

**Biological control of thrips pests (Thysanoptera: Thripidae)
in a commercial greenhouse in Hungary**

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ABSTRACT. Polyphagous thrips, like western flower thrips *Frankliniella occidentalis* and onion thrips *Thrips tabaci*, are major pests in various ornamental and vegetable crops in greenhouses throughout the world. In Hungary, both of these polyphagous thrips species frequently cause severe damage in many greenhouse crops, especially in commercial sweet pepper. Chemical control is not always feasible because of certain ecological characteristics of these thrips species. The commercially available phytoseiid predatory mites like *Amblyseius swirskii* and anthocorid flower bugs like *Orius laevigatus* are often used simultaneously for the biological control of severe thrips infestation in sweet pepper cultivation in Hungary. Our observations demonstrated that the polyphagous thrips assemblages were effectively controlled by the combined release of natural enemies, despite the fact that the establishment of *O. laevigatus* did not seem to be successful in the first year. Overall, the thrips population density remained below the economic threshold in both years. However, the low infestation level of thrips suggests that a single predator release strategy could be applied effectively and still maintain the thrips below the damage threshold in greenhouse sweet pepper.

KEY WORDS: biological control, greenhouse sweet pepper, *Amblyseius swirskii*, *Orius laevigatus*, thrips.

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INTRODUCTION

Polyphagous thrips, like western flower thrips *Frankliniella occidentalis* (PERGANDE, 1895) (Thysanoptera: Thripidae) and onion thrips *Thrips tabaci* LINDEMAN, 1889 (Thysanoptera: Thripidae), are key pests in various ornamental and vegetable crops in greenhouses worldwide. Since *Frankliniella occidentalis* was first reported in Hungary on various cut flowers in 1989 (JENSER & TUSNÁDI 1989), western flower thrips have become the most dangerous thrips pest of protected crops in Hungary (HATALA ZSELLÉR & KISS 1999, MOLNÁR et al. 2008). Thrips infestations can cause serious economic losses by their direct damage and by transmitting Tomato Spotted Wilt Virus (TSWV). Both of these polyphagous thrips species frequently cause severe damage in many greenhouse crops, especially in commercial sweet pepper. Sweet pepper (*Capsicum annuum* L.) is the most important greenhouse crop in Hungary: it is grown on approximately 2 000 hectares. Growers most often produce traditional Hungarian cultivars (HATALA ZSELLÉR 1992, TOMPOS 2006), where the success of plant protection is based on the management of thrips. Chemical control is not always feasible because of certain ecological characteristics of these thrips species: thigmotactic behaviour, high reproductive capacity and tolerance to insecticides. The efficiency of thrips management in sweet pepper could be improved by using predatory arthropods like the predatory mite *Amblyseius swirskii* ATHIAS-HENRIOT, 1962 (Acari: Phytoseiidae) and the flower bug *Orius laevigatus* FIEBER, 1860 (Hemiptera: Anthocoridae), which are economically successful alternatives to chemical pest control within the framework of augmentative biological control (VAN LENTEREN & BUENO 2003). Biological pest control is most economical in long-period forcing, when a growing period can last for up to 8-10 months. Numerous domestic pepper growers have already changed to soilless, nutrient solution cultures, where the cultivar almost exclusively used is the conical, white-fleshed 'HÓ-F1'. This is the favourite cultivar type in Hungary but also the most susceptible to damage by thrips species (MOLNÁR et al. 2008). Therefore in Hungary, key biocontrol agents like *Amblyseius swirskii* and *Orius laevigatus* are widely used for the control of thrips pests by most sweet pepper growers as part of Integrated Pest Management.

Amblyseius swirskii is a beneficial predatory mite, native to the eastern Mediterranean region (RAGUSA & SWIRSKI 1977). Many studies have shown that the omnivorous *A. swirskii* can feed and reproduce on a wide range of food sources, such as greenhouse whitefly (*Trialeurodes vaporariorum* WESTWOOD, 1856), tobacco whitefly (*Bemisia tabaci* (GENNADIUS, 1889)), acrid mite (*Tyrophagus putrescentiae* (SCHRANK, 1781)), broad mite (*Polyphagotarsonemus latus* (BANKS, 1904)), tomato russet mite (*Aculops lycopersici* (MASSEE, 1937)), spider mite (*Tetranychus urticae* KOCH, 1836), chilli thrips (*Scirtothrips dorsalis* HOOD, 1919) and also feed on the pollen of various plant species (MOMEN & EL-

