LECTURES OF WEED SCIENCES & INTEGRATED PEST MANAGEMENT (IPM) SESSIONS
EFFECT OF PHENOLOGY AND RAINFALL ON ALLELOPATHY OF XANTHIUM ITALICUM MOR.

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The cocklebur species become more and more significant weed species in Hungary, where the row crops (such as: corn, sunflower, sugar beet, etc) are endangered by them very badly. Their fast spreading and danger are caused by many reasons: lasted sprouting, reduced sensitivity against many kinds of herbicides, competitive ability, allelopathy, fast initial growth and changes in climate.

Allelopathy is used by many species of plants in the competition with one another, as well as by the cockleburs. But the way and efficiency of this are influenced by many factors. Species with allelopathical effect could have different attitudes under different conditions: they can produce materials with retarding and stimulating effects in different quantities or composition, and the actual conditions of existences (water supply, nutritive supply, quantity and quality of light, proportion of minerals etc.) of the acceptor plants and the mediator agent (i.e. soil) influence the final effect.

Literature

Cocklebur species are spreading, noxious weeds, and get in the centre of interest in several parts of world. However, there are experiments for therapeutic utilization in some country, e.g. against Plasmodium falciparum, Trypanosoma evansi (Joshi et al. 1997; Talakal et al. 1995). Ground cover of cockleburs, especially Xanthium strumarium and X. italicum, became larger and larger in the past decades in Hungary, like warm-philous species (Szőke, 2001). They threaten mainly row crops (sugarbeet, maize, sunflower, soybean) and cause reduce of yield (Bloomberg et al. 1982; Wilson, 1995).

Danger of cockleburs may be explained with continuous emergence, large capability for competition, allelopathy and spreading several pathogens, e.g. they are hosts of beet necrotic yellow vein virus (Dikova, 1997; Kutluk et al. 2000), in addition, plants are toxic at cotyledon stage when seeds contain hydroquinone (Mitch, 1987). Allelopathy of X. strumarium was examined on several cultivated plants: lettuce, maize, soybean (Inam et al. 1987), and extracts were active against Proteus vulgaris, Staphylococcus aureus, Bacillus subtilis, Candida

The allelopathy of plants was caused by several compounds, among others free amino acids, phenols (Elmore, 1980a; Colton and Einhellig, 1980; Elmore, 1980b), which may have several other functions. Their role was studied exposed to stress factors where their level increased in tissues of several species preventing decay of certain plants. (Sircelj et al., 1999; Sanchez et al. 1998; Gilbert et al., 1994; Ashraf et al., 1994; Maggio et al., 2002; Politycka, 1999). Consequently, stress factors can effect on allelopathy, and modify interactions between plants. These allelochemicals can be produced in different ratios and quantities under different conditions (e.g. drought stress, different nutrient supply) modifying the interference between weeds and crops (Dávid and Radócz, 2002).

Materials and Methods

Xanthium italicum and Abutilon theophrasti Medic., from which extracts were made, were grown in the field with different population densities (5 plants per m² and 20 plants per m²). Samples were collected in May at four or five leaves stage and in July (before flowering) before and after rainfall. There were long drought periods before 20 and 15 mm rain. Fresh roots and shoots were cut into small pieces, 4g of fresh biomass was put into 100 ml tap water and left for a day, then extracts were filtered through filter paper. Test plants were cress (Lepidium sativum L.) and sugarbeet (Beta vulgaris var. saccharifera). Experiments were conducted in Petri plates on filter paper with four replications at room temperature used 6ml leachate and 50 seeds of test plant in each dish. Root and shoot growth of cress were measured on the 3rd and 6th day, and germination of sugarbeet was valued on the 6th and 10th day.

Results

Experiments were conducted in May and in July, in both cases samples collected before and after rainfall. There was difference between the effect of root and shoot extracts depending on the phenological stage and rainfall.

Effect on growth of cress in relation to rainfall
Leachates of shoots of cockleburs and velvetleafs collected before rainfall in May reduced root length. Leachates made from roots had no inhibitory effect, in fact, samples after rainfall slightly promoted the growth. (Samples
collected before rain had no significant effect compared to check.) The difference in shoot growth was smaller (Figure 1).

Figure 1. Effect of velvetleaf’s and cocklebur’s leachates on root and shoot growth of cress before and after rainfall in May; A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.

In July there was a larger difference between the effects of extracts made from shoots collected before and after rainfall than in May. Leachates before rainfall strongly retarded root growth, however, after it leachates had no inhibitory effect, in fact, leachate of shoots of low population density stimulated the growth. The effect was similar but slighter on shoot growth. Extracts made from roots had no significant effect on growth of cress except treatment with roots of high population density, which promoted root growth (Figure 2).

Effect on germination of sugarbeet in relation to rainfall
Germination of sugarbeet was examined by treatments with the same leachates. In May, every extract depressed the germination on the 6th day, but extracts made from plants collected after rainfall exerted stronger effect than those made from plants collected before rainfall. The difference was large in the case of cocklebur’s extracts, and small in the case of velvetleaf’s extracts (Figure 3). By the 10th day the difference became smaller for treatments with shoot extracts, but a significant difference was observed in the case of root extracts made before and after rainfall.

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Figure 2. Effect of cocklebur’s leachates on root and shoot growth of cress before and after rainfall in July; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain H = high population density.

Figure 3. Effect of velvetleaf’s and cocklebur’s leachates on germination of sugarbeet on the 6th day before and after rainfall in May; A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.
In July the inhibition of germination was mostly weaker, and the difference was smaller before and after rainfall than in May. The effect of rainfall was significant only in the case of cockleburs’ root extracts of high density population (Figure 4).

Figure 4. Effect of velvetleaf’s and cocklebur’s leachates on germination of sugarbeet on the 6th day before and after rainfall in July; A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain, H = high population density.

Difference caused by phenology
Radicle length of cress was strongly retarded by shoot extract made before rain in July, but the effect was smaller in May. In the case of shoot extracts after rain, no effect was observed in May, and a promotive effect in July. Rainfall influenced effects of root extract differently in May and July (Figure 5).
Figure 5. Effect of cocklebur’s leachates on root growth of cress in May and July; S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.

Trend was similar in the case of shoot growth, but only slight difference was observed between results in May and July.
On the 6th day, germination of sugarbeet was retarded more strongly in May than in July by shoot extracts, and the difference was especially large after rainfall. Root extracts before rain had a similar effect, but after rainfall the germination was significantly lower in May than in July (Figure 6). By 10th day the difference did not decrease in the case of root extracts.

Discussion

The results supported the hypothesis that some environmental factors and the phenological stage of plants play a determining role in interference between individual plants. Results can be altered depending on only the date of sample collection. When interpreting and comparing results of allelopathical experiments, it is recommended to pay attention to environmental conditions, phenological stage, setting of experiment etc.
Figure 6. Effect of cocklebur’s leachates on germination of sugarbeet on the 6th day before and after rainfall in May and July
S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain

References


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The cocklebur species become more and more significant weed species in Hungary, where the row crops (such as: corn, sunflower, sugar beet, etc) are endangered by them very badly. Their fast spreading and danger are caused by many reasons: lasted sprouting, reduced sensitivity against many kinds of herbicides, vigorous competitive ability, allelopathy, fast initial growth and changes in climate. Allelopathy is used by many species of plants in the competition with one other, as well as by the cockleburs. But the efficiency of this is influenced by many factors. Species with allelopathical effect could have different attitudes under different conditions: they can produce materials with retarding and stimulating effects in different quantities or composition, and the actual conditions of existences, of acceptor plants and the mediator agent (i.e. soil) influence the final effect. In this experiments influence of phenology and rainfall were studied on allelopathy of cockleburs in May and July. In July a stronger inhibitor effect was observed on growth of cress before rain than in May, but it disappeared after rain. Inhibition of sugarbeet’s germination was stronger in May than in July, and the difference became larger after rain.
In the past years weed flora has changed significantly. Some species have spreaded while other ones have disappeared. In the course of permanent usage of herbicides weed population of cultivated fields has changed disadvantageously: tolerant and resistant weed breeds have appeared and spreaded (Reisinger 2001). Each crop has own special weed flora, which can be influenced by several factors, for example nutrients (Lehoczky 1995).

Lots of weeds can be found in potato crop. From the point of view of importance Chenopodium album L. is on the first place (Maykuss 1993). This is one of our most spreaded weed breeds, which can be found not only in the fields, but in every crop culture (Ujvárosi 1973). Based on the data of IV. National Weed Survey C. album is on the 4th place (Tóth-Spilák 1998).

