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# **Research** article

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# An overlooked case for a century: taxonomy and systematics of a new Iberian species of *Eumerus* Meigen, 1822 (Diptera, Syrphidae)

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Abstract. The hoverfly genus Eumerus Meigen, 1822 (Eristalinae: Merodontini) comprises 250+ described species, of which 36 are reported from the Iberian Peninsula. The high species diversity linked to the low degree of morphological differentiation between some species, which is even lower in females, leads to a high taxonomic complexity in this genus. The aim of this work is to confirm the morphological and molecular validity of an undescribed species of *Eumerus*, which is widespread in the Iberian Peninsula. The new species is described and compared with similar species. The genitalia of the new species are similar to those of *Eumerus clavatus* Becker, 1923 and *Eumerus uncipes* Rondani, 1850, but also share some features with Eumerus nudus Loew, 1848. The COI-5' barcode is provided for the new taxon and analysed together with those of other named *Eumerus* sequences/species publicly available online. In the light of the morphology and barcoding data, the systematic position of the new species is discussed.

Keywords. DNA, Eristalinae, taxonomy, Spain, Portugal.

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# Introduction

The hoverfly genus Eumerus Meigen, 1822 (Eristalinae: Merodontini) is widely distributed in the Palaearctic, Afrotropical, Oriental and Australasian Regions (Grković et al. 2015; van Steenis et al. 2017). Some species were introduced in other Regions by man, probably inside of plant or plant parts (Pérez-Bañón & Marcos-García 1998), and cause agriculture and horticulture damages (Souba-Dols et al. 2020). Nowadays, the following species are introduced into the New World (Marinoni & Morales 2007; Speight et al. 2013; Garcete-Barrett et al. 2020): Eumerus aurifrons (Wiedemann, 1824), Eumerus figurans Walker, 1859, Eumerus funeralis Meigen, 1822, Eumerus narcissi Smith, 1928, Eumerus obliquus (Fabricius, 1805) and Eumerus strigatus (Fallén, 1817). Within the Palaearctic Region, Eumerus represents one of the most speciose hoverfly genera with more than 160 species confirmed (i.e., Doczkal 1996; Grković et al. 2015; Markov et al. 2016; Ricarte et al. 2017). Although there are some regional identification keys (e.g., Speight et al. 2021), the absence of an updated key to all Palaearctic species of *Eumerus* makes the morphological identification at the species level difficult in this region. In relatively recent times, molecular techniques have proven useful in the species classification of *Eumerus*. Chroni et al. (2017) were the first in generating a phylogeny of Eumerus using molecular markers and proposed a classification into species groups based on their phylogenetical results. At present, ten species groups have been defined based on both molecular and morphological data: (1) Eumerus barbarus group (as Eumerus sulcitibius group in Chroni et al. 2017; van Steenis et al. 2017), (2) Eumerus basalis group (Chroni et al. 2017), (3) Eumerus clavatus group (Grković et al. 2017), (4) Eumerus minotaurus group (Chroni et al. 2017, 2018), (5) Eumerus olivaceus group (Grković 2018), (6) Eumerus obliquus group (Smit et al. 2017; Ricarte et al. 2020), (7) Eumerus ornatus group (Chroni et al. 2017), (8) Eumerus pulchellus group (Chroni et al. 2017), (9) Eumerus strigatus group with the E. bactrianus subgroup (Chroni et al. 2017; Grković et al. 2017, 2019b) and (10) Eumerus tricolor group with E. binominatus subgroup (Chroni et al. 2017; Grković et al. 2017, 2019a).

The most recent checklist of Iberian hoverflies included 31 species of *Eumerus* from mainland Spain plus the Balearic Islands (Ricarte & Marcos-García 2017), of which only one was endemic to Spain, *Eumerus hispanicus* van der Goot, 1966. After 2017, four new species endemic to the Iberian Peninsula have been described: *Eumerus azabense* Ricarte & Marcos-García in Ricarte *et al.* (2018), *Eumerus bifurcatus* van Steenis & Hauser in Grković *et al.* (2019b), *Eumerus gibbosus* van Steenis, Hauser & van Zuijen, 2017 and *Eumerus grallator* Smit in Grković *et al.* (2019a). Moreover, the status of *Eumerus truncatus* Rondani, 1868 has recently been clarified and it has been reported, for the first time, from Spain as well (van Steenis *et al.* 2017). The above-mentioned recent discoveries of *Eumerus suggest* that the diversity and distribution of this genus in the Iberian Peninsula is still far from being fully understood.

