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Occurrence of *Hemerobius stigma* Stephens in pine bast scale (*Matsucoccus* spp.) populations: opportunistic predation or obligatory association?

Abstract - *Hemerobius stigma* Stephens (Neuroptera: Hemerobiidae) occurs naturally in coniferous forests of western and central Europe. Based on field and laboratory observations conducted mainly by European researchers, *H. stigma* has been accurately defined as predator of aphids (Hemiptera: Aphidoidea), often observed on spruce and fir trees. *Hemerobius* species were among several insect predators collected from epidemic populations of *Matsucoccus* (Hemiptera: Matsucoccidae). Collection of *H. stigma* from pine trees infested with *M. pini* Green in the 1970s and *M. feytaudi* Ducasse in the early 1990s suggests for the first time a close relationship between *H. stigma*, *Matsucoccus*, and pine. However, there was no data confirming that these *Hemerobius* spp. feed and successfully develop on *Matsucoccus*. The first evidence of interrelationship between *H. stigma* and *Matsucoccus* was obtained using pheromone traps in field studies conducted in maritime pine (*Pinus pinaster* Ait.) stands in central Portugal which show that *H. stigma* is highly attracted to sex pheromone of maritime pine bast scale *M. feytaudi*. It was also lured by the pheromone of *M. matsumurae* Kuwana, originating in the pine forest of the southern Asia, but not by the sex pheromone of *M. josephi* Bodenh., occurring in the East Mediterranean.
The frequent occurrence of *H. stigma* in aphid colonies, documented as suitable feed for the brown lacewing on the one hand and the strong specific kairomonal response of the lacewing to certain sex pheromone of *Matsucoccus* on the other, raise the question about the essence of the lacewing and the scale predator-prey relationship. To address this question, we tested the capability of *H. stigma* to prey and develop on *Matsucoccus*, using *M. josephi* and *Cinara palaestinensis* Hille Ris Lambers (Hemiptera: Lachnidae) as prey models. Further, we activated traps baited with the sex pheromone of *M. feytaudi* to compare the seasonal population trends of males of *M. feytaudi* with those of adult *H. stigma* and *Elatophilus crassicornis* Reuter (Heteroptera: Anthocoridae). The latter is a specific predator of *M. feytaudi* in Portugal, also attracted to the sex pheromone of both *M. feytaudi and M. matsumurae*. In the laboratory, both *M. josephi* and *C. palaestinensis* have served as appropriate food for *H. stigma*, allowing its complete development for successive generations and verifying its wide range of prey species among hemipteran taxa. However, the seasonal flight pattern of *H. stigma*, as obtained by trap captures was not correlated with that of the *M. feytaudi*

males, whereas a significant correlation was observed between the seasonal flight of *E. crassicornis* and that of males of *M. feytaudi*. A possible explanation to the interrelationship between *Hemerobius stigma* - *Matsucoccus feytaudi* - *pine* is presented and discussed.

Key words: scale insects, sex pheromone, kairomonal response, plurithropic interactions.

INTRODUCTION

Insect enemies of pine bast scales

Most groups of predators associated with Matsucoccidae (Hemiptera) are polyphagous and occur in different sylvan and agricultural ecosystems (the family is not attacked by parasitoids). Mendel *et al.* (1991) and references cited suggested that 18 species of *Elatophilus* (Hemiptera: Anthocoridae) feed in nature solely or primarily on a few species of pine bast scales, *Matsucoccus* spp. However, others believe that they also feed on aphids (Lattin & Stanton, 1993; Covassi et al., 1991). *Elatophilus* spp. colonize pine forests and *E. hebraicus* Péricart, a common monophagous predator of *M. josephi* Bodenh., occurs in brutia pine and Aleppo pine forests of the East Mediterranean (Mendel *et al.*, 1994). Information about *Elatophilus* spp. associated with North American *Matsucoccus* spp. was outlined by Mendel *et al.* (1991) and Lattin & Stanton (1993). Very little experimental work has been done on specialist anthocorid predators. Generally, a close association with a particular insect and a plant species among this group is relatively rare. Mendel *et al.* (1991) drew attention to the fact that the tritrophic system *Elatophilus* - *Matsucoccus* - *Pinus* is particularly interesting as an example of possible co-evolution of the predator - prey - plant complex. Brown lacewings, *Hemerobius* spp. (Neuroptera: Hemerobiidae), are among several insect enemy guilds associated with pine bast scales, *Matsucoccus* spp. (Bean & Godwin, 1955; Covassi *et al.*, 1991; Mendel *et al.*, 1991; Ming *et al.*, 1983; Siewniak, 1976). Many brown lacewings are naturally limited in habitat range (Monserrat & Marin, 1996). Several *Hemerobius* species are associated with conifers, while others are restricted to broadleaves (Aspock *et al.* 1980). Species found predominantly on conifers exhibit prey specificity (New, 1975). They are considered by many as predators of aphids (e.g., Kevan & Klimaszewski, 1987). Typically, *H. stigma* Stephens and *H. nitidulus* Fabricius collected from spruce, fir, and pine in England, have been associated primarily with woolly aphids (Adelgidae) (Laidlaw, 1936; Wilson, 1938). *Hemerobius* spp. were among several insect predators collected from epidemic populations of *Matsucoccus*. Collection of *H. stigma* from pine trees infested with *M. pini* Green in the 1970s and *M. feytaudi* Ducasse in the early 1990s was the first evidence of a close relationship between *H. stigma*, *Matsucoccus*, and pine (Mendel *et al.*, 1997). However, there is no data demonstrating that the lacewing feeds and successfully develops on *Matsucoccus*. Species of *Elatophilus* and

