Impact of net houses on the natural regulation of the populations of *Trialeurodes vaporariorum* (Homoptera: Aleyrodidae) and *Tuta absoluta* (Lepidoptera: Gelechiidae), two major tomato pests in Kenya

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Abstract

The abandonment of insecticide treatments to achieve sustainable crop production calls for a combination of methods to obtain satisfactory pest control. To this end, net houses and biological control are two promising methods, though we suspected that most natural enemies are blocked outside the net houses. In Kenya, tomato crops are particularly threatened by *Trialeurodes vaporariorum* (Westwood 1856) and *Tuta absoluta* (Meyrick, 1917) that are targets of most insecticide treatments in this country. We investigated in an on-farm experiment (i) the abundances of *T. vaporariorum* and *T. absoluta* in open fields and in net houses, (ii) the diversity of arthropod natural enemies of these pests, and (iii) the effect of net houses on the natural regulation. To complete, in the laboratory, we checked the capacity of natural enemies to pass through different net types. We identified several natural enemies, mainly the mirid bug *Nesidiocoris tenuis* (Reuter 1895) that was the most abundant predator for both pests. One parasitoid species was also identified for each pest, i.e. *Encarsia formosa* Gahan (1924) and *Copidosoma* sp. for *T. vaporariorum* and *T. absoluta* respectively. Net houses reduced drastically the pest populations. Predators were less abundant under net houses, while parasitoids did not seem to be affected by the nets. *Encarsia formosa* was shown to be able to pass through the different net types, while *N. tenuis* adults were not. These results raise the potential for augmentative biological control under net houses. Using kairomones to attract natural enemies, or introducing natural enemies under the net houses through inoculative releases, are two approaches to increase the abundance of natural enemies under net houses.

Keywords *Nesidiocoris tenuis* · *Encarsia formosa* · Augmentative biological control · Agroecology

Introduction

Pest control methods can interact synergistically or antagonistically. Yet, to avoid pesticide treatments, several integrated agroecological control methods are required to achieve satisfactory control (Gurr and Kverdaras 2010;

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Kungu et al. 2019). This could involve multi-tool control programs, e.g. physical crop protection (net houses) and biological control, as already implemented in European greenhouses (Calvo et al. 2012; Chailleux et al. 2012). However, studies are required to determine how this combination could be effectively applied in net houses in Africa, where pesticides are still heavily used (De Bon et al. 2014).

Tomato crops in Africa suffer massive damage caused by large caterpillars such as the cotton bollworm *Helicoverpa armigera* (Diatte et al. 2018). Net houses were successfully developed as a control method to protect tomato plants from such large pests (Achieng'a et al. 2016), yet one drawback has been noted—small arthropod pests are able to penetrate and thrive under net houses and destroy the crops if no insecticides are sprayed (Simon et al. 2014). We hypothesize that small pests escape the

control of their natural enemies that might be too big to pass through the net (Majumdar et al. 2015). Moreover, larger pests sometimes also ultimately manage to invade the net houses through the entrances during the coming and going of personnel for agricultural practices (Gogo et al. 2014; Simon et al. 2014). Inside net houses, pests are mostly controlled using chemical insecticides, stressing the need for biological control methods adapted to this context (Nordey et al. 2017; Ramasamy and Ravishankar 2018). Some biological control techniques, such as augmentative biological control, would be facilitated by the net-confined environment.

Major pests threatening tomato production under net houses include the invasive tomato leafminer, *Tuta absoluta* (Meyrick 1917) (Lepidoptera: Gelechiidae) and the worldwide whitefly *Trialeurodes vaporariorum* (Westwood 1856) (Homoptera: Aleyrodidae). *Tuta absoluta* is a devastating tomato pest originating from South America (Guillemaud et al. 2015). Since 2004, it has rapidly spread and caused heavy tomato crop loss throughout the Mediterranean Basin (Desneux et al. 2010), Africa (Mansour et al. 2018; Rwomushana et al. 2019) and Asia (Han et al. 2019). *Trialeurodes vaporariorum* is a sucking pest native to Central and South America (Mound & Halsey 1978). As one of the most important crop pests and virus vectors (Jones 2003), it also causes substantial tomato crop loss in Kenya (Gamarra et al. 2016; Kimathi et al. 2020). Whiteflies are small-sized insects and are suspected to be able to pass through most nets, and *T. absoluta* females can oviposit on nets and subsequently the hatched larvae can produce a silk thread (Desneux et al. 2010) that they use to drop onto the tomato plants (AC personal observation). Many different chemical pesticides are applied under net houses in Kenya to control these two crop pests (Njuguna 2000; Nderitu et al. 2018).

Natural enemies are known for both species, particularly predatory mirid bugs for *T. absoluta* (Zappala et al. 2013), and parasitoid wasps for *T. vaporariorum* (Perić and Dimic 1997; Perić et al. 2008), yet very little information is available on the natural enemies of these pests in Kenya. Natural regulation protagonists and how they are affected by net protection is an essential information for the development of biological control in net houses. We hypothesize that several indigenous arthropod natural enemies are already present in Kenya for both pests, although that contrary to pests, they could be blocked outside the net house. We conducted a field experiment in Kenya to collect natural enemies and compare tomato crops in open fields and under net houses. Then we carried out laboratory tests to assess the net-crossing capacity of the natural enemies.

Material and methods

On farm-experiment

Study site and experimental design

In partnership with Kenyan farmers, a set up was established in 16 farms located in Kirinyaga, Kiambu, Machakos, Kajiado, and Migori in Kenya (Table 1). Farmers were asked to grow tomato plants both in their net house (0.9 mm mesh size, not treated net) and in an open field (at least 50 m² for both situations). However, not all the farmers accepted to grow open field tomato crops. Three monitoring sessions were conducted, one in the cool dry season (June to July 2017, 12 farms), one in the hot dry season (February 2017, three farms), and one in the short rainy season (November 2017, one farm). Indeed, many fields were affected by bacterial wilt during the hot season, preventing us from conducting more monitoring at this season. The monitoring session of each field depended on farmers' cropping season habits. Monitoring and sampling were conducted once per field. Tomato crop development at the time of our monitoring ranged from 2 weeks after transplanting to the fruiting stage.

Pest and natural enemy recording and identification

An entomological field aspirator (Improved Prokopack Aspirator—Model 1419 from John W. Hock company) was used to sample the predators and parasitoids on tomato plants. Ten entire plants, randomly selected, were sampled in each open field and each protected net house.

