Crop Protection 77 (2015) 38–44

Contents lists available at ScienceDirect

Crop Protection

journal homepage: www.elsevier.com/locate/cropro

Flea beetles (Coleoptera: Chrysomelidae, Alticinae) in Bt- (MON810) and near isogenic maize stands: Species composition and activity densities in Hungarian fields

Ágnes Szénási [a], *, Viktor Markó [b]

[a] *Plant Protection Institute, Faculty of Agriculture and Environmental Sciences, Szent István University 2100 Gödöllő Páter K. str. 1, Hungary*
[b] *Department of Entomology, Faculty of Horticultural Science, Corvinus University of Budapest 1118 Budapest Ménesi str. 44, Hungary*

A R T I C L E I N F O

Article history:
Received 2 April 2015
Received in revised form
7 July 2015
Accepted 8 July 2015
Available online 25 July 2015

Keywords:
Flea beetles
Bt maize
MON810
Species composition
Phyllotreta atra
Phyllotreta vittula

A B S T R A C T

Flea beetles (Chrysomelidae, Alticinae) were collected with Pherocon AM yellow sticky traps in maize plots to compare the assemblages from transgenic Bt- (genetic event MON810, producing Cry1Ab protein effective against lepidopteran pests) and near isogenic maize in Hungary. Altogether, 51,348 flea beetle individuals from 26 species were collected. The dominant species were *Phyllotreta atra* (F.) and *Phyllotreta vittula* (Redtenbacher). Their abundances along with other (non-*P. atra* and non-*P. vittula*) flea beetle species showed no significant differences between Bt- and isogenic maize plots. Similarly, no difference was found between Bt maize and isogenic maize plots in the species richness of the flea beetle assemblages.

1. Introduction

The Cry1Ab toxin produced by maize hybrids containing MON810 event is highly effective in controlling target Lepidoptera larvae (*Ostrinia nubilalis* (Hübner), Pyralidae and *Sesamia nonagrioides* (Lefebvre), Noctuidae) and may also affect larvae of other Lepidoptera species (e.g. *Helicoverpa armigera* (Hübner) [Eizaguirre et al., 2010; Kiss et al., 2003] *Mythimna unipuncta* (Haworth) [Eizaguirre et al., 2010; Pilcher et al., 1997]). However, maize fields harbour species rich assemblages of other arthropod groups (Mészáros et al., 1984), including flea beetles, which are present in maize stands in high numbers, occasionally causing damage in both Europe (Vörös and Maros, 2004) and North America (Hoffmann et al., 1995). These phytophagous insects in maize may also serve as prey for predacious species (Árpás et al., 2005). The presence of the Cry1Ab protein in all maize plant parts throughout the whole growing season might affect a number of associated organisms besides the target pests. Herbivores feeding on maize may be exposed to the Cry1Ab protein, e.g., the Cry1Ab toxin was detected in the flea beetle *Chateocnema pulicaria* Melsheimer feeding on MON810 maize (Harwood et al., 2005), although herbivores with different types of mouthparts (chewing, sucking) may ingest different amounts of Bt toxin. Also, predators consuming phytophagous insects containing Cry1Ab toxin may move the toxin into higher trophic levels (Harwood et al., 2005).

European Union legislation requires a pre-market risk assessment of genetically modified crops before commercial use (EC, 2001, 2002; EFSA, 2010; EU, 2013). For that purpose, this study examined potential effects of genetically modified maize expressing the Cry1Ab toxin on non-target, within-maize herbivores, using flea beetles as model species.

Several studies have found no differences in abundance, seasonal activity and assemblage characteristics in several non-target herbivore taxa in Bt (MON810) and isogenic maize (Balog et al., 2010a,b; Bourguet et al., 2002; Daly and Buntin, 2005; Lozzia, 1999; Lozzia and Rigamonti, 1998; Musser and Shelton, 2003; Sehnal et al., 2004). These same authors have raised the importance of longer term field studies to detect possible cumulative effects over several seasons, to determine thresholds for detecting any effects and for selecting suitable species for impact studies. The

---

* Corresponding author.
E-mail address: szenasi.agnes@mkk.szie.hu (Á. Szénási).

http://dx.doi.org/10.1016/j.cropro.2015.07.008

necessity of a more ecological approach in the study of the potential impact of growing GM crops on non-target organisms is raised by Andow et al. (2006, 2013), Arpaia et al. (2014), Lövei and Arpaia (2005), Lövei et al. (2009).

Several studies have been conducted in Hungary on economically important maize pests, secondary pests, other herbivores, and predators including a 10-year maize ecosystem research study (Mészáros et al., 1984). This faunal survey, however, focused primarily on the occurrence of arthropod taxa in the studied maize fields and did not collect data on their abundance. Because the Cry1Ab toxin is expressed in all maize tissues sampling should examine all groups of herbivores in maize that occur at high enough density to permit quantitative comparisons between Bt and isogenic maize.

Our study group, flea beetles are best known as pests of Brassicaceae crops (Bohinc et al., 2013; Sáringer, 1990; Trdan et al., 2008). In contrast, *Phyllotreta vittula* (Redtenbacher) has been recorded as a pest on cereals (including maize) (Fritzsche, 1971; Szeőke, 1997), sugar beet and crucifers (Naibo, 1974), including yellow mustard (Hurej et al., 1997). Vig (1998a) reported a wide host range of *P. vittula* including grasses, maize and Brassicaceae, but damage in Hungary is reported only on cereals, such as maize (Nagy and Deseő, 1969; Szeőke, 1997). *Phyllotreta vittula* has been found damaging maize in Hungary (Gyulai and Garay, 1996; Hinfner and Papp, 1961; Jablonowski, 1906; Nagy and Deseő, 1969; Szeőke et al., 1996; Szeőke, 1997; Szűcs, 1973; Vörös and Maros, 2004), in the former Soviet Union (Arnoldi and Gurjeva, 1960; Scsegolev, 1952), in Germany (Sorauer, 1954), in Poland (Kania, 1962), in the former Czechoslovakia (Hruska, 1962), in France (Leclant, 1977), in the former Yugoslavia (Sekulic et al., 1989) and in Romania (Grozea et al., 2006), making it one of the most important Coleoptera species attacking maize foliage (Sekulic et al., 1989). Leaf-damage by *P. vittula* is most pregnant in spring on the juvenile plants (Sáringer, 1990; Scsegolev, 1951; Szeőke et al., 1996).

