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Highlights

1. SHD olive orchard system requires agronomic practices influencing pest behaviour.
2. The *Bactrocera oleae* infestation percentage was reduced by SHD management.
3. SHD management might benefit *Palpita vitrealis* and *Otiornychus cribricollis*.
4. SHD management affected the minor pests *Liothrips oleae* and *Euphillura olivina*.

Abstract

The increasing use of the super high-density (SHD) olive orchard system requires a careful assessment of its potential impact on the main olive pests. The purpose of this study is to evaluate the SHD effect on *Bactrocera oleae* and other harmful phytophagous species by assessing damages caused by each pest. For three years, in three different sites in Tuscany established with Italian selection or Spanish Arbequina cultivars, the SHD management system was compared to an adjacent traditional olive orchard system in which the same soil and phytosanitary management were applied. Samplings of twigs and fruits from spring to fall together with adult monitoring of *B. oleae*, *Prays oleae*, and *Palpita vitrealis* by pheromone traps were used to determine infestation percentages and insects' population dynamics. *Bactrocera oleae*, *Liothrips oleae*, and *Euphillura olivina* were negatively affected by the SHD olive orchard system, while *P. vitrealis* and *Otiornychus cribricollis* were favoured by this management. *Bactrocera oleae* total infestation was reduced to almost 50% in the SHD system. In contrast, *Palpita vitrealis* showed high infestation levels in more vigorous cultivars. Further studies are required for a complete evaluation of the impact of SHD management on olive pests.

Introduction

Super high-density (SHD) olive growing is a cultivation system with very a high planting density ranging from 1000 to 2500 plants/ha (Lo Bianco et. al., 2021). It has been developed in Spain since the beginning of the 1990s and is currently gaining popularity in the world due to its many advantages, including early entry into production, very low alternation of production, higher productivity, high yields, mechanization of all operations, and the reduction of production costs. In Italy over the last 10 years, an area of 5,000 hectares was covered by this olive cultivation essentially with two varieties for SHD: Arbequina (85%) and Arbosana (10%). Today the varietal offer is much wider: Arbequina accounts for 34% of the SHD surface in Italy, followed by other cultivars such as Oliana, Arbosana, Diana, Tosca, and Maurino selection Vittoria (Sportelli, 2022).

The use of the SHD olive orchard system is also expanding in Tuscany, a region located in the Central Northern part of Italy, where olive tree cultivation is of great importance for its impact on the local

economy, environment, landscape, and biodiversity. Regional olive cultivation covers more than 89,000 hectares of land, with over 15 million plants and as many as 80 cultivars of native olive trees, among which the most widespread are Frantoio, Moraiolo, Leccino, Maurino, and Pendolino (<https://www.regione.toscana.it/produzioni-vegetali/olio-di-oliva>). The key element of the olive/oil regional sector is the high quality of the production: 40% of the olive-growing area is designed for certified productions, such as the Protected Designations of Origin (PDO) and Protected Geographical Indications (PGI) of extra virgin olive oils (Pupo D'Andrea and Sarnani, 2021). The SHD olive orchard system in Tuscany involves the use of both local cultivars, preserving the typical characteristic of the product, and Spanish cultivars highly suitable for SHD.

The olive crop intensification based on an increase in the number of plants per hectare, could have an impact on the main olive pests and diseases. It is worth noting that for the SHD olive orchard system, several factors such as requiring more irrigation and fertilization, the changes in some cultivation practices (e.g. completely mechanized pruning and harvesting), and the adoption of specific cultivars might determine variations in pest behaviour and spread of diseases. Landi et al. (2022) have recently demonstrated in a field experiment, that SHD affected the soil nematode community, with particular regard to the plant-parasitic nematode community. The individuals belonging to the families Telotylenchidae, Paratylenchidae, Meloidogynidae, and Criconeematidae were favoured in the SHD olive orchard system, while Longidoridae, Heteroderidae, and Pratylenchidae were disadvantaged. The assemblage of microarthropods was confirmed to be an effective tool to determine the quality of soils in olive groves and maintenance of soil ecosystem services. Simoni et al., (2021) conducted studies on the edaphic arthropod community in SDH olive groves with different soil management practices, demonstrating how the ecotonal microenvironment between the surface and edaphic habitat was chosen by feeding functional groups (including hemiedaphic, macrosaprophages, polyphages, predators).

To date, specific field research focused on the impact of SHD on well-known and widespread pests of olive orchards is lacking. In Spain, several questionnaires have been posed to technicians, experts in the cultivation of olive groves, and researchers about this topic (Barranco et al., 2008; Jiménez Díaz, 2008). The respondents highlighted that plant density could be a relevant factor in the increase of pests and diseases followed by irrigation and fertilization, while soil management, harvest, and pruning were considered lesser relevant. The expansion of new intensive plantations has been associated with an increase in the incidence of pests and diseases, even if in most cases, observational data prevail over experimental ones, which at times are not conclusive (Rallo et al., 2013).

The aim of the present study was to assess the impact of SHD olive orchards on the key pest *Bactrocera oleae* (Rossi) (Diptera: Tephritidae) and other harmful and secondary pests, particularly,

Prays oleae Bernard (Lepidoptera: Hyponomeutidae), *Palpita vitrealis* (Rossi) (Lepidoptera: Pyralidae), and *Otiorrhynchus cribricollis* Gyllenhal (Coleoptera: Curculionidae). The SHD was compared to traditional (TRA) olive orchard systems in three sites (Tuscany, Italy) characterized by different soils, climatic conditions, and cultivars. In this study, the hypothesis was that *P. vitrealis* and *O. cribricollis* infestation levels could be strongly affected by SHD system due to heavy pruning and high production of plant biomass. Conversely, *B. oleae* and *P. oleae* could be less affected by the changes in management. In detail, the effect of SHD management system was assessed by: (i) damage caused by each pest; (ii) monthly catches by pheromone traps for the main pests; (iii) interactions between sites and the agronomic management.

Materials and Methods

Field sites and experimental design

The three experimental olive groves selected in this study were in Tuscany (Central Italy) in the provinces of Florence (Poggio a Remole) and Siena (Rapolano and Buonconvento) (Table 1).

