Bulletin of Insectology **66** (2): 297-307, 2013
ISSN 1721-8861

Distribution and abundance of species of the genus *Orius* in horticultural ecosystems of northwestern Italy

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Abstract

The genus *Orius* Wolff includes generalist predators revealed to be very effective in thrips control worldwide. During the years 2005-2007, *Orius* species were sampled on some horticultural crops (strawberry, sweet leek, and sweet pepper), and on wild flora surrounding crops to identify alternative host plants in Piedmont, northwestern Italy. In the three-year survey, *Orius horvathi* (Reuter), *Orius laevigatus* (Fieber), *Orius majusculus* (Reuter), *Orius minutus* (L.), *Orius niger* Wolff, and *Orius vicinus* (Ribaut) were collected on crop and non-crop plants in the surveyed area. On pepper, the major number of species was recorded, depending on environmental and cultivation conditions. *O. niger* was the predominant species on strawberries and wild flora, while *O. majusculus* was the primary species on sweet leek. By contrast, *O. laevigatus* was never found on the crops, and rarely collected on wild flora in the areas where it has been usually released. On the wild flora larger amounts of *Orius* were sampled on *Galeopsis tetrahit* L., *Medicago sativa* L., *Malva sylvestris* L., *Matricaria chamomilla* L., *Urtica dioica* L., *Carduus* sp., *Lythrum salicaria* L., *Erigeron annuus* L., and *Trifolium pratense* L., which proved to be important sites for population development and overwintering. The conservation of plant biodiversity as a whole in and near agro-ecosystems is the most reliable way to achieve beneficial insect populations for an effective crop control.

Key words: minute pirate bug, generalist predator, horticultural crops, wild flora.

Introduction

Some species of the genus *Orius* Wolff (Heteroptera Anthocoridae) proved to be effective predators of thrips in both field and greenhouse crops worldwilde. *Orius insidiosus* (Say) and *Orius tristicolor* (White) are two nearctic species widespread in the area of origin of *Frankliniella occidentalis* (Pergande) (Thysanoptera Thripidae), and the first species produced by commercial insectaries and released in Integrated Pest Management (IPM) programmes (Ferragut and Gonzàles Zamora, 1994). Nevertheless in Europe, some palaearctic species proved to be very efficient biological control agents of nearctic *F. occidentalis* and other harmful thrips on crops (van de Veire and Degheele, 1992; Riudavets, 1995; Tavella *et al.*, 1994, 2000). The native anthocorid *Orius laevigatus* (Fieber) showed to be very effective in controlling thrips outbreaks when not disturbed by chemical treatments in different areas of Europe (Villevieille and Millot, 1991; Chambers *et al.*, 1993; Vacante and Tropea Garzia, 1993; Riudavets, 1995; Frescata and Mexia, 1996; Tavella *et al.*, 1991, 1996). This species is widespread along Mediterranean and Atlantic coasts in areas with a marine influence (Péricart, 1972), and it is now produced by many commercial insectaries and largely used in IPM programmes. In Piedmont, a more internal region of northwestern Italy, where *O. laevigatus* is usually released in IPM greenhouses, other *Orius* species are more common on both crop and non-crop plants, such as *Orius niger* Wolff, *Orius majusculus* (Reuter) and *Orius minutus* (L.) (Tavella *et al.*, 1994; Tommasini, 2004; Bosco *et al.*, 2008; Bosco and Tavella, 2008), known as thrips control agents also in other areas of Europe and Middle East (Fischer *et al.*, 1992; van de Veire and Degheele, 1992; Riudavets, 1995; Barbetaki *et al.*, 1999; Castañé and Sanchez, 2006; Bahsi and Tunç, 2008). Indeed, the most important role in preying thrips is usually carried out by the wild *Orius* species, which are clearly well-adapted to climatic conditions (van de Veire and Degheele, 1992; Tavella *et al.*, 2000; Bosco and Tavella 2008; Bosco *et al.*, 2008).

Beside climatic conditions, the host plants play a fundamental role on abundance and composition of *Orius* species. In fact, plants sustain populations of beneficial arthropods by providing them with plant-based foods, preys, sites for hibernation, mating and shelter, and refugia. Vegetation properties should have a particularly high impact on populations of these predators that feed also on pollen (Coll and Ridgway, 1995). On the other hand, plant chemistry and morphological features (plant architecture and leaf morphology) are known to affect directly the behaviour, fitness and reproduction success of anthocorids and, consequently, their predator efficiency (Coll, 1996; Scott Brown *et al.*, 1999; Shaltiel and Coll, 2004; Lundgren and Fergen, 2006; Lundgren *et al.*, 2008, 2009). In such a tritrophic system of plant-prey-predator, the prey density in itself is not univocally correlated with the predator distribution, since plants provide *Orius* spp. with resources besides prey (Coll and Ridgway, 1995; Scott Brown *et al.*, 1999; Ripa *et al.*, 2009; Atakan and Tunç, 2010).

The knowledge of both crop and non-crop plant preferred by *Orius* spp. could be a very useful tool for a correct agro-ecosystem management, in order to attract and augment beneficial insect populations in and around crop fields (Bottenberg *et al.*, 1999). In particular, weed hosts can serve as refugia in absence of the crops and during winter, which is a critical period for insects. *Orius* species are reported to overwinter as adults on a variety of habitats (Péricart, 1972; Elkassabany *et al.*, 1996), and some species show a reproductive diapause induced by photoperiod and temperature (Ruberson *et al.*, 1991; Fischer *et al.*, 1992; van den Meiracker, 1994; Tommasini and van Lenteren, 2003; Bahşi and Tunç, 2008).

The host range and seasonal abundance of anthocorids are essential to understand predatory ecology and evaluate their impact on thrips pests on crops. Therefore, the aim of the research was to assess distribution, host association and abundance of native *Orius* species on some horticultural crops on which thrips are generally a serious threat (i.e., strawberry, sweet leek, and sweet pepper), and on wild plants surrounding cultivations in Piedmont (northwestern Italy).

Materials and methods

The presence and population dynamics of *Orius* spp. were assessed on different horticultural crops (sweet pepper, strawberry, and sweet leek) located in Piedmont (northwestern Italy, 44°17-51'N, 7°30-51'E) during 2005-2007 (table 1). In 2006-2007 *Orius* spp. were sampled also on the wild flora growing in the surroundings of the surveyed crops, to assess the natural host plants alternative to crops in this geographical area.