SAWAY 1993, EL-SHERIF et al. 1999, NOMIKOU et al. 2001, 2002, 2004, MESSELINK et al. 2006, 2008, ARTHURS et al. 2009, VAN MAANEN et al. 2010, PARK et al. 2010, XU & ENKEGAARD 2010, NOMIKOU et al. 2010, LEE 2011). According to BLOCKMANS et al. (2005) and WIMMER et al. (2008) *Amblyseius swirskii* is a promising agent in biological control systems because it predaes, reproduces and develops on western flower thrips and onion thrips as well. Studies have reported the efficiency of *A. swirskii* against *Frankliniella occidentalis* on vegetable crops and also their good control on sweet pepper (VAN HOUTEN et al. 2005, BELDA & CALVO 2006, KUTUK et al. 2011).

Orius WOLFF, 1811 species can feed on various kinds of arthropods but display a certain preference for thrips. Their advantage in thrips control is that they prey on both larval and adult thrips (WITTMANN & LEATHER 1997). The anthocorid bug most commonly used in Europe as an effective biological control agent of western flower thrips is the generalist predator *Orius laevigatus* (WEINTRAUB et al. 2011, MESSELINK et al. 2012).

The aim of this study was to observe whether the combined release of two biological control agents would be sufficient to suppress the phytophagous thrips population in greenhouse sweet pepper.

MATERIAL AND METHODS

Experimental site

The trial was conducted at the Experimental and Research Farm, Faculty of Horticultural Science, Corvinus University, Budapest, in 2010 and 2011. The usual unheated growing season for the production of peppers in a Filclair greenhouse extends from mid-April to November under Hungarian climatic conditions. Sweet pepper seedlings were transplanted onto rockwool slabs in a 1500 m² multi-span Filclair greenhouse on 16 April 2010 and 18 April 2011. This is an air-inflated, double-layer polyethylene greenhouse, 52 m long and 28.8 m wide with a gutter height of 3.75 m. The ventilation is controlled by an automatic system that opens or closes the vents. The *Capsicum annuum* cv. HÓ F1 sweet pepper variety was planted, an indeterminate hybrid variety (white fruited) that is the most widely used variety in soilless cultivation in Hungary (TOMPOS 2006). The sweet pepper plants were grown in twin rows and spaced apart to a density of 4.04 plants per square metre. The plants were trellised to a two-stem pruning system. Plants were watered and fertilized by a drip irrigation system, according to standard grower's practice.

Natural enemies and monitoring

The biological control strategy was based on the release of *Amblyseius swirskii* and *Orius laevigatus* for thrips control and several treatments of 0.1% *Bacillus thuringiensis*

BERLINER, 1915 var. *kurstaki* were applied to control cotton bollworm (*Helicoverpa armigera* HÜBNER, 1809). No other pesticides were applied. Dosing and timing of natural enemy releases were determined by the local distributor and beneficial arthropods were released when the pepper plants started flowering in both years. The release rates and times in both years are shown in Table 1.

Table 1. Timing and release rates for natural enemies in 2010 and 2011.

Release time		Natural enemies	Type of product	Release rate
2010	7 May	<i>A. swirskii</i>	Swirskii Mite Plus™	167/m ²
		<i>O. laevigatus</i>	Thripot™	1.3/m ²
	14 May	<i>A. swirskii</i>	Swirskii-Mite™	33.3/m ²
	20 May	<i>O. laevigatus</i>	Thripot™	0.7/m ²
	19 August	<i>O. laevigatus</i>	Thripot™	1/m ²
2011	12 May	<i>A. swirskii</i>	Swirskii Mite Plus™	83/m ²
	19 May	<i>O. laevigatus</i>	Thripot™	2/m ²

The predatory mites and bugs were produced by Koppert Biological Systems (Netherlands). *Amblyseius swirskii* was packaged in paper sachets (Swirskii-Mite Plus™®) and in 500 ml bottles (Swirskii-Mite™®). The mites were released by hanging the paper sachets in the first branch of the pepper plants to distribute the predatory mites in the crop. When the bottle shipment was applied the mites were sprinkled from the bottle onto the surface of the pepper leaves. The anthocorid bug was supplied in bottles (Thripot™®) and the bugs were released uniformly on the surface of the pepper leaves with the carrier materials.