The three experimental fields were situated on hilly positions; soil texture evidenced a high occurrence of clay. Based on Köppen Climate types, the Florence site was characterized as temperate with a hot summer and without a dry season, while the two sites located in Siena were described as temperate with a dry and hot summers. In all sites, between 2018 and 2020, the period from June to September showed the highest temperatures without relevant differences among the years (maximum temperature 32-33°C in August) (Fig. 1). The lowest rainfall in Poggio a Remole (only 878.6 mm per year) with the minimum in September (10 mm) was recorded in 2018. In the two sites located in Siena province, the yearly lowest rainfall was recorded in 2020 (751.6 mm and 670.8 mm in the Rapolano and Buonconvento areas, respectively); the minimum monthly values were recorded in June 2019 (3.4 and 0.6 mm in the Rapolano and Buonconvento areas, respectively) (fig. 1).

Two adjacent areas belonging to the same farm were selected to compare the infestation of the olive insect pests between the SHD and TRA olive orchard systems. The total experimental area of each farm was 3.0 ha consisting of two plots (1.5 ha per plot for each management), in which three sub-areas (0.5 ha each) were selected for the trials. The SHD olive orchards and the traditional (TRA) plantings covered 1,500 and 400 plants/hectare, respectively. In all sites the olive trees had reached the full-grown stage: TRA orchards had been used for olive cultivation for over 40 years, while the SHD orchards for approximately 10 years. The SHD system differed from the TRA system because cultivars were mainly characterized by their small-medium size. In particular, the Poggio a Remole and Rapolano sites were experimental fields because local small Italian olive cultivars were tested to

preserve the Italian biodiversity and characteristics of oil production (University of Florence and National Council of Research). Conversely, in the Buonconvento site, only the Spanish Arbequina cultivar was grown. In all cases, the varieties used were characterized by a small-medium fruit size. In each site, the same soil and phytosanitary practices were applied under both systems. Mechanical pruning and harvest were used in SHD plots, the hard hedging and topping pruning were performed annually in winter. Instead, in TRA, the light pruning was done by hand each year. The irrigation system was established in the SHD olive orchard management in all sites but never used throughout the year over the three years of the study.

Sampling design and monitoring of main phytophagous insects

Samples from the study fields were collected every two weeks for three years (2018-2020) in each sub-area per management system and site (three samples/management systems/fortnightly/year/location). Random samplings of twigs of 30 cm (10 per sub-area) from spring to fall were carried out during the three years. Fruits (100 per sub-area) were randomly picked from several trees from July to October (once the olives reached the stone hardening stage = BBCH 75) and examined in the laboratory to evaluate potential damage. Male pheromone traps (produced by ISAGRO, Via Caldera, 21 - 20153 Milano) were used for monitoring the three main insect pests: *B. oleae*, *P. oleae*, and *P. vitrealis*. In each sub-area, one trap for each of the monitored species was placed, following the factory instruction to cover the whole investigated area, in April and checked out every two weeks until October. The pheromone dispensers were replaced monthly. To detect, *O. cribricollis* adults, tissue bands (Rincotrap®, Greenagri srl Via M.L. King 55 70124 Bari Italy) were placed around the collar of the olive trunks. During the monitoring activities, minor pests were found to damage the olive plants. The most widespread among them were evaluated.

Laboratory observation

The collected twigs and fruits were examined for each pest investigated. The damage caused by larvae of the jasmine moth (*P. vitrealis*) was measured as the number of attacked leaves/shoots presenting large deep holes or entirely removed parts. The damage caused by the other pests was evaluated as the number of leaves or shoots with typical symptoms over the total number of leaves or shoots. Damages due to *Liothrips oleae* (Costa) (Thysanoptera: Phlaeothripidae), were evaluated by counting shoot and leaf distortions. Fruit damage by *B. oleae*, *P. vitrealis*, and *P. oleae* was evaluated as the ratio between the number of fruits infested by the target pests and the total number of collected fruits. The *P. oleae* anthophagous generation was measured as the number of flowering shoots infested by larvae with respect to the total number of shoots with healthy flowers.

Statistical analysis

A three-way ANOVA was performed to assess the influence of management, site, and year on the main olive orchard pests measured as the percentage of total infestation. Moreover, the trend of *B. oleae*, *P. oleae*, and *P. vitrealis* monthly catches was also evaluated with the same statistical analysis. When the F-test was significant at $P < 0.05$, treatment means were compared using the Student-Newman-Keuls by CoStat statistical software package (<https://www.Cohort.com/costat.html>). In addition, different species compositions were compared for site, management, and their interactions. Olive tree phytophagous insect communities were compared using two-way Permutational multivariate analysis of variance (Permanova) based on Bray-Curtis similarity testing provided by the Past analysis package (Past Statistical Software, 2020, Oslo, Norwegian).

Results

Twelve species of olive pests were identified in twig and fruit samples collected in the three selected sites (Table 2). Two-way Permanova showed significant differences for site ($df = 2$; $F = 19.05$; $p = 0.0001$) and management ($df = 1$; $F = 13.24$; $p = 0.0001$) in the pest community compositions. Moreover, the interaction between site and management was also significant ($df = 2$; $F = 5.20$; $p = 0.0005$). Six species were distributed in all sites, but their frequency was different between the two olive orchard systems. *Bactrocera oleae*, *P. oleae*, *L. oleae*, and *E. olivina* showed the highest distribution percentage values in the TRA olive orchard system, while *O. cribricollis* showed the opposite trend. Only *P. vitrealis* evidenced no differences between the two management systems. Frequency percentages of other species were specific to each site, and, generally never exceeded 30%.

Olive fruit fly Bactrocera oleae

The three-way ANOVA showed that the monthly catches and the total infestation of olive fruits were significantly higher in the TRA olive orchard system than in the SHD one (Table 3; Fig. 2). No difference was found among the sites regarding catches, while, in the Buonconvento site the highest value of total infestation was recorded. The highest number of individuals captured by the pheromone traps was found in 2019, while no difference was reported among the years in terms of total infestation. The interactions between site-management and site-year were significant for total infestation (Table 3).

