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(RESEARCH ARTICLE)

Study of the biological and taxonomic characteristics of the families Megamerinidae, Nemestrinidae and Therevidae (Insecta: Diptera)

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Abstract

Regarding the biology of the Megamerinidae family, their larvae live under the bark of dying or dead deciduous trees where they are predators or scavengers of the larvae of other insects. Adults, which are rarely captured, are found mainly in wooded areas, close to the larval habitat, mainly on trunks and leaves. All known larvae of Nemestrinidae are internal parasitoids of nymphs and adults of locusts and beetle larvae. This makes the nemestrinids have the potential to be used as biological control agents for Mantodea. Nemestrinid adults are often found hovering around flowers where they feed on nectar, emitting a characteristic buzzing sound. The family of Therevidae biology is poorly understood. Adults are diurnal and many can live for more than a month under favorable conditions. They often go out to sunny spots on trails, the males usually waiting for the females to pass. Resting sites are generally specific to different species Larvae feed on other insects, mainly beetles, butterflies, moths and other flies that live in leaf litter, tree holes, or sandy terrains. Study of the biological and taxonomic characteristics of the families Megamerinidae, Nemestrinidae and Therevidae (Insecta: Diptera). The methodological basis of the present work consists of bibliographical research of scientific articles published in national and international academic scientific journals classified by the Coordination for the Improvement of Higher Education Personnel (CAPES). The search criterion for articles was prioritizing articles dealing with the topic. Document analysis was used as a data collection method to gather information on the websites of the University of São Paulo (http://www.usp.br/), Latin American and Caribbean Literature in Health Sciences (http:// www.bireme.br/), Scielo (http://www.scielo.org) and University of Brasília (http://www.bce.unb.br/sistemas/pesq_bibliografica.php).

Keywords: Adult; Larvae; Beatles; Mantodea; Nectar

1. Therevidae Family

The Therevidae are a family of flies of the superfamily Asiloidea commonly known as stiletto flies. The family contains about 1,600 described species worldwide, most diverse in arid and semiarid regions with sandy soils (Figures 1-3) [1].

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Source: https://en.wikipedia.org/wiki/Therevidae

Figure 1 *Thereva nobilitata* (Fabricius 1775), female

Sources: Salvador Vitanza and https://elp.tamu.edu/ipm/bugs/order-diptera-flies/family-therevidae-stiletto-flies/diptera-therevidae-subfamily-thervinae-stiletto-flies-b/

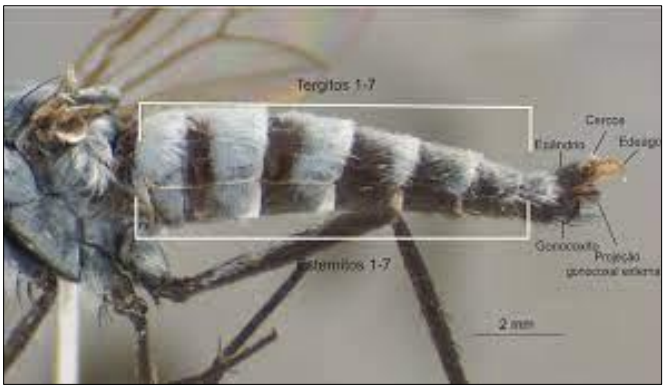
Figure 2 Diptera: Therevidae/subfamily Therevinae-Stiletto Flies (B)

Source: https://www.inaturalist.org/observations/103874231

Figure 3 *Thereva microcephala* Loew, 1847

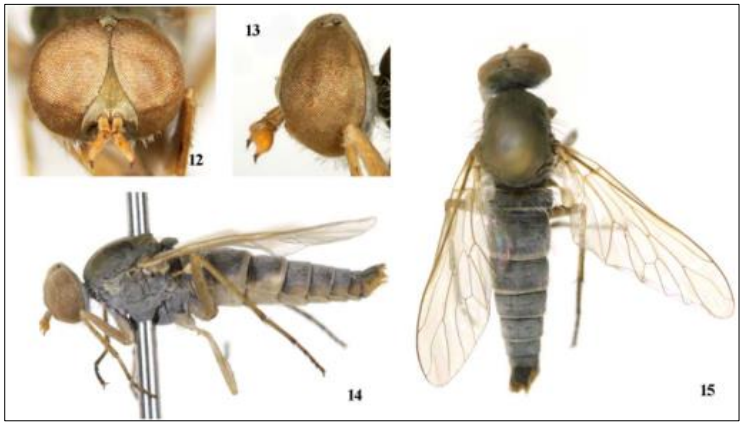
1.1. Description

The Family Therevidae are flies that vary between 3 to 15 mm in length, are generally thin, have quite long legs and the abdomen is frequently pointed at the rear, which is why they are called "stiletto flies". They are closely related to the Asilidae from which they can be distinguished as the vertex between the eyes is flat or slightly convex, while in Asilidae it is concave. They can be distinguished from the more related but much smaller Scenopinidae by the lack of modified setal patches on the dorsum of the second abdominal tergite. In the Scenopinidae these groups of setae occur variably in this segment (Figures 4-7) [1,2].



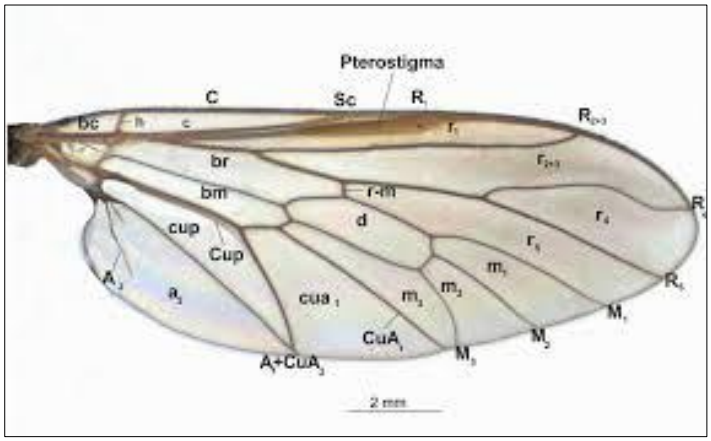
Source: https://www.ppgbas.uema.br/wp-content/uploads/2017/02/DISSERTA%C3%87%C3%83O-DA-RAIANA-04.02.17-Fim.pdf

Figure 4 Morphology and terminology of *Penniverpa* abdomen. Lateral view with dorsal segments (Tergites) and ventrais (Sternites)



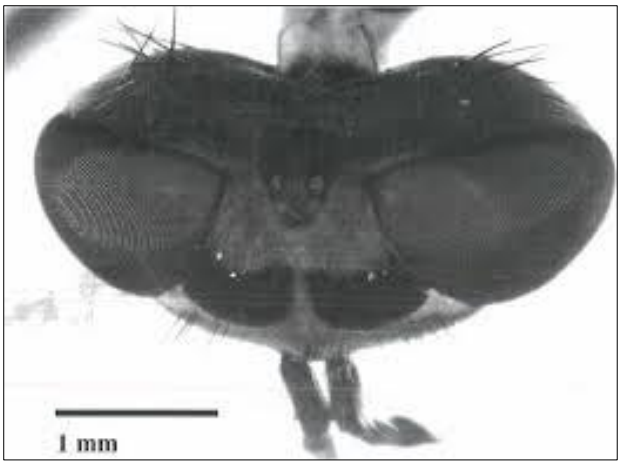
Source: https://zenodo.org/record/213841#.Y8rAwnbMLIU

Figure 5 figs12–15. External morphology of male *Ammothereva flavifemorata* sp. nov. (12). head, frontal view; (13). head, lateral view; (14). habitus, lateral view; (15). habitus dorsal view



Source: https://www.ppgbas.uema.br/wp-content/uploads/2017/02/DISSERTA%C3%87%C3%83O-DA-RAIANA-04.02.17-Fim.pdf

Figure 6 *Penniverpa* wing morphology and terminology. A1, A2, anal vein; a, anal cell; CuA1, CuA2, anterior cubital vein; cua1, cubital-anterior cell; bc, costal basal cell; bm, basal-mid cell; br, basal-radial cell; C, costal vein; c, costal cell; cua1, anterior cubital cell; Cup, posterior cubital vein; cup, cubital cell posterior; d, discal cell; h, transverse humeral vein; M, middle vein; M1, M2, M3, branches of the M vein; m1, m2, m3, middle cell; R, radial vein; R1, R2+3, R4, R5, R vein branches or combinations, for example R2+3; r1, r2+3, r4, r5, radial cell; r-m, mid-radial transverse vein; Sc, subcostal vein; sc, subcostal cell



Sources: Photograph: Holston KC and https://www.zobodat.at/pdf/Fauna-Flora-Rheinland-Pfalz_9_1193-1202.pdf

Figure 7 *Thereva oculata* Egger, 1859, dorsal view of head (female) showing the bipunctate frontal callus. Specimen preserved in alcohol from malaise trap sample collected at RoBstein, 18.V-30. V2000, leg. NIEHUIS

1.2. Biology

The family of Therevidae biology is poorly understood. Adults are diurnal and many can live for more than a month under favorable conditions. They often go out to sunny spots on trails, the males usually waiting for the females to pass. Resting sites are generally specific to different species. Most of the family Therevidae quickly fly short distances to recover when disturbed. Adults are not predators (Figures 8-9) [1,2,3].

