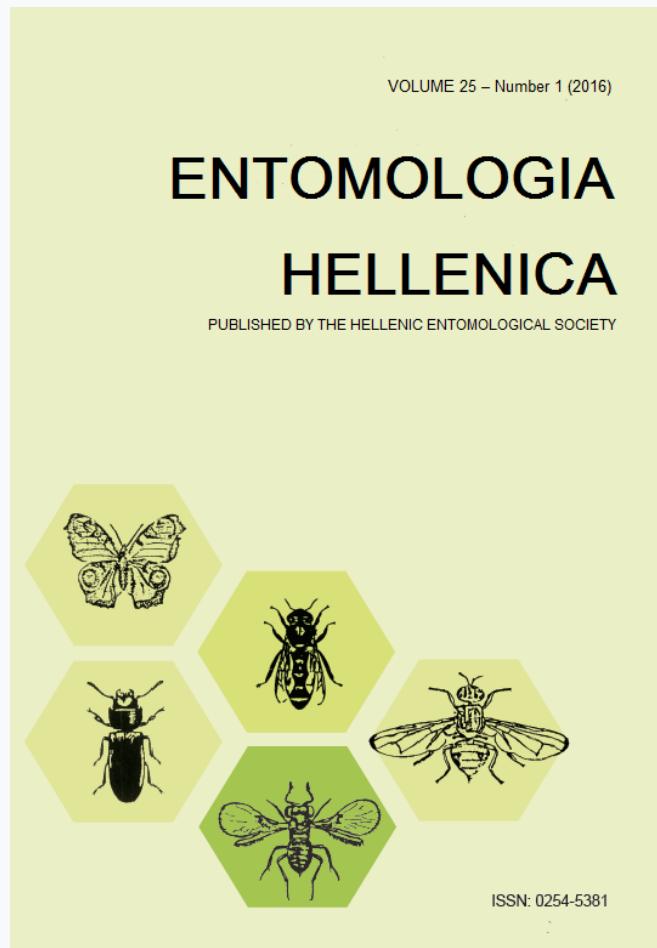


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Effects of sticky traps on *Dacnusa sibirica*, *Diglyphus isaea* and *Nesidiocoris tenuis*

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ABSTRACT

The potential of commonly used sticky traps (yellow, blue and transparent) to attract the two major natural enemies of the leaf miners, *Dacnusa sibirica* Telenga (Hymenoptera: Braconidae) and *Diglyphus isaea* (Walker) (Hymenoptera: Eulophidae) was recorded. In addition, we studied their efficacy to capture the effective predator *Nesidiocoris tenuis* (Reuter) (Hemiptera: Miridae) which in high population levels may cause damage on tomato crop. The captures of *D. sibirica* and *D. isaea* were negligible. Captures of *N. tenuis* adults were significantly higher on yellow and blue colour traps than on transparent traps. The difference of captures between yellow and blue traps was not significant. Furthermore, the effectiveness on *N. tenuis* was much increased when the traps were established next to the tomato plant apex. The results show that the use of coloured sticky traps and release of the leafminer parasitoids may be combined. Furthermore, it was clearly shown that sticky traps may not be compatible with the release of the generalist predator *N. tenuis*; however, yellow and blue traps should be further evaluated for its mass trapping.

KEY WORDS: leafminer parasitoid, mass trapping, tomato.

Introduction

Yellow sticky traps (YST) are well known as a means to monitor the population of several pest species such as whiteflies (Gillespie and Quiring 1992, Park et al. 2011, Lu et al. 2012), thrips (Casey and Parrella 2002, Ranamukhaarachchi et al. 2007, Pizzol et al. 2010), aphids (Straw et al. 2011, Rajabpour and Yarahmadi 2012) and leafminers (Taha et al. 2012) in greenhouse crops for timely application of control measures. The YST have been also evaluated with positive results in the decision making for managing whiteflies in greenhouse crops (Pinto-Zevallos and Vänninen 2013). In addition, mass

trapping using YST has been also considered as a key component for pest management in greenhouse crops (Steiner et al. 1999, Kaas 2005). Lu et al. (2012) showed the high potential of YST to suppress whitefly population in greenhouses.