Olive moth Prays oleae

Prays oleae showed slightly higher catches in the TRA olive orchard system than in SHD one; however, the plant infestation has always been low, with no difference between the two managements (Table 4; Fig. 3). The anthophagous generation caused more damages than the phyllophagous and carpophagous generations even though damages never exceeded 30%, with no differences between the two managements. For site and year, monthly catches also showed significant differences: the highest captures were reported in the Florence site, while the lowest values were recorded in 2020 in all sites. The interactions between site-year were significant for monthly catches and flower infestation.

Jasmine moth* *Palpita vitrealis

The jasmine moth captures showed very low values without differences for site, management, and year (Table 5). By contrast, shoot and fruit infestations evidenced significant differences: twig infestation was higher in the SHD olive orchard system than in the TRA system, while fruit infestation showed the opposite result. Significant differences in infestation were also observed among the sites: high values, concerning both twigs and fruits, were found in the Rapolano (Siena province) site. It is worth noting that, here, the infestation rate was very high, reaching 50% of the infestation in the SHD system (Fig. 4). The interactions between site-management and site-year were significant for twig infestation, and between site-year and management-year for fruit infestation. Moreover, the interaction between site-management-year was significant both for twig and fruit infestation.

Minor pests

Otiorynchus cribricollis infestation was often higher in the SHD system than in the TRA system, even though its damage was negligible: leaf infestation never exceeded 15% (Table 6; Fig. 5). Although *Liothrips oleae* and *Euphyllura olivina* (Costa) (Hemiptera: Psilloidea) were widespread, their infestations were irrelevant as damage during the experimental period. However, in the TRA olive orchard system were found infestation values significantly higher than in the SHD system. The interaction between site-management was significant for the infestation due to *L. oleae* and *E. olivina*. Moreover, the interaction between site-management-year was significant for the infestation caused by *O. cribricollis* and *L. oleae*.

Discussion

Tuscany represents the northernmost Italian area of extensive olive cultivation since this species is characterized by a relatively high critical growth temperature (-10/12 °C) (Longo, 2019). However, in the next future, climate change might shift the biogeographical distribution of this plant northwards

(Rodrigo-Comino et al., 2021), and presumably the findings obtained within this study could rapidly change. In the three years of our investigation, irrigation was never applied due to the local climatic conditions, nevertheless, this practice might be required in the next future.

The selection of different experimental fields, in the same farm/area, allowed the comparison of SHD and TRA olive orchard system characteristics excluding other factors such as microclimate and soil management which were the same in both of the tested conditions. It is known that different soil managements can affect the survival of olive fruit fly pupae in the soil since tillage may partially influence their mortality by moving them into the deeper layers of the soil (Bachouche et al., 2018). The use of different cultivars between the SHD and TRA olive grove systems, depends on the different requirements of SHD system. However, the comparison between the two production systems (SHD vs TRA) was possible despite the different cultivars that have been investigated, at least as far as *B. oleae* was concerned. In fact, most of the varieties considered in our study, both those of new selection for the SHD system (Arbequina) and those already present in the Tuscan olive-growing heritage (Frantoio, Maurino and Leccino), are considered not particularly susceptible to this phytophagous (Burrack and Zalom, 2008; Malheiro et al., 2015). In Bartolini (2008) Pendolino, Leccio del Corno, and Moraiolo cvs, were also recorded as moderately susceptible.

During the three-yearly investigation, in accordance with other authors, the species most frequently found were *B. oleae*, *P. olea*, *P. vitrealis* *L. oleae*, and *E. olivina*, while *Saissetia oleae* (Olivier) (Hemiptera: Coccidae), *Rhynchites cribripennis* Desbrocher (Coleoptera: Curculionoidea), *Rhodocyrthus cribripennis* Desbrocher (Coleoptera: Curculionoidea), and *Lichtensia viburni* Signoret (Hemiptera, Coccoidea) occurred sporadically (Bagnoli, 2022; Del Bene et al., 1997). On the whole, the Permanova test evidenced that the composition of the entire pest community was subject to variations in both site and management in terms of distribution frequency percentage. Although differences for site were not relevant, they indicated that *B. oleae*, *P. oleae*, *O. cribricollis*, and *L. oleae* were more frequent in the Florence site, while *E. olivina* was more frequently detected in the other sites located in the Siena province. Several factors could be responsible for this finding such as different geographic parameters, agronomic practices, and climatic conditions. As regard climate, the two districts differed in the absence of a dry season in Florence. This could have determined better conditions for the development of the most common olive pests in this site. In terms of management, differences were not relevant too. However, the key pest *B. oleae* together with *P. oleae*, *L. oleae*, and *E. olivina* were favoured by the conditions created in the TRA olive orchard system. Conversely, *O. cribricollis* was favoured by the SHD system while the frequency of *P. vitrealis* was not influenced by orchard management. Moreover, differences between the two managements were more evident in the Siena districts that showed more extreme climatic conditions.

The total infestation percentage and the monthly catches of olive pests confirmed the findings of the PERMANOVA test. The key pest *B. oleae* showed double values in the TRA olive orchard system compared with the SHD one. This was contrary to our original hypothesis and some extent related to the fertilization and irrigation impact on *B. oleae* (Rodrigues et al., 2019; Yokoyama and Miller, 2007; Del Rio and Lentini, 2016). These authors have ruled out, that the incidence of the olive fruit fly in SHD olive orchard systems is higher due to irrigation and more conspicuous fertilization required for this management system. In our study in which no irrigation was applied and the fertilization is the same in both managements, many other factors may have contributed to the reduction of olive fruit fly infestation on SHD systems, such as the use of different cultivars characterized by small size or different canopy structures.

