







Scientific Note

Subfamily composition of Ichneumonidae (Hymenoptera: Ichneumonoidea) from Eastern Uruguay

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Abstract. The knowledge of invertebrate diversity in Uruguay is much less as compared to bird and tetrapod fauna, but some important advances have been made within some groups of insects. The Ichneumonidae constitute one of the largest families in the animal kingdom. This family is important because their larvae can be either endo- or ectoparasitoids of larvae or pupae of holometabolous insects as well as Chelicerata. In this study ichneumonid wasps were collected from three environments near the city of Castillos, Rocha Department, Uruguay between December 2014 and December 2016. A total of 5740 Ichneumonidae specimens were collected, representing 19 subfamilies, of which 3685 specimens (64.2%) correspond to three subfamilies: Campopleginae (1533 specimens/26.7%), Ichneumoninae (1303/22.7%) and Cryptinae (849/14.8%); all others subfamilies together represented less than 7.0% of the total specimens. In addition, 32 genera were registered for the first time in Uruguay.

Keywords: Idiobiont, koinobiont, Neotropical, parasitoid wasps, South America.

The Darwin wasps (Ichneumonidae) constitute one of the largest families in the animal kingdom, with a total of 25285 species, classified in 1601 genera and 44 subfamilies (Yu et al. 2016, Klopstein et al. 2019). In the Neotropical region, the ichneumonids are divided in 32 subfamilies and 435 genera (Yu et al. 2016).

Biologically, the family utilize a somewhat different spectrum of hosts, their larvae being endo- or ectoparasitoids of larvae or pupae of holometabolous insects (e.g. Coleoptera, Diptera, Hymenoptera, Lepidoptera, Raphidioptera and Trichoptera) and Chelicerata (Araneae and Pseudoscorpionida) (Wahl & Sharkey 1993). This family is ecologically significant and extremely species-rich, but its tropical diversity is poorly known (Hopkins et al. 2019).

The biodiversity of the ichneumonids in Neotropical region is better studied in Mexico (e.g. Pérez-Urbina et al. 2018), Costa Rica (e.g. Gauld 2000), Guatemala, El Salvador and Nicaragua (e.g. Veijalainen et al. 2014), while considering Central America, and in Peru/Ecuador (Veijalainen et al. 2012), Brazil (Kumagai & Graf 2000, Fernandes et al. 2019b), Chile (Porter 1979) and Argentina (Porter 1975), in South America. Still there exists a knowledge gap about ichneumonids in many countries of Neotropical region, especially in South America.

According to Aldabe et al. (2008), the information regarding invertebrate diversity in Uruguay is much lower compared to bird and tetrapod fauna, but some important advances have been made with some groups of insects, such as Hymenoptera (Zolessi et al. 1989, Castiglioni et al. 2017). In light of the increasing concern about the loss of Uruguayan biodiversity caused by the conversion of natural ecosystems for agriculture in the last few decades (Aldabe et al. 2008), it is extremely important to carry out studies focused on local fauna.

Thus far, a total of 10 subfamilies, 30 genera and 49 species are listed for Uruguay (Yu et al. 2016, Santos & Aguiar 2018, Santos & Hoppe 2018, Supeleto et al. 2019): Anomaloninae (*Parania* Morley, 1913), Banchinae (*Glypta* Gravenhorst, 1829), Campopleginae (*Campoletis* Förster, 1869; *Casinaría* Holmgren, 1859; *Diadegma* Förster, 1869; *Venturia* Schrottky, 1902), Cryptinae (*Aeglocryptus* Porter, 1987; *Chromocryptus* Ashmead, 1900; *Compsocryptus* Ashmead, 1900;