Despite their well-known use in Integrated Pest Management (IPM) programmes, interestingly, their effects on parasitoids of greenhouse pests have been rarely addressed. Hoelmer and Simmons (2008) stated that although YST did caught a significant number of *Eretmocerus* sp. (Hymenoptera: Aphelinidae) were much more attractive to the whitefly *Bemisia tabaci* (Gennadius) (Hemiptera:

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Aleurodidae). Similarly, YST caught high numbers of the target whitefly *Bemisia argentifolii* (Gennadius) in cotton fields and only a few of the non-target parasitoid *Eretmocerus eremicus* Rose & Zolnerowich (Hoelmer et al. 1998). However, Chu et al. (1998) demonstrated that the YST had an adverse effect on the whitefly parasitoids *Encarsia* sp. (Hymenoptera: Aphelinidae) and *Eretmocerus* sp. and to overcome this adverse effect they designed a special trap called “CC trap” which gives the parasitoid adults the chance to escape.

The clarification of the effect of YST on parasitoid population will enhance the effectiveness of pest control in greenhouse crops since mass trapping combined with biological control may enhance whitefly control (Lu et al. 2012). Gu et al. (2008) stated that YST and parasitoids together offered an effective method to suppress *B. tabaci* in a greenhouse whereas, Moreau and Isman (2011) recommended assessing the combination of YST with other control strategies. However, the use of YST traps in mass trapping or in combination with natural enemies of whiteflies may cause disturbance in the use of biological control against other greenhouse pests, such as leafminers. Leafminers are major pests of greenhouse vegetable and ornamental crops. Mass trapping with the use of YST has been proven effective in the control of leafminers in eggplant crops (Durairaj et al. 2007). Similarly, mass trapping can be effective to control leafminers in greenhouses equipped with insect exclusion nets (Civelek et al. 2004), whilst one YST/10m² resulted in effective control for leafminers (Yabas et al. 2000).

Despite the positive evidence and the common use of YST for monitoring or mass trapping for leafminers or other pests, the effects of YST on the major biological control agents of leafminers, i.e. the endoparasitoid *Dacnusa sibirica* Telenga 1935 (Hymenoptera: Braconidae) and the

ectoparasitoid *Diglyphus isaea* (Walker, 1838) (Hymenoptera: Eulophidae) have not been addressed. *Dacnusa sibirica* lays its eggs in young host larvae which continue to feed and pupate. This parasitic wasp is a key element for leafminer control under low temperatures since the optimum temperature for oviposition is 20°C (Minkenberg 1990). The ectoparasitoid wasp *D. isaea* is more effective and lays more eggs at warmer periods (optimum 25°C) (Johnson and Hara 1987, Minkenberg 1990). Considering that the control of leafminers under the IPM is commonly based on these two parasitoids, the attractiveness of YST to leafminer parasitoids remains a gap of knowledge that needs to be investigated.

Nesidiocoris tenuis (Reuter) (Hemiptera: Miridae) is an effective predator of major pests such as whiteflies, thrips, leafminers, spider mites and lepidopteran species when released early in the season or pre-plant released in tomato crops (Torreno 1994, Calvo and Urbaneja 2003, Desneux et al. 2010, 2011, Calvo et al. 2012a,b, Perdikis et al. 2015). Additionally, *N. tenuis* can effectively control the tomato leafminer *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) by preying on eggs and mostly first-instar larvae (Urbaneja et al. 2009, Mollá et al. 2011, Cabello et al. 2012, Calvo et al. 2012b, Sánchez et al. 2014). That makes *N. tenuis* a valuable biological control agent in tomato crops and for this reason is widely used commercially (Symondson et al. 2002, van Lenteren 2012). Nonetheless, *N. tenuis* has been reported to cause necrotic rings in leaf and flower petioles or even injuries on fruits when occurs in high densities (El-Dessouki et al. 1976, Sánchez et al. 2008, Calvo et al. 2009, 2012a,b, Sánchez et al. 2015). The intensity of injury to tomato has been observed to decrease with increased availability of prey (Calvo et al. 2009, Arnó et al. 2010). According to the study of Sánchez (2008),

no yield reduction is expected from *N. tenuis* phytophagy for values <0.65 individuals per leaf, independently of the whitefly abundance, nor for up to 5 individuals per leaf when whiteflies are more than 26 immatures per leaf. When control measures are required against *N. tenuis* mass trapping using YST could offer a valuable alternative strategy to chemical control.

