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Effect of exclusion net timing of deployment and color on *Halyomorpha halys* (Hemiptera: Pentatomidae) infestation in pear and apple orchards

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ABSTRACT

The brown marmorated stink bug, *Halyomorpha halys* (Stål, 1855) (Hemiptera: Pentatomidae), is an invasive pest species native to East Asia that has become the major pest for many crops, causing serious economic damage. The occurrence of this invasive pest leads to an increase in broad-spectrum insecticides applications, often with limited results in *H. halys* management. Exclusion netting based on insect-proof nets is considered an environmentally friendly tactic that may potentially reduce *H. halys* infestation and damage. In a first experiment, during two growing seasons we investigated whether the timing of exclusion netting deployment affected season-long *H. halys* infestation and impact on apple and pear production. A second experiment considered the effect of netting color. Our results confirm that insect-proof nettings are effective in controlling *H. halys* infestations in fruit orchards, also observing an effect of timing of netting deployment on *H. halys* adult infestation, especially in apple orchards. Netting deployment could be delayed until the growing fruit phase on pear orchards, while on apple orchards it should be performed at flower fading phase. Brown marmorated stink bug infestation and damage were also influenced by the netting color, with black nets having higher infestation levels and fruit damage than white ones. Within an integrated pest management framework, early net deployment and the use of clear color nettings should be considered to reduce *H. halys* infestation and its damage in pome fruit crops.

1. Introduction

The brown marmorated stink bug, *Halyomorpha halys* (Stål, 1855) (Hemiptera: Pentatomidae), is an invasive polyphagous species native to East Asia (Lee et al., 2013; Leskey and Nielsen, 2018), which spread to North America (Canada and USA; Hoebeke and Carter, 2003), Chile, and several European countries (Bariselli et al., 2016; Leskey and Nielsen, 2018; Maistrello et al., 2014; Wermelinger et al., 2008). In Italy, the first detection of *H. halys* in a field occurred in Emilia Romagna region in 2012 (Cesari et al., 2015), while in Veneto and Friuli-Venezia Giulia regions it was found for the first time in 2014 (Benvenuto et al., 2015; Maistrello and Dioli, 2014). Nowadays, *H. halys* is present all over the country (Maistrello et al., 2018). In geographical areas where this species is invasive, *H. halys* has become the major pest for many crops, causing substantial economic damage. In the agroecosystem, habitat composition is a key element that drives brown marmorated stink bug population dynamics, influencing crop colonization and, consequently, damage (Tamburini et al., 2023). For instance, in the eastern USA, brown marmorated stink bug infestations caused up to 100% crop loss in apple and peach orchards in 2010 (Leskey et al., 2012). In the north of Italy, *H. halys* caused more than 740 million euros of losses in 2019, 160 million in the Veneto region alone (Italiafruit News, 2020). While being able to feed on plant tissue such as blooms, leaves, trunks, or twigs (Martinson et al., 2013; Scaccini and Pozzebon, 2021), *H. halys* needs to feed on fruit structures to complete its development (Leskey and Nielsen, 2018). This physiology-driven behavior, together with the great dispersal ability and the high reproductive rate, makes this pest of primary importance on fruit crops such as pear and apple (Bariselli et al., 2016; Bergmann et al., 2016). While feeding, *H. halys* injects its enzyme-rich saliva to predigest the plant materials, leading to tissue damage that causes discoloration, depression, suberification of fruits parts, and even their premature drop (Leskey and Nielsen, 2018; Peiffer and Felton, 2014). On the fruit structures of pear and apple, the typical injuries are dimpling or discolored areas with or without a distinct

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depression, suberification in the fruit flesh, fruit abortion, and deformation when early-season punctures occur (Acebes-Doria et al., 2016; Maistrello et al., 2016; Nielsen and Hamilton, 2009). The severity of injuries is influenced by the timing of *H. halys* infestation and plant phenology (e.g., Acebes-Doria et al., 2016; Hedstrom et al., 2014; Joseph et al., 2015; Leskey et al., 2012; Stahl et al., 2020). Late instar nymphs and adults can cause high level of injuries on apples when the infestation occurs late in the season compared to early instar nymph infestations in the first part of the growing season (Acebes Doria et al., 2016). The feeding activity of *H. halys* was associated with increased damage by fruit pathogens such as *Eremothecium coryli* Kurtzman, 1995 (Saccharomycetales: Saccharomycetaceae) yeast (Rice et al., 2014), or *Monilinia* spp. (Helotiales: Sclerotinaceae) fungi (Moore et al., 2019), agents of fruit rot that can decrease yield and fruit quality.

The occurrence of this invasive pest leads to an increase in broad-spectrum insecticides applications, but this strategy is considered not fully effective for *H. halys* control, mainly because of the limited residual effects on this pest and thus requiring other tactics (e.g., Kuhar and Kamminga, 2017; Leskey et al., 2014; Finetti et al., 2021a, 2021b; Finetti et al., 2023). Exclusion strategy based on insect-proof nets is considered a more environmentally friendly tactic than chemical control (Briassoulis et al., 2007; Castellano et al., 2008; Chouinard et al., 2016). Exclusion netting consists of physical barriers that limit the movement of target insects when colonizing the crop. Their effectiveness in excluding pests is well-known for aphids (Dib et al., 2010) and *Drosophila suzukii* (Matsumura) (Diptera: Drosophilidae) in cherry, raspberry, and blueberry crops (Charlot and Weydert, 2017; Cormier et al., 2015). Besides, exclusion netting used for *Cydia pomonella* (L.) (Lepidoptera: Tortricidae) management in apple orchards can block or limit the flight of codling moth and their coupling (Angeli and Rizzi, 2013; Pasqualini et al., 2013; Tasin et al., 2008). This tool can be a potential solution to reduce *H. halys* infestation on plants under netted plots and thus its damage, as reported in North America (Marshall and Beers, 2016) and Italy (Candian et al., 2018, 2021). Indeed, in nectarine orchards in Italy the damage was reduced by up to 78% for netted plants compared to those with insecticidal treatments alone (Candian et al., 2021).

Exclusion netting can affect microclimatic conditions in the orchards depending on the effect of mesh size, photoselective properties and color (i.e., white, grey, black) on temperature, relative humidity and spectral composition of the light transmitted and reflected. These can be desirable effects when fruits' quality and yield are increased (Basile et al., 2012; Ben-Yakir et al., 2012; Retamales et al., 2008; Shahak et al., 2004). However, alteration of microclimatic features (e.g., temperature and relative humidity) can influence *H. halys* biological parameters such as nymph development and adult longevity and fecundity (e.g., Baek et al., 2017; Cira et al., 2018; Fisher et al., 2021; Haye et al., 2014; Khadka et al., 2020; Nielsen et al., 2008; Scaccini et al., 2019, 2020; Stahl et al., 2021). Light conditions affect *H. halys* behavior, as they tend to prefer dark areas (Cullum et al., 2020; Toyama et al., 2011).