Digonocryptus Viereck, 1913; *Dotocryptus* Brèthes, 1919; *Mallochia* Viereck, 1912; *Messatoporus* Cushman, 1929; *Neocryptopteryx* Blanchard, 1947; *Phycitiplex* Porter, 1987; *Trachysphyrus* Haliday, 1836), Diplazontinae (*Diplazon* Nees, 1819), Ichneumoninae (*Matara* Holmgren, 1868; *Thymebatis* Brèthes, 1909; *Trogomorpha* Ashmead, 1900), Ophioninae (*Enicospilus* Stephens, 1835; *Ophion* Fabricius, 1798; *Thyreodon* Brullé, 1846), Phygadeuontinae (*Chirotica* Förster, 1869), Pimplinae (*Calliephialtes* Ashmead, 1900; *Itoplectis* Förster, 1869; *Neotheronia* Krieger, 1899; *Pimpla* Fabricius, 1804; *Tromatobia* Förster, 1869) and Tersilochinae (*Stethantyx* Townes, 1971), and among which, at least nine genera (*Campoletis*, *Campoplex* Gravenhorst, 1829, *Casinaría*, *Diadegma*, *Enicospilus*, *Itoplectis*, *Ophion*, *Stethantyx* [= *Thersilochus*] and *Venturia*) have economic importance as biological control agents in Uruguay (Bentancourt et al. 2009).

In this study we evaluated the diversity and subfamily composition of Ichneumonidae in three environments of Eastern Uruguay for a sampling period of two years using Malaise traps. The Hymenoptera sampling in each of the studied environments was performed every two weeks between December 2014 and December 2016 with two Malaise traps, separated from each other by about 100 m. ETOH 95% GL was used for the preservation of captured insects.

The traps remained active throughout the entire study period in three environments near the city of Castillos, Rocha Department, Uruguay. The environments studied were: a) Natural Field Area (NFA), with low grazing intensity by cattle - Don Bosco / Campo Natural; 34°05'1.07"S, 53°45'43.08"W - 57 m asl; b) Pasture System Area (PSA), area under production under grazing of cattle and sheep - Cardoso / Bosque Campo; 34°05'26.8"S, 53°52'14.4"W - 89 m asl, and c) Integrated Agriculture Area (IAA), areas under integrated livestock production system with winter and summer agriculture - Branaa; 34°02'33.7"S, 53°50'02.7"W - 26 m asl (between December 2014 and December 2015) and Llambí; 34°24'42.2"S, 54°08'10.5"W - 18 m asl (between January and December 2016).

The separation, cataloging and quantification of the insects contained in each sample were carried out in the "Laboratorio

de Entomología” of “Centro Universitario Regional del Este of the Universidad de la República”, Rocha, Uruguay. After the completion of the above mentioned procedures, the parasitic Hymenoptera were sent for identification in the “Laboratório de Sistemática e Bioecologia de Parasitoides e Predadores” of “Instituto Biológico”, in Ribeirão Preto, Brazil. All the Ichneumonidae specimens were pinned, labeled, identified and deposited in the Invertebrate Collection of the Instituto Nacional de Pesquisas da Amazônia, Manaus, Brazil (M. L. Oliveira, curator).

A comparison of the diversity of subfamilies captured in each environment was performed using rarefaction curves by the resampling bootstrap process (to obtain a confidence interval of subfamily wealth). The bootstrap analyses were calculated using EstimateS Win9.1 software (Colwell 2013) using 2,000 randomizations and 95% confidence intervals. Maps were prepared using free website SimpleMappr (www.simplemappr.net).

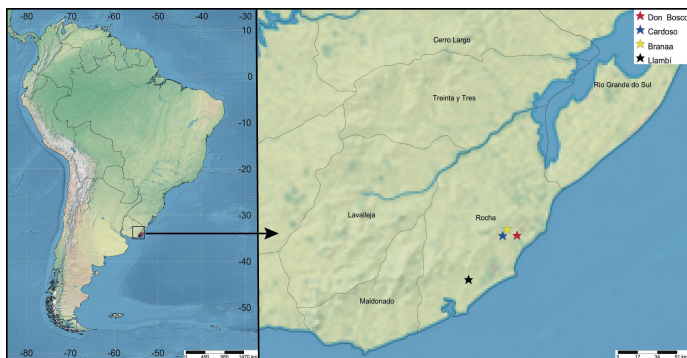


Figure 1. Collection points in the municipality of Castillos, Rocha Department, Uruguay.

A total of 5740 Ichneumonidae specimens were collected, representing 19 subfamilies of which, 3685 specimens (64.2%) were distributed in Campopleginae (1533 specimens/26.7%), Ichneumoninae (1303/22.7%) and Cryptinae (849/14.8%) (Tab. 1), and all other subfamilies together presented less than 7% of total specimens.

