Studies on the damage potential of *Closterotomus trivialis* and *Aphanosoma italicum* on olive fruit setting

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Abstract

Closterotomus trivialis (Costa) (Hemiptera Miridae) is widespread in olive groves in the Mediterranean basin. This species feeds on flowers and it has been reported to reduce fruit setting due to flower abortion. However, there is contradictory evidence and its damage potential has not been clarified. The related species Aphanosoma italicum Costa is also commonly recorded on olive trees in the study area. The aim of this study was to investigate the potential of C. trivialis and A. italicum to impact adversely on olive fruit setting. Their damage potential was investigated at different stages of flower development, as this could constitute a critical factor in the damage incidence of these insects and thus being a key issue in unravelling the puzzle evidence. Shoots on the trees were enclosed in cages into which 3 or 6 adults in spring 2007 or 8 nymphs or adults in spring 2008 were introduced. In 2007 the introduction took place on 29 March. In 2008, the damage potential was investigated in 3 periods extending from early flower emergence to the start of the blooming period (placement of the cages on 7 and 27 March and 18 April). Records showed that in the cages where C. trivialis or A. italicum had been enclosed a high percentage of the fruiting organs dropped (99 and 91%, in each species, respectively). In the controls, the respective percentages were 63 and 64%. This high reduction in fruit setting was mostly attributed to bud abortion. The damage potential of both species was affected by the time of the infestation. Damage of C. trivialis can be severe from the early period of inflorescence development before they develop well and start blooming, whereas A. italicum showed a comparatively lower damage potential. In any case, at the period of blooming damage does not seem to be important. Thus, C. trivialis can cause serious fruit abortion. This knowledge can substantially contribute to the development of appropriate control programs of this pest in olive groves.

Key words: Closterotomus trivialis, Aphanosoma italicum, Miridae, flower abortion.

Introduction

The plant bug species Closterotomus trivialis (Costa) (Hemiptera Miridae Mirinae; previously known as Calocoris trivialis see Rosenzweig, 1997) is an insect commonly recorded on olive and citrus in the Mediterranean region. In the plain of Catania (Sicily, Italy) it is univoltine on citrus and overwinters in the egg stage. In late winter to early spring, eggs hatch and early instars feed on understorey weeds, with Urtica spp. (Urticaceae) and Parietaria officinalis L. (Urticaceae) being the main hosts. Later, larger instars and the emerged adults feed on the vegetative and reproductive parts of olives, but prefer buds and young flowers. This results in damage that may lead to abortion of buds and flowers. At the end of spring females oviposit their eggs, mainly in the wood of the trees exposed after pruning (Barbagallo, 1970).

C. trivialis is common in olive groves in several areas of Greece. It is univoltine and second and third instars are common at the end of February (Drosopoulos, 1993). On the island of Crete early instars of C. trivialis are first recorded at the end of January on weeds. Their population densities are generally much higher on weeds than on olive trees. On Crete the main weedy hosts are Urtica spp., P. officinalis, Mercurialis annua L. (Euphorbiaceae), Sonchus oleraceus L. (Compositae)

and Malva silvestris L. (Malvaceae) (Gerakaki et al., 2007).

C. trivialis has been reported as a serious pest of olive crops in areas of Italy (Barbagallo, 1970; Monaco, 1975). In certain areas of central Greece, Corfu, the western Peloponnesus and Crete, this species has been reported to cause reduced production in olive crops (Yamvrias, 1998; Gerakaki et al., 2007). However, its damage potential was investigated by Drosopoulos (1993) and he concluded that it did not cause significant damage to olive crops.

The following study sought to provide further evidence in deciding on the pest status and damage potential of *C. trivialis* on olives, and if it is found to be pestiferous, to aid in the development of appropriate management strategies for its control.

In our preliminary sampling of olive orchards in certain areas of Peloponnesus another species of mirine plant bug, *Aphanosoma italicum* Costa (Hemiptera Miridae) was also found on olive trees. Although this species has been reported from several regions of the eastern Mediterranean (Wagner, 1974; Tamanini, 1981), its ecology and damage potential are unknown. As a consequence, and as part of a broader aim of examining mirid species assemblages in olive crops, the damage potential of both *C. trivialis* and *A. italicum* were investigated in this study.

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In summary, the aims of this study were: 1) to evaluate the potential of these species to inflict damage on fruit setting of olives and 2) to elucidate whether damage intensity is related to olive tree phenology.

Materials and methods

This study was undertaken in a 1 hectare olive grove of the oil variety "Koroneiki" in the area of Pylos, in southwestern Peloponnesus. "Koroneiki" is the main variety cultivated for olive oil production in Greece. The olive grove was composed of trees with canopies of about 4-5 m in diameter. The standard cultivation practices of the area were applied in this olive grove, although no tilling or insecticidal treatments were applied during the course of the study.

