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Natural enemies associated with *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae) in the state of São Paulo, Brazil

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Abstract

The pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae), occurs widely in Brazil, causing damage to some crops. However, information on beneficial insects acting to control populations of this pest in Brazil is practically non-existent. A survey was conducted in the state of São Paulo. The insect associates of *M. hirsutus* recorded on *Hibiscus rosa-sinensis* L. (Malvales: Malvaceae) were the parasitoid *Gyranoidea indica* Shafee, Alam & Agarwal (Hymenoptera: Encyrtidae) and the predators *Cycloneda sanguinea* (L.), *Cryptolaemus montrouzieri* Mulsant, *Chilocorus nigrita* (F.), *Exoplectra* sp., *Harmonia axyridis* (Pallas), *Tenuisvalvae notata* (Mulsant) (Coleoptera: Coccinellidae), and *Ceraeochrysa* sp. (Neuroptera: Chrysopidae). *Cryptolaemus montrouzieri* was the most commonly collected species on populations of *M. hirsutus* infesting plants of *H. rosa-sinensis*. *Chilocorus nigrita*, *Exoplectra* sp., *H. axyridis*, *T. notata*, and *Ceraeochrysa* sp. were associated for the first time with *M. hirsutus*, and *G. indica* was recorded for the first time in Brazil.

Key Words: invasive species; geographical distribution; biological control; parasitoid; predator

Resumo

A cochonilha rosada do hibisco, *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae), atualmente difundida de Norte a Sul do Brasil, vem causando danos a algumas plantas cultivadas. Entretanto informações sobre insetos atuando sobre o controle das populações dessa praga no país são praticamente inexistentes. Um levantamento foi realizado no Estado de São Paulo, e os inimigos naturais associados à *M. hirsutus* sobre *Hibiscus rosa-sinensis* L. (Malvales: Malvaceae) foram o parasitoide *Gyranoidea indica* Shafee, Alam & Agarwal (Hymenoptera: Encyrtidae) e os predadores *Cycloneda sanguinea* (L.), *Cryptolaemus montrouzieri* Mulsant, *Chilocorus nigrita* (F.), *Exoplectra* sp., *Harmonia axyridis* (Pallas), *Tenuisvalvae notata* (Mulsant) (Coleoptera: Coccinellidae), e *Ceraeochrysa* sp. (Neuroptera: Chrysopidae). *Cryptolaemus montrouzieri* foi a espécie mais comumente coletada sobre populações de *M. hirsutus* infestando plantas de *H. rosa-sinensis*. *Chilocorus nigrita*, *Exoplectra* sp., *H. axyridis*, *T. notata* e *Ceraeochrysa* sp. foram registradas pela primeira vez associadas com *M. hirsutus* e *G. indica* é registrada pela primeira vez no Brasil.

Palavras Chave: espécie invasora; distribuição geográfica; controle biológico, parasitoides; predadores

The pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae), has caused serious damage to many crops in tropical and subtropical regions around the world. *Maconellicoccus hirsutus* has been associated with more than 200 genera of host plants from 76 plant families (Ben-Dov et al. 2015). In Brazil, *M. hirsutus* was first recorded in 2010 on a plant of *Hibiscus rosa-sinensis* L. (Malvales: Malvaceae) in the state of Roraima (Marsaro Júnior et al. 2013) and subsequently in the states of Espírito Santo (Culik et al. 2013), Mato Grosso (Morais et al. 2015), Alagoas (Broglia et al. 2015), Bahia (CEPLAC/CEPEC 2014), and Santa Catarina (Alexandre et al. 2014), totaling a list of 19 host plants for these states. *Maconellicoccus hirsutus* is already causing damage to crops such as cacao (*Theobroma cacao* L.; Malvales: Malvaceae) in Bahia and Espírito Santo (CEPLAC/CEPEC 2014) and soursop (*Annona muricata* L.; Magnoliales: Annonaceae),

star fruit (*Averrhoa carambola* L.; Oxalidales: Oxalidaceae), barbados cherry (*Malpighia glabra* L.; Malpighiales: Malpighiaceae), and cupaçu tree (*Theobroma grandiflorum* [Willd. ex Sprengel] Schumann; Malvales: Malvaceae) in Alagoas (Broglia et al. 2015).

In São Paulo State, *M. hirsutus* was first recorded on *H. rosa-sinensis* in an urban area of São Carlos in 2012 (Morais et al. 2015). Between 2013 and early 2014, Peronti & Martinelli (2014) recorded this pest in 11 municipalities of the state, infesting *Erythrina* spp. (Fabales: Fabaceae), *Ficus pumila* L. (Rosales: Moraceae), and *Eugenia uniflora* L. (Myrtales: Myrtaceae), but mainly *Hibiscus* species in urban areas and some teak, *Tectona grandis* L.f. (Lamiales: Lamiaceae), plantations in the northwest region of the state.