Crop surveys

S w e e t p e p p e r

Field surveys were conducted on two local sweet pepper varieties: the “Cuneo” variety, grown under plastic tunnels in the province of Cuneo, and the “Corno” variety, grown in the open field in the province of Torino. For each variety, three and two commercial farms were surveyed in 2005 and 2006, respectively; seven plots were totally sampled in the two-year period (table 1). Pepper plants were usually transplanted from mid-April to early May, and harvested starting from early July (tunnel) or early August (open field); seeds were provided and selected by farmers themselves, to maintain traditional cultivars. During the growing season, the standard local horticultural practices were used for irrigation, manuring, weed and disease management. Since many pests, such as the European corn borer, thrips,

Table 1. Crop setting (op fld = open field), plot size, pest management (PM) strategies, location and sampling period for the investigated plots of each crop.

Crop	Plot	Crop setting	Plot size (m^2)	PM strategies/ *Orius* release	Location	Year	N. of surveys	Sampling period
Sweet pepper	1	op fld	2300	conv/no	1: 44°51'N, 7°43'E	2005	6	4-Jul/15-Sep
	2	op fld	6000	conv/no	1: 44°50'N, 7°47’E	2005	6	4-Jul/15-Sep
						2006	11	22-Jun/5-Oct
	3	op fld	1800	conv/no	1: 44°50'N, 7°41'E	2005	6	4-Jul/15-Sep
						2006	11	22-Jun/5-Oct
	4	tunnel	213	conv/no	2: 44°33'N, 7°37'E	2005	6	6-Jul/14-Sep
	5	tunnel	600	conv/no	3: 44°41'N, 7°51'E	2005	6	6-Jul/14-Sep
	6	tunnel	300	IPM/no	4: 44°19'N, 7°32'E	2005	6	6-Jul/14-Sep
						2006	11	22-Jun/5-Oct
	7	tunnel	120	IPM/yes	4: 44°20'N, 7°33'E	2006	11	22-Jun/5-Oct
Strawberry	1	tunnel	500	conv/no	5: 44°19'N, 7°36'E	2006	10	14-Jun/11-Oct
	2	tunnel	500	conv/no	5: 44°19'N, 7°36'E	2006	10	14-Jun/11-Oct
	3	tunnel	500	conv/no	4: 44°19'N, 7°33'E	2006	10	14-Jun/11-Oct
	4	tunnel	500	conv/no	4: 44°19'N, 7°33'E	2006	10	14-Jun/11-Oct
	5	tunnel	500	conv/no	6: 44°17'N, 7°30'E	2006	10	14-Jun/11-Oct
	6	tunnel	500	conv/no	6: 44°17'N, 7°30'E	2006	10	14-Jun/11-Oct
	7	tunnel	430	IPM/yes	4: 44°20'N, 7°33'E	2006	10	8-Jun/11-Oct
						2007	11	15-May/3-Oct
Sweet leek	1	op fld	5300	conv/no	1: 44°51'N, 7°42'E	2005	7	24-Jun/15-Sep
						2006	7	6-Jul/2-Oct
	2	op fld	6200	conv/no	1: 44°51'N, 7°42'E	2005	7	24-Jun/15-Sep
						2006	7	6-Jul/2-Oct
	3	op fld	1900	conv/no	1: 44°50'N, 7°42'E	2005	7	24-Jun/15-Sep
						2006	7	6-Jul/2-Oct
	4	op fld	800	conv-bio/no	1: 44°50'N, 7°42'E	2005	7	21-Jul/30-Sep
				conv-bio/no		2006	7	6-Jul/2-Oct
				IPM/no		2007	7	6-Jul/6-Oct
	5	op fld	7300	conv/no	7: 44°38'N, 7°48'E	2005	7	22-Jun/14-Sep
						2006	7	28-Jun/22-Sep
	6	op fld	1700	conv/no	7: 44°38'N, 7°48'E	2005	7	22-Jun/14-Sep
						2006	7	28-Jun/22-Sep
	7	op fld	2000	conv/no	7: 44°38'N, 7°47'E	2005	7	22-Jun/14-Sep

Locations: 1 = Carmagnola (TO); 2 = Levaldigi (CN); 3 = Bra (CN); 4 = Boves (CN); 5 = Peveragno (CN); 6 = Robilante (CN); 7 = Cervere (CN).

aphids and mites, are highly damaging in northwestern Italy, pesticides were occasionally applied or not at all, depending on the pest management strategy adopted by each farmer. Beneficial organisms (i.e., *Bacillus thuringiensis* Berliner, and *Phytoseiulus persimilis* Athias-Henriot) were released in plots 3, 6 and 7; also a release of *O. majusculus* (1.5 adults m^{-2}) was made in plot 7 on June 14. For the artificial releases *O. majusculus* was chosen, because of its natural spread in this geographic area, unlike the other commercially available species *O. laevigatus* (Bosco *et al.*, 2008).

Anthocorids were sampled fortnightly from early July to mid-October in 2005 and every 10 days from late June to early October in 2006 (table 1). Since populations of *Orius* spp. occurred mostly in the flowers of sweet pepper (Shipp *et al.*, 1992), during surveys, five open flowers were randomly collected in five and 10 sectors of each plot in 2005 and 2006, respectively, placed into vials containing 70% ethyl alcohol, and transferred to the laboratory.

Strawberries

Field surveys were carried out on everbearing strawberries in three commercial farms and in an experimental station, all located in the province of Cuneo, in 2006 (table 1). In the commercial farms, strawberries were grown under plastic tunnels using the standard local horticultural practices for irrigation, manuring, weed, disease and pest management. Sampling was conducted in two tunnels for each farm (totally six tunnels), fortnightly from June to October (table 1). Anthocorids were sampled by collecting 10 open flowers in three sectors of each tunnel; then, flowers were placed into vials containing 70% ethyl alcohol, and transferred to the laboratory. In the experimental station, strawberries were always grown in plastic tunnel, applying IPM strategies. Releases of *O. majusculus* (1.5 adults m^{-2} on June 14) and of *P. persimilis* (7 individuals m^{-2}, 3 releases) were made in 2006; one release of *O. majusculus* (1.5 adults m^{-2} on June 27) was made in 2007. Sampling was carried out by collecting 10 open flowers on nine groups of 10 plants in 2006 and five open flowers on six groups of five plants in 2007, every two weeks from May or June until October (table 1).

Sweet leek

Field surveys were carried out in three commercial farms located in Carmagnola (TO) in 2005-2006, and in three and two commercial farms located in Cervere (CN) in 2005 and 2006, respectively (table 1). During the growing season, the standard horticultural practices typical for each area were used: sweet leek plants were sown by growers in plastic tunnels open on the sides in March-April and transplanted to nearby open fields in June-July. Seeds were selected and provided by farmers themselves, to maintain traditional cultivars. In the surveyed crops, chemical pest management was generally applied using broad-spectrum insecticides on a routine basis to control both onion thrips and leek moth. Immediately after transplanting, *Orius* populations were monitored fortnightly by checking subsamples of five adjacent plants chosen randomly at each of 10 points distributed diagonally over the field (50 plants $field^{-1}$). By beating each plant onto a plastic tray (350 × 250 mm) predators were counted, collected with a mouth aspirator, and transferred to the laboratory.

