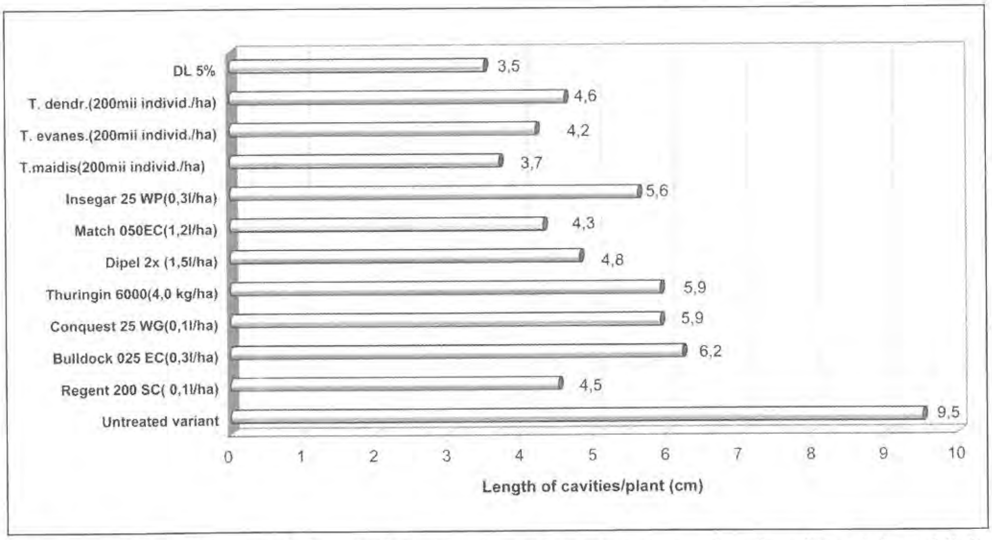

Figure 6 - The influence of chemical and biological treatments on lenght of *Ostrinia nubilalis* Hbn. cavities from the maize plants (Turda, 1998-2000)

The yield obtained was very significant comparatively with untreated variant, at the products: Regent, Match, Dipel, T. maidis, T. evanescens; the efficiency of these products and entomophagues was between 68% – 80% (*figure 7*).
The treatments performed with the 3 species of Trichogramma reduced more strongly the attack of ECB, comparatively with untreated variant, than the products Dipel, Match, Insegar, Thuringin, and than the chemical treatments (Conquest, Bulldock, Regent) (*figure 8*).

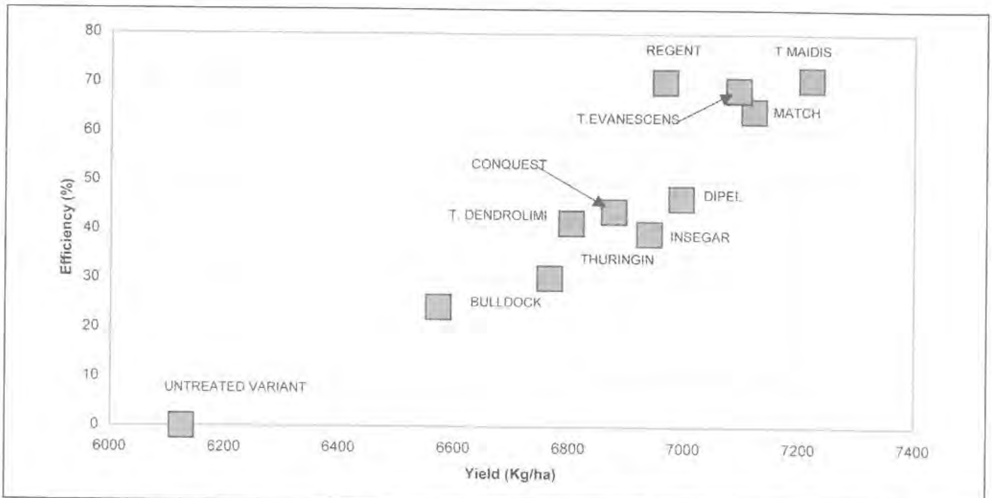

Figure 7 - The yield obtained and the efficiency of treatments in *Ostrinia nubilalis* Hbn. control (Turda 1998-2000)

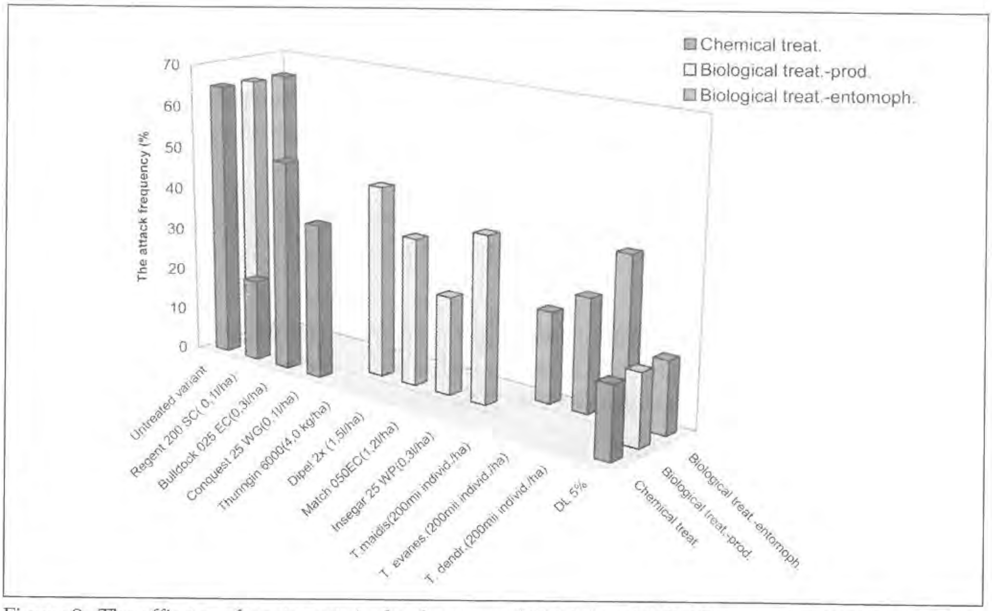

Figure 8 - The effiency of treatments in the decrease of *Ostrinia nubilalis* Hbn. attack (Turda 1998-2000)

Conclusions

1.The frequency and the degree of corn borer attack (*Ostrinia nubilalis* Hbn.) on maize hybrids cultivated at A. R. S. Turda and in the influenced area, between 1998-2000 varied between 35.7-69.3%, recording significant yield losses, which were correlated with the attack frequency and the number of larvae/plant. In this way the yield losses recorded were between 12.7-23.7%.

2.Besides these direct losses which consist in reducing the biological productive potential, it is recorded also indirect losses by breaking the attacked plants, their falling, contributing to multiply the animal pests and making difficult the mechanical harvesting.

3.As assessment methods of chemical and biological treatments in reducing the corn borer population and even the damages, the observation was noted on: the number of holes/plant, the number of larvaes/plant, the number and the length of cavities/stem and the yield obtained.

4.The efficiency of chemical treatments (Regent 200 SC, Bulldock 025 EC, Conquest 25 WG) was between 25-71%, of biological ones using products (Thuringin 6000, Dipel 2x, Insegar 25 WP, Match 050 EC) between 40-65% and with entomophagues (*T.maidis, T.evanescens, T.dendrolimi*) 41-75% regarding the reducing attack of *Ostrinia nubilalis* Hbn.

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CONTROL OF EUROPEAN CORN BORER (*OSTRINIA NUBILALIS* Hübner) IN SEEDCORN PRODUCTION

Anka Ostojčić,
AK «Jasinje», RJ «Kopanica», Kopanica, Croatia

Marija Ivezić, Emilija Raspudić, Mirjana Brmež,
Univ. of J.J. Strossmayer, Faculty of Agriculture in Osijek,
Trg sv. Trojstva 3, 31000 Osijek, Croatia

Abstract

European corn borer *Ostrinia nubilalis* Hübner, (ECB) is a perennial insect pest of corn in Croatia, with infestation levels reaching 50% in many fields the past 10 years. This insect is especially important in seed corn production because of this crop's higher market value. The objective of this study was to determine the efficacy of chemical and biological controls against ECB in seed production fields. Eight treatments were applied to two corn inbred (Bc 492, Bc 592) at two location (Kopanica, Berava) in Croatia in 1994 and 1995. Treatments were 3 systemic organophosphates (Lebaycid, Rogor, and Ekatin 25 EC), 2 pyrethroids (Sherpa and Karate), two formulations of *Bacillus thuringiensis* Berlinier (Biobit XL; WP and FC), and 1 untreated control. Insecticide treatments were applied when at least 2/3 of the plants contained egg masses and at least 1/3 of the plants showed symptoms of injury (shot-holing) in the whorl. Efficacy of the insecticide treatments was evaluated just before harvest by dissecting 50 plants/treatment and counting the number of tunnels, number of live larvae, measuring tunnel length, and grain yield. In 1994, 87% of the plants in the untreated control plots were infested with ECB. The organophosphate treatments reduced the infestation level by 42%, the pyrethroids by 40%, and the Bt treatments by 42%, compared to untreated control. In 1995, 70% of the plants in the untreated control plots were infested with ECB. The organophosphate treatments reduced the infestation level by 32%, the pyrethroids by 30%, and the Bt treatments by 29%, compared to untreated control. In both years, the organophosphate treatments provided the greatest yield protection from ECB. Yields in these treatments were 13-19 % greater than the untreated control. Results from this study show that yields of seedcorn could be significantly increased if ECB population were managed in Croatia.

Key words: *Ostrinia nubilalis* Hübner, seedcorn, insecticides.

INTRODUCTION

The corn production has been organized on ca. 400,000 ha in Croatia, mostly in eastern parts of the country. Hybrids from different FAO groups were grown (100-700) as well as corn for seedcorn production. European corn borer (*Ostrinia nubilalis* Hübner) (ECB) is a perennial pest of corn, but controlling is not implemented in Croatia (Ivezić & Raspudić, 1997, 2001). One of the reasons is the lack of proper mechanization for insecticides' application. The seed corn production has higher market value and any kind of interruptions during the vegetation period can result with lower yield. So, the aim of this research was to determine the efficacy of chemical and biological controls against ECB in seed production fields.

Material and methods

The efficacy of different insecticides in controlling of ECB in seedcorn production was examined in East Croatia, near Osijek (45°32''N, 18°44''E), in AK «Jasinje» - two localities: Kopanica and Berava. Investigation was perfomed in 1994 and 1995, and eight treatments were applied on two corn inbred: Bc 492 and Bc 592.

Previous crop was wheat in both years of survey. Sowing of corn was done when the temperature of the soil reached above 10°C. Before the sow, seed was treated with Semevin 375 KS in the dose 1.3 l/100 kg of seed, and Primextra SC (6l/ha) were incorporated in the soil. Herbicides Parandel EC and Lontrel 300 were applied for atrazin resistant weeds.

Moth fly was detached by light trap (100W), which was placed 4m above ground level. The material was collected every morning from May 15^{th} to 30^{th} of September. The time of appearance of first moths was detected, as well as the number of males and females.

Treatments were three systemic organophosphates (Lebaycid - *fention*, Rogor - *dimetoat*, and Ekatin 25 EC - *timeton*), two pyrethroids (Sherpa - *cipermetrin* and Karate – *lambda cihalotrin*), two formulations of *Bacillus thuringiensis* Berlinier (Biobit XL; WP and FC), and one untreated control. Insecticide treatments were applied when at least 2/3 of the plants contained egg masses and at least 1/3 of the plants showed symptoms of injury (shot-holing) in the whorl. Efficacy of the insecticide treatments was evaluated just before harvest by dissecting 50 plants/treatment and counting the number of tunnels, number of larvae, measuring tunnel length, and grain yield.

Results

Intensity of attack (%) of ECB was determined on examined inbred and treatments and results are shown in *figure 1*.

Average attack intensity in 1994 was 87% in untreated plots, while organophosphate treatments reduced the infestation level by 42%, the pyrethroids by 40%, and the Bt treatments by 42%. In 1995 average attack intensity was 70% in untreated plots, and the organophosphate treatments reduced the infestation level by 32%, the pyrethroids by 30%, and the Bt treatments by 29% compared to untreated control.

Number of tunnels and larvae per plant were determined by dissection of the corn stalk (*figures 2, 3*).

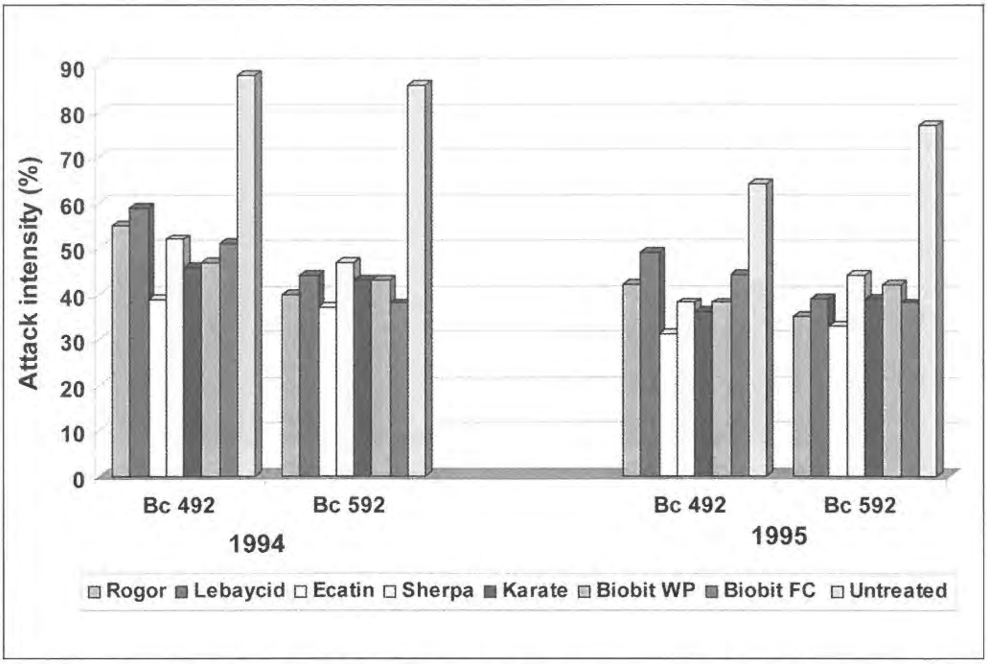

Figure 1 - Intensity of attack of ECB in 1994 and 1995 (average for both localities Kopanica and Berava)

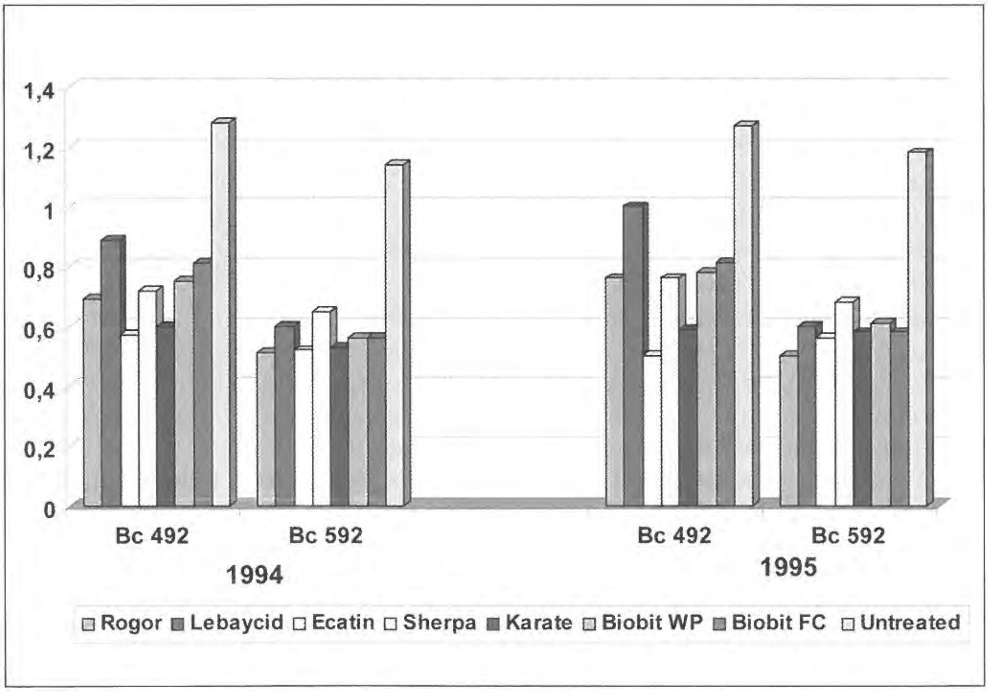

Figure 2 - Number of tunnels per plant in 1994 and 1995 (average for both localities)

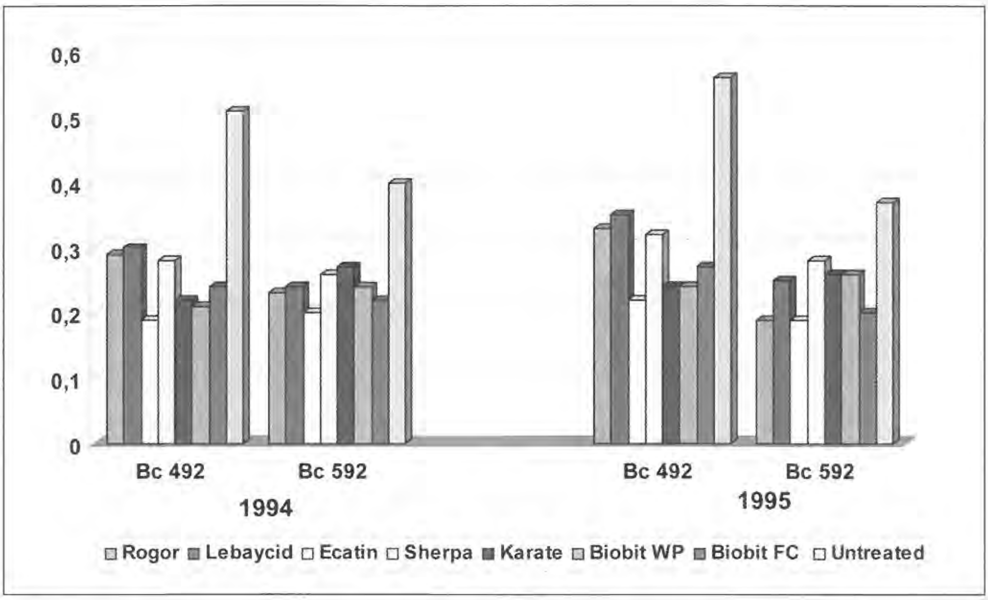

Figure 3 - Number of larvae per plant in 1994 and 1995 (average for both localities)

In both years of investigation it was determined 1.22 tunnels per plant in untreated control, while in treatments the average was 0.66 tunnels per plant. The highest number of tunnels occurred in Bt treatments (0.69), organophosphate treatments had 0.64, and the pyrethroids had 0.65 tunnels per plant.

Average number of larvae in both years of investigation was 0.47 larvae per plant in untreated plot, 0.26 in organophosphate treatments, 0.28 in treatment with phyrethroids, and 0.25 in Bt treatments. The majority of the tunnels and larvae were found under the ear, and the least were found in the ear.

In seedcorn production, the loss in grain yield is very important. In untreated plot the average yield loss on both inbred was 987 kg/ha, while in organophosphate treatments it was 310 kg/ha, in phyrethroids 308 kg/ha, and in Bt treatments 306 kg/ha (*figure 4*). Biological preparations on the base of the *Bacillus thuringiensis* Berliner showed the same efficacy as chemical preparations, and because of their ecological value, they can be recommended (Ivezić *et al.*, 1998, Raspudić *et al.*, 1999).

Ecological factors had the great impact on the grain yield in both years of investigations. In 1994, two very dry periods occurred during the vegetation (in May and July), and in 1995 dryness occurred just in July.

Grain yield showed very significant differences (Lsd 0.01) between untreated plot and treatments in both years of examinations.

Conclusions

The reduction of infestation level with ECB was noticed in all treatments in comparison with untreated plot. The organophosphate treatments reduced the infestation level by 37%, the pyrethroids by 35%, and the Bt treatments by 36%, in comparison with untreated plot. Number of tunnels and larvae were lower on treatments in comparison to untreated plots also. In both years, the organophosphate treatments provided the highest yield protection from ECB. Yields in these treatments were 13-19% higher than the untreated plot.

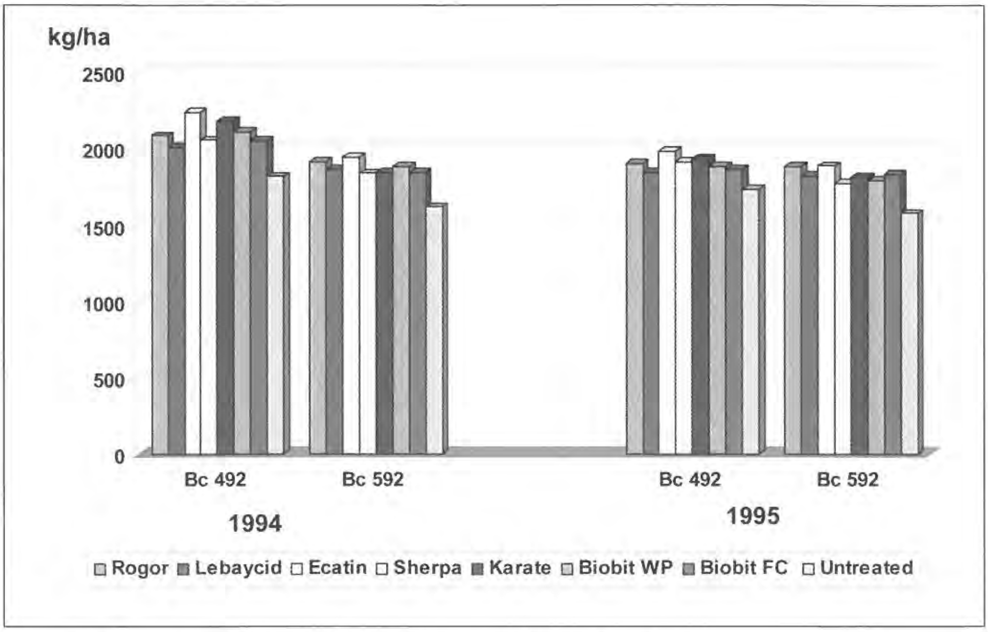

Figure 4 - The grain yield in both years (average for both localities)

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FIELD TRAPPING OF *SESAMIA NONAGRIOIDES* (LEFÈBVRE) (LEPIDOPTERA: NOCTUIDAE) USING MULTICOMPONENT BLENDS OF SEX ATTRACTANTS

Giacinto S. Germinara, Giuseppe Rotundo, Antonio De Cristofaro,
Department of Animal,
Plant and Environmental Sciences,
University of Molise, Via De Sanctis,
I-86100 Campobasso, Italy

Abstract

The capture power of oil traps baited with different sex attractant blends and dispenser types to *Sesamia nonagrioides* (Lefèbvre) males was evaluated in corn fields comparing catches with those obtained by virgin females and light traps.

Captures by synthetic sex attractant mixtures were significantly lower than those obtained using virgin females and similar to that of the light trap.

Preliminary studies carried out in laboratory conditions showed that 12:Ac, even if present in a low ratio in the bait, is released by rubber septa in a higher quantity than other compounds for at least three weeks.

Field test results demonstrate that in Southern Italy maize crops the monitoring of *S. nonagrioides* flight activity can be achieved using oil traps and rubber septa baited with quantities of synthetic sex pheromone components [Z11-16:Ac (75 μg), Z11-16:OH (25 μg), Z11-16:Ald (25 μg), 12:Ac (25 μg)] lower than those actually included in the commercial formulation. Further laboratory and field investigations will be necessary to set up an adequate formulation with an attractant power similar to that of virgin females.

Key words: *Sesamia nonagrioides*, sex pheromone, dispensers, release, field tests.

INTRODUCTION

Sesamia nonagriodes (Lefèbvre) (Lepidoptera: Noctuidae) is an important pest of maize crops in several Mediterranean countries.

Sex pheromone can provide a useful tool for monitoring *S. nonagrioides* populations and for developing biological and/or integrated pest management strategies. Previous studies showed that the sex pheromone of *S. nonagrioides* is a multicomponent blend containing Z11-hexadecenyl acetate (Z11-16:Ac), as the main component, Z11-hexadecenol (Z11-16:OH), Z11-hexadecenal (Z11-16:Ald), dodecanyl acetate (12:Ac), hexadecanyl acetate (16:Ac). The presence of minor components varies according to different populations (Rotundo *et al.*, 1985; Sreng *et al.*, 1985; Mazomenos, 1989; Frerot *et al.*, 1997; Sans *et al.*, 1997; Rotundo *et al.*, 2001); in the

Italian population, a higher ratio of Z11-16:Ald was detected in the effluvia from individual virgin females (Germinara *et al.*, 1998).

On the whole, a pheromone blend evokes an optimal response in the target specimen only when the different components are released in precise quantities and ratios. For this reason, in the case of multicomponent blends, it is not easy to set up a dispenser that assures a natural-like release.

Field trials in Italy demonstrated that self-made oil traps are more efficient than other models (Mastrap, ISAGRO, Italy; Unitrap, Biological Control System Ltd., UK) for monitoring *S. nonagrioides* males (Rotundo *et al.*, 1993).

The response of *S. nonagrioides* males to oil traps baited with different blends of sex pheromone components and dispensers, including the commercially available one, were evaluated in corn fields comparing captures with those obtained by virgin females and light traps.

Materials and methods

Insects – *S. nonagrioides* larvae were collected from maize stalks grown in Battipaglia (Campania Region, Central Italy) during August and September and reared on a meridic diet at 25±2°C, 65±2 r.h. and L16:D8 photoperiod (Germinara *et al.*, 2000).

Pupae were sexed and placed in cylindrical plastic containers (6 cm x 8 cm) until emergence. Two virgin females kept in a cylindrical metallic net container (4 cm x 6 cm) were used to bait oil traps. They were renewed twice a week.

Traps - Oil traps were prepared from plastic waste-paper baskets (lower diameter 20 cm; upper diameter 30 cm; 31 cm high) whose walls were partially cut to produce four windows (16 cm wide; 26 cm high) and a tray on the bottom (5 cm high). The tray was filled with water and oil was added until an oil layer of about 0.5 cm was formed. A plastic pot stand was glued to the upper border (diameter 30 cm) as a lid. An iron wire was suspended in the centre of the lid to situate the bait 5 cm above the oil surface.