Hemerobius are multivoltine. *Elatophilus* spp. oviposit in needles of any pine species but not of other plants (Covassi *et al.*, 1991; Mendel *et al.*, 1995a). *Hemerobius* spp. attach the eggs to the needles (Aspöck *et al.*, 1980). Both larvae and adults of the two genera are predators. The natural distribution of *H. stigma* consists of the pine forests of Western and Central Europe and the West Mediterranean (Aspöck *et al.*, 1980). It was released in Eastern Canada between 1935 and 1938 (together with *H. nitidulus*) for biological control of the balsam woolly aphid (McGugan & Coppel, 1962). Although not recovered in the first years after its introduction, *H. stigma* was reported from Canada and the US in the following years (Garland, 1978a; Miller & Lambdin, 1984a). Its native occurrence in North America, however, is uncertain because of the difficulties to discriminate between *H. stigma* and the Nearctic species *H. stigmaterus* Fitch (Garland, 1978b; Miller & Lambdin, 1984b). The different kairomonal attraction of *H. stigma* and *H. stigmaterus* to the sex pheromones of various pine bast scales favours the separation into two species.

Sex pheromones as long-range kairomones of predators

There are very few reports of insect sex pheromones acting as kairomones attracting predators. Recently, Boo *et al.* (1998) showed that aphid sex pheromone components do attract the polyphagous predator *Chrysopa cognata* McLachlan. We have shown that *E. hebraicus*, a predator of *M. josephi*, is strongly attracted by the sex pheromone of the scale (Fig. 1). The kairomonal activity is not affected by the inactive stereoisomers in the racemic mixture (Zegelman *et al.* 1993; Mendel *et al.* 1995b). This was the first unequivocal demonstration of kairomonal attraction of a predator to a scale insect sex pheromone. The pheromones of *M. matsumurae* Kuwana and *M. feytaudi* (Fig. 1) are also potent kairomones of *E. hebraicus* despite the fact that these two pine bast scales do not occur in its natural range. As expected, these two pheromones do not attract the males of *M. josephi* (Dunkelblum *et al.*, 1996). All

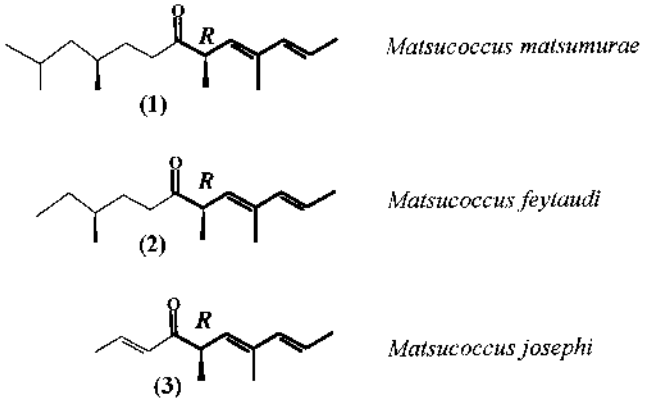

Fig. 1 - *Matsucoccus* sex pheromones.

three *Matsucoccus* pheromones have the same chiral ketodiene moiety, within the same absolute configuration but differ in the remainder of the molecule (Dunkelblum *et al.* 2000, Fig. 1).