During 2005-2007 surveys were also conducted in an experimental field where different pest management strategies were compared: in 2005 and 2006, biological and chemical pest control were applied in different subplots, whereas in 2007 IPM based on supervised control was adopted for the whole plot. Fortnightly surveys were conducted from early (in 2005) or late July (in 2006 and 2007) to late September (in 2005) or early October (in 2006 and 2007) (table 1); anthocorids were sampled as described for the commercial fields. In 2005 and 2006, a total of 30 plants per treatment (biological and chemical control) were checked; in 2007 a total of 60 plants were checked for the whole plot (IPM).

Statistical analyses

Mean numbers of total, adults and nymphs of *Orius* spp. per sampling point, collected in the pepper plots in 2005 and 2006 and in the strawberry plots in 2006, were pooled over time; then, they were log-transformed to achieve homogeneity of variance (Levene) and normality (Shapiro-Wilk), and analyzed by Univariate Analysis of Variance (ANOVA). Means were separated by Tukey's test when ANOVA was significant ($P < 0.05$). To compare the abundance of the different *Orius* species, collected on pepper in 2005 and 2006, and on strawberry in 2006 (plot 7), mean numbers of anthocorids sampled per point and date in each of the investigated plots were log-transformed and analyzed as described above (ANOVA followed by Tukey's test). All statistical analyses were performed using the software SPSS® 20.0 version (SPSS, Chicago, IL, USA).

Wild flora surveys

During 2006-2007, *Orius* spp. were sampled also on the wild flora in Boves (44°20'N, 7°32'E, 580 m a.s.l.), in the surroundings of sweet pepper plot 7 and strawberry plot 7 (table 1). This site was chosen due to its high biodiversity of both crop and non-crop plant species and the low anthropic impact on the agro-ecosystem. In fact, various crops (alfalfa, bean, blueberry, lettuce, strawberry, sweet pepper, tomato, zucchini) were grown, separated each other by uncultivated corridors. Overall, 17 surveys were carried out monthly or bimonthly from late June 2006 until early May 2007, and fortnightly from mid-May 2007 until early October 2007. Anthocorids were sampled on the wild flora by beating, if possible, at least 10 plants for each species (the flowers, when present, or the terminal buds) onto a white plastic tray (350 × 250 mm). *Orius* adults and nymphs were then counted, collected with a mouth aspirator and taken back to the laboratory. The presence of thrips of other possible preys (e.g., aphids or mites) was detected for each sampled plant.

Laboratory analyses

In the laboratory, anthocorids collected on sweet leek and wild flora were killed with ethyl acetate, and stored in vials containing 70% ethyl alcohol. Then, field col-

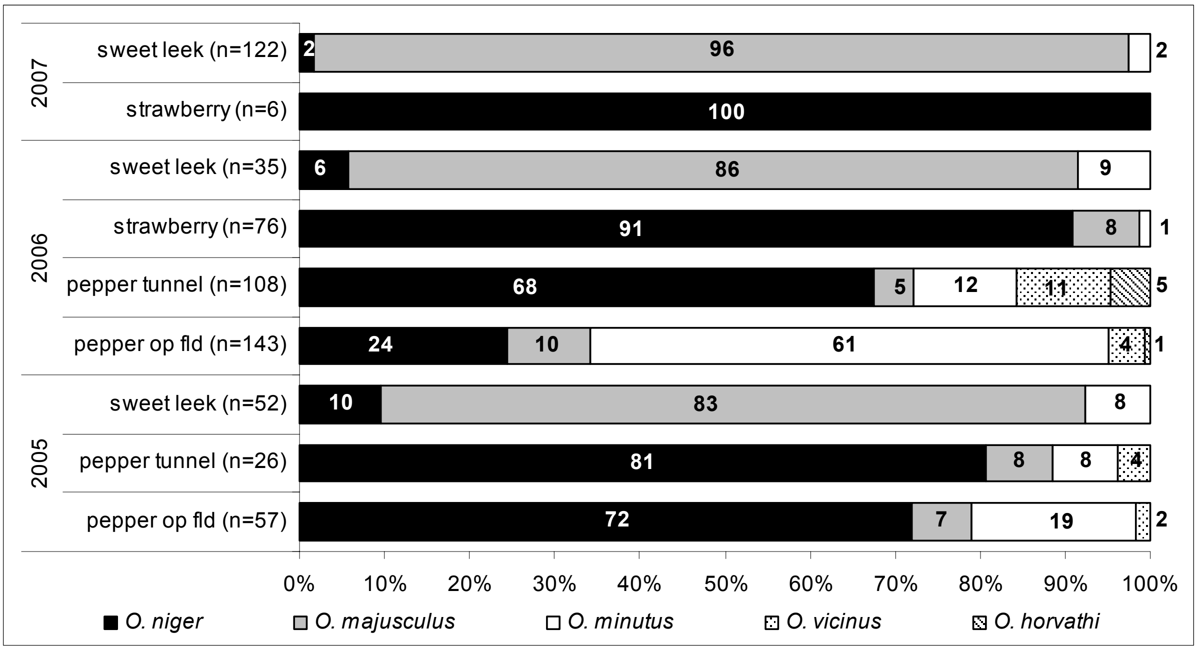

Figure 1. Composition of *Orius* species on the crops surveyed in 2005-2007. Percentages are referred to the total dissected adults for each year and crop (n).

lected pepper and strawberry flowers were dissected, vial content was examined under a stereomicroscope at 40× magnification, and anthocorids were separated and counted. Adults were identified to the species level by comparing, under a stereomicroscope at 160× magnification, the genital clasper in males and the copulatory tube in females according to Péricart (1972). Nymphs were examined for the presence of four macrochetae in the corners of the pronotum, which are present in subgenus *Orius* and absent in subgenus *Heterorius* (Péricart, 1972). The females collected on wild flora in late autumn-winter were dissected to observe the presence of well-developed ova and reproductive structures or extensive fat body, being an indication of non-diapausing and diapausing females, respectively (El-kassabany *et al.*, 1996). All plant species sampled in the hedgerows and wastelands were determined according to Pignatti (1997).