Timing of exclusion netting deployment could represent a critical aspect of their use. On the one hand, early deployment was associated with a negative effect on plants, with a reduction in fruit number and their quality (Elsysy et al., 2019; Kelder et al., 2014). On the other hand, early protection by exclusion netting should ensure high protection against *H. halys* infestation, thus limiting fruit damage. However, no data are available on this aspect; therefore, no robust criteria can be established for the decision on netting deployment timing. For this reason, we investigated whether the timing of exclusion netting deployment affected season-long *H. halys* infestation and its impact on apple and pear production. The effect of netting color was also considered in a second experiment. In these studies, we evaluated seasonal population dynamics of *H. halys* over two growing seasons and its damage on pear and apple fruits at harvest.

2. Materials and methods

The studies were carried out during 2020 and 2021 in organic apple and pear orchards located in Veneto region (north-eastern Italy). The study on the timing of netting deployment was conducted in two pear orchards (orchard A both in 2020 and 2021; orchard B only in 2021) and in two apple orchards (orchard C both in 2020 and 2021; orchard D only in 2021). The study on the effect of netting color was conducted in two apple orchards (orchard E both in 2020 and 2021; orchard F only in 2021).

2.1. Experimental design

2.1.1. Field experiment 1. timing of deployment of insect-proof netting

The pear orchards used in the first study were located in Castelbaldo (pear orchard A, 45°7′7.94″N, 11°28′0.64″E), and the second (B) in Masi (45°7′32.07″N, 11°30′20.90″E). The size of orchard A was 0.8 ha, planted in 2015 with pear cultivars Conference and White William, grafted on Farold40 (spacing: 1.5 m × 3.8 m; 1754 trees per hectare) and trained with a Bibaum architecture with three axes. Orchard B consisted of a 0.7 ha pear orchard of cultivars Conference and Abate Fétel, on Farold40 (spacing: 1.7 m × 3.9 m; 1508 trees per hectare) planted in 2005 and trained with a Bibaum architecture. In both orchards, the rows were oriented in a north-south direction.

The study on the insect-proof netting deployment time was performed also in an apple orchard (C) located in Castelbaldo (45°7′7.94″N, 11°28′0.64″E) and another (D) located in Bosco di Zevio (45°20′19.52″N, 11°9′21.70″E). Orchard C consisted of a 0.5 ha apple orchard of the cultivar Golden Delicious grafted on M9 (spacing: 1.2 m × 3.8 m; 2193 trees per hectare), planted in 2016 and trained with a Spindel architecture. Orchard D consisted of a 0.65 ha apple orchard of Golden Delicious on M9 (spacing: 1.0 m × 3.5 m; 2857 trees per hectare), planted in 2007 and trained with a Spindel architecture. In both orchards, the rows were oriented in a west-east direction.

A single row exclusion netting system was present in all the pear and apple orchards, using the commercially available white Alt'Carpò nets, with a 2.8 × 7.0 mm mesh size. The netting system was closed below the tree canopy, about 50–60 cm from the ground level. Orchard ground-cover vegetation consisted of perennial grasses that were mechanically mulched. Three treatments were compared: (1) un-netted control (CTRL) without netting system, (2) netted plots deployed at flowers fading (BBCH 67, FF), and (3) netted plots deployed at fruit size up to 20 mm phase (BBCH 72, GF). In 2020 a completely randomized design with six replicates per treatment was used, while a randomized block design with nine replicates per treatment (six in orchards A and C, and three in orchards B and D) with two blocks (orchards) was used in 2021. Replicates were constituted by 20 trees per each treatment and cultivar in 2020 and 2021.

In pear orchards (A and B), netting deployment occurred on 15 April in 2020 and on 22 April in 2021 for FF plots, while on 27 April in 2020 and on 12 May in 2021 in the case of GF. In apple orchards (C and D), nets were deployed on 24 April in 2020 and on 4 May in 2021 in the case of FF, while for GF it was on 8 May in 2020 and on 19 May in 2021.

The same fungicides (e.g., calcium polysulfide, sulfur, and copper) were applied during both growing season in all orchards to control the most important pear and apple pathogens. Natural pyrethrins were applied in pear orchards on 21 May and July 17, 2020 and in apple orchards on May 12, 2020. During March and April 2021, spring frosts caused the total loss of pear production and more than 50% losses on apple fruits, and no insecticides were applied in the fruit orchards.

2.1.2. Field experiment 2. color of insect-proof nettings

The apple orchards used for this experiment were located in Bevilacqua (45°12′9.88″N, 11°24′2.89″E) and in Bosco di Zevio (45°20′19.52″N, 11°9′21.70″E), and they were under organic management. Orchard E consisted of a 1.56 ha apple orchard (Golden Orange on

M9; spacing: 1.3 m × 4.0 m; 1923 trees per hectare) planted in 2014 and trained with a Spindel architecture. Orchard F consisted of a 0.77 ha apple orchard (Golden Delicious on M9; spacing: 1.0 m × 3.5 m; 2857 trees per hectare) planted in 2007 and trained with a Spindel architecture. In both orchards, the rows were oriented in a north-south direction.

The apple orchards were covered by Alt'Carpò netting with a single row complete exclusion system, mesh size 2.5 × 2.5 mm closed above the tree canopy at about 50–60 cm from the ground level. In this study, two treatments were compared, i.e., white net (WN) vs. black net (BN). In both orchards (E and F), three replicates per treatment (20 trees for each) were arranged in a randomized block design.

Netting systems (independently of color) were deployed on May 18, 2020, and on 10 May (orchard F) and 19 May (E) 2021. The same fungicides (i.e., sulfur and copper) were applied against pathogens during both seasons in the apple orchards, while insecticides were not applied. In both years, data-loggers (Hobo U23 Pro v2 Temperature/Relative Humidity Data Logger, Onset Computing, Bourne, MA, USA) were installed to measure the minimum, maximum and mean temperature (°C) and relative humidity (%). For each plot and treatment, three data-loggers were randomly placed in the orchard canopy 2 m above the ground level, in plots with white and black nets.

2.2. Insect sampling

Halyomorpha halys abundance was quantified in pear and apple orchards. In the field experiment 1, pear orchards A and B were sampled every 7–10 days from mid-April to mid-August 2020, and every 10–15 days from mid-April to the end of July 2021. In apple orchards C and D, all samplings were performed every 7–10 days from early May to mid-August 2020, and every 10–15 days from May to the end of August 2021. In the field experiment 2, in orchards E and F samplings were performed every 10–15 days from mid-May to the end of August in both 2020 and 2021. In all experiments, samplings started before net deployment. The abundance of *H. halys* in each orchard was estimated by beat sampling, performed by striking pear or apple branches 3 times – a branch at a time totalizing three branches in all – and collecting all the insects falling on a 1 × 1 m white beating sheet placed beneath the tree canopy. Stink bugs that fell on the exclusion net while beating were also counted. *Halyomorpha halys* nymphs (first to the fifth instar) and adults were identified and counted. Beating samplings were performed on four consecutive plants for each replicate (24 plants per treatment in 2020 and 36 in 2021 for the experiment 1, while 24 plants per treatment for the experiment 2).