To record whitefly abundance and parasitism, three leaflets were sampled per plant (bottom, middle, and top of the plant) on the 10 plants in each open field and each protected net house. The number of whitefly nymphs was counted using a binocular microscope. Then, all leaflets were stuck into small pieces of humid floral foam in boxes (10 cm diameter, 9 cm deep) closed with a 160 μm mesh netting and stored at 25 °C to check for parasitoid emergence. Leaflets were kept for 2 weeks, with parasitoid emergence checked every 2 days.

To record *T. absoluta* abundance, three leaves per plant (bottom, middle and top of the plant) on the 10 plants per field were monitored. The presence of larvae in mines was confirmed by shining a torch through the leaves. Leaflets containing larvae were collected in each field when available, and kept in aery boxes (10 cm diameter, 9 cm deep) for 2 weeks, with parasitoid emergence checked every 2 days.

Table 1 Description of the Kenyan tomato farms investigated

Field identifier	Location	Gps coordinates	Tomato variety	Open field/Net house	Insecticides used	Crop state
1	Kirinyaga	S 00.618231 E37.369609	Perseo	OF + NH	Metarhizium	Fruiting
2	Kirinyaga	S 00.618231 E37.369609	Perseo	OF + NH	Metarhizium	Fruiting
3	Kirinyaga	S 00.611750 E 037.388580	KILELE F1	OF + NH	Abamectine	Fruiting
4	Kirinyaga	S 00.602460 E 037.364280	KILELE F1	OF + NH	Imidaclopride + Beta-cyfluthrine, Chlorant-raniliprole, Abamectine	Started harvesting
5	Kirinyaga	S 00.597070 E 037.380050	ANNA F1	OF + NH	Flubendiamide	Fruiting
6	Kiambu	S 01.233250 E 036.664310	BRAVO F1	OF + NH	Metarhizium	Fruiting
7	Kiambu	S 01.143570 E 036.578370	ANNA F1	OF + NH	Metarhizium	Fruiting
8	Machakos	S 01.318040 E 037.226040	EVA F1	OF + NH	Thiamethoxam, meta-laxyl + mancozeb, Mefenoxam + Mancozeb	2 weeks after transplanting
9	Machakos	S 01.385300 E 037.249250	EVA F1	OF + NH	Thiamethoxam, meta-laxyl + mancozeb, Mefenoxam + Mancozeb	3 weeks after transplanting
10	Kajiado	S 01.432100 E 036.682300	TYLKA F1	NH	Neonicotinoides	Flowering
11	Kajiado	S 01.303160 E 036.777720	BRAVO F1	NH	Pyréthrinoïdes	3 weeks after transplanting
12	Kajiado	S 01.435010 E 036.675950	TYLKA F1,ANNA F1	NH	Metarhizium	Fruiting
13	Kajiado	S 01.434130 E 036.676760	EVA F1	OF + NH	Lambda-cyhalothrin, Imidacloprid 250 + Bifen-thrin 50 SC	2 weeks since transplanted
14	Migori	S 00.763980 E 034.585090	RIOGRANDE	OF + NH	No insecticide	Fruiting
15	Migori	S 00.764100 E 034.584900	RIOGRANDE	OF + NH	No insecticide	Flowering
16	Migori	S 00.861280 E 034.610370	CAL J	NH	No insecticide	Fruiting

All pest and natural enemy samples were sorted out by morphotype and placed in ethanol (70%) and sent to the Centre for Biology and Management of Populations (CBGP, Montpellier, France) for identification or confirmation by barcoding. DNA was extracted nondestructively from full specimens using a DNeasy Blood & Tissue kit (Qiagen, Hilden, Germany), according to the manufacturer's protocol. PCR amplifications were performed using standard primers for barcoding (cytochrome c oxidase subunit 1 mitochondrial gene) of invertebrates: LCO1490: 5'-GGTCAACAAATCATAAAGATATTG G-3' (forward primer sequence) and HCO2198: 5'-TAA ACTTCAGGGTGACCAAAAAATCA-3' (reverse primer sequence) (Folmer et al. 1994). PCR reactions were carried out on a Mastercycler® Nexus (Eppendorf, Hamburg, Germany) in a 10 µL PCR mix containing 5 µl of Multiplex Master Mix (Qiagen, Hilden, Germany), 0.8 µl primers (forward and reverse at 2 µM) and 2 µl of DNA. The PCR conditions were as follows: initial DNA denaturation at 94 °C for 15 min, followed by 10 cycles of 30 s each at 94 °C, 1 min at 60–50 °C (touchdown of -1 °C per cycle) and 30 s at 70 °C, followed by 30 cycles of 30 s at 94 °C, 1 min at 55 °C, and 1 min at 72 °C with a final 20 min extension at 72 °C (Haran et al. 2018). The PCR products were sequenced by Eurofins Genomics (http://www.eurofinsgenomics.eu/). All voucher specimens were deposited in the general collection at CBGP. Barcode sequences were aligned and checked using CodonCode Aligner V.3.7.1. (CodonCode Corporation, Centerville, MA, USA). Each sequence was then subjected to molecular identification using the BLAST tool implemented in the NCBI platform (https://www.ncbi.nlm.nih.gov/).

Laboratory experiments

Laboratory experiments were conducted at 25 ± 2 °C and $65 \pm 5\%$ RH. Six-week-old tomato plants from the Kilele F1 tomato hybrid (Syngenta, Nairobi, Kenya) were used in the experiments.

Individual crossing tests

A device was created using two transparent tubes (10 cm long, 5 cm diameter) to check the capability of the main natural enemies, i.e. *Nesidiocoris tenuis* (Reuter, 1895) (Hemiptera: Miridae) and *Encarsia formosa* Gahan (1924) (Hymenoptera: Aphelinidae), to pass through the nets (Deletre et al. 2015). Three types of nets were tested (A to Z Textile Mills

Ltd, Arusha, Tanzania), two types with different mesh sizes (0.9 mm and 0.4 mm, not treated nets) and one-insecticide treated net (0.9 mm mesh size; net impregnated with deltamethrin during the manufacturing process). One tube contained the insects and was closed at the top with the tested net. A second tube contained a trifoliate tomato leaf and was fixed on the top of the first tube (on the mesh side). The end of this second tube was closed with 160 μm mesh netting to retain all insects. The tube containing the insect was covered with aluminum foil and black paper, and the light was placed on the edge of the device. The presence of the leaflet and the light contrast would motivate the insects to cross the net. The set-up was monitored after 30 min to check whether the individuals were able to go through the net or not.

