

Endemic and invasive Coccinellidae associated with maize (*Zea mays* L.) fields, in Manabi province, Ecuador

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Abstract

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Maize (*Zea mays* L.), which is considered an important cereal everywhere, is widely cultivated in different agroecological conditions. Throughout their phenological stages, maize crops are attacked by pests and diseases, and in Ecuador these phytosanitary problems are usually counteracted with applications of pesticides. However, knowledge of beneficial entomofauna is fundamental to guide pest management programs. As part of this objective, the Coccinellidae species present in maize fields in localities (Santa Ana, Colon, and Danzarin) in Manabí province were identified. In each zone, maize plants were sampled from 2018–2019. A total of 2,654 specimens belonging to 14 taxa were collected in this study, of which *Cheilomenes sexmaculata*, *Cycloneda sanguinea*, *Hippodamia convergens*, *Hyperaspis arida*, and *Psyllobora confluens* were found in all the studied areas. *Hyperaspis arida* and *Diomus apollonia* are reported for the first time in Ecuador.

Keywords

biological control, diversity, lady beetles, natural enemies, predators

Introduction

Maize, *Zea mays* L. (Poaceae), is considered the third most important grain crop in the world, and widely cultivated due to its adaptability to different agroecological conditions (SINGH and SINGH, 2018; SULONG et al., 2019). In 2019, approximately 197,204,250 ha were planted worldwide with a production of 1,148,487,291 tons (FAOSTAT,

2019), and used mainly for human consumption and as animal feed (SANTANA et al., 2018). Maize originated in the Americas where it has been an important staple food for many centuries. In Ecuador, maize constitutes one of the main crops with 352,039 ha planted (39.41% of the area sown as temporary crops) and a production of 1,341,468 tons per year (MAG, 2020), which indicates the relevance of this cereal as a food source.

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Throughout its phenological stages, many insects and diseases are known to cause damage to the different parts of the plant and affect its growth and production (SINGH and SINGH, 2018). Some 250 phytophagous species associated with maize plants have been reported worldwide, of which approximately six are considered to be of economic importance, including the aphid species *Rhopalosiphum maidis* (Fitch, 1856) (Hemiptera: Aphididae) (SINGH and SINGH, 2018).

In Ecuador, highly toxic chemical insecticides are continuously used to reduce the damage caused by *R. maidis* and other pest species (NARANJO, 2017), without considering that, in general, the indiscriminate use of pesticides and inappropriate dosages generate resistance problems, as well as ecological imbalances because they interfere with the action of biological control agents (AKTAR et al., 2009).

In integrated pest management programs, the first alternative to be considered is biological control because natural enemies are usually found feeding on insect pests (LUCKMANN and METCALF, 1975). As a fundamental basis, it is necessary to know the diversity of beneficial entomofauna and to establish its role in biological control. Among beneficial insects, predatory coccinellids, commonly known as lady beetles, ladybugs or ladybirds are considered of great importance for agroecosystems and forests, contributing significantly to the control of key pests, such as aphids, whiteflies, mealybugs, and soft-bodied insects in general (GONZÁLEZ, 2015; ZACH et al. 2020). They are taxonomically classified in the superfamily Cucujoidea, family Coccinellidae Latreille, which includes approximately 6,000 species (VANDENBERG, 2002), of which an estimated 1,500 occur in South America and 134 are recorded in Ecuador (GONZÁLEZ, 2015).

Ecuador has a high diversity of agricultural pest species, but it also has a significant ecological richness, as well as a considerable abundance of natural enemies (CASTILLO, 2020). The objective of this study was to identify the species of the family Coccinellidae, both endemic and invasive, present in maize fields in localities of Manabi province, Ecuador, where the highest production of maize is found.

Materials and methods

The present study was carried out in three locations in the province of Manabi, i.e., Santa Ana (coordinates: 01°12'25"N, 80°22'07"E, 60 m), Colon (coordinates: 01°06'39"N, 80°24'57"E, 43 m) and Danzarin (00°55'57"N, 80°27'27"E, 18 m). All localities belong to the tropical dry forest life zone (FAO, 1981).