2.3. Fruit damage caused by Halyomorpha halys

The damage caused by *H. halys* feeding on pears and apples was assessed at harvest time by fruit observation. Fruits were classified into four categories, i.e., undamaged fruit (D0), fruit with damage 1 (D1), fruit with damage 2 (D2), and fruit with damage 3 (D3; Supplementary Fig. S1). In detail, undamaged fruits presented no damage caused by brown marmorated stink bug. D1 corresponded to fruits with up to 10% of their surface damaged, thus representing a low *H. halys* feeding activity that makes the fruit still acceptable on the fresh market. Fruits classified as D2 presented up to 40% of their surface with stink bug damage, making these fruits not marketable. The D3 category was characterized by fruits up to 90% or with 100% of the surface damaged and deformed fruits and with small size.

The percentage of *H. halys* damaged fruits was estimated at fruit harvest for each treatment. In the insect-proof netting deployment timing study, in pear orchard A, 50 fruits per replicate were analyzed on July 30, 2020 for White William and August 11, 2020 for Conference. Fruit damage was not estimated in both A and B pear orchards surveyed in 2021 due to the spring frost event, which caused the complete loss of fruiting structures on this crop. In apple orchard C, the damage assessment was conducted on 20 apples per replicate on August 18, 2020 and 50 fruits per replicate on September 3, 2021. In apple orchard D, 100 fruits per replicate were evaluated for damage assessment on September 3, 2021 (900 apples per treatment). In the study on netting color, 50 fruits per replicate were evaluated for damage assessment in orchard E on August 31, 2020, on 100 fruits per replicate on September 18, 2021 in the same orchard, and on September 3, 2021 in orchard F.

2.4. Data analysis

In the field experiment 1 and for each year, *H. halys* abundance was analyzed with a linear mixed model with a normal error distribution and an identity link function, using a Restricted Maximum Likelihood (REML) repeated measures model with the MIXED procedure of SAS, ver. 9.4 (SAS Institute, 2016). The data of the two crops, pear and apple, were analyzed separately. The number of adults, nymphs, and overall population (nymphs + adults) found in pear and apple orchards were considered dependent variables in three separate models. In these models, treatment (i.e., un-netting, netted plots deployed at flower fading, and netted plots deployed at growing fruits) and time of sampling were considered as categorical independent variables. The effect of all independent variables and their interactions were tested with an F test ($\alpha = 0.05$). Differences among treatments were evaluated with the Tukey's test ($\alpha = 0.05$) to the least-square means. Orchards (only for the 2021 data) and cultivars were considered as random effect terms in the models (Littell et al., 2006). Degrees of freedom (df) were estimated using the Kenward and Roger method. Data on nymphs, adults and the overall population were checked for normality and homoscedasticity and then transformed in log (x + 1), and untransformed data are shown in figures. The SLICE option of the LSMEANS statement was used to test treatment effect variation during observation periods. Model assumptions were evaluated by inspecting diagnostic plots of model residuals.

A similar statistical approach was used for the experiment 2 and a linear mixed model with a normal error distribution and an identity link function, using a REML repeated measures model with the MIXED procedure of SAS, ver. 9.4 (SAS Institute, 2016). The number of adults, nymphs and the overall population (nymphs + adults) found in apple orchards were considered dependent variables in three separate models. In these models, treatment (black or white net) and time of sampling were considered as independent variables, and their effect plus interactions were tested with an F test ($\alpha = 0.05$). Differences among treatments were evaluated with the Tukey's test ($\alpha = 0.05$) to the least-square means. Orchards (only for the 2021 field data collection) were considered as a random effect term in the models (Littell et al., 2006). Data on nymphs, adults and the overall population were checked for model assumption with diagnostic plots of model residuals and then transformed in log (x + 1), and untransformed data are shown in figures. The SLICE option of the LSMEANS statement was used to test treatment effect variation during observation periods.

For every trial, the data prior to starting the experiment was analyzed with a linear mixed model using the MIXED procedure of SAS, ver. 9.4 (SAS Institute, 2016). The treatment was considered as categorical independent variable and the effect was tested with an F test ($\alpha = 0.05$). Differences among treatments were evaluated with with Tukey's test ($\alpha = 0.05$) to the least-square means. Plots and cultivars were considered as a random effect. Degrees of freedom were projected using the Kenward and Roger method. Data were transformed in log (x + 1), and untransformed data are shown in figures. The SLICE option of the LSMEANS statement was used to test treatment effect variation during observation periods.

In both studies, analyses of data on fruit damage were run on the percentage of fruit belonging to each damage category (e.g., D0 to D3) using linear mixed models with the MIXED procedure of SAS, ver. 9.4 (SAS Institute, 2016). For the field experiment 1 only, data from the two crops were analyzed separately. Rates of the four categories were considered as dependent variables in separate models. In these models, treatment (for the experiment 1: CTRL vs FF vs GF; for the experiment 2:

black vs white net) was considered as independent variable, and its effect was tested with an F test (α = 0.05). Differences among treatments were evaluated with the Tukey's test (α = 0.05) to the least-square means. Orchards (only for 2021) and cultivars (only for the experiment 1) were considered as a random effect term in the models (Littell et al., 2006). The Kenward and Roger method was used for degrees of freedom estimation. Data were transformed in arcsine $\sqrt{x}$ prior to the analysis, and untransformed data are shown in figures.

3. Results

3.1. Timing of deployment of insect-proof nets

3.1.1. Pear orchards

Halyomorpha halys individuals were found in all experimental orchards, where stink bugs fluctuated in numbers over the growing season (Fig. 1). Adults were found for the first time on April 23, 2020 and on May 4, 2021, and then observed in all sampling events (Supplementary Fig. S2). Nymphs were observed from June 26, 2020 and from June 17, 2021, until the end of the experiments in both years (Supplementary Fig. S2).