In addition to *P. vittula*, *Chaetocnema aridula* (Gyllenhal) is found in Hungary on maize and other cereals (Sáringer, 1990). The flea beetle *Epitrix cucumeris* (Harris) is an early season pest of maize in Kansas, USA, where chemical controls have occasionally been needed (Wilde et al., 2004). *Chaetocnema pulicaria* is a pest of maize in the USA (Steffey et al., 1999) and an important pest of sweet corn in Iowa (Hoffmann et al., 1995).

In Hungary, dense flea beetle populations occur only in periods of dry, sunny weather (Sáringer, 1990), when young maize plants are especially susceptible to injury if drought stressed (Szeőke et al., 1996; Vörös and Maros, 2004). In older plants, flea beetles reduce foliar area and thus photosynthesis (Szeőke et al., 1996). Under dry conditions, damage increases and plant development is inhibited (Nagy and Deseő, 1969). Among different crops the most conspicuous injury by *P. vittula* is found in maize, mainly on the edges of fields and in unevenly emerged stands. In some cases, the lower 1–4 leaves of certain maize plants are totally destroyed (Gyulai and Garay, 1996).

Arthropod assemblages were surveyed in Bt- and in isogenic maize plots from 2001 to 2003 under an EU-5th framework project. In 2001, significant numbers of flea beetles and feeding damage were observed on the maize plants in the study field. In the spring, adult flea beetles were feeding on the lower 1–5 leaves of maize plants, and caused notable damage in all three years of the study. This taxon was therefore selected for more detailed sampling with Pherocon® AM yellow sticky traps in 2002–2003, although only the species list of flea beetles collected in 2002 has already been published (Kiss et al., 2003).

Since only a few studies (Grozea et al., 2006; Rauschen et al., 2010; Vrablova, 2002) have examined the species composition, dominance, and phenology of flea beetles in maize fields, the aim of our study was to complete and supplement these data.

Cry1Ab protein (MON810 event) shows high specificity to target organisms (*Ostrinia nubilalis*, *Sesamia nonagrioides*) under laboratory studies (Tier1), and variable mortality to larvae of numerous Lepidoptera species (MON810 SO Update, Perry et al., 2010, 2011) (Tier1a and b, Tier2) and by maize hybrids on the field (Tier3) (intended effect). However, only a few in-planta data (unintended effect) are available about flea beetles in connection with secondary metabolites and others (Bak et al., 1999; Nielsen et al., 2001; Verpoorte and Memelink, 2002). The further purpose of this study was to survey for possible un-intended effects of the MON810 maize on flea beetles.

2. Materials and methods

2.1. Experimental site

The two-year (2002 and 2003) field experiment was carried out in an isolated maize stand located near Budapest (GPS, N: 47° 25′; E: 18° 47′), Hungary, surrounded by large stone fruit orchards (apricot, peach and plum). Plots within the experimental field were arranged in a randomized complete block design with six replications. The plots (sized 30 × 30 m) were planted either with Bt maize (DK 440 BTY – transformation event MON810) or with its near isogenic line (DK 440) on chernozem soil. An alley distance of 3 m was used between replications. A retention zone (a pollen capture crop surrounding the entire test field) was established to a maize hybrid of similar maturity ground to the test hybrid, in accordance with the requirements of the release permit.

Maize was planted at a seed rate of 65,000 seeds/ha and was reduced to 50,000 plants/ha after emergence. Sowing was in late April and maize was harvested in mid-October to early November depending on the year.

2.2. Sampling

Flea beetle adults were collected with Pherocon AM yellow sticky traps (Trece Inc. California, USA), since yellow colour has long been known as attractive for flea beetles (Hung and Hwang, 2000; Vincent and Stewart, 1986). Three traps/plot were placed between rows 20 and 21, 7.5 m apart and 7.5 m from the field edge. Traps were fixed to a wooden stick at a height of maize canopy until silking, when they were adjusted to ear height. In 2002, traps were changed weekly, and according to the experiences of the first year, in order to reduce efforts and costs of sampling, in 2003 they were changed fortnightly. Sampling took place from late May to late September in 2002 and from early June to mid-September in 2003. Flea beetle adults were removed by petrol from the traps and submitted to I. Rozner (Museum of Natural History, Budapest) for identification. Collected adults were identified using the key of Warchalowsky (2003).

2.3. Statistical analysis

The catches of all three yellow sticky traps per plot were pooled for analysis. To compare the effect of different maize hybrids on flea beetle abundance and species richness, a repeated measures analysis of variance (ANOVA) was performed. Yearly comparisons were based on the samples collected at corresponding sampling dates (see ‘Maize hybrids’ versus ‘Sampling dates’); Welch's test was used for the analysis of the main effect of ‘Maize hybrids’ and the Greenhouse–Geisser test (with epsilon-adjusted degrees of freedom) was used for the trial factor (sampling dates) and for the ‘Maize hybrids’ × trial interaction effect. The total number of Alticinae species collected during each year was compared with a one-sample t test. A Welch test or (non-parametric) Brunner–Munzel test was performed for comparison of abundances across sampling dates. All statistical analyses were performed with the software package ROPstat (Vargha et al., 2015).

