Non Target Effect of Cry1 Ab and Cry Ab x Cry3 Bb1 Bt Transgenic Maize on Orius insidiosus (Hemiptera: Anthocoridae) Abundance

Santiago A. Palizada¹,², Difabachew K. Belay¹,³, Bamphitlhi Tiroesele¹, Fatima Mustafa⁴, Muhammad Irfan Ullah⁵, Thomas Hunt⁶, Jaime Molina-Ochoa⁷,², Steven R. Skoda⁷,², Pete L. Clark¹ and John E. Foster¹

¹Department of Entomology, Insect Genetics Laboratory, University of Nebraska-Lincoln, 103 Entomology Hall, Lincoln, NE 68583-0816, USA
²Dow Agro Sciences, 18078 N 1500 E Road, Pontiac, IL 61764, USA
³University of Nebraska, Haskell Agricultural Laboratory, 57905 866 Road, Concord, NE. 68728, USA
⁴Simplot -Manager Regulatory Affairs, 5369 W. Irving Street, Boise, Idaho 83706, USA
⁵Department of Agriculture-Bureau of Plant Industry, 692 San Andres S.t., Malate, Manila 1004, Philippines
⁶Universidad de Colima, Coordinación General de Investigación Científica, Centro Universitario de Investigación y Desarrollo Agropecuario, Km 40 autopista Colima-Manzanillo, Tecomán, Colima 28930, México
⁷USDA-ARS-KBUSLIRL Screwworm Research Unit, Kerrville, TX 78028, USA

Abstract

Non-target effects of Cry1 Ab and Cry Ab x Cry3 Bb1 Bt transgenic maize hybrids on Orius insidiosus (Sayi) was studied in Nebraska (Mead, Clay Center, and Concord) during 2007 and 2008. The Bt effect was compared to conventional maize (isoline), conventional maize, and insecticide applications of permethrin (Pounce® 1.5G) and bifenthrin (Capture® 2EC) to control first and second generations of Ostrinia nubilalis (Hübner), respectively. Yellow sticky cards, visual observations, and destructive samplings were used to evaluate O. insidiosus abundance. In 2007 the O. insidiosus abundance was lower on Pounce® 1.5G treated non-Bt isoline maize plots compared to the BT transgenic hybrids at 60 and 90 days after planting (DAP). From visual observations, numbers of O. insidiosus were lower in Pounce® 1.5G treated plots and no adverse effects of the Bt hybrids was observed. In 2008, no significant differences were found among treatments in the sticky card data, but the O. insidiosus population significantly increased, with increasing DAP, where the lowest and highest numbers were recorded at 30 and 120 DAP, respectively. In the visual observation and destructive samplings, numbers of O. insidiosus were lower at Concord compared to other sites. Results from the visual observation data in 2008 also revealed that O. insidiosus abundance was lower on Pounce® 1.5G treated plots compared to other treatments. This study showed no adverse effects of the new BT transgenic hybrids that included stacked resistance genes on O. insidiosus compared to the non-Bt maize hybrids.

Keywords: Orius insidiosus; Non-target effects; BT transgenic maize

Introduction

Insect resistance based on Bacillus thuringiensis (Bt) (Berliner) endotoxins is the most widely used trait following herbicide tolerance in commercial transgenic crops [1]. The deployment of transgenic plants resistant to insects offered expectations as a means of pest control that led to a reduction in pesticide use in intensive cropping systems.

Although the increased global adoption of transgenic crops [2] shows usefulness for many growers and their acceptance in many markets, the imposition of moratoria in several countries reflects skepticism and public concern about a range of issues around transgenics including potential impacts on the environment. Potential adverse effects of transgenics on the environment include effects on non-target species, invasiveness, release or “escape” into the environment, and development of resistance to transgenic products [3]. To address these concerns, governments have authorized regulatory bodies like the U.S. Environmental Protection Agency to regulate the deployment of transgenics requiring environmental risk assessment data as part of the registration process [4].

Orius insidiosus (insidiosus flower bugs) (Hemiptera: Anthocoridae) are generatilist predators which are frequently reported in ecological studies as important non-target organisms in transgenic maize [5-8]. In the Midwest, including Nebraska, O. insidiosus is a common predator in maize (Wright 2004) and soybean fields [9].