In both years, timing of netting deployment influenced pest infestation in pear orchards. The number of *H. halys* adults was influenced by the treatment (2020: $F_{2, 536} = 118.47$, $P < 0.001$; 2021: $F_{2, 756} = 5.59$, $P = 0.004$), the time (2020: $F_{16, 536} = 5.88$, $P < 0.001$; 2021: $F_{10, 756} = 38.86$, $P < 0.001$) and the interaction "time × treatment" (2020: $F_{32, 536} = 2.91$, $P < 0.001$; 2021: $F_{20, 756} = 1.81$, $P = 0.016$). In 2020, netted plots had a lower *H. halys* infestation with netting deployment at FF ($P < 0.001$) and at GF ($P < 0.001$) if compared to CTRL. On the sampling dates prior to netting deployment at growing fruits, adult abundance did not differ among experimental plots ($P = 0.136$), while after netting deployment (i.e., 27 April) and during the season, the pest density was lower in netting treatments as compared to CTRL (Fig. 1). In 2021, *H. halys* adult population was lower on FF treatment if compared to GF ($P = 0.011$) and CTRL ($P = 0.002$). Similarly, prior to netting deployment at at growing fruits the adult abundance did not differ among un-netted plots ($P = 0.841$), while following netting deployment on 12 May, adult infestation was lower only on FF treatment throughout the growing season (Fig. 1). In both years, the abundance of *H. halys* nymphs was influenced by treatment (2020: mean number ± std. err., CTRL = 0.54 ± 0.12, FF = 0.13 ± 0.03, GF = 0.06 ± 0.02; $F_{2, 537} = 14.34$, $P < 0.001$; 2021: CTRL = 0.31 ± 0.11, FF = 0.10 ± 0.06, GF = 0.16 ± 0.07; $F_{2, 756} = 8.86$, $P < 0.001$), with a lower infestation in netted plots than in CTRL ($P < 0.001$ in both years). Netting presence showed a lower infestation of *H. halys* nymphs after netting deployment, with a lower pest density in FF and GF than in CTRL (interaction "time × treatment", 2020: $F_{32, 537} = 1.73$, $P = 0.008$; 2021: $F_{20, 756} = 1.86$, $P = 0.013$). On *H. halys* population (i.e., nymphs and adults taken together), differences were attributed to the treatment (2020: $F_{2, 537} = 103.75$, $P < 0.001$; 2021: $F_{2, 756} = 9.71$, $P < 0.001$), the time (2020: $F_{16, 536} = 6.82$, $P < 0.001$; 2021: $F_{10, 756} = 50.55$, $P < 0.001$) and the interaction "time × treatment" (2020: $F_{32, 537} = 2.75$, $P < 0.001$; 2021: $F_{20, 756} = 1.78$, $P = 0.019$). In 2020, *H. halys* abundance was significantly lower in FF and GF if compared to CTRL ($P < 0.001$). In 2021, pest density was lower in FF than GF ($P = 0.003$) and CRTL ($P < 0.001$).

At fruit harvest, the percentage of undamaged pears (D0) was affected by the treatment ($F_{2, 32} = 99.94$, $P < 0.001$), being higher in FF and GF than in CTRL plots (Fig. 2). No differences among treatments were observed when considering fruits classified as D1, pears with up to 10% of damage ($F_{2, 32} = 0.85$, $P = 0.437$; Fig. 1). Differently, the percentage of pears damaged by *H. halys* classified as D2 (i.e., up to 40% of fruit damage) was influenced by the treatment ($F_{2, 32} = 83.52$, $P < 0.001$). It was lower on FF and GF if compared to CTRL (Fig. 2). Fruits classified as D3, with damage that affected 100% of fruit, were similarly influenced by the treatment ($F_{2, 32} = 13.26$, $P < 0.001$), being less on FF and GF than on CTRL (Fig. 2). In 2021, it was not possible to estimate the damage on pears in both orchards due to the spring frost events that caused the total loss of production.

3.1.2. Apple orchards

In 2020, adults were observed from 1 May to mid-July, while few nymphs were found only in August (Supplementary Figs. S2 and S3). In 2021, adults appeared from 4 May and were continuously observed throughout the season, while nymphs were observed in July and August (Supplementary Fig. S2). *Halyomorpha halys* infestations were affected by the treatment (2020: $F_{2, 96} = 48.65$, $P < 0.001$; 2021: $F_{2, 179} = 42.56$, $P < 0.001$), the time (2020: $F_{15, 96} = 12.48$, $P < 0.001$; 2021: $F_{11, 179} = 6.61$, $P < 0.001$) and the interaction between "time × treatment" (2020: $F_{30, 96} = 4.76$, $P < 0.001$; 2021: $F_{22, 179} = 2.17$, $P = 0.003$). In the samplings prior to netting deployment, adult abundance did not differ among treatments (2020: $P = 0.999$; 2021: $P = 0.998$). A lower abundance of *H. halys* adult was observed in FF and in GF if compared to the CTRL ($P < 0.001$; Fig. 3) and, between the plots with netting, it was lower in FF than GF (2020: $P = 0.015$; 2021: $P = 0.006$; Fig. 3). Differently from adults, nymph infestation was not influenced by the treatment (2020: $F_{2, 96} = 1.54$, $P = 0.219$; 2021: $F_{2, 179} = 3.09$, $P = 0.051$) nor the interaction "time × treatment" (2020: $F_{30, 96} = 0.96$, $P = 0.528$; 2021: $F_{22, 179} = 1.01$, $P = 0.451$) in both years. The time was significant in 2021 ($F_{11, 179} = 4.85$, $P < 0.001$) and not in 2020 ($F_{15, 96} = 0.96$, $P = 0.498$).

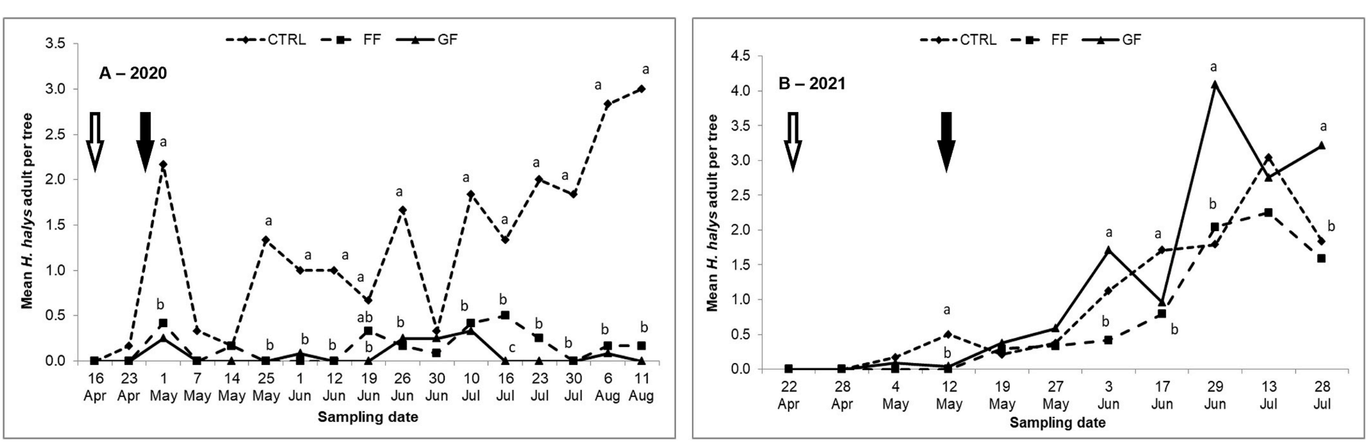

Fig. 1. Number of *Halyomorpha halys* adults observed in pear orchards during 2020 (A) and 2021 (B) in the field experiment 1. CTRL = un-netted control; FF = netting deployed at flower fading; GF = netting deployed at fruit size up to 20 mm phase. The white arrow refers to the period of netting deployment in the FF plots, the black arrow to that in the GF plots. Different letters indicate differences at Tukey's test (α = 0.05).