Sources: ID confirmed by Elias de Bree @ waarneming.nl and https://www.jungledragon.com/image/62590/thereva_nobilitata_-_larva_pupa_fly.html

Figure 8 *Thereva nobilitata* (Fabricius 1775) - larva, pupa, fly collage of shots of a larva of *T. nobilitata* that I kept documenting the development to imago. Top row: Complete larva and a close-up of the head, showing the "spoon"-shaped appendage typical for Therevidae larvae Bottom left: Fresh pupa Bottom center: Mature pupa Bottom right: Resulting imago *T. nobilitata* (male)

Sources: Photo by Andrew Weeks, Cesar Australia and https://cesaraustralia.com/pestfacts/stiletto-flies/

Figure 9 A stiletto fly larva - Therevidae family: Stiletto fly larvae can be recognized by their smooth white to pinkish, legless bodies, which taper at both ends. They have a small head capsule and their bodies appear divided into nineteen segments. Earthworms can be distinguished from stiletto fly larvae by the lack of a head capsule and a highly segmented body (> 100 segments). The body segmentation of wireworm larvae is more reminiscent of stiletto fly larvae, however, the former can be distinguished by the presence of three pairs of legs and a serrated plate with two protrusions at their tail-end

Most species drink water; some feed on nectar, pollen and plant exudates. A few species may even be cannibals and others are phytophagous. Males of some species tend to set on beams of light on trails, usually waiting for females to mate. Some species may even have a very specific substrate for repose, from sand spots, rocks, herbs and leaves to trunks (Figures 10-11) [1,2,3].

Sources: Photo by Andrew Weeks Cesar Australia and https://cesaraustralia.com/pestfacts/stiletto-flies/

Figure 10 True wireworm larva head and legs (left) and serrated plate with two upturned protrusions (right)

Source: https://tasmanianinsectfieldguide.com/hexapoda/insectsoftasmaniadiptera/suborder-brachycera/infraorder-orthorrhapha/therevidae/

Figure 11 Copulation Genus *Anabarhynchus* sp.

White, highly sclerotized larvae are voracious generalist predators of fossorial arthropods active in sandy soils, mulch, underbark debris, tree hole deposits, and other similar loose substrates. When exposed, they writhe violently. The Therevidae can be considered biological control agents of certain actual or potential fossorial pests. Adults can be

collected by taking advantage of the fact that they look for drinking water in streams or small puddles, where they sometimes gather, especially in periods of drought (Figures 12A-12B) [3,4,5].

Sources: https://www.flickr.com/photos/johnhallmen/3788170487 and https://www.dreamstime.com/coastal-silver-stilleto-stiletto-fly-acrosathe-annulata-therevidae-insects-image152146649

Figure 12A Stiletto fly *Thereva nobilitata* (Fabricius 1775) adult feeding nectar on pollen family: Therevidae. **Figure 12B** Coastal Silver-stilleto, stiletto fly, *Acrosathe annulata* (Fabricius, 1805) (Insects: Therevidae)

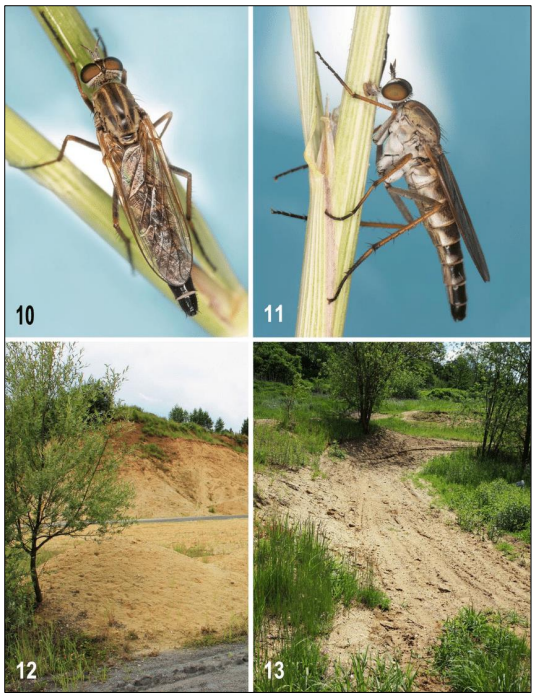
The larvae can be collected by straining the material where they are found and can be raised in the sand by feeding them beetle larvae. They usually occur in dry areas but they can also be found in mountains and tropical humid regions. Larvae feed on other insects, mainly beetles, butterflies, moths and other flies that live in leaf litter, tree holes, or sandy terrains (Figure 13) [4,5,6].

Source: https://www.mapress.com/zt/article/view/zootaxa.4816.3.6

Figure 13 A new bee-mimicking stylet fly with predatory larvae enveloping prey in China discovered on iNaturalist

1.3. Habitat and distribution

The habitat of the Therevidae is more varied than that of other Asiloidea, but as in Asiloidea, preferred ecosystems better suit the larvae, so these insects are more common in thickets of xerophilous plants garrigue and maquis, in deserts and on sandy beaches (Figure 14) [4,5,6].



Sources: Photo by J. Roháček and https://www.researchgate.net/figure/13-Therevidae-and-their-habitats-10-Cliorismia-rustica-Panzer-female-in-dorsal_fig1_353327634

Figure 14 Therevidae and their habitats. (10) - *Cliorismia rustica* (Panzer, 1802) (Panzer), female in dorsal view, body length ca 11 mm; (11) -the same, laterally; (12) -habitat of *C. rustica*, sand heaps in NW part of the Závada sand-pit; (13) –the habitat of *Dialineura anilis* (Linnaeus, 1761), sand places disturbed by vehicles on bottom of the Oldřišov sand-pit

The Therevidae are represented in all zoogeographical regions of the Earth. The Therevinae is present in all continents, with a lower frequency in the eastern region. The Phycinae have spread to the Afrotropical and the Holarctic. The Xestomyzinae are mainly Afrotropical. The Agapophytinae are endemic to the Australasian realm [5,6,7].

1.4. Taxonomy

Members of the Therevidae in general are poorly known and poorly understood. In the world, 840 species are scarcely known, and there are estimated to be more than 2,500. In the Neotropics, 147 species in 19 genera are recognized today, and only in Costa Rica, there are 16 described species in 8 genera. However, this country is rich in species of this family, with an estimated 12 genera containing more than 30 species, many of which have not yet been described [6,7,8].

Therevidae is divided into four subfamilies: Agapophytinae, Phycinae, Xestomyzinae and Therevinae. Worldwide, there are 1,123 described species in 119 genera, and this number represents only 56,15% of the estimated fauna of Therevidae. Neotropical fauna comprises 147 species [7,8,9].

- **Agapophytinae** is characterized by the presence of elongated spots with velvety textures on the ventral surface of the anterior and posterior femur, also present on the posteroventral surface of the gonocoxites (secondarily reduced in *Acraspisa* Kröber, 1912) and also by the presence of three spermathecae attached to the spermatic duct to form a common duct.
- **Phycusinae** has the following characteristics: developed or absent acanthophorites; membranous furcation; abdominal tergites 9–10 separated; three spermathecae and absence of a spermatic sac.
- **Xestomyzinae** is characterized by the set of macro bristles present on the sternite 8 of females used to dig the soil during oviposition reduction of the dorsal apodeme of the aedeagus and the presence of sclerites in the *Dystyphalus* [7,8,9].
- **Therevinae contains** half of the genera and more than 70% of the species described for the world, with 107 species in 25 genera (one extinct) cataloged for the region Neotropical. They have some characteristics that define the group: the presence of scaly macro bristles on the femurs; the presence of two spermathecae and one spermatic sac, and wing spines present in the pupa (Figures 15-23) [7,8,9].

Source: https://www.inaturalist.org/taxa/639871-Agapophytinae/browse_photos

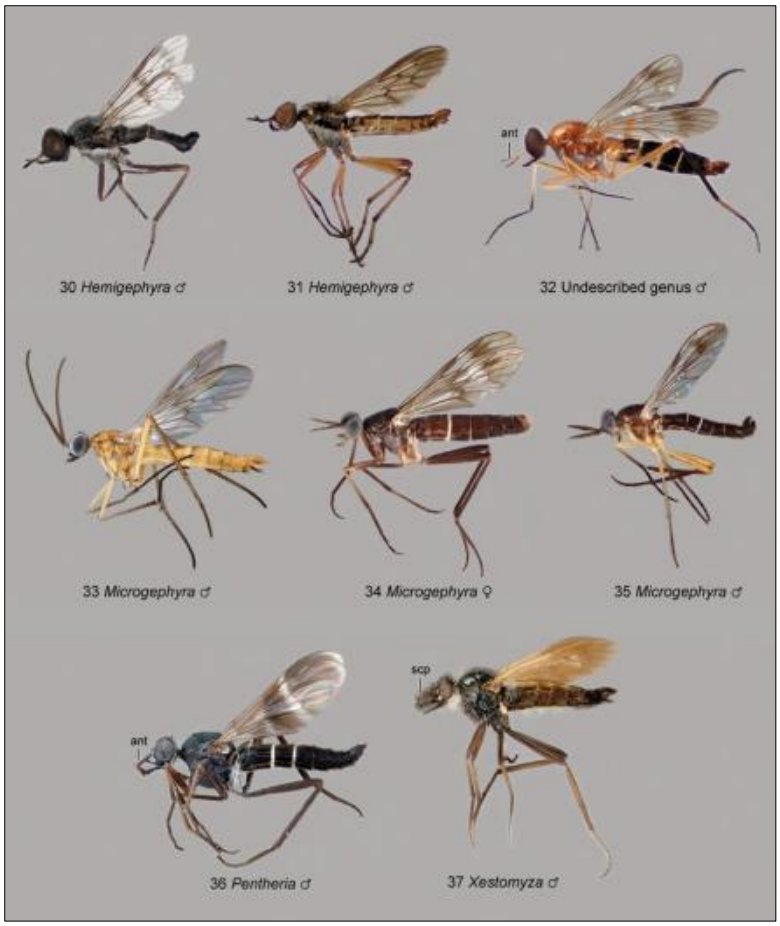
Figure 15 Subfamily Agapophytinae

Sources: Photo 9275639, (c) Valter Jacinto, all rights reserved and https://inaturalist.ca/photos/9275639