Preliminary results from our field tests in Portugal (Mendel *et al.*, 1997) and the US indicate that *Matsucoccus* pheromones attract other *Elatophilus* spp. (e.g., *E. crassicornis* Reuter) and also *Hemerobius* spp. (e.g., *H. stigma, H. stigmaterus*) pointing to a general mode of kairomonal response. Our recent findings that *H. stigma* and *H. stigmaterus* are attracted by *Matsucoccus* pheromones (Table 1) suggest that, like *Elatophilus* spp., members of the genus *Hemerobius* are also closely associated with pine bast scales.

The available data raises the question about the actual food range of *H. stigma*

Table 1 - Intensity of the attraction (strong - +++, weak +, none - 0) of Hemerobius *spp. (Neuroptera: Hemerobiidae) to the sex pheromones of* Matsucoccus *spp. (Hemiptera: Matsucoccidae).*

Matsucoccus species whose sex pheromone was tested	Studied *Hemerobius* species	
	H. stigma	*H. stigmaterus*
M. feytaudi	+++	0
M. matsumurae	+++	0
M. josephi	+	+++

and its possible predator-prey relationship with *Matsucoccus* spp. On the one hand, the brown lacewing is reported to feed on conifer aphids. On the other, the strong kairomonal attraction to the sex pheromones of *Matsucoccus* spp. suggests a close relationship of *H. stigma* with pine bast scales.

In order to address this question, we tested the capability of *H. stigma* to prey and develop on *Matsucoccus* and other Hemiptera species, using as prey models three allopatric species, i.e., *M. josephi, Cinara palaestinensis* and *Cinara laportei* Remaudière (Hemiptera: Lachninae). In additional, we activated also traps baited with the sex pheromone of *M. feytaudi* in order to compare the seasonal population trends of males of *M. feytaudi* with those of adults of *H. stigma* and *E. crassicornis*. The latter species was used as a model for comparison with *H. stigma*.

EXPERIMENTS AND RESULTS

Experiment 1. Feeding range of *Hemerobius stigma*

The ability of *H. stigma* to prey and develop on different Hemiptera was tested in the laboratory using *M. josephi, C. palaestinensis* and *C. laportei*. *H. stigma* completed several generations on all three tested prey species. The experiment showed

that the brown lacewing is able to feed and develop on *M. josephi* and both *Cinara* spp. The faster rate of development was observed with *M. josephi* as prey (Table 2). Present findings, together with other documented information (Table 3), suggest that the prey range of *H. stigma* includes species of Adelgidae, Aphididae and Matsucoccidae.

Table 2 - Rate of development (24-26^{0}C) of Hemerobius stigma.

Prey	Generation time (days)
Matsucoccus josephi	28-30
Cinara palaestinensis	35-37

Table 3 - Prey range of Hemerobius stigma.

Family/Subfamily	Species	Reference
Adelgidae	*Adelges viridis*	Laidlaw, 1936
	Adelges cooleyi	Laidlaw, 1936
	Adelges piceae	Garland, 1978b
	Pineus strobi	Wilson, 1938
Aphididae		
- Lachninae	*Lachnus pini*	Laidlaw, 1936
	Cinara strobi	Miller & Lambdin, 1985
	Cinara palaestinensis	(lab rearing) present study
	Cinara laportei	(lab rearing) present study
- Aphidinae	*Acyrthosiphon pisum*	(lab rearing) Garland, 1980
Matsucoccidae	*Matsucoccus josephi*	(lab rearing, present study)
	M. feytaudi (?)	Covassi *et al.*, 1991

Experiment 2. Seasonal flight pattern of Matsucoccus feytaudi males, and the predators *Hemerobius stigma* and *Elatophilus crassicornis*.

The fact of *H. stigma* and *E. crassicornis* demonstrate a kairomonal response to the sexual pheromone of the maritime pine bast scale, *M. feytaudi*, leads to the hypothesis that both predators have a close relationship with *M. feytaudi*. If this assumption is correct, a dependence relationship between the dynamics of the predators and that of the bast scale is expected. To test it, we examined the seasonal flight pattern of the three species based on the captures obtained in pheromone traps.