The damage potential of *C. trivialis* and *A. italicum* was evaluated in spring 2007 and 2008. Shoots with flowering organs were enclosed in mouslin cages (25 cm diameter and 70 cm length) along with nymphs or adults of each pest species, separately. The experiments were initiated during different stages of inflorescence development. This was designed in such a way because the severity of mirid damage has been related to their feeding on buds or flowers of olives or citrus (Barbagallo, 1970; Monaco, 1975). Cages without mirids were used as controls. The mirids used in these experiments were obtained from wild populations collected in olive orchards of the study area.

To reduce variability, one cage for each treatment and control were established on adjacent shoots from the same twig of a tree. Before the cages were fully enclosed any arthropod contaminants were removed. Each group of cages was placed in a different tree selected at random within the orchard. The orientation of each group was also randomly selected. The cages were placed at a height of 1-1.5 m at the periphery of each tree.

This study commenced on 29 March 2007. At this time the axis of the olive inflorescence was almost completely developed and flower buds were clearly separated [stages (e) and (f) of olive inflorescence, as described by Kitsaki *et al.*, 1999]. The two treatments were represented by three and six adults of *C. trivialis*, enclosed in each cage, and replicated 11 times. On 23 May 2007, about three weeks after the end of the flowering period, the number of buds, flowers and fruits abcissed in each cage was counted.

In 2008, *C. trivialis* and *A. italicum* were introduced in the cages on 7 March, 27 March and 18 April. On the first date, the olive inflorescences were completely covered by bracts and the axis of the inflorescence was visible at about 20% of the stages (a) and (b) according to Kitsaki *et al.* (1999), respectively. On 27 March, the axis of the inflorescence was well developed [stage (c) according to Kitsaki *et al.* (1999)]. On 18 April, separation among buds was clear [stages (e) and (f) of Kitsaki *et al.* (1999) classification of olive inflorescence development], whereas the flowering period had started since approximately 20% of the flowers were in bloom.

On 7 March 2008, 8 nymphs of *C. trivialis* or 8 nymphs of *A. italicum* were introduced to each cage. On

27 March and 18 April, eight adults of *C. trivialis* or *A. italicum* were placed in each cage. For all dates, each treatment was replicated six times, including the controls. The mirid densities used in the treatments were designed to be commensurate with relatively high wild population densities, as found in preliminary sampling of olive trees (Perdikis *et al.*, unpublished data).

The number of buds, flowers and fruits dropped, and the number of fruits on the shoots in each cage was recorded on 12 June 2008.

Statistical analyses

In the results of the experiments for each year, variation among treatments due to the placement of the cages on different trees were examined. To accomplish this aim, this effect of the "tree" factor on the number of damaged organs and the percentage of dropped fruiting organs was investigated using 1-way ANOVA.

The number of buds, flowers and fruits dropped in 2007 was analyzed using one-way ANOVA with the treatment as factor. In 2008, data on the percentage of the total damage induced by *C. trivialis* and *A. italicum* and the percentage of flowers in the dropped organs (buds, flowers and fruits) in each cage were analyzed using a two-way ANOVA. The factors used were the treatment (*C. trivialis*, *A. italicum* and control) and the initiation date of each experiment.

Taking into account the initial number of fruits in each cage, the final number of olives on the shoots in each cage was compared among treatments and dates, so that the damage potential of the phytophagous mirids on olive fruit setting could be determined. This analysis was performed using an ANCOVA with factor the treatment and covariate the initial number of fruits.

Prior to the analyses, data on numbers and percentages were logarithmically and arcsine transformed, respectively. Means were separated using Tukey HSD test. Analyzes were done using the statistical packages SPSS 13.0.1 (SPSS Inc., 2004) and JMP 7.0.1. (SAS Institute, 2007).

Results

In 2007 and 2008 the effect of the tree that treatments had been arranged on the number of damaged organs and the percentage of dropped fruiting organs was not significant (F = 0.45, d.f. = 10,84, p = 0.90 in 2007, F = 0.53, d.f. = 5,48, p = 0.75, in 2008, respectively). This outcome proves that effect of the tree where the different groups of cages had been established was not important and thus it was excluded from further analyses.

In 2007, the treatment effect was not significant on the number of dropped organs (F = 2.61, d.f. = 2,84, p = 0.076) (figure 1).

In 2008, the total number of dropped fruiting organs was not significantly different among treatments and the dates of cage establishment (F = 2.23, d.f. = 2,45, p = 0.11, F = 2.64, d.f. = 2,45, p = 0.08, respectively) (figure 2). A significantly higher total number of fruiting organs was only recorded in the *C. trivialis* treatment compared to the control, in the last date (figure 2).