Figure 16 Subfamily Phycinae

Sources: Photo 46608468, (c) Alan Manson, some rights reserved (CC BY), uploaded by Alan Manson and https://uk.inaturalist.org/photos/46608468b

Figure 17 Subfamily Xestomyzinae

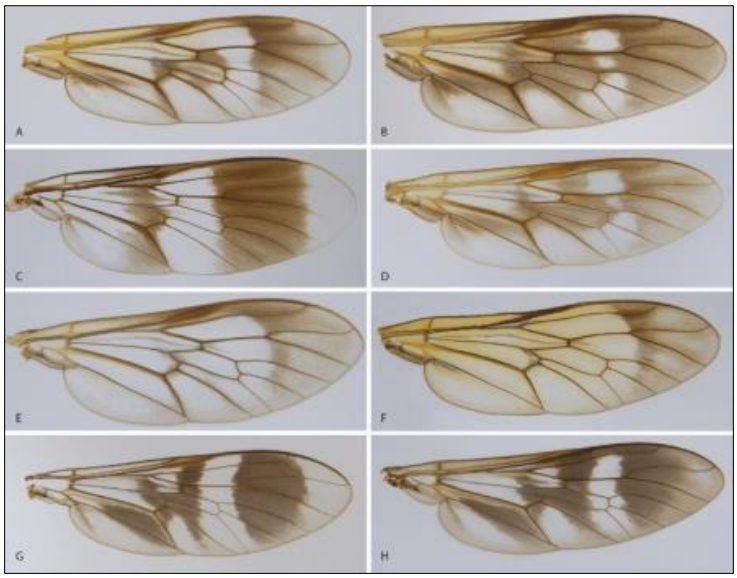


Source: https://www.researchgate.net/figure/37-Habitus-of-Therevidae-lateral-views-30-Hemigephyra-atra-Lyneborg-31-H_fig13_321934448

Figure 18 figs. 30-37. Habitus of Therevidae (lateral views): (30) *Hemigephyra atra* Lyneborg, 1972 ♂; (31) *Hemigephyra braunsi* (Kröber, 1931). ♂; (32) Undescribed genus ♂; (33) *Microgephyra* sp. ♂; (34) *Microgephyra* sp. ♀; (35) *Microgephyra* sp. ♂; (36) *Pentheria* sp. ♂; (37) *Hemigephyra braunsi* (Kröber, 1931) ♂.

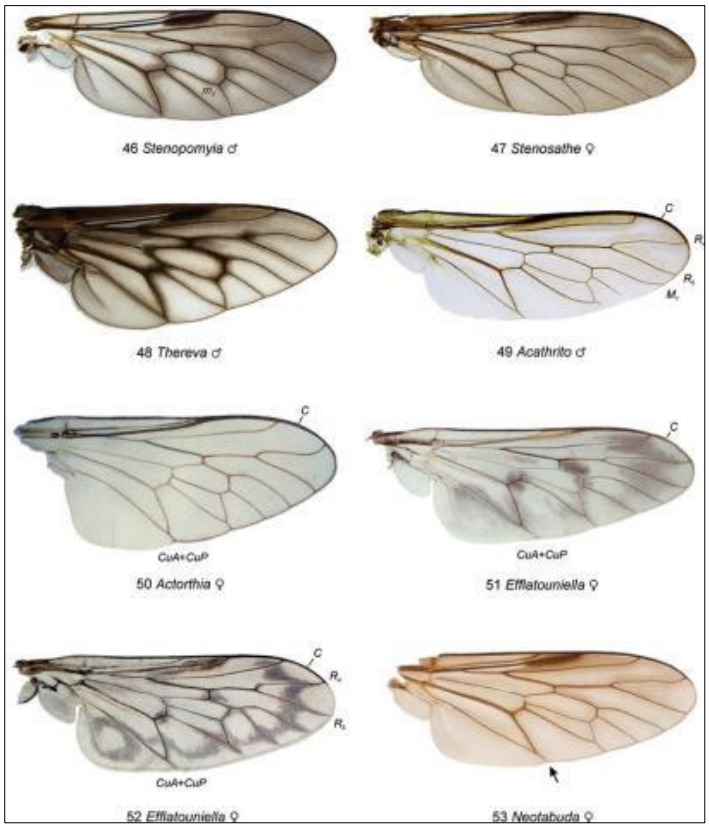
Source: https://zookeys.pensoft.net/article/53587/zoom/fig/19/

Figure 19 *Calophytus chazeaui* sp. nov. (A) An adult male lateral view (B) same, oblique view (C) adult female oblique view (D) same, lateral view. Body length: male



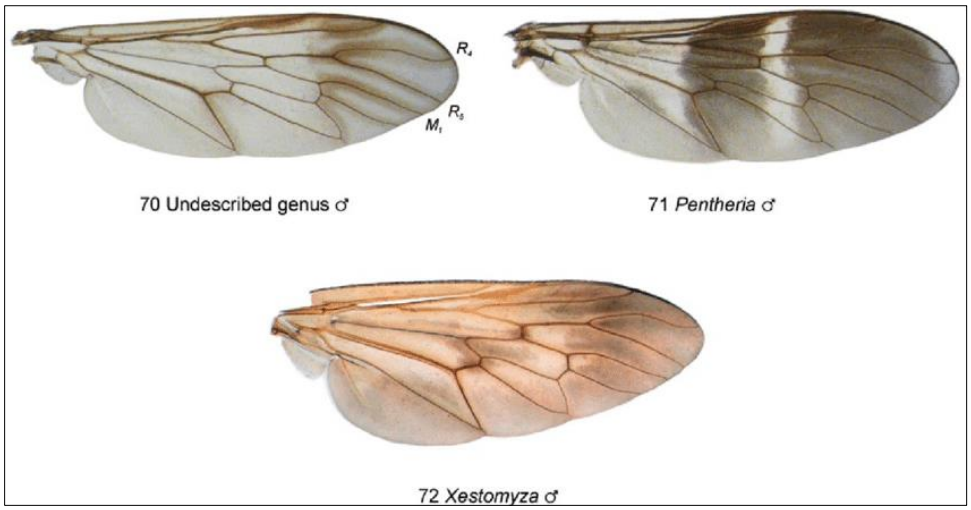
Source: https://zookeys.pensoft.net/article/53587/zoom/fig/17/

Figure 20 (A) Wings of *Calophytus* gen. nov. and *Jeanchazeauia* gen. nov. *Calophytus chazeaui* sp. nov. (B) *Calophytus schlingeri* sp. nov. (C) *Calophytus grandiosus* sp. nov. (D) *Calophytus matilei* sp. nov. And *Calophytus webbi* sp. nov. (F) *Calophytus monteithi* sp. nov. (G) *Jeanchazeauia amo*a sp. nov. (H) *Jeanchazeauia nubilosus* sp. nov. (female)



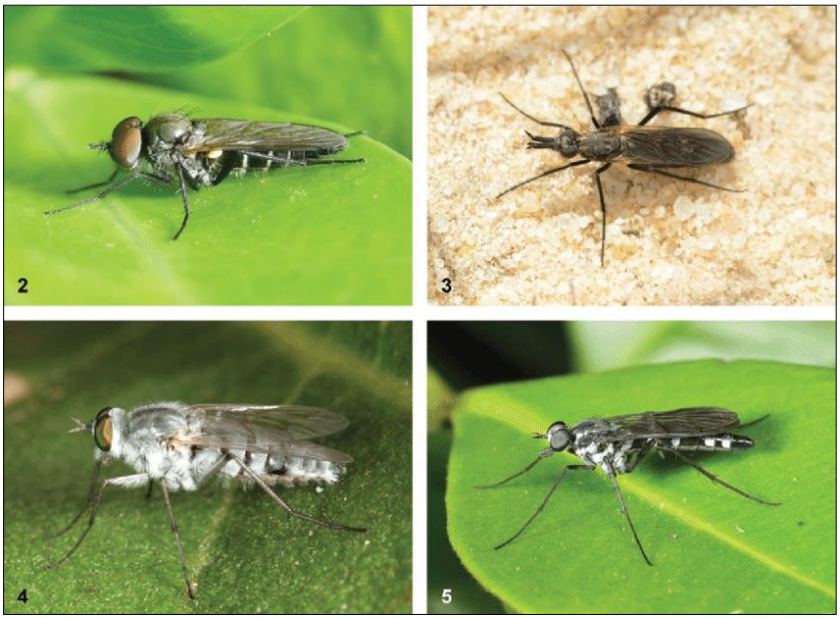
Source: https://www.researchgate.net/figure/46-53-Wings-of-Therevidae-dorsal-views-46-Stenopomyia-sp-47-Stenosathe-sp_fig7_321934448