Orius spp. are important natural enemies of pest insects and mites in many cropping systems such as maize, soybeans, vegetables, and fruit crops [10,11]. Nearly all Orius spp. are preaceous as nymphs and adults. The primary food of Orius spp. consists of small insects and insect eggs, plant pollen, and plant sap [12]. Nymphs and adults of O. insidiosus are commonly found on maize silks and serve as natural enemies of key maize pests such as of Ostrinia nubilalis (Hübner), Helicoverpa zea (Hübner) [13], Spodoptera frugiperda (J.E. Smith) [14], Rhopalosiphum maidis (Fitch) [9,15,16], Frankliniella sp. [12,17,18], spider mites, white flies (Bemisia spp.), and eggs of other insects in the field. O. insidiosus are commercially mass produced and sold as biocontrol agents against pests of glasshouse- grown vegetables and ornamental crops [19].

The potential non-target impact of transgenic maize was studied using O. insidiosus as a key non-target arthropod [5,8,20]. Effective and reliable sampling of O. insidiosus nymphs and adults is important for genetic monitoring of transgenic crops.
in assessing the impact of transgenic corn on non-target organisms, particularly for environmental risk assessments. Previous ecological studies have assessed the non-target effects of transgenic maize by using visual observations, pitfall traps, sticky cards, sweep nets, and beat buckets [7,8,21,22].

Non-destructive (visual observations), yellow sticky card s, and destructive sampling techniques have been used to monitor O. insidiosus nymphs and adults together with aboveground arthropods pests for the non-target impact of transgenic plants [5,6,8,22,23]. These techniques were used to approach the objective of this study, to evaluate and destructive sampling techniques have been used to monitor O. insidiosus abundance compared to insecticide applications and non-transgenic maize.

Materials and Methods

Experimental sites and description

The experiments were conducted during 2007 and 2008 at three geographically different experimental research stations of University of Nebraska-Lincoln. The experimental fields were located at the Agricultural Research and Development Center, Mead, (N41°11.09’ W96°27.411’ in 2008), South Central Agricultural Laboratory, Clay Center, (N40°34.216’ W98°07.958’ in 2007 and N40°34.272’ W98°07.822’ in 2008), and the Northeast Research and Extension Center, Haskell Agricultural Laboratory, Concord, (N42°23.037’ W96°57.193’ in 2007 and N42°23.149’ W96°57.331’ in 2008). Soil types were Sharpsburg silty clay loam, Kennebec silty clay loam, and Butler/Crete silt loam, respectively. The experimental fields at all locations were previously planted with soybeans in a no-tillage system.

Agronomic practices

Plantings were done in a no-till corn system on 10, 11 and 15 May in 2007, and during 19, 20 and 21 May in 2008 at Mead, Clay Center, and Concord, respectively. Fertilizer management, irrigation, and herbicide application were made based on the normal agronomic recommendations of each specific site.

Experimental design and treatments

A randomized complete block design with four replications was used. The treatments were: a) Cry1Ab X CP4 EPSPS maize, b) CP4 EPSPS maize (isoline), c) CP4 EPSPS maize (isoline) plus an insecticide application to control the first generation of O. nubilalis, d) Cry1Ab+Cry3Bb1X CP4 EPSPS maize, e) CP4 EPSPS maize (isoline) plus an insecticide application to control second generation of O. nubilalis, and f) a conventional maize without insecticide application. The Cry1Ab Bt transgenic maize is used to control lepidopteran pests while Cry3Bb1X is used against corn root worms (Diabrotica spp.). The CP4 EPSPS is a genetically engineered glyphosate tolerant maize variety which allows the use of glyphosate as a postemergence herbicide.

In the case of CP4 EPSPS maize plus an insecticide application to control the first generation of O. nubilalis both in 2007 and 2008, permethrin (Pounce® 1.5G) (FMC Corporation, PA) was applied at the recommended rate of 12 oz. /1000 row ft band using an improvised jar shaker applicator at whorl maize stage (V9-V12 growth stages). Bifenithrin (Capture® 2 EC) (Bayer, NJ) was sprayed at the rate of 6.66 ml/ 2 gallons of water using a carbon-gated sprayer for the control of second generation O. nubilalis. Individual plots were 60 square meters. There were 8 rows in each plot with ~400 plants per plot (~50 plants per row). A 3 m spacing between treatments and blocks was planted with conventional corn hybrid.