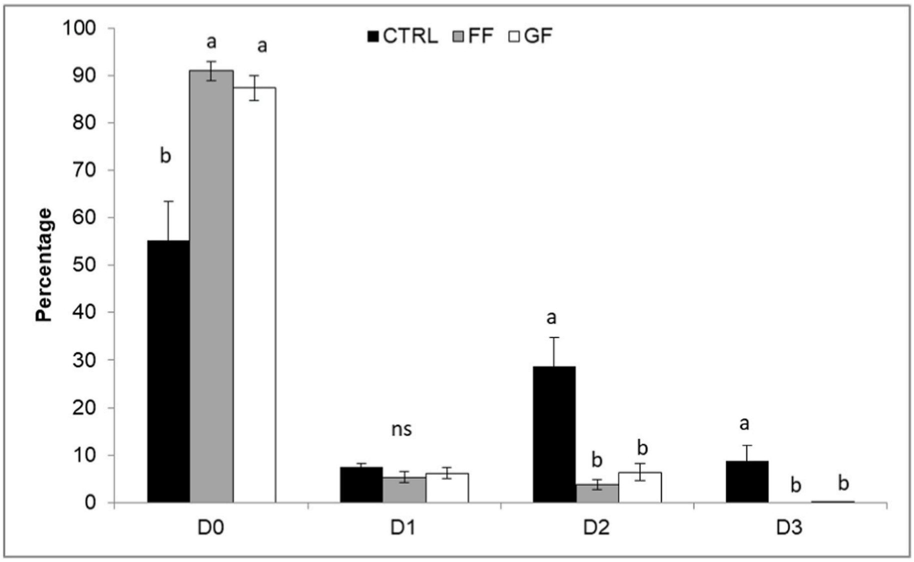

Fig. 2. Percentage of undamaged and damaged fruits (3 classes) caused by *H. halys* estimated at pear harvest in 2020 for the field experiment 1. CTRL = un-netted control; FF = netting deployed at flower fading; GF = netting deployed at fruit size up to 20 mm phase. Four classes: D0 = undamaged fruit; D1 = damage 1 (<10%); D2 = damage 2 (>40%); D3 = damage 3 (>90%). Error bars represent standard error of the mean. Different letters indicate differences at Tukey's test (α = 0.05), while "ns" stands for "not significant".

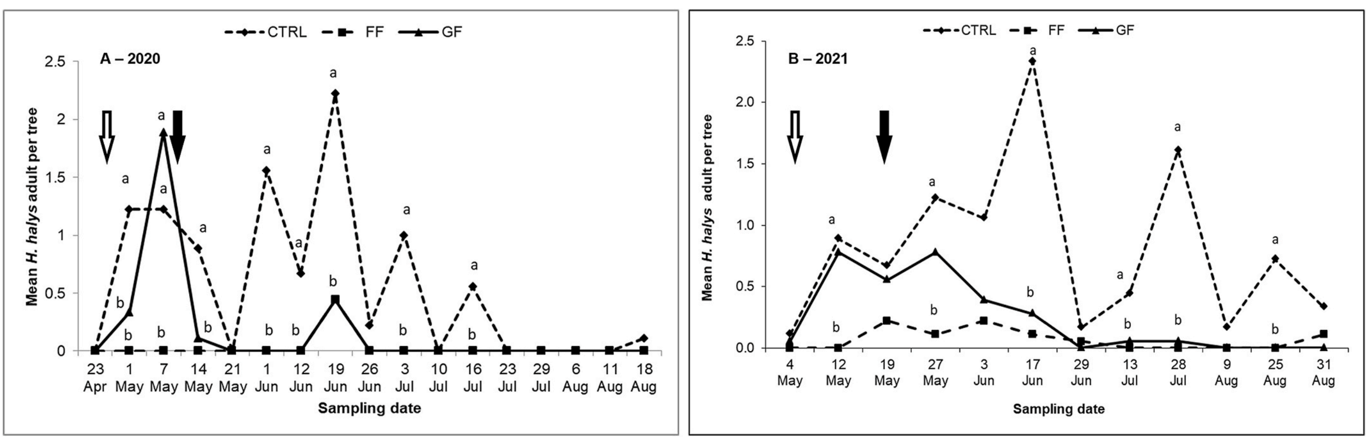

Fig. 3. Number of *Halyomorpha halys* adults observed in apple orchards during 2020 (A) and 2021 (B) in the field experiment 1. CTRL = un-netted control; FF = netting deployed at flower fading; GF = netting deployed at fruit size up to 20 mm phase. The white arrow refers to the period of netting deployment in the FF plots, the black arrow to that in the GF plots. Different letters indicate differences at Tukey's test (α = 0.05).

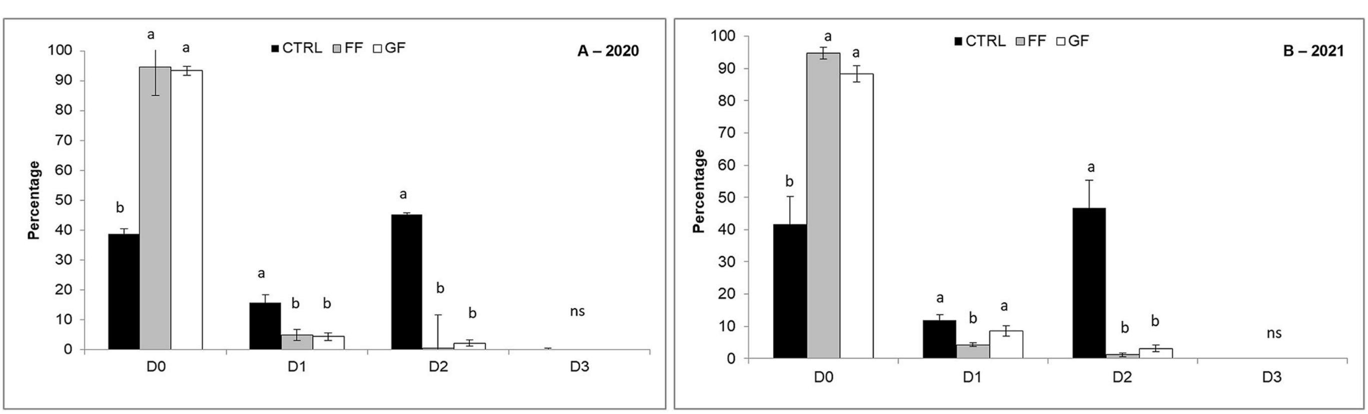

Fig. 4. Percentage of undamaged and damaged fruits (3 classes) caused by *H. halys* estimated at apple harvest in 2020 (A) and 2021 (B) in the field experiment 1. CTRL = un-netted control; FF = netting deployed at flower fading; GF = netting deployed at fruit size up to 20 mm phase. Four classes: D0 = undamaged fruit; D1 = damage 1 (<10%); D2 = damage 2 (>40%); D3 = damage 3 (>90%). Error bars represent standard error of the mean. Different letters indicate differences at Tukey's test (α = 0.05), while "ns" stands for "not significant".