Echinochloa crus-galli (L.) P.B. and Amaranthus spp. have the same signification. The 900 species of the Amaranthaceae family are spreaded all over the world, except the cold zone (Ujvárosi 1973). Amaranthus retroflexus L. is on the 3rd place while Amaranthus chlorostachys Will.d. has the 9th place on the weed dominance order in Hungary (Tóth-Spilák 1998). E. crus-galli is the dangerous weed of several crops. It is a cosmopolitan species (Lehoczky 2002). Based on the results of the IV. National Weed Survey E. crus-galli on the 2nd place (Tóth-Spilák 1993). Similar to potato Solanum nigrum L. is belonging to the Solanaceae family so its weed control is different (Hoffmanné 2002).

Potato needs 4 kg N, 2 kg P₂O₅ and 8 kg K₂O for 1 t potato tubers (Horváth 1997). For 30 t tubers potato needs 150 kg N, 60 kg P₂O₅, 270 kg K₂O, 90 kg CaO and 30 kg MgO (Antal 1996).

At the early stage of development potato cannot compete with the weeds, thus proper control has a special importance. The quantity and forms of the damages caused by weeds can be a lot of kind. At the early stage of development weeds can use the water, light and space but the nutrients (Lehoczky 2002, Lehoczky and Reisinger 2002). Successful weed control has several factors for example the knowing of weed species and selection of effective herbicides (Béres 2000, Reisinger 2000). Competition between
weeds and crop is for the essential factors, for example water, light and nutrients. The significant nutrient usage of the weeds can cause disadvantageous effect on the crops (Lehoczky 1988, 1994, Kazinczi 1993, 1998).

On the effect of competition the quantity and quality of the crop increase that is caused by the lack of the water and nutrient used by weeds. Our aim was the study of the nutrient concentration of the weeds in potato with the examination of the weight of the biomass and nutrient uptake of the plants.

Materials and methods

Our examination on competition was made in a field trial. The experiment was set up on the plots of Veszprém University, Georgikon Faculty of Agriculture, Regional Potato Research Centre. A randomised blocks design with four replicates and 27.6 m² plots was used. Twenty-eight potato tubers cv. „White Lady” were planted in a row. In autumn before planting and at the time of planting, fertilizers were applied. In autumn 200 kg N ha⁻¹ was applied. In spring 80 kg N ha⁻¹, 16 kg P ha⁻¹ and 24 kg K ha⁻¹ were applied at the time of planting. The quantities of the applied fertilizers and the time of application are the part of the crop technology of the Regional Potato Research Centre. On the untreated control plots there was not weed control from the planting of potato (23 April 2002) until harvest. On these plots we could examined the nutrient uptake of potato and weeds. Potato and weed samples were collected on 18 June, after flowering of potato. Weeds were collected as species from 1 m² sampling area of the plots. Fresh and dry weight of the weeds and potato (shoots and tubers) were measured. N, P, K and Ca concentration of the samples was analyzed. Mathematical-statistical analyzes of the experimental data was made by SPSS software.

Results

Twelve weed species were found on the weedy control plots (Table 1). From among these weeds S. nigrum, A. chlorostachys, A. theophrasti, C. album and A. artemisiifolia were the most important species. Weeds with T₄ life cycle form were dominated. There were two perennial weeds on the experimental plots: C. arvensis and L. tuberosus.
Table 1. Weed species occurring on the untreated (weedy) plots

<table>
<thead>
<tr>
<th>Weed species</th>
<th>Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abutilon theophrasti MEDIC.</td>
<td>ABUTH</td>
</tr>
<tr>
<td>Amaranthus chlorostachys WILLD.</td>
<td>AMACH</td>
</tr>
<tr>
<td>Ambrosia artemisiifolia L.</td>
<td>AMBAR</td>
</tr>
<tr>
<td>Chenopodium album L.</td>
<td>CHEAL</td>
</tr>
<tr>
<td>Convolvulus arvensis L.</td>
<td>CONAR</td>
</tr>
<tr>
<td>Echinochloa crus-galli (L.) P.B.</td>
<td>ECHCG</td>
</tr>
<tr>
<td>Lathyrus tuberosus L.</td>
<td>LATTU</td>
</tr>
<tr>
<td>Matricaria inodora L.</td>
<td>MATIN</td>
</tr>
<tr>
<td>Polygonum aviculare L.</td>
<td>POLAV</td>
</tr>
<tr>
<td>Polygonum lapathifolium L.</td>
<td>POLLA</td>
</tr>
<tr>
<td>Setaria glauca (L.) P.B.</td>
<td>SETGL</td>
</tr>
<tr>
<td>Solanum nigrum L.</td>
<td>SOLNI</td>
</tr>
</tbody>
</table>

Fresh weed shoot weight was higher than fresh potato shoot weight (Table 2,3). Comparing the fresh potato weight to the fresh weed weight we have found that the weight of the weeds is 30% of the potato weight. This fact shows that weeds were able to spread in high mass. Comparing the dry weed weight to the dry potato weight we have found that there is a difference between the results of fresh and dry weight. Dry matter of the weeds was more by 9% than the dry weight of potato (Table 2,3).

Table 2. Fresh and dry weight of potato on the untreated (weedy) plots (g m$^{-2}$)

<table>
<thead>
<tr>
<th>Potato</th>
<th>Fresh weight</th>
<th>Dry weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tuber</td>
<td>1650</td>
<td>170</td>
</tr>
<tr>
<td>Shoot</td>
<td>295</td>
<td>52</td>
</tr>
<tr>
<td>Total</td>
<td>1945</td>
<td>222</td>
</tr>
</tbody>
</table>
### Table 3. Fresh and dry weight of the weed shoots (g m\(^{-2}\))

<table>
<thead>
<tr>
<th>Weed species</th>
<th>Fresh weight</th>
<th>Dry weight</th>
<th>Water %</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Solanum nigrum</em> L.</td>
<td>283,8</td>
<td>50,9</td>
<td>82,1</td>
</tr>
<tr>
<td><em>Amaranthus chlorostachys</em> WILLD.</td>
<td>158,8</td>
<td>38,7</td>
<td>75,6</td>
</tr>
<tr>
<td><em>Abutilon theophrasti</em> MEDIC.</td>
<td>102,9</td>
<td>35,8</td>
<td>65,1</td>
</tr>
<tr>
<td><em>Chenopodium album</em> L.</td>
<td>114,6</td>
<td>35,0</td>
<td>69,4</td>
</tr>
<tr>
<td><em>Ambrosia artemisiifolia</em> L.</td>
<td>126,6</td>
<td>30,5</td>
<td>75,9</td>
</tr>
<tr>
<td><em>Polygonum lapathifolium</em> L.</td>
<td>87,9</td>
<td>23,1</td>
<td>73,6</td>
</tr>
<tr>
<td><em>Convolvulus arvensis</em> L.</td>
<td>40,0</td>
<td>9,7</td>
<td>75,6</td>
</tr>
<tr>
<td><em>Echinochloa crus-galli</em> (L.) P.B.</td>
<td>29,8</td>
<td>6,3</td>
<td>78,6</td>
</tr>
<tr>
<td><em>Polygonum aviculare</em> L.</td>
<td>18,4</td>
<td>5,8</td>
<td>68,1</td>
</tr>
<tr>
<td><em>Matricaria inodora</em> L.</td>
<td>11,4</td>
<td>3,2</td>
<td>71,5</td>
</tr>
<tr>
<td><em>Lathyrus tuberosus</em> L.</td>
<td>8,3</td>
<td>1,5</td>
<td>81,3</td>
</tr>
<tr>
<td><em>Setaria glauca</em> (L.) P.B.</td>
<td>8,2</td>
<td>1,3</td>
<td>83,5</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>991,3</strong></td>
<td><strong>242,5</strong></td>
<td>-</td>
</tr>
</tbody>
</table>

Water content of potato tubers and shoot altogether was 88.6% while water content of weeds was 75.6%. In the point of view of competition it is important to know the water content of the weeds. *S. glauca*, *S. nigrum* and *E. crus-galli* content a lot of water while *A. theophrasti*, *P. aviculare* and *C. album* had less water. Nutrient concentration and uptake by the plants were measured. N concentration of the weed shoots was 1.2-2.8% (Fig. 1).
This nutrient could be found in the highest amount in the weeds. In the shoots of *L. tuberosus* N concentration was especially high, but *A. artemisifolia*, *S. nigrum*, *C. arvensis* and *C. album* have a lot of N, too. In the half of the examined weed species N concentration was 1.5-2.0%. N content of potato shoots was 1.91% that is similar to the weeds. P concentration of the weeds was 0.06-0.16% (Fig. 2).
From among the measured nutrients P could be found in the lowest amount in the plants. P concentration of potato tubers and shoots was higher than in the weeds, 0.3%. Similar to the nitrogen, K concentration could be found in high concentration in the plants. The measured K concentration was 0.54-2.43% (Fig. 3).