3. Results

Altogether, 51,348 flea beetle individuals from 26 species were captured by yellow sticky traps during 2002 and 2003 combined. In 2002, 18 flea beetle species were found in both Bt and isogenic maize plots. In 2003, 17 species were found in Bt maize and 14 species were found in isogenic maize (Tables 1 and 2). The highest species richness in 2002 was found from late June to mid-July, and, in 2003, from mid-June to late July. While species richness showed no difference between treatments, the number of captured species was significantly higher in Bt maize than in isogenic maize on 23 July 2002 (Welch $t = 2.739$, $df = 5.0$, $p = 0.041$) and on 18 June 2003 (Welch $t = 2.272$, $df = 9.1$, $p = 0.049$) (Fig. 1).

The most abundant flea beetle species, in all Bt- and isogenic maize plots in both years, were *Phyllotreta atra* and *P. vittula*. Together, these two species represented 99.3% and 99.5% of the total catch in 2002 and 2003 (Table 1). The highest trap catch of *P. atra* and *P. vittula* occurred between 18 June and 16 July depending on the year. In neither year was there a significant difference in abundance of *P. atra* or of *P. vittula* between the Bt and non-Bt maize varieties (Figs. 2 and 3). Neither was any difference observed for the other (non-*P. atra* and non-*P. vittula*) flea beetles (Fig. 4), although in 2002 the abundance of this group was significantly higher in Bt maize on 16 July (Welch $t = -3.213$, $df = 5.8$, $p = 0.019$) and significantly higher in isogenic maize on the 23rd of July (Welch $t = 2.739$, $df = 5.0$, $p = 0.041$) (Fig. 4) (Table 3 and Supplementary Table 1).

Table 1
Species composition of flea beetle assemblages on Bt and isogenic maize (plots) and the individual number of the species (Sóskút, Hungary, 2002–2003, June–September).

	Species/year	2002		2003	
		Bt	Iso	Bt	Iso
1	*Altica* sp.			2	3
2	*Aphthona* sp.			1	
3	*Chaetocnema aridula* (Gyllenhal)	3	3	8	11
4	*Chaetocnema concinna* (Marsham)	5	3	9	4
5	*Chaetocnema hortensis* (Geoffroy)	2	1	10	10
6	*Chaetocnema tibialis* (Illiger)	8	8	29	35
7	*Epitrix pubescens* (Koch)	2		3	
8	*Longitarsus exoletus* (L.)		3		
9	*Longitarsus melanocephalus* (De Geer)		2	2	
10	*Longitarsus obliteratus* (Rosenhauer)	1			
11	*Longitarsus parvulus* (Paykull)	1			
12	*Longitarsus pratensis* (Pancer)		2		
13	*Longitarsus rubiginosus* (Foudras)	3	6		1
14	*Longitarsus succineus* (Foudras)	2	3		
15	*Longitarsus* sp.	1	1	7	1
16	*Phyllotreta aenea* (Illiger)	1	1		
17	***Phyllotreta atra*** (F.)	**4909**	**3998**	**4153**	**3635**
18	*Phyllotreta balcanica* Heikertinger	3			
19	*Phyllotreta cruciferae* (Goeze)	1	3	3	3
20	*Phyllotreta diademata* Foudras		2	1	1
21	*Phyllotreta nemorum* (L.)	1	1	3	4
22	*Phyllotreta nigripes* (F.)	18	19		1
23	*Phyllotreta procera* (Redtenbacher)			1	3
24	*Phyllotreta undulata* Kutschera	3	7	6	
25	***Phyllotreta vittula*** (Redtenbacher)	**4135**	**3842**	**13,243**	**13,150**
26	*Psylliodes luteola* (O.F. Müller)			1	
	No. of species	**18**	**18**	**17**	**14**
	No. of individuals	**9099**	**7905**	**17,482**	**16,862**

Table 2
Mean number of *Phyllotreta atra*, *P. vittula* and other flea beetles (number of individuals in a plot per sampling date, SE). Mean species richness (number of flea beetle species collected in a plot per sampling date, SE) and total species richness (total number of flea beetle species per plot in 2002 or 2003, SE). There was no difference between the maize hybrids in any comparison. For statistical analyses, see Table 3.

	2002		2003	
	Bt	Iso	Bt	Iso
P. atra	68.2 (6.2)	55.5 (5.7)	98.9 (10.6)	86.6 (6.7)
P. vittula	57.4 (1.9)	53.4 (4.5)	315.3 (13.1)	313.1 (8.1)
Other species	0.8 (0.1)	0.9 (0.1)	2.1 (0.2)	1.9 (0.2)
Species rich./date	2.1 (0.1)	2.2 (0.1)	3.4 (0.1)	3.3 (0.1)
Total species rich.	8.3 (0.3)	8.2 (0.5)	9.3 (0.6)	8.5 (0.5)

4. Discussion

In this study species-rich and abundant flea beetle assemblages were found both in the Bt- (Cry1Ab) and in the isogenic maize plots. *Phyllotreta* species were dominant in maize in both years. We found *P. vittula* to be the most abundant species followed by *P. atra*, with the other flea beetles species captured only in small numbers (Table 1). Of the eight Alticinae species collected on maize by sweep netting in Slovakia by Vrablova (2002), all species except two (*Chaetocnema laevicollis* [Thomson] and *Longitarsus pellucidus* [Foudras]) were found in our study as well. *Phyllotreta vittula*, *Chaetocnema tibialis* and *P. atra* were the dominant species in the study in Slovakia, while *P. vittula* and *P. atra* were the dominant species in our study, in both Bt and non Bt maize. *Phyllotreta vittula*, *P. atra* and *Chaetocnema aridula* were also observed on maize in Romania (Grozea et al., 2006). *Chaetocnema tibialis* was also found on maize in both our study and in the USSR (Naibo, 1974). The maize is mentioned as food plant of *C. aridula* in Hungary (Sáringer, 1990) which was detected in our field study as well, but only in low number (Table 1).