For *N. tenuis*, 15 replicates were conducted per instar and per male and female adults. Only one insect was placed per tube, thus altogether, 105 individuals were tested. For *E. formosa*, adults were tested (mix of males and females). Ten individuals were placed in a tube and 10 replicates were carried out, so 100 individuals were tested.

Oviposition crossing test

The capacity of *N. tenuis* females to oviposit on a leaflet in direct contact with the net, by crossing the three types of nets with their ovipositor, was tested. A device was thus created using Petri dishes (9 cm diameter). The bottom and cover of the Petri dishes were separated using the tested mesh type. A piece of young tomato stem was stuck into floral foam and deposited on the mesh (on the cover side). Then one week-old female was introduced at the bottom. After 24 h, the stem was removed and placed in a plastic box closed with a 160 μm mesh netting for 15 days and watered daily. After 15 days, the number of juveniles in the boxes was recorded. Twenty replicates were carried out per net and 20 replicates of the control treatment without net were tested.

Statistical analyses

All analyses were conducted using R software (R Core Team 2020). For the field survey, a principal component analysis (PCA) was performed to describe the link between the two pests and the *N. tenuis* abundance (the main natural enemy detected). For the field survey, the *T. absoluta*, *N. tenuis* and whitefly densities were analyzed with the generalized linear model (GLM) using a Poisson distribution with net presence as factor. Parasitism was analyzed with a GLM using a binomial distribution with the same factor. Net crossing test results of *N. tenuis* were analyzed using a GLM with a quasibinomial law with the net type (mesh size and insecticide treatment) and the instar as factors. In addition, the net crossing proportions per instar and per mesh were compared using chi-squared tests. A GLM with a quasibinomial

law was conducted for the *E. formosa* net cross test, with the net type as factor, and was complemented by pairwise Tukey analyses (post-hoc test using the multcomp package). A value of $p < 0 0.05$ was considered statistically significant and a value of p between 0.05 and 0.1 were considered marginally significant.

Results

Field survey and identifications

In overall 27 samplings conducted, whiteflies were found 19 times (70%) and *T. absoluta* were found 16 times (59%). Whiteflies were morphologically identified to be *Trialeurodes vaporariorum* (Bruno Michel, CBGP, CIRAD). This identification was confirmed by barcoding, the three specimens sequenced [CBGP vouchers JHAR01877/83/85] showing 99–100% identity with a specimen identified as *T. vaporariorum* [GB accession: MN312161.1]. No other whitefly species were found during this field survey.

The dominant predator recorded in the aspirator sampling was *Nesidiocoris tenuis* (Hemiptera, Miridae; CBGP voucher JHAR04152, 99% identity with [GB accession: AB587603.1]), which is an efficient zoophytophagous predator that attacks both *T. absoluta* and *T. vaporariorum* (Urbaneja et al. 2009; Hassanpour et al. 2016). It represented 96.5% (335 individuals) of the individuals we caught and suspected were predatory. Nevertheless, individuals of another Miridae species generally considered as a predator, i.e. *Campylomma leucochila* (Reuter 1905) (Hemiptera, Miridae; CBGP voucher JHAR01863, 100% identity with [GB accession: KT201382.2]) were also recorded. This species constitutes 2.59% (9 individuals) of the potential predatory species we identified.

Two parasitoid species were obtained via field sampling and laboratory rearing: *Encarsia formosa* (Hymenoptera, Aphelinidae; CBGP voucher JHAR01894, 99% identity with [GB accession: MG813797.1]) emerged from *T. vaporariorum*, and *Copidosoma* sp. (Hymenoptera, Encyrtidae; CBGP voucher JHAR01876, 92% identity with [GB accession: MF906707.1]) emerged from *T. absoluta*.

In the aspirator sampling, a few individuals of other parasitoid species were trapped in addition to the previous natural enemies mentioned, probably not attacking *T. vaporariorum* or *T. absoluta,* but rather attacking other minor pests of the sampled crops. We still report this information as a testimony of their presence in this area for further studies. The following were morphologically identified by Gerard Delvare (CBGP, CIRAD, France) to at least the genus level: *Diglyphus isaea* (Walker 1838) (Hymenoptera: Eulophidae), *Opius* sp. (Hymenoptera: Braconidae) and *Maruecesa* sp. (Hymenoptera: Eulophidae) that attacks leafminer