Figure 3. Potassium concentration of the weed shoots, in the % of dry matter

From among the weeds in *S. glauca*, *C. album*, *E. crus-galli* and *S. nigrum* K concentration was very high. In potato shoots K concentration was 3.01% while tubers content 2.10% K. It is known that potato takes up K in the highest amount. Ca concentration of the weeds was between 0.34-1.13% (Fig. 4).

Figure 4. Calcium concentration of the weed shoots, in the % of dry matter
In the shoots of *S. glauca, C. album, S. nigrum* and *A. artemisiifolia* Ca concentration was high. Ca concentration in potato shoots was 3.03% while tubers have 0.97% Ca.

The amounts of nutrients taken up by plants until the examination were counted. Competition for N was very strong weeds have taken up more N by 30% than potato plants. From among weed species *S. nigrum* has taken up the most N, but *A. artemisiifolia* and *A. chlorostachys* have taken up a lot of N, too. These three weed species are „nitrophyl” species.

The amount of P taken up by plants was less than N. On the weedy plots potato has taken up the 57% of P, while the amount of P taken up by weeds was 43%.

Potato plants have taken up K in the highest amount from among the examined nutrients. The effect of competition can be seen in the K uptake of potato. Plants have taken up 7.4 g K altogether from 1 m². Potato has taken up 58% K and weeds have taken up 42% K (Table 4).

Table 4. The amounts of nutrient elements taken up by weeds and potato on the weedy plots (mg m⁻²)

<table>
<thead>
<tr>
<th>Weed species</th>
<th>N</th>
<th>P</th>
<th>K</th>
<th>Ca</th>
</tr>
</thead>
<tbody>
<tr>
<td>SOLNI</td>
<td>1172,8</td>
<td>54,1</td>
<td>711,7</td>
<td>360,1</td>
</tr>
<tr>
<td>ABUTH</td>
<td>700,0</td>
<td>31,2</td>
<td>345,8</td>
<td>121,6</td>
</tr>
<tr>
<td>AMACH</td>
<td>640,0</td>
<td>47,1</td>
<td>547,8</td>
<td>283,6</td>
</tr>
<tr>
<td>AMBAR</td>
<td>510,0</td>
<td>25,5</td>
<td>190,9</td>
<td>190,7</td>
</tr>
<tr>
<td>CHEAL</td>
<td>490,0</td>
<td>44,9</td>
<td>690,9</td>
<td>345,0</td>
</tr>
<tr>
<td>POLLA</td>
<td>330,0</td>
<td>24,0</td>
<td>240,7</td>
<td>121,5</td>
</tr>
<tr>
<td>CONAR</td>
<td>190,0</td>
<td>11,4</td>
<td>112,0</td>
<td>46,5</td>
</tr>
<tr>
<td>ECHCG</td>
<td>106,6</td>
<td>7,9</td>
<td>110,6</td>
<td>40,5</td>
</tr>
<tr>
<td>POLAV</td>
<td>100,0</td>
<td>9,6</td>
<td>49,8</td>
<td>28,6</td>
</tr>
<tr>
<td>LATTU</td>
<td>43,7</td>
<td>1,3</td>
<td>24,9</td>
<td>7,2</td>
</tr>
<tr>
<td>MATIN</td>
<td>40,0</td>
<td>3,2</td>
<td>41,5</td>
<td>14,3</td>
</tr>
<tr>
<td>SETGL</td>
<td>20,0</td>
<td>1,7</td>
<td>33,2</td>
<td>14,3</td>
</tr>
<tr>
<td>Total (weeds)</td>
<td>4309</td>
<td>262</td>
<td>3100</td>
<td>1573</td>
</tr>
<tr>
<td>Potato</td>
<td>3339</td>
<td>303</td>
<td>4278</td>
<td>2333</td>
</tr>
</tbody>
</table>

Based on literature data potato needs 3 kg Ca to 1 t tubers. From among the 12 weed species four species have taken up 70% of the Ca taken
up by all weeds. These species are as follows: *S. nigrum*, *C. album*, *A. chlorostachys* and *A. artemisiifolia*.

**Conclusions**

Based on our experimental data we have found that on the untreated weedy plots the spreading of the weeds was very high. Weed fresh weight was 50% of the potato fresh weight.

Weeds have taken up 4.35 g N, 0.26 g P, 3.10 g K and 1.57 g Ca altogether. These nutrient quantities correspond to 43.5 kg N ha\(^{-1}\), 2.6 kg P ha\(^{-1}\), 31 kg K ha\(^{-1}\) and 15.7 kg Ca ha\(^{-1}\).

**Acknowledgement**

The authors would like to thank Dr. Sándor Horváth for his assistance.

**References**


NEW POSSIBILITIES IN THE SUNFLOWER POSTEMERGENT HERBICIDE APPLICATION

András Horn
Summit-Agro Hungaria Ltd, Budapest

Flumioxazine (PLEDGE-SUMISOYA) is a well known herbicide in the broad leaf-control of soybean and maize (corn) all over the world. Main properties of flumioxazine (PLEDGE) are follows:
- efficacy is independent from the temperature.
- in case of pre- or postemergent application 5-10 mm rainfall after the treatment increases the effect of the herbicide.
- no phytotoxicity is on the following crop. The half-life time (degradation) is 30 days under average conditions. Organic matter content of the soil has limited influence on the herbicide efficacy.
- The product has very low water-dilution capability.

Figure 1. Main weeds in sunflower and effect of flumioxazin on them treated preemergence or postemergence
As a result of a 6 years development work of the trading company Summit-Agro Hungaria, the product was registered in Hungary in sunflower as a preemergent and postemergent herbicide as well.

In Figure 1 the main weeds in sunflower (ranking of the weeds) and the effect of flumioxazine as preemergent and postemergent treatment are summarized.

In Figure 2 are summarized the tankmixture possibilities and dosages in sunflower.

Figure 2. Tankmixture possibilities and dosages in sunflower
AUTUMN APPLICATION OF METSULFURON-METHYL AGAINST APERA SPICA-VENTI (L.) P.B.

Elemér Tóth¹ – István Molnár¹ – István Somlyay¹ – Sándor Bálint¹
– Margit Nagy² – Lajos Szőke²

¹DuPont Hungary Ltd., Budapest
²Crop and Soil Protection Service of Szabolcs-Szatmár-Bereg county, Nyíregyháza, Hungary

Application of autumn weed control against T₁ and T₂ life cycle type weed species is a tradition on some part of Western Europe mainly against Alopecurus myosuroides and Apera spica-venti. In Hungary there is a very limited infection of Alopecurus myosuroides, but Apera spica-venti is a serious problem on acidic soils. Its spreading areas are Szabolcs-Szatmár-Bereg, Somogy, Zala, Vas, Győr-Moson-Sopron and Veszpém counties. In 2003 metsulfuron-methyl (Ally® 20 DF), a new herbicide was registered in winter wheat, winter barley and triticale in Hungary. In Western Europe a lot of farmers are using this herbicide to control black grass (Alopecurus myosuroides), but there is not too much data how it can control loose silky bent (Apera spica-venti). It was the reason, why a trial was carried out using of Ally® 20 DF on Apera infested area in Hungary.

Literature

There cannot be found a publication in the international publication lists which is written in this topic (connection of metsulfuron and Apera spica-venti), but there are some publications speaking about control of this weed species by other active ingredients. Prometryne + simazine was examined at the eastern part of Germany (BUHR et al., 1977). Primary dormancy of seeds of Apera spica-venti and Alopecurus myosuroides was tested by WALLGREN and AVHOLM (1978) and demonstrated that both weed species show a typical winter annual habit with a germination peak in the autumn and little or none during the following spring after the seeds had been “stored” outdoors in the soil. In Poland efficacy of some treatments (MCPA+ dicamba, 2,4-D+ dicamba, amidosulfuron, isoproturon, chlorosulfuron, fenoxaprop-P+ isoproturon) was compared by DOMARADZKI and ROLA (1997). The best efficacy was given by chlorosulfuron. The emerging of loose silky bent on sandy soils was proven by PETERSEN in 1998 in Denmark, where the early drilling resulted in heavy loose silky bent emergence. Even though loose silky bent
is controlled, the greatest yield are achieved after late drilling. Field studies were conducted in Poland during 1998 to study the effectiveness of a combined application of herbicide (Chisel 75 DF)+ growth regulator+ adjuvant mixture. The control of *Apera spica-venti* increased by 4-9 %. There was proven by CIMERMAN and BABNIK (1999) that isoproturon active ingredient acts residually for 2-3 months, residual activity of amidosulfuron is shown for 1-3 weeks against *Apera spica-venti*. Also ROLA et al. (1999) studied the efficacy of some sulfonylurea herbicides (chlorsulfuron + thifensulfuron-methyl, rimsulfuron and triflusulfuron) in tank mixture with adjuvants in winter wheat, corn and sugar beet. KLEM and VANOVÁ (2000) were examined the effect of sulfo sulfuron on *Apera spica-venti* and *Elytrigia (Elymus, Agropyron) repens*.