Sampling methods

O. insidiosus nymphs and adults were monitored using visual observations, and adults with yellow sticky cards, in 2007 and 2008. A destructive sampling technique was added in 2008 to validate the actual nymph and adult counts. Visual observations were made on 20 randomly selected plants from rows 2 and 3 in each plot at reproductive stages, R1 (silking) and R2 (blister) i.e. 80 and 90 DAP, respectively. Nymphs and adults of O. insidiosus were observed on maize ears, and silks were tapped and O. insidiosus falling from the silk were collected with a clean sheet of bond paper underneath to quantitate the number of nymphs and adults. The mean nymph plus adult counts per plant were used for the analysis.

Two yellow sticky cards (23 x 28 cm) per plot (sticky on one side only) (Pherocon® AM, Trécé Inc., Adair, OK) [7,8] were used. The traps were attached to wooden stakes (2.5 x 2.1 x 244 cm) that were placed between rows 5 and 6 and 7 of each plot at the seedling stage (V3). The yellow stick cards were attached on the wooden stakes at 30, 60, 90, and 120 DAP. The cards were folded and clipped with 2 binder clips around the wooden stake facing the maize rows at the canopy level during the vegetative stage and parallel to the ears in the reproductive stages. After 7 days, the yellow sticky cards were collected, sealed in ziplock plastic bags, and brought to the laboratory for quantification. O. insidiosus adults were counted with the aid of a dissecting microscope. The adult counts from the 2 yellow sticky cards were pooled, and mean adult counts per card per day were used for the analysis.

Destructive sampling was done on five randomly selected maize ears from row 4 of each plot at R2. The randomly sampled maize ears were cut from the plant using a knife and kept in a ziplock plastic bag separately and brought to the laboratory for counting. Adults and nymphs of O. insidiosus were counted using a dissecting microscope. Mean number of nymphs and adults of the five ears per plot were pooled for the analysis. Voucher specimens of O. insidiosus were kept at University of Nebraska-Lincoln, Department of Entomology.

Data analysis

Analysis of variance (ANOVA) was performed using SAS's PROC GLM procedure (SAS, 2003) [24]. The level of significance was set at P=0.05. Whenever there was significant interaction among factors (treatment, sampling period, location, season), each factor was analyzed with respect to the levels of the other factor. In the absence of significant interaction, data were pooled. The treatment x site effects generally revealed no significant differences, and these were not presented in the results and discussion. For parameters that showed significant difference among treatments, individual means were separated using the Student’s Newmean Keuls test (SNK).

Results

There was a significant interaction in 2007 between treatments and sampling period for the yellow sticky card data (F=2.29, P=0.0050, df=15,216), so treatments were compared at a specific sampling period. Abundance of O. insidiosus also significantly varied among locations (F=16.72, P<0.0001, df=2,216). Significant differences among treatments were observed at 60 DAP (F=3.48, P=0.0076, df=5, 64) and 90 DAP (F=4.26, P=0.0117, df=5, 64); numbers of O. insidiosus were significantly lower on Pounce® 1.5G treated CP4 EPSPS maize (isoline) compared to the rest of the treatments including the transgenic hybrids.
from each other (SNK, P = 0.05). Means within a column followed by the different letter are significantly different from each other (SNK, P = 0.05). Ns = not significant. Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize

Table 1: Mean number of O. insidiosus (± S E) in BT transgenic maize hybrids and non transgenic insecticide treated and non-treated hybrids during the 2007 cropping season.