Studies on olive tree cultivars susceptibility to *B. oleae* reported ambiguous results (Rebora et al., 2020). Overall, all the cultivars involved in this study were characterized by medium size fruit. It is well known that fruit size is not the only key parameter for susceptibility to olive fruit fly attack, but there are others that contribute such as leaf and fruit VOCs, anthocyanin and glucoside content in the fruit, and fruit epicuticular waxes (Malheiro et al., 2015; Iannotta et al., 2006; Rebora et al., 2020). However, the susceptibility of different olive trees cultivars to *B. oleae* should be considered in light of the possible options made by the olive fruit fly and its ability to adapt (Bagnoli and Iannotta, 2012). The cultivar Arbequina, one of the most used in the SHD olive orchard system, could be less sensitive to olive fruit fly due to the small size of fruits, their green coloration, and the high production of oleuropein, a chemical compound considered one of the resistance factors to olive fruit fly (Kombargi et al., 1998; Mojdehi et al., 2019). Nevertheless, Burrack and Zalom (2008) suggested that female ovipositional preference is not necessarily related to the low capacity of non-attractive varieties to host larval development. In fact, the same authors suggested that Koroneiki olives presented the lowest number of *B. oleae* pupae, without avoiding their development. The other varieties with small fruits, such as Arbequina and Leccino, had low levels of infestation in the field, but high pupal yields (Burrack and Zalom, 2008). Moreover, Rizzo & Caleca (2006) found that fruit colour and olive hardness were important factors in determining the choice of drupes for oviposition by *B. oleae* females. Specifically, green olives were significantly more infested than the reddish and blackish one, instead, hardness played an important role in disfavoured female oviposition until the end of August-half of September before the olives turned darkcoloured.

It may be more interesting to consider the structure of the hedgerow. It is well known that, in summer, the olive fruit fly adults search for canopy areas with a more humid and cooler microclimate (Ricci and Ballatori, 1982). High summer temperatures (monthly average over 26°C with maximum values around 34°C) together with low relative air humidity can increase adult mortality and reduce egg

development (Avidov, 1957; Fletcher et al., 1978; Broufas et al., 2009; Wang et al., 2009). The TRA olive orchard system, characterized by a wider canopy, might represent a more favourable habitat for adult olive fruit flies than the SHD system. In the three monitored years, summer periods were very hot, even in September when adult fly incidence is high. As reported by Connor et al. (2014), the canopy of suitable SHD cultivars has an open structure with narrow and/or low leaf-area-density (LAD). Such a structure allows both good porosity, determining penetration of radiation into canopies for photosynthesis and ventilation, reducing pest/disease outbreaks. However, in all the years suitable for pest activity, the infestation in the study areas always required treatments in both management systems. Burrack and Zalom (2008) argued that flies, although accepting certain varieties as ovipositional substrates more readily than others, are capable of developing in all of them. The same concept could be applied to climate: olive fruit flies are more attracted to the microclimate found in a wider canopy, but they are potentially able to adapt to even worse climatic conditions.

The following pests *P. oleae*, *L. oleae*, and *E. olivina* also found unfavourable microclimates in SHD olive orchard system. Catches of *P. oleae*, one of the most important pests of olive trees in the Mediterranean Basin (Ramos et al., 1998; Caselli and Petacchi, 2021), but with moderate infestations in Tuscany, were slightly higher in the TRA management system, particularly in the Florence site. However, plant infestation was low and with no differences between the two management systems and the conversion to SHD did not change this trend. Although the anthophagous generation, the most dangerous in Tuscany, caused less than 30% of damage, its yield loss was counterbalanced in the SHD by higher number of flowers per hectare than in the TRA (Connor et al., 2014). Damage due to *L. oleae* and *E. olivina* was negligible, even though, they were disadvantaged by the SHD olive orchard management.

In contrast, *O. cribricollis* and *P. vitrealis* confirmed our hypothesis and showed an increased infestation percentage in the SHD olive orchard system compared with the TRA. The greater incidence on vegetation in SHD olive orchards, mainly due to pruning, favoured the infestation of *O. cribricollis* and *P. vitrealis*. In general, the damage was always irrelevant for *O. cribricollis*, while the *P. vitrealis* impact was different among sites. Although the *P. vitrealis* frequency was the same among the sites and between the two management systems, the infestation percentage showed relevant differences. In accordance with Connor et al. (2014), green pruning intensity depends on the vigour of the cultivated genotype. Some appropriate cultivars for the SHD olive orchard system, such as Arbequina, Arbosana, and Koroneiki, show medium-low vigour and slow canopy growth together with early bearing, and high yield efficiency (Camposeo et al. 2008). In the experimental site in the Siena province, in which only Arbequina was established, the infestation of shoots remained low and without differences between the SHD and TRA managements due to the low total plant biomass

produced. Conversely, in the sites in which Italian cultivars characterized by small and medium size were chosen, the infestation increased producing high total plant biomass and compromising olive yields. This was particularly evident in the Rapolano site because vigorous cultivars such as Frantoio and Coreggiolo were established together with more SHD suitable cultivars such as Maurino (Vivaldi et al., 2015; Tombesi and Farinelli, 2017). In fact, the Maurino cultivar was characterized by the low production of plant biomass as well as moderate olive yield (Vivaldi et al, 2015); hence, the necessity to include more productive cultivars.

Conclusions

The SHD olive orchard system had a moderate impact on the main olive pests. The most relevant effect was recorded for the key pest *B. oleae*, whose total infestation was reduced to almost 50% in the SHD system during the three years of the study. However, the intrinsic characteristics of the cultivars should be considered, as well as the possible difference in their productive load. Only *P. vitrealis* could compromise the olive yield if the chosen cultivars produced a large amount of plant biomass. To date, only SHD appropriate cultivars such as Arbequina prevent high levels of infestation. To this end, a complete evaluation of oil quality in cultivars suited to SHD management is necessary to maintain the product characteristics required for certified productions.

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Table 1. Geographical position, climate parameters by SIR/DATI Tuscany (<http://www.sir.toscana.it/consistenza-rete>), soil texture, and olive orchard features of the experimental sites.