Bacillus thuringiensis var. *kurstaki* was applied on 15 June, 11 and 17 July, 2 and 16 August in 2010, and on 9 and 21 July, 4 August in 2011 to prevent damage of the cotton bollworm.

Sampling

The arthropods were collected by picking flowers directly into plastic vials separately containing 70% ethanol. 50 open blooms were randomly chosen and collected every 14 days from the entire area to monitor the thrips and predator populations, 9 and 11 times in 2010 and 2011, respectively. From the samples, thrips and mites were mounted on separate microscope slides for each flower and identified to species using a compound microscope. Adult thrips were identified based on the identification guide of MOUND & KIBBY (1998) and the key of MORITZ et al. (2001). Juvenile stages of thrips were distinguished and the L2

stages were identified using the key by VIERBERGEN et al. (2010). Predatory mites were identified using the key by SWIRSKI et al. (1998). The adults of the bugs collected were identified using PÉRICART'S (1972) guide.

Statistical analysis

Statistical analysis was carried out using IBM SPSS 22 software. The abundance of different arthropod species (*Amblyseius swirskii*, *Orius laevigatus*, thrips species) was monitored during the entire production period and analysed using a one-way repeated measures ANOVA model with species impact as a factor. In order to normalize the data 10 x 20 flowers were selected at random from each sample of 50 flowers. The sum of the individuals observed in the 20 chosen flowers was calculated for all species.

As the assumption of sphericity was violated (2010: $\epsilon = 0.64$, 2011: $\epsilon = 0.64$), we applied Greenhouse-Geisser's degrees of freedom correction (GEISSER & GREENHOUSE 1958) to test for within-subjects effects. Normality of residuals was proven according to skewness and kurtosis (TABACHNICK & FIDELL 2012). Homogeneity of variances was tested by Levene's test ($p > 0.05$) and, depending on its significance level, within-subjects effect analysis was followed by Tukey's or Games-Howell's post hoc test to detect homogeneous groups of species at different observation times. Pairwise comparisons to explore the observation time effects were made using Bonferroni's test.

RESULTS

Proportion of thrips species in sweet pepper flowers

We found 6 thrips species in the sweet pepper flowers in 2010, most frequently *Thrips flavus* SCHRANK, 1776 (41.7%), *Thrips tabaci* (25.3%), *Frankliniella intonsa* TRYBOM, 1895 (22.9 %) (Fig. 1). Adults of *Frankliniella occidentalis* were not found in the flowers at all. In the same greenhouse in 2011, *Thrips tabaci* (90.3%) was the dominant thrips species and the most dangerous pest of sweet pepper, *Frankliniella occidentalis* was not encountered at all throughout the growing season (Fig. 2).

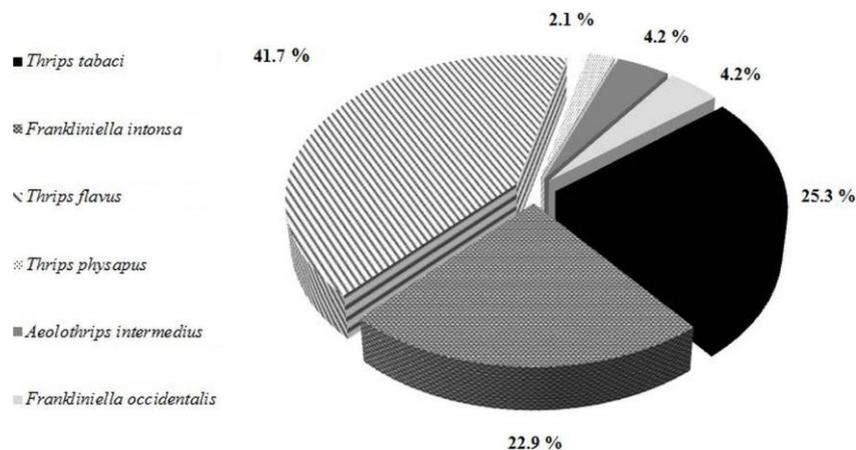


Fig. 1. Proportion of thrips species in flowers of greenhouse sweet pepper in 2010.