Because of its wide host range and its rapid geographic expansion, not only to agricultural land but also to home gardens and forest areas,

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biological control appeared as the most suitable method to manage populations of *M. hirsutus* (Sagarra & Peterkin 1999). Additionally, chemical management is hindered by (1) the waxy deposit on the mealybug bodies, which reduces penetration of the chemical solution (Kairo et al. 2000), and (2) the cryptic behavior of this insect which congregates in protected parts of the plants (Kairo et al. 2000; Chong 2009).

Among the 39 parasitoids and 42 predators of *M. hirsutus* known worldwide (Chong et al. 2015), 27 species are known to the Neotropical region (Culik et al. 2013; Chong et al. 2015). The mealybug destroyer, *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae), and the parasitoids *Anagyrus kamali* Moursi, *Gyranusoidea indica* Shafee, Alam & Agarwal (Hymenoptera: Encyrtidae) are used in large-scale biological control programs against *M. hirsutus* in many parts of the world (Culik et al. 2013; Chong et al. 2015).

In Brazil, only *A. kamali* was previously recorded as a natural enemy of *M. hirsutus*. It emerged from specimens collected on *H. rosa-sinensis* and *Glycine max* (L.) Merr. (Fabales: Fabaceae) in Boa Vista in the state of Roraima, and was probably introduced together with *M. hirsutus* in Brazil (Marsaro Júnior et al. 2013). The objective of this study was to survey species of natural enemies associated with *M. hirsutus* in the state of São Paulo, checking for pest–natural enemy associations known in other countries, and to document new associations.

Materials and Methods

A survey of the natural enemies associated with *M. hirsutus* on *H. rosa-sinensis* was conducted in urban areas of 3 municipalities of São Paulo State. Occasional samplings were carried out in São Carlos (22.0152778°S, 47.8911111°W) and Campinas (22.8554167°S, 47.0569722°W) in Jul 2012 and Jan 2013, respectively. Afterwards, monthly samplings were carried out in Jaboticabal (21.2552778°S, 48.3225000°W) between Mar and Jul 2014.

Infested plant parts were inspected visually for adult predators and, if present, were collected with an aspirator. Coccinellid and chrysopid larvae were placed in Petri dishes covered with muslin netting and fed with nymphs and adults of *M. hirsutus*. The adult insects that emerged were preserved by pinning. To obtain parasitoids of *M. hirsutus*, darkened specimens that showed a decrease in mobility were placed in glass tubes with a cotton lid. The emerging adult parasitoids were subsequently preserved in 100% ethanol.

The mealybugs previously preserved in 75% ethanol were mounted on microscopic slides following the method described by Gullan (1984)

and identified according to morphological characteristics of the adult female as described by Miller (1999) and Miller et al. (2011). The hymenopterous parasitoids were identified using the keys by Noyes & Hayat (1994), the coccinellids were identified according to the keys by Gordon (1985), Araujo-Siqueira & Almeida (2006), and Costa et al. (2008), and the chrysopids were identified by using the keys by Brooks & Barnard (1990).

Voucher specimens were deposited in the following institutions: mealybugs and chrysopids at the Department of Plant Protection, Universidade Estadual Paulista [FCAV/UNESP]; parasitoids at the Department of Ecology and Evolutionary Biology at Universidade Federal de São Carlos [UFSCar]; coccinellids at the Coleção Entomológica Pe. J.S. Moure, Department of Zoology, Universidade Federal do Paraná [UFPR].

Results

All pseudococcids collected were confirmed to be *M. hirsutus*. Natural enemies recovered were the parasitoid *G. indica* (Hymenoptera: Encyrtidae) and the predators *Cycloneda sanguinea* (L.), *C. montrouzieri*, *Chilocorus nigrita* (F.), *Exoplectra* sp., *Harmonia axyridis* (Pallas), and *Tenuisvalvae notata* (Mulsant) (Coleoptera: Coccinellidae) and *Ceraeochrysa* sp. (Neuroptera: Chrysopidae) (Table 1).