The seasonal flight pattern of males of *M. feytaudi* and that of *H. stigma* and *E. crassicornis* was studied in two maritime pine stands, 30 and 40-50 years old, in Portugal, i.e., Sintra (38°47'N, 9°25'W) and Apostiça (38°32'N, 9°8'W). Four 15 cm x 15 cm delta traps were activated in each stand with 400 mg of racemic mixture of the sex pheromone of *M. feytaudi*. The capture was assessed every other week, from February 1998 to February 1999.

The seasonal flight pattern observed (Fig. 2) was studied by means of time series cross correlation analyses (SPSS 10.0). A significant correlation was found between the seasonal flight trends of *M. feytaudi* and *E. crassicornis*, with a time lag of six months at Apostiça stand and a time lag of one and six months at Sintra (Fig. 3). Differences between the two sites may be related to differences on the flight pattern of *M. feytaudi* (Fig. 2). However, no significant correlation was found in both sites between seasonal trends of *M. feytaudi* and *H. stigma* (Fig. 4).

Experiment 3. Occurrence of *Hemerobius stigma* and *Elatophilus crassicornis* in different pine species in Portugal.

To strengthen our finding concerning the different flight pattern relationships between *E. crassicornis / H. stigma* and *M. feytaudi,* demonstrated in experiment 2, we activated traps with racemic mixture of the sex pheromone of *M. feytaudi* in different pine species located in several regions of Portugal, during June, July and August, when adults of both predators are active (Fig. 2). In the Island of Madeira, traps were activated during June only.

Elatophilus crassicornis occurred only in stands of *P. pinaster* Ait., while *H. stigma* was present in all pine species and in all locations (Table 4). The absence of *Elatophilus* in Madeira may be explained by the absence of its single prey guild *Matsucoccus* spp.

DISCUSSION

The strong kairomonal response of *H. stigma* to the sex pheromone of *M. feytaudi* implies a close relationship between the lacewing and the bast scale. On the other hand, we found *H. stigma* feeding and reproducing on other prey species. It is suggested that pine aphids are most probably a primary prey of *H. stigma* wherever *M. feytaudi* is absent (justifying, for example, the abundance of *H. stigma* in the island of Madeira where *Matsucoccus* is absent) or occurs in a very low density. This assumption is also supported by the fact that there is no obvious seasonal population relationship between *H. stigma* and *M. feytaudi* in the areas studied in the Portugal

Table 4 - Occurrence of Hemerobius stigma *and* Elatophilus crassicornis *as determined by their presence (+) or absence (0) in pheromone baited traps exposed in stands planted with several pine species in few areas in Portugal.*

Location	Geographical coordinates	*Pinus* species	*H. stigma*	*E. crassicornis*
Mainland Portugal				
Leiria	39°40´N, 8°55´W	*P. pinaster*	+	+
Península de Setúbal	38°32´N, 9°8'W	*P. pinaster*	+	+
Silves	37°11´N, 8°22'W	*P. pinaster*	+	+
Caparica	38°41´N, 9°11'W	*P. pinea*	+	0
Lousã	40°8´N, 8°3'W	*P. nigra*	+	0
Trás-os-Montes	41°50´N, 6°45´W	*P. sylvestris*	+	0
Madeira Island	32°45´N,16°52'W	*P. pinaster*	+	0