Results

Crop surveys

During field-surveys, 993 specimens of *Orius* spp. (70% adults and 30% nymphs) were overall collected. The dissected adults (n = 625) belonged to five species: *Orius horvathi* (Reuter) (1%), *O. majusculus* (35%), *O. minutus* (20%), *O. niger* (41%), and *Orius vicinus* (Ribaut) (3%). Specific composition was variable depending on the investigated crop and/or year (figure 1). Regardless of the year and geographical location, some species were predominant on one crop: 91% of adults totally collected on sweet leek (n = 209) and on strawberries (n = 82) belonged to *O. majusculus* and *O. niger*, respectively. On the contrary, all five species were sampled on sweet pepper; under tunnels the specific composition was relatively similar in the two years (n = 134), prevailing *O. niger* (70%), followed by *O. minutus* (11%), *O. vicinus* (10%), *O. majusculus* (5%) and *O. horvathi* (4%). Nevertheless, in the open field there was a greater variability in the specific composition, prevailing *O. niger* in 2005 and *O. minutus* in 2006 (figure 1).

Sweet pepper

The mean numbers of total *Orius*, adults and nymphs per sampling point collected during 2005 and 2006 are shown in table 2; the specific *Orius* composition for each plot is shown in figure 2. In 2005, despite no releases were made in any plots, differences in the *Orius* amounts were observed, regardless of the crop setting (open field or tunnel). The highest and the lowest numbers of total and adults were recorded in plot 3 (open field) and in plot 6 (tunnel), respectively (ANOVA, $P \leq 0.01$). On the other hand, the highest and the lowest numbers of nymphs were collected in plot 4 (tunnel) and in plots 1, 2 (open fields) and 6 (tunnel), respectively (ANOVA, $P < 0.0001$) (table 2). Four species were sampled: *O. niger*, *O. majusculus*, *O. minutus* and *O. vicinus* (figures 1 and 2). Among them, *O. niger* was predominant in all the surveyed plots; the mean numbers of *O. niger* adults collected per point and per date were significantly greater than those of other species in plots 2, 3 and 4 [ANOVA: df = 3,16; F = 11.672 (plot 2); 10.126 (plot 3); 5.248 (plot 4); $P < 0.01$; N = 5] (figure 2A).

In 2006, the highest numbers were recorded in plot 7 (tunnel), not surveyed in 2005, where *O. majusculus* was released, whereas the lowest numbers were sampled in plot 6, similarly to what observed in 2005 (ANOVA, $P < 0.0001$) (table 2). During 2006, beside the four species found in 2005, also *O. horvathi* was collected (figures 1 and 2B). As in 2005, *O. niger* was predominant in both surveyed tunnels; particularly in plot 7, despite

Table 2. Mean numbers (N°) per sampling point of *Orius* (total, adults and nymphs), collected during all field surveys (N° of surveys) on sweet pepper plots in 2005 and 2006, and on strawberry plots in 2006. Statistical analyses were performed on log-transformed data which are not shown. Within each column (year and crop), means followed by different letters are significantly different (Tukey's test following ANOVA, P < 0.05). ANOVA results (P, df, F) and SED values are reported.

Crop	Year (N° of surveys)	Plot	N° of total *Orius* ± SE	N° of adults *Orius* ± SE	N° of nymphs *Orius* ± SE
Sweet pepper	2005 (6)	1 op fld	2.20 ± 0.97 bc	1.80 ± 0.80 ab	0.40 ± 0.24 b
		2 op fld	3.40 ± 0.93 abc	3.20 ± 0.86 ab	0.20 ± 0.20 b
		3 op fld	8.20 ± 1.36 a	6.40 ± 1.29 a	1.80 ± 0.58 ab
		4 tunnel	7.00 ± 1.14 ab	2.60 ± 0.93 ab	4.40 ± 0.24 a
		5 tunnel	3.60 ± 1.40 abc	2.40 ± 1.50 ab	1.20 ± 0.37 b
		6 tunnel	0.60 ± 0.40 c	0.20 ± 0.20 b	0.40 ± 0.40 b
		$P_{(5,24)}$	<0.0001	0.010	<0.0001
		F	7.120	3.861	10.004
		SED	0.354	0.427	0.263
Sweet pepper	2006 (11)	2 op fld	7.10±1.23 b	5.80±1.01 b	1.30 ± 0.45 b
		3 op fld	10.60±0.79 b	8.40±1.00 ab	2.20 ± 0.59 b
		6 tunnel	1.50±0.40 c	0.60±0.22 c	0.90 ± 0.28 b
		7 tunnel	21.30±1.52 a	10.20±1.08 a	11.10 ± 1.11 a
		$P_{(3,36)}$	<0.0001	<0.0001	<0.0001
		F	53.750	42.528	24.496
		SED	0.186	0.194	0.252
Strawberry	2006 (10)	1 tunnel	1.33±0.33 b	1.33±0.33 ab	0.00 ± 0.00 b
		2 tunnel	0.33±0.33 b	0.33±0.33 b	0.00 ± 0.00 b
		3 tunnel	0.33±0.33 b	0.00±0.00 b	0.33 ± 0.33 b
		4 tunnel	1.67±0.33 b	1.67±0.33 ab	0.00 ± 0.00 b
		5 tunnel	1.33±0.88 b	1.00±1.00 b	0.33 ± 0.33 b
		6 tunnel	1.67±0.88 b	1.67±0.88 ab	0.00 ± 0.00 b
		7 tunnel	15.11±0.95 a	6.44±0.62 a	8.67 ± 1.35 a
		$P_{(6,14)}$	<0.0001	0.003	<0.0001
		F	10.731	6.061	37.752
		SED	0.372	0.374	0.191

the release of *O. majusculus*, the mean number of *O. niger* adults per point and per date was significantly greater than that of other species (ANOVA: df = 4,45; F = 30.510; P <0.0001; N = 10) (figure 2B). On the contrary, *O. minutus* was predominant in both surveyed fields, with mean number significantly greater than that of other species in plot 3 (ANOVA: df = 4,45; F = 25.950; P < 0.0001; N = 10) and than other species except *O. niger* in plot 2 (ANOVA: df = 4,45; F = 11.824; P < 0.0001; N = 10) (figure 2B).

In both years, larger amounts of adults (over 80% of total *Orius*) and of nymphs (over 55% of total *Orius*) were collected in the open field and under tunnel, respectively. During 2005, adults and nymphs of the subgenus *Orius* (including *O. niger*) prevailed; during 2006, adults and nymphs of the subgenus *Heterorius* (including *O. majusculus*, *O. minutus* and *O. horvathi*) prevailed in the open field, whereas adults and nymphs of the subgenus *Orius* prevailed in tunnels.