For the collection of coccinellids, in each zone, maize fields of approximately 2 ha were sampled weekly from September 10, 2018, to March 25, 2019, using the zigzag transect method, within the crop, entering through the third row of plants to avoid edge effect. A total of 100 plants per field per zone were observed in each of the samplings carried out. Coccinellids were collected using a mouth aspirator and once captured, they were placed in vials filled with

70% ethyl alcohol and taken to the laboratory of the Agency for the Regulation and Phytosanitary and Zoosanitary Control (AGROCALIDAD), in Manta, Ecuador. Part of the insect samples were mounted, and the rest were used to extract the genitalia for identification following the method detailed by GONZÁLEZ (2015).

The identifications were carried out in the Entomology Laboratory of the La Molina National Agrarian University in Lima, Peru, using the diagnostic features of Coccinellidae species from South America given by GONZÁLEZ (2015). Voucher specimens are deposited in the Entomological Collection of the Agrocalidad Laboratory, Manta.

Data analysis

The percentages of abundance were analyzed using a general linear model (GLM). Prior to the analysis, the variable was adjusted to the normal distribution with the function $\sqrt{x+1}$. Means were compared using the Least Squares test ($p < 0.05$). Additionally, to associate the percentages of abundance per species for each studied area, a principal component analysis was conducted. To obtain the principal components, a standardization of the data was executed (mean 0 and standard deviation 1), to avoid that the variables with greater variance dominated the others. Statistical analyses were performed with the Infostat program, version 2018 (DI RIENZO et al., 2018).

Results

A total of 2,654 coccinellid specimens were collected in this study, of which 27.1%, 33.7%, and 39.2% were found in Santa Ana, Colon, and Danzarin, respectively. Fourteen taxa were identified, six in Santa Ana, and a total of 11 in each of Colon and Danzarin (Tables 1–2), belonging to three subfamilies, most of which belonged to the subfamily Coccinellinae (84.17%), tribe Coccinellini (97.76%) (Table 1). *Cheilomenes sexmaculata* (Fabricius, 1781), *Cycloneda sanguinea* (Linnaeus, 1763), *Hippodamia convergens* Guerin-Meneville, 1842, *Hyperaspis arida* GORDON and CANEPARI, 2008, and *Psyllobora confluens* (Fabricius, 1801) (Figs 1–2), were found in all the studied areas (Table 1).

The most abundant species were *Ch. sexmaculata*, *H. convergens* and *H. arida*, detecting differences between localities (Table 2). The highest percentages of abundance for *Ch. sexmaculata* were observed in Colon, while *H. convergens* was superior in Colon and Danzarin and *H. arida* was significantly abundant in Santa Ana ($p < 0.05$). The second predominant group consisted of *Ch. sexmaculata* and *H. convergens* associated with the localities of Colon and Santa Ana, followed by *Coleomegilla maculata limensis* (Philippi & Philippi, 1854), *H. arida* and *C. sanguinea* in Colon, as well as the latter species detected in Danzarin. A fourth group consisted of *P. confluens* (Colon and Santa Ana), *C. maculata limensis* (Santa Ana) and *C. sanguinea* (Santa Ana). The rest of the species by locality reached the lowest percentages of abundance that did

not exceed 1% (Table 2). The associations of these species with the localities studied are shown in Fig. 3.

The principal component analysis corroborated the association of *H. arida* with Santa Ana (Fig. 3) and the association of *Azya* sp., *Diomus* sp., *C. maculata limensis*, *P. confluens* with Colon, as well as the association of the species *Paraneda pallidula guticollis* (Mulsant, 1850), *Diomus apollonia* Gordon, 1999 *Ch. sexmacu-*

lata, *Pentilia insidiosa* Mulsant, 1850 and *Hyperaspis onerata* (Mulsant, 1850) with Danzarin. Additionally, the species, *C. sanguinea*, *H. convergens*, *Brachiacantha darlene* GORDON & CANEPARI, 2014 and *Tenuisvalvae bromelicola* (Sicard, 1925), had a shared relationship with the localities of Colon and Danzarin. The two axes in the graph in Fig. 3 explain 100% of the total variability in the observations.