Fig. 1 PCA mapping of the fields based on the whitefly and *T. absoluta* infestations and *N. tenuis* abundance. (**A**) represents the contribution of the variables to the axis, and (**B**) is the positioning of the fields on the axes
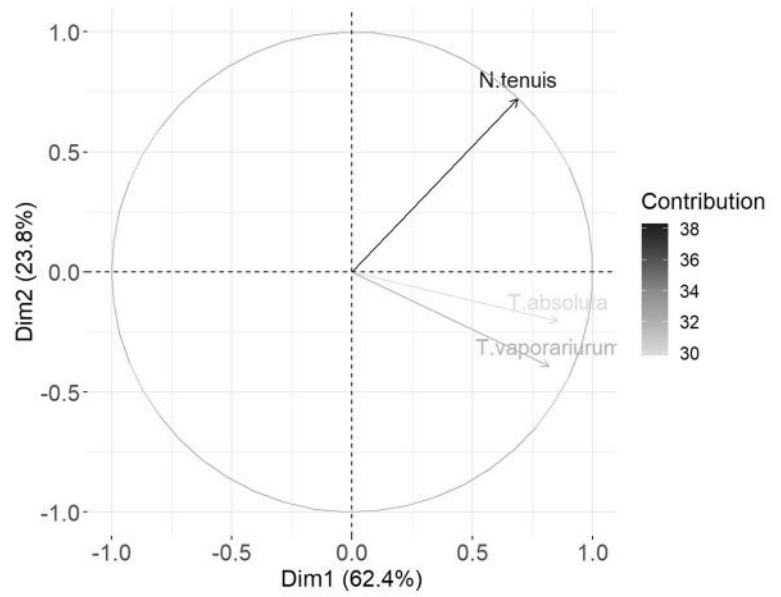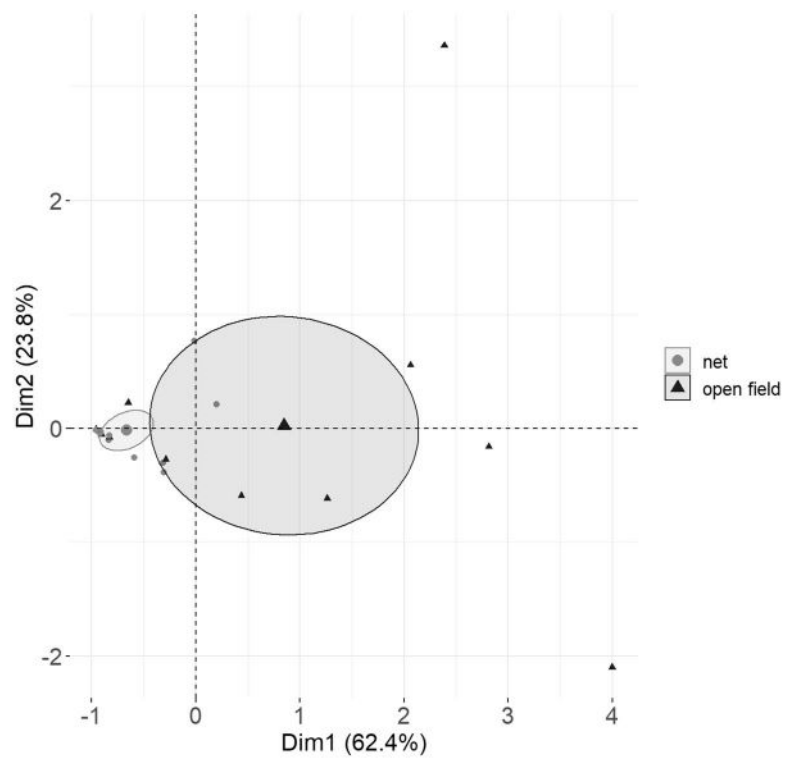
flies, *Aphididus* sp. (Hymenoptera: Braconidae) that attacks aphids, *Oomyzus* sp. (Hymenoptera: Eulophidae) that attacks *Plutella xylostella, Neochrysocharis* sp. (Hymenoptera: Eulophidae) that attacks leafminer flies or microlepidopterans, and *Ceranisus menes* (Walker 1839) (Hymenoptera: Eulophiade) that attacks thrips.

Regarding field infestation and predator presence, PCA mapping of the data collected revealed that *T. absoluta* and whitefly infestation were linked, while *N. tenuis* abundance was not linked to pest infestation (Fig. 1A). We also observed that open field tomato were well segregated from net tomato with respect to the pest infestation and *N. tenuis* abundance (Fig. 1B). PCA shows that: (i) net protected crops hosted fewer pests and predators, (ii) *N. tenuis* was only highly abundant in open fields, and (iii) open fields were more variable in terms of pest and predator abundances (Fig. 1B).

The presence of nets significantly reduced pest infestations ($p < 0.001$ for both pests) but did not completely prevent infestations. Under the nets, 1.02 ± 0.46 (mean ± SE) whiteflies were found per leaflet, while 7.71 ± 4.07 (mean ± SE) were found per leaflet in open fields. Similar differences were observed with *T. absoluta*, 0.12 ± 0.05 (mean ± SE) larvae per leaflet were found under the nets, while 0.69 ± 0.18 (mean ± SE) were found in open fields.

The parasitism ratio was low and not significantly affected by the nets ($p = 0.781$ and $p = 0.360$, for *T. vaporariorum* and *T. absoluta*, respectively). Regarding whiteflies, we found $3.57 \pm 3.57\%$ (mean ± SE) parasitism under the net and $0.06 \pm 0.04\%$ (mean ± SE) in open fields. Regarding *T. absoluta* larvae, we found $2.86 \pm 2.86\%$ (mean ± SE) parasitism under the nets and $3.50 \pm 2.57\%$ (mean ± SE) in open fields. However, cases of parasitism were observed under nets for both pests, highlighting that parasitoids were able to enter the net houses.

Nesidiocoris tenuis was the dominant predator species and was therefore the focus of our statistical analyses. Its population was significantly affected by the net ($p < 0.001$).

Populations were higher in open fields, with 2.57 ± 1.24 (mean ± SE) individuals found per plant, while 0.37 ± 0.26 (mean ± SE) individuals per plant were found under the nets.

Laboratory experiments

Individual crossing tests

The net type, i.e. the mesh size and the treatment of the net with deltamethrin, did not significantly affect the proportion of *N. tenuis* individuals passing through the mesh ($p = 0.338$) (Table 2). However, the ability of *N. tenuis* instars to cross the different net types significantly varied ($p < 0.001$). The first three *N. tenuis* instars were able to cross the untreated 0.9 mm mesh net, while only the first two instars were able to cross the untreated 0.4 mm net and the treated 0.9 mm mesh net (Table 2).

Encarsia formosa adults were able to cross all the three types of nets, but the proportion that passed through the mesh was marginally significantly affected by the net type ($p = 0.096$), with the lowest crossing rate obtained for the untreated 0.4 mm mesh net (Table 2).

Oviposition test

All net types significantly reduced oviposition by *N. tenuis* females ($p < 0.001$), although they were able to oviposit through all nets without any significant differences noted between net types (Fig. 2).

Discussion

We investigated the impact of net houses on tomato crop infestation by two major pests in Kenya, i.e. *T. vaporariorum* and *T. absoluta*, and their natural enemies. Several natural

Table 2 Percentage of *Nesidiocoris tenuis* and *Encarsia formosa* individuals that crossed nets of two different mesh sizes and treated or not with deltamethrin. Letters indicate differences between nets for each insect stage, that were significant ($p < 0.05$) for *N. tenuis* and marginally significant ($p < 0.1$) for *E. formosa*. For *N. tenuis,* crossing percentages were compared using a chi-squared tests ($P < 0.05$), for *E. formosa,* mean ± SE were compared using a Tukey test

Species	Instar	0.4 mm untreated (%)	0.9 mm treated (%)	0.9 mm untreated (%)
N. tenuis	1st nymph instar	80 a	70 a	80 a
	2nd nymph instar	70 a	60 a	60 a
	3rd nymph instar	0 a	0 a	60 b
	4th nymph instar	0	0	0
	5th nymph instar	0	0	0
	Adult females	0	0	0
	Adult males	0	0	0
E. formosa	Adults mixed sex	12 ± 3 a	18 ± 3 ab	23 ± 4 b

enemy species, both predators and parasitoids, were found and identified. In addition, the capacity of the key natural enemies of the two pests to cross the nets was tested.