In the framework of an ongoing revision of the *Eumerus* diversity in the Iberian Peninsula, the aim of this work is to describe a new Iberian species and explore its systematic position based on morphological and molecular evidence.

# Material and methods

# Material examined

The specimens of the new species examined (79 specimens:  $36 \ 3 \ 3 \ 9 \ 9$ ) have been collected during a wide time span, from 1908 to 2021, at different Iberian localities (one specimen from Portugal) and by various collectors. Fresh specimens (caught over the last three years) were collected with hand net and killed by freezing. Additionally, specimens of similar species were examined as well. Examined material is now deposited in the following collections:

CEUA-CIBIO	=	Colección Entomológica de la Universidad de Alicante, Research Institute
		CIBIO, University of Alicante, Spain

- MNCN = Museo Nacional de Ciencias Naturales, Madrid, Spain
- CSCA = California Department of Food and Agriculture, Sacramento, USA

Coll. A. v. Eck = Private collection of André van Eck

All specimens are databased in an internal excel table at CIBIO. A unique bar code label was assigned to each of the specimens belonging to the CEUA-CIBIO collection. A map representing the distribution of the examined specimens was produced with the software QGIS (QGIS Development Team 2020) (Fig. 1).

# **Morphological study**

An initial diagnosis of the new species with a morphological description of the male holotype and the female are given. Then, phenotypic variation, species distribution and biology of the new species are indicated. To complete the new species description, the following measures were taken with the software Leica Application Suite X (LAS X) ® ver. 3.0.4.16529: Body length (l), from the base of the antenna to the posterior margin of tergum IV (Fig. 4A); body width (w), from tegula to tegula; head height (hh), from the top of the ocellar triangle to gena base (Fig. 4C); head width (hw), from one eye external side to the other (Fig. 4D); basoflagellomere ratio (length: width), of which length (bl) is measured from the pedicel apex to the apical margin of the basoflagellomere and width (bw) from the apical apex of the wing (Fig. 4B). Male genitalia were dissected and prepared following Ricarte *et al.* (2012). Photographs of the holotype, one female paratype and male genitalia were taken with a Leica DFC 450 camera attached to a Leica M205 C binocular microscope. Male genitalia were hand drawn from printed photographs and stored in glycerine in plastic microvials. Morphological terminology used in the adult description follows Thompson (1999), except for terms "fossette" and "notopleural sulcus" which follow Dozckal & Pape (2009), and terminology for male genitalia follows Doczkal (1996).



Fig. 1. Distribution map of *E. colladoi* sp. nov. in the Iberian Peninsula.

Species	DNA voucher	Country of origin	Acc. number (Genbank)	Specimen ID (BOLD)	Reference
<i>Eumerus alpinus</i> Rondani, 1857	G1147	Italy	KX083349	_	Grković <i>et al.</i> (2017)
E. amoenus Loew, 1848	FSUNS:E1260	Greece	KY865462	_	Chroni <i>et al.</i> (2018)
<i>E. argyropus</i> Loew, 1848	22	Macedonia	MG559830	_	Chroni <i>et al.</i> (2019)
<i>E. armatus</i> Ricarte & Rotheray in Ricarte <i>et al.</i> (2012)	FSUNS:G1020	Greece	KY865456	-	Chroni <i>et al.</i> (2018)
<i>E. aurofinis</i> Grković, Vujić & Radenković in Grković <i>et al.</i> (2015)	6800	Turkey	KT221012	GBDP31534-19	Grković <i>et al.</i> (2015)
<i>E. azabense</i> Ricarte & Marcos-García in Ricarte <i>et al.</i> (2018)	CEUA00106286	Spain	MG604936	-	Ricarte <i>et al.</i> (2018)
<i>E. banaticus</i> Nedeljković, Grković & Vujić in Grković <i>et al.</i> (2019b)	FSUNS_15558	Romania	MK059990	-	Grković <i>et al.</i> (2019b)
<i>E. barbarus</i> (Coquebert, 1804)	CNC: Dipte- ra:102077	France	JN991987	_	Penney <i>et al.</i> (2012)
<i>E. basalis</i> Loew, 1848	FSUNS:G0992	Greece	KY865448	_	Chroni <i>et al.</i> (2018)
<i>E. bicornis</i> Grković, Vujić & Hayat in Grković <i>et al.</i> (2019b)	FSUNS_16045	Greece	MK060002	_	Grković <i>et al.</i> (2019b)
<i>E. bifurcatus</i> van Steenis & Hauser in Grković <i>et al.</i> (2019b)	PSJA_TS445	Spain	MK060003	_	Grković <i>et al.</i> (2019b)
<i>E. clavatus</i> Becker, 1921	E1327	Serbia	MG559951	_	Chroni <i>et al.</i> (2019)
<i>E. colladoi</i> Ricarte & Aguado-Aranda sp. nov.	CEUA_S01	Spain	OM791818	-	Present work
<i>E. colladoi</i> Ricarte & Aguado-Aranda sp. nov.	CEUA_S03	Spain	OM791819	_	Present work
<i>E. colladoi</i> Ricarte & Aguado-Aranda sp. nov.	CEUA_S04	Spain	OM791820	_	Present work