According to previous observations in Hungary (Sáringer, 1990) *P. atra* feeds on cultivated and wild growing cruciferous plants, on *Reseda* and *Tropaeolum* spp., on *Cleome speciosissima* and pea (Balás and Sáringer, 1982). In summer emerged flea beetle adults maintain their fecundity by feeding on cruciferous crops and weeds as well (Sáringer, 1990). While no data is available on *P. atra* feeding on maize, high number of this species in the experimental maize field may be explained by the dispersion of the newly emerged adults from other crops.

In plant choice tests *P. vittula* preferred grasses (*Agropyron* species), but sufficient feeding and egg laying was observed on Brassicaceae and other Poaceae such as maize (Vig, 1998a). Earlier observations found that *P. vittula* disappeared from the maize fields from early-mid-July, but in our study it remained on the maize plants until the end of August in 2002 and until the middle of September in 2003 (Fig. 3). However, *P. atra* reached its lowest abundance much earlier, by the second decade of July and by the mid-August in 2002 and 2003, respectively (Fig. 2). Flight activity peaks can be explained by the summer appearance of the newly emerged adults (Foster, 1984; Hurej et al., 1997; Naibo, 1974; Sáringer, 1990), as the summer generation adults of *P. vittula* and *P. atra* emerged at the end of June or beginning of July (Kocourek et al., 2002; Naibo, 1974). At this time the young *P. vittula* adults dispersed from cruciferous plants to cereal crops including maize (Leclant, 1977). However, Vig (1998b) found that the newly emerged *P. vittula* adults appeared from late August to mid-September, significantly later than in other studies.

Phyllotreta atra was the most abundant in the first decade of July in both years, while *P. vittula* in mid-July in 2002, and from the mid-June to early July in 2003 (Figs. 2 and 3). These periods overlap with

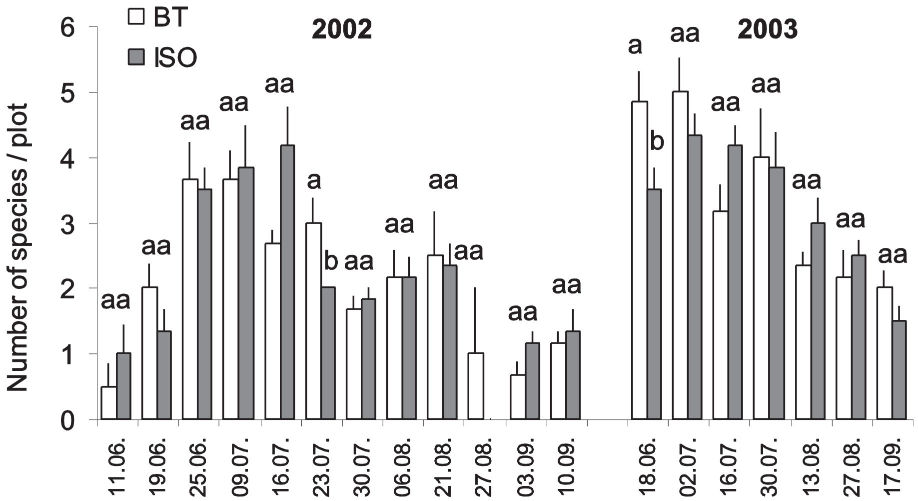

Fig. 1. Mean species richness (number of species/3 traps/plot) of flea beetles collected by yellow sticky traps in Bt (BT) and isogenic (ISO) maize plots in 2002–2003, at Sóskút, Hungary. Vertical lines indicate ± SE. Different letters within a date represents significant differences ($p < 0.05$).

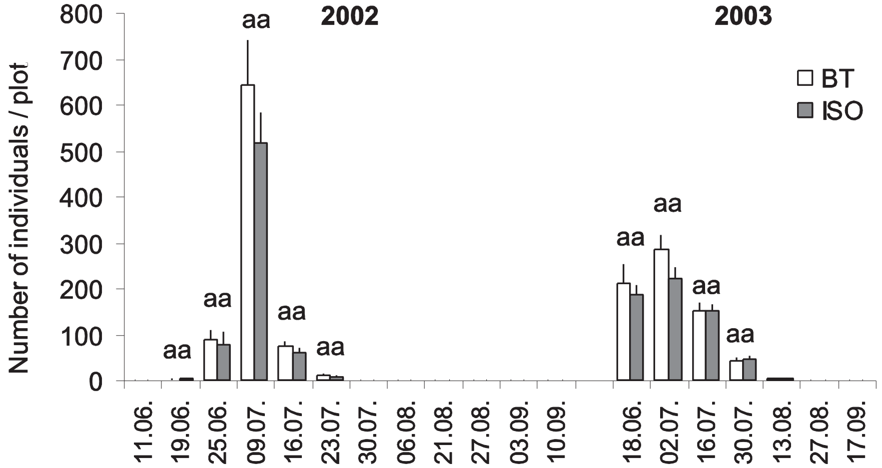

Fig. 2. Mean activity density (number of individuals/3 traps/plot) of *Phyllotreta atra*, collected by yellow sticky traps in Bt (BT) and isogenic (ISO) maize plots in 2002–2003, at Sóskút, Hungary. Vertical lines indicate ± SE. No differences between treatments on the same date were significant.