Table 1. Coccinellidae species found in the sampled localities of Santa Ana, Colon, and Danzarin in Manabí province, Ecuador

Subfamily	Tribe	Genus/Species	Colon	Danzarin	Santa Ana	Total
Coccinellinae	Coccinellini	<i>Hippodamia convergens</i>	+	+	+	1.029
Coccinellinae	Coccinellini	<i>Cheilomenes sexmaculata</i>	+	+	+	876
Scymninae	Hyperaspidini	<i>Hyperaspis arida</i>	+	+	+	383
Coccinellinae	Coccinellini	<i>Cycloneda sanguinea</i>	+	+	+	155
Coccinellinae	Coccinellini	<i>Coleomegilla maculata limensis</i>	+	-	+	118
Coccinellinae	Halyziini	<i>Psyllobora confluens</i>	+	+	+	50
Scymninae	Brachiacanthini	<i>Brachiacantha darlene</i>	+	+	-	18
Scymninae	Hyperaspidini	<i>Tenuisvalvae bromelicola</i>	+	+	-	12
Coccinellinae	Coccinellini	<i>Paraneda pallidula guticollis</i>	+	+	-	6
Scymninae	Cryptognathini	<i>Pentilia insidiosa</i>	-	+	-	3
Scymninae	Diomini	<i>Diomus apollonia</i>	-	+	-	1
Scymninae	Hyperaspidini	<i>Hyperaspis onerata</i>	-	+	-	1
Azyinae	Azyini	<i>Azya</i> sp.	+	-	-	1
Scymninae	Diomini	<i>Diomus</i> sp.	+	-	-	1

The (+) sign represents the presence of the species and the (-) sign its absence.

Table 2. Percentages of the abundance of the Coccinellidae species found in the localities of Santa Ana, Colon, and Danzarin in Manabí province, Ecuador

Species	Colon	Danzarin	Santa Ana
<i>Cheilomenes sexmaculata</i>	21.79 b	45.73 a	29.05 b
<i>Hippodamia convergens</i>	44.36 a	43.77 a	25.14 b
<i>Hyperaspis arida</i>	12.07 c	0.98 f	35.81 ab
<i>Coleomegilla maculata limensis</i>	10.17 c	0.00 g	3.65 de
<i>Tenuisvalvae bromelicola</i>	0.45 f	0.79 f	0.00 g
<i>Psyllobora confluens</i>	3.02 de	0.20 f	2.84 de
<i>Cycloneda sanguinea</i>	6.82 cd	6.67 cd	3.51 de
<i>Brachiacantha darlene</i>	0.89 f	0.98 f	0.00 g
<i>Paraneda pallidula guticollis</i>	0.22 f	0.39 f	0.00 g
<i>Diomus</i> sp.	0.11 f	0.00 g	0.00 g
<i>Diomus apollonia</i>	0.00 g	0.10 g	0.00 g
<i>Pentilia insidiosa</i>	0.00 g	0.29 f	0.00 g
<i>Azya</i> sp.	0.11 f	0.00 g	0.00 g
<i>Hyperaspis onerata</i>	0.00 g	0.10 f	0.00 g

R2: 0.68; CV: 25.57. Mean followed by different letters is significantly different at 0.05 level of significance using Least Square test ($p < 0.05$).

