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Interactions between the citrus bud mite *Aceria sheldoni* (Acari: Eriophyidae) and the lemon host tree in a Mediterranean area

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ABSTRACT — This study evaluated the relation between the citrus bud mite [*Aceria sheldoni* (Ewing)] and lemon trees [(*Citrus limon* (L.) Burm] in two groves, over two seasons in Reggio Calabria (Italy). Random samples of shoots were taken every 10 days and the number of buds, blossoms, fruits, and leaves present on each shoot were counted. Blossoms and fruits were classified as healthy or deformed and the immature and adults stages of the citrus bud mite and associated predatory mites were counted. Observational data indicated that the citrus bud mite does not have a negative influence on the lemon harvest and that the interaction between its populations and the host plant is a mutual symbiosis in which mite populations use the buds as a resource and repay the host plant by positively regulating its productive system. In both lemon groves, three stigmaeid mites [*Zetzellia mali* (Ewing), *Z. graeciana* Gonzalez and *Agistemus collyerae* Gonzalez] were also collected and their density was compared with that of the *A. sheldoni*. Our study highlights the complexity of relations of the citrus bud mite and lemon trees, and stresses the importance of carefully determining the real influence of the citrus bud mite on host plant harvests.

KEYWORDS — Eriophyoidea; Stigmaeidae; predatory mite; host-plant relationship; citrus; ecology

INTRODUCTION

The citrus bud mite (CBM), *Aceria sheldoni* (Ewing), commonly infests lemon, *Citrus limon* (L.) Burm., and, in some regions of the world, also sweet orange *Citrus sinensis* Osbeck (var. Navels, Valencia) and other species and varieties of citrus (Schwartz and Riekert, 1967; Schwartz, 1972; Jeppson *et al*., 1975; Talhouk, 1975; Harty *et al*., 2004). The mite lives and reproduces inside the buds, where it feeds on embryonic tissue and on the growing or mature fruits. Mite infestations on flowering and wood buds result in morphological alterations of the shoots, leaves, buds, blossoms, and fruits (Boyce and Korsmeier, 1941; Ebeling, 1959; Jeppson *et al*., 1975). In the tissue of infested buds, there is an increase in phenol and a simultaneous decrease in auxin activity, with alterations in the activity of these compounds as well as that of ribonuclease (RNAase; Ishaaya and Sternlicht, 1969; Ishaaya and Sternlicht, 1971). Distortion of the fruits is significantly correlated with that of the blossoms from which they develop; most deformed blossoms and fruits fall and the degree of abscission increases significantly with the level of distortion. The increase in abscission of blossoms and fruits is likely to depend on a decrease in auxin activity and other biochemical anomalies in infested axil-

lary buds (Phillips and Walker, 1997). Although attack of the embryonic tissues of fruit buds results in the distortion of fruit, the mites do not attack the fruit once it has emerged (Walker *et al.*, 1992). It is believed that the presence of one to three mites per bud reduces the growth activity and production of the plant (Ishaaya and Sternlicht, 1969).

The control of *A. sheldoni* throughout the citrus-growing regions of the world is traditionally assigned to a variety of chemicals and a detailed account is given by McCoy (1996), Childers *et al.* (1996), and Vacante (2010). In Sicily (Italy), Vacante and Nucifora (1984) established that even infestation levels of 70 % of the buds do not cause any economic damage. Similarly, in California studies conducted by Hare *et al.* (1999) suggested that the mite is not a lemon pest as detrimental as it might first appear.

Therefore, the main aim of this study was to investigate interactions between CBM and its main host plant, lemon trees, through analysis of correlations between number of healthy or deformed tissues and mite densities. In addition, information on associated predatory mites along the period of study is presented. Possible economic effect of CBM on the lemon harvest in the studied area was discussed.