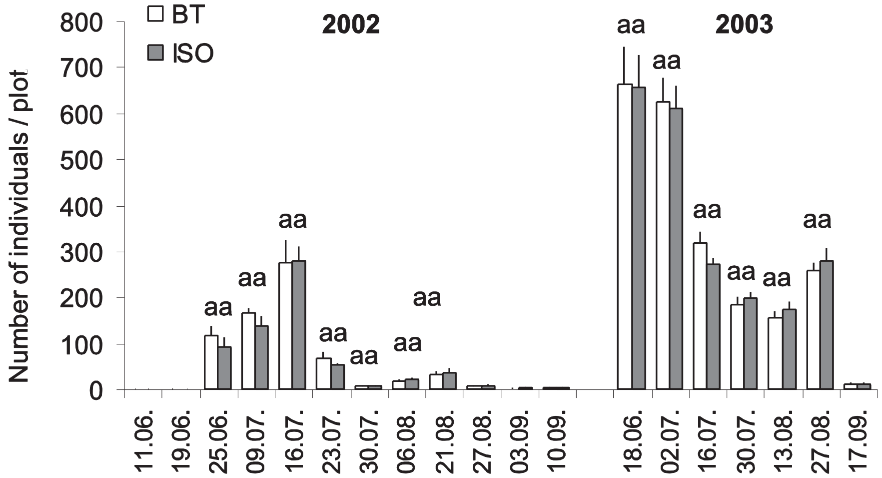

Fig. 3. Mean activity density (number of individuals/3 traps/plot) of *Phyllotreata vittula*, collected by yellow sticky traps in Bt (BT) and isogenic (ISO) maize plots in 2002–2003, at Sóskút, Hungary. Vertical lines indicate ± SE. No differences between treatments on the same date were significant.

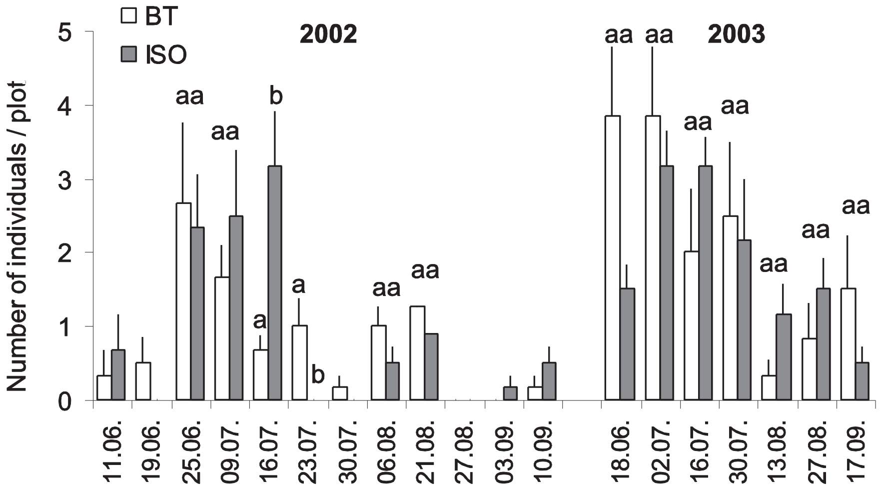

Fig. 4. Mean activity density (number of individuals/3 traps/plot) of Alticinae other than *P. atra* or *P. vittula*, collected by yellow sticky traps in Bt (BT) and isogenic (ISO) maize plots in 2002–2003, at Sóskút, Hungary. Vertical lines indicate ± SE. Different letters within a date represents significant differences ($p < 0.05$).

Table 3
Statistical analyses (repeated measures ANOVA; Welch test for the effect of 'maize hybrids', Geisser–Greenhouse test for the effect of 'sampling dates' and 'maize hybrids' × 'sampling date' interaction) of the abundance of *Phyllotreta atra*, *P. vittula*, other (non-*P. atra* and non-*P. vittula*) flea beetle species and species richness of flea beetles per sampling date. One sample t test for the total species richness of flea beetles. See also Table 2.

	Maize			Epsilon	Date			Maize × date		
	df	*F*	*p*		*df*	*F*	*p*	*df*	*F*	*p*
2002										
P. atra	1; 9.9	1.130	0.3129	0.1	1.1; 11.0	87.118	0.0000	1.10; 10.96	1.038	0.3384
P. vittula	1; 6.7	0.351	0.5729	0.170	1.9; 18.7	65.888	0.0000	1.87; 18.72	0.270	0.7525
Other species	1; 10.0	0.663	0.4344	0.270	3.0; 29.7	7.862	0.0005	2.97; 29.65	2.067	0.1265
Spec. rich./date	1; 9.4	0.211	0.6565	0.404	4.0; 40.4	16.606	0.0000	4.04; 40.37	1.518	0.2148
Total spec. rich.	10	0.194[a]	0.8501	–	–	–	–	–	–	–
2003										
P. atra	1; 8.4	0.833	0.3868	0.355	2.1; 21.3	87.044	0.0000	2.13; 21.28	1.152	0.3376
P. vittula	1; 8.3	0.018	0.8970	0.323	1.9; 19.4	73.534	0.0000	1.94; 19.36	0.180	0.8305
Other species	1; 8.4	0.833	0.3868	0.355	2.1; 21.3	87.044	0.0000	2.13; 21.28	1.152	0.3376
Spec. rich./date	1; 9.4	0.265	0.6187	0.594	3.6; 35.7	13.268	0.0000	3.57; 35.65	1.927	0.1337
Total spec. rich.	10	1.000[a]	0.3409	–	–	–	–	–	–	–

[a] *t* values (one sample t test).

the first observed damage by *P. vittula* in different maize fields in Hungary, namely from the end of June to early July (Nagy and Deseő, 1969) and in Romania from mid-May to early July (Grozea et al., 2006). In 2003 the abundance of *P. vittula* was much higher than in 2002 (Table 1, Fig. 3) which might be due to the drought, since a mass reproduction of the flea beetles can occur under such weather conditions (Sáringer, 1990).