Nesidiocoris tenuis, the most abundant predator found, is a well-known predatory species that feeds on eggs and larvae of *T. absoluta* (Urbaneja et al. 2009), and on nymphs of *T. vaporariurum* (Hassanpour et al. 2016). It is also known to be present in 17 African countries (CABI 2019a) and to feed on *T. absoluta* in invaded areas (Sylla et al. 2016; Boualem et al. 2012; El Arnaouty and Kortam 2012). In this study, its abundance was linked neither to *T. absoluta* nor whitefly abundance, which might possibly be explained by its zoophytophagous and polyphagous diet. However, this also highlighted the impact of factors other than the prey resource on its abundance, such as insecticide treatments (Arnó and Gabarra 2011), crop diversity (Thomine et al. 2020), climate or agroecological zones (Sridhar et al. 2012), but these factors could not be tested with our sampling design. In our study, most of the pesticides used were not selective, and it is known that organic insecticides can also negatively affect mirids (Biondi et al. 2012).
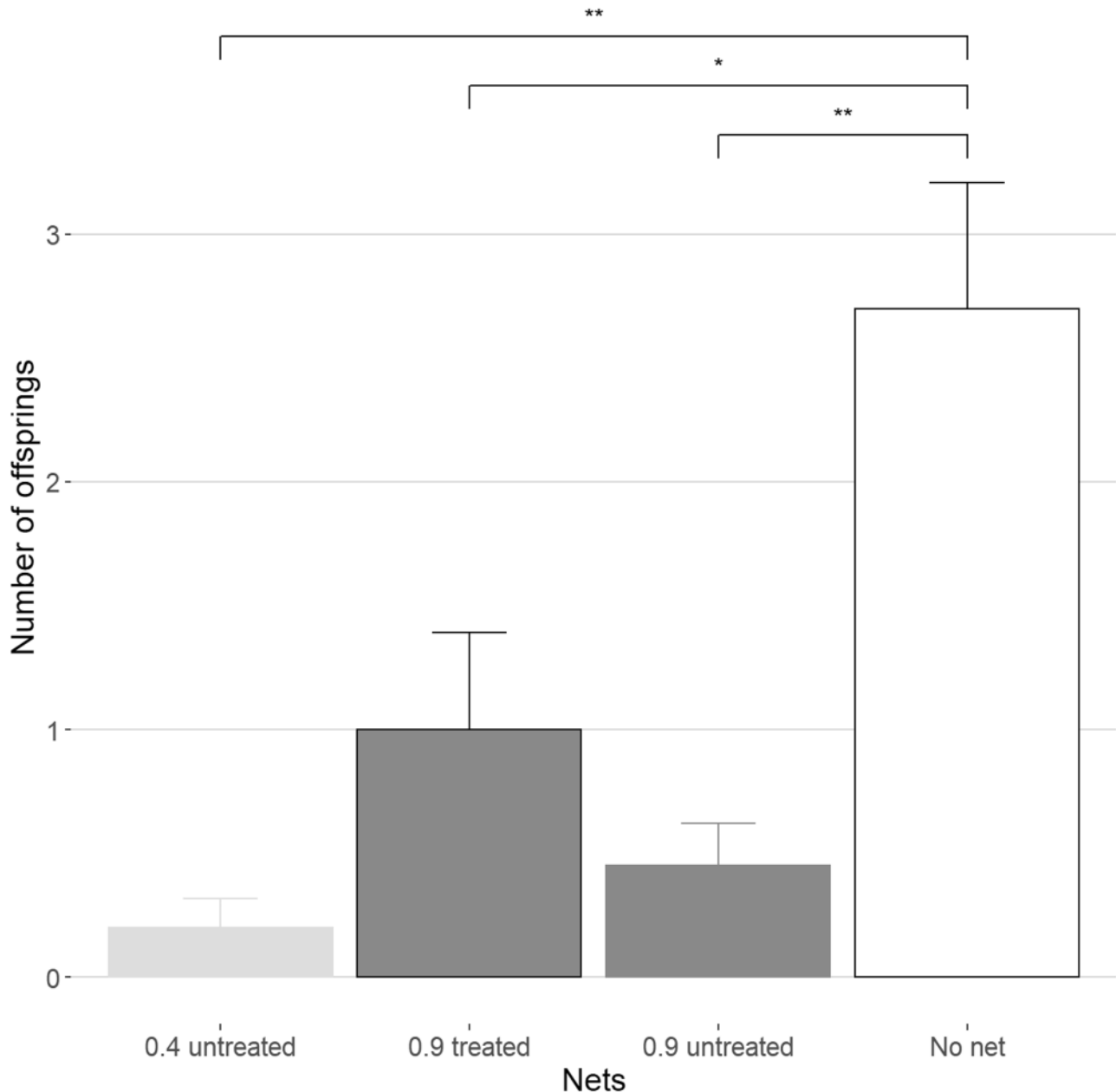

Fig. 2 Ovipostion (mean ± SE) of *N. tenuis* females through nets of two different mesh sizes, treated or not with deltamethrin, and a control (no net). Stars indicate significant differences using a Tukey test (**: P < 0.01 and *: P < 0.5)