**Table 1** (continued on next two pages). List of the species used in the molecular analyses. New sequences generated for this work are highlighted in bold.

Species	DNA voucher	Country of origin	Acc. number (Genbank)	Specimen ID (BOLD)	Reference
<i>E. colladoi</i> Ricarte & Aguado-Aranda sp. nov.	CEUA_S06	Spain	OM791821	-	Present work
<i>E. colladoi</i> Ricarte & Aguado-Aranda sp. nov.	CEUA_S52	Spain	OM791822	_	Present work
<i>E. consimilis</i> Šimić & Vujić, 1996	FSUNS_11489	Montenegro	MK060004	_	Grković <i>et al.</i> (2019b)
<i>E. crassus</i> Grković, Vujić & Radenković in Grković <i>et al.</i> (2015)	FSUNS:G3025	Greece	KY865452	_	Chroni <i>et al.</i> (2018)
<i>E. erythrocerus</i> Loew, 1858	FSUNS:EU104	South Africa	MH550072	_	Vujić <i>et al.</i> (2019)
E. feae Bezzi, 1912	415E04	Africa (no further details)	KT624230	_	Jordaens <i>et al.</i> (2015)
<i>E. flavitarsis</i> Zetterstedt, 1843	FSUNS:E164	Russia	LT882603	-	Vujić <i>et al.</i> (2018)
<i>E. funeralis</i> Meigen, 1822	BIOUG<- CAN>:08TT- ML-0912	Canada	HQ581894	_	Ratnasingham & Hebert (2007)
<i>E. grandis</i> Meigen, 1822	FSUNS:G1919	Serbia	MN401271	-	Likov <i>et al.</i> (2020)
<i>E. karyates</i> Chroni, Grković & Vujić in Chroni <i>et al.</i> (2018)	FSUNS:06557	Greece	KY865458	_	Chroni <i>et al.</i> (2018)
<i>E. lyneborgi</i> Ricarte & Hauser in Ricarte <i>et al.</i> (2020)	CSCA 19G345	Namibia	MN717174	_	Ricarte <i>et al.</i> (2020)
<i>E. macquarti</i> Ferguson, 1926	CNC:Dipte- ra:102088	Australia	JN991988	-	Penney <i>et al.</i> (2012)
<i>E. minotaurus</i> Claussen & Lucas, 1988	FSUNS:E0787	Greece	KY865451	_	Chroni <i>et al.</i> (2018)
<i>E. montanum</i> Grković, Radenković & Vujić in Grković <i>et al.</i> (2017)	FSUNS_11499	Montenegro	MK060005	-	Grković <i>et al.</i> (2019b)
<i>E. niveitibia</i> Becker, 1921	FSUNS:E0613	Greece	MG604934	_	Ricarte <i>et al.</i> (2018)
<i>E. nudus</i> Loew, 1848	CEUA_S64	Spain	OM791823	_	Present work

Table 1 (continued). List of the species used in the molecular analyses. New sequences generated for this work are highlighted in bold.