Figure 21 figs. 46-53. Wings of Therevidae (dorsal views): (46) *Stenopomyia* ♂; (47) *Stenosathe* ♀; (48) *Thereva* ♂; (49) *Acathrite* ♂; (50) *Actorthia* ♀; (51) *Efflatouniella* ♂; (52) *Efflatouniella* ♀; (53) *Neotabuda* ♀ (arrow indicates extent of costal vein). Abbreviations: C-costal vein; CuA+CuP-anterior branch of cubital vein + posterior branch of cubital vein; M 1-first branch of media; m 3-third medial cell; R 4-upper branch of the third branch of radius; R 5-lower branch of the third branch of radius



Source: https://www.researchgate.net/figure/72-Wings-of-Therevidae-dorsal-views-70-Undescribed-genus-71-Pentheria-sp_fig10_321934448

Figure 22 figs. 70-72. Wings of Therevidae (dorsal views): (70) Undescribed genus ♂; (71) ♂; (72) *Xestomyza* ♂



Sources: Photographs © S.A. Marshall and https://www.researchgate.net/figure/5-Photographs-of-living-Afrotropical-Therevidae-2-Acathrito-lindneri-Lyneborg-South_fig2_321934448

Figure 23 figs.2-5. Photographs of living Afrotropical Therevidae: (2) *Acathrito lindneri* Lyneborg, 1963 (South Africa); (3) *Xestomyza lugubris* Wiedemann, 1830 (South Africa); (4) *Irwiniella tomentosa* (Becker, 1904) (Namibia); (5) *Stenopomyia angulata* Lyneborg, 1866 (Madagascar)

Objective

The objective of the manuscript is to study the taxonomic biological characteristics of the families Megamerinidae, Nemestrinidae and Therevidae (Insecta: Diptera).

2. Methods

The methodological basis of the present work consists of bibliographical research of scientific articles published in national and international academic scientific journals classified by the Coordination for the Improvement of Higher Education Personnel (CAPES). The search criterion for articles was prioritizing articles dealing with the topic. Document analysis was used as a data collection method to gather information on the websites of the University of São Paulo (http://www.usp.br/), Latin American and Caribbean Literature in Health Sciences (http:// www.bireme.br/), Scielo (http://www.scielo.org) and University of Brasília (http://www.bce.unb.br/sistemas/pesq_bibliografica.php).

3. Select papers

3.1. Study 1

3.1.1. Checklist of species from Mato Grosso do Sul

Therevidae. Regarding therevid species from Mato Grosso do Sul, only one record was obtained within the literature published up to 2010: *Cyclotelus kroeberi* (Cole, 1960), a species included in the subfamily Therevinae. The state of Amazonas (AM) presents the greater species richness (eight species), followed by Paraná (PR), Santa Catarina (SC) and Rio de Janeiro (RJ), each state with six recorded species [10].

The estimated fauna of therevids for Mato Grosso do Sul is at least eight species since many other species are recorded for the surrounding states: *Henicomyia flava* Lyneborg, 1972, *Brachylinga ornata* (Krober, 1911), *Cyclotelus silacrusus* Irwin & Webb, 1992 and *Anolinga melanothrix* Gaimari & Irwin, 2000) for São Paulo; *H. flava* for Mato Grosso and *H. flava* Goiás. All of these species are likely to also occur in Mato Grosso do Sul [10].

From Brazil, 30 species and 11 genera of Therevidae are recorded. The state of Amazonas (AM) presents the greater species richness (eight species), followed by Paraná (PR), Santa Catarina (SC) and Rio de Janeiro (RJ), each state with six recorded species. The estimated fauna of therevids for Mato Grosso do Sul is at least eight species since many other species are recorded for the surrounding states: *H. flava*, *Brachylinga ornata* (Kröber, 1911), *C. beckeri*, *C. silacrusus* and *A. melanothrix* for São Paulo; *H. flava* and *L. yneborge* for Mato Grosso and *H. flava* for Goiás. All of these species are likely to also occur in Mato Grosso do Sul [10].

3.2. Study 2

Penniverpa has 13 described species, two with an exclusively Nearctic occurrence, nine exclusives to the Neotropics and two commons to both regions. Three species are known for Brazil: *Penniverpa* has 13 described species, two with an exclusively Nearctic occurrence, nine exclusives to the Neotropics and two commons to both regions. Three species are known for Brazil: *Penniverpa* has 13 described species, two with an exclusively Nearctic occurrence, nine exclusives to the Neotropics and two commons to both regions [11].

Three species are known for Brazil: *Penniverpa* has 13 described species, two with an exclusively Nearctic occurrence, nine exclusives to the Neotropics and two commons to both regions. Three species are known for Brazil: *Penniverpa alvatra* Irwin & Webb, 1992 and *Penniverpa dives* (Schiner, 1868) for the North region, and *Penniverpa parvula* (Kröber, 1911) for the Brazilian Northeast [11].

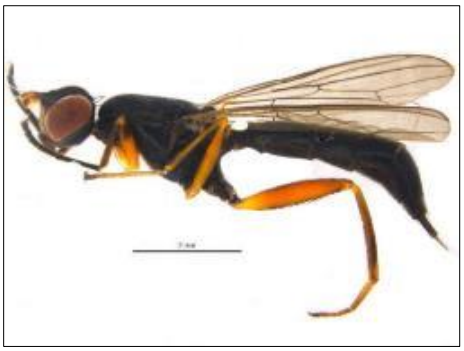
The genus is characterized by the absence of bristles on the forehead of males; the absence of shiny calli on the face and antennal base; the presence of bristles around the central depression. From the sternum; absence of bristles on the posterior surface of the middle thigh; widely open m3 cell and spots on the ventral surface of the hypoproct, in addition to the presence of a sinuous sclerite in short, spiny “S” shape. alvatra Irwin & Webb 92 and *P. dives* for the North region, and *P. parvula* for the Brazilian Northeast [11].

4. Megamerinidae Family

The Megamerinidae are a small family of Acalyptera Diptera. It has an average size between 6 and 10 mm. It is an elongated species, with a shiny black body and pale to reddish femurs. The head is small and spherical and the antennae are short with pubescent edges. The ocellis are present, while ocellar bristles, postocular bristles, front orbitals, interfrontals and vibrissae are absent [12,13].

4.1. Description

The wings are brownish, without spots. The costal vein is continuous and the vein subcostal is fully developed; cross-vein BM-Cu is present and the cup cell is closed. In the tibias, it is missing the preapical dorsal bristle. This species, together with the previous characters, is very easy to recognize by presenting dilated posterior femurs and two rows of spines on the ventral face (Figures 24- 25) [13,14].



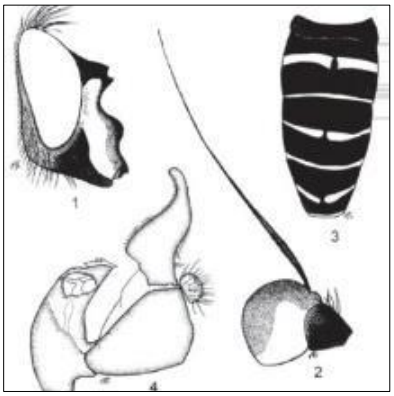
Source: https://eol.org/pages/747767

Figure 24 *Megamerina dolium* (Fabricius 1805)

Source: https://www.naturespot.org.uk/species/tachypeza-nubila

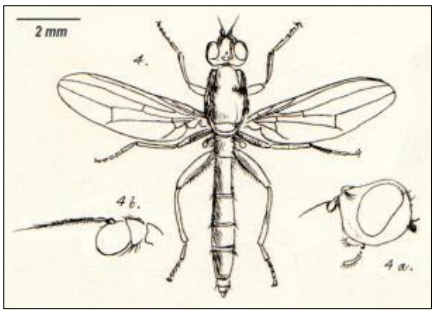
Figure 25 *Tachypeza nubila* (Meigen, 1804). Un Megamerinidae forestier. Description: Length 2.5 to 3.5 mm. A small dark brown looking fly. The front legs appear to have swollen thighs and are slightly more yellowy brown. The wings have a dark shading. Habitat: Usually you will find them on tree trunks (both standing and felled trees) and they can hide under loose bark or stones as well when feeling threatened. When to see it May to October. Life History: When threatened they usually try to run away (very quickly for a fly!), flying away is usually the last resort. UK Status. Widespread but under recorded in Britain.VC55 Status in Leicestershire and Rutland not known

At first glance, due to its general appearance, this species can be confused with the Diptera of the *Loniceracera* Meigen of the family Psilidae or some species of Hymenoptera of the family Ichneumonidae. However, they are very easily distinguished by present all the latter very long antennae. Hymenoptera pieces of evidence have four wings, while Diptera has two; however, when the wings are folded and close to the body, this difference is not useful for separating them (Figures 26-27) [15,16,17].