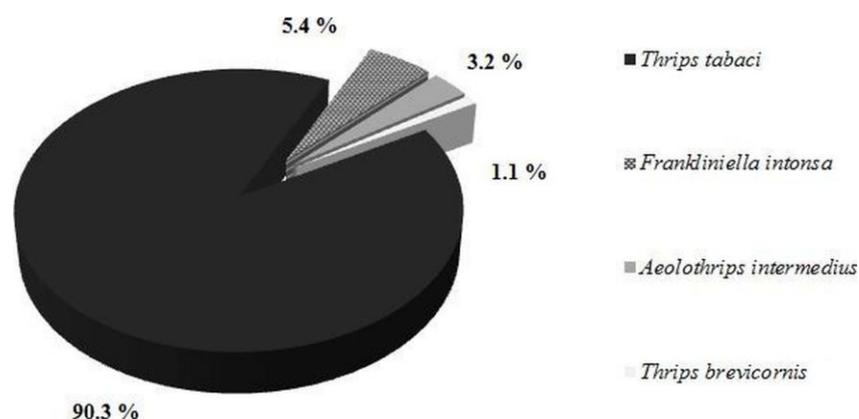


Fig. 2. Proportion of thrips species in flowers of greenhouse sweet pepper in 2011.

Trend of the predatory arthropods and thrips assemblages

The effect of observation time was significant ($F_{\text{time}}(5.14; 138.8)=30.22; p<0.001$) in 2010. The density of thrips species was nearly 0.1 thrips per flower from the beginning of sampling and remained at this level until the middle of August. The abundance of predatory arthropods reached its maximum in June and July, after which it started to decline steeply, and on 12 August we did not find any predatory bug in the samples (Fig. 3). The density of thrips started to rise slightly in late August (0.5 per flower) followed by an increase in the abundance of *Amblyseius swirskii*, but the population density of the predatory bug remained at a low level despite its repeated introduction on 19 August; this release was made because the predator had apparently not established itself (Fig. 3).

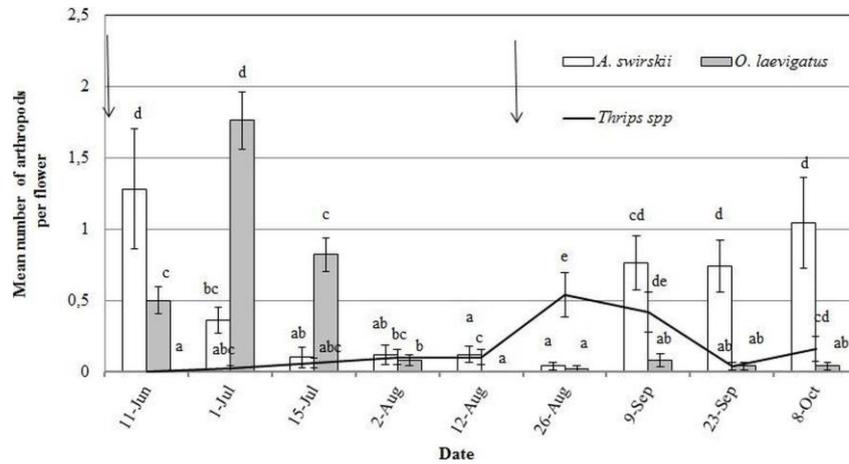


Fig. 3. Trend and mean (\pm SE) numbers of the natural enemies and thrips species in flowers of sweet pepper plants in 2010. Standard error bars are indicated. Arrows indicate the predator release dates. Different letters indicate significant difference (Bonferroni's, $p < 0.05$).