The aim of this study was to investigate whether the two major natural enemies of the leaf miners *D. sibirica* and *D. isaea* could be entrapped on different sticky traps (yellow, blue and transparent) and on the other hand to search whether those traps could attract the potential pest, *N. tenuis*.

Materials and Methods

The experiments were conducted in an organic tomato (hybrid "Formula F1") greenhouse of 50x50m located at Marathonas, Attiki (Greece). The plants were about 2.3m in high bearing tomato fruits and inflorescences. During the experimentation period the temperature inside the greenhouse was automatically controlled in a range of 18 to 25°C and the photoperiod was natural.

Experimental procedure

The effects of commercially available yellow and blue sticky traps (Horiver and Horiver-TR, Koppert B.V., The Netherlands, respectively) on *D. isaea* and *D. sibirica* were evaluated. As a control, a transparent PVC leaf (0.4mm thick) covered with entomological glue in both sides was used. The dimensions of all traps were 10x25 cm (width: length). The comparative attraction was recorded by placing traps of each colour (yellow, blue and transparent) alternately in a circle around the release point of the parasitoids. In the course of the experiments a high

population of *N. tenuis* was spontaneously established in the greenhouse. Thus, simultaneously to the effects of the traps on the parasitoids their effects on *N. tenuis* were investigated as well.

On October 20th 2012 two circles with traps were created, each circle was 12m in diameter and the distance between them was 12m. The distance between traps was about 2m. Six traps were placed at a height of 1.5 m with color sequence in each circle (Figure 1). Each sticky trap was hanging on an individual rope between tomato plants at a distance of 0.3m from the plant canopy.

Another experiment was initiated on 27th of November 2012, by placing 18 traps, 6 from each colour in a large single circle 15m in diameter. The traps were placed at the height of 1.5m. At the same time, 6 traps (2 of each colour) were placed in an additional concentric circle (12m diameter) at a height of 2.5m (Figure 2). The average distance between two neighboring traps was about 2.6m for the large and about 3m for the smaller circle at 2.5m height. The traps were removed 7 days after their establishment. The individuals of parasitoids (*D. sibirica*, *D. isaea*) or *N. tenuis* were counted on each sticky trap, under a stereoscope.

Release of parasitoids

In the first experiment, 250 adults of *D. sibirica* (Minusa, Koppert B.V., The Netherlands) and 250 adults of *D. isaea* (Miglyphus, Koppert B.V., The Netherlands) were released at the center of each circle at a height of 1.5 m. In the second trial, 500 adults of *D. sibirica* and 500 of *D. isaea* were again released at the center of the two concentric circles. The bottles were uncapped and the wasps were left undisturbed to exit till the last one. In all cases the parasitoids were released between 16:00 and 16:30 pm.