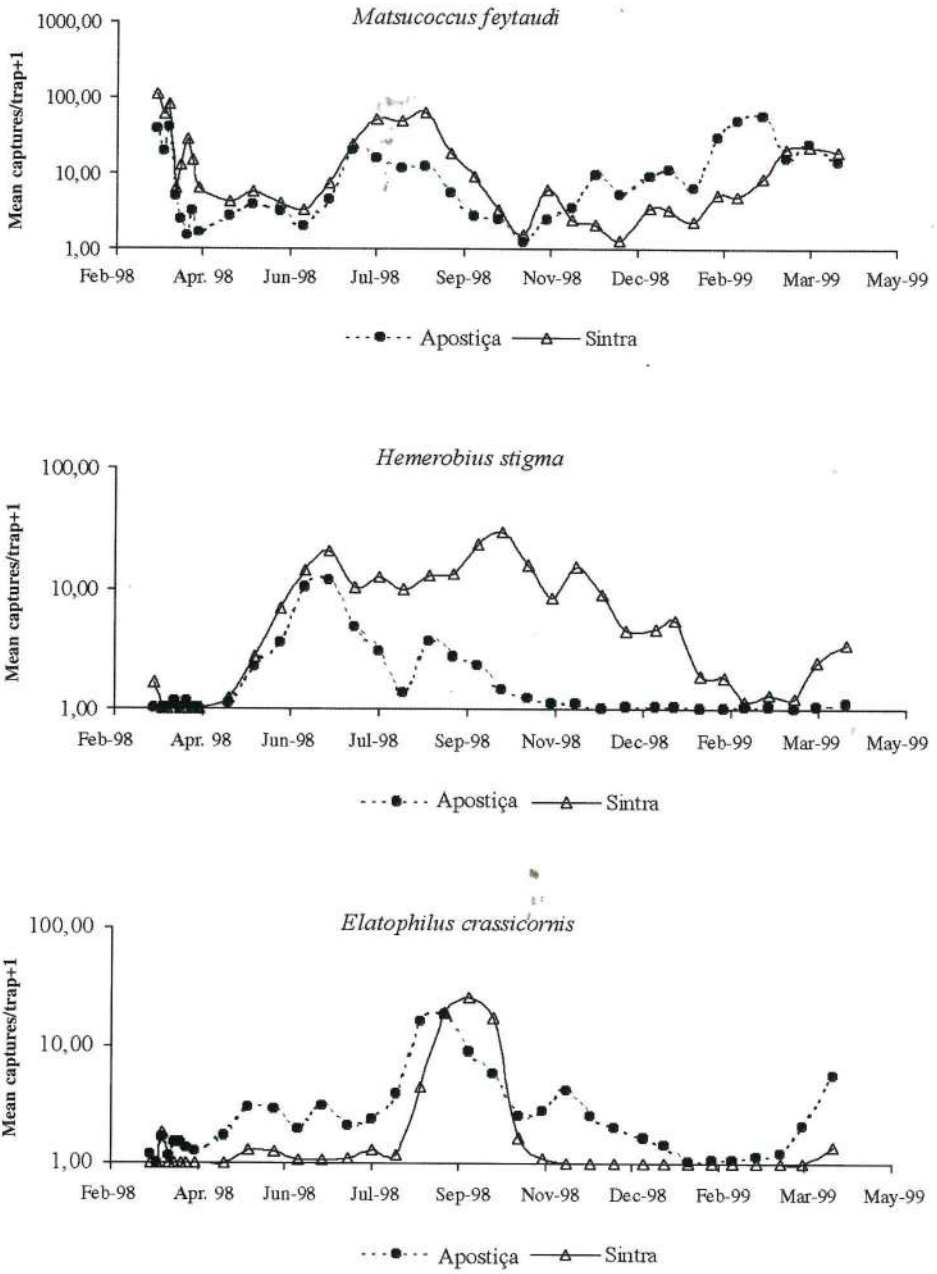

Fig.2 - Seasonal flight pattern of *Matsucoccus feytaudi, Hemerobius stigma* and *Elatophilus crassicornis* observed in two maritime pine stands in Portugal.

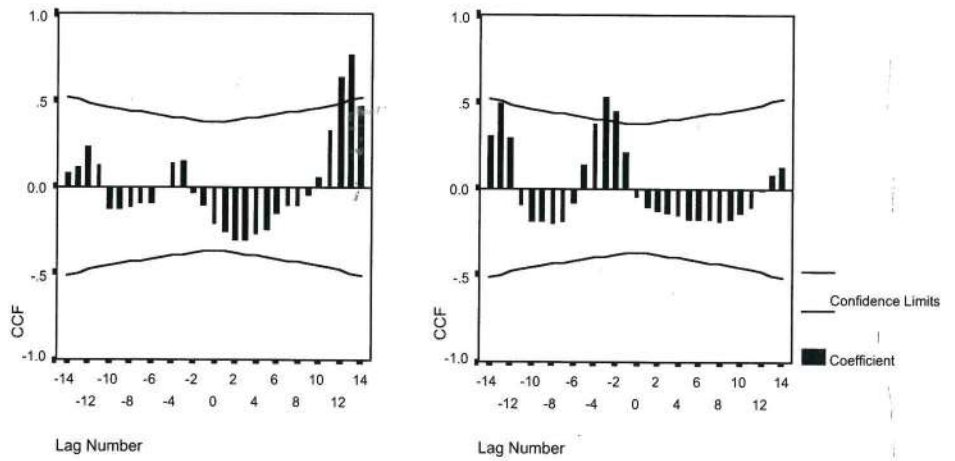

Fig. 3 - Time autocorrelation coefficients between catches of *Matsucoccus feytaudi* males and adults *Elatophilus crassicornis* in two maritime pine stands, in Portugal (Apostiça left and Sintra right); a lag time equals 2 weeks.