Light traps (150 W) were similar to that described in Parenzan and De Marzo (1981).

Dispensers - Rubber septa dispensers were used to test different synthetic blends (Novapher, Novara, Italy). Rubber septa were also loaded with single chemicals, cut longitudinally into pieces (from 1/2 to 1/32) and different portions of them stapled together were used as lure.

Two types of double-matrix dispensers were prepared in order to reduce the release rate of 12:Ac. The first type (rubber-rubber) is described in Bratti *et al.* (1987). The second (polyethylene-rubber) consists of a high density polyethylene vial (0.35 ml) containing a cylindrical (22 mm x 3 mm) piece of expanded black rubber. It was prepared by filling the vial with Z11-16:Ac (900 µg), Z11-16:OH (100 µg) and Z11-16:Ald (100 µg) and impregnating the rubber with 12:Ac (100 µg) in hexane.

All dispensers and blends were compared with the commercial formulation [a rubber septum loaded with Z11-16:Ac (100 µg), Z11-16:OH (100 µg), Z11-16:Ald (100 µg), 12:Ac (100 µg)] (Novapher, Novara, Italy). Dispensers were aged for at least 6 days before laboratory or field use (McDonough and Butler, 1983).

Field - Tests were carried out, over a four year period (from 1998 to 2001), in maize fields (hybrid LG 11) in Battipaglia (Campania Region, Southern Italy) from the end of August to the end of September. Oil traps were installed at the height of 1 m on metallic poles placed 30 m apart along the border of the field. Light traps were fixed to the soil 150 m apart. Each treatment was repeated three times.

Traps were checked once a week and insect species determined by examining the male genitalia. Captures were expressed as males/trap/week and data analysed statistically (ANOVA) using the Duncan's multiple range test.

Collection of sex pheromone components from rubber dispensers - Rubber septa dispensers baited with Z11-16:Ac (450 µg), Z11-16:OH (50 µg), Z11-16:Ald (50 µg) and 12:Ac (50 µg) were exposed to an air flow of 0.7 m/s at room temperature (23±2°C).

Quantities of various synthetic sex pheromone components released from rubber septa after different times of aeration (1 h, 1 day, 1, 2, 3 and 4 weeks) were determined collecting them on the surface of glass capillary tubes.

The dispenser was suspended in a 6 ml conical glass container, using a spiral steel holder, in which dried (silica gel) and filtered (charcoal) synthetic air flowed (90 ml/min) for 30 min at 28°C.

Airborne volatiles were adsorbed onto 2 glass capillaries (200 mm long; i.d. 1 mm) kept at about 2°C and mounted on the outlet of a glass container (i.d. 1 mm). Each glass capillary and the glass container end were rinsed with twice distilled hexane (2 µl x 2) containing 100 ng of Z11-heptadecenyl acetate (Z11-17:Ac) as an internal standard. The remaining rinse volume (1,5-2 µl) was directly injected into GC for analysis.

GC analysis - Measurements were taken using a Fisons 9000 series chromatograph equipped with a splitless injection system. The column was a SPB-5 (30 m x 0.32 mm i.d., 0.25 µm film thickness, Supelco Inc., Bellefonte, USA). Conditions were: carrier gas, helium at 20 psi; temperature programme, 70°C for 2 min, up to 200°C at 10°C/min, up to 240°C at 5°C/min, 240°C for 5 min, up to 260°C at 10°C/min, at 260°C for 10 min; injector and detector temperature, 250°C. The quantity of each compound was calculated based upon the peak area, and calibrated by comparing it with that of Z11-17:Ac. Three replicates of dispenser were tested.

RESULTS

1998 - The blend containing Z11-16:Ac, Z11-16:OH, Z11-16:Ald and 12:Ac in the ratios of 69/8/15/8 (1300 µg and 650 µg) allowed to catch mean numbers of *S. nonagrioides* males (13.87 and 10.60 trap/week) which were not significantly different (P=0.05) from those (11.00 and 10.87/trap/week) obtained by the 76/8/8/8 blend (1200 µg and 600 µg) (*table 1*).

1999 - Male captures corresponding to decreasing doses (from 1200 µg to 150 µg) of synthetic blend containing the four compounds in the ratio of 76/8/8/8 were not significantly different (P=0.05) among them (*table 1*).

Table 1 - Male captures of *S. nonagrioides* during 1998 (25. VIII - 29 IX) and 1999 (23 VIII - 27 IX) using oil traps baited with different sex attractant blends adsorbed into rubber septa or virgin females and light traps

Year	Attractants (μg)				Males/trap/week (Mean ± S.D.)
	Z11-16:Ac	Z11-16:OH	Z11-16:Ald	12:Ac	
1998	900	100	100	100	11.00 ± 4.13 **b**
	450	50	50	50	10.87 ± 3.38 **b**
	900	100	200	100	13.87 ± 3.23 **b**
	450	50	100	50	10.60 ± 5.15 **b**
	Unbaited control				0.67 ± 0.68 **a**
	Virgin females (n. 2)				43.00 ± 12.18 **c**
	Light				12.18 ± 9.95 **b**
1999	900	100	100	100	24.66 ± 9.11 **b**
	450	50	50	50	27.71 ± 11.22 **b**
	225	25	25	25	26.04 ± 9.56 **b**
	112.5	12.5	12.5	12.5	25.00 ± 4.34 **b**
	Unbaited control				0.33 ± 0.58 **a**
	Virgin females (n. 2)				74.25 ± 20.81 **c**
	Light				25.07 ± 9.35 **b**

2000 - Oil traps baited with 1 + 1/16 + 1/8 + 1/16 or 1 + 1/32 + 1/16 + 1/32 portions of rubber septa loaded with 1 mg of Z11-16:Ac, Z11-16:OH, Z11-16:Ald and 12:Ac respectively caught a number of males significantly (P=0.05) greater than other combinations but not significantly different from those obtained using the commercial formulation (*table 2*).

Table 2 - Male captures of *S. nonagrioides* (25. VIII - 29 IX.2000) using oil traps baited with combinations of rubber septa portions containing single sex pheromone components (1 mg) or virgin females and light traps

Portion of rubber septa containing single sex pheromone components				Males/trap/week (Mean ± S.D.)
Z11-16:Ac	Z11-16:OH	Z11-16:Ald	12:Ac	
1	1/4	1/2	1/4	2.75 ± 0.50 **b**
1	1/8	1/4	1/8	4.25 ± 1.26 **b**
1	1/16	1/8	1/16	12.38 ± 3.88 **c**
1	1/32	1/16	1/32	13.71 ± 2.05 **c**
Commercial formulation				12.13 ± 3.09 **c**
Unbaited control				0.93 ± 0.60 **a**
Virgin females (n. 2)				36.67 ± 9.26 **d**
Light				12.83 ± 1.73 **c**

The release rate of pheromone components by rubber septa were studied in laboratory conditions (*figure 1*). The amounts of 12:Ac, collected in 30 min, ranged from 60.3 ng after 1 h to 1.8 ng after four weeks. Quantities of Z11-16:Ac ranged from 8.6 ng to 3.6 ng, those of Z11-16:Ald from 1.2 ng to 0.3 ng and those of Z11-16:OH from 0.8 ng to 0.4 ng. Z11-16:Ac was released in quantities higher than other compounds during the fourth week (*figure 1*).

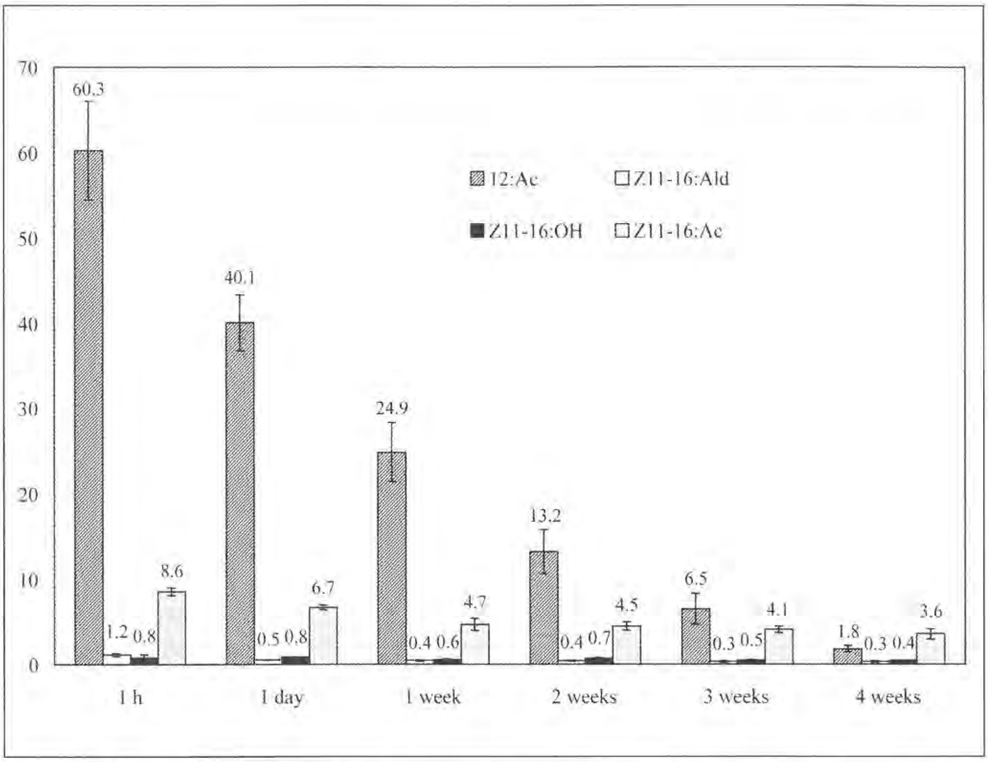

Figure 1 - Mean quantities (ng) of synthetic sex attractants collected from a rubber septum (n=3) loaded with Z11-16:Ac (450 µg), Z11-16:OH (50 µg), Z11-16:Ald (50 µg) and 12:Ac (50 µg), after different periods of aeration (0.7 m/s) at 23±2°C. Vertical lines indicate standard deviation

2001 - Double substrate dispensers allowed, on the average, male captures not significantly different (P=0.05) from those by rubber septa (*table 3*).

During the four years the attractant power of oil traps baited with different synthetic pheromone blends was always significantly lower (P=0.05) and about three-four times less, than that of the oil traps baited with virgin females and similar to that of the light trap (*tables 1, 2, 3*).

Traps baited with synthetic attractants caught a significant number of other moth species, particularly *Trachea atriplicis* L., *Mythimna unipuncta* (Haworth) and *Discestra trifolii* Rottenberg.

Table 3 - Male captures of S. *nonagrioides* (24. VIII - 28.IX.2001) using oil traps baited with the commercial attractant blend absorbed into different dispensers or virgin females and light traps

Attractants (μg)				Dispensers	Males/trap/week (Mean ± S.D.)
Z11-16:Ac	Z11-16:OH	Z11-16:Ald	12:Ac		
900	100	100	100	rubber septa	46.83 ± 7.49 **b**
900	100	100	100	rubber-rubber	32.17 ± 4.86 **b**
900	100	100	100	rubber-polyethylene	35.83 ± 10.02 **b**
Unbaited control					0.67 ± 0.68 **a**
Virgin females (n. 2)					150.50 ± 16.97 **c**
Light					44.50 ± 11.63 **b**

Conclusions

The different field tests did not allow a significantly higher capture power than that obtained by rubber septa containing Z11-16:Ac (900 μg), Z11-16:OH (100 μg), Z11-16:Ald (100 μg), and 12:Ac (100 μg) (commercial formulation). The attractant power of the oil trap did not increase when (i) the quantity of Z11-16:Ald was doubled in respect to the commercial blend, (ii) the bait dose ranged from 1200 μg to 150 μg and (iii) portions of rubber septa containing individual sex pheromone components were used.

The blend released by rubber septa was different from the one introduced according to the hypothetical composition of *S. nonagrioides* virgin female pheromone. During the first three weeks, 12:Ac was released in the highest quantity even if present only at an 8% ratio; this finding could explain the different attractant power among synthetic pheromone blends and virgin females. However, double substrate dispensers prepared in order to reduce the release of 12:Ac did not increase male captures.

The lower attractiveness of a synthetic pheromone blend (Sans *et al.*, 1997) in comparison to that of virgin females was also reported by Ameline and Frérot (2001).

Results confirmed the complexity of setting up a multicomponent pheromone dispenser and they lead to the undertaking of more detailed laboratory and field investigations regarding the release of *S. nonagrioides* sex pheromone from various other substrates.

Our field studies show that in Southern Italy maize crops the monitoring of *S. nonagrioides* flight activity can be achieved using oil traps and rubber dispensers baited with quantities of synthetic sex pheromone components [Z11-16:Ac (75 μg), Z11-16:OH (25 μg), Z11-16:Ald (25 μg), 12:Ac (25 μg)] lower than those actually included in the commercial formulation.

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Authors have equally contributed to the present paper.

SEX PHEROMONE OF THE ITALIAN POPULATION OF *SESAMIA CRETICA* (LEDERER) (LEPIDOPTERA: NOCTUIDAE)

Giuseppe Rotundo, Giacinto S. Germinara, Antonio De Cristofaro,
Department of Animal, Plant and Environmental Sciences,
University of Molise, Via De Sanctis,
I-86100 Campobasso, Italy

Paola Riolo,
Department of Agricultural and Environmental Biotechnology,
University of Ancona, Via Brecce Bianche,
Monte Dago, I-60100 Ancona, Italy

Abstract

Two active compounds were detected from *Sesamia cretica* (Lederer) male moth antennae analysing by gas chromatography (GC) linked to electroantennographic detection (EAD) ovipositor washings, the compounds present on the surface of the pheromone gland and effluvia by virgin females.

The compounds were identified as cis-9-tetradecenol (Z9-14:OH) and cis-9-tetracedecenyl acetate (Z9-14:Ac) by comparing their retention times, mass spectra and EAG activities with synthetic standards.

Preliminary field tests showed that the major component, Z9-14:OH, is essential for attraction. Oil traps baited with rubber septa dispensers containing 1 mg of Z9-14:OH caught the highest number of males. The addition of Z9-14:Ac (from 75% to 25%) to the corresponding alcohol caused significant reductions in trap catches.

Key words: *Sesamia cretica*, sex pheromone, GC-EAD, GC-MS, EAG, field tests.

INTRODUCTION

The dura stem borer *Sesamia cretica* (Lederer) (Lepidoptera:Noctuidae) is a polyphagous and serious pest of maize crops in several Mediterranean and Northern Africa areas. Even if the presence of this pest was recorded in different Italian region, its real geographical distribution is not clear.

A field study carried out in Sudan to evaluate the attractant power of synthetic compounds for cotton pests showed that traps baited with a 3:1 blend of cis-9-tetradecenol (Z9-14:OH) and cis-9-tetracedecenyl acetate (Z9-14:Ac) allowed for the capturing of a high number of *S. cretica* males while traps baited with Z9-14:OH or Z9-14:Ac captured fewer males (Arsura *et al.*, 1977). The 3:1 blend of sex attractants used to monitor the Italian population of *S. cretica* captured a low number of males (Riolo *et al.*, 2001).

The aim of the present study was to identify the compounds produced by virgin females of *S. cretica* able to elicit an antennal response from males and to evaluate in field the attractant power of the synthetic compounds for the male moths.

MATERIALS AND METHODS

Insects - Diapausing larvae of *S. cretica* were collected from maize stems near Ancona (Marche Region, Central Italy) during March 2001. Pupae were sexed and individually kept in plastic containers (30 cm x 20 cm x 8 cm) at 25±2°C, 65±5% r.h. and L16:D8 photoperiod until emergence. Adults were separated daily and fed with a 10% sucrose solution.

Pheromone collection - From 30 virgin females, during the first scotophase and after 15 min of calling behaviour, the abdominal tips were excised and extracted 2 at time with hexane (15 µl per tip) for 5 min at room temperature. The extract obtained was then concentrated to a final volume of 30 µl (1 female equivalent per µl = 1 FE/µl) using a slow flow of nitrogen and stored at -20°C until used. Additional extracts were prepared from individual abdominal tips and added of 10 ng of Z3-16:Ac for quantitative GC analysis.

Solid phase micro-extraction (SPME) was performed by using a fused silica fibre covered with polydimethylsiloxane (PDMS; external thickness of 100 µm) placed into a SPME holder (Supelco Inc. Bellefonte, USA). The fibre was gently rubbed (5 min) on the pheromone gland of a calling female extruded by a slight pressure on the abdomen. The retained substances from the fibre were desorbed inside the injector of a gas chromatograph at 250°C (4 min). Prior to each extraction, the fibre was exposed for 30 min at 250°C in an additional split/splitless inlet.

Airborne volatiles from individual virgin females were collected in cold glass capillary tubes. A calling female was introduced in an adequately shaped pipette tip that could allow the external extrusion of the last urites and then suspended by a polyurethane foam (PUF) support (cleaned with hexane for 12 h in a Soxhlet extraction apparatus) onto a 6 ml conical glass container in which dried (silica gel) and purified (active carbon) synthetic air flowed (70 cc/min) for 15 min at 28°C.

Effluvia were adsorbed onto two 200 mm-long glass capillaries (i.d. 1 mm) kept at about 2°C and mounted at the outlet of the glass container (i.d. 1 mm). Capillaries and the glass container were connected with small pieces of Teflon tube with the glass surfaces in contact.

Each glass capillary was rinsed with redistilled hexane (2 µl x 2) containing 10 ng of Z3-hexadecenyl acetate (Z3-16:Ac) added as an internal standard. The remaining rinse volume (1,5 –2 µl) was directly injected into GC for the different analyses. Prior to the extraction, the glass container was rinsed with hexane and dried at 250°C for 12 h; Teflon tube, pipette tip and PUF support were replaced.

Coupled Gas Chromatography-Electroantennographic Detection (GC-EAD) - Hexane and SPME extracts and airborne volatiles collected from single virgin females were analysed by GC-EAD. Measurements were carried out using a Fisons 9000 series chromatograph equipped with a splitless injection system. The column was a SPB-

5 (30 m x 0.32 mm i.d., 0.25 µm film thickness, Supelco Inc., Bellefonte, USA). Conditions were: carrier gas, helium at 20 psi; make up, helium at 15 ml/min; temperature programme, 60°C for 2 min, up to 280°C at 10°C/min, 280°C for 15 min; injector and detector temperature, 250°C. The effluent from the column was split between a flame ionisation detector (FID) and the EAG detector in a ratio of 1:1. The EAG equipment was furnished by Syntech Laboratories, Hilversum, The Netherlands.

Gas chromatography-mass spectrometry (GC-MS) – Analyses were performed with a Fisons 8000 series gas chromatograph linked to a MD 800 (Fisons) quadrupole mass detector. Injections were done in splitless mode. Capillary column, carrier gas, temperature programme and injector conditions were the same used in GC analysis. Mass spectra were obtained at 70 eV with the ion source at 200°C.

The GC-EAD active compounds were tentatively identified by comparison with spectra from authentic samples.

Gas chromatography (GC) - A Fisons 8000 Top series fitted with a FID and a splitless injector was employed to quantify compounds present in the different extracts. The column and GC conditions were the same used in GC-EAD. The quantities of compounds were determined based on the peak areas and calibrated by comparison with that of 10 ng of Z3-16:Ac for the solvent and effluvia extracts. For SPME extracts peak areas were compared with those of 10 ng of standard compounds (Z9-14:OH or Z9-14:Ac).

Electroantennography (EAG) - The EAG technique was similar to that used in previous studies (Rotundo *et al.*, 1984, Den Otter *et al.*, 1996, De Cristofaro *et al.*, 2000).

Antenna from 1-day-old males was excised at the base and few distal segments were cut off.

Monounsaturated (cis and trans) 14-carbon acetates and alcohols hexane solutions (100 ng/µl) were used as stimuli. Dose response curves were calculated applying Z9-14:OH and Z9-14:Ac hexane solutions (10 µl) from 10 pg/µl to 10 µg/µl. Z11-16:OH (10 µl of a 100 ng/µl solution) was used as reference stimulus to correct for changes in EAG response. Synthetic compounds were supplied by IPLO-DLO, Wageningen, The Netherlands.

EAG responses recorded from 10 different antennae were normalised according to Van Der Pers (1981).

Field tests - Field tests were carried out in maize fields grown near Ancona during 2001 from the 17th of July to the 29th of August.

The treatments established were: oil traps baited with rubber septa dispensers (Novapher, Novara, Italy) containing 1 mg of synthetic Z9-14:OH or Z9-14:Ac or their mixtures (3:1, 1:1, 1:3), and unbaited oil trap. Treatments were replicated three times.

Traps installed at the height of 1 m on metallic poles and placed 40 m apart along the border of the field, were checked once a week. Insect species were determined by examining the male genitalia. Captures were expressed as males/trap/week and data analysed statistically (ANOVA) using the Duncan's multiple range test.

RESULTS

GC-EAD analyses of the hexane and SPME extracts and effluvia from living females detected the presence of two EAG active peaks at the retention times of 17.54 min (A) and 19.05 min (B) respectively (*figure 1*).

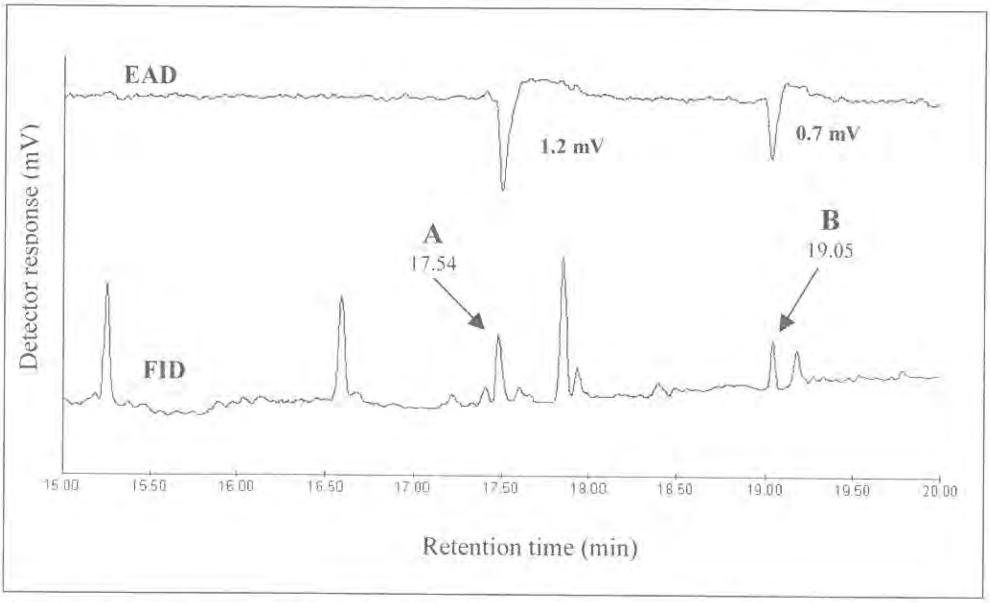

Figure 1 - Simultaneously recorded gaschromatogram (FID) and electroantennographic detector (EAD) responses of S. cretica males to a pheromone gland extract (1 FE)

In the capillary column SPB-5, peak A appeared at the same retention time of Z9-14:OH and peak B to that of Z9-14:Ac.

Mass spectrum of compound A showed diagnostic peaks at m/z 212 (M+), 194 (M-H_2O), 41 ($C_3H_5^+$) and 31 (CH_2OH^+). The spectrum of compound B presented characteristic ions at m/z 254 (M^+), 194 (M^+- CH_3COOH), 61 ($CH_3COOH_2^+$), 43 (CH_3CO^+), 41 ($C_3H_5^+$). This information suggested that compound A and B are monounsaturated 14-carbon alcohol and acetate respectively.

Comparative GC-MS revealed that mass fragment patterns of compound A and B were identical to that ones of synthetic Z9-14:OH and Z9-14:Ac.

EAG responses of *S. cretica* males to Z isomers of 14-carbon acetates and alcohols were greater than those to E isomers (*figures 2* and *3*). In addition, EAG response to Z9-14:OH was significantly higher than that to other Z isomers of monounsaturated 14-carbon alcohols (*figure 2*).

Similarly, EAG response to Z9-14:Ac was higher than the responses elicited by other Z acetate isomers (*figure 3*). Dose dependent responses were evoked by the two compounds. Z9-14:OH elicited EAG responses higher than those to Z9-14:Ac at the different doses, and showed a lower threshold dose (10^{-6} mg) when compared to the corresponding acetate (10^{-4} mg).