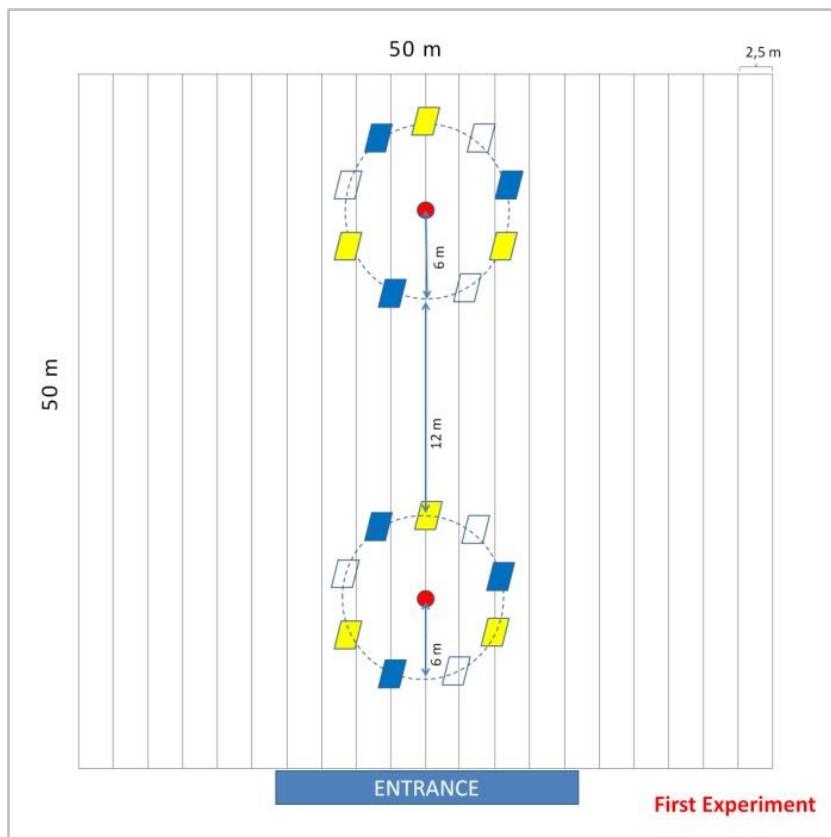


FIG. 1. In this figure the schematic representation of the trap establishment in the first experiment is shown. The traps were placed in two circles (6m radius each) and the distance between them was 12m. The traps were placed at a height of 1.5m. The red dot indicates the center of each circle where the parasitoids had been released. The yellow, blue or without colour rectangles indicate the respective yellow, blue and transparent sticky traps.

Monitoring of *N. tenuis* population densities

On the day of the traps' establishment and for both experiments, the population of the spontaneously established population of *N. tenuis* on the plants was recorded. The number of its nymphs and adults was counted at the upper 4 totally expanded leaves of a lateral stem at a height of about 1.5-2.0 m, on each of 14 plants in the first, and 20 plants in the second trial. In the sampling, tomato plants close to the traps had been randomly selected.

Statistical analysis

The results were analyzed using ANOVA with fixed factors "circle" and the "trap colour" (first experiment) and the "trap colour", for the inner (smaller) circle, in the second experiment. Comparisons between means were performed with Tukey's HSD test ($P < 0.05$). At the case of outer (larger) circle at the second experiment the assumption of equality of variances was not fulfilled, so the Kruskal-Wallis test was used. Analyses were conducted with the statistical package JMP (v. 12.0, SAS Institute 2015).