Strawberries

The mean numbers of total *Orius*, adults and nymphs per sampling point collected in 2006 are shown in table 2. The highest values were recorded in plot 7, in which *O. majusculus* was released. Here, total *Orius* and nymphs were significantly greater than in all other plots (ANOVA, P < 0.0001), whereas adults were significantly greater than in plots 2, 3 and 5 (ANOVA, P < 0.01) (table 2).

The three species *O. niger*, *O. majusculus* and *O. minutus* were sampled (figure 1); among them, *O. niger* was predominant in all the surveyed plots. In particular, in plot 7, despite the release of *O. majusculus*, the mean number of *O. niger* adults collected per point and per date (0.61 ± 0.07) was significantly greater than the mean numbers of both *O. majusculus* (0.02 ± 0.01) and *O. minutus* (0.01 ± 0.01) (ANOVA: df = 3,32; F = 91.642; P < 0.0001; N = 9). Overall, nymphs were collected in larger amounts than adults (57% of total *Orius*). Adults and nymphs of subgenus *Orius* prevailed on those of the subgenus *Heterorius*.

Sweet leek

During field surveys in 2005-2006 a small number of *Orius* spp. was sampled in the six commercial fields: on average 0.02 and 0.01 anthocorids per plant were collected in Carmagnola (plots 1, 2 and 3) and Cervere (plots 4, 5 and 6), respectively. In the experimental field (plot 7), specimens of *Orius* spp. were rarely collected: on average, 0.04 and 0.08 anthocorids per plant in 2005, and 0.03 and 0.01 anthocorids per plant in 2006 were sampled in biological and chemical treatments, respec-

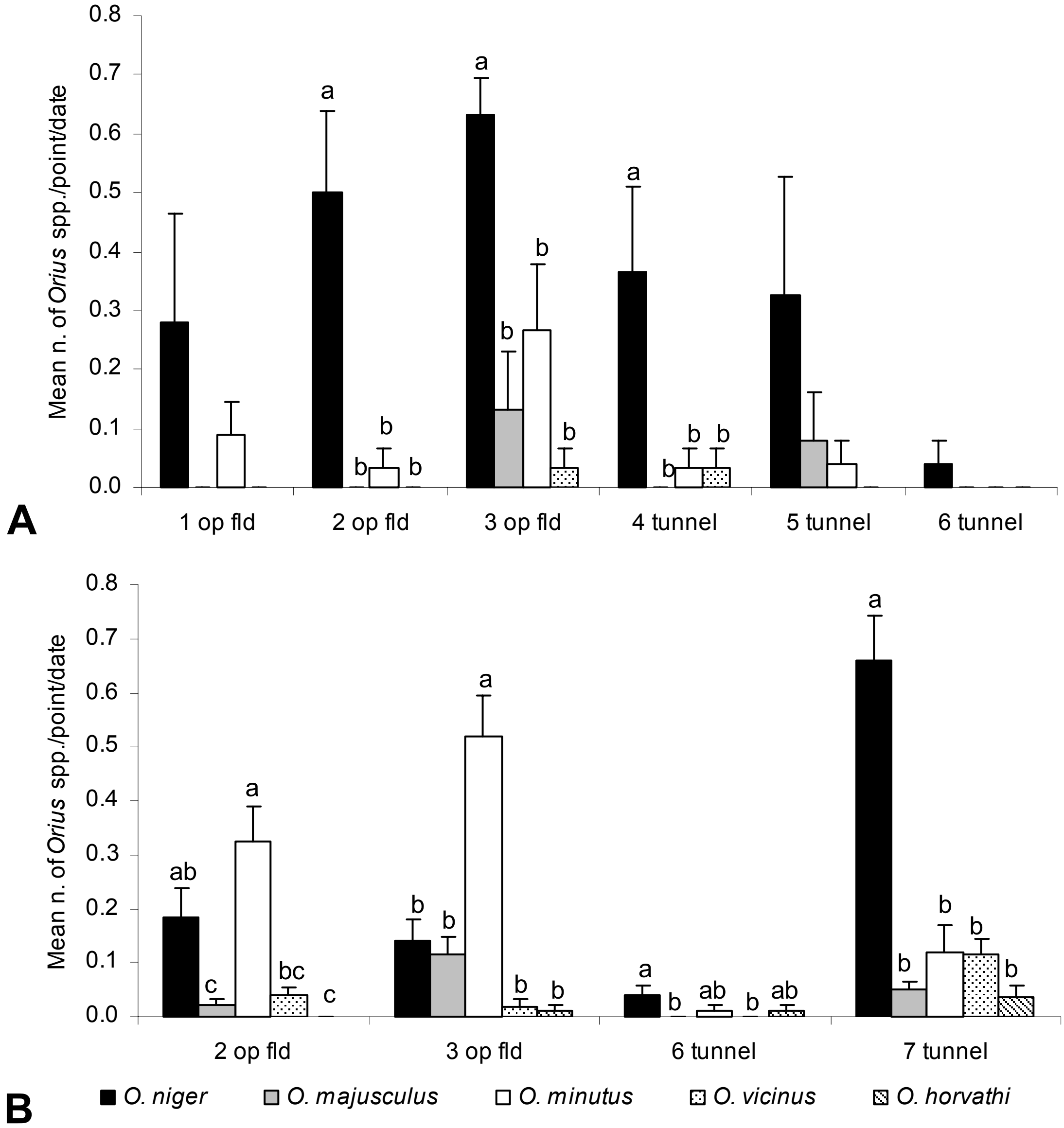

Figure 2. Mean numbers (± SE) of *Orius* spp. adults collected per sampling point and date on pepper plots investigated during 2005 (A) and 2006 (B). Within each plot, bars labelled with different letters are significantly different (Tukey's test following ANOVA, P < 0.05). Statistical analyses were performed on log-transformed data which are not shown.

tively. In 2007, when integrated pest control was applied, larger amounts of *Orius* were collected than in the previous two years. On average, 0.39 anthocorids per plant were recorded, with maximum values of 1.3 and 1.2 anthocorids per plant collected on September 5 and 20, respectively, when leek plants were carefully inspected at the upper part of the shaft of basal leaves.

Overall, the three species *O.niger*, *O. majusculus*, and *O. minutus* were sampled. During the two-year surveys, in the commercial plots (1-6) 95.3% and 4.7% of the dissected adults (n = 43) belonged to *O. majusculus* and *O. niger*, respectively, while in the experimental plot (7) 90% of dissected adults (n = 166) belonged to *O. majusculus*, and the remaining to *O. minutus* (6%), and *O. niger* (4%). At the same, the few nymphs collected (4 in the commercial plots and 3 in the experimental plots) belonged to the subgenus *Heterorius*.