Parasitoids were identified for both pests, despite parasitism being low. Indeed, the natural parasitism of both pests is generally found to be low when no practices are set up to improve it (Kinyanjui et al. 2021; Abbes et al. 2014; Nannini et al. 2006). In addition, farmers were allowed to pursue the practices they usually set up, involving insecticides, yet it is known that parasitoids are susceptible to insecticides (Yarahmadi et al. 2018; Abbes et al. 2015) including entomopathogenic fungus (Oreste et al. 2016). We still identified the parasitoid *E. formosa,* that is widely used for biological control of *T. vaporariorum,* firstly used successfully in England in 1926 (Speyer 1927). By 1988, *E. formosa* was estimated to have been used on 3,800 ha of greenhouse area worldwide for biological control of *T. vaporariorum* (van Lenteren and Woets 1988). It has been authorized for biological control in Kenya since 2003 (Songa 2003). This might explain why this was the only parasitoid we found despite other parasitoids are known to develop on *T. vaporariurum* and to be able to coexist with *E. formosa* when sharing a host (Bográn et al. 2002). This can also owe to the sampling size that might not be sufficient to detect rare species, or do not exhaustively represent whitefly instars, thus biasing the results (*E. formosa* preferentially attack third and fourth instars (Nell et al. 1976)). We also identified *Copidososma* sp. that is associated with *T. absoluta* in South America (Desneux et al. 2010). Contrary to most of *T. absoluta* parasitoids (Chailleux et al. 2014), *Copidosoma* sp. is a polyembrionic endoparasitoid belonging to the egg-larval parasitoid guild (Salas Gervassio et al. 2016). This genus is known to be present throughout Africa (CABI 2019b) and the species *Copidosoma koehleri* Blanchard (1940) is known to be present in Kenya (Guerrieri and Noyes 2005).

Our study is the first formal report of the presence of *N. tenuis* and *E. formosa* in Kenyan fields. Both of these natural enemies are commercially produced and sold worldwide. It is unclear if these natural enemies are indigenous to Kenya or if they were introduced for biological control in industrial horticultural crops. This is quite likely with regard to *E. formosa*, which has been produced and sold all over the world for decades, while acclimation and establishment in the environment has not been investigated everywhere. Besides, the fact that both natural enemies are easily mass-reared is crucial toward their use in augmentative biological control programs. Indeed, *E. formosa* is mass-reared on *T. vaporariorum* on tobacco plants (Scopes 1969), and plantless rearing of *N. tenuis* fed on *E. kuhenialla* eggs is being developed (De Puysseleyr et al. 2013).

Our results showed that net houses reduced the pest population but did not entirely prevented neither pest infestation nor the establishment of natural enemies. Predators were less abundant under net houses, while parasitoids did not seem to be affected by the nets. This was probably due to differences in the size of the natural enemies, i.e. parasitoids were smaller than the predators found in this

survey. Moreover, our laboratory experiments confirmed this hypothesis, showing that *E. formosa* was even able to pass through the 0.4 mm mesh net, while only *N. tenuis* nymph, which are wingless and therefore less mobile, were able to cross the nets. Still, the 0.4 mm mesh reduced the percentage of parasitoids crossing the net, probably owing to the visual barrier created by the small mesh, that superimpose to the physical barrier. Indeed, *E. formosa* uses visual stimuli to locate its host (Romeis and Zebitz 1997), thus the visual barrier may have decreased parasitoid motivation to cross the net.

Conclusion and recommendations

Net houses are a first step toward pesticide reduction but are not sufficient, while indigenous natural enemies have the potential to complement the tomato protection under nets. Our results highlighted two main needs warranting further use of natural enemies for tomato protection in Kenya. First, the need to identify the factors other than prey abundance affecting *N. tenuis* populations, so as to boost its abundance in open fields and make them more reliable. Second, there is a need to find ways to increase the abundance of natural enemies, either by (i) using kairomones to attract natural enemies (Ayelo et al. 2021), or (ii) introducing natural enemies under net houses through inoculative releases, as has been done for *N. tenuis* (Calvo et al. 2012), or through inondative releases, as has been done for *E. formosa* (van Lenteren et al. 1996).

Declarations

Conflict of interest statement On behalf of all authors, the corresponding author states that there is no conflict of interest.

References

Abbes K, Biondi A, Zappalà L, Chermiti B (2014) Fortuitous parasitoids of the invasive tomato leafminer *Tuta absoluta* in Tunisia. Phytoparasitica 42(1):85–92

Abbes K, Biondi A, Kurtulus A, Ricupero M, Russo A, Siscaro G, Chermiti B, Zappala L (2015) Combined non-target effects of insecticide and high temperature on the parasitoid *Bracon nigricans*. PLoS One 10(9):e0138411

Achieng'a FC, Kasina M, Mbugi J, Wasilwa L, Kariuki F, Kipyab P, Martin T (2016) Low cost insect proof and micro-climate regulating Agronets are effective in managing infestation of American

Bollworm (*Helicoverpa armigera* Hubner.) on field tomatoes in Nairobi and Kiambu counties, Kenya. In 2nd biennial international conference on enhancing sustainable agricultural production and marketing systems (p 340)

Arnó J, Gabarra R (2011) Side effects of selected insecticides on the *Tuta absoluta* (Lepidoptera: Gelechiidae) predators *Macrolophus pygmaeus* and *Nesidiocoris tenuis* (Hemiptera: Miridae). J Pest Sci 84(4):513–520

Ayelo PM, Pirk CWW, Yusuf AA, Chailleux A, Mohamed SA, Deletre E (2021) Exploring the kairomone-based foraging behaviour of natural enemies to enhance biological control: a review. Front Ecol Evol 9:143

Biondi A, Desneux N, Siscaro G, Zappalà L (2012) Using organic-certified rather than synthetic pesticides may not be safer for biological control agents: selectivity and side effects of 14 pesticides on the predator *Orius laevigatus*. Chemosphere 87(7):803–812

Blanchard EE (1940) Apuntes sobre Encirtidos Argentinos. An Soc Cient Argent 130(3):106–128

Bográn CE, Heinz KM, Ciomperlik MA (2002) Interspecific competition among insect parasitoids: field experiments with whiteflies as hosts in cotton. Ecology 83(3):653–668

Boualem M, Allaoui H, Hamadi R, Medjahed M (2012) Biologie et complexe des ennemis naturels de *Tuta absoluta* à Mostaganem (Algérie). EPPO Bull 42:268–274

CABI (2019a) *Nesidiocoris tenuis* (tomato bug). https://www.cabi.org/isc/datasheet/16251. Accessed 24 Jan 2020

CABI (2019b) *Copidosoma*. https://www.cabi.org/isc/datasheet/15708. Accessed 24 Jan 2020

Calvo FJ, Lorente MJ, Stansly PA, Belda JE (2012) Preplant release of *Nesidiocoris tenuis* and supplementary tactics for control of *Tuta absoluta* and *Bemisa tabaci* in greenhouse tomato. Entomol Exp Appl 143(2):111–119