MATERIALS AND METHODS

Research was carried out from January 2004 to november 2005, in Reggio Calabria Province, Italy. Two lemon plots were examined each covering 1.5 ha; the first in Catona (latitude 38°.1038, longitude 15°.4059; 30 m above sea level) and the second in Salice (latitude 38°.1132, longitude 15°.3957; 50 m above sea level), with an age of 20 and 10 years, respectively. Both lemon groves were planted with the 'Monachello' lemon variety, with an average plant height of 2.50 m and a density of 500 plants/ha. Neither of the two plots have been treated with chemicals over the 5 years previous to the study, and no other treatments were carried out during the research period. Both orchards were fertilized in the spring of 2004 and 2005 with 2 kg/plant of N, P, and K-based trivalent inorganic fertilizer (20:10:10). The citrus orchard in Catona was trimmed in May 2004.

Samplings were carried out on a constant number of plants (14 at Catona and 16 at Salice), chosen at random. Four sectors were identified on each plant (North-East, South-East, South-West, North-West) and in each one a random destructive sampling of three shoots (12 per plant) was carried out every 10 days on the most recently emerged shoots with an average length of 15 – 16 cm. At Catona, 168 buds were sampled each time, and at Salice 192 buds.

Using a stereoscope, the samples were observed in the laboratory on the same day that they were picked and the number of buds, blossoms, fruits, and leaves on each individual shoot was counted. The blossoms and fruits were categorized as being healthy (not deformed) or deformed in terms of their morphology. The immature and adult stages of *A. sheldoni* and the other mites were counted and identified from four randomly selected buds and fruits on each shoot. Data analysis was carried out on the average number of buds, healthy and deformed blossoms, healthy and deformed fruits and/or shoot. The average number of mites found in buds and fruits were also investigated.

Relations between the variables examined were interpreted using Pearson correlation coefficients (r), verifying significance with the Fisher-Snedecor F Test. To support parametric correlation analysis and to compare the respective levels of significance, relations between the different variables were evaluated by nonparametric analysis with Kendall's correlation coefficient τ (tau)-*b*. Following verification of the conditions of validity of the individual correlations, relations between the variables studied were analyzed to inferring about host-relationship interactions between the citrus bud mite and lemon trees. Average lemon per plant production was also calculated in both experimental areas.

RESULTS AND DISCUSSION

In both areas, the CBM were consistently found in the buds and on the fruits (Fig. 1), as well as