Site	Geographical position		Climate parameters			Soil texture USDA	Orchard features	
	Coordinates	Altitude a.s.l. (m)	Köppen Climate types*	Mean air temperature (°C)	Mean annual precipitation (mm)		Olive tree cultivar	Soil management
Florence Poggio a Remole	43.800183 N; 11.403579 E	260	Cfa	14.7	940	Clay	SHD: Leccio del Corno, Tosca, Diana TRA: Frantoio, Leccino, Moraiolo	Conventional tillage Mineral fertilization
Siena Rapolano	43.275739 N; 11.603974 E	280	Csa	14.5	880	Silty Clay Loam	SHD: Maurino selection Vittoria, Frantoio, Leccino, Moraiolo, Pendolino, Leccio del Corno, Correggiolo TRA: Frantoio, Leccino, Moraiolo, Pendolino	Green cover Organic fertilization
Siena Buonconvento	43.0940 N; 11.2316 E	147	Csa	14.7	890	Clay Loam	SHD: Arbequina TRA: Correggiolo, Pendolino, Leccino	Conventional tillage Mineral fertilization

* Cfa – Temperate, no dry season, hot summer; Csa – Temperate, dry summer, hot summer

Table 2. Distribution frequency percentage of sampled phytophagous from May to October 2018, 2019, and 2020 in three sites in Tuscany.

Species	Florence (Poggio a Remole)		Siena (Rapolano)		Siena (Buonconvento)	
	SHD	TRA	SHD	TRA	SHD	TRA
<i>Bactrocera oleae</i>	75.9	77.8	56	64.8	40.7	63
<i>Prays oleae</i>	79.6	94.4	81.5	85.2	74.1	79.6
<i>Palpita vitrealis</i>	88.9	88.9	88.9	88.9	77.8	77.8
<i>Liothrips oleae</i>	90.7	96.3	75.9	85.2	72.2	88.9
<i>Otiorrhynchus cribricollis</i>	92.6	96.3	94.4	70.4	87	70.4
<i>Rhynchites (=Haplorhynchites) cribripennis</i>	0	0	33.3	72.2	1.9	0
<i>Rhodocyrthus cribripennis</i>	0	1.9	0	0	0	0
<i>Euphillura olivina</i>	3.7	7.4	1.9	31.5	9.3	22.2
<i>Saissetia oleae</i>	0	0	0	0	0	1.9
<i>Lichtensia viburni</i>	0	0	5.6	1.9	0	1.9
<i>Methriochroa latifoliella</i>	5.6	5.6	3.7	13	0	0
<i>Dasineura oleae</i>	1.9	0	0	0	0	0

Table 3. Effects of site, management, and year on monthly catches and total fruit infestation of *Bactrocera oleae*. Averages and standard errors are reported (n=3).

	Monthly catches	Total infestation %
Site		
Florence (Poggio a Remole)	18.4±3.05 a	3.2±0.64 b
Siena (Rapolano)	12.1±2.36 a	6.7±1.46 b
Siena (Buonconvento)	12.2±2.27 a	11.9±2.08 a
Management		
SHD	9.0±1.56 b	4.0±0.83 b
TRA	19.4±2.46 a	10.5±1.54 a
Year		
2018	12.9±2.21 b	7.4±1.35 a
2019	22.7±3.55 a	8.5±1.97 a
2020	7.0±1.05 b	5.8±1.28 a
Main effects:		
Site	df = 2 F = 2.23 p = 0.11	df = 2 F = 9.57 p = 0.0001
Management	df = 1 F = 13.95 p = 0.0002	df = 1 F = 16.07 p = 0.0001
Year	df = 2 F = 10.77 p = 0.00001	df = 2 F = 0.90 p = 0.41
Interactions:		
SxM	df = 2 F = 1.84 p = 0.16	df = 2 F = 3.56 p = 0.03
SxY	df = 4 F = 0.55 p = 0.70	df = 4 F = 3.23 p = 0.01
MxY	df = 2 F = 0.99 p = 0.38	df = 2 F = 0.53 p = 0.59
SxMxY	df = 4 F = 0.39 p = 0.81	df = 4 F = 1.76 p = 0.14

Table 4. Effects of the site, management, and year on monthly catches and total fruit infestation of *Prays oleae*. Averages and standard errors are reported (n=3).

	Monthly catches	Flower infestation %
Site		
Florence (Poggio a Remole)	122.3 ±16.54 a	26.1±4.72 a
Siena (Rapolano)	64.6±8.42 b	19.2±5.27 a
Siena (Buonconvento)	49.7±6.46 b	17.8±3.69 a
Management		
SHD	61.9±8.52 b	17.8±3.43 a
TRA	95.8±10.43 a	24.3±4.02 a
Year		
2018	107.2±14.18 a	23.3±5.06 a
2019	84.5±12.55 a	15.6±4.52 a
2020	45.0±6.21 b	24.2±4.16 a
Main effects:		
Site	df = 2 F = 12.61 p = 0.00001	df = 2 F = 1.51 p = 0.23
Management	df = 1 F = 7.43 p = 0.007	df = 1 F = 2.43 p = 0.13
Year	df = 2 F = 8.52 p = 0.0003	df = 2 F = 1.74 p = 0.19
Interactions:		
SxM	df = 2 F = 0.07 p = 0.94	df = 2 F = 1.28 p = 0.29
SxY	df = 4 F = 3.32 p = 0.011	df = 4 F = 8.72 p = 0.0001
MxY	df = 2 F = 1.26 p = 0.29	df = 2 F = 0.60 p = 0.55
SxMxY	df = 4 F = 0.77 p = 0.54	df = 4 F = 0.53 p = 0.71

Table 5. Effects of site, management, and year on monthly catches, twig, and fruit infestations of *Palpita vitrealis*. Averages and standard errors are reported (n=3). PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).