Ichneumoninae, Campopleginae and Cryptinae were the most abundant subfamilies, corresponding to 64% of the total sampled in Don Bosco and Cardoso (Figs. 2A and 2B). In Branaa, Campopleginae, Ichneumoninae and Phygadeuontinae were the most abundant, representing approximately 60% of the total (Fig. 2C). In Llambí, Campopleginae represented 66% of the total (258 specimens), and the other subfamilies were sampled with less than 25 specimens each, presenting a disproportional scenario compared to the other environments (Fig. 2D).

For the subfamily rarefaction curves, in 49 samplings, the three environments presented similar diversity (Fig. 3A). However, NFA presented the asymptote at 15 samples, indicating that all subfamily fauna was effectively collected in this environment (17 subfamilies). The highest diversity was observed (19 subfamilies) in PSA, although the curve tended to be asymptotic (3 singletons were observed). In IAA (Branaa + Llambí), there was also a tendency to reach an asymptote with 17 subfamilies (2 singletons).

To verify the quality of the samples in IAA, data were analyzed using the number of individuals from each location sampled. Thus, according to Fig. 3B, we found that for both Branaa and Llambí, sampling for only one year in each environment was inadequate, leading to few collected specimens, and a low diversity being observed in relation to other environments.

To date, a total of 10 following subfamilies have been listed for Uruguay: Anomaloniinae, Banchinae, Campopleginae, Cryptinae, Diplazontinae, Ichneumoninae, Ophioninae, Phygadeuontinae, Pimplinae and Tersilochinae (Yu et al. 2016). Here in, we increased the number of listed subfamilies by almost 90%, recognizing nine new subfamilies for this country: Cremastinae, Ctenopelmatinae, Labeninae, Mesochorinae, Metopiinae, Nesomesochorinae, Orthocentrinae, Tryphoninae and Rhysinae, totaling 19 subfamilies.

Table 1. Ichneumonidae subfamilies collected during two years at four sites in the municipality of Castillos, Rocha Department, Uruguay.

Subfamily	Don Bosco	Cardoso	Branaa	Llambí	Total
Anomaloniinae	20	54	4	2	80
Banchinae	164	115	4	6	289
Campopleginae	521	654	100	258	1533
Cremastinae	149	55	6	7	217
Cryptinae	431	341	53	24	849
Ctenopelmatinae	2	0	0	1	3
Diplazontinae	1	12	7	18	38
Ichneumoninae	781	410	95	17	1303
Labeninae	1	4	0	0	5
Mesochorinae	29	16	3	5	53
Metopiinae	63	43	2	1	109
Nesomesochorinae	49	6	1	0	56
Ophioninae	57	224	12	16	309
Orthocentrinae	36	26	6	8	76
Phygadeuontinae	204	92	68	18	382
Pimplinae	144	121	27	11	303
Tersilochinae	21	9	0	0	30
Tryphoninae	46	9	48	1	104
Rhysinae	1	0	0	0	1
Total	2720	2191	436	393	5740

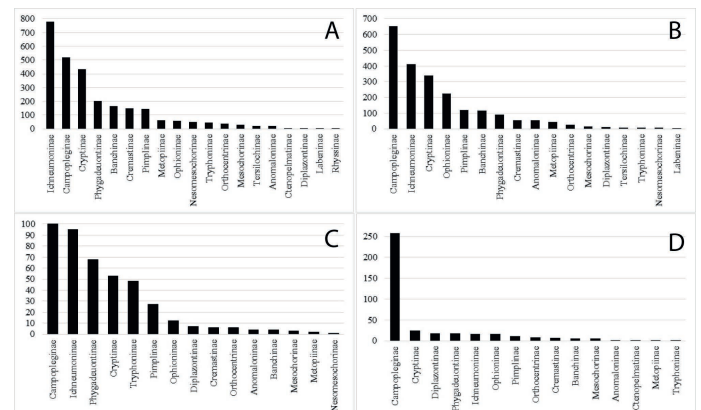


Figure 2. Abundance of Ichneumonidae subfamilies collected by each site: A. Don Bosco; B. Cardoso; C. Branaa; D. Llambí.

These numbers are still far less than those recorded for the southern region of Brazil: 24 subfamilies, 129 genera and 354 species (Fernandes et al. 2019a), which most likely should share part of their fauna, due to the similarity of some of the environments and biomes (e.g. Pampa). However, we listed a larger fauna record when compared to the state of Rio Grande do Sul, Brazil (bordering state with Uruguay), which has 12 subfamilies, 29 genera and 48 species (Fernandes et al. 2019a).