When considering all *H. halys* individuals, nymphs and adults, significant effects of the treatment (2020: $F_{2,\ 96} = 46.28$, $P < 0.001$; 2021: $F_{2,\ 179} = 41.61$, $P < 0.001$), the time (2020: $F_{15,\ 96} = 9.56$, $P < 0.001$; 2021: $F_{11,\ 179} = 5.21$, $P < 0.001$) and of the interaction "time × treatment" (2020: $F_{30,\ 96} = 3.60$, $P < 0.001$; 2021: $F_{22,\ 179} = 1.90$, $P = 0.012$) were found. In netted plots there was a reduction in *H. halys* abundance (FF and GF, $P < 0.001$) if compared to un-netted ones for both years. Moreover, *H. halys* infestation was always lower in FF than GF (2020: $P = 0.028$; 2021: $P = 0.005$).

When considering fruit damage at harvest, the percentage of undamaged apples differed among treatments (2020: $F_{2,\ 24} = 220.4$, $P < 0.001$; 2021: $F_{2,\ 50} = 57.25$, $P < 0.001$) and was higher in FF and GF plots than in CTRL ones (Fig. 4). Concerning damage categories, differences among treatments on fruits classified as D1 were also observed (2020: $F_{2,\ 24} = 35.90$, $P < 0.001$; 2021: $F_{2,\ 50} = 21.26$, $P < 0.001$), and in comparisons with CTRL plots there were fewer D1 apples in FF and GF, and in FF plots ($P < 0.001$) in 2020 and 2021, respectively.

Treatment influenced the amount of fruit in D2 category (i.e., up to 40% of fruit damage) for both years (2020: $F_{2,\ 24} = 220.5$, $P < 0.001$; 2021: $F_{2,\ 50} = 55.24$, $P < 0.001$), with fewer D2 fruits in FF and GF plots if compared to CTRL ones (Fig. 4). Fruits classified as D3 were however not influenced by the treatment (2020: $F_{2,\ 24} = 1.00$, $P = 0.383$; 2021: $F_{2,\ 50} = 1.00$, $P = 1.000$; Fig. 4).

3.2. Color of insect-proof nets

Halyomorpha halys population was continuously observed in the apple orchards throughout the two seasons. Its population density was variable among the years and during the season, with a higher infestation of adults and nymphs in 2020 than in 2021. Nymphs were generally observed from mid-June to the end of samplings, while adults from May onwards (Supplementary Fig. S4).

The infestation by *H. halys* adults was significantly affected by net color (2020: $F_{1,\ 90} = 197.23$, $P < 0.001$; 2021: $F_{1,\ 214} = 15.27$, $P < 0.001$) and the interaction "time × color" in 2020 ($F_{14,\ 90} = 8.40$, $P < 0.001$; Fig. 5). The interaction "time × color" was not significant in 2021 ($F_{9,\ 214} = 1.60$, $P = 0.115$; data not shown). The effect of time was significant in both years (2020: $F_{14,\ 90} = 7.07$, $P < 0.001$; 2021: $F_{9,\ 214} = 5.99$, $P < 0.001$). *Halyomorpha halys* adults were more abundant in BN plots than in WN ones, and this was observed in 2020 on dates following netting deployment (Fig. 5). Similarly to adults, nymphs infestation was influenced by the net color (2020: $F_{1,\ 96} = 278.49$, $P < 0.001$; 2021: $F_{1,\ 214} = 16.92$, $P < 0.001$), and the interaction "time × color" in 2020 ($F_{14,\ 90} = 15.85$, $P < 0.001$) but not in 2021 ($F_{9,\ 214} = 1.54$, $P = 0.136$). The effect of time was significant in both years (2020: $F_{14,\ 90} = 21.10$, $P < 0.001$; 2021: $F_{9,\ 214} = 3.46$, $P < 0.001$). In both years, nymph abundance did not differ among experimental plots prior to netting deployment (2020: $P = 0.878$; 2021: $P = 0.456$). More nymphs were observed in plots covered by black nets (BN) if compared to those with white net (WN), and this difference was observed on all sampling dates (Fig. 5). The effect of netting color observed on adults and nymphs was also reflected on overall population level in both years (color, 2020: $F_{1,\ 90} = 431.03$, $P < 0.001$; 2021: $F_{1,\ 214} = 29.26$, $P < 0.001$; time, 2020: $F_{14,\ 90} = 12.72$, $P < 0.001$; 2021: $F_{9,\ 214} = 3.04$, $P = 0.002$; time × color, 2020: $F_{14,\ 90} = 14.90$, $P < 0.001$; 2021: $F_{9,\ 214} = 1.45$, $P = 0.169$), with a higher number of *H. halys* in BN than WN ($P < 0.001$).

The percentage of undamaged fruits at harvest was different between netting colors (2020: $F_{1,\ 10} = 765.26$, $P < 0.001$; 2021: $F_{1,\ 21} = 62.20$, $P < 0.001$) with a higher percentage of undamaged fruits in WN than in BN plots (Fig. 6). Differences among treatments on fruits classified as D1 were observed only in 2021 ($F_{1,\ 21} = 46.26$, $P < 0.001$), and not in 2020 ($F_{1,\ 10} = 0.29$, $P = 0.599$; Fig. 6). The percentage of damage in 2021 was lower in WN than in BN ($P < 0.001$). Concerning the D2 category, more fruits with symptoms were observed in BN than in WN (2020: $F_{1,\ 10} = 298.59$, $P < 0.001$; 2021: $F_{1,\ 21} = 32.56$, $P < 0.001$; Fig. 6). Effect of netting color was found on D3 category only in 2020 ($F_{1,\ 10} = 311.92$, $P < 0.001$), but not in 2021 ($F_{1,\ 21} = 3.90$, $P = 0.061$). In 2020, the percentage of D3 apples was lower in WN than BN ($P < 0.001$; Fig. 6).