Species	DNA voucher	Country of origin	Acc. number (Genbank)	Specimen ID (BOLD)	Reference
<i>E. obliquus</i> (Fabricius, 1805)	427E02	Africa (no further details)	KT624209	-	Jordaens <i>et al.</i> (2015)
<i>E. ornatus</i> Meigen, 1822	E0801	Greece	KX083388	_	Grković <i>et al.</i> (2017)
E. ovatus Loew, 1848	FSUNS:G3015	Italy	MG604933	_	Ricarte <i>et al.</i> (2018)
<i>E. pannonicus</i> Ricarte, Vujić & Radenković in Markov <i>et al.</i> (2018)	08910	Serbia	KY272852	_	Grković <i>et al.</i> (2017)
<i>E. phaeacus</i> Chroni, Grković & Vujić in Chroni <i>et al.</i> (2018)	FSUNS:G0278	Greece	KY865446	_	Chroni <i>et al.</i> (2018)
E. pulchellus Loew, 1848	10064	Greece	KY865464	_	Chroni <i>et al.</i> (2018)
<i>E. punctifrons</i> Loew, 1857	CNC:Dipte- ra:102104	Morocco	JN991989	-	Penney <i>et al.</i> (2012)
<i>E. pusillus</i> Loew, 1848	UOTA_MEL123655	Greece	MG560014	_	Chroni <i>et al.</i> (2019)
<i>E. simplex</i> Ferguson, 1926	BIOUG01905-C12	Australia	-	DPAST560-11	Ratnasingham & Hebert (2007)
E. sinuatus Loew, 1855	FSUNS:G0271	Serbia	KY865455	_	Chroni <i>et al.</i> (2018)
<i>E. sogdianus</i> Stackelberg, 1952	06548	Greece	KX083384	_	Grković <i>et al.</i> (2017)
E. strigatus (Fallén, 1827)	42-504	Germany	KX083379	-	Grković <i>et al.</i> (2017)
<i>E. sulcitibius</i> Rondani, 1868	G3034	Greece	KX083387	-	Grković <i>et al.</i> (2017)
<i>E. torsicus</i> Grković & Vujić in Grković <i>et al.</i> (2015)	E0746	Greece	KT221006	-	Grković <i>et al.</i> (2015)
<i>E. tricolor</i> (Fabricius, 1798)	FSUNS:G3018	Italy	KY865450	_	Chroni <i>et al.</i> (2018)
<i>E. turanicus</i> Stackelberg, 1952	ZFMK- TIS-8000880	Georgia	MN621965	_	Mengual <i>et al.</i> (2020)
<i>E. uncipes</i> Rondani, 1850	12	Greece	KX083363	_	Grković <i>et al.</i> (2017)
<i>E. vestitus</i> Bezzi, 1912	-	India	MH178365	_	Annoj <i>et al.</i> (2020)
Platynochaetus setosus (Fabricius, 1794)	MZH:Y1711	France	MH521921	_	Vujić <i>et al.</i> (2019)

**Table 1** (continued). List of the species used in the molecular analyses. New sequences generated for this work are highlighted in bold.

# **Molecular study**

DNA was extracted both from one mesoleg and one metaleg of five paratypes (2 males and 3 females) and one male of *Eumerus nudus* Loew, 1848 using the DNeasy Blood & Tissue Kit (Qiagen, Valencia, CA) following the manufacture's protocol for animal tissues. PCR amplifications of the 5' end region of the Cytochrome *c* oxidase subunit I (COI-5') gene were performed with the universal primers LCO1490 (5'-GGTCAACAAATCATAAAGATATTGG-3') and HCO2198 (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') (Folmer *et al.* 1994). All amplifications were carried out in a total volume of 25  $\mu$ l containing 1× of Buffer reaction, 0.4 mM of dNTPs, 0.2  $\mu$ M of each primer, 2 mM of MgCl<sub>2</sub> and 1 unit of NZYTaq II DNA polymerase. Thermocycler conditions followed Grković *et al.* (2019b). PCR products were visualized with an electrophoresis process in a 1% agarose gel and sequenced at Macrogen Inc. (Macrogen, Spain).

The COI-5' barcode sequences obtained were edited by eye with the program Sequencher ver. 5.4.6 (Gene Codes Corporation 2017). Then, COI-5' sequences for each species of *Eumerus* available at the public repositories GenBank and BOLD (Ratnasingham & Hebert 2007) were downloaded (see Table 1). We excluded barcodes of *Eumerus figurans* Walker, 1859 and *Eumerus superbus* Shannon, 1927 as the pictures of the specimens assigned to them do not appear to correspond to these species. First alignment was performed automatically with MAFFT online service (Katoh *et al.* 2017) which was checked by eye with the program AliView ver. 1.25 (Larsson 2014). The final matrix had a length of 658 bp. We employed DAMBE7 (Xia 2018) to determine levels of variation for the first and third codon positions. We carried out a Neighbor-Joining (NJ) and a Maximum Likelihood (ML) analyses in MEGA7 (Kumar *et al.* 2016) both with 1000 bootstrap replications. The NJ analysis was conducted using the Maximum Composite Likelihood model. The ML analysis was performed using a General Time Reversible (GTR) model with invariant sites (+I) and gamma distribution (+G) which was the best evolutionary model proposed by the Corrected Akaike Information Criterion (AICc, 5038.996) using the software jModelTest ver. 2.1.10 (Darriba *et al.* 2012). The resulting trees were rooted based on a *Platynochaetus setosus* (Fabricius, 1794) sequence.