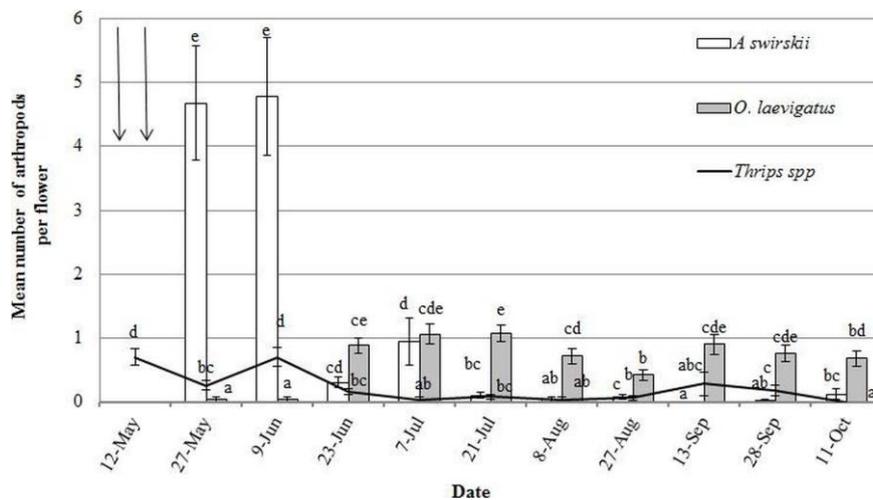


Fig. 4. Trend and mean (\pm SE) numbers of the natural enemies and thrips species in flowers of sweet pepper plants in 2011. Standard error bars are indicated. Arrows indicate predator release dates. Different letters indicate significant difference (Bonferroni's, $p < 0.05$).

The effect of time ($F_{\text{time}}(5.73; 154.79) = 49.29$; $p < 0.001$) was significant in 2011. When the predators were released in the middle of May the polyphagous thrips had already been present in the crop at an infestation level of 0.7 thrips per flower. Soon after the release of

predators the thrips population started to decline slowly (from 0.7 to 0.01 per flower) and the number of individuals remained very low from the end of July until the end of the growing season. The predatory mite population attained a peak of 4.78 individuals per flower within 4 weeks of the release and the predatory bug population density reached a level of approximately 1 bug per flower (Fig. 4). From early June to the end of July the abundance of *A. swirskii* dropped to approximately 1 mite per flower and remained at a very small level (0.1 per flower) for the rest of the season, with the exception of the sampling on 13 September, when no predatory mite was found in the pepper flowers. The predatory anthocorid bug population increased a month following its release, after which the population density remained at or slightly below 1 bug per flower.

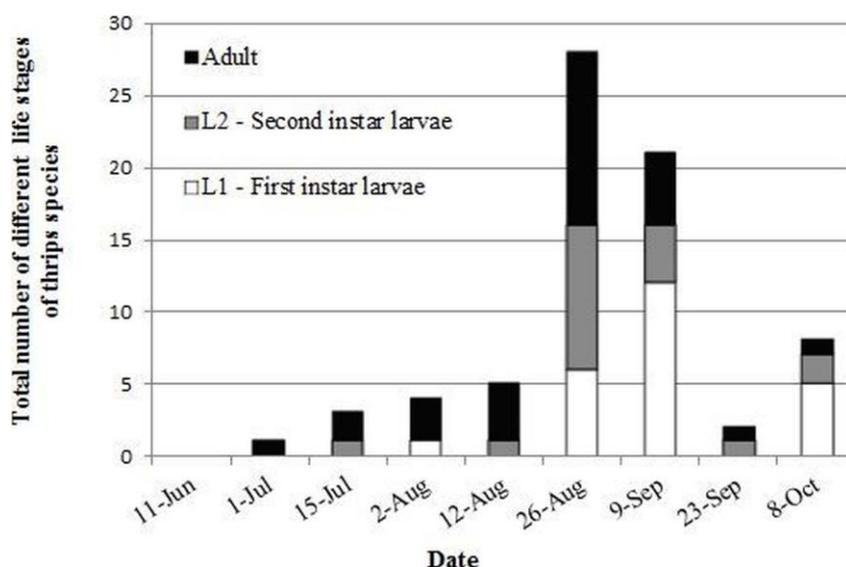


Fig. 5. Population density of different life stages of thrips in 2010.

Abundance of different developmental stages of thrips

Orius predatory bugs can feed on every feeding stage of thrips but the predatory mites feed primarily on their first instar larvae. In 2010 this vulnerable larval stage was absent or its proportion in the thrips population was extremely low until the end of August, when more first instar larvae were observed in the samples (Fig. 5). Overall, the population density of thrips as available prey for predators was fairly low in 2010. Every feeding stage of thrips was present in most of the samples in 2011, but the proportion of adults was significant until the third decade of June. The overall population density of thrips was

somewhat bigger at the beginning of sampling than at its peak in 2010, then it dropped to a significantly lower infestation level for the rest of the study (Fig. 6).