An effect of netting color was found on the maximum daily temperature in 2021 ($F_{1,\ 47} = 807.08$, $P < 0.001$), that was higher in WN plots than in BN ones ($F_{11,\ 47} = 2.86$, $P = 0.006$). No differences were however observed in 2020 ($F_{1,\ 55} = 1.23$, $P = 0.273$). No difference were found for the minimum daily temperature (2020: $F_{1,\ 56} = 2.00$, $P = 0.163$; 2021: $F_{1,\ 47} = 0.22$, $P = 0.640$; Table 1), while the mean daily temperature resulted higher under WN than BN, in 2020 ($F_{1,\ 56} = 5.95$, $P = 0.018$) but not in 2021 ($F_{1,\ 47} = 0.01$, $P = 0.968$; Table 1). In both years, the minimum (2020: $F_{1,\ 56} = 17.34$, $P < 0.001$; 2021: $F_{1,\ 47} = 41.73$, $P < 0.001$), maximum (2020: $F_{1,\ 56} = 22.5$, $P < 0.001$; 2021: $F_{1,\ 47} = 20.54$, $P < 0.001$) and mean relative humidity (2020: $F_{1,\ 55} = 43.65$, $P < 0.001$; 2021: $F_{1,\ 48} = 84.92$, $P < 0.001$) were influenced by the color of net, with a higher relative humidity recorded in BN than in WN plots (Table 1).

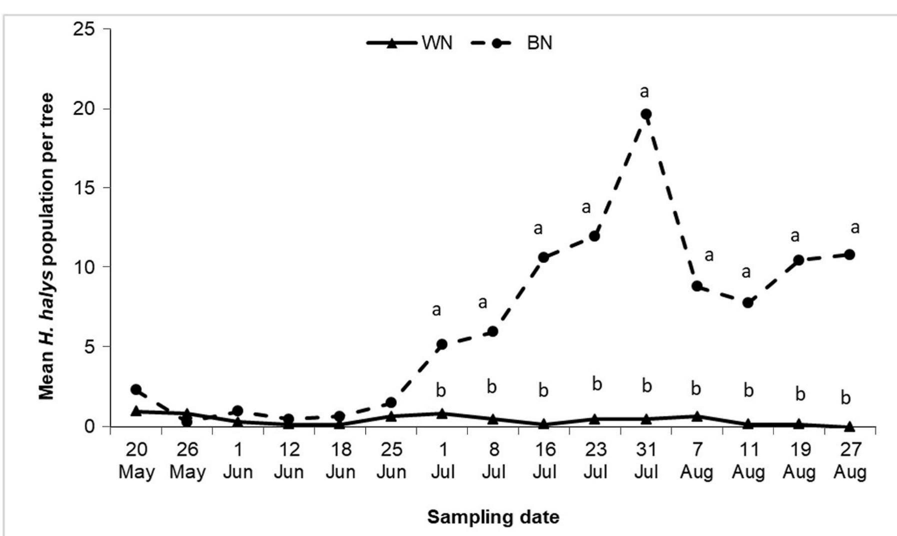

Fig. 5. *Halyomorpha halys* overall population observed in the apple orchard (2020) in the field experiment 2. BN = black netting; WN = white netting. Different letters indicate differences at Tukey's test (α = 0.05).

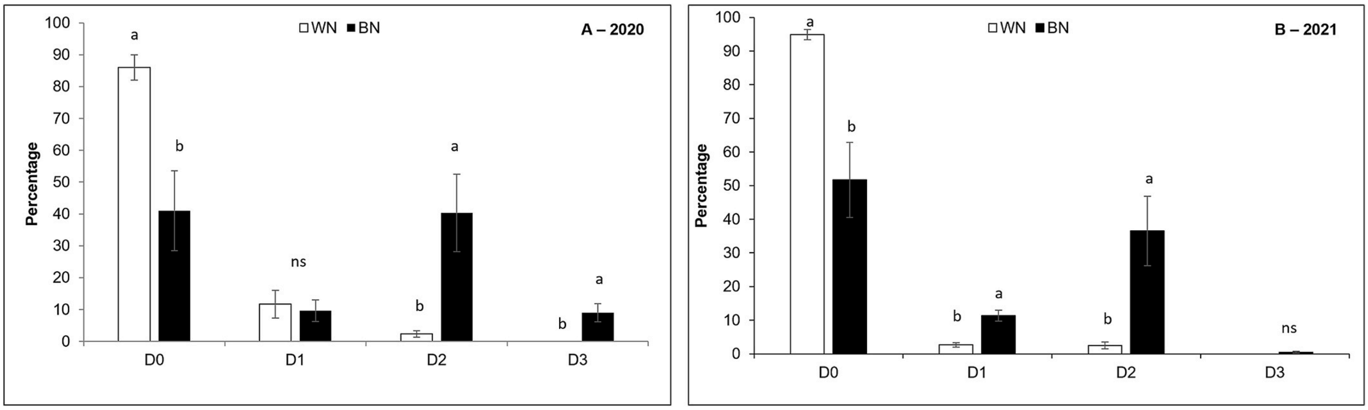

Fig. 6. Percentage of undamaged and damaged fruits caused by *H. halys* estimated at apple harvest in 2020 (A) and 2021 (B) in the field experiment 2. BN = black netting; WN = white netting. Four classes: D0 = undamaged fruit; D1 = damage 1 (<10%); D2 = damage 2 (>40%); D3 = damage 3 (>90%). Error bars represent standard error of the mean. Different letters indicate differences at Tukey's test (α = 0.05), while "ns" stands for "not significant".

Table 1
Mean of temperature (°C) and relative humidity (%): minimum, maximum and mean, observed in the apple orchard (2020–2021) in the field experiment 2. "±" represents the standard error of the mean. Different letters indicate differences at Tukey's test (α = 0.05), while "ns" stands for "not significant".

Year	Net color	Temperature (°C)			Relative humidity (%)		
		minimun	maximun	mean	minimun	maximun	mean
2020	**White**	16.7 ± 0.37 **ns**	30.1 ± 0.42 **ns**	23.4 ± 0.40 **a**	51.5 ± 0.91 **b**	96.0 ± 0.43 **b**	74.6 ± 0.86 **b**
	Black	16.4 ± 0.38 **ns**	29.9 ± 0.45 **ns**	23.0 ± 0.40 **b**	54.6 ± 0.95 **a**	97.9 ± 0.32 **a**	77.7 ± 0.80 **a**
2021	**White**	17.1 ± 0.24 **ns**	32.2 ± 0.31 **a**	24.1 ± 0.27 **ns**	44.9 ± 0.92 **b**	86.3 ± 0.56 **b**	66.4 ± 0.78 **b**
	Black	17.0 ± 0.23 **ns**	30.6 ± 0.37 **b**	24.1 ± 0.26 **ns**	48.7 ± 0.81 **a**	89.1 ± 0.60 **a**	69.3 ± 0.73 **a**

4. Discussion

In invaded areas, *H. halys* is causing economic losses, especially on pome fruit crops, and insect-proof netting has been proposed as an effective pest management tool (Candian et al., 2018; Caruso et al., 2019). In these studies, the authors evaluated the impact of a netting system on key fruit pests and also on invasive species such as *H. halys*. Here we performed experiments to optimize the use of this tool in pome fruit crops.