**Materials and Methods**

The efficacy of metsulfuron-methyl against *Apera spica-venti* was tested by Plant and Soil Protection Service of Szabolcs-Szatmár-Bereg county in Nagyhalász, Hungary in 2002/2003 season. Date of treatments were the following: autumn postemergence treatment: November 18, 2002, spring postemergence treatment April 15, 2003. The trial was treated by a “Szolnok” type sprayer machine, water volume was 300 litre/ha, pressure 2 bar. Plot size: 20 m\(^2\), number of replications: 4.

In Nagyhalász there is a brown forest type of soil, which has 1.81 % organic matter, this soil is acidic: pH(KCl) = 5.13.

The crop before winter wheat was in the 2001/2002 season was also winter wheat, the stubble-field peeling was done at July 26, 2003, then the soil preparation before sowing was carried out at October 8, 2003. Altogether – because the owner of field had some machinery capacity problem- the quality of soil preparation could be only sufficient.

Sowing of winter wheat was made at October 21, 2003, variety was GK Petur. In October the trial territory got 78 mm precipitation, after it *Apera spica-venti* started to emerge. So this fact resulted that by the time of the autumn postemergence treatment the phenology of *Apera spica-venti* was 1-3 (4) leaves stage, *Tripleurospermum inodorum* (*Matricaria inodora*) cotyledon- 2 leaves stage. Winter wheat was by the time of autumn postemergence treatment 1-3 leaves stage, by the time of spring postemergence treatments in tillering. By spring postemergence treatments *Apera spica-venti* individuals were foundable from 1 leaves stage unto tillering, *Tripleurospermum inodorum* (*Matricaria inodora*) was in 2-4 (6) leaves stage.
In four weeks after the autumn postemergence treatments the territory got 33 mm precipitation, in two weeks after the spring postemergence treatments 10 mm, but in four weeks also 33 mm precipitation. Assessment were done three times: the first assessment at April 1, 2003; second assessment at May 6, 2003; third assessment at July 6, 2003 (just before harvesting).

**Results**

Effect of Ally® 30 g/ha + Trend™ 0,1 % autumn postemergence treatment gave against in autumn emerged *Apera spica-venti* plants excellent weed control and this treatment could prohibit the spring emerge of this weed species.

Autumn application of Ally® 30 g/ha + Trend™ 0,1 % treatment proved its excellent efficacy against loose silky bent (Figure 1), and ensured 100 % efficacy against *Tripleurospermum inodorum* in case of all of the three assessments.

In the springtime a new flush of *Apera spica-venti* started to emerge, and the spring was very windy, that were the reasons, why the spring postemergence treatments were made at April 15. That time some individuals of loose silky bent emerged in autumn were already after tillering.
The effect of spring application of Ally® 30 g/ha + Trend™ 0.1 % treatment controlled well the less developed *Apera spica-venti* plants among the autumn emerged individuals three weeks after the treatment, plants of loose silky bent, which were more developed than 4-5 leaves stage by the spring treatment, were damaged, but some of them were regenerated in July. There could be establish that spring application of metsulfuron-methyl + non-ionic surfactant in this case could not give enough (more than 90 %) efficacy against the heterogen *Apera spica-venti* population, but showed better result than the standard treatments (amidosulfuron + jodosulfuron + mefenpir-diethyl + rape-oil 300 g + 1 l/ha and triasulfuron 15 g/ha). Metsulfuron-methyl could give longer effect than the standard treatments (Figure 2).

Figure 2.

Winter wheat weed control trial against *Apera spica-venti*.

Autumn use of Ally® 30 g/ha + Trend™ 0.1 % treatment showed an excellent weed control against *Apera-spica-venti* and it could keep that efficacy up to harvesting (Figure 3). The product is registered in Hungary from 3 leaves stage up to end of tillering of winter wheat, winter barley and triticale crops.
Figure 3. Nagyhalász, Eastern Hungary, 2003.

left: untreated           right: Ally® 30 g/ha + Trend™ 0.1 % (autumn treatment)

References


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Summary

AUTUMN APPLICATION OF METSULFURON-METHYL AGAINST APERA SPICA-VENTI (L.) P.B.

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Autumn application of metsulfuron-methyl 30 g f.p./ha + non-ionic surfactant 0.1 % was examined against Apera spica-venti in Hungary. The treatment showed an excellent weed control against loose silky bent and it could keep that efficacy up to harvesting. The product is registered in Hungary from 3 leaves stage up to end of tillering of winter wheat, winter barley and triticale crops.
Acetochlor (Ac, 2-chloro-N-ethoxymethyl-N-(2-ethyl-6-methylphenyl) acetamide) is a chloroacetanilide herbicide applied preemergence to control many annual grass and certain broadleaf weeds in maize (Zea mays L.). Maize exhibits marginal tolerance to the acetochlor. Therefore, the use of safeners is necessary to overcome the phytotoxic symptoms such as shoot and root growth reduction as well as distorted leaves and coleoptiles as consequences of the acetochlor treatment.

Safeners are chemical agents that increase the tolerance of crop plants to herbicides without affecting the weed control efficacy. They appear to induce a set of genes that encode enzymes and the biosynthesis of cofactors involved in the herbicide detoxication (Gatz, 1997). Glutathione S-transferase isoenzymes (GSTs) and endogenous glutathione (GSH) play a vital role in chloroacetanilide herbicide detoxication by GSH conjugation. Safeners of various chemical classes were found to induce the activity of GSTs and the level of GSH in the protected plants (Davies and Caseley, 1999). The dichloromethyl ketal MG-191 (2-dichloromethyl-2-methyl-1,3-dioxolane) is highly active used in the safening of maize against thiocarbamate and to a lesser extent chloroacetamide herbicides (Jablonkai, et al., 2001). MG-191 similarly to the other safeners antagonizes the growth inhibition of the acetochlor by eliminating the shoot-twisting and leaf-rolling injuries of maize seedlings but appears to be less effective in the root system even (Jablonkai, 1991).

In order to further clarify the mode of action of MG-191 and analogous molecules the significance of GST and GSH enhancement in safening maize against the herbicide acetochlor, the relationship of structure to safening efficacy, GSH and GST inducibility was examined using halogenated acetals (1a-l), ketals (2a-k) and acetamides (3a-d) with emphasis on the roots (Figure 1).
Figure 1. Examined halogenated acetals (details in the text)

\[
\begin{align*}
1a-l & : \quad & \begin{array}{c}
\text{R}^1 \quad \text{R}^2 \\
\text{O} \quad \text{O} \\
\text{H} \\
\text{C} \quad \text{XYCH}
\end{array} \\
2a-k & : \quad & \begin{array}{c}
\text{R}^1 \quad \text{R}^2 \\
\text{O} \quad \text{O} \\
\text{H} \\
\text{C} \quad \text{XYCH}
\end{array} \\
3a-d & : \quad & \begin{array}{c}
\text{R}^1 \quad \text{R}^2 \\
\text{N} \\
\text{XYCH} \quad - \quad \text{C}
\end{array}
\end{align*}
\]

**Materials and Methods**

*Chemicals*

Open-chain dichloromethyl acetals and ketals were synthesized from dichloroacetaldehyde, 1,1-dichloroacetone, and 1,1-dichloroacetophenone (Dutka, 1991). Cyclic acetals and ketals were prepared from diethyl acetal and ketal of dichloroacetaldehyde and 1,1-dichloroacetone by transacetalisation. Acetamides were synthesised by haloacetylation of amines using standard Schotten-Baumann conditions. Crude reaction products were purified by either distillation or silica gel column chromatography. Acetochlor was purified by column chromatography from the commercial product. \([\text{Carbonyl-}^{14}\text{C}]\text{acetochlor (sp. act. 37 MBq /mmol)}\) was a sample prepared previously (Jablonkai and Hatzios, 1991).