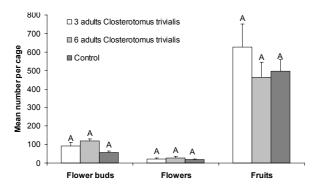


Figure 1. Mean number (± SE) of flower buds, flowers and fruits found dropped on 23 May 2007, in each cage where 3 or 6 adults of *C. trivialis* had been enclosed on 29 March on olive trees. Capital different letters indicated significant differences among treatments within each fruiting organ.

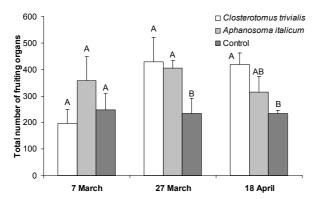


Figure 2. Mean number (± SE) of fruiting organs (flowers and fruits) recorded on 12 June 2008 in each cage where olive shoots had been enclosed together with nymphs or adults of *C. trivialis* or *A. italicum*. In each cage established on March 7, 8 nymphs and in each cage established on March 27 and April 18, 8 adults of each species had been introduced. Capital different letters indicated significant differences among treatments within each date.

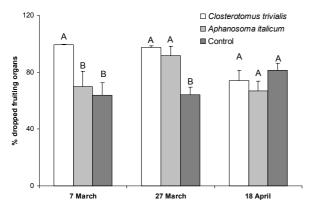


Figure 3. Mean percentage (± SE) of fruiting organs (flowers and fruits) that found dropped on 12 June 2008, in each cage where olive shoots had been enclosed together with nymphs or adults of *C. trivialis* or *A. italicum*. In each cage established on March 7, 8 nymphs and in each cage established on March 27 and April 18, 8 adults of each species had been introduced. Capital different letters indicated significant differences among treatments within each date.

The treatment showed a significant effect on the percentage of fruiting organs found dropped in the cages (F = 8.67, d.f. = 2,45, p < 0.001) (figure 3). This effect was due to the higher percentage observed in the cages where C. trivialis and A. italicum had been enclosed, in comparison to the control. The effect of date was marginally not significant (F = 3.06, d.f. = 2.45, p = 0.056). There was also a significant interaction between the factors treatment and date (F = 4.77, d.f. = 4.53, p < 0.01). In the first date, 7 March, the percentage of dropped organs in the cages with C. trivialis was significantly higher than that of A. italicum, with the latter being similar to the control (figure 3). In the second date, 27 March, in the cages where C. trivialis or A. italicum had been enclosed, the percentages of dropped organs were similar to each other and both were significantly higher than the control. However, on the last date, 18 April, no significant differences were recorded among the treatments. In the cages where C. trivialis had been enclosed, the percentages were $99 \pm 1\%$ in the first and $97 \pm 1\%$ in the second date, in average. In the cages with A. italicum, the highest average percentage was recorded in the second date $(91 \pm 6\%)$ (figure 3). In the first and the second date, in the control cages, the average percentages were 63 ± 8 and $64 \pm 6\%$, respectively.

A significant effect of the treatment was recorded on the percentage of flowers in the fruiting organs dropped in each cage (F = 5.81, d.f. = 2,45, p < 0.001). The effect of date was marginally insignificant (F = 2.44, d.f. = 2,45, p = 0.057), whereas the interaction between treatment and date was significant (F = 2.65, d.f. = 4,45, p = 0.045). This percentage was significantly higher in the case of C. trivialis than the control in the first and the second date of cage establishment. In the case of A. italicum, in the first and the second date of cage establishment, this percentage was similar to that of C. trivialis and the control treatments, despite being much higher than the control. In the third sampling this percentage was similar among treatments (figure 4).

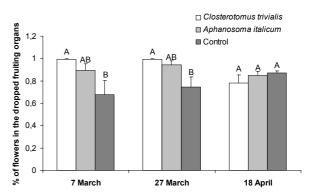


Figure 4. Mean percentage (± SE) of flowers in the fruiting organs (flowers and fruits) that found dropped on 12 June 2008, in each cage where olive shoots had been enclosed together with nymphs or adults of *C. trivialis* or *A. italicum*. In each cage established on March 7, 8 nymphs and in each cage established on March 27 and April 18, 8 adults of each species had been introduced. Capital different letters indicated significant differences among treatments within each date.