## Results

#### Taxonomy

Class Insecta Linnaeus, 1758 Order Diptera Linnaeus, 1758 Family Syrphidae Rondani, 1856 Subfamily Eristalinae Newman, 1834 Tribe Merodontini Edwards, 1915 Genus *Eumerus* Meigen, 1822

*Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov. urn:lsid:zoobank.org:act:C63F1D95-7528-4679-9705-FAA919D7CDDD Figs 2–7

# Diagnosis

Medium sized (6.5–9.5 mm) black species (Fig. 2), slightly larger than *E. strigatus*, which can be distinguished from similar species by the black pilosity on vertical triangle (Fig. 3), the absence of a baso-ventral ridge in the metatibia (Fig. 5B), the shape of sternum IV with a pair of black spinose valves (Fig. 6) and the general morphology of male genitalia (Fig. 7).

# Etymology

This species is named after the Spanish entomologist Juan Gil Collado for his important contribution to the knowledge of the Iberian hoverflies (Gil-Collado 1930). The specific epithet '*colladoi*' refers to his second surname and should be treated as a noun in the genitive case.

## Material examined

#### Holotype

SPAIN • ♂; Alicante, Alcoleja, Sierra de Aitana, Puerto de Tudons, on *Thapsia villosa*; 18 Jun. 2020; A. Ricarte leg.; CEUA00109561; CEUA-CIBIO.

## **Paratypes**

PORTUGAL • 1 ♂; Mértola, NE of town; 29S618-416; 22 May 2004; A. v. Eck leg.; coll. A. v. Eck.

SPAIN • 1 3; Málaga, Yunquera, Parque Natural Sierra de las Nieves, Puerto Saucillo; 36°43'22.9" N, 04°57'59.8" W; 1181 m a.s.l.; 21 May 2010; M.A. Marcos García leg.; en ('hovering around') piornal ('broom'); CEUA 00107843; CEUA-CIBIO • 1 3; Ciudad Real, Parque Nacional de Cabañeros; 2–23 Aug. 2004; A. Ricarte leg.; Malaise trap maA1; CEUA 00109557; CEUA-CIBIO • 1 3; Ciudad Real, Parque Nacional de Cabañeros, Finca Garbanzuelo; 27 Jul. 2021; P. Aguado leg.; CEUA 00109777; CEUA-CIBIO • 1 3; León, Sena de Luna; 3 Sep. 2010; F. Fresno leg.; N° Cat. 56574; MNCN • 1 3; León, Rodiezmo; 27 Aug. 2010; F. Fresno leg.; N° Cat. 57041; MNCN • 1 3; Salamanca, Espeja,



**Fig. 2.** *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov. **A**. Holotype,  $\mathcal{E}$ , dorsal view (CEUA-CIBIO). **B**. Holotype,  $\mathcal{E}$ , lateral view. **C**. Paratype,  $\mathcal{P}$ , dorsal view (CEUA-CIBIO). **D**. Paratype,  $\mathcal{P}$ , lateral view view (CEUA-CIBIO). Scale bars = 1 mm.



**Fig. 3.** *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov. **A**. Holotype,  $\Diamond$  (CEUA-CIBIO), head, general view. **B**. Paratype,  $\Diamond$  (CEUA-CIBIO), head, general view. **C**. Holotype,  $\Diamond$ , head, lateral view. **D**. Paratype,  $\Diamond$  (CEUA-CIBIO), head, lateral view. **E**. Holotype,  $\Diamond$ , antenna, lateral view. **F**. Paratype,  $\Diamond$  (CEUA-CIBIO), antenna, lateral view. Abbreviation: fs = fossette. Scale bars: A–C = 500 µm; D–F = 250 µm.