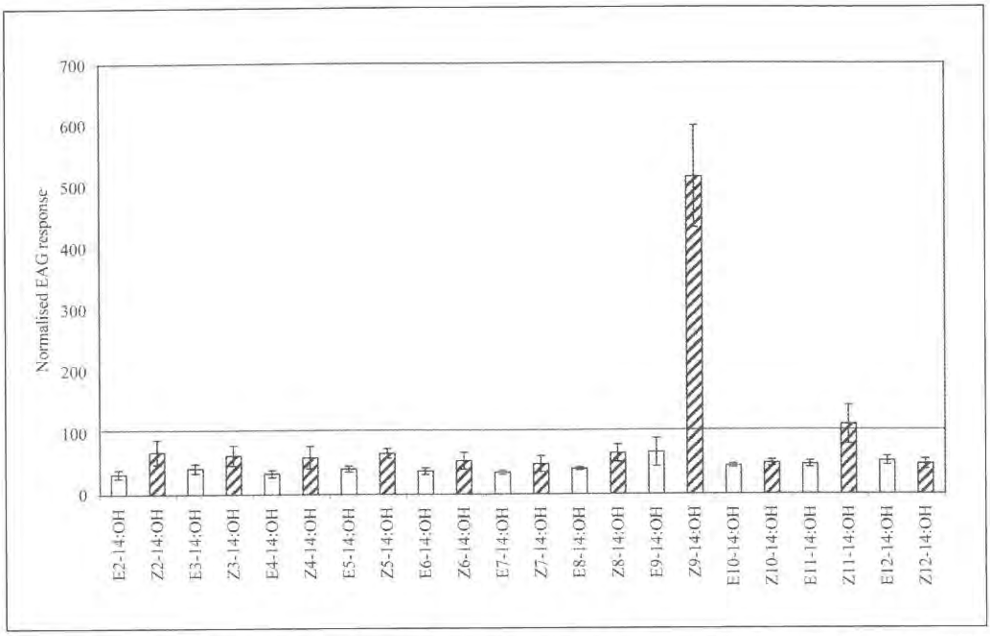

Figure 2 - Normalised EAG response of S. cretica males to Z and E isomers of monounsaturated 14- carbon alcohols. Reference stimulus: Z11- 16: OH (1 µg). Vertical lines indicate standard deviation

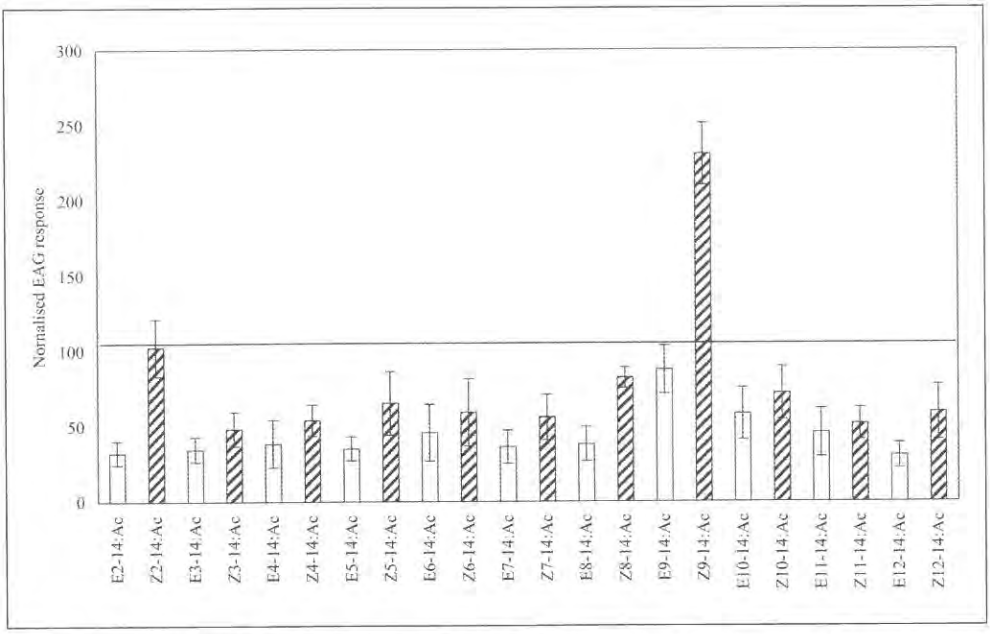

Figure 3 - Normalised EAG response of S.cretica males to Z and E isomers of monounsaturated 14-carbon acetates. Reference stimulus:Z11-16:OH (1 µg).Vertical lines indicate standard deviation

In an abdominal tip, on the average, 5.06±1.81 ng of Z9-14:OH and 4.75±3.60 ng of Z9-14:Ac were found by hexane extraction and 3.19±0.79 ng/tip of Z9-14:OH and 1.51 ±0.18 ng/tip of Z9-14:Ac using SPME. In analysing effluvia from individual virgin females (n=10) it was calculated that a female of *S. cretica* releases, on the average, 0.81±0.51 ng/min of Z9-14:OH and 0.40±0.38 ng/min of Z9-14:Ac. Substances were collected only in the first capillary.

During field tests, 112 males of *S. cretica* were captured. Oil traps (n=3) baited with 1000 µg of Z9-14:OH caught a number of males significantly (P=0.05) higher than other treatments. The 1:3 blend of Z9-14:Ac and Z9-14:OH allowed to catch a higher number of males than the other mixtures. Male captures by Z9-14:Ac alone or mixed with Z9-14:OH in the ratios of 3:1 or 1:1 were not significantly different (P=0.05) than those obtained by unbaited oil traps (*table 1*).

Attractants (µg)		**Male captures**
Z9-14:Ac	**Z9-14:OH**	
1000	0	8 **a**
750	250	6 **a**
500	500	14 **ab**
250	750	19 **b**
0	1000	58 **c**
Unbaited (control)		7 **a**

Values followed by the same letter are not significantly different for P=0.05 (Duncan's multiple range test)

Table 1 - Male captures of S. cretica by oil traps (n=3) baited with different sex attractants

Conclusions

GC-EAD analysis of different extracts showed that virgin females of *S. cretica* produce two compounds that elicit antennal responses from conspecific males. These compounds were identified as Z9-14:OH and Z9-14:Ac by GC and GC-MS analyses.

EAG studies support the results of chemical analyses. In fact, the male EAG responses to Z9-14:OH and Z9-14:Ac were significantly higher than that to Z and E isomers of the corresponding monounsaturated 14-carbon alcohols and acetates.

Z9-14:Ac and Z9-14:OH were also identified from the female sex pheromone gland of many other species of Lepidoptera (Arn *et al.*, 2000). In the genus *Sesamia* they are secondary components of the sex pheromone blend of *S. calamistis* (Hampson) (Zagatti, 1988).

Our preliminary field studies showed that Z9-14:OH is essential for the attraction of *S. cretica* male moths. A synergistic effect on male attraction by Z9-14:Ac was not apparent.

Recently an antagonistic effect of Z9-14:Ac on the attraction of the Mediterranean corn borer, *S. nonagrioides* (Lefèbvre) to its sex pheromone was observed in field trapping studies (Germinara 1998; Rotundo *et al.*, in press). The results of this study suggest that Z9-14:Ac, acting as inhibitor to the congener *S. nonagrioides*, probably play a role in the reproductive isolation of the two species. A similar antagonistic effect of Z9-14:Ac has been found in *Ostrinia nubilalis* (Hübner) (Glover *et al.*, 1989) living on the same host plant.

On the contrary to what was observed in the Sudan population (Arsura *et al.*, 1977) the 3:1 blend of Z9-14:OH and Z9-14:Ac was less attractive than Z9-14:OH to males of the Italian population of *S. cretica*.

Further studies will be necessary to determine the quantities of sex attractants to load into an adequate dispenser able to assure a natural-like sex pheromone release.

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Authors have equally contributed to the present paper.

OSTRINIA NUBILALIS POPULATION LEVELS IN NORTHEASTERN ITALY: LONG-TERM DATA AND PRACTICAL CONSIDERATIONS

Lorenzo Furlan, Vincenzo Girolami,
Department of Agronomy, Entomology,
University of Padova, Agripolis,
via Romea 16, Legnaro PD

Pavan Andrea, Bianchi Michele,
Freelances

Abstract

Population levels of and the pressure on maize crops by *Ostrinia nubilalis* Hb. (ECB) were estimated in Northeastern Italy between 1982 and 2000. At harvest, the number of larvae and tunnels, the total stalk tunneling per plant, were recorded. The ECB flight period was studied using light traps and Hartstack traps baited with Isagro E/Z caps placed in large maize field and compared with the presence of egg clusters estimated by observing 1000 plants twice per week. In addition in 2000, the effect of insecticide treatments on ECB populations was evaluated.

First generation damage was unusually low, but larval injury was found on most of the plants sampled after the second generation. Low parasitisation rates by *Trichogramma* spp. and *Lydella thompsoni* were observed. The peak of egg presence usually occurred in the second ten days of August, about 15 days after the peak flight of ECB moths. Only light traps recorded significant moth captures, while sex pheromone traps did not provide reliable data. Treatments applied every 8 – 10 days only slightly improved the level of control compared with one treatment applied at the most appropriate time. Light traps and observation of the eggs only gave reliable information on ECB population development.

Key words: *Ostrinia nubilalis*, Northeastern Italy, population level, seasonal flight, egg presence, parasitism rate, damage.

Introduction

In Northeastern Italy, *Ostrinia nubilalis* Hb. (ECB) is usually present at high levels and regarded as a serious problem by most farmers. Consequently, ECB population levels and behaviour have been studied for many years. A summary of data collected between 1982 and 2000 is given.

MATERIALS AND METHODS

Observations were carried out in at least three fields (Eraclea and San Donà di Piave, Veneto region) from mid June to mid September in 1982, 1983, 1991, 1999, and 2000. The fields were planted with maize hybrids considered medium sensitive to ECB attack (Padano, Asgrow in 1982, 1983; Juanita (Pioneer) and Paolo (Dekalb) in 1991; Laramis, Androdek and Altdek (Dekalb) in 1999; H785 (HGD) in 2000). First generation population levels were estimated by inspecting at random at least 5000 plants per each field and by counting all those showing signs of larval feeding. At harvest, the number of larvae and tunnels per plant, the length of stalk tunneling were recorded by inspecting at random at least 100 plants per each field.

Egg parasitisation rate was evaluated by counting the number of black egg clusters from which *Thrichogramma* spp. adults had emerged. The larval parasitisation rate was evaluated by counting the number of *Lydella thompsoni* pupae present in maize stems at harvesting.

The ECB flight period was studied using a Blacklight trap equipped with a 15-watt bulb and 2 Hartstack traps baited with Isagro E/Z caps. Traps were inspected at least twice per week. The numbers of moths caught were compared with the presence of egg clusters estimated by observing 1000 plants chosen at random twice per week. In addition in 2000, the effect of insecticide treatments on ECB populations was evaluated.

Three treatments were compared: insecticide (Decis, 1 kg/ha, 200 liters of water per hectare) treatment every 8 – 10 days, insecticide treatment at the peak of egg-hatch; and untreated. Treatments were applied using a high clearance sprayer. Four replications were included; the individual plots were maize field strips 20 m wide and 400 m long. Data were analyzed by ANOVA. The data on number of larvae and tunnels were transformed to $(x+0.5)^{1/2}$. Treatment means were separated by Duncan's New Multiple Range Test.

RESULTS

Population levels and parasitisation rate over the years

First generation damage was usually low, but larval injury was found on most of the plants sampled following the second generation. The number of larvae per plant ranged from 0.6 (2000) to about 2 in most of the other years. The number of tunnels per plant ranged from 0.6 (2000) with a total stalk tunneling of 0.12 cm/plant to 10.3 (1999) with a total stalk tunneling of 37 cm per plant. An intermediate total stalk tunneling length value (6 cm/plant) was recorded in 1983. The highest egg parasitisation rate (by *Trichogramma* spp.) was observed in 1982 (33%), while in other years percentage parasitism ranged between 20 and 24%. At the site with the highest parasitism rate, one plant in five contained *Lydella thompsoni* pupae.

Table 1 - ECB infestation level in Veneto region from 1982 to 2000. At least 1000 plants from at least 3 fields were sampled in each year

	plants damaged by ECB 1st generation larvae (%)	plants damaged by ECB 2nd generation larvae (%)	number of ECB tunnels/plant	number of ECB larvae/plant	egg clusters paratisized by Thrichogramma sp %	*Lydella thompsoni pupae/plant*
1982	70	95	2,1	1.6	33	0,05
1983	62	100	1,8	2.1	24	0,02
1991	60	91	2,2	1,9	20	0,1
1999	55	100	10,3	1,7	20	0,2
2000	25	60	0,6	0,6	23	0

Biology and swarming period

The peak of egg presence usually occurred in the second ten days of August, about 15 days after the peak of moth flight. Data are in agreement with those collected in other localities of Northeastern Italy in the same or previous years (Barbattini *et al.*, 1986; 1988; Burgio and Maini, 1994; Maini and Burgio, 1991; Zangheri, 1969). The peak occurred several days after the period when most of treatments had been applied based on normal farm practices (e.g. *figure 1*). Only light traps recorded significant moth captures, while the sex pheromone traps did not provide reliable data (no more than 10 moths captured per season). Light traps and observations of eggs only gave reliable information on ECB population development.

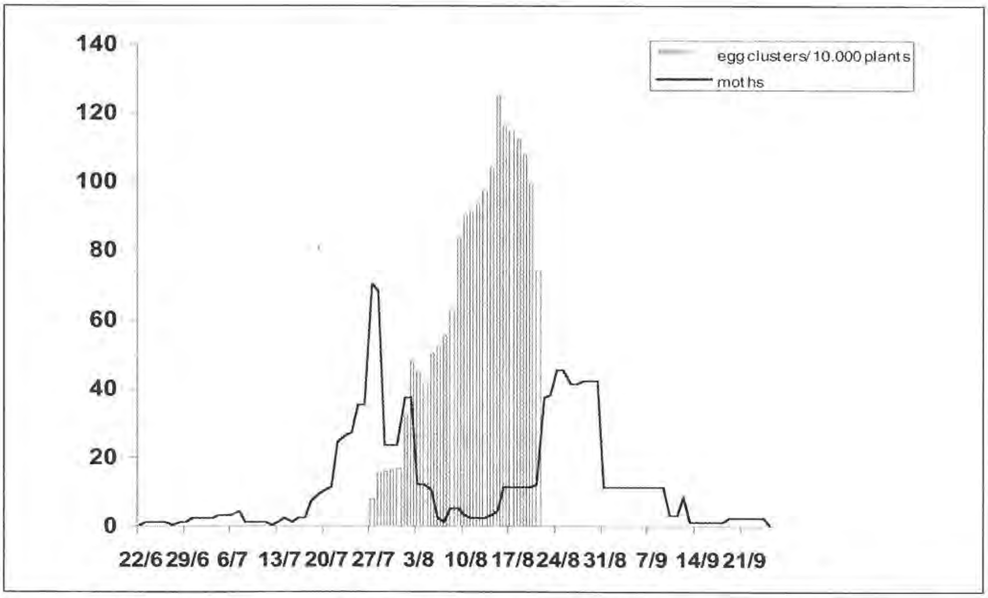

Figure 1 - Swarming pattern of ECB moths captured by light trap in comparison with egg presence in 2000 in Eraclea

Population containment

Treatments applied every 10 days only slightly improved the level of control compared with one treatment applied at the most appropriate time. In 2000 where ECB pressure was low on maize, treatments did not improve the yield.

Table 2 - Effect of different control strategies on ECB population and maize yield. Eraclea, 2000. Means followed by the same letter in a vertical row are not different at P=0.05

Treatments	Tunnels/90 plants	Damaged ears/90 plants	Larvae/40 plants	Dry matter tons/ha
Test	23.2a	46.5a	22.7a	22.6a
One at egg peak	14.5b	42.0a	19.5a	18.6a
Six (every 10 days)	12.2b	22.7b	11.7a	19.2a

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A NEW PRODUCT CRUISER 350 FS FOR MAIZE SEED TREATMENT AGAINST *TANYMECUS DILATICOLLIS* GYLL.

A. Bărbulescu, I. Voinescu, D. O. Sadagorschi,
A. Penescu, C. Popov, S. Vasilescu,
Research Institute for Cereals and Industrial Crops,
8264 Fundulea-Călărasi, Romania
e-mail : fundulea@ns.ricic.ro; Phone/fax: 401-311.07.22

Abstract

Maize leaf weevil *Tanymecus dilaticollis* is the most dangerous soil pest of maize, the first ranged spring field crop in Romania. This pest is able to induce high yield losses and even compromise crop. The seed dressing with carbofuran products provide satisfactory protection of this crop against the pest. This method replaced the use of organochlorine insecticides applied, for many years, as dusts, which contributed to environmental pollution and the destruction of useful fauna. However, the very high toxicity of carbofuran to man and animals, as well as a tendency for *T. dilaticollis* to develop tolerance to this compound, necessitated the development of seed treatments using less toxic products, such as Cruiser 350 FS.

The experiments were done in laboratory, under artificial infestation, and in field, in plots cropped successively over the last three years with maize, thus favouring pest reproduction. The efficacy of the Cruiser 350 FS, a thiamethoxam - based product, was compared with a carbofuran - based product, as a standard. Evaluation of chemicals was done in relation to values of attack intensity, and the percentage of plants escaping pest attack for field trials only. The behaviour of maize hybrids to Cruiser 350 FS was examined in several localities under field conditions. The percentage of emerged plants was assessed in treated and untreated plots.

Trials performed revealed very good results obtained by the use of Cruiser 350 FS, providing satisfactory protection of maize crop against *T. dilaticollis*. As differences between the rates are not high, that means from economic standpoint the dose of 9 l/t seed is most interesting. Being a product with much lower toxicity than older alternatives it is possible for unauthorized persons to apply this seed treatment, which is not permitted when using carbofuran. Almost all maize hybrids trialled showed good tolerance to the Cruiser 350 FS, even at the high rate of 20 l/t seed.

INTRODUCTION

Maize leaf weevil (*Tanymecus dilaticollis*) is the most dangerous soil pest in the early vegetation phases of maize, the first ranged spring field crop in Romania (Paulian *et al.*, 1969; Paulian, 1973). The pest has a restricted distribution, and is mainly limited to Romania and neighbouring countries to the east, south and west. It mainly occurs in the south, south - east and east of the country, while in the north and centre of the country it has no economic significance.

Though it is considered to be polyphagous, *T. dilaticollis* exhibits preference for maize, a crop which provides optimal development for the larvae and is the most preferred food by adults (Bărbulescu & Voinescu, 1998). Due to this fact, the traditional practice of cropping maize after maize for several consecutive years greatly contributes to the reproduction of this insect and thus to an increase in its population. In a not too distant past, populations of this insect sometimes attained densities above 60 adults/m^2.

As a result of attack caused by adults, even before emergence of plants above soil surface, high harvest losses can frequently be recorded, sometimes compromising not only maize crops, but also sunflower and sugarbeet. Paulian (1972) showed a yield loss of 34% maize grains where pest density ranged between 25 - 30 individuals/m^2.

Out of more than 3 millions ha cropped with maize, about one third of area requires chemical treatment against the maize leaf weevil. The control of this soil pest was based, in a long period of time, nearly exclusively on organochlorine insecticides, applied as dusts (Paulian, 1972). Unilateral, irrational and excessive use of these treatments resulted in environmental pollution and the destruction of beneficial fauna. *T. dilaticollis* also became tolerant to some chemicals. These reasons determined the replacement of organochlorine insecticides, as they provided only a very low extent of outbreak limitation, at a high cost.

The very good results obtained in controlling this pest by seed treatment with products based on carbofuran determined the promotion and generalisation of this method, which is modern, efficient, economic and less polluting. As a result, insecticides applied as powders were given up towards the end of the 1980s (Bărbulescu *et al.*, 1988, 1989).

The high toxicity of carbofuran to man and animals, as well as possible onset of tolerance of maize leaf weevil to this product, imposed the improvement of seed treatment methods using less toxic products, however able to protect maize crops against soil pests.

MATERIALS AND METHODS

The experiments were carried out both in the laboratory and the field, in three localities. The Cruiser 350 FS was tested at several dosages/t seed, using a carbofuran-based product, registered in this country for treatment of maize seeds against this pest, as a standard.

In the laboratory, under controlled environment conditions, the efficacy of chemical treatment of maize seed was established at a density of seven adults per plant, except for the first trial in 1999, where four adults per plant were used. Infestation with adults previously collected in the open, was performed at the beginning of plant emergence above the soil surface. Attack rating was done four to six days after infestation. Each plant was assessed on a scale of 1-9, where 1 represented an unattacked plant and 9 a completely destroyed plant.

In the field, the experiments were carried out in plots cropped successively over the last three years with maize, thus favouring pest reproduction. To avoid migration of *T. dilaticollis* adults from one plot to another, the experimental plots were laterally isolated with a 2 m wide strip sown with pea, a plant repellent to this insect (Paulian, 1972; Paulian & Popov, 1977). Evaluation of chemicals was done having in view values of attack intensity, as assessed at the end of the maximum attack period by rating plants on the scale of 1-9, and the percentage of plants escaping pest attack.

The behaviour of maize hybrids to the Cruiser 350 FS was examined in several localities under field conditions using the rate of 10 l/t seed and 20 l/t seed. Sowing was as early as possible in spring. The percentage of emerged plants was assessed in treated and untreated plots.

Results and discussions

In order to fully understand the role of chemical treatment of seeds in protecting plants from attack by maize leaf weevil, it is considered as welcomed to outline some features specific to this species. Economic significance is dependent on attack by overwintering adults, after their appearance at the soil surface. Larvae do not induce obvious damage.

The fact that this insect overwinters in the adult stage in the soil, at 40 - 60 cm depth or more, while its occurrence in the upper soil layers takes place by the second half of March or at the begging of April. Thus during plant emergence, even in the case of a very early sowing, the whole adult population is present at soil surface, and ready to start attacking young plants even before their appearance at the soil surface. If this period of plant emergence corresponds to warm weather, with temperatures above 20°C and without rainfall, the danger of attack from this pest is very high.

It results in the need for high solubility of the insecticide used for seed dressing, a rapid translocation of the active ingredient within the germinating seed and in the young plants, so that the lethal dose for the pest occurs at the time of attack onset. From this point of view, it is worth noting that Cruiser 350 FS applied to the seed is rapidly taken up by roots of germinating seedlings respectively, and is translocated to the cotyledons and leaves (Senn *et al.*, 1998).

When analysing data regarding the efficacy of the chemical treatment of maize seed (*figure 1*) against attack by maize leaf weevil under laboratory conditions, it is noted that for all experimental doses of Cruiser 350 FS good results have been obtained, similar or better, compared to the standard. Certain differences of values referring to the attack intensity are to be remarked, as depending on the product dose. The lowest attack values were recorded with the highest active ingredient amount per tone of seed. When assessed in terms of adult density per plant, the values of attack intensity were, generally, similar to all trials performed at seven adults/plant, but these values were lower in the case of trials done using four adults/plant.

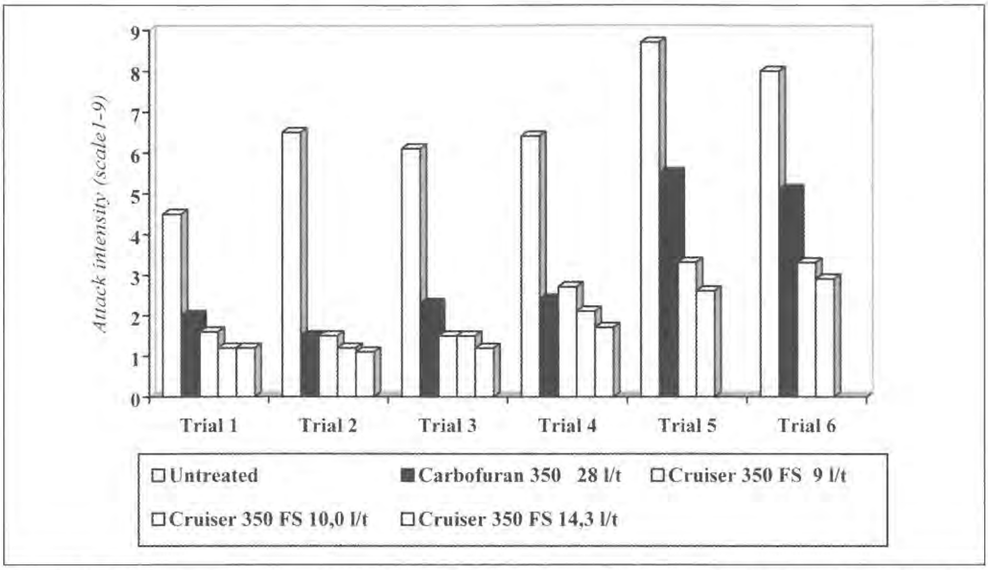

Figure 1 - Efficacy of Cruiser 350 FS against *Tanymecus dilaticollis* in maize under laboratory conditions (trials 1-4 in 1999, trials 5-6 in 2000)

Referring to the control experiments with maize leaf weevil under field conditions, it is noted that, due to different climate conditions, the intensity of attack varied from one locality to another, and from year to year. The heaviest outbreaks occurred at Valu Traian in 1999. Therefore, the attack values recorded differed to a certain extent depending on the pest infestation level. In general, attack values for Cruiser 350 FS, in terms of both attack intensity and percentage of plants escaping damage were much reduced compared to those in the untreated check, and similar or even less compared to those of the standard product.

When considering the attack values recorded for different experimental rates of Cruiser 350 FS, it was noted that a dose of 9 l/t seed provided a lower efficacy under a heavy outbreak. Good results were obtained for 10 and 14.3 l/t seed, respectively (*table 1*).

Table 1 - Efficacy of Cruiser 350 FS against *Tanymecus dilaticollis* in maize, under field conditions*

Treatment	Dose l/t seed	Attack intensity (1 - 9)			Saved plants(%)		
		1998	1999	2000	1998	1999	2000
Untreated	-	6.1	6.5	5.4	64	59	73
Carbofuran 350 (std)	20	2.9	4.2	3.2	96	80	98
Cruiser 350 FS	9	3.3	3.8	2.7	91	87	99
Cruiser 350 FS	10	3.0	3.2	2.4	94	94	100
Cruiser 350 FS	14.3	2.8	2.9	-	96	95	-

* means of 3 localities

Particular attention has also been paid to data referring to the tolerance of various maize hybrids usually used in this country (*table 2*). From this point of view, almost all hybrids tested showed good tolerance to Cruiser 350 FS at a rate of 10 and 20 l/t seed. Nevertheless, in the northern area of the country (Suceava), the germination of early maize hybrids was affected.

Table 2 - Behaviour of some maize hybrids to the seed treatment with Cruiser 350 FS

VARIANT	Treatment	Fundulea		Secuieni		Valu Traian	
		Emerged pl. %	% from check	Emerged pl. %	% from check	Emerged pl. %	% from check
NEPTUN	1	79	100	72	100	79	100
	2	92	116	59	82	78	99
	3	88	111	56	78	76	96
LSD = 5 %		25,02		13,62		11,62	
OVIDIU	1	83	100	82	100	58	100
	2	89	107	85	104	68	117
	3	89	107	84	102	65	112
		8,06		10,84		9,73	
PALTIN	1	84	100	56	100	70	100
	2	84	100	55	98	87	124
	3	89	106	51	91	86	123
LSD = 5 %		18,07		15,01		5,19	
RAPID	1	85	100	72	100	84	100
	2	96	113	77	107	70	83
	3	89	105	78	108	61	73
LSD = 5 %		12,78		27,80		6,89	
SOIM	1	86	100	62	100	74	100
	2	85	99	66	106	81	109
	3	82	95	66	106	76	103
LSD = 5 %		11,12		4,72		16,95	
OCTAVIAN	1	87	100	61	100	79	100
	2	93	107	69	113	80	101
	3	89	102	69	113	77	97
LSD = 5 %		19,46		27,52		18,07	
RAPSODIA	1	83	100	84	100	70	100
	2	93	112	84	100	73	104
	3	88	106	79	94	71	101
LSD = 5 %		19,74		18,84		15,56	
FUNDULEA 376	1	70	100	73	100	72	100
	2	86	123	76	104	67	93
	3	86	123	76	104	72	100
LSD = 5 %		6,67		5,56		9,45	
VULTUR	1	93	100	65	100	80	100
	2	93	100	64	98	75	94
	3	86	92	67	103	71	89
LSD = 5 %		15,84		2,24		25,29	
CAMPION	1	86	100	81	100	66	100
	2	81	94	86	106	80	121
	3	82	95	73	90	71	107
LSD = 5 %		21,96		15,29		3,50	
RIVAL	1	89	100	82	100	72	100
	2	98	110	81	99	86	119
	3	91	102	76	93	84	117
LSD = 5 %		16,40		13,90		11,39	
GRANIT	1	85	100	66	100	82	100
	2	89	105	76	115	84	102
	3	88	103	67	101	82	100
LSD = 5 %		24,74		21,40		13,90	
ROBUST	1	86	100	80	100	77	100
	2	90	105	74	92	78	101
	3	82	95	70	87	74	96
LSD = 5 %		19,46		24,18		17,79	

T1 - TMTD (std.)