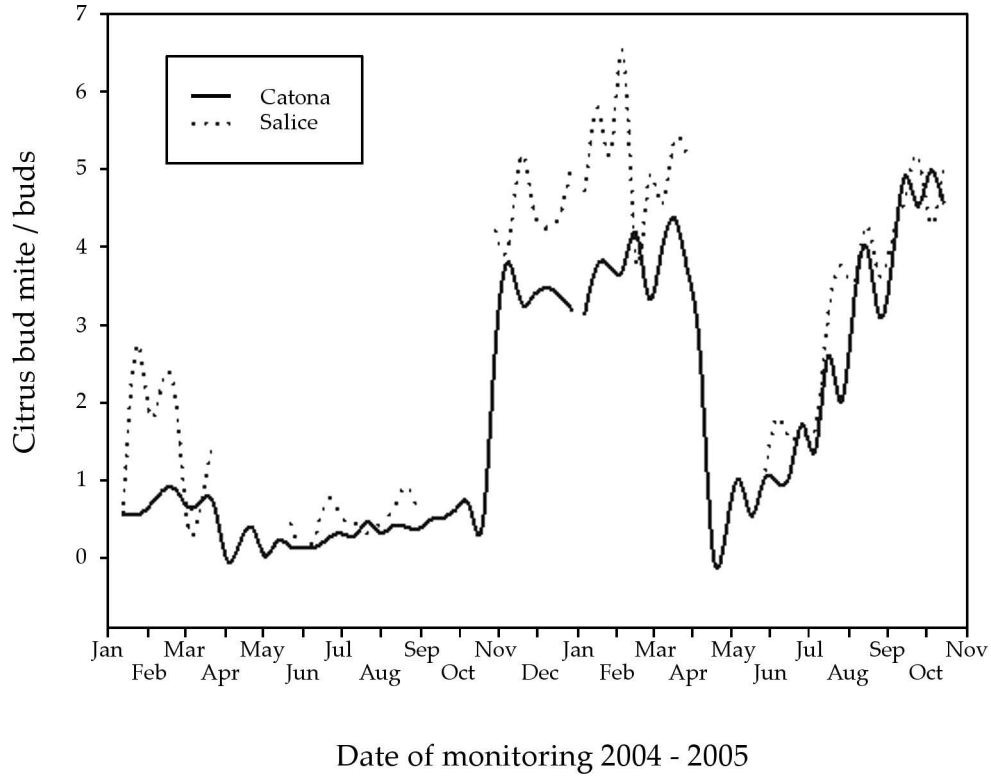


FIGURE 1: Number of CBMs per bud at two lemon orchards in Reggio Calabria Province, Italy, from January 2004 to November 2005.

some stigmaeid mites, including *Zetzellia mali* (Ewing) and two other species (*Z. graeciana* Gonzalez and *Agistemus collyerae* Gonzalez).

In both areas, the highest density of *A. sheldoni* in buds and on fruits occurred during vegetative standstill. The maximum densities of *A. sheldoni* recorded in winter at Catona on fruits and in buds was equal to 17.31 mites/fruit (12 January 2004) and 6.51 mites/bud (6 February 2005), whereas at Salice, 5.73 mites/fruit (16 February 2004) and 4.98 mites/bud (4 October 2005) were counted.

In terms of the density of mites per bud and the average number of buds per shoot, Table 1 shows that there is no uniform pattern or significant difference; thus, at the mite population levels recorded in this study, the mites do not have a negative influence on the number of buds on a shoot. In both areas, the correlations between the density of mites per bud and the average number of leaves and blossoms per shoot were negative. Generally, therefore, an increase in the number of blossoms per shoot was linked with a reduction in *A. sheldoni* density probably as a result of its migration throughout the tree crown in search of new buds and fruits to colonize.

The correlations between the mite density per bud and the average number of the fruits per shoot were different in the two areas, being positive at Catona and negative at Salice, although there was no significant difference. This could be connected with the different flushes of the crops.

The density of mites per bud and the average number of deformed blossoms per shoot were negatively correlated. Nevertheless, no significant correlation was found at Salice. Thus, the explanation given for the blossoms above might also be relevant

TABLE 1: Correlations between CBM densities and lemon host plant variables examined at two orchards in Reggio Calabria Province Italy from January 2004 to November 2005[a].

Variables	Catona					Salice				
	N	Pearson correlation		Kendall's tau-*b*		N	Pearson correlation		Kendall's tau-*b*	
		R	P	T	P		R	P	T	P
CBM/bud and buds/shoot	65	0.237	0.057	0.291	0.002	51	-0.250	0.076	-0.066	0.5
CBM/bud and leaves/shoot	65	-0.256	0.040	-0.130	0.126	51	-0.339	0.015	-0.195	0.05
CBM/bud and blossoms/shoot	65	-0.375	0.002	-0.365	<0.001	51	-0.179	0.209	-0.166	0.154
CBM/bud and fruits/shoot	65	0.141	0.263	0.029	0.738	51	-0.152	0.288	-0.050	0.613
CBM/bud and deformed blossoms/shoot	65	-0.333	0.007	-0.316	0.002	51	-0.179	0.209	-0.166	0.154
CBM/bud and deformed fruits/shoot	65	0.469	<0.001	0.414	<0.001	51	0.699	<0.001	0.605	<0.001
CBM/bud and CBM/fruit	59	0.672	<0.001	0.433	<0.001	50	0.452	0.001	0.529	<0.001
Healthy blossoms and fruits/shoot	65	-0.855	<0.001	-0.522	<0.001	62	-0.872	<0.001	-0.542	<0.001
Healthy and deformed blossoms/shoot	65	0.808	<0.001	0.771	<0.001	65	0.959	<0.001	0.958	<0.001
Deformed blossoms and fruits/shoot	65	-0.678	<0.001	-0.377	<0.001	62	-0.819	<0.001	-0.532	<0.001
Deformed blossoms and deformed fruits/shoot	65	-0.309	0.012	-0.345	0.001	62	-0.288	0.02	-0.323	0.02
Healthy and deformed fruits/shoot	65	0.344	0.005	0.213	0.015	62	0.192	0.135	0.164	0.131
CBM/bud and S/bud	65	0.799	<0.001	0.554	<0.001	51	0.803	<0.001	0.606	<0.001
CBM/fruit and S/fruit	55	0.845	<0.001	0.688	<0.001	57	0.617	<0.001	0.703	<0.001