Campanarios de Azaba; 2 Aug. 2011; García, Ramírez, Moreno leg.; Malaise trap 19; CEUA 00109556; CEUA-CIBIO • 1 ♀; Salamanca. Topas, Castillo del Buen Amor; 11 Jun. 1978; Mª A. Marcos leg.; CEUA 00018013; M<sup>a</sup> A. Marcos det. as *Eumerus strigatus*; CEUA-CIBIO • 1  $\mathcal{Q}$ ; Salamanca, Montemayor del Río; 29 Apr. 1980; Mª A. Marcos leg.; CEUA 00018004; Mª A. Marcos det. as Eumerus strigatus; CEUA-CIBIO • 1 ♂; Salamanca, Vallejera de Riofrío, Puerto Vallejera; 1202 m a.s.l.; 26 Sep. 1977; S. Fdez. Gayubo leg.; CEUA 00017757; Mª A. Marcos-García det. as Eumerus amoenus; CEUA-CIBIO • 1  $\delta$ ; same locality and altitude as for preceding; 24 Aug. 1980; M.A. Marcos leg.; CEUA 00017758; M<sup>a</sup> A. Marcos-García det. as *Eumerus amoenus*; CEUA-CIBIO • 2 33; Alicante, Agres, Caveta del Voltor; 1200 m a.s.l.; 29 May 2001; Pérez Bañón, Marcos-García & Rojo leg.; CEUA 00109554 to 0010955; CEUA-CIBIO • 5 ♂♂; Alicante, Agres, Foia Ampla; 1060 m.; 29 May 2001; Pérez Bañón, Marcos-García & Rojo leg.; CEUA 00109771 to 00109775; X. Mengual det. as Eumerus amoenus in Mengual (2005); CEUA-CIBIO • 1 👌; same locality and altitude as for preceding; 17–31 Jul. 2001; Pérez Bañón, Marcos-García and Rojo leg.; Malaise trap; CEUA 00109552; X. Mengual det. as Eumerus amoenus in Mengual (2005); CEUA-CIBIO • 1 3; same locality and altitude as for preceding; 23 Apr.–15 May 2002; Pérez Bañón, Marcos-García and Rojo leg.; CEUA 00109553; Malaise trap; X. Mengual det. as Eumerus amoenus in Mengual (2005); CEUA-CIBIO • 1 3; Alicante, Agres, Font Retura; 900 m a.s.l.; 30 Aug.- 2 Sep. 2001; Pérez Bañón, Marcos-García and Rojo leg.; Malaise trap; CEUA00109776; X. Mengual det. as *Eumerus amoenus* in Mengual (2005); CEUA-CIBIO • 1 Å; same locality as for holotype; 6 Jun. 2020; A. Ricarte leg.; CEUA S01; CEUA 00107848; CEUA-CIBIO • 1 9; same locality as for holotype; 6 Jun. 2020; A. Ricarte; CEUA 00109558; CEUA-CIBIO • 1  $\bigcirc$ ; same locality



**Fig. 4.** Measurements of *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov. **A**, **C–D**. Holotype,  $\bigcirc$  (CEUA-CIBIO). B. Paratype,  $\bigcirc$  (CEUA-CIBIO). Abbreviations: bl = basoflagellomere length; bw = basoflagellomere width; hh = head height; hw = head width; l = body length; tg = tegula; w = body width; wl = wing length. Scale bars: A–B = 1 mm; C–D = 500 µm.