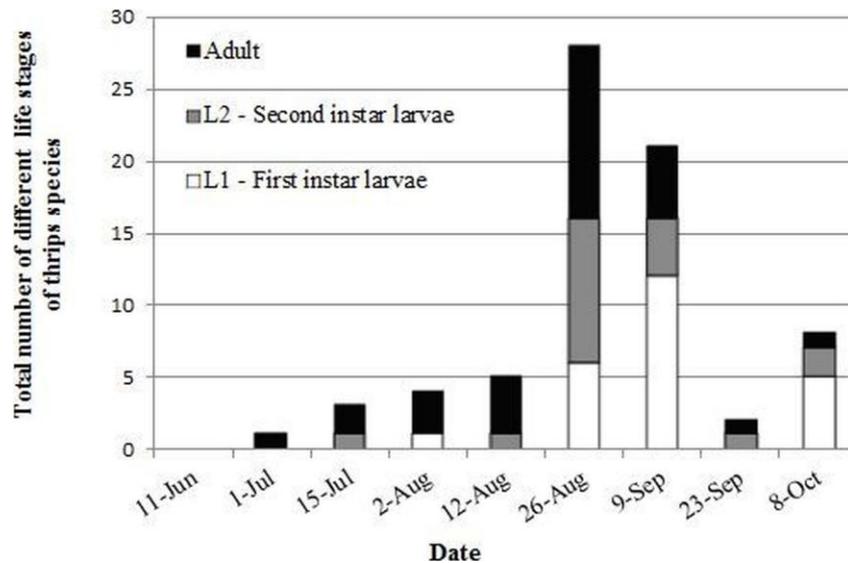


Fig. 6. Population density of different life stages of thrips in 2011.

DISCUSSION

The overall population density of the thrips assemblages was almost identical in both years and thrips damage to pepper fruits was negligible. But the composition of the thrips assemblages in sweet pepper flowers differed considerably between the years of this study. The species composition in the immediate surroundings of the greenhouse may have been influenced by climatic conditions. The weather conditions were far from normal in 2010: that year was much more humid with extreme levels of precipitation, which could have affected the build-up of thrips populations in the vicinity of the greenhouse. In 2011 weather conditions were much closer to the usual, i.e. hot, dry days for most of the summer. The higher temperature and dry conditions in 2011 could have favoured *T. tabaci* more than the other species, which were more dominant in 2010, resulting in the absolute dominance of onion thrips. On the basis of OROSZ's (2006) surveys in Hungary, *Frankiniella occidentalis* and *Thrips tabaci* occur frequently on numerous weed species (e.g. *Taraxacum officinalis* (L.) WEBER EX F.H. WIGG, *Stellaria media* (L.) VILL., *Trifolium* spp.) growing near the greenhouses, and *Thrips tabaci* larvae were observed on *Stellaria media* and

Capsella bursa-pastoris (L.) MEDIK. from the end of April, so they could well have infested greenhouse crops from these weed reservoirs.

A continuously reproducing population of *Orius laevigatus* was not successfully established on pepper in 2010, possibly due to the lack of sufficient thrips prey (<0.1 thrips per flower) in the first half of the season; moreover, this species appeared to have abandoned the greenhouse by the beginning of August. Then the thrips outbreak in the flowers rose to its peak (0.54 thrips per flower) until suppressed by *Amblyseius swirskii* returning from the foliage. This observation is confirmed by the result of SANCHEZ & LACAZA (2002), namely, that the population of *Orius* spp. remained at a low level or declined when prey were scarce. We hypothesize that the re-establishment of the predatory mite population in sweet pepper flowers was possible only in the absence of the more competitive predatory bug. The predatory bug was unable to build up an appropriately-sized population and thus contribute to the suppression of thrips pests because of its very small density and slower development. In contrast, the predatory mites in the absence of available thrips prey were able to feed on other arthropod species, for instance on indifferent dust mites present in the crop on the underside of the leaves, next to the main veins, and pepper pollen grains, which were also present on the foliage. They could potentially migrate to the flowers from these reservoirs in the absence of *Orius laevigatus*.