[a]Abbreviations: CBM, Citrus bud mite; S, stigmaeids.

here. A highly significant positive correlation was found between the density of mites per bud and the average number of deformed fruits per shoot, confirming that damage to the fruits occurs in the bud tissue (Fig. 2 a, b).

The correlation between the healthy and deformed blossoms per shoot was highly positive. The deformed blossoms and fruits per shoot in both sites were marked by a highly negative correlation, related to the fact that the number of deformed blossoms was positively correlated with the number of healthy blossoms, and that the overall extent of the flowering was negatively correlated with that of setting. As in the case of blossoms, a positive correlation, although non-significant, was also observed for healthy and deformed fruits. The same reasoning as given above also explains the negative correlation between deformed blossoms and deformed fruit.

In addition, < 1 % of fruits were deformed at Catona, with a highest recorded value of 0.074 % recorded across both years. The average weight of fruits from October 2004 to March 2005 ranged from 182.59 g to 222.65 g and net production free from attack harvested in 2004 and 2005 totaled 90 and 70 kg/tree, respectively.

The percentage of deformed fruits throughout the period of study at Salice was constantly lower than 1 %, with a highest observed value of 0.12 %, whereas the average weight of the fruit was between 180 and 200 g. Net production free from attack harvested in 2004 and 2005 was 30 and 38 kg/tree, respectively.

The results found make it possible to create a hypothetical behaviour model of mite populations, which could help to interpret their ecological relation with the host plant. Such an understanding would be useful in defining the actual level of harm caused to the fruit and could guide adoption of technically supported control strategies. Given that the mite infestation of bud embryonic tissues (Ishaaya and Sternlicht, 1969) leads to damage of the resulting blossoms and fruits, data from the current study suggests that *A. sheldoni* density in a bud