*Safener activity of experimental molecules*

Seeds of maize (Gazda Martonvasar, Hungary) were soaked in water and planted in plastic cups (6 cm diameter, 9 cm deep, 3 seeds/cup) containing air-dried foundry sand (250 g, OH-4 type). Treatment solutions (50 ml) containing safener (50 µM) and/or acetochlor (50 µM) were applied to each cup. Seeds were placed 2 cm deep. The plants were grown in a growth room (temperature: 23 ± 1 °C; relative humidity: 60 ± 5 %; light intensity: 10 klux; light period: 16 h per day). The plants were watered three times a week to bring the weight of cups to 300 g. Plants were harvested two weeks after the treatment shoot and root lengths measured. The experiment was carried out twice with four replicates.

*Plant materials and enzyme isolation*

For GST activity analyses seeds (25) of maize were placed in Petri dishes (18.5 cm in diameter) on two layers of filter-paper wetted by aqueous
solution (20 ml, 50 µM) of chemicals studied. The dishes were placed in a germination thermostat. The seedlings were grown in the dark for 5 days at 27 °C. Five-day-old seedlings were thoroughly washed with tap water and separated roots were homogenized in a mortar and pestle using quartz sand then extracted with 5 volumes of cold Tris-HCl buffer (100 mM, pH 7.5) containing 2 mM EDTA, 1 mM dithiothreitol and 5 % (w/v) polivinyl polypirrolidone. The homogenates were filtered through two layers of Miracloth and the filtrates were centrifuged at 10,000 x g for 20 min at 4 °C. The supernatants were brought to 80% (NH₄)₂SO₄ to 80 % saturation and centrifuged at 10,000 x g for 20 min at 4 °C. Aliquots of the protein precipitates were resuspended in potassium phosphate buffer (20 mM pH 6.5) and desalted by gel filtration (Sephadex G25, medium) before use for enzymatic studies.

For determination of GSH contents root tissues of etiolated seedlings were grown as described earlier. Tissues were frozen and homogenized in liquid nitrogen and extracted with 4 volumes of 70% ethanol The homogenates were centrifuged at 10,000 x g for 20 min at 4 °C and the supernatants were collected.

**Analysis of GSTs and GSH**

Glutathione S-transferase activities of desalted enzymes were determined with CDNB (1-chloro-2,4-dinitrobenzene) and [carbonyl-¹⁴C]acetochlor (Ac) substrates. GST(CDNB) activities were determined spectrophotometrically (340 nm) and expressed as nmol product formed per second (nkat) per mg protein (Dixon et al., 1998a). GST(Ac) activities of the samples were determined by liquid scintillation counting radioassaying the conjugate formed in the reaction of [carbonyl-¹⁴C]acetochlor (0.75 mM) and GSH (10.0 mM) mediated by the desalted enzymes at 37 °C in 30 min. The GST(Ac) activity was expressed as pmol conjugate per second (pkat) per mg protein. Protein contents of the extracts were determined spectrophotometrically using a Coomassie Brilliant Blue reagent with bovine serum albumin as reference protein.

Non-protein thiol (GSH) content of alcoholic supernatant was measured spectrophotometrically (412 nm) using DTNB reagent (Jablonkai and Hatzios, 1991).

**Results and Discussion**

Safening experiments were carried out in sand at a relatively high pre-emergence acetochlor rate (2.4 kg/ha) and at high moisture content. Under these conditions the herbicide is extremely phytoxic and no complete
protection can be achieved. The protection of shoots by bromoacetaldehyde diethyl acetal (1b) was moderate while the monochloroacetal 1a and the dichloroacetals (1c-l) exhibited poor or no safening activity (Table 1). Among dialkyl ketals having increasing alkyl chain length (2a-d) the highest safening activity was observed for the diethyl (2a) and dipropyl (2b) derivative. In general, cyclic ketals (2e-k) were effective safeners.

Derivatives having 1,3-dioxolane (2f), dioxane (2g) and dioxepane (2i) ring in their structure were the most active molecules. Interestingly, dioxacycloalkanes with 8- and 9-membered ring were still active in safening in the shoot zone. The safening activity of ketals also exceeded that of acetals against the thiocarbamate EPTC (Dutka, 1991). Among amides the marketed safener dichloroacetyl-diallylamide (dichlorimid, 3a) was highly protective and decreasing the number of halogens and allyl groups yielded less active molecules. The root growth inhibition of the acetochlor was less antagonised by the ketals 2a-k and the amides 3a-d as compared to safening the shoots of maize seedlings (Table 1). However the protective action of the acetals 1c-l in the root zone was superior to that in shoot zone and exceeded the root safening activity of the ketals and the amides. The higher activity of the acetals in the roots can be explained by their increased root uptake and reduced mobility towards shoots. In an early study the ketal type MG-191 (2f) was found extremely mobile after root application and its enhanced mobility to the root-absorbed acetochlor was considered as an important factor in its safening efficacy (Jablonkai, 1991).

The GSH content of the root tissues of safener-treated plants was only slightly affected by treatment with either acetals or ketals as compared to that of untreated control (Figure 1). The only exception was the moderately safening 2e cyclic ketal that triggered out a 2.4-fold increase. Cyclic acetals were inhibitory of GSH biosynthesis in roots. Among amides a high degree of induction was observed for the less effective safener 3b. It seems that no correlation exists between the elevation of GSH content in roots and the shoot safening efficacy of these molecules. On the other hands the elevation of GSH content has been observed for a number of safeners (Davies and Casely, 1999).
Table 1. Structure and safening activity of acetals, ketals and amides towards the acetochlor in maize

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ᵃ based on shoot length; protection (%) = 100 x [(herbicide + safener)] / [control - herbicide]; shoot lengths 14 DAT: control, 27.9±5.3 cm, acetochlor, 3.1±0.3 cm
ᵇ based on root length; root lengths 14 DAT: control, 9.5±1.2 cm, acetochlor, 6.2±1.2 cm
Figure 1. GSH content in the roots of maize seedlings pretreated with the acetochlor and the experimental safeners

GST(CDNB) activity of the root tissues was hardly affected by pretreatment with safeners (Figure 2). The acetals 1a-c were inhibitory while treatment with 1k and 1l cyclic acetals showing no safening activity to the shoots and some activity to the roots resulted in 2- and 4-fold increase in this isoenzyme activity. In general, no relationship can be shown between their effects on GST(CDNB) activity and shoot or root safening efficacy. GST(CDNB) activity associated with the safener inducible ZmGSTF1-2 isozyme was increased by the dichlormid in roots and shoots of maize (Dixon et al., 1997) and only marginally effected by the MG-191 (Jablonkai et al., 2001).

GST(Ac) activity in the roots was enhanced by both protective and less effective structures as compared to that of untreated control (Fig. 3). The treatment with the herbicide acetochlor in itself doubled this isoenzyme activity and exceeded the effects of the acetals. Among ketals the safener MG-191 (2f) induced the highest increase in the enzyme activity while no elevation was shown for the molecule 2i which posseses with the same safening activity in the shoot zone. For the amides the degree of induction on this isozyme activity appears to be parallel with their shoot safening potential. A correlation between GST(Ac) induction and safening activity of amides exists only in the shoot tissues (data not shown). These findings indicate that the mode of action of the acetals and ketals is likely differs from that of the amides.
Figure 2. GST(CDNB) activity in the roots of maize pretreated with the acetochlor and the experimental safeners

Figure 3. GST(Ac) activity in the roots of maize pretreated with the acetochlor and the experimental safeners

Pretreatment of maize seedlings with acetochlor resulted in a very high degree of induction of the enzyme activity indicating that the induction of GST isoforms by both chloroacetanilides and their safeners is based on a similar mechanism.
The exact mechanism of the safener-mediated enhancement of GST activity is not completely understood. GSTs are induced by a diverse range of chemicals and accompanied by the production of active oxygen species. Thus the connection between safener-mediated protection of crops and oxidative stress tolerance has been suggested (Theodoulou et al., 2003). Many GSTs are effective not only in conjugating electrophilic substrates but also function as glutathione peroxidases. Safeners may induce GST expression by mimicking oxidative insult (Dixon et al. 1998b). Our results indicate that safener structure plays a decisive role in specific expression of GSTs mediating the detoxication of chloroacetamide herbicides and seems to be tissue specific. Since no strong correlation between the degree of induction of levels of GSH and GST isoforms and the safener activity was found the mode of action of safeners is a more complex process than simply promoting the metabolism of herbicides.