| Treatment | Sampling periods (days after planting) |
|-----------|----------------------------------------|
|           | 30 days | 60 days | 90 days | 120 days |
| CP4 EPSPS maize | 0.05 ± 0.02 | 0.88 ± 0.15a | 0.81 ± 0.11a | 1.14 ± 0.09 |
| Pounce® 1.5G | 0.05 ± 0.02 | 0.46 ± 0.09b | 0.45 ± 0.07b | 1.57 ± 0.18 |
| Cry1Ab x Cry3Bb1 | 0.018 ± 0.01 | 0.75 ± 0.13ab | 0.77 ± 0.09a | 1.28 ± 0.27 |
| Capture® 2 EC | 0.036 ± 0.01 | 0.93 ± 0.19a | 0.82 ± 0.12a | 1.17 ± 0.18 |
| Conventional corn | 0.01 ± 0.01 | 0.65 ± 0.12ab | 0.85 ± 0.08a | 1.19 ± 0.16 |

Means within a column followed by the same letter are not statistically different from each other (SNK, P = 0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.

Table 2: Mean number of O. insidiosus (± S E) in BT transgenic maize hybrids and non transgenic insecticide treated and non-treated hybrids during the 2008 cropping season.

| Experimental Site | Season |
|-------------------|--------|
|                   | 2007               | 2008               |
|                   | Sticky card | Visual observation | Sticky card | Visual observation | Destructive sampling |
| Clay Center       | 0.94 ± 0.07a   | 0.25 ± 0.03c       | 0.66 ± 0.08b | 1.74 ± 0.73a       | 2.93 ± 0.20a       |
| Concord           | 0.70 ± 0.05b   | 0.77 ± 0.05a       | 0.83 ± 0.13a | 0.37 ± 0.03b       | 0.66 ± 0.11c       |
| Mead              | 0.53 ± 0.05c   | 0.58 ± 0.04b       | 0.54 ± 0.06b | 1.63 ± 0.16a       | 1.81 ± 0.13b       |

Means within a column followed by the different letter are significantly different from each other (SNK, P = 0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.

Figure 1: Mean number of O. insidiosus (± SE) in BT transgenic maize hybrids and non-transgenic insecticide treated and non-treated hybrids during the 2007 cropping season in Nebraska. Bars followed by different letter are significantly different from each other (SNK, P=0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.

During the 2008 cropping season, the sticky card data showed no significant differences among treatments (F=2.13, P=0.0624, df=5,274). However, there was significant differences among sampling periods (F=255.56, P<0.0001, df=3,216) and locations (F=9.17, P<0.0001, df=2,216). O. insidiosus abundance significantly increased with DAP and the highest (1.95 O. insidiosus per sticky card per day) was recorded at 120 DAP and no O. insidiosus recorded at 30 DAP (Figure 2).

During the 2008 cropping season, the sticky card data showed no significant differences among treatments (F=54.4, P<0.0001, df=5, 54). Similar to the sticky card data, O. insidiosus populations were significantly lower on Pounce® 1.5G treated plots than the rest of the treatments (Figure 1). Moreover, data not shown adverse effect of the Bt transgenic hybrids compared to the isoline and conventional counterparts (Figure 1).

Citation: Palizada SA, Belay DK, Tiressele B, Mustafa F, Ullah MI, et al. (2013) Non Target Effect of Cry1 Ab and Cry Ab x Cry3 Bb1 Bt Transgenic Maize on Orius insidiosus (Hemiptera: Anthocoridae) Abundance. Entomol Ornithol Herpetol 2: 107. doi:10.4172/2161-0983.1000107

Figure 2: Mean number of O. insidiosus (± SE) in BT transgenic maize hybrids and non-transgenic insecticide treated and non-treated hybrids at different sampling periods using a yellow sticky card trapping method during the 2008 cropping season in Nebraska. Bars followed by different letter are significantly different from each other (SNK, P=0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.