as for holotype; 29 May 2020; A. Ricarte leg.; CEUA 00109559; CEUA-CIBIO • 3  $\bigcirc$  ; same collection data as for holotype; CEUA 00109560; CEUA 00109562 to 00109563; CEUA-CIBIO • 3 QQ; same locality as for holotype; 20 Jun. 2020; A. Ricarte leg.; CEUA S02, CEUA 00107847; CEUA 00109564 to 00109565; CEUA-CIBIO; Coll. A. v. Eck • 3 QQ; same locality as for holotype; 20 Jun. 2020; Z. Nedeljković leg.; CEUA 00109566 to 00109568; CEUA-CIBIO • 1  $\bigcirc$ ; same locality as for holotype; 10 Sep. 2020; Z. Nedeljković leg.; CEUA 00109569; CEUA-CIBIO • 2 QQ; same locality as for holotype; 16 May 2021; Z. Nedeljković leg.; CEUA S52, CEUA 00107928; CEUA 00109570; CEUA-CIBIO • 2 ්ථ; same locality as for holotype; 26 May 2021; Z. Nedeljković leg.; CEUA 00108052 to 00108053; CEUA-CIBIO; CSCA • 5 ♀♀; same locality as for holotype; 26 May 2021; Z. Nedeljković leg.; CEUA 00108050 to 00108051; CEUA 00108067 to 00108069; CEUA-CIBIO; CSCA  $\cdot$  1  $\Im$ ; same locality as for holotype; 11 Jun. 2021; Z. Nedeljković leg.; CEUA 00108054; CEUA-CIBIO • 1 9; Alicante, Alcoleja, Sierra de Aitana, Mas del Piscul; 29 May 2019; M.A. Marcos García leg.; CEUA 00109854; CEUA-CIBIO • 1 ♀; same locality and date as for preceding; A. Ricarte leg.; CEUA 00109855; CEUA-CIBIO • 1  $\Im$ ; same locality as for preceding; 20 Jun. 2016; G.J. Souba Dols leg.; INV09526; CEUA 00111027; Ricarte det. as *Eumerus* sp. ; CEUA-CIBIO • 6  $\bigcirc \bigcirc \bigcirc$ ; same locality as for preceding; 26 May 2021; I. Ballester leg.; CEUA 00109860 to 00109865; CEUA-CIBIO • 1 ♂; Alicante, Benimaurell; 7 May 2007; Marcos-García & Ricarte leg.; CEUA 00029572; CEUA-CIBIO • 1 3; Alicante, la Vall d'Ebo; 38°48'16.60" N, 0°11'59.57" W; 555 m a.s.l.; 26 May 2021; P. Aguado leg.; CEUA 00108079; CEUA-CIBIO • 1 ♀; same collection data as for preceding; A. Ricarte leg.; CEUA 00108075; CEUA-CIBIO • 1 ♀; same locality as for preceding; 38°48′16.47″ N, 0°11′42.97″ O; 546 m a.s.l.; 20 May 2021; A. Ricarte leg.; on Thapsia villosa; CEUA 00109853; CEUA-CIBIO • 3 99; Alicante, la Vall de Laguar, Venta del Collao; 38°46'10.91" N, 0°9'4.21" W; 762 m a.s.l.; 26 May 2021; A. Ricarte leg; CEUA 00108059; CEUA 00108073 to 00108074; CEUA-CIBIO • 1 ♂; Alicante, Cocentaina, Font de la Penya Banyà; 4 Jul. 2021; I. Ballester leg.; en ('on') Rubus sp.; CEUA 00108072; CEUA-CIBIO • 1 3; Valencia, Bocairent, Mas del Parral; 900 m a.s.l.; 5-19 Jun. 2001; Malaise trap; Pérez Bañón, Marcos-García and Rojo leg.; CEUA 00109851; X. Mengual det. as Eumerus amoenus in Mengual (2005); CEUA-CIBIO • 1 ♂; same locality and altitude as for preceding; 30 Jul.–13 Aug. 2002; Malaise trap; Pérez Bañón, Marcos-García and Rojo leg.; CEUA 00109852; X. Mengual det. as Eumerus amoenus in Mengual (2005); CEUA-CIBIO • 1 9; Valencia, Bocairent, Sierra de Mariola, Pla d'Aparici, on *Thapsia villosa*; 3 Jun. 2020; A. Ricarte leg.; CEUA S06; CEUA 00107844; CEUA-CIBIO • 1 ♀, Valencia, Sª Mariola, Bocairent, Font del Mas dels Arbres, on *Thapsia villosa*; 3 Jun. 2020; A. Ricarte leg.; CEUA 00109850; CEUA-CIBIO • 2 QQ; Valencia, Alt del Portell, Bocairent, Vall Albaida; 837 m a.s.l.; 12 Jun. 2021; Z. Nedeljković leg.; CEUA 00108056 to 00108057; CEUA-CIBIO • 1 ♂; same collection data as for preceding; CEUA 00108058; CEUA-CIBIO • 1 3; same locality, altitude and date as for preceding; A. Ricarte leg.; CEUA 00108055; CEUA-CIBIO • 1 ♂; Valencia, S<sup>a</sup> Mariola, Bocairent, Font del Mas dels Arbres, campo de Thapsia villosa; 12 Jun. 2021; Z. Nedeljković leg.; CEUA 00108078; CEUA-CIBIO • 2  $\bigcirc$ ; same collection data as for preceding; CEUA 00108077 to 00108076; CEUA-CIBIO • 1  $\Diamond$ ; same locality and date as for preceding; A. Ricarte leg.; CEUA 00108071; CEUA-CIBIO • 1 &; Sierra Espuña; 37.866° N, 1.447° W; 732 m a.s.l.; 7 Oct. 2020; A. Ricarte, Z. Nedeljković and P. Aguado leg.; CEUA S03; CEUA 00107846; CEUA-CIBIO • 1 ♀; same collection data as for preceding; CEUA S04; CEUA 00107842; CEUA-CIBIO • 1 3; El Escorial; 14 Jun. 1922; Dusmet leg.; • 1 3; El Pardo; Jun. 1908; Arias leg.; MNCN • 1 3; Villaviciosa de Odón, Villaviciosa; 14 Apr. 1926; Dusmet leg.; J. Gil Collado det. as Eumerus amoenus in Gil-Collado (1930); MNNC.