References


**Summary**

**INDUCTION OF MAIZE GLUTATHIONE S-TRANSFERASES BY HALOACETAL, HALOKETAL AND HALOAMIDE SAFENERS**

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The herbicide safener MG-191 and its acetal and ketal analogues as well as mono- and dichloroacetamides were tested for their ability to alleviate toxicity of acetochlor to maize. The differential enhancement of the GSH content and the expression of GST isoforms was studied in roots of maize. Our results demonstrate that the safener structure affects the specific expression of GSTs mediating the detoxication of acetochlor. No correlation was found between the degree of induction of GSH and GSTs and the safening activity.
LECTURES OF
ENTOMOLOGICAL &
ECOLOGICAL SESSION
THE LACEWING FAUNA (NEUROPTERA: CHRYSOPIDAE) OF DEBRECEN

(Summary)

András Bozsik

Department of Plant Protection, University of Debrecen, Debrecen, Hungary

Studies were conducted in the botanical garden and the surrounding experimental area of the Faculty of Agricultural Sciences in Debrecen to assess structure parameters of the occurring lacewing assemblage(s). Localities were sampled by sweeping net from early April until early October in 1996-2003.

In the sample area at least 12 chrysopid species have been captured among which 7 were found relatively constantly. Sibling species of the Chrysoperla carnea complex (Chrysoperla affinis (kolthoffi), Chrysoperla carnea s. str., Chrysoperla lucasina) were the most common species, comprising 75-96 % of the total, followed by individuals of Dichochrysa prasina, Chrysopa pallens, Chrysopa formosa, Chrysopa perla, Chrysopa viridana, Chrysopa phyllochroma, Chrysopa nigricostata and Chrysotropia ciliata specimens occurred only rarely and singly. The assemblages were characterized by low diversity and high similarity values. The original and processed data stress the scarcity of the lowland lacewing fauna.
According to Günther (1975), the Spilostethus equestris L. is indigenous from southern England to Siberia, and from central Sweden to the Mediterranean areas. It is less common in the north of Central Europe; it prefers areas with warmer climate. It likes staying both on the ground or in the flowers of various plants. It especially likes the tame poison or swallowwort (Vincetoxicum officinale MNCH., syn.: Cynanchum vincetoxicum (L.) PERS.).

Data on the life history of the S. equestris in Hungary is extremely scarce, despite the fact that the species is widespread in our country. Information on its nourishing plants and habits can only be found in the publications of Horváth (1984; 1986; 1987b; 1989; 1999) and Bujáki and Horváth (1992). Horváth (1984) inferred the nourishing plant specialization of the species from the total nitrogen contents of various plant seeds, such as those of the Asclepias syriaca L., and the sunflower. Amino-acid composition in the seeds of the A. syriaca is the most similar to that of the sunflower, soy or peanut. It may be assumed that its similarity to the sunflower seed is the cause of the fact that the S. equestris willingly sucks the ripening achenes of the sunflower, too (Horváth, 1984).

Material and Methods

Our study was carried out in the districts of Bácsalmás („free” of A. syriaca) and Katymár (heavily infected with the A. syriaca) between 25 and 31 August 2001. The two districts were significantly different in respect of infection with A. syriaca.

While the A. syriaca only occurred sporadically in the forest stripes and in the edges of the industrial (hybrid propagating) sunflower fields in the Bácsalmás district, in the Katymár district the occurrence of the weed was 20-25 specimen/m².

The damage caused by S. equestris was investigated in the selected land strips as follows: ten randomly selected, adjacent sunflower discs were
examined at every 10 meters (10 locations in total) from the edge of the land strip towards the centre.

By carrying out these investigations we tried to assess the extent and frequency of the damage caused by the *S. equestris*; partly as a function of the number of the achenes damaged in a disc, and partly of the occurrence of the *A. syriaca*. We also wanted to understand the cause of the „greening of the seed inside the achene”, which phenomenon adversely affects the export market position of the striped, alimentary purpose species (hybrids).

Our investigations aimed at finding satisfactory answers both in respect of the market „condition” of the export consignments of the striped, alimentary purpose seeds, and the germination value (germ %) of hybrid sunflower propagation.

**Results and Discussion**

Both Table 1 and Table 2 show clearly the difference in the damage caused by the *S. equestris* that difference originates from the extent of infection with the *A. syriaca* (the main nourishing plant of the *S. equestris*). The occurrence of this insect significantly increased in the areas that were infected with the *A. syriaca*. Overwintering of the adults was relatively unhindered due to the mild winters in recent years.

The data in the tables make it unambiguous that the severe damage caused by plant bugs is prominent primarily in the 40 m wide strips of the land strips (Table 1 and 2). The settlement of, and characteristic damage caused by the bugs can best be observed in these stripes. This applies to the damage characteristics of both the Katymár district (which is more infected with the *A. syriaca*) and the less infected areas (Bácsalmás district). At the same time, it has also been proved, that now the damage caused by the *S. equestris* (which species was earlier referred to as a typical “pest of the edges” of land strips) (Horváth, 1989; 1991) is no longer restricted to the edges of the land strips. Although to a decreasing extent, but it is present at a distance of 100 metres from the edge of the land strip, that is worth paying attention. It is especially true, as the *S. equestris* may be a vector of the cucumber mosaic virus (*Cucumber mosaic Cucumber virus*) or other viruses, the presence of which can be demonstrated on the *A. syriaca* (Horváth, 1980; 1981). This is also supported by the observations of recent years that refer to the spreading of the damage caused by pathotype B of the cucumber mosaic virus (CMV), and the *sunflower chloratic virus* (SuCMoV) (Salamon, 2002, Lenardon et al., 2001). The severe damage caused by the bug locally (in spots within a strip of land) usually occurs after the fall of tubular flowers (about the middle or end of August).
### Table 1. Occurrence of achenes damaged by *Spilostethus equestris* L. (number of specimen) Katymár, 2001

<table>
<thead>
<tr>
<th>Number</th>
<th>1 (10m)</th>
<th>2 (20m)</th>
<th>3 (30m)</th>
<th>4 (40m)</th>
<th>5 (50m)</th>
<th>6 (60m)</th>
<th>7 (70m)</th>
<th>8 (80m)</th>
<th>9 (90m)</th>
<th>10 (100m)</th>
<th>Total, pcs</th>
<th>Mean of 10 discs</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>47</td>
<td>50</td>
<td>36</td>
<td>20</td>
<td>20</td>
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<td>16</td>
<td>15</td>
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<td>20</td>
<td>10</td>
<td>21</td>
<td>16</td>
<td>319</td>
<td>31.9</td>
</tr>
<tr>
<td>3</td>
<td>65</td>
<td>50</td>
<td>38</td>
<td>30</td>
<td>15</td>
<td>25</td>
<td>12</td>
<td>16</td>
<td>20</td>
<td>20</td>
<td>291</td>
<td>29.1</td>
</tr>
<tr>
<td>4</td>
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<tr>
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<td>44</td>
<td>47</td>
<td>48</td>
<td>17</td>
<td>29</td>
<td>10</td>
<td>14</td>
<td>20</td>
<td>26</td>
<td>351</td>
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<td><strong>515</strong></td>
<td><strong>427</strong></td>
<td><strong>416</strong></td>
<td><strong>228</strong></td>
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<td><strong>189</strong></td>
<td><strong>142</strong></td>
<td><strong>3171</strong></td>
<td><strong>31.71</strong></td>
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</table>
Table 2. Occurrence of achenes damaged by *Spilostethus equestris* L. (number of specimen) Bácsalmás, 2001

<table>
<thead>
<tr>
<th>Number</th>
<th>Number of damaged achenes per location (10 discs from each location)</th>
<th>Total, pcs</th>
<th>Mean of 10 discs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>(10m)</td>
<td>(20m)</td>
<td>(30m)</td>
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<tr>
<td>1</td>
<td>26</td>
<td>23</td>
<td>26</td>
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<tr>
<td>2</td>
<td>32</td>
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<td>3</td>
<td>18</td>
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<td>4</td>
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<td>23</td>
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<td>9</td>
<td>18</td>
<td>17</td>
<td>10</td>
</tr>
<tr>
<td>10</td>
<td>23</td>
<td>20</td>
<td>8</td>
</tr>
<tr>
<td><strong>Total:</strong></td>
<td><strong>224</strong></td>
<td><strong>204</strong></td>
<td><strong>196</strong></td>
</tr>
</tbody>
</table>
Stinging by the imago is the most common at the connection point (coronula) of the tubular flower and the achene. At this point a vulnerable, poorly protected surface is formed, that lasts for 2 to 3 days. Here the bug is able to penetrate into the internal substance quite easily with its powerful stinging/sucking mouth organ. However, greening of the contents (the seed) hardly ever occurs at this point – which problem may especially be a crucial “technical” problem with the striped, alimentary purpose hybrids during acceptance of export consignments –, contrary to the damage caused at the shoulder part of the achene, which mostly (in 70% to 80% of the cases) results in the greening of the seed (chlorophyll formation started by the effect of sunshine, which penetrates through the large injuries).