Data on the number of olives that finally developed on the shoots in each treatment are shown in figure 5. An analysis of covariance using the initial number of fruiting organs in the cages as a covariate, showed that the number of olive fruits in each cage was significantly different among the treatments in each date but not in the last one (F = 9.93, d.f. = 2,17, p < 0.01, F = 6.13, d.f. = 2,17,p < 0.01, but F = 1.29, d.f. = 2,17, p = 0.30, in each date, respectively) (figure 5). On the first date, 7 March, the number of fruits in the cages with C. trivialis was almost eliminated and significantly lower than that of A. italicum and the control, with those in the A. italicum cages being similar to the control (figure 5). In the second date, 27 March, in the cages with C. trivialis, again the number of fruits was almost eliminated and significantly lower number of fruits was found than in the control. In the first and the second date, the number of fruits in the cages with A. italicum, were similar to either the C. trivialis or the control. On the last date, 18 April, no significant differences were recorded among the treatments.

Discussion

The results demonstrated that *C. trivialis* is capable of inducing damage on the fruit setting of olive trees due to flower abortion. This damage can be extremely serious as almost complete flower abortion might occur (figures 2, 3 and 4). Thus, previous reports (Barbagallo, 1970; Monaco, 1975; Yamvrias, 1998; Gerakaki *et al.*, 2007) mentioning that damages might be of high level are supported by our results.

In addition to *C. trivialis*, another very little studied species, *A. italicum*, proved capable of reducing fruit setting, although it was not proved to be significant in comparison to the controls. Thus, *A. italicum* has potential as an olive pest, and its population densities might be taken into account in the development of control programs in olive orchards.

A major finding of our study was that damage severity produced depends on the phenological stage of the olive inflorescence at the time of infestation. In the case of *C. trivialis*, exposure of the inflorescence in the early stages of development could result in considerable reduction in fruit setting. This potential was not found to be significant in the early stages of inflorescence development or commencement of flowering (figures 1, 3 and 5). Specifically, in the experiments where the cages had been established on 7 and 27 March it was shown that *C. trivialis* can cause a high reduction on the number of fruits that remain on the trees (figures 3 and 5) and this most likely indicates a serious damage potential on the yield.

Therefore, aiming to optimize control strategies of *C. trivialis*, it is advisable to perform beating-tray samplings at regular intervals at critical periods, extending from the early emergence of flower buds until the period before flowering starts, as within this period olive trees are more susceptible.

In the study of Drosopoulos (1993) fruit setting of olive trees of the table olive variety "Amphissis" was compared between exposed and caged branches to naturally occurring populations of the pest in 3 olive orchards.

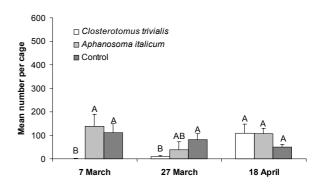


Figure 5. Mean number (± SE) of fruits on the shoots on the shoots that were recorded on 12 June 2008, in each cage where olive shoots had been enclosed together with nymphs or adults of *C. trivialis* or *A. italicum*. In each cage established on March 7, 8 nymphs and in each cage established on March 27 and April 18, 8 adults of each species had been introduced. Capital different letters indicated significant differences among treatments within each date.

According to the results, no difference was recorded in the fruit setting between the exposed or unexposed shoots and it was thus concluded that *C. trivialis* should not be considered as a pest of olives. However, the population densities of the pest were not recorded and the results are therefore not comparable to our study. Additionally, there could be different susceptibility between olive varieties (e.g., "Koroneiki" vs. "Amphissis") but this requires further investigation.

Generally, the infestation time has been shown to be a major factor in the damage severity caused by hemipteran species. Fruits of pistachios in California were more susceptible to damage by *Leptoglossus clypealis* L. (Hemiptera Coreidae) in June than in July (Bolkan *et al.*, 1984). A number of hemipteran species were proved to be successive in damaging pistachio fruits in a period lasting from April to September (Michailides *et al.*, 1987).

In the evaluation of the damage levels, the potential of the olive tree to compensate fruit drop due to the increase of the weight of the remained olives, has to be considered. This potential was proved to reach about 10% (Neuenschwander *et al.*, 1980). However, further studies focusing on the potential of recovery of olive drop at the early stages of fruit development could largely contribute to a more thorough understanding of the damage level. In addition, in our experiments, the experimental enclosure of insects may have increased rates of herbivory on shoots on the one hand but also reduced their longevity, due to factors, such as experimental conditions or restriction of access to wild host plants.

In conclusion, the current study showed that *C. trivialis* can cause serious reduction in fruit setting of olive trees. In comparison, *A. italicum* showed a comparatively lower damage potential. The damage severity depends on the phenological stage of the trees at the time of infestation. However, this finding is undoubtedly influenced by the pest densities used in our experiments, and we recommend that in future studies, damage severity

should be assessed against a wider range of population densities of both target species. We also believe that the potential compensatory responses of the olive trees, the effect of olive variety, and experimentation under more realistic conditions, need also to be investigated.

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