Figure 3: Abundance of O. insidiosus (mean ± SE) in BT transgenic maize hybrids and non-transgenic insecticide treated and non-treated hybrids during the 2008 cropping season in Nebraska using a visual observation sampling technique. Bars followed by the same letter are not statistically different from each other (SNK, P>0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.
In the visual observations in 2008, there was a significant three-way interaction among sampling periods, locations (sites), and treatments (F=18.23, P<0.0001, df=12,108). Therefore, treatments were compared for each location separately at a specific sampling period. At Clay Center, significantly lower numbers of *O. insidiosus* were recorded from Pounce® 1.5G treated plots compared to the other treatments (Table 3). Similarly, at Concord, abundance of *O. insidiosus* was significantly higher in Cry1Ab and Cry1Ab X Cry1Bb hybrid compared to the non-transgenic isolate treated with Pounce® 1.5G to control first generation of *O. nubilalis*. Moreover, at 90 DAP, a higher number of *O. insidiosus* was recorded from Capture® 2 EC treated plots compared to Pounce® 1.5G treated plots (Table 3). At Mead, *O. insidiosus* abundance was also significantly lower in Pounce® 1.5G treated plots than the other treatments at 80 DAP, and there were no significant differences among treatments at 90 DAP (F=0.87, P=0.5263, df=5,15). The overall treatment effect in the visual observations of 2008 season indicated that significantly lower numbers of *O. insidiosus* were recorded from Pounce® 1.5 G treated isolate than other treatments including the Bt transgenic hybrids (Figure 3). Moreover, *O. insidiosus* abundance showed a similar trend in the destructive sampling where Orius counts were significantly lower in Pounce® 1.5G treated plots than the Bt transgenic hybrids, the non-transgenic isolate, conventional maize, and Capture® 2 EC sprayed conventional maize (Figure 4).

**Discussion**

Visual observations, yellow sticky cards and destructive sampling techniques revealed the same trend of significantly fewer mean adult counts of *O. insidiosus* on CP4 EPSPS maize plus Pounce® 1.5G for the control of first generation *O. nubilalis* at R2 (blister) stage. Neither Bt transgenic maize hybrids had observable effects on populations of *O. insidiosus* in all sampling techniques used in the study. *O. insidiosus* nymphs and adults were fewer on insecticide treated CP4 EPSPS maize. These findings support previous ecological studies on non-target predators that transgenic maize does not have a significant negative effect on the predator *O. insidiosus*, but our results differ with those previously reported because we obtained significant differences in the sampling techniques [7,8,20,22,25,26].

The results of our study suggested that visual observation, yellow sticky cards, and destructive sampling are effective in monitoring abundance of *O. insidiosus* in non-target studies. These results corroborate other ecological field studies on non-target arthropods of transgenic maize. Al-deeb et al. [5] used visual counts of *O. insidiosus* in Bt and non-Bt maize fields at three locations in Kansas to show that Bt maize does not have significant effects on *O. insidiosus*. Musser et al. [7] also recommended the use of field counts of immature and adults, because these counts are accurate, have no associated supply costs, and can be made quickly. In a similar study using yellow sticky cards, Pilcher et al. [8] showed that significantly higher numbers of adult *O. insidiosus* preferred the early planting date of Bt hybrids during the first *O. nubilalis* generation. The variation in *O. insidiosus* population abundance among the three sites may be due to slight variation in biotic and abiotic factors [22]. Moreover, development of *O. insidiosus* is very dependent on temperature [12], and availability of food supply [19,27].

In conclusion, our findings support non-target arthropod ecological

![Figure 4: Abundance of *O. insidiosus* (mean ± SE) in BT transgenic maize hybrids and non-transgenic insecticide treated and non-treated hybrids during the 2008 cropping season in Nebraska using a destructive sampling technique. Bars followed by the same letter are not statistically different from each other (SNK, P<0.05). Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.](image-url)

| Treatments                  | Clay Center | Concord | Mead    |
|-----------------------------|-------------|---------|---------|
|                             | 80 days     | 90 days | 80 days | 90 days | 80 days | 90 days |
| Cry1Ab                      | 2.06 ± 0.19a| 1.09 ± 0.16b| 0.34 ± 0.03a| 0.53 ± 0.13ab| 2.18 ± 0.27a| 0.76 ± 0.13b|
| CP4 EPSPS Maize             | 2.00 ± 0.31a| 1.88 ± 0.04a| 0.19 ± 0.05ab| 0.44 ± 0.14ab| 2.85 ± 0.44a| 0.75 ± 0.19a|
| Pounce® 1.5G               | 0.85 ± 0.22b| 1.00 ± 0.12b| 0.09 ± 0.02ab| 0.23 ± 0.06a| 1.29 ± 0.27c| 0.51 ± 0.19a|
| Capture® 2 EC               | 2.54 ± 0.36a| 1.88 ± 0.08a| 0.30 ± 0.07ab| 0.74 ± 0.11a| 3.33 ± 0.09a| 0.91 ± 0.17a|
| Cry1Ab x Cry3Bb1            | 2.30 ± 0.15a| 1.71 ± 0.04a| 0.34 ± 0.07a| 0.49 ± 0.12ab| 2.70 ± 0.38a| 0.74 ± 0.04a|
| Conventional Maize          | 1.94 ± 0.17a| 1.63 ± 0.19a| 0.29 ± 0.08ab| 0.51 ± 0.04ab| 2.94 ± 0.19a| 0.73 ± 0.07a|