Source: https://www.researchgate.net/figure/3-Megamerina-dolium-1-head-front-view-2-body-lateral-view-3-body-dorsal-view_fig1_334506505

Figure 26 *Megamerina dolium* (Fabricius, 1805): (1) - head (2) - abdomen (3) - antenna, (4)-terminalia male



Sources: From Walker (1853) and https://www.delta-intkey.com/britin/dip/www/megameri.htm

Figure 27 *Megamerina dolium* (Fabricius, 1805). *M. dolium*, with head viewed from the side (4a), and antenna (4b)

4.2. Biology

Regarding its biology, the larvae of this species live under the bark of dying or dead deciduous trees where they are predatory or necrophagous of the larvae of other insects. The Adults, which are rarely caught, are found mainly in wooded areas, near the habitat of the larvae, mostly on trunks and leaves (Figure 28) [15,16,17].

Source: https://sciendo.com/pdf/10.2478/v10210-011-0018-3

Figure 28 *Megamerina dolium* (Fabricius, 1805) and its habitat: (a) the female collected in Portugal; (b) Habitat where this specimen was collected

4.3. Taxonomy

Currently, 13 species of Megamerinidae are known to worldwide, of which only 2 have been cited from the region Palearctic and, as already mentioned, 1 from Europe, where is widely distributed. However, regarding the Iberian Peninsula, this family was known only from Andorra [18].

Genus: *Megamerina* and *Protexara* (Figure 29).

Source: https://ukrbin.com/show_image.php?imageid=46775

Figure 29 Genus *Megamerina*

Only one species is known from Europe, *Megamerina dolium* (Fabricius, 1805) Spain: Barcelona: Cabrils (Can Tolrá), male. The only specimen cited was captured thanks to a Malaise trap that was set by the author next to a pool surrounded by dense forest vegetation. The specimen has been identified by the author and is preserved in 70º alcohol in his private collection (Figure 30) [19].

Source: http://v3.boldsystems.org/index.php/Taxbrowser_Taxonpage?taxid=677267

Figure 30 Genus *Protexara*

5. Studies carried out and selected

5.1. Study 1

5.1.1. Megamerinidae in Portugal.

Megamerina dolium (Fabricius) [20].

Megamerina dolium Portugal: Porto, Vila Nova de Gaia, Avintes, Parque Biológico de Gaia, collected with a vial while moving on a leaf of *Rubus* sp. in a shaded area. The specimen is preserved in 70% ethanol and deposited in the author's private collection. The site where the specimen was found is situated in the valley of the river Febros, a tributary of the river Douro, and is included within the area of Parque Biológico de Gaia, an urban park and an environmental education center created in 1983 that occupies an area of 35 hectares [22].

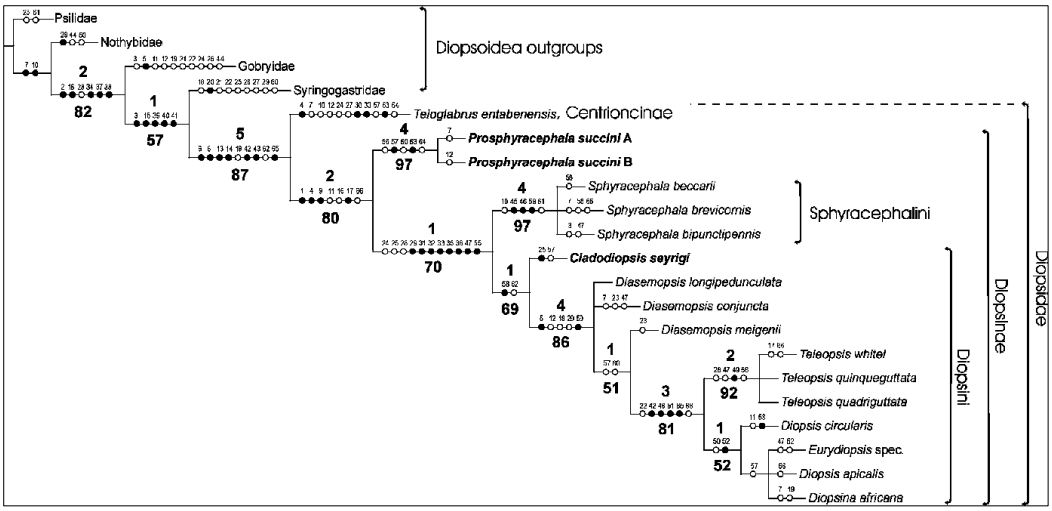
An important portion of the park is forested, with the following predominant tree and herb species: *Quercus robur* L., (Fagaceae), *Alnus glutinosa* (L.) (Betulaceae), *Salix* spp., *Betula* sp., *Castanea sativa* Mill. (Fagaceae), *Sambucus nigra* L., (Adoxaceae), *Urtica* sp. and *Rubus* sp. Given the biology of the species, it is worth keeping decomposing tree and dying trees wherever they lie, so that the species can complete its life cycle and survive in the area successfully. Of course, this would significantly aid also the survival of many more saproxylic insects (Figure 31) [23].

Source: Darwyn Sumner

Figure 31 *Megamerina dolium* (Fabricius, 1805): Have observed this species on a couple of occasions. At first glance it appears and moves like a dark ichneumon, weaving through brambles and not resting long. Locating likely ovipositing sites and staking those out might prove a good tactic

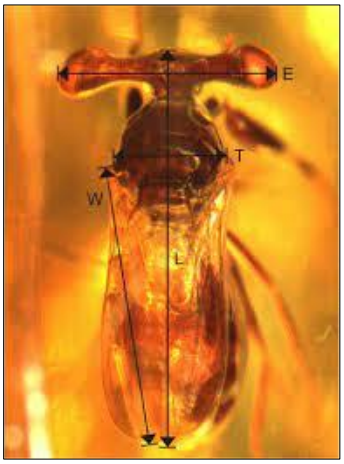
5.2. Study 2

Diopsoidea superfamily: The Megamerinidae are a family of flies (Diptera) with approximately 11 species in three genera Palearctic. They are small and marked by an elongated abdomen with basal constriction. The family has been variously placed in the past within the superfamilies Diopsoidea, Nerioidea, and more recently in the Opomyzoidea, but evolutionary relationships remain unclear (Figures 32-33) [24].



Source: https://www.semanticscholar.org/paper/Baltic-amber-fossils-reveal-early-evolution-of-in-Kotrba/cdf042840dd18129279f4fdcb909a9c5a390a01f/figure/5

Figure 32 Phylogeny based (Diopsoidea) on cladistic analysis of 65 morphological characters (strict consensus from 16 most parsimonious trees, length=136; CI=0,66; RI=0,76; slow optimization). Solid circles: unique character changes, open circles: homoplastic changes, character numbers above circles; Bremer support values given above the branches, bootstrap values 450% below



Source: https://www.semanticscholar.org/paper/Baltic-amber-fossils-reveal-early-evolution-of-in-Kotrba/cdf042840dd18129279f4fdcb909a9c5a390a01f/figure/5

Figure 33 Diopsoidea in Baltic amber (Natural History Museum Krakow). Arrows indicate the measurement of eye span (E), body length (L), thorax width (T), and wing length (W)

The biology of immature Megamerinidae is poorly understood, but the larvae living under bark or decaying vegetation. Regarding their biology, the larvae of this species live under the bark of dying or dead deciduous trees where they are predatory or necrophagous of the larvae of other insects. The Adults, which are rarely taken, are found mainly on trunks and leaves. Currently, 13 species of Megamerinidae are known to worldwide, of which only 2 have been cited from the region Palearctic and, as already mentioned, 1 from Europe, where is widely distributed. However, as to the Iberian Peninsula, this family was known only from Andorra. Therefore, the species that mentioned below represents the first date of this family for Spain [25].

Megamerina dolium (Fabricius, 1805) Spain.

The only specimen that is cited was captured thanks to a trap Malaise that was installed by the author next to a swimming pool surrounded by dense forest vegetation. The specimen has been identified by the author and is preserved in 70º alcohol in his collection in particular. Family, genus and species were new to Spain [25].

5.3. Study 3

The Zoological Survey of India identified 37 families in Maharashtra state. Platypezidae, Pyrgotidae, Rhagionidae, Scatopsidae, Sciomyzidae, Sciaridae, Therevidae and Ulidiidae, bringing the number of Diptera families from Maharashtra to 55 [26,27].

Of the 64 families identified in the Western Ghats, considerable diversity (78%, n = 50) was recorded in the MMR. Two of the 11 unrecorded families from the Western Ghats (Ghorpade 2011), Neriidae and Megamerinidae, were identified in this study, bringing the diversity of Diptera from WG to 66 families [26,27].

Families with the lowest species diversity in the world (<200 species) found in the MMR were Megamerinidae (about 15 species worldwide), Neriidae (about 120 species worldwide), Celyphidae (about 120 species worldwide world) and Diopsidae with less than 200 species worldwide. Eighteen Therevidae species (>1,000 species worldwide) have been recorded from India, of which only one species has been documented from Maharashtra [26,27].