These results are close to those obtained by Kumagai & Graf (2000, 2002), who sampled 18 and 20 subfamilies, respectively, in southern Brazil. However, the taxa identification in a specific level can probably reveal a more realistic diversity in these environments.

The same goes for the diversity found in the environments sampled in this study. The IAA (Branaa + Llambí) areas had relatively low abundance compared to the other two environments, and only 15 subfamilies were collected in each of them (Tab. 1). Most likely the fauna found in this environment may be associated with agricultural pests present in these agroecosystems. In the interior of conventional monocultures, the composition of parasitoids is affected by simplification of the landscape, and other disturbances; and in agroecosystems, it is determined by the associated vegetation, the

abundance, and the time of parasitoid colonization (Greathead 1986).

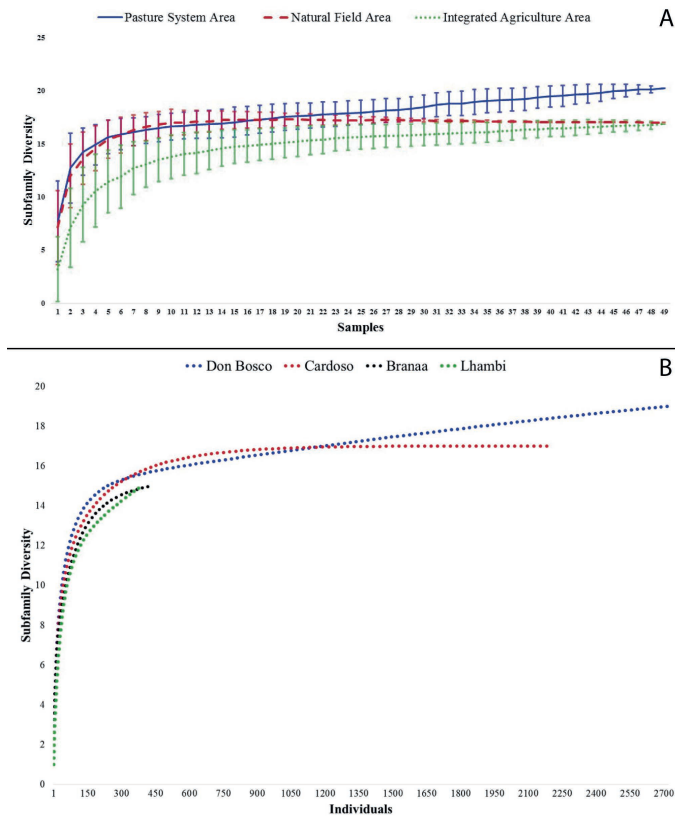


Figure 3. A. Subfamily diversity by Samples; B. Subfamily diversity by Individuals.

In Llambí, a large disparity was observed regarding the collection of subfamily Campopleginae (66% of the total). This can indicate that some ichneumonids benefit from the agricultural environment because their hosts are agricultural pests, such as *Venturia* Schrottky, 1902 and *Microcharops* Roman, 1910 (collected in this survey, Tab. 2); both are generally abundant in agroecosystems. *Venturia* attack Pyralidae larvae (Lepidoptera) and *Microcharops* are important biological controllers of *Anticarsia gemmatilis* Hübner, 1818 (Noctuidae) and *Alabama argillacea* (Hübner, 1823) (Erebidae) (Gauld 1984, Gupta 1987).

In this study, 48 genera (Tab. 2) have been recognized so far, 32 of them being new records for Uruguay. Considering the 32 new records in this study, and the previously reported genera from the literature (30), the fauna of this country is currently represented by 62 genera distributed in 19 subfamilies.

This estimate is higher than that recorded from the state of Rio Grande do Sul (bordering northern Uruguay with Brazil), where only 29 genera have been recorded (Fernandes et al. 2019b). However, in the southern region of Brazil (composed by the states of Paraná, Santa Catarina and Rio Grande do Sul), 129 genera have been recorded (Fernandes et al. 2019b), as well as in Argentina (bordering country at the west), 140 genera of Ichneumonidae has been recorded so far (Yu et al. 2016). Thus, we estimate that Uruguay may probably have a total of 100-150 genera in its territory.