Cryptolaemus montrouzieri (larvae and adults) was the most commonly observed predator associated with colonies of *M. hirsutus* in the state of São Paulo, mainly on plants heavily infested by this pest. The remaining species of predators were generally in the adult stage, and the parasitoid *G. indica* was sporadically collected. Larval stages of the coccinellid *C. nigrita* and the green lacewing *Ceraeochrysa* sp. were also observed on a hedge of hibiscus plants in Jaboticabal. *Chilocorus nigrita*, *Exoplectra* sp., *H. axyridis*, *T. notata*, and *Ceraeochrysa* sp. were recorded for the first time associated with *M. hirsutus*, and *G. indica* was recorded for the first time in Brazil (Table 1).

Discussion

COLEOPTERA

Chilocorus nigrita is indigenous to the Indian sub-continent and Indo-China and has been introduced into many regions for biological control purposes (Ponsonby 2009). Currently, *C. nigrita* is distributed

Table 1. Parasitoids and predators of *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae) from 3 municipalities in the state of São Paulo, Brazil.

Natural enemies	Origin	Material studied
Coleoptera: Coccinellidae		
<i>Chilocorus nigrita</i> *	Oriental	10-I-2013, Campinas, SP, Marsaro Júnior AL col. (1 ex.); VI-2014, Jaboticabal, SP, Alexandrino JG col. (6 ex.).
<i>Cryptolaemus montrouzieri</i>	Australasian	6-XII-2012, São Carlos, SP, Peronti ALBG col. (8 ex.); 10-I-2013, Campinas, SP, Marsaro Júnior AL col. (4 ex.); III-2014, IV-2014, V-2014, Jaboticabal, SP, Alexandrino JG col. (20 ex.)
<i>Cycloneda sanguinea</i>	Neotropical	6-XII-2012, São Carlos, SP, Peronti ALBG col. (1 ex.)
<i>Exoplectra</i> sp.*	Neotropical	10-I-2013, Campinas, SP, Marsaro Júnior AL col. (1 ex.)
<i>Harmonia axyridis</i> *	Palaearctic	VI-2014, Jaboticabal, SP, Alexandrino JG col. (4 ex.)
<i>Tenuisvalvae notata</i> *	Neotropical	10-I-2013, Campinas, SP, Marsaro Júnior AL col. (1 ex.)
Neuroptera: Chrysopidae		
<i>Ceraeochrysa</i> sp.		III-2014, Jaboticabal, SP, Alexandrino JG col. (2 ex.)
Hymenoptera: Encyrtidae		
<i>Gyranusoidea indica</i> **	Oriental	VII-2014, Jaboticabal, SP, Alexandrino JG col. (4 ex.)

*First association with *M. hirsutus*.

**First report in Brazil.

in Bangladesh, Brazil, Chagos Archipelago, China, Fiji, Ghana, India, Indonesia, Madagascar, Malaysia, Mauritius, Myanmar, Oman, Pacific, Seychelles, South Africa, Sri Lanka, Thailand, Western Samoa, and Yemen (Poorani 2004).

Recorded prey of *C. nigrita* are: Aleyrodidae (1 species), Aphididae (2 species), Psyllidae (1 species), Monophlebidae (1 species), Pseudococcidae (3 species), Asterolecaniidae (2 species), Coccidae (10 species), and Diaspididae (26 species) (Samways 1984). In South Africa, *C. nigrita* occasionally feeds on young mealybugs, and in the laboratory it sometimes feeds on young *Planococcus citri* (Risso) (Pseudococcidae), but not on the adults (Samways 1984). In northeastern Brazil, *C. nigrita* was first recorded in Recife, state of Pernambuco (Samways 1989) and was observed preying on *Diaspis echinocacti* (Bouché) (Hemiptera: Diaspididae) infesting several cacti species in the states of Alagoas and Pernambuco (Lima & Gama 2001). Also, 1st-instars (crawlers) of this mealybug were phoretic on *C. nigrita* adults, which contributed to the spread of this pest (Lima & Gama 2001).

Cryptolaemus montrouzieri is native to the Australasian Zoogeographic Region, but now has a world-wide distribution due to its introduction in many countries for biological control purposes. This beetle is one of the most widely used biological control agents and has been used to target more than 16 pest species (Kairo et al. 2013). *Cryptolaemus montrouzieri* is a polyphagous predator with prey documented from at least 8 hemipteran families, including scale insects (Coccoidea). Most recently, it was introduced in parts of the Caribbean and Central and South America for control of the pink hibiscus mealybug *M. hirsutus* (Kairo et al. 2013). In Brazil, *C. montrouzieri* was introduced in 1997 from Chile to target *Praelongorthezia praelonga* (Douglas) (Hemiptera: Ortheziidae) and *Dysmicoccus brevipes* (Cockerell) (Hemiptera: Pseudococcidae), which are pests of citrus and pineapple, respectively (Vilela et al. 2000). *Cryptolaemus montrouzieri* was initially documented in Cruz das Almas (Bahia) and Jaboticabal (São Paulo) and is reported in other parts of Brazil.