The predatory mites were present in sweet pepper flowers after their high density introduction for a shorter or a longer period in 2010 and 2011, respectively. Then they seemed to disappear from the flowers, despite reaching a peak population density of about 5 mites per flower and the initial thrips abundance being already 0.7 thrips per flower in 2011. Their disappearance coincided with the build-up of the predatory bug population in both years. Since the population density of first instar thrips larvae did not decline at this time in 2011, it is unlikely that the disappearance of *Amblyseius swirskii* was driven by the lack of available prey in June. We hypothesize that the increasing presence of the intra-guild predator *Orius laevigatus* is the more likely cause of the decline of the predatory mite population in the flowers. Whether the predatory mites were consumed by the bigger generalist predatory bug or were outcompeted in the flowers and forced to migrate to the foliage remains unclear: our study did not provide any satisfactory evidence regarding this. However, CALVO et al. (2012) came to the same conclusion: they observed larger *Orius laevigatus* populations in the plots where *Amblyseius swirskii* was released, possibly due to intra-guild predation by the anthocorid. In addition, URBANEJA et al. (2003) pointed out that the release of *Neoseiulus cucumeris* (OUDEMANS, 1930) helped *Orius laevigatus* to become established by providing a supplementary food source, because the anthocorid bug may prey on the predatory mites in the flowers where the bug resides if no other food sources are available. Moreover, CHOW et al. (2010) suggested that *Orius* will switch to feeding on the most abundant food source. Some other authors also reported the risk following the

combined release of anthocorids and phytoseiid mites because the bug could be an intraguild predator (JANSSEN et al. 1998, VENZON et al. 2001, SKIRVIN et al. 2007, CHOW et al. 2008, SHAKYA et al. 2009, CHOW et al. 2010). In addition, we recorded that *Orius laevigatus* nymphs and adults are capable of preying on *Amblyseius swirskii* under laboratory conditions. We therefore conclude that the continuous presence of the predatory bugs in the sweet pepper flowers in 2011 most likely prevented the re-establishment of *A. swirskii* population during the remainder of the pepper production period.

Commercially available phytoseiid predatory mites like *Amblyseius swirskii* and anthocorid flower bugs like *Orius laevigatus* are often used simultaneously for the biological control of severe thrips infestations in sweet pepper cultivation in Hungary. Our observations demonstrated that the mass proliferation of thrips was effectively prevented by the combined release of natural enemies and was independent of the species composition of Thysanoptera. In both years, its population density was below (mostly <0.5 per flower) the estimated economic threshold range of 0.7-2.1 adults or nymphs per flower (PARK et al. 2007). The release ratio and timing was applied with the doses recommended by the distributor and there was no need to use pesticides at all. However, this low infestation level of thrips suggests that a single predator release strategy (presumably *Amblyseius swirskii*) could have suppressed thrips effectively and maintained their numbers below the threshold in greenhouse sweet pepper, but we have no satisfactory evidence to substantiate this. WEINTRAUB et al. (2011) did not find a significant difference in thrips density in sweet pepper between treatments combining *Orius laevigatus* and *Amblyseius swirskii*, although a significant improvement was observed when *Amblyseius swirskii* was combined with *Orius laevigatus* in thrips control compared with *Amblyseius swirskii* released alone or over the untreated plot. The capability of both predators to suppress thrips populations alone in sweet pepper was demonstrated by BELDA & CALVO (2006), WEINTRAUB et al. (2011), VAN HOUTEN et al. (2005), CHAMBERS et al. (1993), TAVELLA et al. (1996) and BOSCO et al. (2008). On the other hand, standard grower's practice recommends the combined release strategy for the sake of reducing the incidence of the Tomato Spotted Wilt Virus (CALVO et al. 2012).

In conclusion, the combined use of these two predatory species could offer better security in the management of thrips pests: if one of them is not established successfully in the crop, then the other may still provide efficient control. If there are some other arthropod species damaging the foliage of sweet pepper, e.g. spider mites, broad mites or whiteflies, the presence of *Amblyseius swirskii* might prove even more useful. In the absence of these pests the release of *A. swirskii* may be unnecessary, provided the establishment of *Orius laevigatus* in the crop is successful. In this scenario the more effective predator of anthophilous thrips, *O. laevigatus*, could force *Amblyseius swirskii* out of the flowers or

even consume it. However, if the establishment of *Orius laevigatus* is unsuccessful, *Amblyseius swirskii* can respond to increasing thrips pressure by recolonizing the flowers.

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