T2 - Cruiser 350 FS 10 l/t

T3 - Cruiser 350 FS 20 l/t

In order to have a general idea on the importance of the seed treatment as a best way to assure a satisfactory protection of the maize crop against *Tanymecum dilaticollis*, the evolution of chemical control methods of this pest in maize crop (*figure* 2), and the reduction in the total quantity of chemical products by seed treatment of maize crop for controlling the pest (*table* 3) are presented. It can be noted that seed treatment represents the most efficient, economic and less polluting method for protection of maize crop against *Tanymecus dilaticollis* attack.

TREATED SURFACE (Ha)	800,000-1,200,000		15	20,000	100,000-300,000	400,000-750,000	
Stage	Generalization	Testing of efficacy	Verification of efficacy	Introduction in production	Extension	Generalization	Testing of efficacy
Year	1960 -1985	1977-1978	1979	1980	1981	1990 - 2000	1998 -2000
Product	Organochlorine insecticides	Carbofuran products					Thiamethoxam product
Method	Soil treatment (powders)	Seed treatment					

Figure 2 - Evolution of chemical control methods of *Tanymecus dilaticollis* in maize crop. Yearly cultivated surface: 3.0 – 3.3 mio ha

Table 3 - Reduction in the total quantity of chemical products by seed treatment of maize crop for controlling *Tanymecus dilaticollis* (calculated for 800,000 ha)

Method	Product	Commercial product		Active ingredient	
		Tons	%	Tons	%
Soil treatment	Organochlorine insectices	24,000	100	1.920	100
Seed treatment	Carbofuran products	560	2.3	196	10.2
	Thiamethoxam product	200	0.83	70	3.64

CONCLUSIONS

Tanymecus dilaticollis is the most dangerous soil pest of maize crop in Romania. Chemical seed treatment provides satisfactory protection of maize crop against this pest. This method determined replacement of organochlorine insecticides applied as dusts, which contributed to environmental pollution and the destruction of beneficial fauna. The Cruiser 350 FS ensures suitable protection of maize crop, which is generally similar or better to that given by the standard product, carbofuran. The Cruiser 350 FS being a product with much lower toxicity than older alternatives, this affords the possibility of applying this seed treatment by the unauthorized persons, which is not allowed when using carbofuran. Almost all maize hybrids trialled showed good tolerance to the Cruiser 350 FS at the experimental rates used. The seed treatment represents the best way for protection of maize crop against *Tanymecus dilaticollis* attack.

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CONTROL OF WIREWORMS IN SOME FIELD CROPS BY SEED TREATMENT IN ROMANIA

Constantin Popov, Alexandru Bărbulescu, Constantin Roibu, Alexandru Al. Alexandri, Vera Preoteasa,
Research Institute for Cereals and Industrial Crops,
8264 Fundulea-Calarasi, Romania
e-mail : fundulea@ns.ricic.ro; Phone/fax: 401-311.07.22

Abstract

In Romania, the main species of *Elateridae* are: *Agriotes ustulatus* (40.1%), *A. obscurus* (17.3%), *A. sputator* (11.3%), *A. flavicornis* (10.9%), *A. pilosus* (6.9%), *A. lineatus* (3.8%), *Limonius pilosus* (1.8%), *Synaptus filiformis* (1.6%), *Selatosomus latus* (1.5%). Annually, the wireworms attack 800,000 ha aproximately. The recorded densities in field crops vary from 5-10 larvae/m^2 up to 70-100 larvae/m^2 which determine attack levels up to 25-30% in wheat and barley crops and up to 40-65% in maize and sunflower crops. At present, the seed chemical treatment method is the most important, efficacy, economic and little polutant measure for the control of wireworms in field crops. At the winter spiked cereals, the seed treatment was made with products based on *lindan* and one or more fungicides (*carbendazim, carboxine, diniconazol, miclobuthanil, prochloraz, tebuconazol, tiophanat methyl, thiram, triadimenol*). In this case, a good simultaneous protection to both wireworms and some specific pathogens (*Tilletia spp.* and *Fusarium spp. in* wheat crops or *Ustilago nuda* and *Pyrenophora graminea* in barley crops) was assured.

At the spring row-crops (maize and sunflower), different products based on the folowing active ingredients: *acetamiprid, bifentrin, carbofuran, fipronyl, furatiocarb, imidachlopride, thiamethoxam,* have been used. The efficacy, up to 80% permitted their registration and promoting in production in Romania.

Key words: wireworms, field crops, insecticides, insectofungicides, seed treatment.

INTRODUCTION

Wireworms (*Elateridae, Coleoptera*) present a high harmful effect for many field crops. In Romania, the wireworms are spread in all the agricultural areas, especially in hilly regions where their densities are high (Bărbulescu and Popov, 1995, 1999; Paulian *et al.*, 1974; Perju *et al.*, 1971; Popov, 1996; Radu and Grecea, 1965). The heavy and excessively moist soils (podzols, river meadows, Danube meadow), as well as strong fodder grasslands, encourage this pest (Bărbulescu and Popov, 2000; Perju and Mare, 1984; Manole *et al.*, 1998, 1999; Popov *et al.*, 1996a, 1998; Trotus *et al.*, 1994a).

Because of its specific mode of life, in which the harmful stage, larva respectively, live and eat exclussively into soil, on germinated grains or on seedlings, the possibilities of the attack prevention are limited enough.

The wireworms, especially in spring crops, are able to induce significant yield losses, even compromise crops (Manole *et al.*, 1993; Mărgărit *et al.*, 1990).

The crop protection both maize and sunflower, with reduced number of plants/m^2 and spiked cereals, represents an important technological priority (Bărbulescu and Popov, 1999, 2000; Popov *et al.*, 1996b; Rotaru, 2000; Trotus *et al.*, 1994b).

At present, the best results for the control of these pests, are obtained by seed treatment before sowing, with an insecticid or insectofungicid, differentiated as active ingredient or rate, from a crop to another.

The seed chemical treatment is the most important, efficacy, economic and little polutant measure for field crops protection against both seed or soil born phatogens as will as soil pests including wireworms.

The paper presents the obtained data regarding the control of wireworms in the wheat, barley, maize and sunflower crops by seed treatment.

MATERIALS AND METHODS

The researches were performed in different localities during 1994-1999, under experimental conditions and in large production plots. For larvae collecting, the soil sampling (25/25 cm area up to 30 cm depth) were used. The insect adults were collected using net method.

For seed treatment experiments with different products in each crop (wheat, barley, maize and sunflower), the most cultivated hybrids and cultivars in the area, were used.

At wheat and barley crops, *lindan* – based products (Lindan 400 C: *lindan* 40%; Lindan HC SC: *lindan* 666 g/l) or mixture of *lindan* with different fungicides (Gammavit 85 PSU: *lindan* 35% + *carboxine* 25% + *thiram* 35%; Masterlin: *lindan* 50% + *tebuconazol* 1.5%; Miclodan Extra 45 PUS: *lindan* 40% + *miclobuthanil* 5%; Procarb L PUS: *lindan* 33% + *carbendazim* 26% + *prochloraz* 7%; Protilin Al 81 PUS: *lindan* 35% + *thiram* 40% + *prochloraz* 6%; Sumidan: *lindan* 500g/l + *diniconazol* 10g/l; Supercarb T 80: *lindan* 35% + *carbendazim* 15% + *thiram* 30%; Supercarb T 585 SC: *lindan* 250 g/l + *carbendazim* 11 g/l + *thiram* 225 g/l; Tirametox 90 PTS: *lindan* 35% + *tiophanat methyl* 20% + *thiram* 35%; Tirametox 625 SC: *lindan* 250 g/l + *tiophanat methyl* 150 g/l + *thiram* 225 g/l; Trialin MT PTS: *lindan* 40% + *tiophanat methyl* 10% + *triadimenol* 10%; Vitalin 85 PTS: *lindan* 35% + *carboxine* 25% + *thiram* 25%) were experimented.

At maize and sunflower crops the following active ingredients were used: *acetamiprid* 70% (Mospilan 70 WP), *bifentrin* 200 g/l (Semafor 20 ST), *carbofuran* 350 g/l (Carbodan 35 ST; Carbofuran 350; Diafuran 35 ST; Furadan 35 ST), *fipronyl* 250 or 500 g/l (Cosmos 250 FS; Cosmos 500 FS), *furatiocarb* 40% (Promet

400 CS), *imidachloprid* 60 or 70% (Gaucho 600 FS; Gaucho 70 WP), *thiamethoxam* 350 g/l (Cruiser 350 FS).

Evaluation of chemicals was done in relation to values of larvae density and attack frequency.

The experimental data have been statistically calculated.

Results and discussions

Annually, wireworms attack about 800,000-1,000,000 ha, especially the crops from Transylvanian Plain and Plateau, Moldavian Plateau, as well as the hilly regions from Muntenia and West Plain (Bărbulescu *et al.*, 1995; Manole *et al.*, 1993, 1999; Mãrgãrit *et al.*, 1988; Popov *et al.*, 1999; Trotus *et al.*, 1994a). Usually, in field crops, the larvae density of different *Elateridae* species varies form 5-10 larvae/m^2, up to 25-40 larvae/m^2, and isolatedly, can exceeds 100 larvae/m^2 (*table 1*).

As the recent researches data show, 60 species of *Elateridae* which belong to 12 subfamilies and 20 genera, have been identified (Manole *et al.*, 1999). The most important species from the viewpoint of both infested area and larvae density/m^2 are: *Agriotes* species, such as *Agriotes ustulatus, Agriotes obscurus, Agriotes ustulatus v. flavicornis, Agriotes sputator, Agriotes pilosus, Agriotes lineatus* and *Agriotes gurgistanus* and other species as *Synaptus filiformis, Selatosomus latus, Melanotus crassicollis* and *Athous hirtus.*

Table 1 - Populations level of wireworm, in field crops in Romania

Agriculturale zone	Locality(County)	Density (larvae/m^2)
Danubian Plain	Malu-Mare (Dolj)	10 - 27
Danubian Plateau	Babeni (Valcea)	12 - 35
Transylvanian Plain and Plateau	Dumbravita (Brasov)	25 - 104
Moldavian Plain	Secuieni (Neamt)	23 - 59
Moldavian Plateau	Bobdanita (Vaslui)	15 - 76
West Romanian Plain	Recas (Timis)	23 - 64

In the field crops (*table 2*), *Agriotes ustulatus* species is the most spreaded one (40.09%), following *Agriotes obscurus* (17.30%), *Agriotes sputator* (11.26%), *Agriotes ustulatus* var. *flavicornis* (10.87%). Moderate or accidental presences were registered for: *Agriotes pilosus* (6.90%), *Agriotes lineatus* (3.85%), *Limonius pilosus* (1.80%), *Synaptus filiformis* (1.55%), *Selatosomus latus* (1.46%) and *Agriotes gurgistanus* (1.38%). The obtained data show that maize, sunflower, winter wheat and barley, alfalfa, clover, as well as potato and sugar beet were the most affected crops. Leguminous crops for grains (soybean, bean, pea or chick-pea), had a moderate or accidental presence of the pests.

Table 2 - Percentage of the main *Elateridae* species, in fieldcrops, in Romania

Species %		Agroecosistem of field crops						
		Winter spiked cereals (wheat, barley)	Row-crops (maize, sunflower)	Leguminous for grains (soybean, pea, bean, chick-pea)	Sugar beet	Potato	Forage crops (alfalfa, clover)	Vegetable fields
Agriotes ustulator	40,09	++++	++++	++	+++	++++	++++	+
Agriotes obscurus	17,30	+++	++++	+++	++	+++	++	+
Agriotes sputator	11,26	+++	+++	+	++	++	+++	-
Agriotes flavicornis	10,87	++	+++	+	++	++	+++	+
Agriotes pilosus	6,90	++	+++	+	++	++	+++	-
Agriotes lineatus	3,85	++	++++	-	++	+	+	-
Limonius pilosus	1,80	+	++	+	+	+	+	-
Synaptus filiformis	1,55	+	++	-	+	+	+	-
Selatosomus latus	1,46	++	+	+	++	+	+	-
Agriotes gurgistanus	1,38	++	+++	-	+	+	+	-
Alte specii	3,54	+	+	-	+	+	+	-

Note: (-) no presence; (+) accidental presence; (++) moderate presence; (+++) high presence; (++++) significant presence

The evolution of the chemical control methods of wireworms from field crops is synthetically presented in *table 3*. During 1950-1990, the prevention of damages produced by wireworms, was mainly performed, by powder application with products on the basis of *DDT, HCH* or *metil chloride*, at the rate of 20-50 kg/ha. When these products such as Duplitox 5+3, Heclotox 3, PEB + Lindan 5+3 or Lindatox 3 PP, were prohibited, at the end of the 1980's, other methods for crop protection against these dangereous pests, replaced organochlorine insecticides applied as powder, because they provided only a low extent of outbreak limitation, at a high lost.

On the basis of numerous studies, the prevention of wireworm attack from the main field crops, wheat, barley, maize, sunflower, is exclusively realized by seed treatment before sowing, with specially conditioned specific products.

At winter spiked cereals, the seed treatment was made with various insecto-fungicides which content *lindan* and different fungicides, their efficacy to control the wireworms being about 85%.

In the case of winter wheat a good simultaneous protection against both harmful insects, inclusively wireworms and specific pathogens, as common bunt or scab was assured. The data from *table 4* show that, in different experimental stations, during 1994-1998, the attack frequency of wireworms varied from 9.0% (Secuieni, 1994) up to 31.6% (Oradea, 1998). The average of the attack frequency per year and stations, was 10.7% in 1994-1995, 20.6% in 1995-1996, 12.1% in 1996-1997 and 20.0% in 1997-1998. Under these conditions, the mean values of efficacy ranged between 79.1% and 83.7% in 1994-1995; 83.6% and 89.5% in 1995-1996; 83.4% and 88.2% in 1996-1997; 83.0% and 88.5% in 1997-1998, the differences between the tested products being insignificant.

Table 3 - Evolution of the chemical methods for prevention and control of wireworms, in field crops, in Romania

Variant	Comercial product(composition)	Rate	Crops
Powder *)	DUPLITOX 5 +3 (*DDT +HCH*)	25 – 30 kg/ha	different field crops
	HECLOTOX 3 (*HCH*)	40- 50 kg/ha	
	PEB + LINDAN 5+3 (*Metil Clor+Lindan*)	25 – 30 kg/ha	
	LINDATOX 3 PP (Lindan)	25 – 30 kg/ha	
Granules	SINORATOX 5 G (*lindan*)	20 – 30 kg/ha	different field crops
	SINOLINTOX 10 G (*dimetoat+lindan*)	15 - 20 kg/ha	
Seed treatment **)	*Lindan* – based products	900 g a.i./t	wheat, barley
	INSECTOFUNGICIDES	900 – 1000 g a.i./t	
	Carbofuran – based products	25 - 28 l/t	maize, sunflower
	CRUISER 350 FS	9 – 10 l/t	
	GAUCHO 600 FS	6 – 10 l/t	
	COSMOS 250 and 500	5,0 and 2.5 l/t	
	SEMAFOR 20 ST	2 – 3,5 l/t	

Note: * Products forbidden before 1992

** Products presented in the table with the results, which are used at the moment

In order to control the wireworms from wheat crops, the following products: Gammavit 85 PSU - 3.0 kg/t; Masterlin - 2.0 kg/t; Miclodan Extra 45 PUS - 2.5 kg/t; Procarb L PUS - 3.0 kg/t; Protilin Al 81 PUS - 3.0 kg/t; Sumidan - 1.8 kg/t; Supercarb T 80 PTS - 3.0 kg/t; Supercarb T 585 SC - 3.75 kg/t; Tirametox 90 PTS - 3.0 kg/t; Tirametox 625 SC - 3.75 kg/t; Trialin MT - 2.5 kg/t; Trialin 50 - 2.5 kg/t; Vitalin 85 PTS - 3.0 kg/t, were registred and promoted in production in our country.

For winter barley the tested insectofungicides showed the same good simultaneous protection against both the soil pests, as wireworms, and the specific pathogens which cause loose smut or barley leaf stripe (*table 5*). In the experimental period the attack frequency of wireworms varied from 6.5% (Oradea 1994) up to 22.5% (Oradea 1995). The yearly means of the attack frequency were 10.5% in 1994-1995; 14.9% in 1995-1996; 15.1% in 1996-1997 and 13.7% in 1997-1998. Under these conditions, the mean values of efficacy varied, between 77.5 and 88.3% in 1994-1995; 89.5 - 91.4% in 1995-1996; 86.9 - 89.4% in 1996-1997 and 84.0 - 90.0% in 1997-1998.

On the basis of the good results in controling the wireworms from barley crops, the following products: Gammavit 85 PSU - 3.0 kg/t; Masterlin - 2.0 kg/t; Miclodan Extra 45 PUS - 2.5 kg/t; Protilin Al 81 PUS - 3.0 kg/t; Sumidan - 1.8 kg/t; Vitalin 85 PTS - 3.0 kg/t, were registered and promoted in production in Romania.

Table 4 - Efficacy of some insecticides and insectofungicides applied as seed treatment against wireworms in winter wheat crops

Experi-mental year	Variant	Rate kg,l/t	Efficacy %			
			Secuieni	Pitesti	Oradea	Average
1994-95	**Untreated** (% attack)		**9,0**	**12,4**	**10,7**	**10,7**
	Tirametox 90 PTS	3,0 kg	77,4	83,3	89,0	83,2
	Gammavit 85 PSU	3,0 kg	77,8	81,5	88,6	82,6
	Miclodan 50 PTS	3,0 kg	67,9	86,8	82,7	79,1
	Supercarb T 80	3,0 kg	77,3	84,7	89,2	83,7
	Vitalin 85 PTS	3,0 kg	67,7	83,0	89,5	79,7
	Lindan 400 SC	2,5 l	78,6	80,6	81,4	80,2
1995-96	**Untreated** (% attack)		**26,0**	**11,5**	**24,3**	**20,6**
	Tirametox 90 PTS	3,0 kg	73,5	92,0	91,1	85,5
	Gammavit 85 PSU	3,0 kg	85,8	91,5	91,1	89,5
	Procarb L	3,0 kg	74,9	91,8	90,9	85,8
	Supercarb T 80	3,0 kg	70,8	91,5	93,3	85,2
	Vitalin 85 PTS	3,0 kg	70,4	92,5	91,1	84,7
	Lindan 400 SC	2,25 l	72,5	90,3	88,1	83,6
	Lindan HC SC	1,35 l	73,9	92,0	93,5	86,6
1996-97	**Untreated** (% attack)		**10,2**	**14,5**	**11,6**	**12,1**
	Procarb L	3,0 kg	88,0	86,3	89,4	87,9
	Tirametox 625 SC	3,75 l	78,9	87,6	83,7	83,4
	Lindan HC SC	1,35 l	74,3	90,3	89,3	84,6
	Miclodan Extra 45	2,5 kg	83,7	89,1	90,3	87,7
	Protilin Al 81 PUS	3,0 kg	85,0	88,4	91,3	88,2
	Trialin MT	2,5 kg	85,3	88,5	85,9	86,6
	Trialin 50	2,5 kg	86,7	87,2	87,6	87,2
1997-98	**Untreated** (% ttack)		**12,6**	**16,0**	**31,6**	**20,0**
	Masterlin PTS	2,0 kg	81,0	84,0	84,0	83,0
	Tirametox 625 SC	3,75 l	80,0	87,0	83,0	83,3
	Supercarb 585 SC	3,75 l	84,0	87,0	87,0	86,3
	Lindan HC SC	1,35 l	87,4	89,0	89,1	88,5
	Trialin MT	2,5 kg	83,0	85,0	85,0	84,3
	Trialin 50	2,5 kg	85,0	87,0	86,0	85,0
	Protilin Al 81 PUS	3,0 kg	86,4	86,9	88,3	86,7
	Sumidan	1,8 l	87,0	90,0	86,4	87,8

Additionally, for treatment of wheat and barley seeds, *lindan*–base products formulated as concentrated suspension, Lindan 400 SC at the rate of 2.25 l/t and Lindan HC SC-1.35 l/t gave good results against wireworms. These products can be applied, as the second treatment, on a seed already treated with a specific fungicide for wheat or barley.

At spring crops, the attack of wireworms can be extremely dangereous, especially for maize and sunflower, in the first vegetation stages. Wireworm attack is typical in moist and cold springs, with deep sowing which delays the germination and growing of the plants.

Under these conditions, even one larva/m^2 can causes losses. From this viewpoint, the crop protection by both cultural and chemical methods (seed treatment) can be assured.

Table 5 - Efficacy of some insecticides and insectofungicides applied as seed treatment against wireworms in winter barley crops

Experi-mental year	Variant	Rate kg,l/t	Efficacy %			
			Secuieni	Pitesti	Oradea	Average
1994-95	**Untreated** (% attack)		**10,5**	**14,5**	**6,5**	**10,5**
	Gammavit 85 PSU	3,0 kg	88,4	87,1	81,3	85,6
	Miclodan 50 PTS	3,0 kg	70,8	86,4	75,6	77,6
	Vitalin 85 PTS	3,0 kg	87,2	86,4	91,3	88,3
1995-96	**Untreated** (% attack)		**9,3**	**12,8**	**22,5**	**14,9**
	Gammavit 85 PSU	3,0 kg	89,2	91,3	92,5	91,0
	Vitalin 85 PTS	3,0 kg	90,5	91,3	92,5	91,4
	Lindan 400 SC	2,25 l	89,2	90,4	88,9	89,5
	Lindan HC SC	1,35 l	90,5	90,0	91,5	90,7
1996-97	**Untreated** (% attack)		**15,0**	**9,2**	**21,2**	**15,1**
	Miclodan Extra 45	2,5 kg	87,0	84,9	89,0	86,9
	Protilin Al 81 PUS	3,0 kg	88,5	88,0	91,7	89,4
	Lindan HC SC	1,35 l	87,0	89,1	87,8	87,9
	Lindan 400 SC	2,25 l	86,5	87,4	90,0	88,0
1997-98	**Untreated** (% attack)		**16,2**	**11,7**	**13,3**	**13,7**
	Masterlin PTS	2,0 kg	86,6	83,4	81,9	84,0
	Protilin Al 81 PUS	3,0 kg	88,6	89,5	92,1	90,0
	Sumidan	1,8 l	83,7	89,0	84,7	85,8
	Lindan HC SC	1,35 l	87,2	87,9	87,9	87,7

In *table 6*, the obtained results in the maize seed treatment are presented. During the experimental period, the maize crops from the above mentioned experimental stations presented an attack frequency level of 15.0% - at seeds, and 12.2% - at collum, in 1995; 22.4%/16.9% in 1996; 17.3%/16.6% in 1997 and 29.8%/28.2% in 1998. Although at the average level (years and stations), the attack values at both seed and collum are closely, with the remark that the seed attack was higher, however at the individual level, the seed attack as compared with that of collum was greater differentiated. Thus, cases when seed attack was higher in comparison with collum attack (Secuieni 1995; 1998) or, on the contrary, seed attack was more reduced (Pitesti 1995, Suceava 1998) stand out. Very high levels for both attack stages (Suceava 1996; Pitesti 1998) are mentioned, too. Under these conditions, the tested insecticides for wireworm control assured a partial protection of plant. On this basis, in order to control wireworms in maize crops, the following products: Carbodan 35 ST - 28 l/t; Carbofuran 350 - 28 l/t; Cosmos 250 FS - 5.0 l/t; Cosmos 500 FS - 2.5 l/t; Cruiser 350 FS - 9.0 l/t; Diafuran 35 ST - 28 l/t; Furadan 35 ST - 28 l/t; Semafor 20 ST – 2.0 l/t; Gaucho 600 FS - 7.0 l/t; Promet 400 CS - 25.0 l/t, gave satisfactory results and were registered and promoted in our country.

Regarding the sunflower crops, the data on the attack of wireworm are presented in *table 7*. It is to notice that in some years, the seed attack (Oradea and Pitesti, 1997) or the collum attack (Pitesti, 1996) exceeded 50%. At the average (year and stations), the frequency of attack was 23.2% at seed and 14.1% at collum in 1995; 48.7%/31.3% in 1996; 24.4%/17.9% in 1997 and 24.3%/23.0% in 1998. The obtained data show a satisfactory efficacy in wireworm control. For sunflower, the following products:

Carbodan 35 ST-28 l/t; Carbofuran 350-28 l/t; Cosmos 250 FS-5.0 l/t; Cosmos 500 FS-2.5 l/t; Cruiser 350 FS-10.0 l/t; Diafuran 35 ST-28 l/t; Furadan 35 ST-28 l/t; Gaucho 600 FS-10 l/t; Semafor 20 ST-3.5 l/t assured protection of the plants and were registred and promoted in protection in Romania.