6. Family Nemestrinidae

Source: http://www.entomologia.cl/TricophthalmaSp1.html

Figure 28 Nemestrinidae Family *Trichophtalma* sp.: This is one of several species of Nemestrinidae. They are very common during spring and summertime in the Central and Southern areas of the country. There are many pretty species in Chile. One of the most striking features of this family is the ability to stop in the air, as for looking where else to go

Source: http://www.ispot.org.za

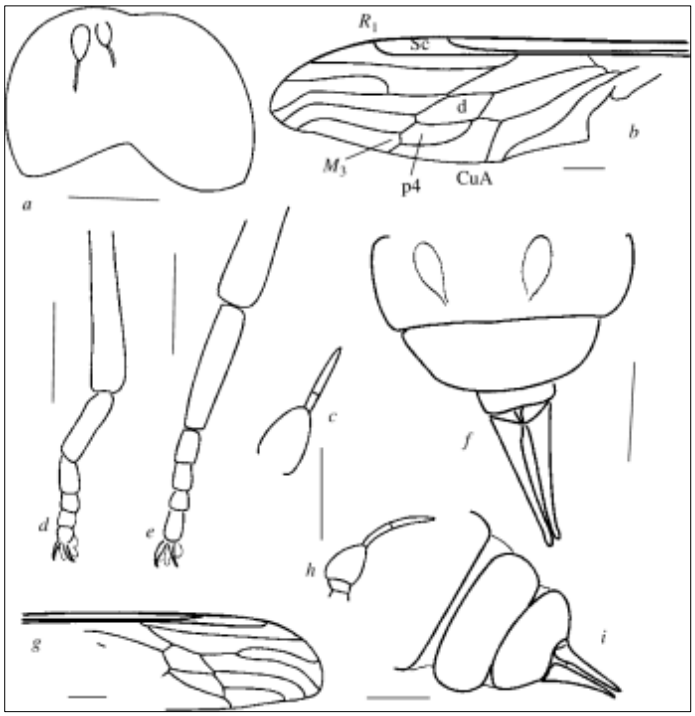
Figure 29 Nemestrinidae: The irises in the photo above seem to have landing strips that help guide the fly in. Work conducted by Dennis Hansen when he was at the University of Kwazulu-Natal discovered that if you paint over these strips, the flies cannot find the nectaries at the base of the tube and many people have studied these groups, as they are amazing examples of co-evolution with very good models correlating corolla (the petals of the flower) length and proboscis

The Nemestrinidae (tangle-veined flies) are a widespread family of moderate to large-sized rather stout and compact files. This family is represented by about 300 species and 26 genera worldwide, including more than 80 species from

the Palaearctic region and more than 50 species from the Afrotropical region. They are widespread throughout the globe, but there are some areas of higher concentration: from the Mediterranean to Turkistan, eastern Australia, southern Africa, Chile and Argentina (Figures 28-29) [28,29].

6.1. Description

These adults can be easily distinguished by their wings which are typically longer than the body, with veins appearing tangled, with a composite diagonal vein commencing from vein R1 and traversing diagonally the two outer wings margins, and with characteristic apical veins running parallel to the hind margin of the wing and terminating anterior to its apex (Figures 31-36) [30,31,32].



Source: https://www.semanticscholar.org/paper/A-Revision-of-the-Nemestrinid-Flies-(Diptera%2C-by-a-Mostovski/2a81d9a517e31a39595288b65bb73e28b3eebf06

Figure 31 Species of the genus *Archinemestrius*: (a) *Archinemestrius karatavicus* Rohdendorf, head; (b–f) *Archinemestrius mimas* sp. nov., holotype PIN, no. 2997/3496, female: (b) wing, (c) antenna, (d) mid leg, (e) hind

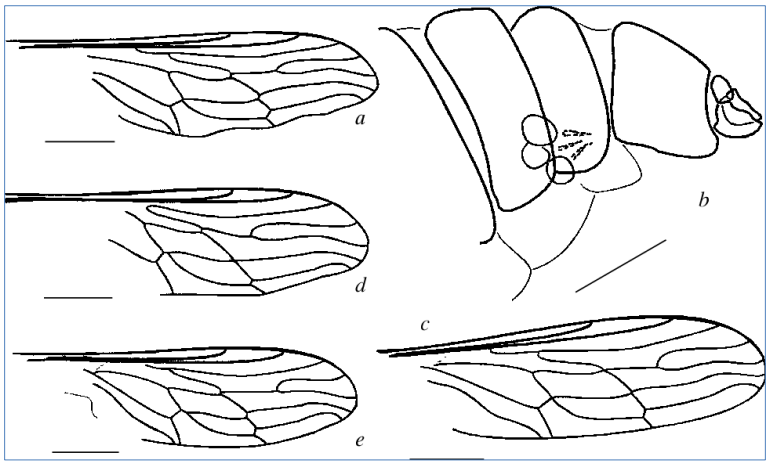
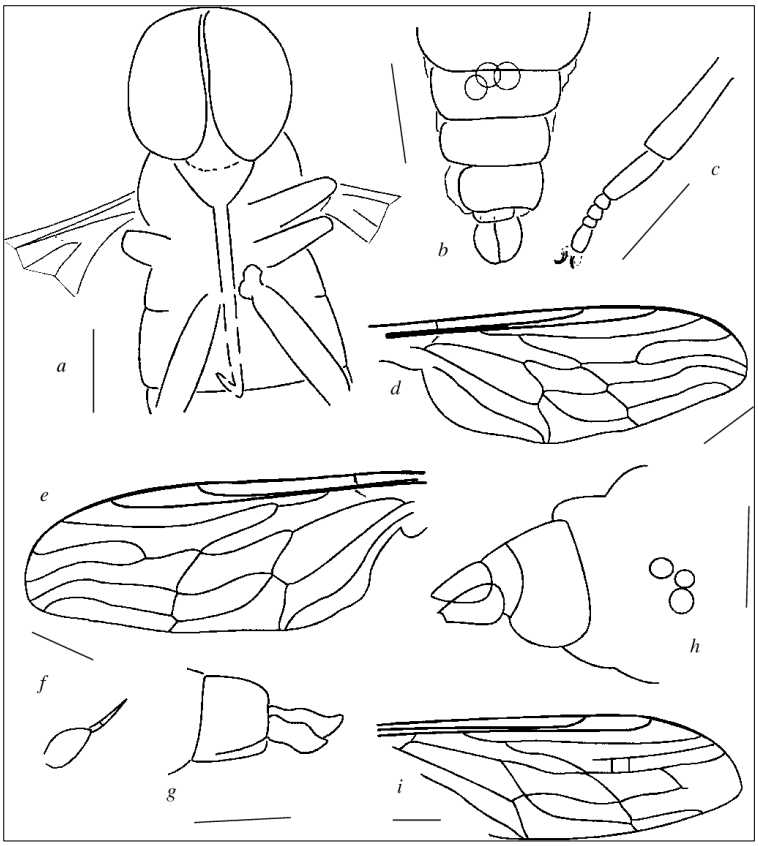
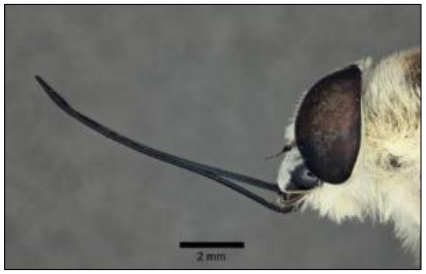


Figure 32 Species of the genus *Protonemestrius*: (a–c) *Protonemestrius martynovi* Rohdendorf, 1968: (a) holotype, wing; (b) specimen, abdominal apex; (c) wing of same; (d) *Protonemestrius bequaerti* Rohdendorf, 1968 holotype, wing; (e) *Protonemestrius handlirschi* Rohdendorf, 1968, wing



Source: https://www.semanticscholar.org/paper/A-Revision-of-the-Nemestrinid-Flies-(Diptera%2C-by-a-Mostovski/2a81d9a517e31a39595288b65bb73e28b3eebf06

Figure 33 New species of upper Jurassic nemestrinids: (a–d) *Protonemestrius rohdendorfi* sp. nov., holotype (a) forebody, (b) abdominal apex, (c) apex of metatibia with tarsus, (d) wing; (e–g) *Protonemestrius rasnitsyni* sp. nov., holotype: (e) wing, (f) antenna, (g) abdominal apex; (h–i) *Aenigmestrinus mirabilis* sp. nov., holotype: (h) abdominal apex, (i) wing



Source: https://www.nhm.ac.uk/natureplus/blogs/diptera-blog/tags/nemestrinidae.html

Figure 34 Their mouthparts. medium proboscis. Hey, vary from rudimentary, reduced, short, medium and long - in some instances very long. In fact, one species of Nemestrininae called *Moegistrorhynchus longirostris* (Grantsau, 1805) has the longest mouthpart in relation to body size of any insect

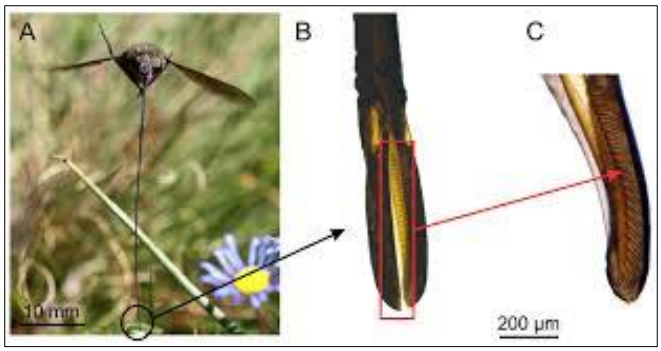
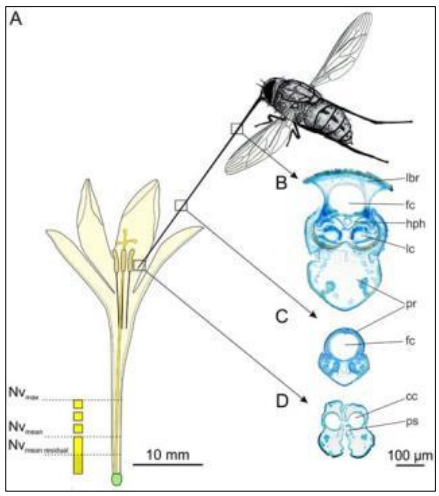