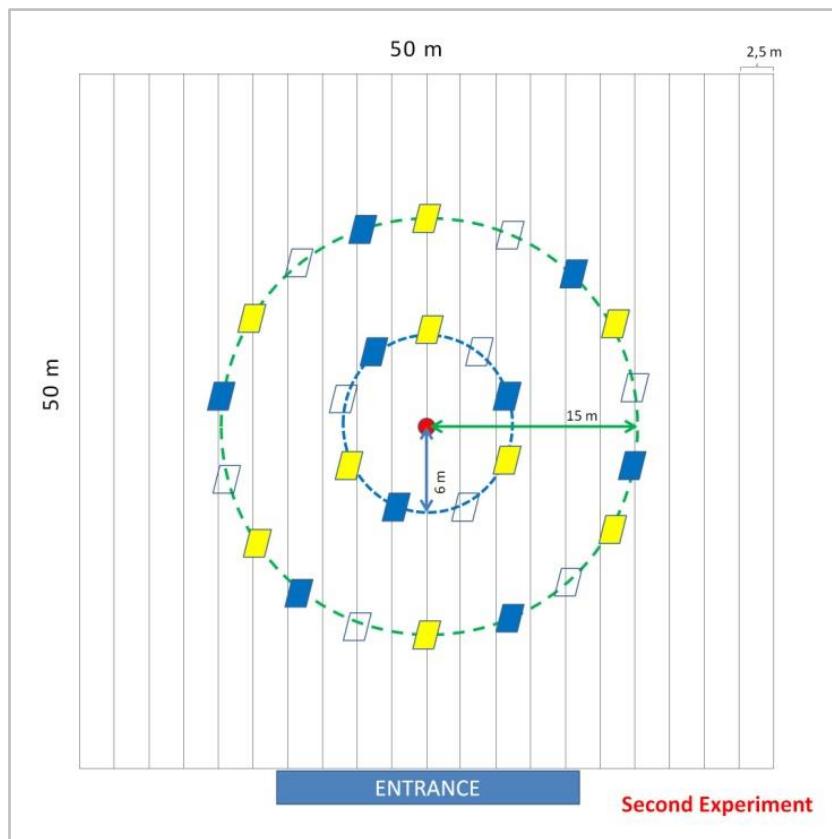


FIG. 2. In this figure the schematic representation of the trap establishment in the second experiment is shown. The traps were placed in two concentric circles. The inner circle was 6m radius and the outer 15m radius. The traps were placed at a height of 1.5m in the inner and 2.5m in the outer circle. The red dot indicates the center of the circles where the parasitoids had been released. The yellow, blue or without colour rectangles indicate the respective yellow, blue and transparent sticky traps.

Results

A very low number of individuals of *D. sibirica* or *D. isaea* were found trapped (7 *D. sibirica* in the first and 1 in the second experiment) whereas 2 individuals of *D. isaea* were found in the second experiment.

In the sampling of the tomato plants in the first experiment, 3.7 ± 0.4 and in the second 9.3 ± 0.9 *N. tenuis* individuals (nymphs and adults) were recorded on average, per stem. On the tomato plants

only whiteflies and *T. absoluta* larvae were present but at very low numbers.

In the first experiment, the numbers of *N. tenuis* on traps was not different between the two circles ($F=1.39$, $df=1,17$, $P=0.26$) (Figure 3). However, coloured traps caught a significantly higher number of *N. tenuis* adults in comparison with transparent traps ($F=11.92$, $df=1,17$, $P<0.001$). Yellow traps were more attractive for *N. tenuis* capturing 150.0 ± 37.8 and 86.3 ± 5.9 adults on average per trap, for the first and the second circle, respectively ($F=6.32$, $df=1,8$, $P<0.05$ and

$F=14.98$, $df=1,8$, $P<0.05$, respectively). Blue traps caught 85.7 ± 28.9 and 81.7 ± 15.3 adults of *N. tenuis*, and the sticky

transparent traps caught only 12.0 ± 0.6 and 17.3 ± 5.4 adults of *N. tenuis* on average for each circle, respectively.

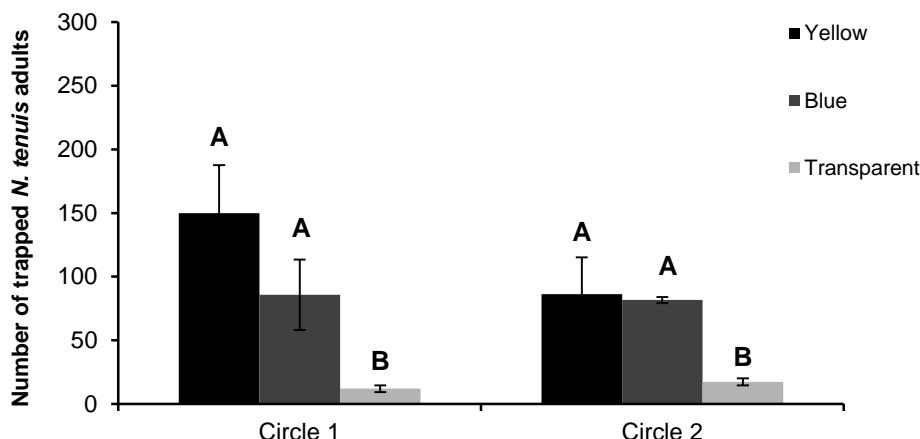


FIG. 3. Experiment 1: captures of *Nesidiocoris tenuis* (mean \pm s.e.) on yellow, blue and transparent sticky traps arranged in two circles (Circle 1 and Circle 2) at a height of 1.5 m in a tomato greenhouse after 1 week of exposure. [Means followed by the same capital letter are not significantly different at each treatment (ANOVA, Tukey's HSD, $P<0.05$)].