Wild flora surveys

From late June 2006 until early October 2007, *Orius* specimens were collected on 27 of the 50 sampled plant species; on the remaining 23 plants where anthocorids were never collected, 17 species were found only once or twice, whereas the following plants were sampled at least in three dates: *Silene inflata* (Salisb.) (50 plants totally checked), *Capsella bursa-pastoris* (L.) Medicus (145), *Rubus* sp. (50), *Conyza canadensis* (L.) Cronq. (40), *Senecio vulgaris* L. (75), and *Taraxacum officinale* Weber (30) (figure 3). On average from 3 to 8 *Orius* specimens 10 beating^{-1} per sampling date were collected on: *Galeopsis tetrahit* L., *Medicago sativa* L., *Malva sylvestris* L., *Matricaria chamomilla* L., *Urtica dioica* L., *Carduus* sp., *Lythrum salicaria* L., *Erigeron annuus* L., and *Trifolium pratense* L. (table 3). Moreover, in a single date relatively high numbers of *Orius* spp. were sampled on *Diplotaxis* sp. (14 specimens on June 27), *Plantago lanceolata* L. (10 specimens on July 11), and *Rumex acetosa* (Miller) Krause (6 specimens on August 9). Regardless of the presence of anthocorids, thrips were observed on all the sampled plant species, while aphids were recorded on *Chenopodium album* L., *Amaranthus retroflexus* L., *Vicia* sp., and *Leuchantemum* sp. However, no evident connection was found between the presence/amounts of anthocorids and of preys on the sampled plants (data not shown).

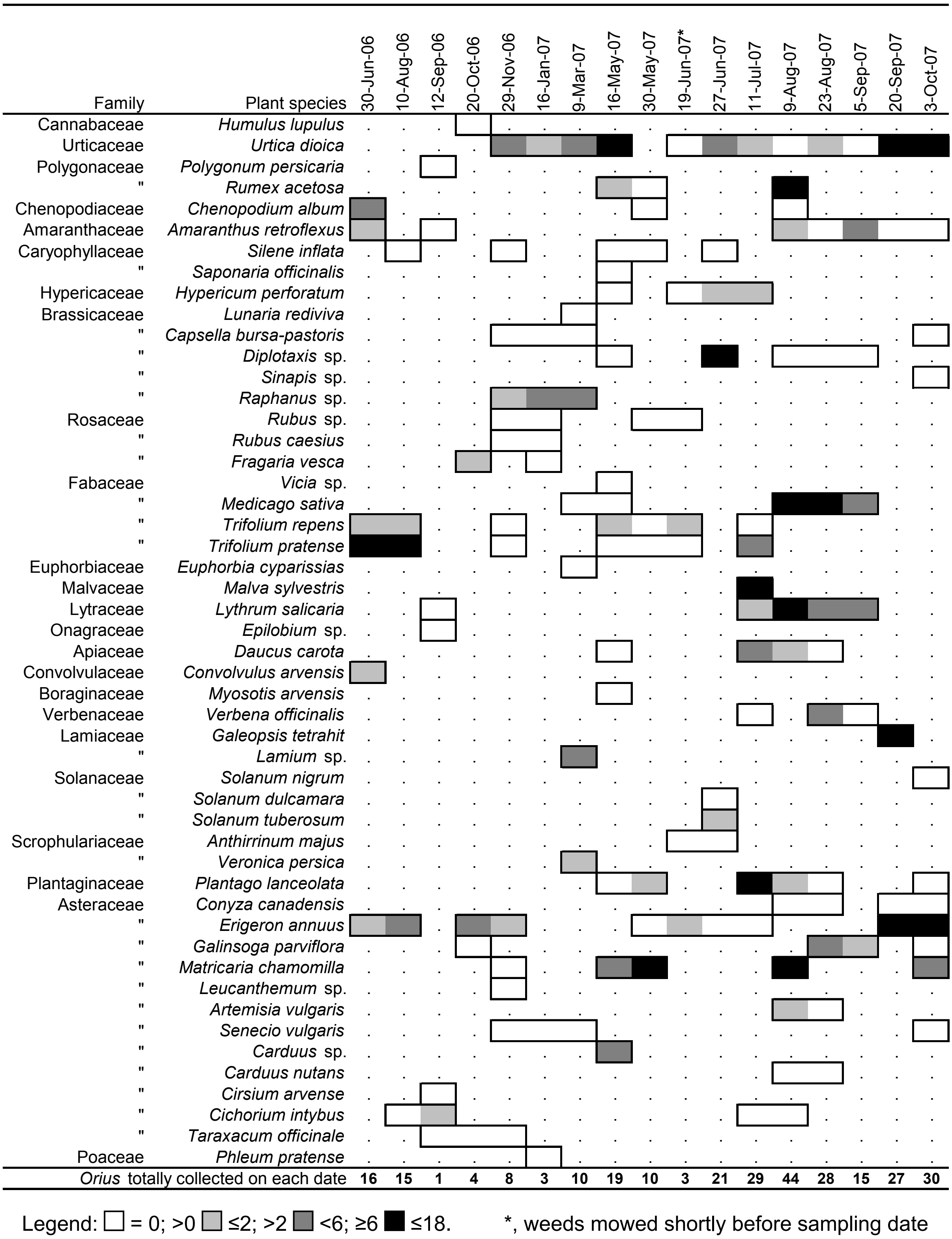

Figure 3. Presence and abundance of *Orius* (adults plus nymphs) collected on the 50 wild plant species surveyed between June 2006 and October 2007. Colours and values are referred to mean total numbers 10 beating^{-1} per date.

Table 3. Abundance and composition of *Orius* spp. on the wild flora. Number of surveys, mean numbers of *Orius* (total, nymphs and adults) per sampling of 10 plants, and percentages of *Orius* species on dissected adults (*O. lae.* = *O. laevigatus*; *O. nig.* = *O. niger*; *O. maj.* = *O. majusculus*; *O. min.* = *O. minutus*) are reported for each plant species.