The adults carry on their maturation nourishment on the achene, and also on the axis inflorescentiae, the torus, and squame of the sunflower. At the reverse side of the disc, scars are formed of the injuries, which typically ulcerate, and later get suberized. Such wounds may facilitate the penetration of various harmful fungi (the *Rhizopus arrhizus* FISHER, in the first place). In the course of our study, we could also observe an “unusual” method of laying eggs of the *S. equestris*. This species usually lays eggs in the soils. However, on 27 August 2001, some specimen gave up the “normal” way, and laid their eggs among the achenes, and in the area sheltered by the outermost row of achenes, the periclinium and by some squame, too. (This had also been observed on one occasion earlier, on 11 August 1991. This phenomenon, by all means, contributes new information to the biology of the *S. equestris*).

Our observations have also made it clear that the classification of the *S. equestris* as an “auxiliary pest” has significantly changed in the years past. This species is playing a more and more dominant role among the plenty of pests of the sunflower. Studies on germination biology carried out prior to, or concurrently with, our studies have proved, among others, that the damage caused by these plant bugs may be significant – though they do not attack the germs directly –, because various species of fungi (*Alternaria* sp., *Botrytis cinerea*, etc.) may accumulate in the suction area, that have well-known harmful effects on the germ (Horváth-Bujáki, 1991).

**Acknowledgements**

The authors wish to express their gratitude for valuable data contributed by Dr Andrea Freese researcher (Commonwealth Institute of Biological Control, Delemont, Switzerland), and Professor Brigitta Sillentulberg (Department of Zoology, University of Stockholm, Sweden).
References


Summary

SPILOSTETHUS [= LYGAEUS] EQUESTRIS L., (HETEROPTERA: LYGAEIDAE), A PEST OF SUNFLOWER

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¹Faculty of Horticulture, College of Kecskemét, Kecskemét, Hungary
²Franciska BT., Kecskemét, Hungary

Western European, and especially German-based multinational food processing companies tend to have “alimentary purpose” sunflower hybrids and varieties grown in Hungary. This “commercial” product is highly preferred as substitute of walnuts, or as filling in chocolate, bread, other bakery products, or baked into the top layer thereof.

In early 1990s German companies rejected several truckloads of export goods due to “greening of the seed in the achene (kernel)”. Almost 90% of the rejected consignments originated from the sandy ridge of the region between the rivers Danube and Tisza, which is severely infected with the Asclepias syriaca L. weed. Field investigations revealed unambiguously that the greening of the seed in the achene had been caused by the (Spilostethus [= Lygaeus] equestris L.), that is a plant bug species. This species is able to cause extensive damages and discontinuities in the so-called shoulder part of the achene with its powerful stinging-sucking mouth organ. In the damaged area of intensive chlorophyll formation begins due to the solar effect, which is the clear cause of the greening of the seed in the achene.

In our investigations we studied the industrial purpose sunflower plantations in two neighbouring areas (Bácsalmás and Katymár) in respect of the damage caused by the S. equestris. While in Bácsalmás (that district was less infected with A. syriaca) we found a damage of decreasing intensity (8.85 damaged achenes per sunflower disc) as advancing towards the centre of the plantation, while in the area of Katymár (this area was severely infected with A. syriaca), this value was almost three times as high, reaching 31.71 damaged achenes per disc. Though the numbers are not very high in themselves – assuming that a well-built disc contains 1100 to 1200 seeds –, but the damage may result in significant fall of quality, or even prevent exporting.

Our extensive research into the causes of the greening of the sunflower seeds made it clear that the S. equestris has a significant influence on the contents of the seeds it has damaged: it increases the proportion of linoleic acid (C₁₈₂) by about 2.5%. That affects the lasting quality of the achene unfavourably (Horváth and Bujáki, 1991). For this reason, a comprehensive study of the biology of the S. equestris became necessary for the developing possible protection methods. Such studies also represented the beginning of the investigation of any external or endogenous factors, that might obstruct the successful use of alimentary purpose sunflower hybrids or varieties in the food industry.
EFFECT OF SOME ORGANOPHOSPHATE INHIBITORS ON ACETYLCHOLINESTERASE IN FORFICULA AURICULARIA ADULTS (DERMAPTERA: FORFICULIDAE)

A. Bozsik\(^1\) – E. Haubrure\(^2\) – Ch. Gaspar\(^2\)

\(^1\)Department of Plant Protection, University of Debrecen, Debrecen, Hungary
\(^2\)Zoologie générale et appliquée, Faculté des Sciences agronomiques de Gembloux, Gembloux, Belgium

The importance of biological control and IPM in the agricultural production seems to become greater and greater. However, the introduction and use of beneficial organisms as biocontrol agents is difficult in practice, because of their sensitivity to pesticides. Possible solutions can be the utilisation of pesticides which are harmless to biological agents (Hassan, 1989) or to find (collect/select) tolerant or resistant strains of natural enemies (Grafton-Cardwell and Hoy, 1985). These goals may be achieved only with the study of the effects of pesticides on beneficial species.

Our aim in the present study was to analyze the detrimental effects of some organophosphorous insecticides on acetylcholinesterase(s) (AChE) of adult common earwig, *Forficula auricularia* L., an efficient polyphagous predator which feeds on aphids, codling moth, and scales (Helsen *et al.*, 1998, Schruft *et al.*, 1995). In addition, this analysis may contribute to the better knowledge of acetylcholinesterase(s) of the earwigs barely studied from this point of view.

**Materials and Methods**

*Insects*

*F. auricularia* adults derive from a field population collected in the experimental area of the Gembloux Agricultural University (Gembloux, Belgium) in 1995. Captures were obtained by sweeping net. Insects were used for analyses immediately after captures.

*Insecticides*

All insecticide active ingredients were purchased: paraoxon, malaoxon, carbaryl, diazinon, schradran (Riedel-de Haën, Belgium).
**Inhibition experiments**

*Preparation of homogenates*

20 adult earwigs were decapitated then their heads homogenized with 2 ml cold HST buffer (Tris HCl 1 mM, pH 8; NaCl 1 M; Triton X-100 1% wt/vol; MnCl₂ 1 mM; CaCl₂ 1mM) in a motor-driven, Teflon pestle and glass tube tissue grinder. This homogenate was filtered through a thin layer of glass wool and centrifuged 15 min at 15300 g and 4 °C. Immediately after centrifugation the supernatant was used for AChE assay.

*AChE assay*

AchE activity in homogenates of earwig heads was estimated with the procedure of Ellman *et al.*, (1961) using a Shimadzu UV-160A spectrophotometer. The assay medium consisted of 10 µl of the following solution (36.6 mg DTNB (5-5-dithiobis 2-nitrobensoic acid) and 15 mg NaHCO₃ in 10 ml phosphate buffer (0.1 M, pH 7.2)), sufficient Tris HCl buffer (1mM, pH 8) to give a final reaction volume of 1 ml, 10 µl acetone solution of paraoxon or malaoxon, 80 µl supernatant, 10 µl acetylthiocholine iodide (100 mM). Acetylthiocholine iodide was added only after a preincubation for 1 min to the other reaction components being in a cuvette. Then it was mixed rapidly and the change in absorbance was measured continuously at 412 nm for 1 min. Each enzyme assay included 9-15 cuvettes (two containing untreated (control) homogenates, 5-11 treated homogenates and two blanks). The blanks contained the same components except that substrate was omitted and instead of the pesticide acetone was added. At least duplicate analyses at each of 5-10 concentrations of pesticides were carried out.

**Surface contact treatment**

*Chemicals*

Parathion (range of 0.00028, 0.00083, 0.0025, 0.0075, 0.03, 0.12 %); the acetone solution of the active ingredients (300 µl/disc) was applied on Whatman paper discs. After the solvent evaporated, the disc was placed into a plastic petri dish with 8 cm diameter. 10 adults were immobilized to facilitate handling by exposing them briefly to carbon dioxide and then placed in each dish. There were 2 dishes per concentration. The test animals remained in the dish until a stable mortality resulted. The number of paralyzed or dead individuals was recorded after 1, 2, 4, hours, 1, 2,…days. Data were analyzed by probit analysis (Finney, 1971) with a program that incorporates Abbot's (1925) correction for natural mortality. All tests were conducted in the laboratory at 22-25 EC, 50-70 % RH, and under a 15:9 (L:D) photoperiod.
Results and Discussion

One population sample of *F. auricularia* was tested against the following classical AChE inhibiting insecticides: paraoxon, malaoxon, carbaryl, diazinon and scharadran. Schradran showed the slightest efficiency. It was so small that it was not possible to determine its I$_{50}$ value. As an approximate value it can be indicated that the I$_{50}$ value must be considerably over 10$^{-3}$ M which is the highest usable concentration. The other inhibitors’ I$_{50}$ and K$_{i}$ values (Table 5, 6), except that of paraoxon, were - regarding its order of magnitude - very similar to those assessed on ladybird (*Coccinella septempunctata*) and lacewing (*Chrysoperla carnea* s.l. (Bozsik et al., 1996). As to the paraoxon, earwig’s AChE responded extremely susceptibly to it. Its I$_{50}$ and K$_{i}$ values represented extraordinarily higher efficiency than the corresponding values measured on ladybird and lacewing.