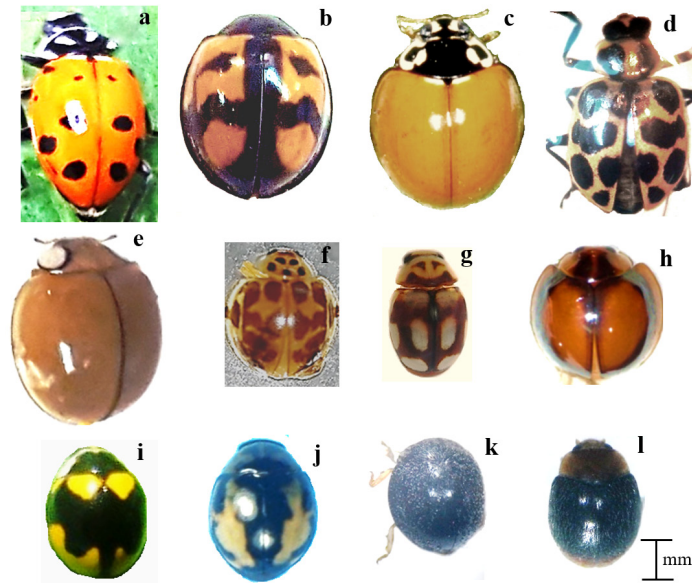


Fig 1. Coccinellidae species present in Ecuador and observed in maize fields. **a.** *Hippodamia convergens* (Guerin-Meneville), **b.** *Cheilomenes sexmaculata* Fabricius, **c.** *Cycloneda sanguinea* Linnaeus, **d.** *Coleomegilla maculata limensis* Philippi & Philippi., **e.** *Paraneda pallidula guticollis* (Mulsant) **f.** *Psyllobora confluens* (Fabricius), **g.** *Brachiacantha darlene* Gordon & Canepari **h.** *Pentilia insidiosa*. Mulsant, **i.** *Tenuisvalvae bromelicola* (Sicard), **j.** *Hyperaspis onerata* (Mulsant), **k.** *Azya* sp., **l.** *Diomus* sp.

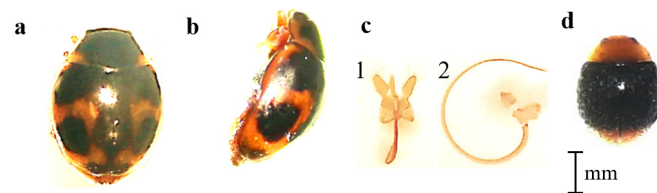


Fig. 2. Coccinellidae species identified for the first time in Ecuador. **a.** *Hyperaspis arida* GORDON & CANEPARI, 2008 (dorsal view); **b.** *Hyperaspis arida* GORDON & CANEPARI, 2008 (♀) (lateral view); **c1.** tegmen, **c2.** penis, **d.** *Diomus apollonia* Gordon, 1999 (♀) (dorsal view). Scale bar: 1 mm.