It is noteworthy that part of the specimens analyzed in the present study still awaits generic identification (mainly Cryptinae, Campopleginae, Ichneumoninae and Phygadeuontinae). Most likely with a larger sampling effort and greater depth in taxonomic studies, this diversity may increase considerably in the coming years, which may be able to corroborate our estimates.

Table 2. Ichneumonidae identified during two years at four sites in the municipality of Castillos, Rocha Department, Uruguay. The symbol “*” was used to highlight the first records for Uruguay.

Subfamily	Genus
Anomaloninae	<i>Parania</i> Morley, 1913
Banchinae	<i>Syzeuctus</i> Förster, 1869*
Campopleginae	<i>Casitaria</i> Holmgren 1859
Campopleginae	<i>Dusona</i> Cameron, 1901*
Campopleginae	<i>Microcharops</i> Roman, 1910*
Campopleginae	<i>Venturia</i> Schrottky, 1902
Cre mastinae*	<i>Eiphosoma</i> Cresson, 1865*
Cre mastinae*	<i>Pristomerus</i> Curtis, 1836*
Cre mastinae*	<i>Temelucha</i> Förster, 1869*
Cre mastinae*	<i>Trathala</i> Cameron, 1899*
Cre mastinae*	<i>Xiphosomella</i> Szépligeti, 1905*
Cryptinae	<i>Dotocryptus</i> Brèthes, 1919
Cryptinae	<i>Messatoporus</i> Cushman, 1929
Diplazontinae	<i>Diplazon</i> Nees, 1819
Ichneumoninae	<i>Joppocryptus</i> Viereck, 1913*
Labeninae*	<i>Grotea</i> Cresson, 1864*
Labeninae*	<i>Labena</i> Cresson, 1864*
Metopiinae*	<i>Chorinaeus</i> Holmgren, 1858*
Metopiinae*	<i>Colpotrochia</i> Holmgren, 1856*
Metopiinae*	<i>Exochus</i> Gravenhorst, 1829*
Metopiinae*	<i>Trieceus</i> Townes, 1946*
Metopiinae*	<i>Triclistus</i> Förster, 1869*
Metopiinae*	<i>Hypsicera</i> Latreille, 1829*
Mesochorinae*	<i>Mesochorus</i> Gravenhorst, 1829*
Nesomesochorinae*	<i>Nonnus</i> Cresson, 1874*
Ophioninae	<i>Alophophion</i> Cushman, 1947*
Ophioninae	<i>Enicospilus</i> Stephens, 1835
Ophioninae	<i>Ophion</i> Fabricius, 1798
Ophioninae	<i>Thyreodon</i> Brullé, 1846
Orthocentrinae*	<i>Megastylus</i> Schiødte, 1838*
Orthocentrinae*	<i>Orthocentrus</i> Gravenhorst, 1829*
Phygadeuontinae	<i>Chirotica</i> Förster, 1869
Pimplinae	<i>Acrotaphus</i> Townes, 1960*
Pimplinae	<i>Calliephialtes</i> Ashmead, 1900
Pimplinae	<i>Clistopyga</i> Gravenhorst, 1829*
Pimplinae	<i>Eruga</i> Townes, 1960*
Pimplinae	<i>Flacopimpla</i> Gauld, 1991*
Pimplinae	<i>Hymenoepimecis</i> Viereck, 1912*
Pimplinae	<i>Itopectis</i> Förster, 1869
Pimplinae	<i>Neotheronia</i> Krieger, 1899
Pimplinae	<i>Pimpla</i> Fabricius, 1804
Pimplinae	<i>Polysphincta</i> Gravenhorst, 1829*
Pimplinae	<i>Tromatobia</i> Förster, 1869
Pimplinae	<i>Zatypota</i> Förster, 1869*
Pimplinae	<i>Zonopimpla</i> Ashmead, 1900*
Tersilochinae	<i>Stethantyx</i> Townes, 1971
Tryphoninae*	<i>Netelia</i> Gray, 1860*
Rhyssinae*	<i>Epirhyssa</i> Cresson, 1865*

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Authors' Contributions

R.I.R.L., N.W.P., J.P.B. and E.C. planned and conceived the experiments, and contributed with the sample preparation. D.R.R.F. and D.G.P. prepared and identified the material and wrote the manuscript. All authors have discussed the results and contributed to its final version.

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