We found no adverse effect of Bt-maize (MON810 event) on flea beetle assemblages or species, or on their abundance or species richness. In Germany, *Chaetocnema* spp., *Longitarsus* spp., *Phyllotreta* spp. and *Psylliodes chrysocephala* L. flea beetle species were collected by sweep net and *Phyllotreta* spp. by panicle samples in a 3-year field study that compared MON88017 Bt with near isogenic maize. Similar to our results, *P. vittula* was the most abundant flea beetle species and no significant difference was found between the Bt and non-Bt maize plots (Rauschen et al., 2010). In the same study, however, no flea beetle adults were observed with these sampling methods from MON810 and near isogenic maize (Nobilis) (Rauschen et al., 2010). *Chaetocnema pulicaria* was among the predominant herbivores in MON810 maize hybrid in Maryland, USA (Dively, 2005) and in Georgia (Daly and Buntin, 2005). In the Maryland study, in agreement with our findings, flea beetles showed no difference between Bt and non-Bt maize plots (Dively, 2005). In the Georgia study, the number of flea beetles was higher in Bt than non-Bt maize, suggesting that they were not adversely affected by the Bt maize (Daly and Buntin, 2005). While no specific mechanism is known that would lead us to expect that plant-incorporated Bt toxins would affect any non-target phytophagous arthropods without specific Bt protein binding sites, it is unclear whether the Bt effect on flea beetles in the study of Daly and Buntin (2005) was real or a statistical, or simply a sampling artefact.

Acknowledgements

This research was supported by EU-5th framework programme (Contract No: QLK3-CT-2000-00547) and by Research Centre of Excellence-9878/2015/FEKUT (Szent István University). The authors thank M. Perczel and Sóskút Fruct Kft. for technical support and to I. Rozner (Hungarian Natural History Museum Budapest) for identifications.

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.cropro.2015.07.008.

References

Andow, D.A., Lövei, G.L., Arpaia, S., 2006. Ecological risk assessment for Bt crops. Nat. Biotechnol. 24, 749–751.
Andow, D.A., Lövei, G.L., Arpaia, S., Wilson, L., Fontes, E.M.G., Hilbeck, A., Lang, A.,

Nguyễn, V.T., Pires, C.S.S., Sujii, E.R., Zwahlen, C., Birch, A.N.E., Capalbo, D.M.F., Prescott, K., Omoto, C., Zeilinger, A., 2013. An ecologically-based method for assessing the risks to biological diversity from genetically-engineered plants. J. Biosaf. 22, 141–156.

Arnoldi, L.W., Gurjeva, E.L., 1960. Coleoptera. In: Spravocsnik, Izd. Naszekomüe vredjascsie kukuruze v SSSR. AN SSSR. Moskva-Leningrad, pp. 115–160 (in Russian).

Arpaia, S., Messéan, A., Birch, N.A., Hokannen, H., Härtel, S., van Loon, J., Lovei, G.L., Park, J., Spreafico, H., Squire, G.R., Steffan-Dewenter, I., Tebbe, C., van der Voet, H., 2014. Assessing and monitoring impacts of genetically modified plants on agro-ecosystems: the approach of AMIGA project. Entomologia 2, 79–86.

Árpás, K., Tóth, F., Kiss, J., 2005. Foliage-dwelling arthropods in Bt-transegenic and isogenic maize: a comparison through spider web analysis. Acta Phytopathol. Entomol. Hung. 40, 347–353.

Bak, S., Olsen, C.E., Petersen, B.L., Møller, B.L., Halkier, B.A., 1999. Metabolic engineering of p-hydroxybenzylglucosinolate in *Arabidopsis* by expression of the cyanogenic CYP79A1 from *Sorghum bicolor*. Plant J. 20, 663–671.

Balás, G., Sáringer, Gy, 1982. Pests of Horticultural Plants. Akadémiai Kiadó, Budapest, pp. 488–490 (in Hungarian).

Balog, A., Kiss, J., Szekeres, D., Szénási, Á., Markó, V., 2010a. Rove beetle (Coleoptera: Staphylinidae) communities in transgenic Bt (MON810) and near isogenic maize. Crop Prot. 29, 567–571.

Balog, A., Szénási, Á., Szekeres, D., Kiss, J., 2010b. Staphylinids (Coleoptera: Staphylinidae) in genetically modified maize ecosystems: species densities and trophic interactions. IOBC/WPRS Bull. 52, 9–15.

Bohinc, T., Košir, I.J., Trdan, S., 2013. Glucosinolates as arsenal for defending Brassicas against cabbage flea beetle (*Phyllotreta* spp.) attack. Žemdirb. Agric. 100, 199–204.

Bourguet, D., Chaufaux, J., Micoud, A., Delos, M., Naibo, B., Bombarde, F., Marque, G., Eychenne, N., Pagliari, C., 2002. *Ostrinia nubilalis* parasitism and the field abundance of non-target insects in transgenic *Bacillus thuringiensis* corn *(Zea mays)*. Environ. Biosaf. Res. 1, 49–60.

Daly, T., Buntin, G.D., 2005. Effect of *Bacillus thuringiensis* transgenic corn for lepidopteran control on nontarget arthropods. Environ. Entomol. 34, 1292–1301.

Dively, G.P., 2005. Impact of transgenic VIP3A x Cry1Ab lepidopteran-resistant field corn on the nontarget arthropod community. Environ. Entomol. 34, 1267–1291.

EC, 2001. Directive 2001/18/EC of the European Parliament and of the Council of 12 March 2001 on the Deliberate Release into the Environment of Genetically Modified Organisms and Repealing Council Directive 90/220/EEC. L 106/1 of 4/17/2001.

EC, 2002. Council decision of 3 October 2002 Establishing Guidance Notes Supplementing Annex VII to Directive 2001/18/EC of the European Parliament and of the Council on the Deliberate Release into the Environment of Genetically Modified Organisms and Repealing Council Directive 90/220/EEC. L 280/27 of 10/18/2002.

Eizaguirre, M., Madeira, F., López, C., 2010. Effects of Bt maize on non-target lepidopteran pests. IOBC/WPRS Bull. 52, 49–55.