The pesticide active ingredients’ efficiency order against the earwig AChE was the following: paraoxon > carbaryl > malaoxon > diazinon > schradran. Comparing the efficiency of the surface contact effect of parathion on earwig with that on *C. septempunctata* (Bozsik et al., 1996), the earwigs were much more tolerant to parathion than the ladybirds (Table 3).

Table 1. Susceptibility of AChE of *Forficula auricularia* Adults from the Experimental Orchard of the Gembloux Agricultural University (Gembloux) to Paraoxon and Malaoxon

<table>
<thead>
<tr>
<th>Earwig</th>
<th>Paraoxon</th>
<th>Malaoxon</th>
</tr>
</thead>
<tbody>
<tr>
<td>Population</td>
<td>I$_{50}$</td>
<td>K$_{i}$</td>
</tr>
<tr>
<td>Ge95</td>
<td>3.6 x 10$^{-6}$</td>
<td>193188.3</td>
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Table 2. Susceptibility of AChE of *Forficula auricularia* adults from the Experimental Orchard of the Gembloux Agricultural University (Gembloux) to Diazinon and Carbaryl (Abbreviations see Table 1)

<table>
<thead>
<tr>
<th>Earwig</th>
<th>Diazinon</th>
<th>Carbaryl</th>
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</thead>
<tbody>
<tr>
<td>Population</td>
<td>I$_{50}$</td>
<td>K$_{i}$</td>
</tr>
<tr>
<td>Ge95</td>
<td>3.4 x 10$^{-3}$</td>
<td>206.7</td>
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Table 3. Surface Contact Effect of Parathion on *Forficula auricularia* adults (Gembloux)

<table>
<thead>
<tr>
<th>Time of evaluation (h)</th>
<th>LC$_{50}$ (%) (95% FL)</th>
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<td>96</td>
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<td>144</td>
<td>0.0293</td>
</tr>
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<td>0.0176 – 0.0542</td>
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</table>

References


152
Summary

EFFECT OF SOME ORGANOPHOSPHATE INHIBITORS ON ACETYLCHOLINESTERASE IN FORFICULA AURICULARIA ADULTS (DERMAPTERA: FORFICULIDAE)

A. Bozsik¹, E. Haubruge² and Ch. Gaspar²
¹Department of Plant Protection, University of Debrecen, Debrecen, Hungary
²Zoologie générale et appliquée, Faculté des Sciences agronomiques de Gembloux, Gembloux, Belgium

In vitro enzyme activity of head homogenates of Forficula auricularia collected from the experimental area of the Gembloux Agricultural University and in vivo surface contact treatments with organophosphorous active ingredients on the same species were investigated. The in vitro studies based on the Ellman method showed that the acetylcholinesterase (AChE) of the studied F. auricularia population sensitivity order to the inhibitors was as follows here: paraoxon, carbaryl, malaoxon, diazinon, schradran. Comparing the I⁰⁰ and Kᵢ values with those of measured in Coccinella septempunctata and Chrysoperla carnea s.l. it seems that the last two species are more tolerant to the AChE inhibitors acting in vitro then the earwig.
SPECIES SPECTRUM OF FLEA BEETLES
(*PHYLLOTRETA* SPP., COLEOPTERA, CHRYSMELIDAE) ATTRACTED TO ALLYL ISOTHIOCYANATE-BAITED TRAPS IN HUNGARY

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⁴Debrecen University, Centre for Agricultural Science, Debrecen, Hungary

There are about 250 flea beetle spp. (Coleoptera, Chrysomelidae, Halticinae) present in the Carpathian basin, of which first of all those species cause agricultural damages, which live on cruciferous plants (mainly *Phyllotreta* spp.). In case of univoltine species most important damages are caused in the spring by the overwintering adults, on the one hand by feeding on leaves of seedlings, and on the other by propagating plant pathogens.

In the control of flea beetles the application of a trap would be very useful, which could detect the occurrence of overwintering adults in early spring, could monitor their flight pattern and could give estimates of the size of the populations of the overwintering generation, and also of the next generation of adults which appear later in the summer.

It has long been known that some plant volatiles, i.e. in the case of cruciferous plants isothiocyanates, which derive as secondary metabolites from the decomposition of non-volatile glucosinolates (feeding and oviposition stimulants), play the role of food attractants in the case of certain *Phyllotreta* spp. The objective of our present study was to investigate the species spectrum of flea beetles which respond to allyl isothiocyanate (which has been described as one of the most potent attractants for certain flea beetles) in Hungary, so that the applicability of this attractant for plant protection purposes can be evaluated.

At all experimental sites our traps baited with allyl isothiocyanate caught large numbers of *Phyllotreta cruciferae* Goeze. Catches in baited traps were always significantly higher than in unbaited ones. Our results confirm earlier reports on the attractiveness of this compound towards this species described earlier from other parts of Europe and from North America (Canada). This species is one of the most important pest flea beetles in Hungary.
The second most frequently recorded species captured was *P. vittula* Redtb. in our experiments. Traps with allyl isothiocyanate clearly caught more than unbaited ones showing a strong attraction by this compound. No previous reports on allyl isothiocyanate attracting this species has been published. Regularly significantly more beetles were caught in baited traps from *P. procera* Redtb. as well. Attraction by this compound has not been published before for this species either.

In a test conducted at Pusztaszabolcs, in a rape field, baited traps caught sizeable numbers of *P. balcanica* Heikert while no beetle were captured in unbaited traps. Similar results were obtained in case of *P. nodicornis* Marsham, in another test at Nadap. No mention of allyl isothiocyanate attracting these spp. was found in previous literature.

In the case of *P. undulata* Kutsch. although significantly more beetles were caught in baited traps than in unbaited ones in a test at Budakalász, which may be an indication for the attractivity of allyl isothiocyanate towards this species, however, due to the overall low numbers caught this should be confirmed in future tests.

Apart from *Phyllotreta* spp., specimens of the close relative *Psylliodes chrysocephala* L. were also captured in significantly larger numbers in baited than in unbaited traps, indicating that allyl isothiocyanate may play a role in the chemical communication of also this species. Scientists from the UK have already reported that certain isothiocyanates evoked an electrophysiological response on the antennae of *P. chrysocephala*, however, to the best of our knowledge ours is the first report on the field activity of the compound.

In some tests sizeable numbers of *Chaetocnema concinna* Marsh. were also captured. In this case however there were no differences between the catches of baited and unbaited traps, showing that allyl isothiocyanate did not influence the behavior of this species. This is not suprising, since the species does not feed on cruciferous plants.

As for relative abundance of these flea beetle species, in the catch by traps with allyl isothiocyanate ca. 80-90% of specimens belonged to *P. cruciferae*; 10-15% were *P. vittula*, or *P. procera*. Other species occurred only in small percentages, depending on experimental site. It is surprising, that of such, important pest species like *P. atra* Fabr., *P. undulata* or *P. nemorum* L. only very low numbers occasionally were captured. Further studies are needed to decide whether this was caused by the fact that allyl isothiocyanate is not attractive towards these spp., or whether these species were present in very low population densities at the test sites. Results are summered up in the Table 1.
Table 1. Species spectrum of flea beetles (*Phyllotreta* spp.) attracted to allyl isothiocyanate baited traps

<table>
<thead>
<tr>
<th>Species captured:</th>
<th>Attractive activity of allyl isothiocyanate:</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. cruciferae</em></td>
<td>has been known from literature; confirmed in this study</td>
</tr>
<tr>
<td><em>P. vittula</em></td>
<td>was discovered in this study</td>
</tr>
<tr>
<td><em>P. procera</em></td>
<td>was discovered in this study</td>
</tr>
<tr>
<td><em>P. balcanica</em></td>
<td>was discovered in this study</td>
</tr>
<tr>
<td><em>P. nodicornis</em></td>
<td>was discovered in this study</td>
</tr>
<tr>
<td><em>P. undulata</em></td>
<td>was discovered in this study; needs further confirmation</td>
</tr>
<tr>
<td><em>P. chrysocephala</em></td>
<td>was discovered in this study; electrophysiological activity has been known before</td>
</tr>
<tr>
<td><em>C. concinna</em></td>
<td>was not observed in our studies</td>
</tr>
</tbody>
</table>

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