Plant species	N° of surveys	N° survey^{-1} total	nymphs	adults	Dissected adults of *Orius* spp. N°	*O. lae.*	*O. nig.*	*O. maj.*	*O. min.*
Urtica dioica	12	4.0	0.4	3.6	61	11%	85%	2%	2%
Rumex acetosa	3	2.3	0.7	1.6	4	25%	75%	0%	0%
Chenopodium album	3	1.0	0.0	1.0	1	0%	100%	0%	0%
Amaranthus retroflexus	7	1.0	0.0	1.0	7	0%	86%	14%	0%
Hypericum perforatum	4	0.6	0.0	0.6	3	0%	0%	33%	67%
Diplotaxis sp.	5	2.8	0.0	2.8	11	0%	100%	0%	0%
Raphanus sp.	3	2.9	0.0	2.9	8	25%	50%	25%	0%
Fragaria vesca	2	0.5	0.0	0.5	1	0%	100%	0%	0%
Medicago sativa	5	7.4	1.6	5.8	28	14%	68%	14%	4%
Trifolium repens	7	1.0	0.3	0.7	5	0%	80%	20%	0%
Trifolium pratense	7	3.0	0.1	2.9	15	7%	93%	0%	0%
Malva sylvestris	1	6.0	0.0	6.0	3	0%	100%	0%	0%
Lythrum salicaria	5	3.6	1.8	1.8	9	11%	78%	11%	0%
Daucus carota	4	1.6	0.4	1.2	6	0%	100%	0%	0%
Convolvulus arvensis	1	1.0	0.0	1.0	1	0%	100%	0%	0%
Verbena officinalis	3	1.3	1.3	0.0	0	0%	0%	0%	0%
Galeopsis tetrahit	1	8.0	4.0	4.0	4	0%	100%	0%	0%
Lamium sp.	1	2.5	0.0	2.5	5	0%	80%	0%	20%
Solanum tuberosum	1	2.0	0.0	2.0	2	0%	100%	0%	0%
Veronica persica	1	0.5	0.0	0.5	1	0%	100%	0%	0%
Plantago lanceolata	6	2.0	0.0	2.0	9	0%	78%	22%	0%
Erigeron annuus	10	3.4	0.8	2.6	17	0%	88%	12%	0%
Galinsoga parviflora	4	1.0	0.0	1.0	3	0%	33%	33%	33%
Matricaria chamomilla	5	4.7	0.8	3.9	19	0%	95%	5%	0%
Artemisia vulgaris	2	0.5	0.0	0.5	1	0%	100%	0%	0%
Carduus sp.	1	4.0	1.0	3.0	3	0%	100%	0%	0%
Cichorium intybus	4	0.3	0.3	0.0	0	0%	0%	0%	0%

A total of 298 *Orius* specimens were collected on the wild flora, of which 83% were adults belonging to *O. laevigatus* (7%), *O. majusculus* (7%), *O. minutus* (3%), and *O. niger* (83%). All the four *Orius* spp. were found only on *U. dioica* and *M. sativa*. Except on *Hypericum perforatum* L. and *Galinsoga parviflora* Ruiz *et* Pavon, *O. niger* was the most abundant species, and the only one sampled on 11 wild plants. Overall, *O. niger* was collected on 24 plant species, *O. majusculus* on 11 species, *O. laevigatus* on 6 species, and *O. minutus* on 5 species (table 3). Nymphs represented 17% of total *Orius*, and were collected on 13 plant species, particularly on *G. tetrahit*, *L. salicaria*, *M. sativa* and *Verbena officinalis* L. On this last plant, and on *Cichorium intybus* L., no adults but only nymphs were found (table 3). Overall, nymphs were collected from May until the beginning of October, and never in late autumn and winter (i.e., October 20 and November 29 in 2006, January 16 and March 9 in 2007). Nevertheless in this period, when wild plants were rarer, adults were sporadically sampled even if in lower mean numbers (except on June 19, 2007, when wild flora was mowed) on *U. dioica*, *Raphanus* sp., *Fragaria vesca* L., *Lamium* sp., *Veronica persica* Poiret, and *E. annuus* (figure 3). Moreover, *U. dioica* and *Raphanus* sp. were the only host plants observed on January 16, 2007: plants of both species were clustered near piles of rubble and compost.

From late autumn to late winter, all the females collected on wild flora were dissected to assess their reproductive status. On November 29, 2006, two females of *O. niger* displayed well-developed ova and reproductive structures rather than extensive fat body, whereas the other females (17 *O. niger*, 7 *O. laevigatus*, 1 *O. majusculus*) showed only fat body. On January 16, 2007, two females of *O. niger* displayed well-developed ova, whereas the remaining three females (1 *O. niger*, 2 *O. laevigatus*) showed only fat body. On March 9, 2007, seven of eight collected females of *O. niger* displayed well-developed ova; on the contrary, two females of *O. majusculus* and one female of *O. minutus* presented only fat body. Starting from May, most females of all *Orius* species presented well-developed ova and reproductive structures, and nymphs were collected again until October 3, 2007. On this last sampling date, the nine females of *O. niger* collected on *U. dioica*, *E. annuus*, and *M. chamomilla*, showed only fat body and no well-developed ova.

Discussion and conclusions

Results of field surveys support the preference of the sampled *Orius* species for a specific crop, irrespective of the year and geographical location. Therefore, on strawberries *O. niger* was the primary species (91%), also in the plots where *O. majusculus* was released, consistent with other previous studies in northern Italy (Tavella *et al.*, 1994; Tommasini, 2004). Moreover, the nymphs belonged mainly to subgenus *Orius* (76%), which includes *O. niger* beside *O. laevigatus*, the latter never found on strawberries. In the same way, *O. majusculus* seems to be more related with the sweet leek, where it was predominant (91%). Although the species was already reported on leek (Bosco and Tavella, 2010), this record is unusual considering the paucity of predators generally assumed on this crop (Vierbergen and Ester, 2000), even when grown using biological control (den Belder *et al.*, 2003). On the other hand, on sweet pepper, that is considered one of the favourite horticultural crops by many *Orius* spp. (Coll and Ridgway, 1995; Scott Brown *et al.*, 1999; Tommasini, 2004), the largest specific variability was recorded.

O. vicinus and *O. horvathi* (subgenus *Heterorius*), mostly found on trees feeding on aphids and spider mites (Péricart, 1972), were rarely collected on sweet pepper in Italy and Spain (Riudavets and Castañé, 1994; Tommasini, 2004). Although in our study they were collected on sweet pepper in larger amounts than in previous studies, their presence and role in controlling thrips on horticultural crops in northern Italy seem to be secondary to those of the other three species. In both years, *O. niger* was the predominant species in plastic tunnels (70%), corroborating what was observed in the same geographic area (Bosco *et al.*, 2008). In open fields species composition was more variable in the two years, and different from what previously observed in another geographic area of Piedmont (northwestern Italy) in pepper open fields (Bosco and Tavella, 2008). Cultivations in the open field are obviously more exposed to local climate conditions (i.e., temperatures, rainfall, humidity), which can play an important role in determining composition and relative abundance of *Orius* species. From May to August, in the investigated area, mean temperatures were similar during 2005 and 2006 (21.8 and 21.1 °C, respectively). On the contrary, rainfalls and relative humidity were notably lower in 2005 (39.8 mm and 47.9%) than in 2006 (66.1 mm and 57.4%). Both *O. majusculus* and *O. minutus* are more hygrophilous and less adapted to high temperatures than *O. niger*. For example, *O. majusculus* is particularly widespread on vegetation along rivers and especially in the swamps (Péricart, 1972); in Italy it is present in the coldest areas (i.e., Piedmont), decreasing its relative abundance from northern to central Italy (Tommasini, 2004).