EFSA, 2010. Guidance on the environmental risk assessment of genetically modified plants. EFSA J. 8 (1879), 1–111.

EU, 2013. Commission Implementing Regulation (EU) No 503/2013 on applications for authorisation of genetically modified food and feed in accordance with Regulation (EC) No 1829/2003 of the European Parliament and of the Council and amending Commission Regulations (EC) No 641/2004 and (EC) No 1981/2006. OJL 157, 8.6.2013, 1–48.

Foster, G.N., 1984. Monitoring and forecasting outbreaks of turnip flea beetles in the west of Scotland, 1978–1983. In: Proc. Conf., Dundee Univ. 19–22 March, pp. 222–227.

Fritzsche, R., 1971. Pflanzenschädlinge. In: Käfer, Bd. 7. Neumann Verlag, Leipzig.

Grozea, I., Badea, A.M., Prunar, F., Adam, F., 2006. Diversity of maize pests and their damaging effect in an agroecosystem from the western side of the country. Agron. Res. Mold. 3, 15–22.

Gyulai, P., Garay, A., 1996. The appearance of new or almost forgotten old insect pests in Hungary. Növényvédelem 32, 175–179 (in Hungarian).

Harwood, J.D., Wallin, W.G., Obrycki, J.J., 2005. Uptake of Bt endotoxins by nontarget herbivores and higher order arthropod predators: molecular evidence from a transgenic corn agroecosystem. Mol. Ecol. 14, 2815–2823.

Hinfner, K., Papp, Zs, 1961. Pathogens and Pests on Cereals. In: Növényvédelmi Zsebatlasz 3–4. Mezőgazdasági Kiadó, Budapest (in Hungarian).

Hoffmann, M.P., Kirkwyland, J.J., Davis, P.M., 1995. Spatial distribution of adult corn flea beetles, *Chaetocnema pulicaria* (Coleoptera, Chrysomelidae), in sweet corn and development of a sampling plan. J. Econ. Entomol. 88, 1324–1331.

Hruska, J., 1962. Pests of maize. In: Monograph on Maize. Statni zemedel. naklad, Praha, pp. 667–727 (in Bohemian).

Hurej, M., Preiss, G., Debek, J., 1997. Species composition and occurrence of flea beetles on yellow mustard in Lower Silesia, Poland. Pol. Pismo Entomol. 66, 311–317 (In Polish).

Hung, C.C., Hwang, J.S., 2000. Influence of cylinder-type sticky traps baited with different mustard oil lures on *Phyllotreta striolata* (Coleoptera: Chrysomelidae). Chin. J. Entomol. 20, 201–214.

Jablonowski, J., 1906. Animal Pests of Sugar Beet. Cukorgyárosok Országos Egyesülete, Budapest (in Hungarian).

Kania, Cz, 1962. Szkodliwa entomofauna kukurydzy obserwowana w okolicach Wroclawia w latach 1956-59. Cześć II. Wroclaw Pol. Pismo Entomol. Ser. 3 (3–4), 183–217 (in Polish).

Kiss, J., Szentkirályi, F., Tóth, F., Szénási, Á., Kádár, F., Árpás, K., Szekeres, D., Edwards, C.R., 2003. Bt Corn: Impact on non-targets and adjusting to local IPM systems. In: Lelley, T., Balázs, E., Tepfer, M. (Eds.), Ecological Impact of GMO Dissemination in Agro-ecosystems. Facultas, Wien, pp. 157–172.

Kocourek, F., Láska, P., Jarošík, V., 2002. Thermal requirements for flight of six species of flea beetle of the genus *Phyllotreta* (Coleoptera: Chrysomelidae). Plant Prot. Sci. 38, 76–80.

Leclant, F., 1977. Pest control methods for maize in France. Ann. Appl. Biol. 87, 270–276.

Lozzia, G.C., 1999. Biodiversity and structure of ground beetle assemblages (ColeopteraCarabidae) in Bt corn and its effects on non target insects. Boll. Zool. Agrar. Bachic. II. 31, 37–58.

Lozzia, G.C., Rigamonti, I.E., 1998. Preliminary study on the effects of transgenic maize on nontarget species. IOBC/WPRS Bull. 21, 171–180.

Lövei, G.L., Andow, D.A., Arpaia, S., 2009. Transgenic insecticidal crops and natural enemies: a detailed review of laboratory studies. Environ. Entomol. 38, 293–306.

Lövei, G.L., Arpaia, S., 2005. The impact of transgenic plants on natural enemies: a critical review of laboratory studies. Entomol. Exp. App. 114, 1–14.

Mészáros, Z., Ádám, L., Balázs, K., Benedek, I.L., Draskovits, Á.D., Kozár, F., Lövei, G., Mahunka, S., Meszleny, A., Mihályi, K., Nagy, L., Papp, J., Papp, L., Polgár, L., Rácz, V., Ronkay, L., Soós, Á., Szabó, S., Szabóky, Cs, Szalay-Marzsó, L., Szarukán, I., Szelényi, G., Szentkirályi, F., 1984. Result of faunistical studies in Hungarian maize stands. Acta Phytopathol. Acad. Sci. Hung. 19, 65–90.

Musser, F.R., Shelton, A.M., 2003. Bt sweet corn and selective insecticides: their impacts on sweet corn pests and predators. J. Econ. Entomol. 96, 71–80.

Nagy, B., Deseő, K.V., 1969. Data on damages caused by *Phyllotreta vittula* Redtb. Coleopt. Halticinae. Növényvédelem 5, 97–104 (in Hungarian).

Naibo, B., 1974. Damage by the flea beetle *Phyllotreta vittula* Redt. on maize. Rev. Zool. Agric. Pathol. Veg. 73, 70–72.