Table 6 - Efficacy of some insecticides applied as seed treatment against wireworms in maize crops

Experi-mental year	Variant	Rate l/t	Attack frequency %							
			Suceava		Pitesti		Secuieni		Average	
			seed	collum	seed	collum	seed	collum	seed	collum
1995	**Untreated**		**10,7**	**3,9**	**9,6**	**29,4**	**24,6**	**3,4**	**15,0**	**12,2**
	Furadan 35 ST	28,0	2,2	0,5	2,1	10,7	10,3	0,3	4,9	3,8
	Carbofuran 350	28,0	2,3	0,6	2,3	11,3	11,2	0,3	5,3	4,1
	Cosmos 500 FS	2,5	1,3	0,4	2,6	10,0	12,2	0,3	5,4	3,6
1996	**Untreated**		**42,8**	**31,2**	**9,3**	**13,0**	**15,0**	**6,5**	**22,4**	**16,9**
	Carbodan 35 ST	28,0	21,6	11,1	3,7	1,3	7,6	1,3	10,9	4,6
	Promet 400 CS	25,0	17,3	7,9	2,9	1,4	7,3	1,4	9,1	3,6
	Cosmos 250 FS	5,0	19,3	9,6	4,7	1,3	5,0	1,0	9,7	3,9
	Diafuran 35 ST	28,0	20,2	10,7	3,3	1,2	6,4	1,3	9,9	4,4
1997	**Untreated**		**4,6**	**23,8**	**25,2**	**12,0**	**22,0**	**14,0**	**17,3**	**16,6**
	Semafor 20 ST	2,0	2,2	3,6	13,4	3,2	10,3	3,7	8,6	3,5
	Carbofuran 350	28,0	1,8	3,7	10,6	4,0	8,5	3,9	7,0	3,9
	Gaucho 600 FS	6,0	2,9	2,2	5,6	3,6	10,5	5,0	6,3	3,6
	Cruiser 350 FS	9,0	2,1	1,9	3,1	2,7	6,0	5,0	3,7	3,2
1998	**Untreated**		**9,3**	**22,7**	**42,0**	**54,0**	**38,0**	**8,0**	**29,8**	**28,2**
	Furadan 35 ST	28,0	5,5	2,2	4,4	4,8	20,2	3,1	10,0	3,7
	Cruiser 350 FS	9,0	2,5	2,0	6,0	4,0	20,0	5,0	9,5	3,6
	Gaucho 600 FS	6,0	4,3	2,7	6,8	5,4	12,7	3,0	7,9	3,7

Table 7 - Efficacy of some insecticides applied as seed treatment against wireworms in sunflower crops

Experi-mental year	Variant	Rate l/t	Attack frequency %							
			Oradea		Pitesti		Secuieni		Average	
			seed	collum	seed	collum	seed	collum	seed	collum
1995	**Untreated**		**7,9**	**23,1**	**8,0**	**13,7**	**48,0**	**5,5**	**23,2**	**14,1**
	Furadan 35 ST	28,0	2,0	3,2	1,8	5,2	16,5	1,2	6,8	3,2
	Carbofuran 350	28,0	2,3	2,8	2,0	4,7	20,3	1,7	8,2	3,1
	Cosmos 500 FS	2,5	0,8	1,8	1,7	2,7	22,0	2,1	8,2	1,4
	Semafor 20 ST	3,5	1,7	1,4	3,0	6,0	20,1	1,7	8,3	3,0
	Mospilan 70	12,5	1,9	1,1	1,7	4,3	3,0	2,5	2,2	2,6
1996	**Untreated**		**64,0**	**31,7**	**55,0**	**53,0**	**27,0**	**9,2**	**48,7**	**31,3**
	Carbodan 35 ST	28,0	21,6	5,2	3,7	1,3	7,6	1,3	10,9	2,6
	Diafuran 35 ST	28,0	23,9	6,0	15,0	4,5	9,0	3,1	15,9	3,2
	Cosmos 250 FS	5,0	26,4	2,0	17,0	5,2	8,7	2,8	17,3	3,3
1997	**Untreated**		**25,7**	**20,7**	**12,6**	**17,0**	**35,0**	**16,0**	**24,4**	**17,9**
	Furadan 35 ST	28,0	5,2	3,9	2,5	1,5	12,0	4,2	6,6	3,2
	Carbofuran 350	28,0	4,8	3,7	3,6	2,0	8,5	3,9	5,6	3,2
	Gaucho 600 FS	10,0	5,0	2.5	2,5	7,2	14.0	4,3	7,2	4,6
	Cruiser 350 FS	10,0	2,5	1,2	2,2	1,1	15,2	4,1	6,6	2.1
1998	**Untreated**		**26,7**	**20,4**	**25,1**	**35,3**	**21,0**	**13,2**	**24,3**	**23,0**
	Furadan 35 ST	28,0	6,0	3,7	8,0	10,2	3,8	3,7	5,9	5.8
	Carbodan 35 ST	28,0	4,8	4,0	6,5	9,3	4,2	4,0	5,2	5,7
	Cruiser 350 FS	10,0	7,2	4,8	11,6	9,3	9,4	4,2	9,1	6,1
	Gaucho 600 FS	10,0	6,2	3,2	7,1	9,2	9,0	1,7	7,4	4,7

CONCLUSION

In Romania, the main *Elateridae* species are: *Agriotes ustulator* (40.1%), *A. obscurus* (17.3%), *A. flavicornis* (10.9%), *A. pilosus* (6.9%), *A. lineatus* (3.8%), *Limonius pilosus* (1.8%), *Synaptus filiformis* (1.6%), *Selatosomus latus* (1.5%).

The registered densities of the wirevorms in field crops vary from 5 -10 larvae/sq m up to 70-100 larvae/sq m, which determine an attack level up to 31.6% in wheat; 22.5% in barley; 42.8% at seed and 54.0% at collum, in maize and 64.0% at seed and 53.0% at collum, in sunflower.

On the bases of the good results obtained in controling wireworms the following products are registred in our country:

For wheat: Gammavit 85 PSU - 3.0 kg/t; Lindan 400 SC - 2.25 l/t; Lindan HC SC - 1.35 l/t; Masterlin - 2.0 kg/t; Miclodan Extra 45 PUS - 2.5 kg/t; Procarb L. PUS - 3.0 kg/t; Protilin Al 81 PUS - 3.0 kg/t; Sumidan - 1.8 l/t; Supercarb T 80 PTS - 3.0 kg/t; Supercarb T 585 SC - 3.75 l/t; Tirametox 90 PTS - 3.0 kg/t; Tirametox 625 SC - 3.75 l/t; Trialin MT - 2.5 kg/t; Trialin 50 - 2.5 kg/t; Vitalin 85 PTS - 3.0 kg/t.

- For barley: Gammavit 85 PSU - 3.0 kg/t; Lindan 400 SC - 2.25 l/t; Lindan HC SC - 1.35 l/t; Masterlin - 2.0 kg/t; Miclodan Extra 45 PUS - 2.5 kg/t; Protilin Al 81 PUS - 3.0 kg/t; Sumidan - 1.8 l/t; Vitalin 85 PTS - 3.0 kg/t.

- For maize: Carbodan 35 ST - 28.0 l/t; Carbofuran 350 - 28 .0 l/t; Cosmos 500 FS - 2.5 l/t; Cosmos 250 FS - 5.0 l/t; Cruiser 350 FS - 9.0 l/t; Diafuran 35 ST - 28.0 l/t; Furadan 35 ST - 28.0 l/t; Gaucho 600 FS - 6.0 l/t; Promet 400 CS - 25.0 l/t; Semafor 20 ST - 2.0 l/t.

- For sunflower: Carbodan 35 ST - 28.0 l/t; Carbofuran 350 - 28.0 l/t; Cosmos 500 FS - 2.5 l/t; Cosmos 250 FS - 5.0 l/t; Cruiser 350 FS - 10.0 l/t; Diafuran 35 ST - 28.0 l/t; Furadan 35 ST - 28.0 l/t; Gaucho 600 FS - 10.0 l/t; Semafor 20 ST - 3.5 l/t.

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RESEARCHES REGARDING INFLUENCE OF ROUNDUP TREATMENT AND ROUNDUP READY CORN CULTIVATION ON USUAL FAUNA

Ioan Rosca,
University of Agricultural Sciences and Veterinary Medicine
Bucharest, Bdl. Marasti nr. 59, sector 1, Bucharest, Romania

Ioan Sabau,
Monsanto Romania SRL,
str. Barbu Vacarescu, nr. 164, sector 2,
Bucharest, Romania

Abstract
The new technology of corn cultivation, based on using of genetically modified corn hybrids, through inducing herbicides (glyphosate) resistance, have offered to us the possibility to observe if there are some influence of this new technology on *Lumbricus terestris* L. or on surface fauna captured in Barber traps in field. Captured species in Barber traps has shown that, taking into consideration number of Arachnids, Miriapoda or Insects, there are not significant differences between these taxons in experimental plots and no significantly differences between different corn cultivation techniques. It was concluded that herbicide treatment or cultivation of Roundup Ready, corn, or Bt corn, genetically modified, has no influence on *Lumbricus terestris* or main species captured on Barber traps.

Key words: Genetically modified corn, useful fauna.

INTRODUCTION

In the USA, CEE, and also in other parts of the world, the transgenic plants have been authorized by authorities and cultivated over a large scale, after 1996, especially maize, cotton, and soybean. Romania tries to follow the world tendencies regarding the law regulations over the cultivation of this type of plants. Needless to underline the advantages of the cultivation of such plants, that has produced a sheer revolution in the agriculture of many countries. Though there are peoples who, sometimes with good intentions, other times, without knowledge of all data of the problem, or covering mean interests, create to large public the impression that the use of such type of plants originates potential danger for the environment. The new technology of maize cultivation, based on the use of the new technology of the genetic modified maize, through the creation of the resistance to the herbicide (glyphosate), determines the absolute control of all species of annual and perennial weeds. The new technology Roundup Ready shows that the herbicide Roundup Ready is applied post emergent either during one single application of

2-4 l/ha (depending on the degree of infection and domination of weed species) until the faze of maximum 8 leaves, or during two essential treatments. In both situations the quantity of 5 l/ha should not be exceeded, the herbicide is recommended to be applied in 100-150 liters of water per hectare, through using of a reduced volume nozzle. The influence of this technology over the trophic chain from the frame of agrocenosis for maize is a continue concern for both the factors interested in the extension of the surfaces cultivated in maize under this technology, the farmers who are no more to perform expensive mechanical or chemical work for weed fighting, and the organizations, societies or people interested in environment preservation. The experiences performed for this contract included the study of epigeic fauna in maize cultivation, knowing the fact that any treatment with pesticides has a more or less influence over the structure of this fauna and also over the quantity of individuals from the same species, which are to be found in the ecosystem of maize cultivation.

METHODS

In 2000, influence of herbicide or corn hybrid resistant to herbicide on *Lumbricus terestris* L. was observed in experimental plots in field through probes of 25 or 50 cm depth, or on epigeic fauna captured on Barber traps. Experiences were done at Didactic Experimental Farm « Moara Domneasca », not so far from Bucharest and have 2 objectives:

- Studying of ROUNDUP treatment influence in plots with ROUNDUP READY corn (AW 641–RR) and check (AW 641) on night crawler (*Lumbricus terestris* L.), by sampling at 25 and 50 cm deepest.
- Evaluation of epigeic fauna, captured in 10 BARBER soil traps/plot, by 9 times weekly opened for 24 hours, during May 23 –September 15 IX - 2000.

In 2001 the experiments were performed in the field, within the demonstrative lots sowed at The Station for Researches for Irrigated Cultures-Marculesti. The experimental variants pursued, which were placed in the field over significant surfaces in conformity with the plan of experimentation established by the company Monsanto Romania SRL at The Station for Researches for Irrigated Cultures–Marculesti regarding different systems of Roundup Ready maize cultivation were studied in comparison with the witness, from the point of view of the structure and evolution of the useful entomofauna and of the epigeic fauna in the ecosystem of maize cultivation. The variants were composed by 32 rows of 40 m length with 0.7 m distance between the rows. There have been performed mainly surveys and periodical harvests from field of witness maize with Roundup Ready conventionally sowed and minimal works for the soil processing applied either through "no tillage" or "minimum tillage". The experimental variants were sowed in meadow. Variants: 1) "No tillage", corn AW 641 RR; 2) "No tillage", corn DK 512 Bt; 3) "No tillage", corn AW 641; 4) "Minimum tillage", corn AW 641 RR; 5) "Minimum tillage", corn DK 512 Bt; 6) "Minimum tillage", corn AW 641. The quantitative and qualitative establishment of the epigeic fauna, captured in traps on the ground type BARBER, filled with 2/3 phormalin solution 4%, installed in three repetitions, the resulted surface is 136.0248 cm^2 or 0.0136024 m^2 in the studied variants, opened 8 times for 48 hours during 9 VI-5 IX – 2001. The samples captured were harvested, kept in phormalin and determined in laboratory. Taking into account that not all the samples could be determined until the species level, the framing was done until the level of gender, family, order or class. The experimental data were statistically calculated either through the test ANOVA, or the test F. The experience was sowed on 27 April. The variants 1 and 4 (AW 641 RR) were herbicide on 6 April with cu 4 l/ha and on 18 May with 2.5 l/ha. Variants 2, 3, 5 and 6 were herbicide on 2 May by usual technology.

Results

In 2000 it was concluded that herbicide treatment or cultivation of Roundup Ready, corn genetically modified, has no influence on *Lumbricus terestris* (*table 1*) or main species captured on Barber traps (*table 2*). There are not differences especially regarding the useful terrestrial fauna.

Table 1 - Number of *Lumbricus terestris* L. in field

Data of sample	Variant	No. exemplars of *Lumbricus terestris* L.	
		Depth 25 cm	Depth 50 cm
17/V/2000	AW 641	65	16
	AW 641-RR	70.25	4
7/VI/2000	AW 641	22.25	29
	AW 641-RR	23.25	16
21/IX/2000	AW 641	33.75	12
	AW 641-RR	44.25	20

Table 2 - Density/ m^2 of *main* captured species Barber on (24 h)

Useful Fauna	AW 641-RR	AW 641
Arachnids	580.5	840.2
Miriapoda	19.5	13
Pyrrochoris apterus	1878.5	1527.5
Harpalus distinguendus	32.5	39
Pterostichus cupreus	45.5	65
Pterostichus vulgaris	448.5	507
Pterostichus melas	110.5	71.5

In 2001, 1447 specimens were captured in total, appertaining to the following classes: Annelid – 2 (0.14%); Crustacean – 243 (16.79%); Miriapoda – 14 (0.97%); Arachnidan – 46 (3.18%); Insect – 1142 (78.92%). It is important to underline the fact that all the specimens of the crustaceans appertain to the *Oniscus asellus* species, and that Annelid to the *Lumbricus terrestris* species; the other specimens could not be determined and were sent to the specialists for determination. From insects, the captured specimens appertain to the orders: Orthoptera – 46 (4.03%); Dermaptera – 19 (1.66%); Heteroptera – 6 (0.53%); Homoptera – 10 (0.88%); Hymenoptera – 253 (22.15%); Coleoptera – 783 (68.56%); Lepidoptera – 12 (1.05%); Diptera – 13 (1.14%). From Orthoptera, 37 belong to *Gryllus desertus* species, 8 to *Dociostaurus maroccanus* and one larva of Acridid's undetermined. The 19 specimens of Dermaptera belong to one single species (*Forficula auricularia*), while the heteropterous specimens belong: 2 to *Eurygaster integriceps* species and 4 to *Graphosoma lineatum* species. The ten specimens of Homoptera belong to the species *Macrosteles laevis*. Of the 253 specimens appertaining to the order Hymenoptera (ants, wasps, bumble bees), 234 belong to ants; the individuals comprised in the genre *Formica* (77), *Lasius* (22) and *Camponotus* (140), considering that there were incidentally captured species *Dolerus haematodes* (2), *Apis melifica* (7), *Andrena* sp. (1) and representants of the Ichneumonidae (4) family. Out of the total of 758 specimens of Coleoptera captured, 2 (0.25%) belong to family Cicicndelidae, 30 (3.83 %) to family Histeridae – *Hister cadaverinus*, 1 (0.13%) to family Elateridae – *Agriotes lineatus*, 4 (0.51%) to family Coccinelidae – *Coccinella 7 punctata*, 714 (91.19%) to family Carabidae, 1 (0.13%) to family Staphylinidae - *Staphylinus ophthalmicus*, 26 (3.32%) to family Anthicidae - *Formicomus pedestris*, 1 (0.13%) to family Tenebrionidae – *Opatrum sabulosum* and 4 (0.51%) to family Scarabaeidae. Taking into account that the representives of Carabidae family were the most numerous, the analysis of the captured specimens shows the fact that 689 (96.5%) specimens belong to genre *Pterostichus*, 12 (1.68%) to *Carabus* genre, 9 (1.26%) to species *Amara aenea*, 3 (0.42%) to genre *Harpalus* and 1 (0.14%) to species *Zabrus tenebrioides*. Taking into account the fact that it is possible that some specimens belonging to good flyers have incidentally fallen into the ground traps, the fact that a series of specimens could not be determined until the level of species, for the study of the eventual differences among the 6 variants of experimental cultivation only 11 species were taken into account for the epigeic fauna and are found in may variants in a significant number [*Gryllus capmestris* (Orthoptera-Grillidae), *Forficula auricularia* (Dermaptera), *Formica* sp., *Lasius* sp., *Camponotus* sp. (Hymenoptera-Formicidae), *Pterostichus vulgaris, Pterostichus cylindricus, Pterostichus melas, Pterostichus aterrimus* (Coleoptera-Carabidae), *Hister cadaverinus* (Coleoptera-Histeridae) and *Formicomus pedestris* (Coleoptera-Anthicidae)].

The obtained results confirmed through either statistical calculation or through ANOVA test, show that there are no significant differences between the variants (rows) ($F=0.22<F$ crit=2.4), but there are significant differences between the specimens of taxons captured in traps (columns) ($F=14.06>F$ crit=2.02). There could not have been compared the number of specimens belonging to *Pterostichus vulgaris* species (403 captured exemplars) to less abundant species as *P. aterrimus* (5 captured exemplars).

The test F was applied to establish whether there are significant differences regarding the following couples of experimental variants: AW 641 RR ("No tillage"/"Minimum tillage"); DK 512 Bt ("No tillage"/"Minimum tillage"); AW

Table 3 - Total number of the specimens captured in the Barber traps for the 11 taxons, representatives for the epigeic fauna

VARIANTA	*Gryllus capmestris*	*Forficula auricularia*	*Formica sp.*	*Lasius sp.*	*Camponotus sp.*	*Pterostichus vulgaris*	*Pterostichus cylindricus*	*Pterostichus melas*	*Pterostichus aterrimus*	*Hister cadaverinus*	*Formicomus pedestris*
"No tillage", AW 641 RR	8	7	6	3	8	79	1	60	0	1	3
"No tillage", DK 512 Bt	4	3	4	9	24	41	15	54	1	6	4
"No tillage", AW 641	3	2	18	7	18	35	0	44	4	10	3
"Minimum tillage", AW 641 RR	10	3	9	0	64	60	6	38	0	4	10
"Minimum tillage", DK 512 Bt	10	0	16	3	18	70	8	27	0	5	5
"Minimum tillage", AW 641	2	4	24	0	8	118	17	11	0	4	1
TOTAL CAPTURED SPECIMENS	**37**	**19**	**77**	**22**	**140**	**403**	**47**	**234**	**5**	**30**	**26**

	Gryllus capmestris	*Forficula auricularia*	*Formica sp.*	*Lasius sp.*	*Camponotus sp.*	*Pterostichus vulgaris*	*Pterostichus cylindricus*	*Pterostichus melas*	*Pterostichus aterrimus*	*Hister cadaverinus*	*Formicomus pedestris*
"No tillage", AW 641 RR	8	7	6	3	8	79	1	60	0	1	3
"Minimum tillage", AW 641 RR	10	3	9	0	64	60	6	38	0	4	10
"No tillage", DK 512 Bt	4	3	4	9	24	41	15	54	1	6	4
"Minimum tillage", DK 512 Bt	10	0	16	3	18	70	8	27	0	5	5
"No tillage", AW 641	3	2	18	7	18	35	0	44	4	10	3
"Minimum tillage", AW 641	2	4	24	0	8	118	17	11	0	4	1
"No tillage", AW 641 RR	8	7	6	3	8	79	1	60	0	1	3
"No tillage", AW 641	3	2	18	7	18	35	0	44	4	10	3
"Minimum tillage", AW 641 RR	10	3	9	0	64	60	6	38	0	4	10
"Minimum tillage", AW 641	2	4	24	0	8	118	17	11	0	4	1

641 ("No tillage"/"Minimum tillage"); "No tillage" (AW 641 RR/AW641) and "Minimum tillage" (AW 641 RR/AW641). Regarding the existing differences in the maize cultivation over the 11 taxons taken into account, as shown in *table 3*, there are no significant or slight differences between the analyzed couples.

Conclusions

Roundup herbicide treatment or using of genetically modified corn Roundup Ready, are not harmful for main useful fauna and night crawler (*Lumbricus terestris* L.) in field in 2000.

It was also concluded that herbicide treatment or cultivation of Roundup Ready, corn or Bt corn genetically modified, has no influence on main species captured on Barber traps. There are not differences especially regarding the useful terrestrial fauna (*Gryllus capmestris*, *Forficula auricularia*, *Formica* sp., *Lasius* sp., *Camponotus* sp., *Pterostichus vulgaris*, *Pterostichus cylindricus*, *Pterostichus melas*, *Pterostichus aterrimus*, *Hister cadaverinus* and *Formicomus pedestris*).

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ABUNDANCE AND SPATIAL DISTRIBUTION OF SPIDER MITES (ACARI: TETRANYCHIDAE) POPULATIONS ON CORN FIELDS (PORTUGAL)

M. M. Pereira,
Escola Superior Agrária de Beja, Apartado 158,
7800-Beja, Portugal; e-mail: mmpereira@mail.pt;

A. M. Mexia,
Instituto Superior de Agronomia, Tapada da Ajuda,
1399-Lisboa, Portugal

Introduction

The spider mites *Tetranychus* spp. are an important maize pest in some areas of Europe and have potential to develop high populations that may result in significant yield reductions. Usually, there are three species associated to maize agro-ecosystem: *T. urticae* Koch, *T. cinnabarinus* and *T. turkestani* Ugarov & Nikolski. Once spider mites infested a crop, they may increase in abundance for several generations, unless they were controlled by predators and acaricides (Wilson, 1993).

In Portugal, like in many other countries that produce corn, the corn borers are, until now, the most important pest problem. However, lately, and probably as a consequence from the increasing number of insecticides treatments against corn borers (and also others moths), the spider mites that belong to "*Tetranychus* complex" have increased in importance, due to the kill of their natural enemies.

In 1998, a field study was conducted in order to evaluate the abundance and spatial distribution of those mites in three maize fields. So, the primary objectives of this study were to determine the distribution of mite densities along the stalk axis and the infestation intensity for each field sampled, throughout the observation period.

Methodology

The selected fields were sampled on a weekly basis, from tassel to grain filling growth stages. This is the period when spider mites population can increase rapidly and reach high peaks. Each field sampling consisted of three groups of fifty leaves randomly selected and removed from each third of the plants (lower, middle and upper thirds); the samples, identified with the field, date and the third of plant sampled, were carefully observed under a dissecting microscope; the number of adults per leaf and the number of infested leaves were recorded.

It was also recorded the leaf damage using a 1-10 rating scale: 1= 1-10% of the leaf area damage by mite feeding to 10= 91-100% of the leaf area damaged (Archer & Bynum, 1993).

Because the vertical distribution of spider mites *Tetranychus* spp. is largely determined by the phenological stage of crop growth, maize growth stage was recorded each week. With the data obtained, the average rate of plant damage was calculated for each field under study.

RESULTS AND CONCLUSIONS

In all fields we have found adults and leaves damaged by mites feeding and web (*figure 1*).

The maximum adult mite densities were substantially different in each field sampled. Field 2 was the most infested (with more than 3000 adults counted in 150 leaves), but in all three the greater density occurred last week of August (sample 4) (*figure 1*).

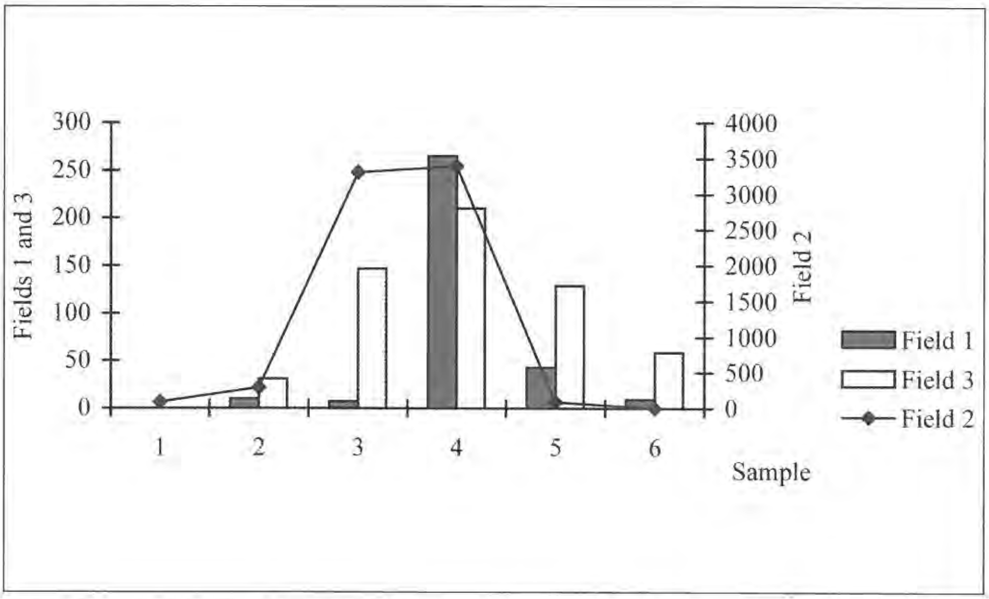

Figure 1 – Total of spider mites adults

As expected, the number of adults increased until sample 4 (end of August) and then it decreased (*figure 1*), probably because spider mites seek out for new habitats, when the food source becomes more dry and less desirable.

Mites disperse up the plant and, as the crop matures, the mites are found closer the tassel. The number (or percent) of leaves infested at each third (*figures 2-4*) usually increased on each successive sample date. These trends were particularly evident at Field 1 (*figure 2*), where the lower third was completely dry at the end of the crop cycle (samples 5 and 6).

Also, on any given sample date more leaves were infested than the preceding one, reaching 50 %, or more, in sample 6 (middle of September).