Figure 35 The long-proboscid fly *Prosoeca marinusi* Barraclough sp. nov. (Nemestrinidae) and morphology of the proboscis tip; (A) Proboscis in feeding position; (B) Proboscis tip formed by pair of labella; (C) Median view of labellum shows pseudotrachea canals for nectar uptake through apical end of proboscis



Sources: https://www.mdpi.com/2075-4450/12/4/371 and (https://www.geogebra.org/m/se2cwx9s, accessed on 16 April 2021)

Figure 36 Nectar accessibility for *Prosoeca marinusi* Barraclough, 2018 from (A) an average *Babiana vanzijliae* F. Bolus (Iridaceae) flower. Nectar heights calculated using the Geo Gebra tool from measured quantities, modeled with a mean inner diameter and average tube length (n = 41). Maximal nectar volume (Nvmax) resulted in a nectar height of approximately up to half-length. Mean nectar volume before fly activity (Nvmean, n = 44) fills nectar tube up to 20% of tube length. The mean residual nectar volume after the fly visited (Nvresidual, n = 39) in the proximal was 10%. (B) Cross section of the proximal proboscis. (C) Proboscis cross-section of the distal proboscis that is composed only of the premium. (D) Cross section of the labella at the tip of the proboscis. cc: collecting canal; fc: food canal; hph: hypopharynx; lbr: labrum; lc: lacinia; pr: prementum; ps: pseudotrachea

6.2. Biology

All known larvae of Nemestrinidae are internal parasitoids of nymphs and adults of locusts and beetle larvae. This makes the nemestrinids have the potential to be used as biological control agents for Mantodea. Nemestrinid adults are often found hovering around flowers where they feed on nectar, emitting a characteristic buzzing sound. (Figures 37-38) [33,34,35,36].

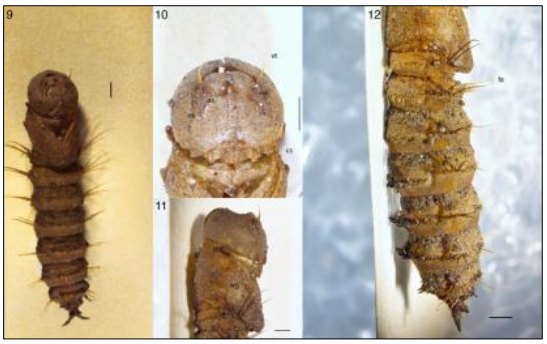


Figure 37 Pupa of *Hirmoneuropsis luctuosa* (Philippi, 1865). (9). Lateral view; (10). Head in frontal view (vt, vertical tubercle; as, antennal sheaths; es, epicranial suture; cs, cephalothorax suture); (11). Head and thorax in lateral view (ths, thoracic spiracle; wg, wing); (12). Abdomen in lateral view (fs, fringe spines)

Sources: https://www.brisbaneinsects.com/brisbane_Tabanoidea/Nemestrinidae.htm and https://www.brisbaneinsects.com/brisbane_Tabanoidea/Nemestrinidae.htm

Figure 38 Nemestrinidae spatial flay

6.3. Taxonomy

The family Nemestrinidae (Diptera, Brachycera, Asiloidea). The family Nemestrinidae is divided into 3 subfamilies: Hirmoneurinae, Nemestrininae and Trichopsideinae (Figures 39-41) [38, 39,40,41].

Sources: Photo 106863817, (c) Brin Steven, some rights reserved (CC BY-NC), uploaded by Brian Steven and https://www.inaturalist.org/photos/106863817

Figure 39 Subfamily Hirmoneurinae

Source: https://v3.boldsystems.org/index.php/TaxBrowser_Taxonpage?taxid=83217

Figure 40 Subfamily Nemestrininae

Sources: Photo 127368530, (c) Candi Welliver, all rights reserved, uploaded by Candi Welliver and https://guatemala.inaturalist.org/taxa/176715-Neorhynchocephalus

Figure 41 Subfamily Trichopsideinae

Some Genus: *Archinemestrius* Rohdendorf, 1968, *Eohirmoneura* Rohdendorf, 1968, *Florinemestrius* Ren, 1998, *Hyrmophlaeba* Rondani, 1864, *Neorhynchocephalus* Lichtwardt, 1909, *Palembolus* Scudder, 1878, *Prohirmoneura* Handlirsch, 1906, *Prohirmoneura* Handlirsch, 1906, *Trichogionemestrius* Westwood 1839 (Figures 42-49) [42,43.44].

Source: https://zookeys.pensoft.net/article/70743/element/7/0/Nemestrinus/

Figure 42 Genus *Nemestrinus*, male type (A) dorsal view (B) lateral view (C) frontal view (D) ventral view (E) labels (Zmhb)

Source: https://www.inaturalist.org/taxa/921513-Hirmoneura-obscura/browse_photos

Figure 43 Genus *Hirmoneura*

Source: https://v3.boldsystems.org/index.php/Taxbrowser_Taxonpage?taxid=83218

Figure 44 Genus *Trichophthalma*

Source: https://www.biodiversidadvirtual.org/insectarium/Neorhynchocephalus-tauscheri-%28Fischer-1812%29-img489218.html

Figure 45 Genus *Neorhynchocephalus*

Source: https://eol.org/pages/79145

Figure 46 Genus *Prosoeca*

Source: https://www.mapress.com/zt/article/view/zootaxa.5196.1.8

Figure 47 Genus *Atriadops*

Source: https://www.biodiversidadvirtual.org/insectarium/Nemestrinus-perezii-%28Dufour-1850%29-img561155.html and https://www.inaturalist.org/taxa/902026

Figure 48 Genus *Nemestrinus*

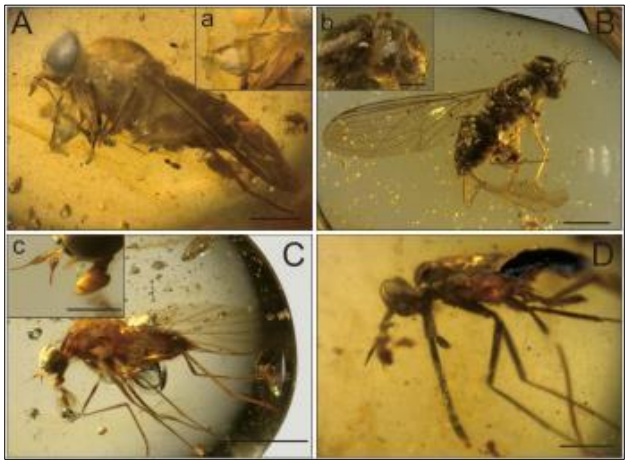
Sources: Photo 21949231, (c) Scott Simmons, all rights reserved, uploaded by Scott Simmons and https://guatemala.inaturalist.org/taxa/176715-Neorhynchocephalus

Figure 49 Genus *Neorhynchocephalus*

Nemestrininae genera are geographically segregated in South America and Australia (genus *Trichophthalma*), southern Africa (*Prosoeca* and *Stenobasipteron*) and the Palearctic region (*Nemestrinus* and *Stenopteromyia*, with a branch that, later, reached Africa, giving rise to *Moegistorhynchus*. In South America, several species of Nemestrinidae belonging to the subfamilies Hirmoneurinae (represented by the genus *Hirmoneura*) and Trichopsideinae (*Neorhynchocephalus* spp.) have also been described (Figures 50-51) [45,46].

Source: https://en.wikipedia.org/wiki/Moegistorhynchus

Figure 50 Genus *Moegistorhynchus*



Source: https://www.frontiersin.org/articles/10.3389/fpls.2017.00631/full

Figure 51 Fossil Source: Four types of mouthparts in mid-Cretaceous Burmese amber. (A) Tabanidae, (a) Mouthparts, (B) Nemestrinidae (b) Mouthparts (C) Bombyliidae (c) mouthparts (D) Zhangsolvidae with a long proboscid

7. Selected studies

7.1. Study 1

Trichophthalma occurs in Australia and South America and is the only case in the family of a genus with such a disjunct (typically Gondwanian) distribution. This genus has 45 Australian species and 21 species described for southern South America (Argentina, Chile and Uruguay) (Figure 52) [47,48,49]

Source: https://www.flickr.com/photos/insectcollection/4152531738/

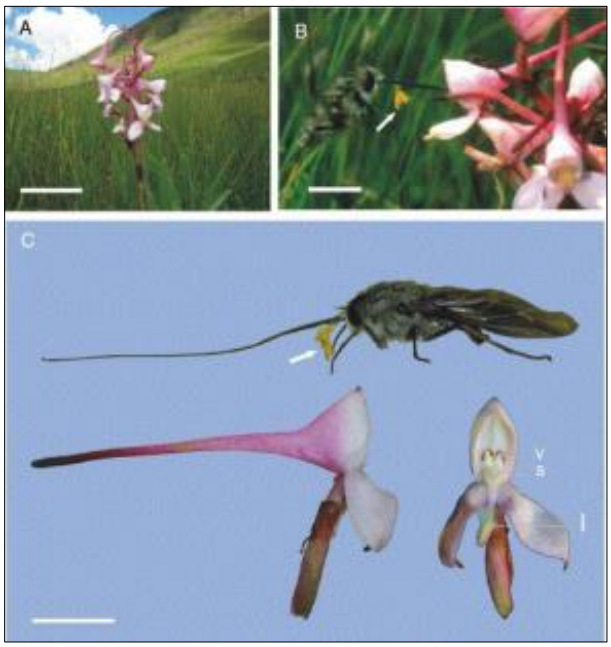
Figure 52 Genus *Trichophthalma*

In spite of its primitiveness, *Trichophthalma* shows a rather specialized character (shared by all Nemestrininae): the proboscis is always well developed and it can range between 1.4 to 7times the height of the head (as measured by being sometimes longer than the whole insect, as in *Trichophthalma scalaris* Bigot 1881, of Chile. Species with long proboscides are able to reach nectar hidden in deep flower tubes, which has led to remarkably specialized mutualisms (Figure 53) [50,51,52].