Chailleux A, Desneux N, Seguret J, Khanh HDT, Maignet P, Tabone E (2012) Assessing European egg parasitoids as a mean of controlling the invasive South American tomato pinworm *Tuta absoluta*. PLoS One 7(10)

Chailleux A, Desneux N, Arnó J, Gabarra R (2014) Biology of two key Palaearctic larval ectoparasitoids when parasitizing the invasive pest *Tuta absoluta*. J Pest Sci 87(3):441–448

De Bon H, Huat J, Parrot L, Sinzogan A, Martin T, Malezieux E, Vayssieres JF (2014) Pesticide risks from fruit and vegetable pest management by small farmers in sub-Saharan Africa. A review. Agron Sustain Dev 34(4):723–736

De Puysseleyr V, De Man S, Höfte M, De Clercq P (2013) Plantless rearing of the zoophytophagous bug *Nesidiocoris tenuis*. Biocontrol 58(2):205–213

Deletre E, Mallent M, Menut C, Chandre F, Martin T (2015) Behavioral response of *Bemisia tabaci* (Hemiptera: Aleyrodidae) to 20 plant extracts. J Econ Entomol 108(4):1890–1901

Desneux N, Wajnberg E, Wyckhuys KA, Burgio G, Arpaia S, Narváez-Vasquez CA, Pizzol J (2010) Biological invasion of European tomato crops by *Tuta absoluta*: ecology, geographic expansion and prospects for biological control. J Pest Sci 83(3):197–215

Diatte M, Brévault T, Sylla S, Tendeng E, Sall-Sy D, Diarra K (2018) Arthropod pest complex and associated damage in field-grown tomato in Senegal. Int J Trop Insect Sci 38(3):243–253

El Arnaouty SA, Kortam MN (2012) First record of the mired predatory species, *Nesidiocoris tenuis* Reuter (Heteroptera: Miridae) on the tomato leafminer, *Tuta absoluta* (Meyrick)(Lepidoptera: Gelechiidae) in Egypt. Egypt J Biol Pest Control 22(2):223

Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Mol Marine Biol Biotechnol 3:294–299

Gahan AB (1924) Some new parasitic Hymenoptera with notes on several described forms. Proc US Nat Mus 2517(65, Art. 4):1–23

Gamarra H, Carhuapoma P, Mujica M, Kreuze J, Kroschel J (2016) Greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood 1956), in Pest distribution and risk atlas for Africa: Potential, global and regional distribution and abundance of agricultural and horticultural pests and associated biocontrol agents under current and future climates, ed. by Kroschel J, Mujica N, Carhuapoma P and Sporleder M. International Potato Center (CIP), Lima, pp 154–168

Gogo EO, Saidi M, Itulya FM, Martin T, Ngouajio M (2014) Eco-friendly nets and floating row covers reduce pest infestation and improve tomato (*Solanum lycopersicum* L.) yields for smallholder farmers in Kenya. Agronomy 4(1):1–12

Guillemaud T, Blin A, Le Goff I, Desneux N, Reyes M, Tabone E, Lombaert E (2015) The tomato borer, *Tuta absoluta*, invading the Mediterranean Basin, originates from a single introduction from Central Chile. Sci Rep 5(1):1–5

Guerrieri E, Noyes J (2005) Revision of the European species of *Copidosoma Ratzeburg* (Hymenoptera: Encyrtidae), parasitoids of caterpillars (Lepidoptera). Syst Entomol 30(1):97–174

Gurr GM, Kvedaras OL (2010) Synergizing biological control: scope for sterile insect technique, induced plant defences and cultural techniques to enhance natural enemy impact. Biol Control 52(3):198–207

Han P, Bayram Y, Shaltiel-Harpaz L, Sohrabi F, Saji A, Esenali UT, Lu ZZ (2019) *Tuta absoluta* continues to disperse in Asia: damage, ongoing management and future challenges. J Pest Sci 92(4):1317–1327

Haran J, Delvare G, Vayssieres JF, Benoit L, Cruaud P, Rasplus JY, Cruaud A (2018) Increasing the utility of barcode databases through high-throughput sequencing of amplicons from dried museum specimens, an example on parasitic hymenoptera (Braconidae). Biol Control 122:93–100

Hassanpour M, Bagheri M, Golizadeh A, Farrokhi S (2016) Functional response of *Nesidiocoris tenuis* (Hemiptera: Miridae) to *Trialeurodes vaporariorum* (Hemiptera: Aleyrodidae): effect of different host plants. Biocontrol Sci Technol 26(11):1489–1503

Jones DR (2003) Plant viruses transmitted by whiteflies. Eur J Plant Pathol 109(3):195–219

Kimathi RH, Wilisiani F, Mashiko T, Neriya Y, Miinda AE, Nishigawa H, Natsuaki T (2020) First report of tomato chlorosis virus infecting tomato in Kenya. Sci Afr 7:e00286

Kinyanjui G, Khamis FM, Ombura FLO, Kenya EU, Ekesi S, Mohamed SA (2021) Distribution, abundance and natural enemies of the invasive tomato leafminer, *Tuta absoluta* (Meyrick) in Kenya. Bull Entomol Res 1–16

Kungu M, Deletre E, Subramanian S, Fiaboe KK, Gitonga L, Lagat ZO, Martin T (2019) A new mite IPM strategy: predator avoidance behaviour resulting from the synergetic effects of predator release and acaricide-treated nets. Pest Manag Sci 75(4):979–985

Majumdar AZ, Chambliss A, Mastin W, Carpenter S (2015) High tunnel pest exclusion system: Laboratory and field experiences. J NACAA 8(1)

Mansour R, Brévault T, Chailleux A, Cherif A, Grissa-Lebdi K, Haddi K, Tonnang HE (2018) Occurrence, biology, natural enemies and management of *Tuta absoluta* in Africa. Entomol Gen 38(2):83–112

Meyrick E (1917) Descriptions of South American micro-lepidoptera. Trans Entomol Soc Lond 65:1–52

Mound LA, SH Halsey (1978) Whiteflies of the World: A systematic catalogue of the Aleyrodidae (Homoptera) with host plant and natural enemy data. British Museum (N.H.) and Wiley, Chichester, UK

Nannini M, Manca L, Giorgini M (2006) Natural parasitism of *Bemisia tabaci* and *Trialeurodes vaporariorum* in an horticultural area of Sardinia, Italy. IOBC WPRS Bull 29(4):59