Means within a column followed by the same letter are not statistically different from each other (SNK, P=0.05). Ns = not significant. Pounce® 1.5G and Capture® 2 EC was applied on CP4 EPSPS maize.

**Table 3:** Abundance of *O. insidiosus* (mean ± S E) in BT transgenic maize hybrids and non transgenic insecticide treated and non-treated hybrid s at C lay Center, Concord, and Mead in Nebraska during the 2008 cropping season using a visual observation sampling technique.

Citation: Palizada SA, Belay DK, Tirosele B, Mustafa F, Ullah MI, et al. (2013) Non Target Effect of Cry1 Ab and Cry Ab x Cry3 Bb1 Bt Transgenic Maize on *Orius insidiosus* (Hemiptera: Anthocoridae) Abundance. Entomol Ornithol Herpetol 2: 107. doi: 10.4172/2161-0883.1000107
field studies that Cry1Ab, and Cry1Ab+Cry3Bb1 maize have no impact on O. insidiosus populations. However, the pyrethroid insecticide (Pounce® 1.5G) applications to control target pests significantly affected non-target natural enemies of the target pests.

Acknowledgements

We would like to thank Bill McCormick, Terry DeVries, Gerald Echtenkamp, Karl Brauer, Rosana Serikawa, Erica Lindroth, Kanhoporn Tangtrakulwanich, and summer student technicians for assistance during the corn growing seasons. We also thank the Fulbright-Philippine Agriculture Scholarship Program, Department of Agriculture-Bureau of Plant Industry, Monsanto Company, and the University of Nebraska-Lincoln for their financial assistance.