Source: https://commons.wikimedia.org/wiki/Category:Trichophthalma

Figure 53 *Trichophthalma scalaris* Bigot 1881

Trichophthalma is a diverse genus within the temperate forests of southern South America. These forests extend over a narrow strip from ca. 35 to 55º S latitude over much of southern Chile and the eastern slope of the Patagonian Andes in Argentina (Figures 54-55) [53,54].



Source: https://www.researchgate.net/profile/Klaus-Lunau/publication/255923738/figure/fig5/AS:669430820401155@1536616153128/Prosoeca-ganglbaueri-visiting-the-flowers-of-Zalusianskya-microsiphon-Scrophulariaceae.ppm

Figure 54 Inflorescence of *Disa scullyi* Bolus 1885 (Orchidaceae) in grassland habitat in the Cape Drakensberg mountains. Scale bar (applicable to inflorescence): 50 mm; (B) long-proboscid fly *Prosoeca ganglbaueri* Lichtwardt, 1910 probing a flower of *Disa scullyi* Bolus 1885. *Pollinaria* (arrowed) are visible on its proboscis. (C) *P. ganglbaueri* positioned next to flowers of *D. scullyi*. *Pollinaria* (arrowed) are attached to the proboscis. The two entrances to the galea are visible in the front view of a *D. scullyi* flower. Abbreviations: v = viscidium, s = stigma, l = lip. Mutualisms of Nemestridae

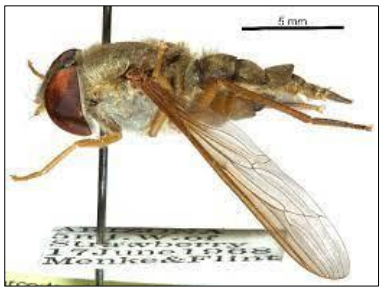
Source: https://www.researchgate.net/figure/Prosoeca-ganglbaueri-visiting-the-flowers-of-Zalusianskya-microsiphon-Scrophulariaceae_fig5_255923738

Figure 55 *Prosoeca ganglbaueri* Lichtwardt, 1910 visiting the flowers of *Zalusianskya microsiphon* Sibthorpii. (Scrophulariaceae). abb. 6*: P. ganglbaueri* and Z. *microsiphon*

In this region, there is a strong rainfall gradient which encompasses a striking shift in vegetation in less than 150 km in an east-west direction from xeric desert shrubland to grass shrub-steppe, leading to a low stature tree cover and finally closed canopy forest. As part of a community-level survey in Patagonia (Argentina), we focused on the diversity and distribution of flower-visiting Nemestrinidae and their floral specificity along the rainfall gradient mentioned above [55,56].

7.2. Study 2

Adults (Nemestrinidae) frequently visit flowers, feeding upon the nectar. Nemestrinid larvae are internal parasitoids of other insects such as Orthoptera, Coleoptera and Mantodea. The species of *Hirmoneuropsis* Bequaert develop in larvae of scarabaeid beetles (Figure 56) [57].



Source: https://v3.boldsystems.org/index.php/Taxbrowser_Taxonpage?taxid=82585

Figure 56 Genus *Hirmoneuropsis*

Knowledge of the immatures of Neotropical species is poor immatures only of four species are known *Neorhynchocephalus sackenii* (Williston, 1880) *Neorhynchocephalus sulphureus* (Wiedemann, 1830), the pupal skin of *Hirmoneuropsis articulata* (Philippi, 1820), and the larvae of Falleniinae, which form a respiratory tube that tube is lacking in the larva of *Hirmoneura obscura* (Wiedemann in Meigen, 1820) (Figures 57-58) [58].

Source: https://en.wikipedia.org/wiki/Neorhynchocephalus_sackenii

Figure 57 *Neorhynchocephalus sackenii* (Williston, 1880)

Source: https://www.researchgate.net/figure/Left-wing-of-Neorhynchocephalus-sulphureus_fig1_279190167

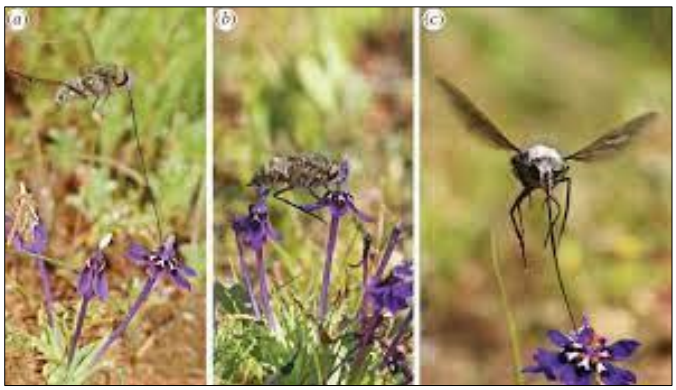
Figure 58 *Neorhynchocephalus sulphureus* (Wiedemann, 1830)

7.3. Study 3

The idea that a syndrome of floral traits predicts pollination by a particular functional group of pollinators remains both controversial and widely used because it allows plants to be quickly assigned to pollinators. Testing the idea requires demonstrating that there is an association between floral traits and pollinator type.

A test was carried out in the Cape Floristic Region, South Africa, studying the pollination of eight plant species from six spring-blooming families that have odorless, actinomorphic, upward-facing flowers with orbicular petals all in the same plane. The petals are bright white with purple-red nectar guides. The tubes are short and contain small volumes of concentrated nectar, except for *Disa fasciata* Lindl. 1838 (Orchidaceae). Pollinators were photographed and captured, pollen loads were analyzed and pollination networks were constructed [59].

Consistent with the pollination syndrome hypothesis, species with the defined syndrome shared a small group of pollinators. The most frequent pollinators belonged to a clade of four species of entangled flies with relatively short proboscis *Prosoeca*, while functionally similar Bombyliidae and Tabanidae played minor roles. Among the four species of *Prosoeca*, only *Prosoeca westermanni* (Wiedemann 1821) was described, a result that shows our lack of knowledge about pollinators (Figures 59-60) [59].



Source: https://www.researchgate.net/figure/The-long-proboscid-nemestrinid-fly-Prosoeca-sp-nov-foraging-at-flowers-of-the-iris_fig1_51526831

Figure 59 The long-proboscid nemestrinid fly *Prosoeca* sp. nov. foraging at flowers of the iris *Lapeirousia oreogena* Schltr. ex Goldblat (Iridaceae). (a) Hovering fly navigating its proboscis towards the narrow perianth tube containing the nectar. (b) Feeding fly with proboscis fully inserted (partly visible through the thin-walled tube), pressing its head against the anthers and stigma of the flower. (c) Departure from a flower with pink UV-fluorescent dye applied to anthers; the fly has picked up dye grains on its fronts

Source: https://www.mindat.org/paleoimg.php?id=887846

Figure 60 figs. 1–4. Photographs of live adult *Prosoeca marinusi* Barraclough sp. nov. in the field in the Nieuwoudtville area, Northern Cape Province. (1). Approaching a flower of *Lapeirousia oreogena* (Schltr. ex Goldblatt (Iridaceae) with elongate proboscis in the feeding position. (2). Taking up nectar from the long, slender flower spur of *L. oreogena*. (3). Visiting a flower of *Babiana framesii* L. (Iridaceae); note the dense covering of pollen on the thorax and head. (4). Basking on a rock with the proboscis in the resting position

8. Conclusion

The larvae of Trevisidae can be collected by straining the material where they are found and can be raised in the sand by feeding them beetle larvae. They usually occur in dry areas but they can also be found in mountains and tropical humid regions. Larvae feed on other insects, mainly beetles, butterflies, moths and other flies that live in leaf litter, tree holes, or sandy terrains. Regarding the biology of the Megamerinidae Family, their larvae live under the bark of dying or dead deciduous trees where they are predators or scavengers of the larvae of other insects. Adults, which are rarely captured, are found mainly in wooded areas, close to the larval habitat, mainly on trunks and leaves. All known larvae of Nemestrinidae are internal parasitoids of nymphs and adults of locusts and beetle larvae. This makes the nemestrinids have the potential to be used as biological control agents for Mantodea. Nemestrinid adults are often found hovering around flowers where they feed on nectar, emitting a characteristic buzzing sound.

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