	Monthly catches	Twig infestation %	Fruit infestation %
Site			
PR	2.3 ±0.59 a	4.2±0.59 b	1.0±0.30 b
Rap	1.7±0.29 a	23.4±1.63 a	3.70±0.97 a
Buo	1.3±0.25 a	4.1±0.50 b	5.3±0.95 a
Management			
SHD	1.5±0.26 a	13.0±1.35 a	2.6±0.52 b
TRA	2.0±0.39 a	8.2±0.75 b	4.1±0.80 a
Year			
2018	2.1±0.58 a	8.9±1.25 b	6.4±1.15 a
2019	1.7±0.31 a	10.7±1.39 ab	2.4±0.61 b
2020	1.4±0.25 a	12.2±1.42 a	1.2±0.34 b
Main effects:			
Site	df = 2 F = 1.69 p = 0.19	df = 2 F = 157.30 p = 0.00001	df = 2 F = 9.90 p = 0.0001
Management	df = 1 F = 1.35 p = 0.25	df = 1 F = 22.70 p = 0.00001	df = 1 F = 3.54 p = 0.06
Year	df = 2 F = 0.64 p = 0.53	df = 2 F = 3.59 p = 0.03	df = 2 F = 16.27 p = 0.00001
Interactions :			
SxM	df = 2 F = 0.33 p = 0.72	df = 2 F = 19.34 p = 0.00001	df = 2 F = 0.96 p = 0.38
SxY	df = 4 F = 1.27 p = 0.28	df = 4 F = 12.07 p = 0.00001	df = 4 F = 3.43 p = 0.01
MxY	df = 2 F = 0.18 p = 0.84	df = 2 F = 1.74 p = 0.18	df = 2 F = 5.92 p = 0.003
SxMxY	df = 4 F = 0.32 p = 0.86	df = 4 F = 3.67 p = 0.006	df = 4 F = 3.07 p = 0.02

Table 6. Effects of site, management, and year on leaf infestation caused by *Otiorrhynchus cribricollis* and infestation of twigs caused by *Liothrips oleae* and *Euphillura olivina*. Averages and standard errors are reported (n=3). PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).

	<i>O. cribricollis</i> infestation %	<i>L. oleae</i> infestation %	<i>E. olivina</i> infestation %
Site			
PR	4.1 ±0.05 a	1.6±0.02 a	0.7±0.31 b
Rap	2.9±0.05 a	0.9±0.01 a	8.9±2.62 a
Buo	4.2±0.07 a	1.7±0.02 a	3.9±1.23 b
Management			
SHD	5.4±0.66 a	1.2±0.12 b	0.7±0.26 b
TRA	2.1±0.21 b	1.6±0.13 a	8.4±1.89 a
Year			
2018	3.9±0.05 a	1.9±0.02 a	2.1±0.77 a
2019	3.2±0.05 a	1.7±0.01 a	4.9±1.50 a
2020	4.0±0.07 a	0.7±0.01 b	6.6±2.46 a
Main effects:			
Site	df = 2 F = 1.49 p = 0.23	df = 2 F = 8.87 p = 0.0002	df = 2 F = 7.27 p = 0.0001
Management	df = 1 F = 24.08 p = 0.00001	df = 1 F = 5.51 p = 0.02	df = 1 F = 19.02 p = 0.00001
Year	df = 2 F = 0.58 p = 0.56	df = 2 F = 22.14 p = 0.00001	df = 2 F = 2.16 p = 0.12
Interactions:			
SxM	df = 2 F = 2.44 p = 0.09	df = 2 F = 4.64 p = 0.01	df = 2 F = 8.30 p = 0.0004
SxY	df = 4 F = 2.06 p = 0.09	df = 4 F = 2.18 p = 0.07	df = 4 F = 0.61 p = 0.66
MxY	df = 2 F = 1.22 p = 0.30	df = 2 F = 0.30 p = 0.72	df = 2 F = 2.19 p = 0.12
SxMxY	df = 4 F = 2.74 p = 0.03	df = 4 F = 2.37 p = 0.05	df = 4 F = 0.72 p = 0.58



Figure 1. Mean temperature and monthly rainfall measured in the three sites for three years. Med, mean temperature; Min, minimum temperature average; Max, maximum temperature average.