References

1. O’Callaghan M, Glare TR, Burgess EP, Malone LA (2005) Effects of plants genetically modified for insect resistance on nontarget organisms. Annu Rev Entomol 50: 271-292.
2. James C (2008) Global status of commercialized biotech/GM crops: 2007. Ithaca, NY:ISAAA. Retrieve at http://www.org/main.htm. Accessed on June 08, 2008.
3. Conner AJ, Glare TR, Nap JP (2003) The release of genetically modified crops into the environment. Part II. Overview of ecological risk assessment. Plant J 33: 19-46.
4. Naranjo S, Head G, Dively GP (2005) Impact of transgenic VIP3A x Cry1Ab lepidopteran-resistant field corn on the non-target arthropod community. Environmental Entomology 34: 1267-1291.
5. Al-deeb AM, Wilde GE, Higgins R (2001) No effect of Bacillus thuringiensis corn and Bacillus thuringiensis on predator Orius insidiosus (Hemiptera: Anthocoridae). Environmental Entomology 30: 625-629.
6. Jasinski JR, Eisley JI, Young CE, Kovach J, Willson H (2003) Select non-target arthropod abundance in transgenic and non-transgenic field crops of Ohio. Environmental Entomology 32: 407-413.
7. Musser FR, Nyrop JP, Shelton AM (2004) Survey of predators and sampling method comparison in sweet corn. J Econ Entomol 97: 136-144.
8. Pitcher CD, Rice ME, Obrycki JJ (2005) Impact of transgenic Bacillus thuringiensis corn and crop phenology on five non-target arthropods. Environmental Entomology 34: 1302-1316.
9. Brosius TR, Higley LG, Hunt TE (2007) Population dynamics of soybean aphid and biotic mortality at the edge of its range. J Econ Entomol 100: 1268-1275.
10. Jarvis JL, Guthrie WD (1987) Ecological studies of the European corn borer (Lepidoptera: Pyralidae) in Boone County, Iowa. Environmental Entomology 16: 50-58.
11. Bush L, Kring TJ, Ruberson JR (1993) Suitability of greenbugs, cotton aphids and Heliothis virescens eggs for development and reproduction of Orius insidiosus. Environ. Exp. Appl. 67: 217-222.
12. Ruudavets J (1995) Predators of Frankliniella occidentalis (Perg.) and Thrips tabaci Lind: a review. In: A.J.M. Loomans, J.C. van Lenteren, M.G. Tommansi, S. Maini, J. Ruudavets (eds.), Biological Control of Thrips Pests. Wageningen, pp. 43-87.
13. Wright RJ (2004) Minute pirate bugs. Biological Control News formerly Midwest Biological Control News. Vol. 1: 1. http://www.entomology.wisc.edu/mbcn/mbcn.html
14. Isenhour DJ, Layton RG, Wiseman BR (1990) Potential of adult Orius insidiosus (Hemiptera: Anthocoridae) as a predator of the fall armyworm, Spodoptera frugiperda (Lepidoptera: Noctuidae). BioControl 35: 269-275.
15. Fox TB, Landis DA, Cardoso FF, DiFonzo CD (2004) Predators suppress Aphis glycines Matsumura population growth in soybean. Environmental Entomology 33: 608-618.
16. Rutledge CE, O’Neil RJ, Fox TB, Landis DA (2004) Soybean aphid predators and their use in integrated pest management. Ann. Entomol. Soc. Am. 9: 240-248.
17. Baez I, Reitz SR, Funderburk JE (2004) Predation by Orius insidiosus (Heteroptera: Anthocoridae) on the life stages and species of Frankliniella flower thrips (Thysanoptera: Thripidae) in pepper flowers. Environmental Entomology 33: 662-670.
18. van den Meiracker RAF, Ramakers PMJ (1991) Biological control of the western flower thrips Frankliniella occidentalis, in sweet pepper, with the anthocorid predator Orius insidiosus. Med. Fac. Landbouw. Rijksuniv.Gent. 56:241-249.
19. Copping LG (2004) A World Compendium: The Manual of Biocontrol Agents. 3rd Ed. Of the BioPesticide Manual. BCPC Pub. Hampshire, UK, 702pp.
20. Ahmad A, Wilde GE, Whitworth RJ, Zolnerowich G (2006) Effect of corn hybrids expressing the coleopteran-specific cry3Bb1 protein for corn rootworm control on aboveground insect predators. J Econ Entomol 99: 1085-1095.
21. De la Pozza M, Pons X, Farinos GP, Lopez C, Ortego F, et al. (2005) Impact of farm-scale Bt maize on abundance of predatory arthropods in Spain. Crop Prot. 24: 667-684.
22. Head G, Moar W, Eubanks M, Freeman B, Ruberson J, et al. (2005) A multyear, large-scale comparison of arthropod populations on commercially managed Bt and non-Bt cotton fields. Environmental Entomology 34: 1267-1286.
23. Dively GP (2005) Impact of transgenic VIP3A x Cry1Ab lepidopteran-resistant field corn on the non-target arthropod community. Environmental Entomology 34: 1267-1291.
24. Copping LG (2004) A World Compendium: The Manual of Biocontrol Agents. 3rd Ed. Of the BioPesticide Manual. BCPC Pub. Hampshire, UK, 702pp.
25. Ahmad A, Wilde GE, Whitworth RJ, Zolnerowich G (2006) Effect of corn hybrids expressing the coleopteran-specific cry3Bb1 protein for corn rootworm control on aboveground insect predators. J Econ Entomol 99: 1085-1095.
26. Rose R, Dively GP (2007) Effects of insecticide-treated and Lepidopteran-active Bt transgenic sweet corn on the abundance and diversity of arthropods. Environ Entomol 36: 1254-1268.
27. Sabelis MW, van Rijn PCU (1997) Predation by Insects and Mites. In “Thrips as Crop Pests” (T. Lewis, Ed.). CAB-International, London, pp. 259-354.