In the second experiment the effect of coloured traps was also significant both for the lower and the upper circle ($F=11.16$, $df=2,5$, $P<0.05$ and $\chi^2=12.32$, $df=2$, $P<0.01$, respectively) (Figure 4). Captures were significantly higher on yellow and blue colour traps than on transparent ones at both circles. However, there was no difference between the captures on yellow vs. blue traps at each level.

Discussion

Coloured traps had no negative effects on the parasitoids of leafminers *D. sibirica* and *D. isaea* despite of releasing them at high numbers and very close to the traps. Although in the greenhouse the tomato plants had no infestation of leafminers, *D. isaea* adults can survive for 6 days without food source (host or non-host diet) (Zhang et al. 2011), so after their release they should have been able to search for host; nonetheless only very few of them had

been entrapped. Thus, although more extensive and replicated experiments should be conducted under variable conditions, the information gathered in the present work indicates that the yellow and blue traps may not have a significant adverse effect on both parasitoid species. Accordingly, earlier studies have shown that YST can be compatible with the use of whitefly parasitoids (Gu et al. 2008, Moreau and Isman 2011, Lu et al. 2012). Our results support those studies and indicate that the potential to combine biological control and mass trapping in the control of leafminers can be further evaluated in IPM programmes in greenhouses.

Interestingly, the results clearly showed the potential of yellow but also blue traps for capturing *N. tenuis*. Furthermore, it was shown that high numbers were caught when the traps were established next to the tomato apex, most likely because *N. tenuis* numbers are higher there (Perdikis et al. 2014).

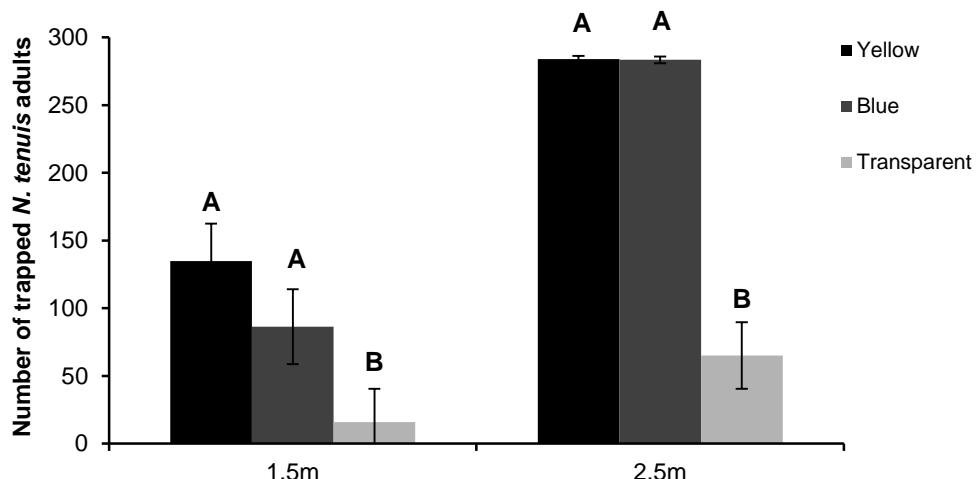


FIG. 4. Experiment 2: captures of *Nesidiocoris tenuis* (mean \pm s.e.) on yellow, blue and transparent sticky traps placed at 1.5 m and 2.5 m after 1 week of exposure. [Means followed by the same capital letter are not significantly different at each treatment (Kruskal-Wallis and ANOVA, Tukey's HSD test, for each circle, respectively, $P < 0.05$)].