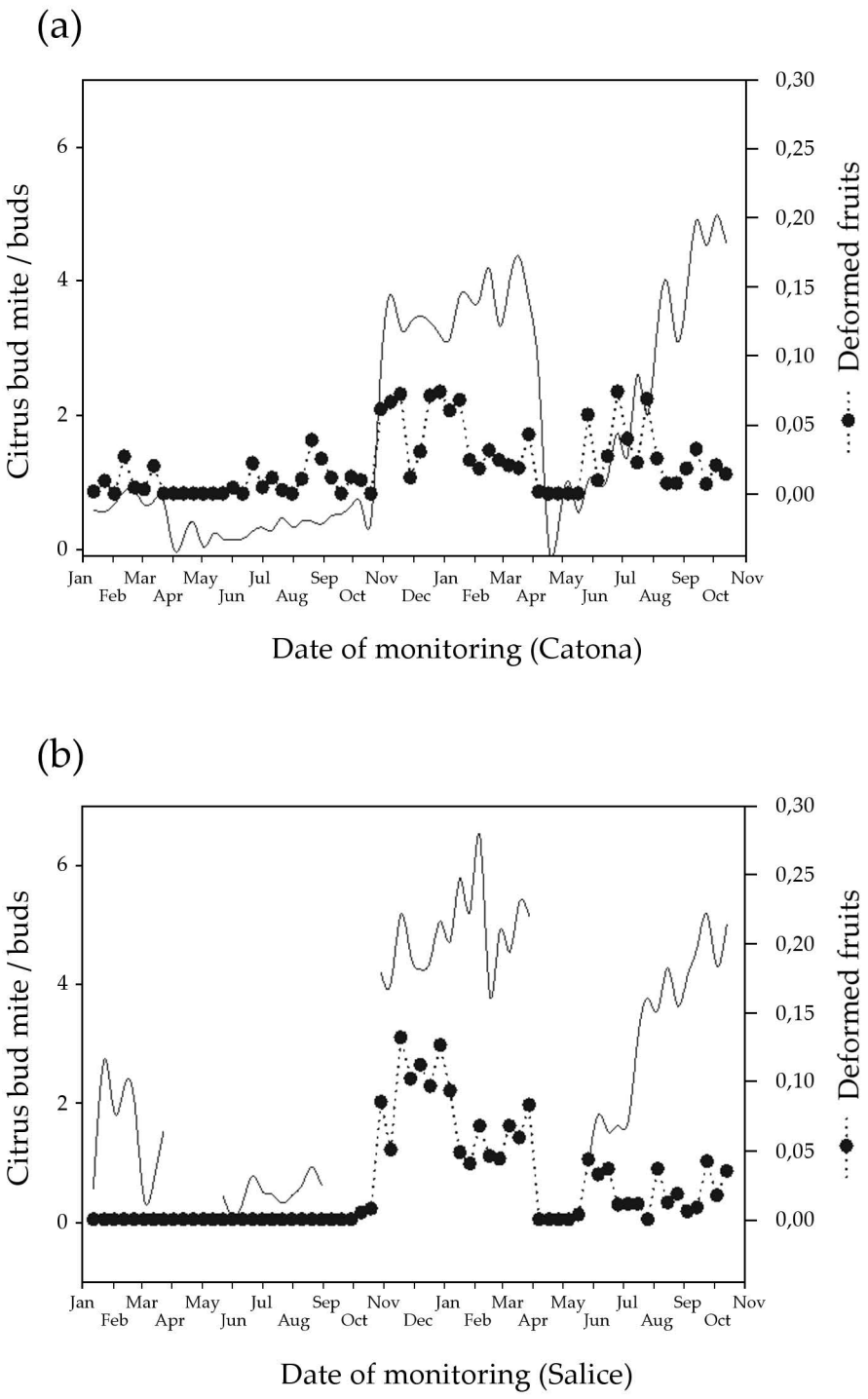


FIGURE 2: Relation between number of CBMs and the rate of deformed fruits in two lemon orchards in Reggio Calabria Province (a. Catona; b.Salice), Italy, from January 2004 to November 2005.

is a quantitative regulatory factor of flowering. The negative correlation found between blossoms and fruits confirms several typical aspects of the physiology of flowering and highlights that intense flowering does not constitute the necessary presupposition for an optimal setting. A high density of CBM does not result in an increase in deformed blossoms, as consolidated in plant protection practices. Therefore, each increase in the population density of this pest is not associated with an increase in the risk of damage and a consequent reduction in the density of healthy blossoms. The positive correlation found between healthy and deformed blossoms and between healthy and deformed fruits indicates that their quantitative relation depends on the action of the mites already inside the bud and that their degrees are linked to the extent of flowering. The mite acts by means of the deformation of blossoms and fruits, all of which fall except for a few deformed fruits that manage to develop on the trees and which constitute evidence of the mite infestation. Indirect confirmation of this interpretation can be found in citrus groves where an normal production of lemons is connected with a setting percentage of no higher than 7 % (Reed, 1919), and a maximum percentage of deformed blossoms of 25 %, similar to that recorded in the current study, represents an adaptive process that favour fructification. Taking into account that in both areas: i) the percentage of deformed lemons was < 1 % of the total; ii) that optimal average production can reach 50 000 kg/ha and that, over the past 3 years in the areas of highest value (Syracuse, Sicily); and that iii) lemons are sold at an average price of 0.35 Euro/kg, it can be estimated that the total product loss is < 500 kg, equal to 175 Euro/ha, a figure that is much lower than the cost of any treatment with petroleum oil (500 Euro/ha).

CBM densities in the buds and on fruits were positively correlated and relates to the behavior of mite populations, which develop first on the buds and then gradually move onto the fruits. However, the mite does not abandon buds on which the density is synchronous with that on the fruits. In a previous study on lemon in Sicily, the population densities of the mites recorded during spring on fruits during the vegetative growth period were modest compared with those observed on the same fruits during the coldest months (Vacante, 1986). The maximum densities of *A. sheldoni* were recorded in winter at each site. The fruit, similar to the bud, represents an overwintering site and the presence of mites on it does not cause any damage.