Cycloneda sanguinea is a Neotropical species that is currently distributed in Argentina, Bolivia, Brazil (Amapá, Amazonas, Pará, Tocantins, Paraíba, Piauí, Bahia, Mato Grosso, Distrito Federal, Goiás, Mato Grosso do Sul, Minas Gerais, Espírito Santo, Rio de Janeiro, São Paulo, Paraná, Santa Catarina, Rio Grande do Sul), Chile, Colombia, Costa Rica, Ecuador, Guatemala, Honduras, Mexico, Nicaragua, Panama, Paraguay, Peru, Uruguay, USA, and Venezuela (Araujo-Siqueira & Almeida 2006). This species feeds primarily on aphids but is a generalist predator (Araujo-Siqueira & Almeida 2006). Michaud & Evans (2000) observed this coccinellid and *C. montrouzieri* (adults and larvae) preying upon *M. hirsutus* on *Hibiscus* shrubs in Puerto Rico.

Exoplectra is a Neotropical genus with 14 species reported in Brazil. *Exoplectra* species feed mainly on aphids and whiteflies (Costa et al. 2008), but *E. miniata* (Germar) was observed feeding on extrafloral nectaries of *Inga edulis* Mart. (Fabales: Fabaceae) in Nova Friburgo, Rio de Janeiro, Brazil (Almeida et al. 2011). *Exoplectra erythrogaster* Mulsant was recorded feeding on *Mimosicerya hempelii* (Cockerell) (Hemiptera: Coelostomidiidae) infesting *Cassia fistula* L. (Fabales: Fabaceae) in São Paulo, SP, Brazil (Mariconi & Zamith 1960). Oliveira et al. (2008) reported that *E. erythrogaster* preyed on *Protortonia navesi* Fonseca (Hemiptera: Monophlebidae), a pest of cassava that is native to central Brazil.

Harmonia axyridis is an Asiatic polyphagous coccinellid and has been used successfully as a biological control agent of aphids around the world (Koch 2003). In the absence of aphids, *H. axyridis* may feed on prey such as Tetranychidae, Psyllidae, Coccoidea, Curculionidae, and Lepidoptera or plant material, damaged fruits, nectar, and pollen, all of which can be used to complete its development (Berkvens et al. 2008). Xie et al. (2004) observed *H. axyridis* associated with *Phenacoc-*

cus azaleae Kuwana (Hemiptera: Pseudococcidae) on bunge prickly-ash tree *Zanthoxylum simulans* Hance (Sapindales: Rutaceae) in China.

In the late 1990s, *H. axyridis* was first introduced in South America in Mendoza Province, Argentina, to control aphids affecting peach orchards (Saini 2004). According to Kondo & Gonzalez (2013), *H. axyridis* was introduced to Colombia in 1989 or earlier, making it the second oldest record of the species in South America after the deliberate releases of the species in Mendoza, Argentina in 1986 (García et al. 1999). In Brazil, it was probably accidentally introduced and first detected in Curitiba, Paraná State, in 2002, feeding on 2 species of aphids (Almeida & Silva 2002). In the same municipalities, Martins et al. (2009) collected *H. axyridis* associated with potential prey species of aphids, scales, and psyllids that may be alternate prey sources, although most of the prey species were aphids. *Harmonia axyridis* was recorded in the states of São Paulo, Minas Gerais, and Goiás (Hartherreiten-Souza et al. 2012).

Tenuisvalvae notata is native to South America and a predator of mealybugs (Dreyer et al. 1997a,b). Recently, it was reported in the semi-arid region of Pernambuco, Brazilian State, associated with *Ferrisia virgata* (Cockerell) (Hemiptera: Pseudococcidae) on cotton and with *Dactylopius opuntiae* (Cockerell) (Hemiptera: Dactylopiidae) on cactus forage. In South America, it is distributed in Bolivia, Brazil (Amapá, Bahia, Mato Grosso do Sul, Rio de Janeiro, Rondônia, São Paulo), Colombia, and Paraguay (Barbosa et al. 2014). In 1980, it was introduced into Africa to control the cassava mealybug, *Phenacoccus manihoti* Matille-Ferrero (Hemiptera: Pseudococcidae) (Herren & Neuenschwander 1991; Chakupurakal et al. 1994).