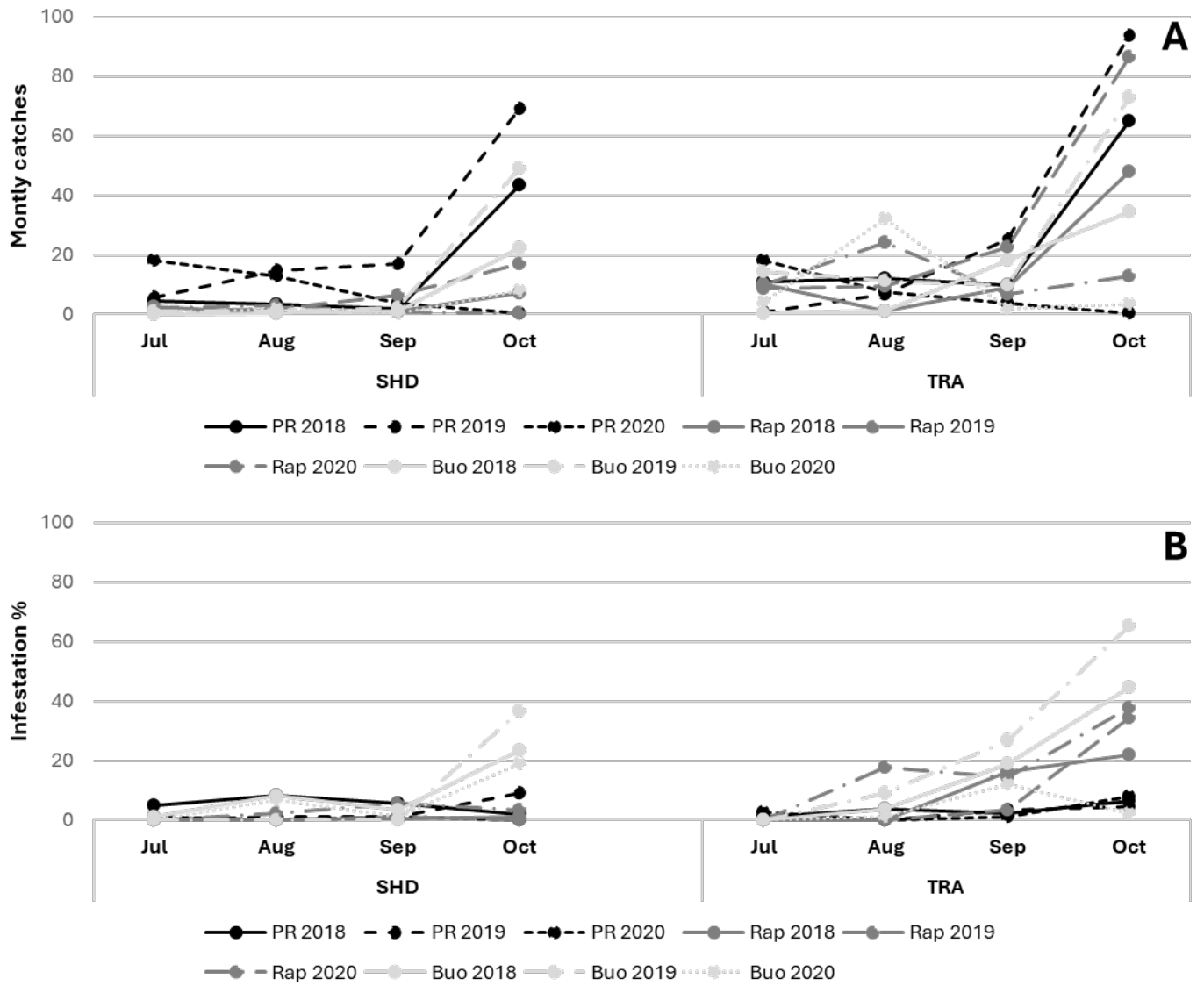


Figure 2. Captures of *Bactrocera oleae* males in pheromone traps/month (A) and total infestation percentage (B) from July to October between 2018 and 2020 in the three Tuscany sites. PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).

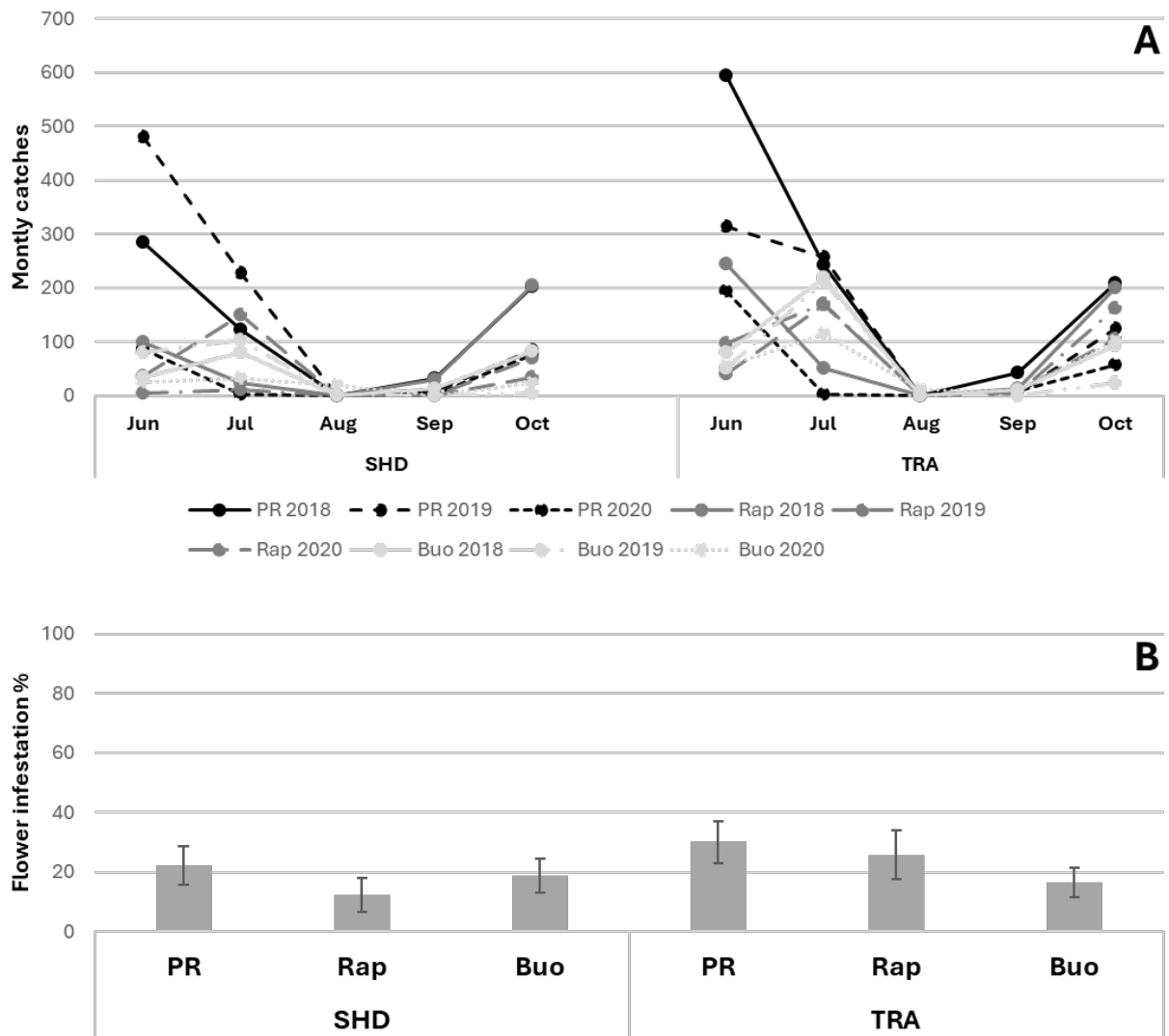


Figure 3. Captures of *Prays oleae* males in pheromone traps/month (A) from June to October and flower infestation percentage (first fortnight of June) (B) between 2018 and 2020 in the three Tuscan sites. PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).