Nielsen, J.K., Hansen, M.L., Agerbirk, N., Petersen, B.L., Halkier, B.A., 2001. Responses of the flea beetles *Phyllotreta nemorum* and *P. cruciferae* to metabolically engineered *Arabidopsis thaliana* with an altered glucosinolate profile. Chemoecology 11, 75–83.

Perry, J.N., Devos, Y., Arpaia, S., Bartsch, D., Gathmann, A., Hails, R.S., Kiss, J., Lheureux, K., Manachini, B., Mestdagh, S., Neemann, G., Ortego, F., Schiemann, J., Sweet, J.B., 2010. A mathematical model of exposure of non-target Lepidoptera to Bt-maize pollen expressing Cry1Ab within Europe. Proc. R. Soc. B Biol. Sci. 277, 1417–1425.

Perry, J.N., Devos, Y., Arpaia, S., Bartsch, D., Gathmann, A., Hails, R.S., Kiss, J., Lheureux, K., Manachini, B., Mestdagh, S., Neemann, G., Ortego, F., Schiemann, J., Sweet, J.B., 2011. The usefulness of a mathematical model of exposure for environmental risk assessment. Proc. R. Soc. B Biol. Sci. 278, 982–984.

Pilcher, C.D., Rice, M.E., Obrycki, J.J., Lewis, L.C., 1997. Field and laboratory evaluations of transgenic *Bacillus thuringiensis* corn on secondary lepidopteran pests (Lepidoptera: Noctuidae). J. Econ. Entomol. 90, 669–678.

Rauschen, S., Schaarschmidt, F., Gathmann, A., 2010. Occurrence and field densities of Coleoptera in the maize herb layer: implications for environmental risk assessment of genetically modified Bt-maize. Transgen. Res. 19, 727–744.

Sáringer, Gy, 1990. Flea beetles (Halticinae spp.). In: Jermy, T., Balázs, K. (Eds.), Handbook of Plant Protection Zoology 3/A. Akadémiai Kiadó, Budapest, pp. 296–307 (in Hungarian).

Scsegolev, V.N., 1952. Opregyeligyej naszekomüch po povrezsdennijam kulturnüh rasztenij. Szelhozgiz, Moskva-Leningrad (in Russian).

Scsegolev, V.N., 1951. Agricultural Entomology. Mezőgazdasági Kiadó, Budapest (in Hungarian).

Sehnal, F., Habuštová, O., Spitzer, L., Hussein, H.M., Růžička, V., 2004. A biannual study on the environmental impact of Bt maize. IOBC/WPRS Bull. 27, 147–160.

Sekulic, R., Camprag, D., Keresi, T., Tallósi, B., 1989. A contribution to the knowledge of some species of Coleoptera in corn fields in Yugoslavia. Acta Phytopathol. Entomol. Hung. 24, 189–193.

Sorauer, P., 1954. Handbuch der Pflanzenkrankhetiten. Berlin-Hamburg, Verlag für Landw. Gartenb. Forstwesen.

Steffey, K., Rice, M.E., All, J., Andrew, D.A., Gray, M.E., Van Duyn, J.W. (Eds.), 1999. Handbook of Corn Insects. Entomological Society of America, Lanham, MD, p. 164.

Szeőke, K., Gáborjányi, R., Kobza, S., Rátainé, Vida R., 1996. Plant protection of sweet corn. Növényvédelem 32, 459–466 (in Hungarian).

Szeőke, K., 1997. Damage of millet flea-beetle on spring barley and maize. Növényvédelem 33, 33–34 (in Hungarian).

Szűcs, J., 1973. Damage of barley flea beetle on maize. Magy. Mezőgazd. Inf. 48, 35 (in Hungarian).

Trdan, S., Vidrih, M., Valič, N., Laznik, Ž., 2008. Impact of entomopathogenic nematodes on adults of *Phyllotreta* spp. (Coleoptera: Chrysomelidae) under laboratory conditions. Acta Agric. Scand. Sect. B Soil Plant Sci. 58, 169–175.

Vargha, A., Torma, B., Bergman, L.R., 2015. ROPstat: a general statistical package useful for conducting person-oriented analyses. J. Pers. Oriented Res. 1, 87–98.

Verpoorte, R., Memelink, J., 2002. Engineering secondary metabolite production in plants. Curr. Opin. Biotechnol. 13, 181–187.

Vig, K., 1998a. Host plant selection by *Phyllotreta vittula* (Redtenbacher, 1849). In: Proceedings of the Fourth International Symposium on the Chrysomelidae. Proc. 4th Int. Symp. Chrysomelidae. Proc. Symp. Firenze, 1996 Italy, XX Int. Congr. Entomol., Torino, Mus. Reg. Sci. Nat., pp. 233–251.

Vig, K., 1998b. Data on the biology of *Phyllotreta vittula* (Redtenbacher, 1849) (Coleoptera: Chrysomelidae: Alticinae). Meded. Fac. Landbouwkd. Toegep. Biol. Wet. Univ. Gent. 63, 357–363.

Vincent, C., Stewart, R.K., 1986. Influence of trap color on captures of adult crucifer-feeding flea beetles. J. Agric. Entomol. 3, 120–124.

Vörös, G., Maros, P., 2004. Droughty 2003 – grave plant protection problems in maize fields of county Tolna. Növényvédelem 40, 287–292 (in Hungarian).

Vrablova, M., 2002. Herbivorous Insect Species Associated with Pigweeds (Amaranthus spp.) in Slovakia and Their Potential Exploitation in Biological Control (Ph.D. thesis). Slovak Agricultural University, Nitra.

Warchalowsky, A., 2003. The Leaf-beetles of Europe and the Mediterranean Area. Natura optima dux Foundation, Warszawa.

Wilde, G., Roozeboom, K., Claassen, M., Janssen, K., Witt, M., 2004. Seed treatment for control of early-season pests of corn and its effect on yield. J. Agric. Urban Entomol. 21, 75–85.