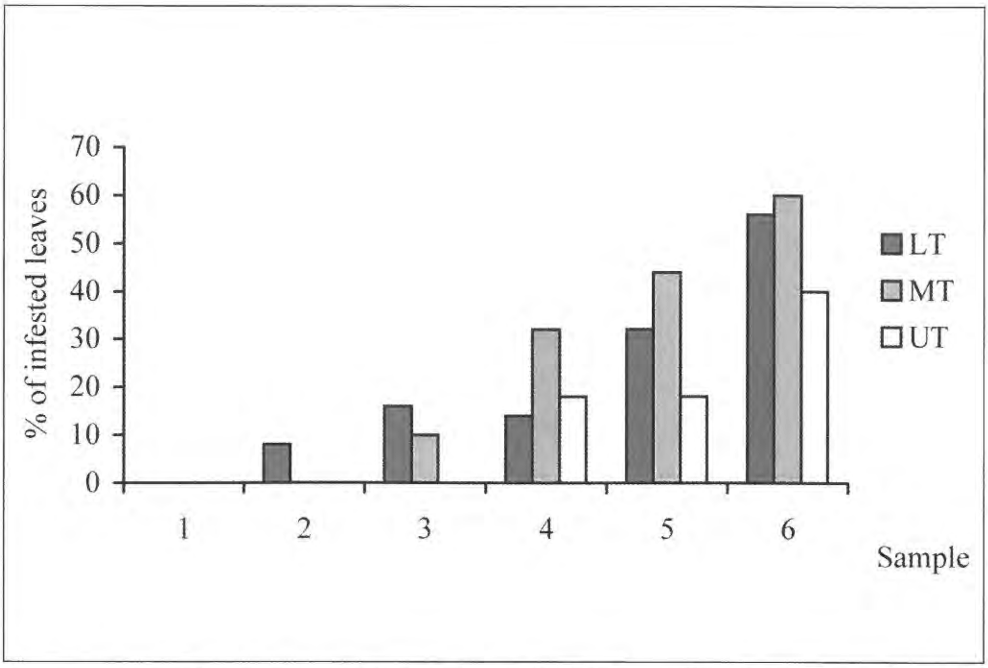

Figure 2 – Infested leaves (%) – Field 1 LT – lower third; MT – middle third; UT – upper third

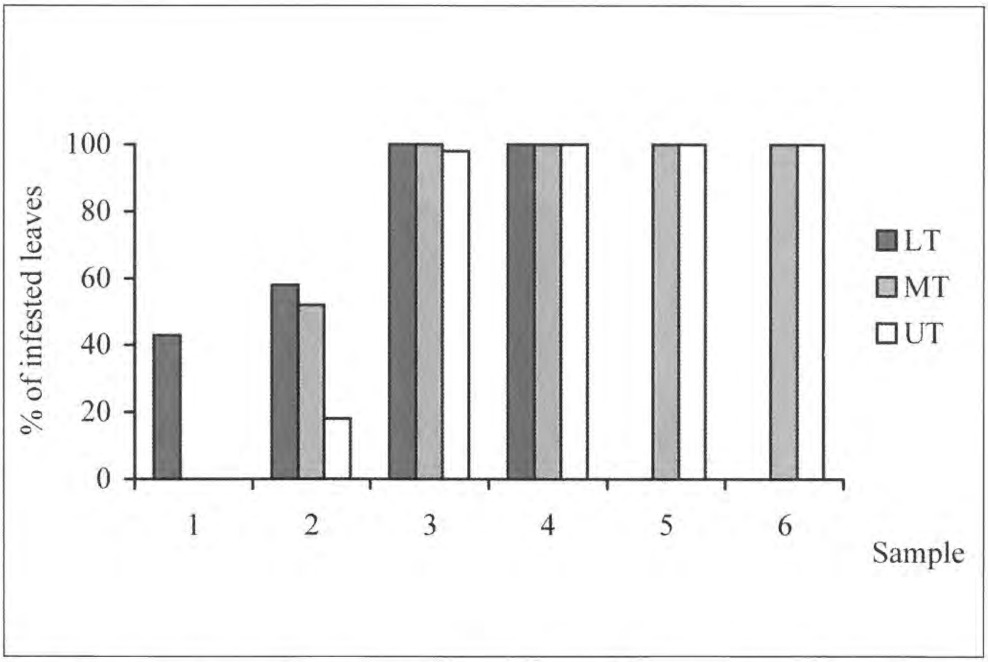

Figure 3 – Infested leaves (%) – Field 2 LT – lower third; MT – middle third; UT – upper third

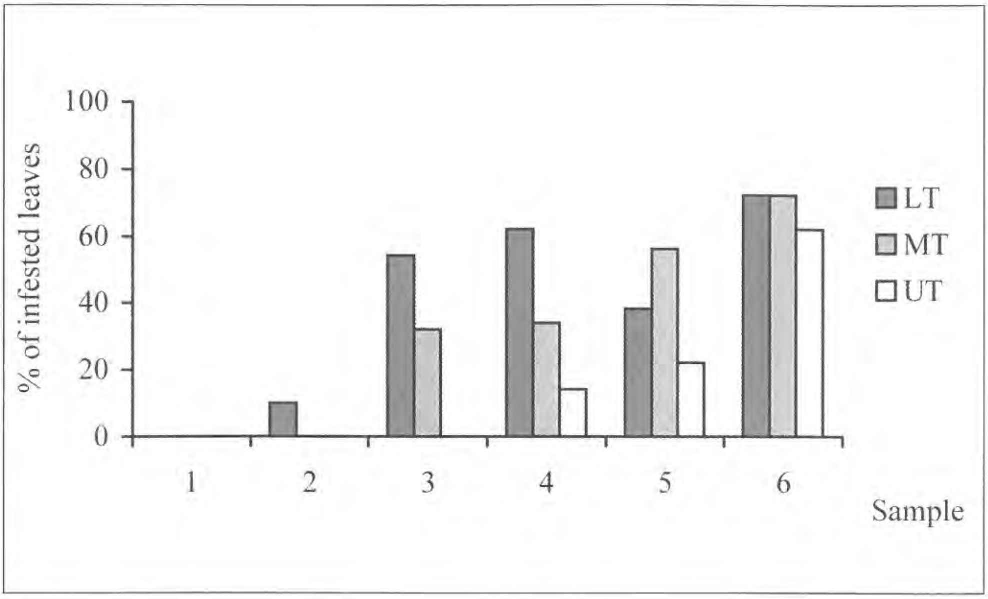

Figure 4 – Infested leaves (%) – Field 3 LT – lower third; MT – middle third; UT – upper third

The Field 2 was always the most infested field (*figure 5*) and also with the grater total of adults (*figure 1*), probably because it was localized near a bean field.

With this field study we could establish the distribution of mites densities throughout the observation period and we confirmed the importance of this pest in the area of the study.

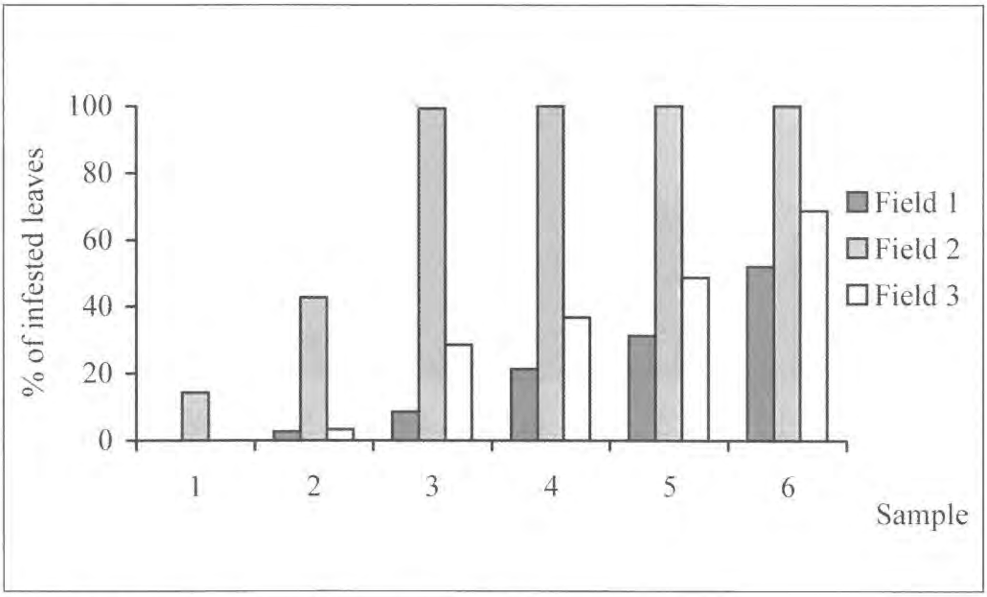

Figure 5 – Average of infested leaves (%) per field

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SEASONAL APPEARANCE OF CEREAL AND MAIZE APHIDS IN GREECE AND FACTORS ASSOCIATED WITH BYDV EPIDEMIOLOGY IN MAIZE

J. A. Tsitsipis, J. T. Margaritopoulos,
Laboratory of Entomology and Agricultural Zoology,
Department of Crop and Animal Production, University of Thessaly,
Fytokou Str. 38 446, Nea Ionia, Magnesia, Greece

N. I. Katis, I. Smyrnioudis,
Plant Pathology Laboratory, Faculty of Agriculture,
Aristotle University of Thessaloniki, 54 006 Thessaloniki, Greece

R. Harrington,
Department of Plant and Invertebrate Ecology,
IACR Rothamsted Experimental Station, Harpenden Herts.
AL5 2JQ, United Kingdom

Abstract

The epidemiology of BYDV in Greece and its economic importance in cereal crops have not been studied in detail. In this study, the seasonal appearance of aphid species, the life cycle category of *Rhopalosiphum padi* L. and *Sitobion avenae* (Fabricious) and various factors affecting the virus epidemiology were examined. Aphid population monitoring was done by means of suction traps in four regions of Greece (Thessaloniki, Velestino, Kopaida and Koroivos) during 1996-98. The species *Metopolophium dirhodum* (Walker), *R. padi*, *Rhopalosiphum maidis* (Fitch), and *S. avenae*, major pests of maize and known vectors of BYDV, were captured in all regions and years. During 1996-1997, higher aphid populations were observed in Thessaloniki than in other regions with an outbreak in 1997 (12433 alatae captured). On the other hand, in 1998 the highest number of aphids was observed further south in Kopaida. Differences as well as similarities were observed in the pattern of captures. However, in all regions a peak in flight intensity of aphid vectors was observed during May and June. Probably, this suggests a migration from wheat and/or barley to maize crops. Flight activity was observed during the whole summer period suggesting a high risk of virus spread. In addition, a significant number of alatae was captured during September and October probably due to the aphid migration to winter crops. The life cycle category of the examined clones, originated from northern Greece, contributes to the former. All clones of *R. padi* and *S. avenae* examined were found non-holocyclic and overwintered in weeds or winter cereal crops. This also suggests a high risk for virus infection in summer maize crops. An important factor affecting BYDV spread is aphid movement. Drought stress and high temperatures were found to promote aphid dispersal and virus transmission. Moreover, the predator *Coccinella septempunctata* L. had a similar effect on aphid movement and virus transmission. On the other hand, the parasitoid *Aphidius rhopalosiphi* DeStefani-Perez did not affect aphid movement. Moreover, the parasitoids had a negative effect on virus spread. Lastly, the effect of BYDV on growth and yield of the maize varieties "Manelis", "Polaris", "Oulis", "Maltos", "Atlantis" and "Damon" was examined. Virus infection did not affect growth and yield of the varieties "Manelis", "Polaris", "Oulis", and "Maltos".

Key words: Aphids, BYDV, Greece, Maize, Wheat.

INTRODUCTION

Barley yellow dwarf virus (BYDV) and cereal yellow dwarf virus (CYDV) are economically important diseases of cereals world-wide. The causal agent is a complex of viruses belonging to the Luteoviridae family (Adams *et al.*, 1998) which are transmitted in a persistent manner (Ossiannilsson 1966) by more than 25 species of aphids (Blackman *et al.*, 1990). The known host range of the viruses includes more than 150 species of the family Poaceae including many cereal crops such as: wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), maize (*Zea mays* L.), oats (*Avena sativa* L.), rye (*Secale cereale* L.), and rice (*Oryza sativa* L.) (Slykhouis *et al.*, 1967, Gould & Shaw 1983). BYDV/CYDV is known to occur in Greece (Panayotou 1980, Ivanovic *et al.*, 1995) but its epidemiology and its economic importance in cereal crops have not been studied in detail. It is well known that many biotic and abiotic factors affect the virus epidemiology, especially the seasonal appearance and population fluctuation of the aphid vectors. In this study, the seasonal appearance of cereal and maize aphid, the life cycle category of *R. padi* and *S. avenae* and various factors affecting the virus epidemiology were examined. The information obtained from this study could be useful in the development of an optimal control strategy for the disease.

MATERIALS AND METHODS

Aphid populations monitoring. The seasonal appearance of the cereal and maize aphids was studied by four Rothamsted type suction traps in four regions of Greece: Thessaloniki in the north, Velestino and Kopaida in Central and South Central Greece, and further south in Koroivos. Aphid populations were monitored during the years 1996-1998 and the traps operated continuously. The traps were inspected daily and/or twice a week and alatae were transferred to the lab for identification. Collected aphids were stored in plastic vials containing two volumes of ethyl alcohol and one volume of lactic acid 75% w/w (Eastop & Van Emden 1972). Aphid identification was based on the keys of Jacky & Bouchery (1980), Taylor (1984) and Remaudière and Seco Fernández (1990).

Life cycle type category. Forty clones of *R. padi* and 19 clones of *S. avenae* were tested. Two *R. padi* and one *S. avenae* holocyclic clones were collected from U.K. to be used as controls. The others were collected from maize and winter cereals in northern Greece in 1997 and 1998. The clones were reared for a minimum of one month at 18°C and L16:D8 and subsequently exposed to short day conditions (SD, L8:D16) at 13°C. The life cycle category was determined by rearing the clones for three generations under SD.

Factors affecting movement of cereal aphids and BYDV transmission. The movement of cereal aphids and BYDV transmission at a range of temperatures and drought stress level was examined. Combinations of three different levels of drought-stress, a moderate (-1.5MPa), a high (-1.9Mpa) drought-stress level and an unstressed control and three different temperatures, (15°C, 10°C and 5°C), were investigated. Durum wheat seedlings (cv. Sifnos) were planted in seed trays (60 cm x 34 cm x 8 cm), 65 plants per tray. When the seedlings were three weeks old, ten viruliferous adult apterous aphids were placed on the middle plant of each tray. Observations were made four times every day for four days. Then, the aphids were discarded and the plants were maintained for four to six weeks before being tested by TAS-ELISA. In another experiment the effect of natural enemies of *R. padi* on the spread of PAV by aphids was examined. Particularly, the parasitoid *A. rhopalosiphi* and the predator *C. septempunctata* were used. A number of seed trays (36 cm x 22 cm x 8 cm) with 33 plants per tray were placed in controlled conditions. Viruliferous aphids and coccinellids or parasitoids were released and the plants were treated with an insecticide (or not) two days, one and two weeks after the release (each time a different tray was treated). Virus infections were evaluated 3-5 weeks later using the TAS-ELISA method.

Resistance of maize varieties in BYDV infections. The effects of a field BYDV-PAV isolate from Greece on growth and yield (plant height, dry weight, no. seeds per ear, seed weight per ear, weight of individuals seeds) of six maize varieties (Manelis, Polaris, Oulis, Maltos, Atlantis and Damon) were examined.

Results and discussion

The data from suction traps revealed that *R. padi, R. maidis, M. dirhodum,* and *S. avenae* were the most abundant species in all regions and years. Different population densities were observed among the four regions depending on the year. During 1996-1997, higher aphid populations were observed in Thessaloniki than in other regions with an outbreak in 1997 (12,433 alatae captured). On the other hand, in 1998 the highest number of aphids was observed further south in Kopaida region (*figure 1*). Differences as well as similarities among the regions were observed in the pattern of captures. In all regions a peak in flight intensity of aphid vectors was observed during May and June. Probably, this suggests a migration from wheat and/or barley to maize crops. Flight activity was observed during the whole summer period suggesting a high risk of virus spread. In some cases, a significant number of alate aphids were captured during September and October, probably due to the aphid migration to winter crops (*figures 2-5*).

The experiment life cycle type category revealed that *S. avenae* and *R. padi* overwinter mainly as parthenogenetic morphs in weeds or winter crops in regions of

North Greece. There were not any holocyclic clones among the examined ones. Thrirty six *R. padi* and 17 *S. avenae* clones were found androcyclic (produced virginopae and some males). Two clones of *R. padi* and one of *S. avenae* had completely lost their ability to produce sexual morphs. However, it is worth mentioning that more clones should be examined from the same and other regions in order to obtain a complete picture about the overwintering strategy that species invest in Greece.

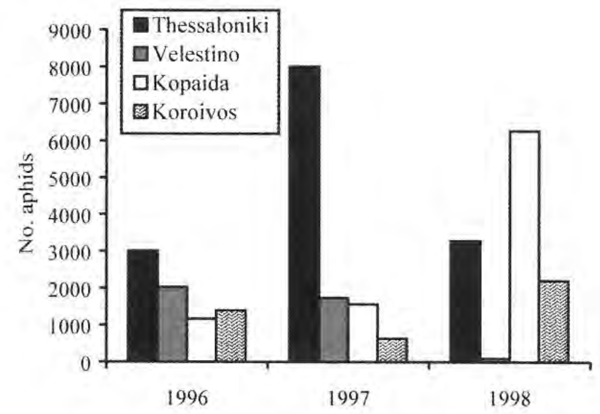

Figure 1 – Alate aphids, BYDV vectors, caught in Rothamsted type suction trap in four regions of Greece

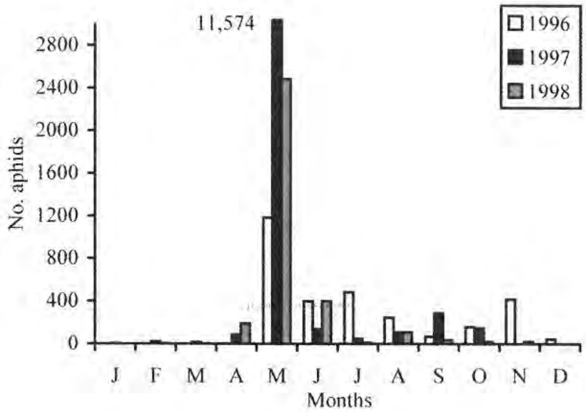

Figure 2 – Number of alate aphids, BYDV vectors, caught in Rothamsted type suction trap in Thessaloniki

Regarding the factors that affect movement of aphids and BYDV transmission, an increase in the number of plants visited by aphids and infected by the virus at higher water stress and temperature was observed. Drought-stress had no effect on aphid movement or dispersal of the virus at 10°C and 5°C. A higher proportion of the visited plants were infected when plants were stressed than when not stressed and at 15°C than at lower temperatures. Therefore, it seems that drought stress is of considerable importance in aphid dispersal and virus spread, but only at higher temperature (Smyrnioudis *et al.*, 2000a). The presence of the parasitoid *A. rhopalosiphi* decreased the aphid movement and the number of plants infected by the virus. On the other hand the predator *C. septempunctata* increased aphid dispersal and virus spread even after two days from the time of release (Smyrnioudis *et al.*, 2000b).

A variation on the effect of BYDV on growth and yield of the maize varieties examined was observed. The responses of the maize varieties "Manelis", "Polaris", "Oulis", and "Maltos" were similar. The virus infection did not affect the yield and growth of these four varieties. In "Atlantis" the weight of the seeds and number of seeds were slightly affected by virus infection. Moreover, in "Damon" the weight of single seeds was slightly affected by BYDV infection.

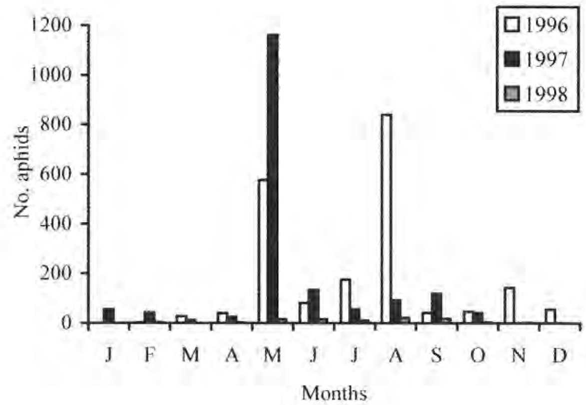

Figure 3 – Number of alate aphids, BYDV vectors, caught in Rothamsted type suction trap in Velestino

In conclusion, the data concerning the seasonal appearance and population fluctuations of the aphid species examined suggest that there is a high risk of virus spread during the period of May to October. Specifically, two periods are of primary importance in the spread of BYDV, May-June when aphids migrate from wheat to maize and autumn due to aphid migration to winter crops. Therefore, early seeding for maize in spring and late for wheat in autumn should be considered in order to avoid synchronisation between the sensitive stages of the crops and the alatae migration. In addition, many other factors are of primary importance in virus epidemiology. The anholocyclic way of life of *R. padi* and *S. avenae* in the region examined suggests a high risk of virus infection. Most of the aphid vectors of BYDV-PAV in Greece can overwinter in crops thus having the potential to spread virus during this period and in spring. The abundance of aphids will depend on the weather during the winter. Low temperature and high rainfall will cause the reduction in numbers of the aphids for the rest of the season and, as a result, reduce the spread of BYDV. On the other hand, mild winters will favour the aphids and, as a result, increase the spread of BYDV. Drought stress, temperature and natural enemies affect aphid dispersal and virus spread. The combination of drought-stress and high winter temperature would seem ideal for BYDV/CYDV outbreaks. Higher winter temperatures due to global warming may increase the risk of epidemics. In the parasitoid experiment described here there was not an increase or decrease in aphid movement but there was a reduction in the number of plants infected with the virus, which indicates that the parasitoids had an negative effect on virus spread. The importance of predators in reducing aphid populations may be overridden by dislodging aphids even with fewer aphids in the predator-exposed cages. Therefore, the abundance and degree of activity of aphid predators may be an important aspect of the epidemiology of aphid transmitted viruses. Maize is likely to be a source of inoculum for cereals in irrigated areas in Greece but not likely to cause a direct economic damage. New varieties should be tested, to ensure tolerance to BYDV/CYDV. Finally, the former information should be considered in the development of an optimal control strategy for the disease.

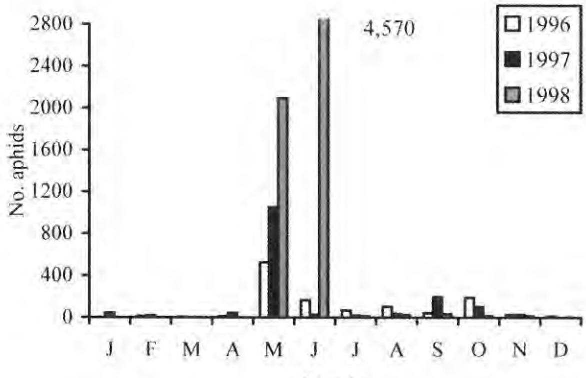

Figure 4 – Number of alate aphids, BYDV vectors, caught in Rothamsted type suction trap in Kopaida

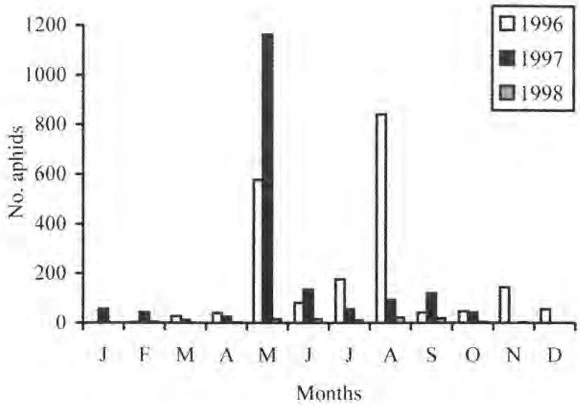

Figure 5 – Number of alate aphids, BYDV vectors, caught in Rothamsted type suction trap in Koroivos

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BLACK CUTWORM ALERT PROGRAMME IN ITALY

Lorenzo Furlan, Sergio Zangheri,
Department of Agronomy, Entomology, University of Padova,
Agripolis-Via Romea 16-35020 Legnaro PD, Italy
e-mail: lorenzo.furlan@inwind.it

Stefano Barbieri,
Veneto Agricoltura, Corte Benedettina, via Roma 34, Legnaro PD

Susanna Lessi, Irene Delillo, Adriano Barbi,
A.R.P.A.V., Meteorological Center of Teolo,
via Marconi 55, 35037 Padova

Francesco Brichese,
Regione Veneto

Abstract

Agrotis ipsilon (BCW) has caused severe damage to maize in some years in Italy. Therefore a black cutworm alert programme able to inform farmers in a timely manner about the presence of black cutworm economic populations has been tested in Veneto region. The procedure suggested by Corn Belt of USA Universities and Extension Services was implemented in Veneto region beginning in 1991. Sex pheromone traps were placed in maize fields of the different Veneto provinces beginning in late February of each year. Air and soil temperatures, information about direction and strength of winds were supplied by A.R.P.A.V. Degree days (development zero = 10.4 °C) were calculated beginning with the first captures on the traps. Ten maize fields in the area where males had been captured were investigated every 2 days beginning in late April of each year.

Very low numbers of moths were observed in some years. In those years, no black cutworm damage on maize plants was observed. The alert programme gave reliable results by using soil surface temperatures in calculating Degree Day accumulations, while the use of air temperature underestimated the DDA. In most years, the first fourth larval instars were observed some days before the date forecasted by the model (176 DDA for 50% of 4th instar larvae in the population). It was possible to forecast the period of larval damage thus making effective the rescue treatments where threshold had been met. Information was transmitted through a specific bulletin issued by A.R.P.A.V., Internet, newspapers and television channels.

Key words: *Agrotis ipsilon*, Alert Programme, maize.

INTRODUCTION

Agrotis ipsilon Hufnagel, (Lepidoptera, Noctuidae) (BCW) represents a serious problem in Europe (Bues *et al.*, 1990; Hachler, 1988, 1989); in Italy it has caused severe damage to maize and other crops (thousands of hectares were destroyed) in some years particularly in 1971 and 1983 (Zangheri and Ciampolini, 1971; Zangheri *et al.*, 1984). Almost yearly, local outbreaks can cause reductions of corn stand or even the need for replanting (Furlan, 1989). Usually the farmers realize the extent of damage too late and rescue treatments become ineffective. Studies carried out in Italy demonstrated that the irregular outbreaks of the Noctuid in Northern Italy could usually be associated with the oviposition of migrant females at the beginning of spring (Zangheri *et. al.*, 1998). Therefore, a black cutworm alert programme that can inform the farmers about the presence of black cutworm economic populations in real time has been tested in the Veneto region.