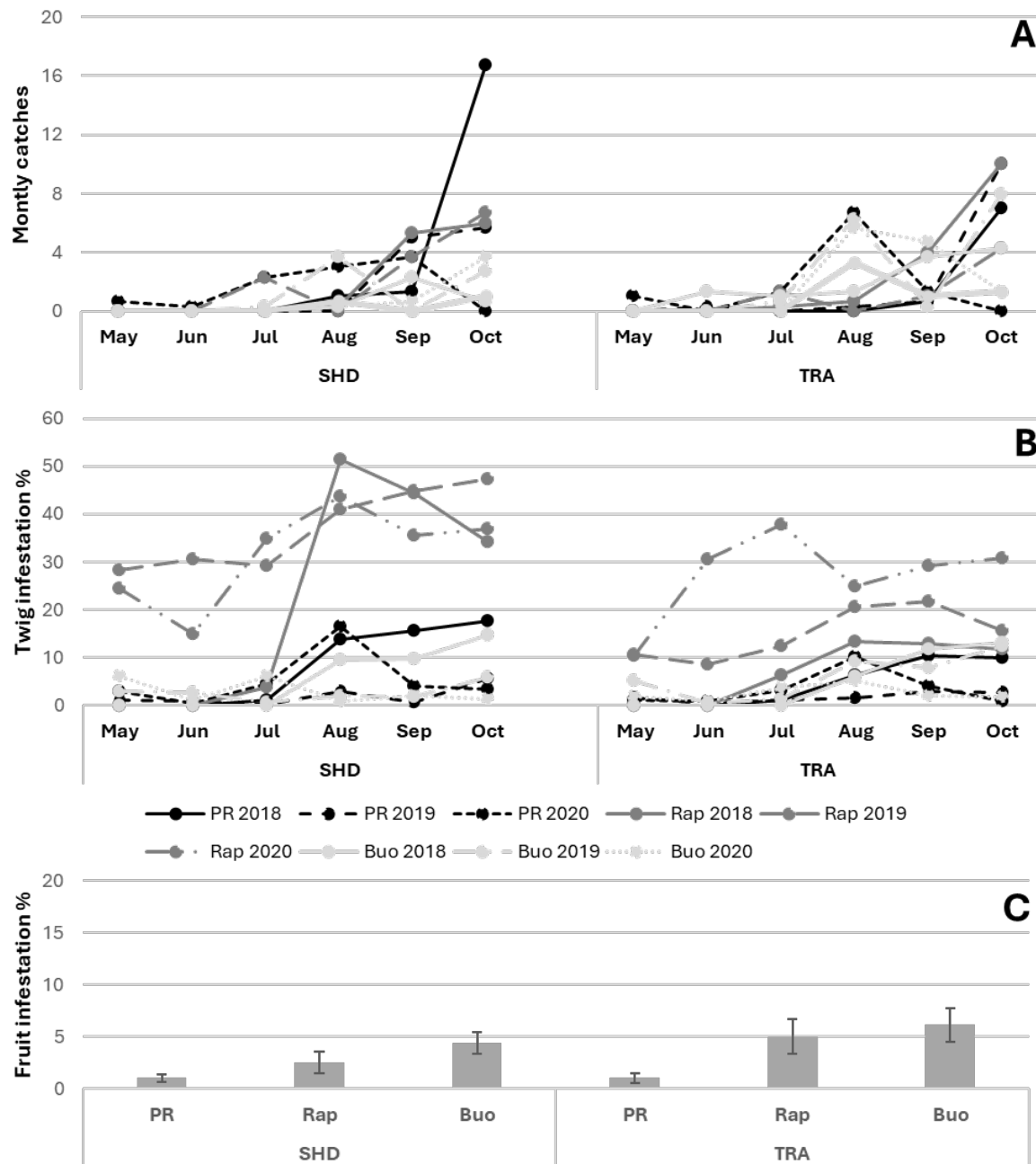


Figure 4. Captures of *Palpita vitrealis* males in pheromone traps/month (A) from May to October and twig (B) and fruit infestation percentages (C) between 2018 and 2020 in the three Tuscany sites. PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).

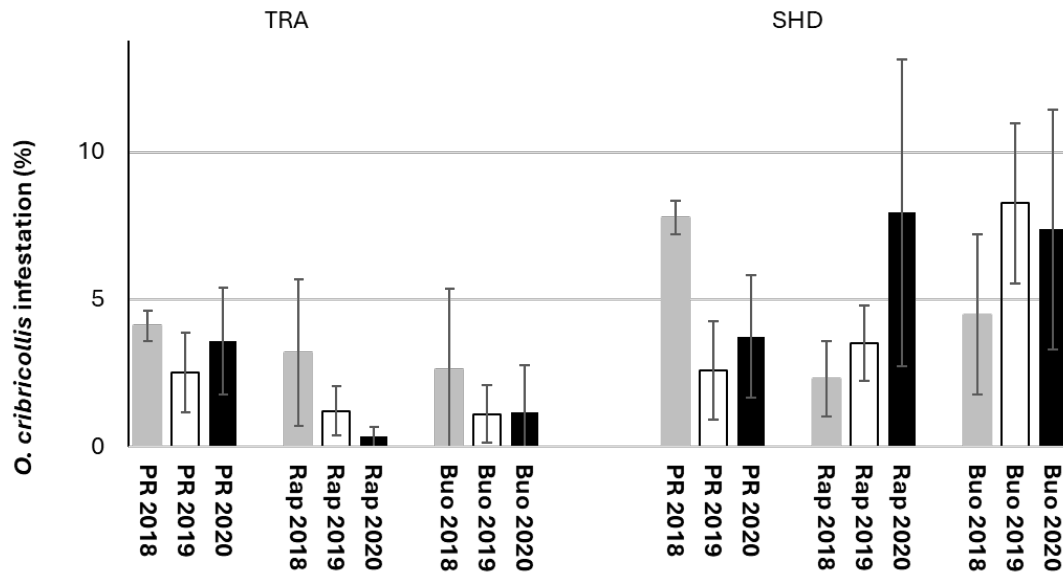


Figure 5. Leaf infestation caused by *Otiorhynchus cribricollis* from May to October between 2018 and 2020 in the three Tuscany sites. Averages and standard errors are reported (n=3). PR: Poggio a Remole (Florence); Rap: Rapolano (Siena); Buo: Buonconvento (Siena).