On the horticultural crops, the most important role in preying thrips was carried out by the wild *Orius* species. In fact, on both strawberries and pepper, in the plots where releases of *O. majusculus* were made, *O. niger* was significantly more abundant than all other species. This propensity of a better crop colonization by the wild species was already observed on pepper when *O. laevigatus* was artificially introduced (Bosco *et al.*, 2008). Although in the present study *O. majusculus* was chosen instead of *O. laevigatus*, because of its natural spread in this geographic area, the introduced anthocorids struggled to reproduce and establish on the crop. Nevertheless, *Orius* populations were significantly higher in the plots where *O. majusculus* was released, probably due to the adopted pest management strategy and to the reduction of insecticide applications which allowed the spontaneous colonization by wild species. As already observed (Bosco *et al.*, 2008), pest control strategies play a more important role than the release itself.

On the wild flora, *O. niger* was the predominant species (83%). Beside *O. majusculus* and *O. minutus*, also *O. laevigatus* was sampled (7%) in the experimental site of Boves, in the province of Cuneo, whereas it was never found on wild flora in the province of Turin (Bosco and Tavella, 2008; Bosco *et al.*, 2008). However, this record is consistent with what observed in previous field surveys in Piedmont (Tommasini, 2004). In northwestern Italy *O. laevigatus* is largely adopted in IPM or organic programmes, even if this species is not widespread in alpine zones, including Piedmont (Péricart, 1972). In the experimental site, wild plants were integrated with vegetable crops, where this predator has been occasionally introduced. It then becomes difficult to distinguish between a presence, however sporadic, but natural of *O. laevigatus* in Piedmont, and a presence due to the migration of individuals released in adjacent crops. Indeed, *O. laevigatus* was never found on the investigated crops in the three-year survey.

In previous research *O. insidiosus* nymphs proved to live longest on the plant species that were more frequently used for oviposition (Lundgren *et al.*, 2008). Thus the presence and abundance of different life stages on a plant species (either crop or non-crop) can suggest whether the plant is suitable for *Orius* reproduction besides providing them with food (preys and plant-derived food), sites for hibernation, shelter, and refugia. For oviposition insect females should choose plants that bestow the greatest fitness for developing offspring, and in several cases non-crop plants are preferred over crop plants by predatory insects (Coll, 1996; Lundgren and Fergen, 2006; Lundgren *et al.*, 2008). Nevertheless, in our study very high numbers of nymphs were often found on sweet pepper and strawberry, while they were rarely collected on leek.

These findings are important tools to plan a correct management of the agro-ecosystem in order to enhance integrated thrips control on crops. On strawberries and pepper *Orius* spp. can reproduce and develop, therefore neighbouring wild plants are important mainly in attracting and augmenting natural enemy populations, besides serving as alternative plants in absence of crops. On the contrary, sweet leek, which is a non-flowering crop, can host *Orius* populations providing a prey source, but it is not a suitable host for reproduction; thus the presence of alternative host plants in the surroundings is crucial to support early crop colonization by predators. Indeed, the major *Orius* numbers on sweet leek were found in the experimental field, a small plot of about 800 m^2 surrounded by ecological corridors. Similarly Shaltiel and Coll (2004) found that *Anthocoris nemoralis* F. preferred to oviposit on non-crop plants surrounding pear orchards, and recommended intercropping with favourite host trees to increase predation within the orchards. The concept that vegetational diversity enhances conservation of natural enemies within farmland and their potential as biological control agents is corroborated by the evidence that the highest numbers of *Orius* spp. were recorded in the agro-ecosystem of Boves, a site characterized by high biodiversity and presence of ecological corridors. Similarly, recent studies showed that *O. insidiosus* was more abundant in diversified or intercropped plots than in monocultures (Lundgren *et al.*, 2009; Bickerton and Hamilton, 2012).

Orius species overwinter as adults; some species, such as the nearctic *O. insidiosus* and *O. tristicolor* (Ruberson *et al.*, 1991; van den Meiracker, 1994), and the palaearctic *O. majusculus* (Fischer *et al.*, 1992; van den Meiracker, 1994) and *O. minutus* (Ito and Nakata, 1998) undergo reproductive diapause under photoperiodic stimuli. On the contrary, photoperiod did not induce reproductive diapause in *O. niger* (van de Veire and Degheele, 1992; Bahsi and Tunc, 2008), which could survive and reproduce under winter conditions in Turkey (Bahsi and Tunc, 2008). In the present study, even if it was not aimed at assessing reproductive diapause in the collected *Orius* species, some observations on adults sampled on wild flora in winter are worth considering. Indeed the presence of mature eggs in *O. niger* females in mid-winter suggests that this species does not enter hibernatory reproductive diapause in northwestern Italy.

Concerning the presence of prey, abundance of *Orius* spp. did not correspond to the abundance of thrips on weeds, as already observed (Atakan, 2010; Atakan and Tunc, 2010). The most beneficial plant species were: *G. tetrahit*, *M. sativa*, *M. sylvestris*, *M. chamomilla*, *U. dioica*, *Carduus* sp., *L. salicaria*, *E. annuus*, *T. pratense*, *Diplotaxis* sp., *P. lanceolata*, and *R. acetosa*, where larger *Orius* amounts were collected. In particular, *G. tetrahit*, *L. salicaria*, *M. sativa*, *V. officinalis* and *C. intybus*, on which a high proportion of nymphs was found, can affect positively predator development and augmentation, while *U. dioica*, *Raphanus* sp., *F. vesca*, *Lamium* sp., *V. persica* and *E. annuus* carry on an important role as overwintering vegetation. Therefore, these predators probably respond differently to various resources offered by different plant species, as assessed in studies focusing on the effects of plant subsidies on *O. insidiosus* fitness (Pumariño *et al.*, 2012). Hence the conservation of plant biodiversity as a whole in and near agro-ecosystems is the most reliable way to achieve beneficial insect populations for an effective crop control.

Acknowledgements

Particular thanks to Michele Baudino and Roberto Giordano of CReSO of Boves for technical assistance in the fields. Research was supported by grants of Regione Piemonte.

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Received May 14, 2013. Accepted September 20, 2013.