MATERIALS AND METHODS

The procedure suggested and used by Universities and Extension Services in Corn Belt of USA (Archer and Musick, 1977, 1980; Showers *et al.*, 1985, 1986; Showers, 1997; Troester *et al.*, 1982; Von Kaster and Showers, 1984) was implemented in Veneto region beginning in 1991. Sex pheromone traps (Hartstack baited with lures prepared by INRA and Plant Protection Institute of Budapes, at least 10 to 35 per year) were placed in maize fields of the different Veneto provinces from late February of each year. Air and soil temperatures, information about direction and strength of winds were supplied by A.R.P.A.V. Degree days (development zero = 10.4 °C, Luckmann *et al.*, 1976) were accumulated (average of maximum-minimum method) beginning with the first captures on traps. These captures occurred just after strong winds coming from Southern regions. 176 DDA were considered for 50% of 4th instar larvae in the population. Tens maize (and sometimes sugar beet and sunflower) fields (0,5 to 2 hectars each) in the area where males had been captured were investigated every 2 days from late April of each year to when the 4th instar larvae started damaging the young plants. Each field was scouted by choosing 8 random areas of 20 m X 10 maize rows per field and observing all plants. Plants with typical black cutworm damage were individually checked and all the larvae found near the collars on each plant were collected and determined to species. At least 20 to 200 larvae per year were collected on damaged plants. The larval instar of captured BCW was estimated by using the head capsule data of Archer and Music (1977).

RESULTS

Moth captures greatly varied in different years and in different provinces of the region (*table 1*). Very low numbers of moths were observed in some years, especially when no or weak southern winds occurred; in these years (1992, 1993, 1995, 1997) no black cutworm damage on maize plants was observed. In contrast in 1998, first significant migrant flight was observed between 5 and 12 April just after strong southerly winds had occurred. Soil temperatures were constantly higher than air temperatures and thus reach 176 DD 2-3 days earlier (*table* 2).

Table 1 - Synthetic results of the implementation of the Black Cutworm Alert programme in Veneto over the last 11 years

Year	First month captures	First significant flight	Flight level	4th instar first larvae	peak of 4th instar larvae	Forecast date for 176 DD	Damage level
1991	6 March	21-26 March	medium	NO larvae found			very low
1992	1 April	3-6 April	low	NO larvae found			NO DAMAGE
1993	29 March	6 April	low	NO larvae found			NO DAMAGE
1994	4 March	23 - 26 March	medium	5 May	7-8 May	8-13 May	medium
1995	11 March	NO	very low	NO larvae found			NO DAMAGE
1996	18 March	3 April	medium	2 May	6-8 May	9-11 May	medium
1997	24 March	NO	very low	NO larvae found			NO DAMAGE
1998	16 March	5-12 April	high	13 May	15-17 May	8-13 May	medium
1999	26 March	6 April	low	10 May	14 May	5-10 May	low
2000	29 March	29 March - 5 April	medium	4 May	8 May	4-8 May	low
2001	2 March	17 March	medium	29 April	1-2 May	5-9 May	medium

Flight level: *very low* means less than 1 moth/trap/day at the peak; *low* means 1 to 5 moths/trap/day at the peak; *medium* means 5 to 10 moths/trap/day at the peak; *high* means > 10 moths/trap/day at the peak

Damage level: *very low* means no or less than 0.1% of plants damaged in all the fields; *low* means no replanted fields and number of plants damaged 0.1 to 2%; *medium* means 0.5 to 5% of sampled surface replanted and in other fields number of plants damaged 2 to 30%.

Table 2 - BCW Alert programme. Differences between the number of days to reach 176 Degree Days forecast using soil temperatures and that forecast using air (2 m) temperatures in different localities of Veneto region in different years

year	Vazzola	Sorgà	Portogruaro	Legnaro	average annual differences	average difference 1996-2001	standard deviation 1996-2001
	0	2	8	5	3,75		
1996	5	4	3	2	3,5		
1998	2	4	2	3	2,75		
1999	2	6	2	-1	2,25		
2000	2	2	2	-2	1		
2001	3	-	4	0	2,33	2,60	2,24

Since the 1st-3rd *A. ipsilon* instars live off young weed and crop plants close to soil surface, soil temperatures were chosen to implement the model. When conspicuous larval populations established model performances varied significantly (*table 1*) as was observed by Kaster and Showers, 1984. In some years (1998, 2000) the prediction matched very well to the field observations. In most cases, the first 4th instar larvae were found a few days (3-7) before the date forecast by BCW Alert programme. The differences were smaller taking into consideration the peak of larval presence (year 1994, 1996, 2001). These data are in agreement with those of Story *et. al.*, 1984 who observed that 4^{th} and 5^{th} BCW instars may be present in fields when < 125 degree-days (sine wave method) or 100 degree-days (average of maximum-minimum method) have accumulated since 1 January using developmental data of Luckmann *et al.*, 1976. Only in one year (1999) was the predicted date slightly earlier than actual findings in the field, but in this case the population level was very low so that it is likely that the first larvae that moulted to 4^{th} instar were not found.

Conclusions

Moth capture levels by Hartstack traps can reliably indicate the years at risk from BCW. The BCW alert programme gave more reliable results by using soil surface temperatures in calculating Degree Days accumulation, while the use of air temperatures underestimated the DDA. The model tends to indicate 4^{th} instar larvae presence in corn fields some days later than actual findings. Some years the prediction was quite correct, while in other years the first fourth instar larvae were observed 3 to 7 days before the date forecast by the model (176 DDA for 50% of 4^{th} instars larvae in the population). Therefore, scouting in high-risk maize fields must begin about 7 days earlier than the model predicts to make sure that the first outbreaks can be detected in time. Taking this into consideration, it was possible to forecast the period of larval damage. This allowed for effective rescue treatments where thresholds had been reached or exceeded. Information was transmitted through specific bulletins issued by A.R.P.A.V., Internet, newspapers and television channels.

Acknowledgements to Mr. Gilbert Pavanello and Dr. Renzo Converso for BCW monitoring at different sites.

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IWGO

INTERNATIONAL WORKING GROUP ON *OSTRINIA* AND OTHER MAIZE PESTS

The International Working Group on Ostrinia and Other Maize Pests is an International Working Group dealing worldwide with all matter of maize pests.

One of the oldest Working Groups within Global IOBC is certainly the International Working Group on *Ostrinia* (IWGO). The group was started during an International Congress in Moscow in 1968, but the roots reach back to the USA regional project on Ostrinia already begun in 1951. IWGO was established through this USA regional project. The founders of the group were D. HADZISTEVIC (Yugoslavia), who had the original idea of founding a group for international cooperation, H.C. CHIANG (USA), who brought the ideas of the USA regional project into the group, I.D. SHAPIRO (Russia of the USSR), T. PERJU (Romania), C. KANIA (Poland) and B. DOLINKA (Hungary). All were well known researchers or maize-breeders. The group was originally organized so that each member country had an official "member" representative and all other participants were classified as "associate members." Researchers who took part in meetings from time to time were called "guest members".

The original idea of IWGO was to exchange inbred lines within the group and test these lines for resistance against the most important maize pest throughout the world, the European corn borer (ECB), *Ostrinia nubilalis* Hubner. The results of this breeding program were to be made available to all member countries. Up to now, three synthetics resistant breeding lines to ECB have been developed and released (IWGO 1, 2, and 3, both late and early). Most of the results of this testing program were published earlier by IWGO. As the membership of the group increased, interests in other areas of ECB research expanded. A pheromone project was established by France (INRA). The influence of other pests of maize became more and more important over time and colleagues from Asia wanted to include problems with *Ostrina furnacalis* and southern European members brought research work concerning *Sesamia nonagroides* into the group. The appearance of *Diabrotica virgifera virgifera* in Europe (Serbia) in 1992 became a further matter of discussion within the group. This appearance was so important that even a subgroup within IWGO was founded in 1996. Additionally, *Elateridae* (wireworms) were a topic of discussion by several member countries. Subsequently, corn borer biology and host response were also studied. More recently biological control has been emphasized. Therefore, within the last several years IWGO has become more and more a working group on all maize pest problems. No longer are members just considering the exchanging of inbred lines for testing for resistance to the European corn borer.

The group has held 20 annual (since 1980 biannual) meetings in one of the member states. Several publications have been released and some are still partly available. Since 1981 "IWGO - NEWSLETTER" has been published. This has been a way to link the members and to establish a permanent record of the activities of the working group, distribute information about the members, and to publish the abstracts of papers presented at the congresses. After several meetings proceedings of the papers presented were issued .

Prof. H.C. CHIANG was the first president (convenor) of the group and held this position until 1982. The group elected P. ANGLADE (Bordeaux, France) as the new president in 1982 and he served until 1993. Since 1994, Harald K. BERGER (Vienna, Austria) has been the convenor of this international group. As the group and the topics discussed and researched grew, the necessity of the nomination of vice presidents (Sub- or Co - Convenors) came up. Prof. Dr. Les LEWIS (Iowa State University; USA) became Vice Convenor (Vice President) and Prof. Dr. Rich EDWARDS Convenor of the *Diabrotica* subgroup, which was established in 1996.

IWGO, which became a Global-IOBC Working Group in the meantime, is now a well established large international working group which now deals with all matters of maize pests and pest resistance. The group is open to all scientists with interest in working within an international group (with familiar and personal contact among the members).

VENETO REGIONAL PHYTOSANITARY SERVICE

The Regional Phytosanitary Service was founded in Verona in 1923, as a technical organ of the Italian Agriculture Ministry. Afterwards, in 1931, it became the Royal Observatory for the Diseases of the Plants and nowadays it's one of the structures of the Veneto Region.

The Regional Phytosanitary Service duties are:

- to certify, all over the regional area, the plant products in import and export both in EU and extra EU market to forbid the marketing and the diffusion of infected or contaminated material;
- to coordinate the compulsory struggles against plant parasites and pathogens;
- to carry out labs analysis on plants, plant products and soil samples for the monitoring of fungus, bacterium, virus, nematode worms, bugs and acarus
- research and study of new IPM techniques, in addition to technical and specialist assistance, to protect the qualities of the productions, to respect the environment and the health of farmers and of consumers;
- to be a reference for the protection of the forests and the public and private parks and gardens.

VENETO AGRICOLTURA

Veneto Agricoltura is the Veneto Region's institute that promotes and realizes interventions for the modernization of the agricultural structures, for the development of aquaculture and fishing, with particular reference to the activities of research and testing of the agricultural, forestry and agro industrial sectors and as a support to the market.

Veneto Agricoltura promotes process and product innovation of the competent sectors, also through the agricultural divulgation and rural activity and delivers specialist services for exploitation and marketing of regional typical products; promotes and organizes quality certification activities of alimentary products; improves rational employment of environmental resources and applied search activity, testing, information and vocational training and it is especially alert to the biodiversity preservation through the management of regional forest nurseries, nature reserves and Veneto Region's property forest.

Scientific Commitee

Harald K. Berger – Convenor of IWGO – Federal Office and Research Centre for Agricolture (A)

C. Richards Edwards – Convenor of Subgroup Diabrotica – Purdue University, Department of entomology (USA)

Jozsef Ziss – Szent Istvan University, Department of Plant Protection (HU)

Stefan Vidal – Georg-August-University Goettingen, Institute for Plant Pathology and Plant Protection (DE)

Lorenzo Furlan – Department of Agronomy, Entomology, University of Padua (I)

Vincenzo Girolami - Department of Agronomy, Entomology, University of Padua (I)

Bruno Minello – Veneto Regional Phytosanitary Service (I)

Marco Vettorazzo – Veneto Regional Phytosanitary Service (I)

Luigino Disegna – Azienda Regionale Veneto Agricoltura (I)

Giovanni Chillemi - Azienda Regionale Veneto Agricoltura (I)

Organizing Commitee

Harald K. Berger – Convenor of IWGO – Federal Office and Research Centre for Agricolture (A)

Lorenzo Furlan – Department of Agronomy, Entomology, University of Padua (I)

Marco Vettorazzo – Veneto Regional Phytosanitary Service (I)

Franco Norido – Azienda Regionale Veneto Agricoltura (I)

Stefano Barbieri – Azienda Regionale Veneto Agricoltura (I)

Francesca Grandis – Azienda Regionale Veneto Agricoltura (I)

Silvia Ceroni – Azienda Regionale Veneto Agricoltura (I)

Vanna Funes - Azienda Regionale Veneto Agricoltura (I)

Name AGAZZI GIORGIO
Structure OSSERVATORIO MALATTIE PIANTE

Address VIA BEATO ODORICO 13
Postal code - City PORDENONE *Country* ITALIA
Phone 0434 529330 *Fax* 0434 520570
E-mail omp.pn@regione.fvg.it

Name AGNES ANDREA
Structure SETTORE AGRICOLTURA PROVINCIA DI NOVARA

Address VIA DOMINIONI, 4
Postal code - City 28100 NOVARA *Country* ITALY - I
Phone *Fax*
E-mail andreaagnes.novara@reteunitaria.piemonte.it

Name ALLARA MANUELA
Structure PLANT PROTECTION SERVICE, FAO

Address VIALE DELLE TERME DI CARACALLA
Postal code - City 00100 ROMA *Country* ITALY - I
Phone 06 57052907 *Fax* 06 57056227
E-mail Manuela.Allara@fao.org

Name BACA FRANJA
Structure MAIZE RESEARCH INSTITUTE "ZEMUN POLJE"

Address ST. BAJICA 1
Postal code - City 11185 BELGRADE-ZEMUN *Country* YUGOSLAVIA - YU
Phone ++381113756707 *Fax* ++381113756707
E-mail FBACA@MRIZP.CO.YU

Name BARBIERI STEFANO
Structure AZIENDA REGIONALE VENETO AGRICOLTURA - SETTORE DIVULGAZIONE TECNICA FORMAZIONE PROFESSIONALE

Address VIA ROMA 34
Postal code - City 35020 LEGNARO (PD) *Country* ITALY - I
Phone +39 (0) 498293947 *Fax* +39 (0) 498293909
E-mail stefano.barbieri@venetoagricoltura.org

Name BARBULESCU ALEXANDRO
Structure RESEARCH INSTITUTE FOR CEREALS AND INDUSTRIAL CROPS

Address NICOLAE TITULESCU str., 1 - DISTRICT CALARASI
Postal code - City 8264 FUNDULEA *Country* ROMANIA - RO
Phone +401-3110722 *Fax* +401-3110722
E-mail fundulea@cons.incerc.ro

Name	BAUFELD PETER		
Structure	FEDERAL BIOLOGICAL RESEARCH CENTRE OR AGRICOLTURE AND FORESTRY-DEPARTMENT FOR NATIONAL AND INTERNATIONAL PLANT HEALTH		
Address	STAHUSCLORFE DAMM 81		
Postal code - City	D - 14532 KLEINMACHNOW	*Country*	GERMANY - D
Phone	+4933203-48(276)	*Fax*	+4933203-48(385)
E-mail	p.baufeld@bba.de		

Name	BAYAR KHOSBAYAR		
Structure	ST. ISTVAN UNIVERSITY DEPARTMENT OF PLANT PROTECTION		
Address	PATER K. 1		
Postal code - City	H 2103 GODOLLO	*Country*	HUNGARY - H
Phone		*Fax*	
E-mail	khosbayar@excite.com		

Name	BERBEROVIC HUSEIN		
Structure	FACULTY OF AGRICULTURE SARAJEVO		
Address	ZUIAJA OD BOSNE 8		
Postal code - City	71000 SARAJEVO	*Country*	BOSNA I HERZEGOVINA BiH
Phone	38733653033	*Fax*	38733667429
E-mail	RNUMIC@UTIC.NET.BA		

Name	BERGER HARALD K.		
Structure	BUNDESAMT UND FORSCHUNGSZENTRUM FL		
Address	SPARGELDFELDSTRASSE 191		
Postal code - City	A - 1226 WIEN	*Country*	AUSTRIA - A
Phone	+43-1-732-16/2002	*Fax*	+43-1-732-16/2108
E-mail	hberger@bfl.at		

Name	BERTOSSA MARIO		
Structure	SWISS FEDERAL RESEARCH STATION FOR PLANT PRODUCTION		
Address	CENTRO DI CADENAZZO		
Postal code - City	6594 CONTONE - TICINO	*Country*	SWITZERLAND - CH
Phone	0041 918502034	*Fax*	0041 918502039
E-mail	mario.bertossa@rac.admin.ch		

Name	BORIANI LUCA		
Structure	CENTRO AGRICOLTURA AMBIENTE S.R.L.		
Address	VIA DI MEZZO LEVANTE, 2233		
Postal code - City	40014 CREVALCORE - BO	*Country*	ITALY - I
Phone	051/981698	*Fax*	
E-mail	Lboriani@caa.it		

Name	BORIANI MARCO		
Structure	LOMBARDY REGION, PHYTOSANITARY SERVICE		
Address	PIAZZA IV NOVEMBRE, 5		
Postal code - City	20100 MILANO	*Country*	ITALY - I
Phone	0267658013	*Fax*	0267658019
E-mail	marco_boriani@regione.lombardia.it - maalces.bor@tiscalinet.it		

Name	BRMEZ MIRJANA		
Structure	FACULTY OF AGRICOLTURE IN OSIJEK		
Address	TRG SU.TROJSTVA 3		
Postal code - City	31000 OSIJEK	*Country*	CROATIA - HR
Phone	++38531224-280	*Fax*	++38531207-017
E-mail	mirjanam@suncokret.pfos.hr		

Name	BROWN TOM		
Structure	FLORIDA FOOD PRODUCTS		
Address	P.O. Box 1300		
Postal code - City	EUSTIS FLORIDA 32727-1300	*Country*	USA
Phone	410 848 7758	*Fax*	
E-mail	schroderçqis.net		

Name	BUTRON GOMEZ ANA MARIA		
Structure	MISION BIOLOGICA DE GALICIA (CSIC)		
Address	APTDO 28		
Postal code - City	86080 PONTEVEDRA	*Country*	SPAIN - SP
Phone	986854800	*Fax*	986841362
E-mail	csgpoamb@cesga.es		

Name	CAMOZZA MARTA		
Structure	AZIENDA REGIONALE VENETO AGRICOLTURA - SETTORE DIVULGAZIONE TECNICA FORMAZIONE PROFESSIONALE		
Address	VIA ROMA 34		
Postal code - City	35020 LEGNARO (PD)	*Country*	ITALY - I
Phone	+39 (0) 498293904	*Fax*	+39 (0) 498293909
E-mail	marta.camozza@venetoagricoltura.org		

Name	CASTAGNOLI FILIPPO		
Structure	AGRONOMIST FREE LANCE		
Address	VIA DAMIANO CHIESA 3/1		
Postal code - City	31021 - MOGLIANO VENETO (TV)	*Country*	ITALY - I
Phone	041-5902195	*Fax*	
E-mail	fc0740@libero.it		

Name	CASTANERA PEDRO		
Structure	CSIS, CIB,		
Address	VELAZQUEZ, 144		
Postal code - City	28006 MADRID	*Country*	SPAIN - SP
Phone	+34-91-5611800	*Fax*	+34-91-5627518
E-mail	castan@cib.csic.es		

Name	CHILLEMI GIOVANNI		
Structure	AZIENDA REGIONALE VENETO AGRICOLTURA		
Address	VIA ROMEA - AGRIPOLIS		
Postal code - City	35020 LEGNARO (PD)	*Country*	ITALY -
Phone	041-8293711	*Fax*	049-8293815
E-mail	giovanni.chillemi@venetoagricoltura.org		

Name	COLOMBI LUIGI		
Structure	SERVIZIO FITOSANITARIO		
Address	VIA S. FRANSCINI, 17		
Postal code - City	6500 BELLINZONA - CANTON TICINO	*Country*	SWITZERLAND - CH
Phone	0039 091 8143586	*Fax*	
E-mail			

Name	COTA EJUP		
Structure	PLANT PROTECTION INSTITUTE		
Address	SHKOZET		
Postal code - City	DURRES	*Country*	ALBANA - AL
Phone	+3555222182	*Fax*	+3555222182
E-mail	imb@anep.al.eu.org		

Name	DE CRISTOFARO ANTONIO		
Structure	DIPARTIMENTO DI SCIENZE ANIMALI, VEGETALI E DELL'AMBIENTE		
Address	VIA DE SANCTIS		
Postal code - City	86100 CAMPOBASSO	*Country*	ITALY I
Phone	0874/404686	*Fax*	0874/404685
E-mail	DECRIST@UNIMOL.IT		

Name	DERKACH VOLODIMIR		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			

Name DERRIDJ SYLVIE
Structure INRA-UNITE DE PHYTOPHARMACIE ET DES MEDIATEURS CHIMIQUES

Address CEDEX ROUTE DE SAINT-CYR
Postal code - City 78026 VERSAILLES *Country* FRANCE - F
Phone 33130833164 *Fax* 33130833119
E-mail derridj@versailles.inra.fr

Name DI BERNARDO ANDREA
Structure DEPARTMENT OF AGRONOMY, ENTOMOLOGY UNIVERSITY OF PADUA

Address AGRIPOLI - VIA ROMEA 16
Postal code - City 35020 LEGNARO (PD) *Country* ITALY - I
Phone 349 5093931 *Fax*
E-mail andrea.dibernardo@libero.it

Name DISEGNA LUIGI
Structure AZIENDA REGIONALE VENETO AGRICOLTURA

Address VIA ROMEA - AGRIPOLIS
Postal code - City 35020 LEGNARO (PD) *Country* ITALY - I
Phone 041-8293711 *Fax* 049-8293815
E-mail luigino.disegna@venetoagricoltura.org

Name DOBRINCIC RENATA
Structure UNIVERSITY OF ZAGREB, FACULTY OF AGRICULTURE, DEPT. FOR AGRICUL.ZOOLOGY

Address SVETOSIMUNSKA 25
Postal code - City 10000 ZAGREB *Country* CROATIA - HR
Phone 385-1-2393-805 *Fax* 385-1-2393-970
E-mail rdobrincic@agr.hr

Name DONANTONI LANFRANCO
Structure AGRONOMIST FREE LANCE

Address VIA ROSSINI, 10
Postal code - City 31021 MOGLIANO VENETO (TV) *Country* ITALIA - I
Phone 335261500 *Fax* 0422380699
E-mail ldonant@tin.it

Name EDWARDS C. RICHARD
Structure PURDUE UNIVERSITY

Address 1158 SMITH HALL, W. LAFAYETTE
Postal code - City INDIANA 47907 - 1158 *Country* USA
Phone I-765-494-4562 *Fax* I-765-494-2152
E-mail rich_edwards@entm.purdue.edu

Name EKE JSTUAN
Structure MINISTRY OF AGRICULTURE

Address P.O.6.1
Postal code - City 1860 BUDAPEST *Country* HUNGARY - H
Phone 36-1301-4015 *Fax* 36-1301-4644
E-mail

Name ESTER ALBERT
Structure APPLIEDE RESEARCH FOR ARABLE-FARMING AND FIELD PRODUCTION OF VEGETABLES (PAV)

Address P.O. BOX 430
Postal code - City 8200 AK LELYSTAD *Country* NETHERLANDS - NL
Phone 0031320291633 *Fax* 0031320230479
E-mail A.ESTER@PAV.AGRO.NL

Name FERRARI ROBERTO
Structure CENTRO AGRICOLTURA AMBIENTE ASSOCIAZIONE

Address VIA DI MEZZO LEVANTE 2233
Postal code - City 40014 CREVALCORE BO *Country* ITALIA - I
Phone *Fax*
E-mail rferrari@caa.it

Name FESTIC HUSEIN
Structure FACULTY OF AGRICULTURE SARAJEVO

Address ZMAJA OD BOSNE 8
Postal code - City 71000 SARAJEVO *Country* BOSNA I HERZEGOVINA BiH
Phone 38733653033 *Fax* 38733667429
E-mail RNUMIC@UTIC.NET.BA

Name FRAUSIN CARLO
Structure O.M.P.