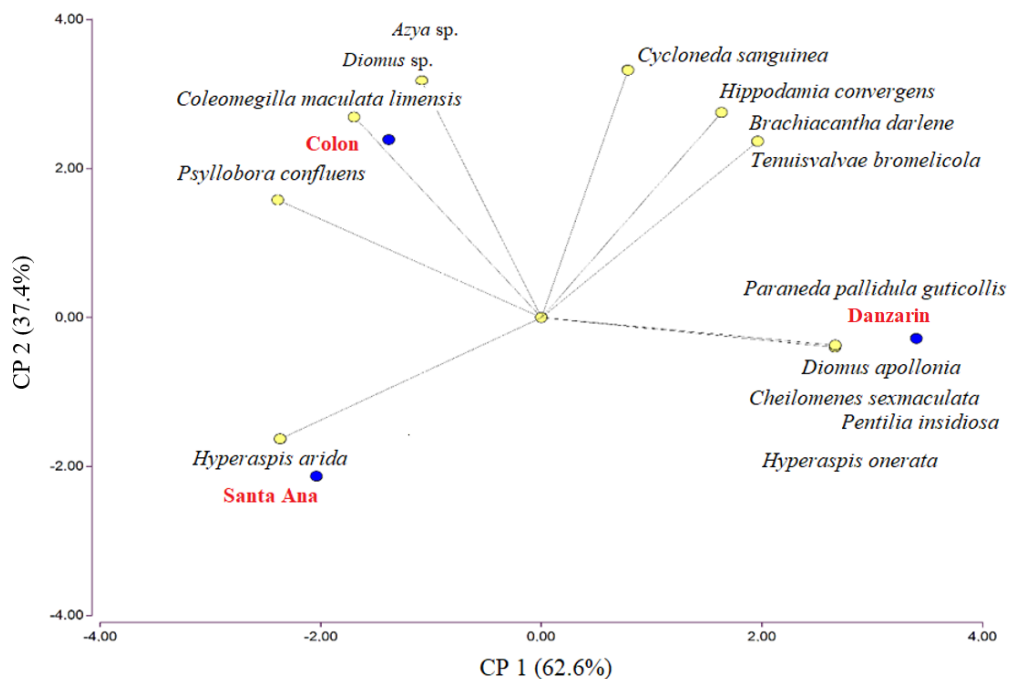


Fig. 3. Principal component analysis of Coccinellidae species present in the sampled localities.

Discussion

The importance of the species found in this study as key predators of some pests has been previously noted. GONZÁLEZ (2019) pointed out that *H. convergens* is of North American origin and that in the last 50 years, it has colonized South American countries. *Hippodamia convergens* has been associated with the aphid *Rhopalosiphum padi* (Linnaeus, 1758) (Hemiptera: Aphididae) in commercial maize fields from six locations in South Dakota (ELLIOT et al., 2002) and considered an important predator of the aphid species *Melanaphis sacchari* (Zehntner, 1897) on sorghum in Mexico (PROVISOR-BERMÚDEZ and LÓPEZ-MARTÍNEZ, 2016). A review study carried out by WEBER and LUNDGREN (2009) evaluating the trophic ecology of Coccinellidae mentioned *H. convergens* preying on nymphs of *Bemisia tabaci* Gennadius, 1889, and eggs of *Pectinophora gossypiella* (Saunders, 1844) (Lepidoptera: Gelechiidae), under field and laboratory conditions. In Ecuador, the presence of *H. convergens* has been reported since 1957 (CASTILLO et al., 2020). Additionally, in 1978, 24 million individuals were imported from USA to Ecuador for the control of *Icerya purchasi* (Maskell, 1878) (Hemiptera: Monophlebidae) (MOLINEROS, 1984). More recently, *H. convergens* was found preying on *Aphis gossypii* Glover, 1877, in a cucumber field in Ecuador (ROMERO et al., 2019).

Cheilomenes sexmaculata is a species native to Asia, also recorded in Oceania and was introduced to several countries in South America, such as Colombia, Chile, Ecuador, Peru, and Venezuela (CORNEJO and GONZÁLEZ, 2015; GONZÁLEZ, 2016; KONDO et al., 2015; RAMÍREZ et al., 2017). In Ecuador, it has occasionally been found feeding on leaf nectaries of *Talipariti tiliaceum* var. *pernambucense* (Arruda) Fryxell (Malvaceae) (CORNEJO and GONZÁLEZ, 2015). Additionally, it has been observed preying on nymphs of the Asian citrus psyllid, *Diaphorina citri* Kuwayama, 1908, on orange jasmine, *Murraya paniculata* (L.) Jack and *Citrus* spp. in Colombia and Ecuador (CHÁVEZ et al. 2017; 2019; ERRÁEZ et al., 2020; KONDO et al., 2015; 2018), and individuals of *A. gossypii* on cucumber in Ecuador (ROMERO et al., 2019). CASTILLO and MIRÓ (2013) pointed out that, in Tumbes, Peru (border area with Ecuador), this species was detected in 2011, feeding on various species of aphids (Hemiptera: Aphididae) i.e., *R. maidis* on maize, *Aphis craccivora* Koch, 1854, on cowpea beans, *Toxoptera aurantii* (Boyer de Fonscolombe, 1841), on citrus and cacao, *Aphis nerii* Boyer de Fonscolombe, 1841, on pink laurel and the soft scale, *Philephedra* sp. (Hemiptera: Coccidae), on ornamental croton. This shows the high potential of *Ch. sexmaculata* as a natural enemy of aphid species (including *R. maidis*) and other soft-bodied insects, which consumption capacity has been studied under laboratory conditions (PANDI et al., 2012).