Arnó et al. (2010) reported that 88.5% of *N. tenuis* population was observed on the upper part of the plant. These results indicate an alternative way useful in the effort to reduce the numbers of this mirid when reaching high levels. This is a valuable alternative to chemical control, since yellow and blue traps are widely applied and can be rather easily incorporated in IPM strategies against this pest. On the other hand, our results show that the use of yellow or blue sticky traps may not be compatible with the release of *N. tenuis*, as reported for other useful generalist mirid predators, which are trapped on them (Ingegno et al. 2009, Stansly et al. 2009). The potential of coloured sticky traps to control *N. tenuis* has been overlooked according to our literature review.

In conclusion, during our experiments we were able to combine the coloured traps and leafminer parasitoids without recording any adverse effect on the

number of the natural enemies. Additionally, we showed that a relatively high number of *N. tenuis* individuals were caught on these traps indicating the potential of these traps to be used as an alternative means to control *N. tenuis*, when necessary. Further experiments are needed to clarify the role of traps in integrated pest control.

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Επίδραση κολλητικών παγίδων στο *Dacnusa sibirica*, *Diglyphus isaea* και *Nesidiocoris tenuis*

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ΠΕΡΙΛΗΨΗ

Οι κολλητικές παγίδες χρώματος χρησιμοποιούνται ευρέως σε θερμοκηπιακές καλλιέργειες για την παρακολούθηση αλλά και την μαζική παγίδευση πολλών εχθρών όπως οι αλευρώδεις, οι αφίδες, οι θρίπες και οι φυλλορύκτες. Όμως, η επίδραση των κολλητικών παγίδων στους φυσικούς εχθρούς όπως παρασιτοειδή και αρπακτικά έχει μελετηθεί ελάχιστα. Στην παρούσα μελέτη, διερευνήθηκε η δυνατότητα των κολλητικών παγίδων (κίτρινου και μπλε χρώματος) να προσελκύνουν τους δυο κυριότερους φυσικούς εχθρούς των φυλλορυκτών, το *Dacnusa sibirica* Telenga (Hymenoptera: Braconidae) και το *Diglyphus isaea* (Walker) (Hymenoptera: Eulophidae) και η επίδρασή τους στην παγίδευση του *Nesidiocoris tenuis* (Reuter) (Hemiptera: Miridae). Το τελευταίο είναι ένα αποτελεσματικό πολυφάγο αρπακτικό όμως σε περιπτώσεις που ο πληθυσμός του αυξηθεί πολύ μπορεί να προκαλέσει ζημιά στην καλλιέργεια της τομάτας. Οι συλλήψεις του *D. sibirica* και του *D. isaea* ήταν αμελητέες. Οι συλλήψεις των ενηλίκων του *N. tenuis* ήταν σημαντικά υψηλότερες στις παγίδες με κίτρινο και μπλε χρώμα συγκριτικά με τις διαφανείς κολλητικές παγίδες. Η διαφορά των συλλήψεων μεταξύ του κίτρινου και του μπλε χρώματος δεν ήταν σημαντική. Επιπλέον, οι συλλήψεις του *N. tenuis* αυξήθηκαν περαιτέρω όταν οι παγίδες τοποθετήθηκαν δίπλα στην κορυφή των φυτών. Τα αποτελέσματα δείχνουν ότι η χρήση κολλητικών παγίδων χρώματος και η εξαπόλυτη παρασιτοειδών μπορούν να συνδυαστούν στο πλαίσιο προγραμμάτων ολοκληρωμένης αντιμετώπισης των φυλλορυκτών. Επιπλέον, τα αποτελέσματα δείχνουν ότι η χρήση τους φαίνεται να μην είναι συμβατή με εξαπολύσεις του *N. tenuis* αλλά από την άλλη πλευρά, θα μπορούσαν να αξιολογηθούν περαιτέρω στη μαζική παγίδευσή του, όταν αυτή απαιτείται.