Address VIA BEATO ODORICO, 13
Postal code - City 33170 PORDENONE *Country* ITALIA - I
Phone 0434529331 *Fax* 0434520570
E-mail carlo.frausin@regione.fvg.it

Name FUNES VANNA
Structure AGRONOMIST FREE LANCE

Address VIA BERTOLINI, 16
Postal code - City 31100 TREVISO *Country* ITALIA - I
Phone 328 5765157 *Fax*
E-mail funesvanna@libero.it

Name FURLAN LORENZO
Structure DEPARTMENT OF AGRONOMY, ENTOMOLOGY UNIVERSITY OF PADUA

Address AGRIPOLIS - VIA ROMEA, 16
Postal code - City 35020 LEGNARO (PD) *Country* ITALIA - I
Phone 042152721 *Fax* 042153945
E-mail lorenzo.furlan@inwind.it

Name GALLUZZO ROBERT
Structure

Address VIA MORO, 18
Postal code - City 33100 UDINE *Country* ITALY - I
Phone 0432 504027 *Fax* 0432 511996
E-mail robertogalluzzo@virgilio.it

Name GATTI BATTISTA
Structure GROUPE LIMAGRAIN GENETICS RECHERCHE

Address VIA MATTEOTTI, 2/A - LOC.CIGNONE
Postal code - City 26020 CORTE DE' CORTESI-CR *Country* ITALY - I
Phone 0372 925003 *Fax* 0372 925017
E-mail battista.gatti@limagrain.com

Name GERBER COREY
Structure PURDUE UNIVERSITY

Address 1158 SMITH HALL. WEST LAFAYETTE
Postal code - City INDIANA 47907-1158 *Country* USA
Phone (765)494-4912 *Fax* (765)494-2152
E-mail cgerber@entm.purdue.edu

Name GERMINARA GIACINTO SALVATORE
Structure DIPARTIMENTO DI SCIENZE ANIMALI, VEGETALI E DELL'AMBIENTE

Address VIA DE SANCTIS
Postal code - City 86100 CAMPOBASSO *Country* ITALY - I
Phone 0874/404684 *Fax* 0874/404685
E-mail GERMINARA@UNIMOL.IT

Name GERVASINI EUGENIO
Structure S.F.R. LOMBARDIA

Address V.LE RAIMONDI, 56
Postal code - City 22070 VERTEMATE CON MINOPRIO (CO *Country* ITALIA - I
Phone 031320502 *Fax* 031887179
E-mail egervasini@tiscalinet.it

Name	GIROLAMI VINCENZO		
Structure	DEPARTMENT OF AGRONOMY, ENTOMOLOGY UNIVERSITY OF PADUA		
Address	AGRIPOLIS - VIA ROMEA, 16		
Postal code - City	35020 LEGNARO (PD)	*Country*	ITALIA - I
Phone	+39 (0) 498272808	*Fax*	
E-mail	vigiro@agripolis.unipd.it		
Name	GOMBOC STANISLAV		
Structure	BIOTECHNICAL FACULTY, AGRONOMY DEPARTMENT		
Address	JAMNIKARDEVA 101		
Postal code - City	SI-1000 LJUBLANA	*Country*	SLOVENIA - SLO
Phone	++386-142-31-161	*Fax*	++386 1423 1088
E-mail	stanislav.gomboc@bf.uni-lj.si		
Name	GRANDIS FRANCESCA		
Structure	AZIENDA REGIONALE VENETO AGRICOLTURA - SETTORE DIVULGAZIONE TECNICA FORMAZIONE PROFESSIONALE		
Address	VIA ROMA 34		
Postal code - City	35020 LEGNARO (PD)	*Country*	ITALY - I
Phone	+39 (0) 498293943	*Fax*	+39 (0) 498293909
E-mail	francesca.grandis@venetoagricoltura.org		
Name	GRITSYUK VOLODYMYR		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			
Name	HATALA ZSELLER IBOLYA		
Structure	PLANT HEALTH AND SOIL CONSERVATION STATION OF CSONGRAD COUNTY		
Address	P.O.BOX 99		
Postal code - City	6801 HODMEZOVASARHELY	*Country*	HUNGARY- H
Phone	36-62- 246 611	*Fax*	36-62-246 036
E-mail	hzsi-csnta@fki.gov.hu		
Name	HUMMEL E. HANS		
Structure	INSTITUT FUR PHYTOPATHOLOGIE UND ANGE-WANDTE ZOOLOGIE BIOL. UND BIOTECHI-NISCHER PFANZENSCHULTZ JUSTUS-LIEBIG-UNIVERSITAT		
Address	HEINRICH BUFF-RING 26-32		
Postal code - City	35392 GIESSEN	*Country*	GERMANY - D
Phone	+49-641-99-37600	*Fax*	+49-641-99-37609
E-mail	hans.e.hummel.@agrar.uni-giessen.de		

Name	HUREMOVIC ZIJO		
Structure	FACULTY OF AGRICULTURE SARAJEVO		
Address	ZUIAJA OD BOSNE 8		
Postal code - City	71000 SARAJEVO	*Country*	BOSNA I HERZEGOVINA BiH
Phone	38733653033	*Fax*	38733667429
E-mail	RNUMIC@UTIC.NET.BA		
Name	IATSYNINE VENIAMINE		
Structure	KRASNODAR LUKYANENKO RESEARCH INSTITUTE OF AGRICULTURE		
Address			
Postal code - City	350012, KRASNODAR - 12	*Country*	RUSSIA - RU
Phone	(8612)564225	*Fax*	(8612)562274
E-mail	kniish@online.ru		
Name	IGRC BARCIC JASMINKA		
Structure	FACULTY OF AGRICOLTURE DEPT. FOR AGRICUL. ZOOLOGY		
Address	SVETOSIMUNSKA 25		
Postal code - City	10000 ZAGREB	*Country*	CROATIA - HR
Phone	385-1-2393-808	*Fax*	385-1-2393-970
E-mail	rdobrincic@agr.hr		
Name	IMGRABEN HANSJORG		
Structure	REGIERUNGSPRASIDIUM FREIBURG		
Address	PFLANZENSCHUTZDIENST		
Postal code - City	D-79098 FREIBURG	*Country*	GERMANY - D
Phone	00497612081303	*Fax*	00497612081236
E-mail	hansjoerg.imgraben@RPF.BWL.de		
Name	IMREI ZOLTAN		
Structure	PLANT PROTECTION INSTITUTE, HUNGARIAN ACCADEMY OF SCIENCES		
Address	PF 102		
Postal code - City	H-1525 BUDAPEST	*Country*	HUNGARY - H
Phone		*Fax*	
E-mail			
Name	IVANOVA EVTIMOVA IVANKA		
Structure	CENTRAL LABORATORY FOR PLANT QUARANTINE BULGARIA		
Address	GORNOBANSKI BLVD. 120		
Postal code - City	SOFIA 1330	*Country*	BULGARIA - BG
Phone	+359223355	*Fax*	+359223381
E-mail	clkr@bgnet.bg		

Name IVEZIC MARIJA
Structure FACULTY OF AGRICOLTURE - UNIVERSITY OF J.J.STROSSMAYER IN OSIJEK

Address TRG SV.TROJSTVA 3
Postal code - City HR - 31000 OSIJEK *Country* CROATIA - HR
Phone +38531224246 *Fax* +38531207017
E-mail mivezic@suncokret.pfos.hr

Name KARABATSAS KONSTANTINOS
Structure UNIVERSITY OF THESSALY - LABORATORY OF ENTOMOLOGY AND AGRICULTURAL ZOOLOGY

Address FYTOKOU STR. 38 446
Postal code - City NEA IONIA MAGNESIA *Country* GREECE - GR
Phone *Fax*
E-mail kkarabat@agr.uth.gr

Name KARIC NEDZAD
Structure FACULTY OF AGRICULTURE SARAJEVO

Address ZUIASA OB BOSNE 8
Postal code - City 71000 SARAJEVO *Country* BOSNA I HERZEGOVINA - BiH
Phone 0038733653033 *Fax* 0038733667425
E-mail RNUMIC@UTIC.NET.BA

Name KERESI TATJANA
Structure FACULTY OF AGRICULTURE, INST. FOR PLANT PROTECTION AND ENV.

Address TRG D. OBRADOVICA 8
Postal code - City 21000 NOVI SAD *Country* YUGOSLAVIA - YU
Phone +38121450-616 *Fax* +38121450616
E-mail keresi@polj.ns.ac.yu

Name KISS JOZSEF
Structure ST. ISTVAN UNIVERSITY - DEPARTMENT OF PLANT PROTECTION

Address PATER KAROLY U. 1
Postal code - City H-2103 GODOLLO *Country* HUNGARY - H
Phone *Fax*
E-mail jkiss@gikk.gau.hu

Name KOMAROMI JUDIT
Structure DEPT. OF PLANT PROTECTION SZENT ISTVAN UNIVERSITY

Address
Postal code - City 2100 GODOLLO *Country* HUNGARY- H
Phone 36 28 410 200/1771 *Fax* 36 28 522 077
E-mail jkiss@gikk.gau.hu

Name KUHLMANN ULRICH
Structure CABI BIOSCIENCE CENTRE SWITZERLAND

Address RUE DES GRILLONS, 1
Postal code - City CH-2800 DELEMONT *Country* SWITZERLAND - CH
Phone +41-32-421 4882 *Fax* +41-32-421 4871
E-mail U.Kuhlmann@cabi-bioscience.ch

Name LANFRANCHINI FABIO
Structure FEDERAZIONE ITALIANA COLTIVATORI DIRETTI

Address VIA RAVIZZA, 10
Postal code - City 28100 NOVARA *Country* ITALY - I
Phone 0321 674210 *Fax* 0321 674200
E-mail vedi Mazzini Nicoletta

Name LEFEBVRE DU PREY VINCENT
Structure AVENTIS CROPSCIENCE ITALIA

Address PIAZZALE TURR, 5
Postal code - City 21000 MILANO *Country* ITALIA - I
Phone 02/31913325 *Fax* 02/31913299
E-mail vincent.lefebvre-du-prey@aventis.com

Name LIFRIERI LUIGI
Structure ISTITUTO DI ENTOMOLOGIA-FACOLTA' DI AGRARIA - BO

Address VIA A. MURRI, 39
Postal code - City 40139 BOLOGNA *Country* ITALY - I
Phone 051/345925 *Fax* 051/2096105
E-mail LUIGILIFRIERI@YAHOO.IT

Name MAINI STEFANO
Structure DISTA UNIVERSITA' DI BOLOGNA

Address VIA FILIPPO RE, 6
Postal code - City 40100 BOLOGNA *Country* ITALY - I
Phone 051/2091558 *Fax*
E-mail Smaini@agrsci.unibo.it

Name MAJDANCIC MEHO
Structure FACULTY OF AGRICULTURE SARAJEVO

Address ZUIAJA OD BOSNE 8
Postal code - City 71000 SARAJEVO *Country* BOSNA I HERZEGOVINA BiH
Phone 38733653033 *Fax* 38733667429
E-mail RNUMIC@UTIC.NET.BA

Name	MALVAR ROSA ANA		
Structure	MISION BIOLOGICA DE GALICIA (CSIC)		
Address	APTDO 28		
Postal code - City	86080 PONTEVEDRA	*Country*	SPAIN - SP
Phone	34986854800	*Fax*	34986841362
E-mail	csgpormp@cesga.es		

Name	MAZZINI NICOLETTA		
Structure	FEDERAZIONE ITALIANA COLTIVATORI DIRETTI		
Address	VIA RAVIZZA, 10		
Postal code - City	28100 NOVARA	*Country*	ITALY - I
Phone	0321 674210	*Fax*	0321 674200
E-mail	cold.novara@iol.it		

Name	MELNIK PAVEL		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address			
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone		*Fax*	
E-mail			

Name	MICHELATTI GIACOMO		
Structure	SERVIZIO FITOSANITARIO REGIONE PIEMONTE		
Address	C.SO GROSSETO 71/6		
Postal code - City	10147 TORINO	*Country*	ITALY - I
Phone	339/1275979	*Fax*	011/4323710
E-mail	Entomologia@regione.piemonte.it		

Name	MILEVOJ LEA		
Structure	BIOTECHNICAL FACULTY, AGRONOMY DEPARTMENT		
Address	JAMNIKAOJEVA 101		
Postal code - City	SVN-1000 LJUBLJANA	*Country*	SLOVENIA - SLO
Phone	00386 14321 161	*Fax*	00386 14231 088
E-mail	lea.milevoj@bf.uni.lj.si		

Name	MINELLO BRUNO		
Structure	SERVIZIO FITOSANITARIO REGIONE VENETO		
Address	VIALE DELL'AGRICOLTURA 1/A		
Postal code - City	37060 - BUTTAPIETRA (VR)	*Country*	ITALY - I
Phone	045-8676511	*Fax*	945-8676937
E-mail	fitosanitario@regione.veneto.it		

Name	MOESER JOACHIM		
Structure	INST. PLANT PATHOLOGY AND PLANT PROTECTION - GEORG-AUGUST-UNIVERSITY GOETTINGEN		
Address	GRISEBACHSTR 6		
Postal code - City	D-37077 GOETTINGEN	*Country*	GERMANY - D
Phone	+49551393730	*Fax*	+495513912105
E-mail	jmoeser@gwdg.de		

Name	MOLNAR ZANOS		
Structure			
Address	VERHALOM 12-16		
Postal code - City	1023 BUDAPEST	*Country*	HUNGARY - H
Phone	+36 13450677	*Fax*	+36 13260407
E-mail	zanos.molnar@ea.monsanto.com		

Name	MOLNAR ZOLTAN		
Structure	I.C.P.C. BRASOV		
Address	STR. PRINCIPALA NR.1312		
Postal code - City	4215 LAZAREA JUD. HARGHITA	*Country*	ROMANIA - RO
Phone		*Fax*	
E-mail	Mozika33@yahoo.com		

Name	MOVCHAN OLEKSIY		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			

Name	MUCSI KRISZTINA		
Structure	CSONGRAD COUNTY PLANT HEALTH AND SOIL PROTECTION SERVICE		
Address	P.O. BOX 99		
Postal code - City	H-6800 HODMEZOVASARHELY	*Country*	HUNGARY - H
Phone	+36-62 246-611	*Fax*	+36-62 246-032
E-mail	KMUCSI@FREEMAIL.HU		

Name	MULLER J FRANCK		
Structure	INRA-UNITE' DE PHYTOPHARMACIE ET MEDIATEURS CHIMIQUES-PLANT INSECT LAB.		
Address	ROUTE DE ST CYR		
Postal code - City	78 026 VERSAILLES	*Country*	FRENCH - F
Phone		*Fax*	
E-mail	frankmuller@gmx.net - muller@versailles.inra.fr		

Name MURESAN FELICIA
Structure AGRICULTURAL RESEARCH STATION OF TURDA

Address STR. AGRICULTURII,27 JUD.CLUY
Postal code - City 3350 TURDA *Country* ROMANIA - RO
Phone *Fax*
E-mail vedi Zoltan Molnar

Name MYKYTYK NADEJDA
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name NORIDO FRANCO
Structure AZIENDA REGIONALE VENETO AGRICOLTURA - SETTORE DIVULGAZIONE TECNICA FORMAZIONE PROFESSIONALE

Address VIA ROMA 34
Postal code - City 35020 LEGNARO (PD) *Country* ITALY - I
Phone +39 (0) 498293919 *Fax* +39 (0) 498293909
E-mail franco.norido@venetoagricoltura.org

Name OLEINIK STANISLAV
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name OMELYUTA VICTOR P.
Structure INSTITUTE OF PLANT PROTECTION OF UAAS HEAD OF DEPARTMENT QUARANTINED PEST

Address VASYLKOYSKAJA 33
Postal code - City UA 03002 KIEV 22 *Country* UKRAINA
Phone *Fax*
E-mail nikol@mail.kar.net

Name ONDRUSZ ILONA
Structure STATE STUD-ESTATE CORPORATION

Address
Postal code - City 5820 MEZOHEGYES *Country* HUNGARY - H
Phone 30 263 8887 *Fax*
E-mail turcasan.istvan@menesbirtok.hu

Name PAJMON ALES
Structure AGRICOLTURAL INSTITUTE OF SLOVENIA

Address HACQUETOVA 17
Postal code - City 1000 LUBIANA *Country* SLOVENIA - SLO
Phone +386 1 2805204 *Fax* +386 1 2805255
E-mail ALES.PAJMON@KIS-H2.si

Name PALAGESIU JOAN
Structure UNIVERSITY OF AGRICULTURE AND VETERINARY MEDICINE - BANATULUI, DEPARTMENT OF ENTOMOLOGY

Address CALEA ARADULUI 119
Postal code - City 1900 TIMISOARA *Country* ROMANIA - RO
Phone *Fax* 00 40 056 200296
E-mail

Name PALEKAR N.V. SHANK
Structure SYNGENTA BIOTECHNOLOGY INC.

Address 3054 CORNWALLIS ROAD, P.O. BOX 12257 - RESEARCH TRIANGLE PARK
Postal code - City NC 27709 - 2257 *Country* USA
Phone 0919 541 8642 *Fax* 0919 541 8585
E-mail shank.palekar@syngenta.com

Name PARKER WILLIAM E.
Structure ADAS WOODTHORNE

Address
Postal code - City WOLVERHAMPTON WV6 8TQ *Country* UNITED KINGDOM - UK
Phone +441902693271 *Fax* +441902693166
E-mail Bill.Parker@adas.co.uk

Name PEREIRA MARIA MARGARIDA
Structure ESCOLA SUPERIOR AGRARIA DE BEJA RUA PEDRO SOARES

Address APARTADO 158
Postal code - City 7801-901 BEJA *Country* PORTUGAL - PT
Phone 284314300 *Fax* 284388207
E-mail mmpereira@mail.pt

Name PEREZ FARINOS GEMA
Structure CSIC, CIB,

Address VELAZQUEZ, 144
Postal code - City 28006 MADRID *Country* SPAIN - SP
Phone 34-91-5611800 *Fax* 34-91-5627518
E-mail gpfarinos@cib.csic.es

Name	PERSHING JAY		
Structure	MONSANTO		
Address	700 CHESTERFIELD PLLCWY NORTK BB1J,		
Postal code - City	ST. LOUIS, NO43198	*Country*	USA
Phone	636-737-6548	*Fax*	636-737-5127
E-mail	jay.c.pershing@monsanto.com		
Name	PETRIS GIOVANNI		
Structure	OSSERVATORIO MALATTIE DELLE PIANTE		
Address	VIA CACCIA 17		
Postal code - City	33100 - UDINE	*Country*	ITALY - I
Phone	0432-555166	*Fax*	0432555163
E-mail	omp.ud@regione.fvg.it		
Name	PIRANI CLEMENTE		
Structure	REGIONE DEL VENETO - GIUNTA REGIONALE		
Address	VIA UDINE, 35		
Postal code - City	30027 SAN DONA' DI PIAVE-VE	*Country*	ITALY - I
Phone	0421 52361	*Fax*	
E-mail	clemente.pirani@regione.veneto.it		
Name	POPOV CONSTANTIN		
Structure	RESEARCH INSTITUTE FOR CEREALS AND INDUSTRIAL CROPS.		
Address			
Postal code - City	FUNDULEA	*Country*	ROMANIA - RO
Phone	+401-311.07.22	*Fax*	+401-311.07.22
E-mail	fundelea@cons.incerc.ro		
Name	PRINCZINGER GABOR		
Structure	MINISTRY OF AGRICULTURE AND REGIONAL DEVELOPMENT		
Address	55. P.O.BOX 1		
Postal code - City	H -1860 BUDAPEST	*Country*	HUNGARY - H
Phone	/36/1/301-4501	*Fax*	/36/1/301-4644
E-mail	prigabor@westel900.net		
Name	RASPUDIC EMILIJA		
Structure	FACULTY OF AGRICOLTURE, UNIVERSITY OF J.J. STROSSMAYER IN OSIJEK		
Address	TRG SU.TROJSTVA 3		
Postal code - City	31000 OSIJEK	*Country*	CROATIA - HR
Phone	385 31 224 280	*Fax*	385 31 207 017
E-mail	remilija@suncokret.pfos.hr		

Name RIPKA GEZA
Structure CENTRAL SERVICE FOR PLANT PROTECTION AND SOIL CONSERVATION OF BUDAPEST
Address P.O. BOX 340
Postal code - City H1519 BUDAPEST, PF340 *Country* HUNGARY - H
Phone 36-1-309-1032 *Fax* 36-1-246-2942
E-mail novved@elender.hu

Name ROSCA IOAN
Structure UNIVERSITY OF AGRICOLTURAL SCIENCES AND VETERINARY MEDICINE
Address BDL. MARASTI 59 - SECTOR 1
Postal code - City BUCHAREST *Country* ROMANIA - RO
Phone 40 1 2242576 *Fax* 00 401 2242266 (FAX UNIV) 00
E-mail rosca@cons.incerc.ro

Name ROY ANNE SOPHIE
Structure EPPO
Address 1 RUE LE NOTRE
Postal code - City 75016 PARIS *Country* FRANCE - F
Phone 33 1 45 207794 *Fax* 33 1 42 248543
E-mail hq@eppo.fr

Name SADLYK ALLA
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE
Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name SANTAMARIA GIORGIO
Structure AGRONOMIST FREE LANCE
Address VIA TRENTO, 14
Postal code - City MESTRE - VENEZIA *Country* ITALY - I
Phone 041 929453 *Fax*
E-mail GISHWS@TIN.IT

Name SIVCEV IVAN
Structure INSTITUTE FOR PLANT PROTECTION AND ENVIRONMENT
Address BANATSKA 33 YU
Postal code - City 11080 ZEMUN *Country* YUGOSLAVIA - YU
Phone 38111611762 *Fax* 38111669860
E-mail ibsivcev@eunet.yu

Name	SIVICEK PETER		
Structure	THE CENTRAL CONTROL AND TESTING INSTITUTE OF AGRICULTURE		
Address	HANULOVA 9/A		
Postal code - City	84429 BZATISLAVA 42	*Country*	SLOVAK REPUBLIC - SK
Phone	00421764462085/087	*Fax*	0421764462084
E-mail	uksup@internet.sk		
Name	SOROKA VASYL		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			
Name	SZABO' ZOLTAN		
Structure	STATE STUD-ESTATE CORPORATION		
Address			
Postal code - City	5820 MEZOHEGYES	*Country*	HUNGARY - H
Phone	68 466 015	*Fax*	
E-mail	turcasan.istvan@menesbirtok.hu		
Name	TINLAND BRUNO		
Structure	MONSANTO		
Address	AV. TERVUREN 270-272		
Postal code - City	1150/BRUSSELS	*Country*	BELGIO - BE
Phone	0032/2-7764922	*Fax*	0032/2-7764676
E-mail	BrunoTinland@Monsanto.com		
Name	TOEPFER STEFAN		
Structure	CABI BIOSCIENCE CENTRE SWITZERLAND		
Address	RUE DES GRILLONS, 1		
Postal code - City	CH-2800 DELEMONT	*Country*	SWITZERLAND - CH
Phone	0041/32/4214885	*Fax*	0041/32/4214871
E-mail	stoepfer@gmx.ch		
Name	TOLLEFSON JON J.		
Structure	DEPARTMENT OF ENTOMOLOGY IOWA STATE UNIVERSITY		
Address	17 INSECTARY BLDG.		
Postal code - City	AMES, IA 50011-3140	*Country*	USA
Phone	(515) 294-8044	*Fax*	(515) 294-4431
E-mail	tolly@iastate.edu		

Name TOTH BERTALAN
Structure BIOMARK LTD

Address NAGYSANDOR G. U. 10
Postal code - City 1046 BUDAPEST *Country* HUNGARY - H
Phone 0036 13699455 *Fax* 0036 13794629
E-mail tothb.biomark@euroweb.hu

Name TOTH FERENC
Structure DEPT. OF PLANT PROTECTION SZENT ISTVAN UNIVERSITY

Address PATER KAROLY U. 1
Postal code - City 2103 GODOLLO *Country* HUNGARY - H
Phone 36 28 410 200/1771 *Fax* 36 28 522 077
E-mail tothf@fau.gau.hu

Name TOTH MIKLOS
Structure PLANT. PROT. INST. HUNGARIAN ACCADEMY OF SCIENCES

Address PF 102
Postal code - City H-1525 BUDAPEST *Country* HUNGARY - H
Phone +36-1-3769555 *Fax* +36-1-3769729
E-mail H2371TOT@ELLA.HU

Name TSITSIPIS JOHN A.
Structure RESEARCH COMMITTEE, UNIVERSITY OF THESSALY, DEPARTMENT OF CROP AND ANIMAL PRODUCTION UNIVERSITY OF THESSALY

Address FYTOKOU STR.
Postal code - City 38 446 NEA IONIA MAGNESIA *Country* GREECE - GR
Phone +3042193190 *Fax* +3042193286
E-mail tsitsipi@uth.gr

Name TUSKA TAMAS
Structure DEPARTMENT OF PLANT PROTECTION ST. ISTVAN UNIVERSITY,

Address PATER KAROLY U. 1
Postal code - City H 2103 GODOLLO *Country* HUNGARY - H
Phone *Fax*
E-mail jkiss@gikk.gau.hu

Name VETTORAZZO MARCO
Structure SERVIZIO FITOSANITARIO REGIONE VENETO

Address VIA POERIO 34
Postal code - City 30173 VENEZIA-MESTRE *Country* ITALY - I
Phone 041-2795701 *Fax* 041-2795703
E-mail serv.fitove@regione.veneto.it

Name	VICINANZA I. SONIA		
Structure	AMERICAN EMBASSY		
Address	APHIS BOLTZMANNGASSE 16		
Postal code - City	A-1091 WIEN	*Country*	AUSTRIA - A
Phone	+43 1 31339 2497	*Fax*	+43 1 31339 2913
E-mail	SONIA.VICINANZA@APHIS.USDA.GOV		
Name	VIDAL STEFAN		
Structure	INST. PLANT PATHOLOGY-UNIVERSITY GOETTINGEN -		
Address	GRISEBALHSTR 6		
Postal code - City	D-37077 GOETTINGEN	*Country*	GERMANY - D
Phone	+49551399744	*Fax*	+495513912105
E-mail	SVIDAL@GWDG.DE		
Name	WENNEMANN LUDGER		
Structure	INSTITUTE OF PHYTOPHATOLOGY AND APPLIED ZOOLOGY BIOLOGICAL AND BIOTECHNICAL PLAN PROTECTION		
Address	HEINRICH BUFF-RING 26-32		
Postal code - City	D-35392 GIESSEN	*Country*	GERMANY - D
Phone	+49(641)99-3 7606	*Fax*	+49 (641)99-3 7609
E-mail	ludger.wennemann@agrar.uni-giessen.de		
Name	WILDE GERALD		
Structure	DEPT. OF ENTOMOLOGY KANSAS STATE UNIVERSITY		
Address			
Postal code - City	MANHATTAN KS 66506	*Country*	USA
Phone	785-532-4729	*Fax*	785-532-6232
E-mail	Gwilde@OZNet.KSU.Edu		
Name	WILHELM RONALD		
Structure	AVENTIS CROPSCIENCE		
Address	INDUSTRIEPARK HOCHST, H 872		
Postal code - City	65926 FRANKFURT AM MAIN	*Country*	GERMANY - D
Phone	49 69 305 80160	*Fax*	49 69 305 7342
E-mail	Ronald.Wilhelm@aventis.com		
Name	YAKOBCHYUK VASYL		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			

Name YAKOVETS PETRO
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name YRYNYAK ALLA
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name YUSHCHUK OSTAP
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone *Fax*
E-mail

Name YUSHCHUK TARAS
Structure GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE

Address STR. KOLOSKOVAYA, 7
Postal code - City UA - 03138 KIEVE *Country* UKRAINA - UA
Phone 0038044 2647707 *Fax* 0038044 2648902
E-mail

Name ZANIRATO MARCO
Structure PIONEER HI-BRED ITALIA S.R.L.

Address VIA GIUSEPPINA, 39
Postal code - City 26030 MALAGNINO - CR *Country* ITALY - I
Phone 0372 441511 *Fax* 0372 441544
E-mail marco.zanirato@pioneer.com

Name ZHANG FENG
Structure AGRICULTURAL PEST RESEARCH CABI BIOSCIENCE CENTRE SWITZERLAND

Address RUE DES GRILLONS
Postal code - City CH-2800 DELEMONT *Country* SWITZERLAND - CH
Phone 41-32-421 4883 *Fax* 41-32-421 4871
E-mail F.ZHANG@CABI-BIOSCIENCE.CH

Name	ZHELEZNYAK VITALIY		
Structure	GENERAL STATE INSPECTION ON QUARANTINE OF PLANT IN THE UKRAINE		
Address	STR. KOLOSKOVAYA, 7		
Postal code - City	UA - 03138 KIEVE	*Country*	UKRAINA - UA
Phone	0038044 2647707	*Fax*	0038044 2648902
E-mail			


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CiP
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