NEUROPTERA

Ceraeochrysa species are distributed from southeast Canada to Argentina, and it is the largest genus of Neotropical Chrysopinae (Brooks & Barnard 1990). Freitas et al. (2009), in the last review of the genus, recognized 63 species in the Neotropics, 21 of which occur in Brazil (Freitas & Penny 2001). According to Sosa & Freitas (2010), larvae of *Ceraeochrysa* species feed on aphids, thrips, whiteflies, mites, and neonatal larvae of Lepidoptera in various types of agroecosystems. However, other authors have associated these species as predators of coccoids. De Bortoli et al. (2005) demonstrated that this species had potential as a control agent of *Selenaspidus* sp. (Hemiptera: Diaspididae) and *Coccus* sp. (Hemiptera: Coccidae). *Ceraeochrysa paraguayaria* (Navás) preferred the armored scale *Selenaspidus* sp. (De Bortoli & Murata 2006), and *Ceraeochrysa cincta* (Schneider) was observed on colonies of *Leptococcus eugeniae* (Miller & Denno) (as *Plotococcus eugeniae*) (Hemiptera: Pseudococcidae) (Eisner & Silberglied 1988). Eight species of Chrysopidae were associated with *M. hirsutus*, but of these only *Chrysoperla carnea* (Stephens) is known in Brazil (Chong et al. 2015).

HYMENOPTERA

Gyranoidea indica was described from India and introduced into Australia, Egypt, Guyana, several Caribbean countries (Noyes 2015), and California (USA) (Goolsby et al. 2002). In conjunction with the parasitoid *A. kamali* and the predator *C. montrouzieri*, *G. indica* has been widely used in biological control programs against the pink hibiscus mealybug. *Gyranoidea mirzai* (Agarwal) is associated with *M. hirsutus*, but it is restricted to the Oriental region (Chong et al. 2015). *Gyranoidea indica* has also been recorded as a parasitoid of other pseudococcids, including *F. virgata*, *Nipaeococcus viridis* (Newstead), and *Pseudococcus longispinus* (Targioni Tozzetti) (Sharaf & Meyerdirk 1987; Meyerdirk & Warkentin 1999; Abd-Rabou 2001). Evans et al. (2012) reported *A. kamali* and *G. indica* associated with *M. hirsutus* on San Andres Island, Colombia, and provided information and illustrations to differentiate the 2 species.

GENERAL DISCUSSION

Mutual occurrence of coccinellids and potential prey insects on a given plant does not necessarily indicate a prey–predator relationship, which must be positively observed in the field and/or tested in the laboratory (Hodek & Honek 1996). Of the insects collected on *H. rosa-sinensis* infested by *M. hirsutus* in the state of São Paulo, the predators *C. montrouzieri* and *C. sanguinea* and the parasitoid *G. indica* have already been recorded as natural enemies of this pest in other countries. Of the new associated species, only *T. notata* has Pseudococcidae as main prey or host. *Harmonia axyridis* is a generalist predator that will take coccoids as prey but feeds mainly on aphids. Species of *Exoplectra* are associated with the scale insect families Coelostomidiidae and Monophlebidae, and *Ceraeochrysa* species are generalist predators.

Secondary predators, or those species that will take prey beyond the species typically preferred, can be important in pest management. For example, *Nesticoccus sinensis* Tang (Hemiptera: Pseudococcidae) and *Eriococcus transversus* Green (synonym *Rhizococcus transversus*) (Hemiptera: Eriococcidae) are important pests of bamboo forests in southern Jiangsu (China), and the strategy used to control these pests involved using oil seed rape planted among the bamboo to attract aphids, which in turn attracted *Harmonia obscurisignata* (Liu) and *H. axyridis* (Xu & Wu 1989). After rape harvest, the coccinellids dispersed to the bamboo and fed on scales, reducing the scale population by 97 to 99%.

In our collections from some plants infested by the pink hibiscus mealybug, we observed aphids and/or other species of coccoids such as *Aphis gossypii* Glover (Aphididae), *Phenacoccus solenopsis* Tinsley (Pseudococcidae), *Parasaissetia nigra* (Nietner) (Coccidae), and *Pinaspis* sp. (Diaspididae), and it is possible that some coccinellids documented in association with *M. hirsutus* were initially attracted to the plant because of the presence of one of these other pests. Furthermore, ladybeetles may provide some suppression when the pest population is high, but not in lower populations because the beetles require large numbers of mealybugs to survive (Hoy et al. 2002), in which case other parasitoids and predators may provide some control.

In our observations, it was clear that *C. montrouzieri* was prevalent on large populations of *M. hirsutus* on *H. rosa-sinensis* in the state of São Paulo. However, additional ecological work in the field and tests in the laboratory are needed before proposing good management strategies against this pest in the state of São Paulo or other regions of Brazil.

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