Nderitu PW, Muturi J, Otieno M, Arunga EE, Mattias J (2018) Tomato leaf miner (*Tuta absoluta*)(Meyrick 1917)(Lepidoptera: Gelechiidae) prevalence and farmer management practices in Kirinyanga County, Kenya. J Entomol Nematol 10(6):43–49

Nell HW, Lelie LSVD, Woets J, Lenteren JV (1976) The parasite-host relationship between *Encarsia formosa* (Hymenoptera: Aphelinidae) and *Trialeurodes vaporariorum* (Homoptera: Aleyrodidae) II. Selection of host stages for oviposition and feeding by the parasite 1. Z Angew Entomol 81(1–4):372–376

Njuguna NT (2000) Assessment of the level of resistance to Bifenthrin, Cypermethrin and Methomyl insecticides in *Bemisia tabaci* (Gennadius) and *Trialeurodes vaporariorum* (Westwood) whitefly populations from selected sites in Kenya (Doctoral dissertation, Kenyatta University)

Nordey T, Basset-Mens C, De Bon H, Martin T, Déletré E, Simon S, Parrot L, Despretz H, Huat J, Biard Y, Dubois T, Malézieux E (2017) Protected cultivation of vegetable crops in sub-Saharan Africa: limits and prospects for smallholders. A review. Agron Sustain Dev 37(6):1–20

Oreste M, Bubici G, Poliseno M, Tarasco E (2016) Effect of *Beauveria bassiana* and *Metarhizium anisopliae* on the *Trialeurodes vaporariorum-Encarsia formosa* system. J Pest Sci 89(1):153–160

Perić P, Dimic N (1997) Natural enemies of *Trialeurodes vaporariorum* Westwood in Yugoslavia. Plant Prot 48(1):67–71

Perić P, Marčić D, Stamenković S (2008) Natural enemies of whitefly (*Trialeurodes vaporariorum* Westwood) in Serbia. In IV Balkan Symposium on Vegetables and Potatoes 830 (pp 539–544)

R Core Team (2020) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/. Accessed 24 Jan 2020

Ramasamy S, Ravishankar M (2018) Integrated pest management strategies for tomato under protected structures. In Sustainable management of arthropod pests of tomato. Academic Press, pp 313–322

Reuter OM (1895) Ad cognitionem Capsidarum. III. Capsidae ex Africa boreali. Revue d'Entomologie, Caen 14:131–142

Reuter OM (1905) Ad cognitionem Capsidarum Aethiopicarum. III. Öfversigt af Finska Vetenskapssocietetens Förhandlingar 47(21):9

Romeis J, Zebitz CPW (1997) Searching behaviour of *Encarsia formosa* as mediated by colour and honeydew. Entomol Exp Appl 82(3):299–309

Rwomushana I, Beale T, Chipabika G, Day R, Gonzalez-Moreno P, Lamontagne-Godwin J, Tambo J (2019) Tomato leafminer (*Tuta absoluta*): Impacts and coping strategies for Africa, 58p. CAB International, Wallingford

Salas Gervassio NG, Luna MG, Lee S, Salvo A, Sánchez NE (2016) Trophic web associated with the South American tomato moth *Tuta absoluta*: implications for its conservation biological control in Argentina. Agric for Entomol 18(2):137–144

Scopes NEA (1969) The economics of mass rearing *Encarsia formosa*, a parasite of the whitefly *Trialeurodes vaporariorum*, for use in commercial horticulture. Plant Pathol 18(3):130–132

Simon S, Komlan FA, Adjaïto L, Mensah A, Coffi HK, Ngouajio M, Martin T (2014) Efficacy of insect nets for cabbage production and pest management depending on the net removal frequency and microclimate. Int J Pest Manag 60(3):208–216

Songa W (2003) Kenyan regulations for importation of biological control agents. Registration for biocontrol agents in Kenya 75

Speyer ER (1927) An important parasite of the greenhouse whitefly. Bull Entomol Res 17:301–308

Sridhar V, Jayashankar M, Vinesh LS (2012) Population dynamics of Zoophytophagous mirid bug, *Nesidiocoris tenuis* (Reuter)(Heteroptera: Miridae) and its prey, *Bemisia tabaci* Genn. (Homoptera: Aleyrodidae) on tomato (*Solanum lycopersicum* Mill). Pest Manag Hortic Ecosyst 18(1):35–38

Sylla S, Brévault T, Streito JC, Diarra K (2016) First record of *Nesidiocoris tenuis* (Reuter)(Heteroptera: Miridae), as a predator of the tomato leaf miner, *Tuta absoluta* (Meyrick)(Lepidoptera: Gelechiidae). Senegal Egypt J Biol Pest Control 26(4):851–853

Thomine E, Jeavons E, Rusch A, Bearez P, Desneux N (2020) Effect of crop diversity on predation activity and population dynamics of the mirid predator *Nesidiocoris tenuis*. J Pest Sci 93:1255–1265

Urbaneja A, Montón H, Molla O (2009) Suitability of the tomato borer *Tuta absoluta* as prey for *Macrolophus pygmaeus* and *Nesidiocoris tenuis*. J Appl Entomol 133(4):292–296

van Lenteren JC, van Roermund HJ, Sütterlin S (1996) Biological control of greenhouse whitefly (*Trialeurodes vaporariorum*) with the parasitoid *Encarsia formosa*: How does it work? Biol Control 6(1):1–10

van Lenteren JC, Woets J (1988) Biological and integrated pest control in greenhouses. Annu Rev Entomol 33:239–269

Walker F (1838) L.—Descriptions of British Chalcidites. J Nat Hist 1(6):449–454

Walker F (1839) Monographia chalciditum. H. Baillière 1:333

Westwood JO (1856) A. vaporariorum. Gardener's Chron 852

Yarahmadi F, Salehi Z, Lotfalizadeh H (2018) Compatibility of insecticides and *Elachertus inunctus* Nees (Hymenoptera: Eulophidae) for controlling *Tuta absoluta* Meyrick (Lepidoptera: Gelechiidae) in greenhouse condition. J Plant Dis Prot 125(4):357–364

Zappala L, Biondi A, Alma A, Al-Jboory IJ, Arno J, Bayram A, Desneux N (2013) Natural enemies of the South American moth, *Tuta absoluta*, in Europe, North Africa and Middle East, and their potential use in pest control strategies. J Pest Sci 86(4):635–647