Our results confirm that insect-proof nettings are effective in controlling *H. halys* infestations in fruit orchards. We also observed an effect of timing of netting deployment on *H. halys* adult infestation, especially in apple orchards. Deployment of insect-proof netting at flower fading phase reduced the colonization by overwintering adults and ensured the reduction of damage at harvest. In pear orchards, the results confirmed the efficacy of insect-proof netting in the control of *H. halys* infestation and damage, but no effect of timing was found. Our results show that the netting deployment could be delayed until the growing fruit phase on pear orchards, while to minimize *H. halys* damage, it should be performed at flower fading phase on apple. The differences between the two crops are probably related to different phenology and the orchard invasion pattern of the pest. In particular, the different phenology of crops, pear flowering and start of fruit growing being earlier than apple. In north-eastern Italy, pear flowers from the end of March to mid-April (Fideghelli et al., 2009), while apple flowers from mid-April to mid-May (Fideghelli et al., 2008). In Central Europe as well as in northern Italy, *H. halys* adults start to emerge in high numbers from overwintering sites during March–early April, generally increase in May and peak in late spring, keeping exiting from overwintering sites until early June (Bergh et al., 2017; Bosco et al., 2019; Costi et al., 2017; Haye et al., 2014). In field observations, Lee et al. (2013) found that *H. halys* often disperses among different habitats in search of preferred hosts, and this includes its dispersal from overwintering sites to host plants early in the spring. There is usually a lag between the period in which *H. halys* disperses from overwintering sites and when it appears in agricultural crops such as fruit orchards. It is suspected that during this early spring period, adults utilize host trees within the forest edge habitats for early season feeding and oviposition (Leskey and Nielsen, 2018; Rice et al., 2014). Then *H. halys* adults disperse from wild hosts to suitable crops following availability of fruiting structures (Bosco et al., 2019; Martinson et al., 2015). In the early stage of the season, when pears are in the flowering and growing fruit phases, the presence of *H. halys* adults are still at low levels, while an earlier deployment of nets can make a difference in apple fruits that have a later development. Pest dispersal, as proposed for other insects, is an important factor when studying its population dynamics (Jennersten, 1988; Kennedy and Storer, 2000; Stinner et al., 1983), in order to enhance management tactics against an invasive and damaging pest (Hughes and Dorn, 2002; Zhang et al., 2009). The behavior of *H. halys* also depends on environmental features at certain temperatures. *Halyomorpha halys* have a high flight activity when the air temperatures are steadily above 15 °C (Lee et al., 2013; Lee and Leskey, 2015) that the study area are typically observed from the end of April, mid-May. In this phase the pest pressure is higher compared to earlier in the season (i.e., 50% of overwintering adults; Bergh et al., 2017; Bosco et al., 2019; Haye et al., 2014). Additionally in this period of the season, apple trees could become attractive to *H. halys* (Leskey et al., 2012; Nielsen and Hamilton, 2009), and thus the deployment of insect-proof netting could subsequently reduce its infestation and damage in the orchard.

Insect-proof netting is a tactic for managing *H. halys* in fruit orchards (Candian et al., 2018, 2021), and net efficacy could be influenced by the netting system color, with black having higher infestation levels and fruit damages than white. These differences can be related to the different attraction of white and black colors to *H. halys*, which prefer cool and dark areas (Bae et al., 2019; Cullum et al., 2020; Hancock et al., 2019; Lee et al., 2014a, 2014b; Toyama et al., 2011). White and black netting strongly differ in light-reflectance, and this could induce differences in microclimatic conditions (i.e., temperature and humidity; Castellano et al., 2008). It is known that temperature and humidity can influence insect pest population dynamics (Chown et al., 2011).

Concerning temperature, the range is an important factor, but also the length of time of temperature exposure could affect the *H. halys* population dynamics (Baek et al., 2017; Haye et al., 2014; Nielsen et al., 2008). Our results suggest that minimum temperatures do not differ under white or black nettings. However, the maximum temperatures reached during summer was higher under white as compared to black nettings, especially in 2021, when it reached 32.2 °C under white nettings and 30.6 °C for black ones. Prolonged periods of temperatures above 30 °C are detrimental to *H. halys* development and survival, and at 35–40 °C the impact on the population can be notable, acting on both egg hatching, nymph and adult survival (Aigner and Kuhar, 2016; Baek et al., 2017; Fisher et al., 2021; Haye et al., 2014; Nielsen et al., 2008; Scaccini et al., 2019). Consequently, high temperatures measured under white nets could have contributed to reducing the *H. halys* infestation in field conditions. In addition to temperature, relative humidity was also influenced by the color of nettings, resulting higher in plots covered with a black net than in those covered with a white net. Relative humidity can impact on *H. halys* nymph survival and egg hatching, for instance in the case of low relative humidity (i.e., <30%; Fisher et al., 2021; Khadka et al., 2020; Stahl et al., 2021), but the levels of humidity observed here were always above critical thresholds for insect survival.

Net color can also influence the pruductive aspects of the crop. Tree physiology and fruit quality are influenced by the shading factor of a net, and crop radiation requirements may lead to differences in color, weight, size of the fruit, and internal quality. The selection of the best net color to adopt thus depends on the cultivated crop/cultivar, expected production, location of the area and its climatic conditions, and the biotic and abiotic issues to counteract (Manja and Aoun, 2019).

Adopting netting systems requires a cost-benefit analysis considering the high installation and maintenance costs (e.g., Del Fava et al., 2017; Ebbenga et al., 2019; Johnson et al., 2020). If, on the one hand, the proper netting deployment showed decreased infestation and damage caused by *H. halys*, on the other hand, a single-row netting system could lead to difficulties in management for pesticide spray and fruit harvest. In previous studies, direct and indirect estimated costs in raspberries and grapes exceed $6000 per acre (almost $15,000 ha^{-1}; Leach et al., 2016; Ebbenga et al., 2019). An economic analysis is thus required for different field systems, which has to include reducing the cost of insecticide applications against pests such as the brown marmorated stink bug. For this purpose, deployment time and netting color must be considered in fruit orchards.

5. Conclusions

Halyomorpha halys requires an integrated management approach where different tactics are combined to keep the infestation level below the economic injury level. Insect-proof exclusion netting represents a key pest control tactic in fruit orchards that helps to reduce chemical use, but it requires high initial financial investment and management effort, and maximization of its efficiency is critical. Based on the result obtained here, we can conclude that the efficacy of these tactics can be maximized by an early deployment in apple orchards at the flower fading phase, and this can be delayed until the growing fruit phase on pear orchards. White or clear color nettings should be preferred over black or dark colored ones since the lower *H. halys* infestation and damage.

Author contributions

DF, DS, and AP planned and designed the research. DF, VL, and GG performed the experiments and analyzed the data. DF, DS, and AP wrote the original draft of the manuscript. DF, DS, and AP reviewed and edited the final version of the manuscript. AP supervised the project and provided funding. All authors have read and agreed to the final version of the manuscript.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.cropro.2023.106331.

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