These results are in agreement with those observed by Jeppson *et al*. (1958), who reported a higher incidence of attack of the fruits than the buds, but contrasts with those observed by Walker *et al*. (1992). Nevertheless, although the adaptation of mite populations to buds can be explained by the utilization of a profitable resource, this is not the case in terms of the fruits. Indeed, the natural fall of the fruits from the tree, as well as their harvest, result in a reduction of the mite population and, thus, it would acts as a regulating mechanism of *A. sheldoni* density.

The natural enemies of the CBM includes some pathogens, such as the fungus *Hirsutella thompsoni* Fisher var. synematosa, (Searle, 1973; McCoy, 1981, 1996a,b; Sosa Gomez and Nasca, 1983), and various predatory mites. These include different stigmaeid mites, some cheyletid mites, and a few phytoseiid mites (Sternlicht, 1970; Searle, 1973; Vacante, 1986; Searle and Smith Meyer, 1998; Matioli *et al.*, 2002; Bonsignore and Vacante, 2012; Vacante, 2015 in press).

A positive correlation was found between the populations of stigmaeids (*Zetzellia mali, Z. graeciana*, and *Agistemus collyerae*) and the CBM populations in the buds and on the fruits in the current study. According to Santos (1982), when confronted with a high prey density, stigmaeids appear to exhibit a reduced predation rate, although these effects are not permanent. On the fruits, stigmaeids were observed to associate with tydeids (*Orthotydeus* spp., *Pronematus* sp.) and phytoseiids [*Amblyseius stipulatus* Athias Henriot and *A. degenerans* (Berlese)], and, although direct predation of *A. sheldoni* was not observed, their presence may make the system more complex, at least as regards the leaves and fruits. The results show that stigmaeids do not have a strong impact on prey populations and their population dynamics follow that of the CBM, but

is not resolutive. This limit was reported for the same species on citrus in Italy (Vacante 1986) and for other stigmaeids in South Africa (Searle, 1973; Searle and Smith Meyer, 1998) and Israel (Sternlicht, 1970).

Although the CBM develops on various citrus species, it usually prefers lemon because of its large buds, which offer greater protection [it also prefers some orange varieties ('Valencia' and 'Navel'), for the same reason]. Jeppson *et al*. (1975) also reported the relations between bud size in lemon and attack by *A. sheldoni*. The mite is common in coastal citrus-growing areas and is probably present in every lemon-growing area of the world that has an adequate relative humidity (RH) for its development. During the year, varying population peaks was recorded from one season to another depending on various ecological factors (growth of new vegetation, climatic trends, etc.) and the highest densities are observed in the buds during the coldest months, with the lowest densities occurring in spring-summer (Schwartz, 1975b; Vacante, 1986; Vacante *et al*., 2007). The population dynamics of *A. sheldoni* appear to be stable compared with those of other mites (Searle, 1978). As soon as populations of the CBM become exposed during new bud growth, they move on in search of new buds, spreading over the tree. The eggs hatch in 3 – 14 days and the length of a generation (egg to egg) is 12 – 33 days; hatching is optimal at 25 °C and 98 % RH but reduces at lower RH (35 % – 40 %). The threshold for embryonic development is at a lower RH (35 % – 40 %) (Sternlicht, 1970).

The results of the current study show that the role of the CBM on citrus plants is complex. The characteristic spatial scale imposed by the individual biology of the CBM requires further research to examine its effects on lemon trees without natural enemies.

To conclude, the results of the current study indicate that the action of the CBM does not negatively influence the productivity of lemon and that its relation with the host plant was characterized by mutual symbiosis, in which its populations use buds as a resource and help the host plant to regulate its productive system. This phenomena has not yet been reported for other eriophyoid mites. *Aculops pelekassi* (Keifer) and *Phyllocoptruta oleivora* (Ashmead), also present in Italy (de Lillo, 2004), do not have any regulatory effect. Further investigations are necessary to overcome the limitations that have influenced this research and to compare plants with and without CBM under controlled conditions to fully confirm the results obtained.

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