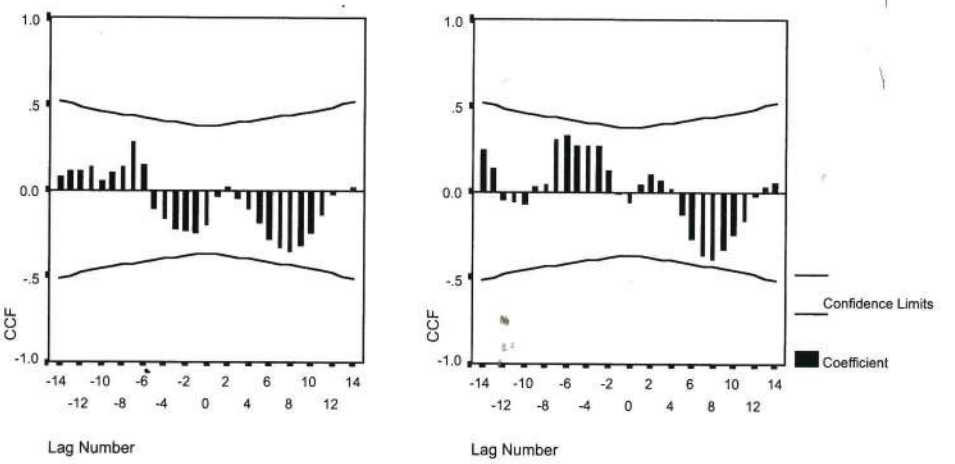

Fig. 4 - Time autocorrelation coefficients between catches of *Matsucoccus feytaudi* males and adults *Hemerobius stigma* in two maritime pine stands, in Portugal (Apostiça left and Sintra right). a lag time equals 2 weeks.

mainland. This puzzling information further emphasizes the enigma regarding the behavioural attractiveness of *H. stigma* to the sex pheromone of the maritime pine bast scale.

Hemerobius stigma displayed a specific kairomonal response to the sex pheromone of *M. feytaudi* and to that of *M. matsumurae,* but not to the sex pheromone of *M.*

josephi. *Hemerobius stigmaterus,* indigenous to Western USA, is attracted to sex pheromone of *M. josephi,* but not to the sex pheromone of the two former *Matsucoccus* species (unpublished data). These data further supports the idea of coevolution between species of both genera. The lack of close relationships between predator and the maritime pine bast scale leads us to assume that *H. stigma* may have coevolved with other *Matsucoccus* species, i.e., *M. pini,* whose sex pheromones have not been identified as yet. The large natural distribution of *H. stigma*, which primarily overlaps the natural distribution of *M. pini* (Aspöck et al., 1980) and the host plants, *Pinus sylvestris* L. and *P. nigra*, is consistent with the hypothesis.

It is suggested the association of *Hemerobius* species with *Matsucoccus* first occurred in northern Asia. *Hemerobius stigma* (or its ancestors) could spread together with *P. sylvestris* and *M. pini* (or their ancestors). *Pinus sylvestris* is widespread in Eurasia and has been isolated from other pine of the *Sylvestris* subsection in eastern Asia and North America since the Tertiary Period (Kremenetski et al., 1998). That may explain the wide distribution of *H. stigma* in Europe (Aspöck et al., 1980). With the scarcity of *Matsucoccus* population, *H. stigma,* as other *Hemerobius* species associated with pine, may have evolved feeding on other Hemiptera colonizing pine, i.e., Adelgidae and Aphididae, which occur in high densities compared with members of the Matsucoccidae. The occurrence of high *Matsucoccus* populations are recent events and often resulting from introductions by humans into areas comprising susceptible genotypes. We suggest that the response to the sex pheromone of *Matsucoccus* has remained as an ancient characteristic whose current function is more linked to the ability of *H. stigma* to find host trees or habitats for the guilds of its prey, rather than a specific prey. Similar situations are reported in the literature for coccinellids and their aphid preys. For example, the species *Anatis ocellata* (L.) is attracted to the aphid habitat by odours from pine needles rather than from the aphids on the pine (Kestaen, 1969). Adults of *Coccinella septempunctata* L., rather than responding to aphid honeydew or odours produced by the aphids themselves, use the pheromone trails of aphid-tending ants as a means of finding its prey (Greany & Hagen, 1981).

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