Hyperaspis arida (Fig. 2a and b) was found in Ecuador in this study for the first time with high percentages of abundance and conspicuously associated with the town of Santa Ana (Fig. 3). GORDON and CANEPARI (2008) reported that *H. arida* could be confused with *Hyperaspis festiva* Mulsant, 1850, however, the shape of the male genitalia

separates both species. *Hyperaspis arida* was hitherto restricted to Peru (CASTILLO and MIRÓ, 2013; GONZÁLEZ, 2007; JUÁREZ-NOÉ and GONZÁLEZ-CORONADO, 2019) where it has been observed feeding on the whitefly, *Aleurodicus juleikae* Bondar, 1923 (Hemiptera: Aleyrodidae) on mango, and several species of scale insects, including mealybugs on some fruit trees (CASTILLO and MIRÓ, 2013).

Cycloneda sanguinea is native to the New World and is found in all South American countries (GONZÁLEZ, 2015). It has been found in Ecuador feeding on *D. citri* on *M. paniculata* and *Citrus* spp. (Chavez et al., 2017; 2019) and *A. gossypii* on cucumber (ROMERO et al., 2019). Like *H. convergens* in Peru, this species has been associated with *R. maidis* on maize, as well as feeding on several species of aphids on citrus trees (CASTILLO and MIRÓ, 2013). Adults and larvae of *H. convergens*, *C. maculata*, and *C. sanguinea* feed on various phytophagous insects such as mites, aphids, scales, mealybugs, eggs and young lepidopteran larvae, including the armyworm, *Spodoptera frugiperda* Smith (Lepidoptera: Noctuidae) in maize fields in the Americas (FAO, 2018).

Psyllobora confluens is distributed in most South American countries (GONZÁLEZ, 2015), but little is known about its biology. CIVIDANES et al. (2007) carried out a laboratory study in which they demonstrated that *P. confluens* can survive and complete its development feeding on the fungus, *Erysiphe cichoracearum* De Candolle, which causes powdery mildew on okra, cucurbits, and ornamental plants. Additionally, it has been found associated with spider mites (Acari: Tetranychidae), whiteflies of the genus *Aleurodicus* (Hemiptera: Aleyrodidae), and some mealybugs species on Musaceae, as well as with the phytopathogenic fungus, *Oidium* sp., on tamarind.

Although *C. maculata limensis* was not present in Danzarin, its percentages of abundance ranged between 3–10% in Santa Ana and Colon (Tables 2). *C. maculata limensis* is found in Peru, Chile, and Ecuador (GONZÁLEZ, 2015). In Peru, it is reported as the most common and important coccinellid species on rice, possibly preying on lepidopteran eggs, as well as in maize fields attacked by *R. maidis* (CASTILLO and MIRÓ, 2013). In Ecuador, *C. maculata limensis* has been associated with *A. gossypii* in cucumber fields (ROMERO et al., 2019).

Diomus apollonia Gordon, 1999 has hitherto only been reported from Para, Brazil (GONZÁLEZ, 2015) and is herein reported for the first time in Ecuador from specimens collected in a maize agroecosystem in Danzarin. There have been no studies that refer to its biology and potential in biological control.

Although these coccinellids are found on maize plants cultivated in localities that belong to the same life zone, the differences in diversity and abundance could be associated with the specificities of each crop. For another beetle species, *Premnobius cavipennis* Eichhoff, 1878 (Coleoptera: Curculionidae), it has been indicated that differences in abundance among localities for the same host plant could be related to the characteristics of each site, such as plant diversification and agroecosystem management, among other factors, which, together with environmental

factors, determine its distribution and abundance (AVEROS et al., 2021).

This study of maize agroecosystems in Manabí province provides a list of coccinellids that have a potential as biological control agents of some pests, and reports for the first time the presence of *H. arida* and *D. apollonia* in Ecuador.

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