## Additional examined material of other species of *Eumerus*

#### Eumerus amoenus Loew, 1848

SERBIA • 1 ♂; Dubašnica, Klisura Lazareve Reke; 21 Aug. 1997; Vujić leg.; CEUA 00017760; A. Vujić det.; CEUA-CIBIO • 1 ♀; Malinik, Malinik; 26 Jul. 1997; Dević leg; CEUA 00017763; A. Vujić det.; CEUA-CIBIO.

SPAIN • 1 ♂; Ciudad Real, Parque Nacional de Cabañeros; 1–24 Aug. 2004; A. Ricarte leg.; Malaise trap maJ1; CEUA 00084834; A. Ricarte det.; CEUA-CIBIO.

TUNISIA • 1 ♂; Djerba, Houmt Souk, 5 km S of Midoun; M. Hauser leg.; CEUA 00017759; M. Hauser det.; CEUA-CIBIO.

# Eumerus chrysopygus Sack, 1941

SOUTH KOREA • 1 ♂; Chungnam Keumsan Nami-myeon Pohyeonsa Pohyeonsa; 36°03.494 N, 127°27.225 E; 1–8 Jun. 2005; P. Tripton leg.; Malaise; CEUA 00103565; M. Hauser det.; CEUA-CIBIO.



**Fig. 5.** Metalegs of species of *Eumerus* Meigen, 1822. **A–B**. *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov., holotype, ♂ (CEUA-CIBIO). C. *Eumerus amoenus* Loew, 1848, ♂ (CEUA-CIBIO). Scale bars: 500 µm.

#### Eumerus clavatus Becker, 1921

GERMANY • 1 ♂; D-BW Pforzheim Niefern MV91 Lattenold MF; 26 Jun. 1992; Schmid-Egger leg.; D. Doczkal det.; CSCA • 1 ♀; same locality and collector as for preceding; 10 Sep. 1992; D. Doczkal det.; CSCA.

#### Eumerus nudus Loew, 1848

SPAIN • 1 ♂; Alicante, Tibi, S<sup>a</sup> del Maigmó, observatorio; 1176 m a.s.l.; 26 May 2021; A. Ricarte leg.; CEUA S64; CEUA 00111026; Z. Nedeljković det.; CEUA-CIBIO.

#### Description

## Male

MEASUREMENTS (mm). Holotype (3): l = 8.02; w = 2.05; h = 1.91; hw = 2.54; wl = 5.82.

HEAD. Eye contiguity virtually of the same length (0.31 mm) as the height of the frontal triangle (0.33 mm). Eye pilosity consisting of rather short, scarce and pale pile, specially conspicuous on the ventral part of eye. Face and frontal triangle (including lunules) densely and white pollinose, covered with white pile (Fig. 3A). Vertical triangle black and with long, erect, black pile (Fig. 3C), pollinose at its anterior apex and with two extremely reduced pollinose spots which are located behind posterior ocelli. Ocellar triangle isosceles. Occiput black and purple iridescent (under artificial white lightning), with pile shorter than those on the vertical triangle; occiput white pollinose along the eye margin. Scape and pedicel dark brown; pedicel with black pile which are longer on the ventral side. Basoflagellomere almost square (ratio length:width = 1:1.1), convex dorsally and slightly concave ventrally, dark brown except baso-ventrally, with a lunulate black fossette at the distal margin. Pedicel and basoflagellomere



**Fig. 6.** *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov., paratype,  $\mathcal{O}$  (CEUA-CIBIO), sternum IV. Abbreviation: vs = valves. Scale bar: 250  $\mu$ m.

sparsely white pollinose (pollinosity more obvious in dorsal view and under artificial white lighting). Arista black (Fig. 3E).

THORAX. Mesonotum and lateral sides of the thorax black. Anterior and posterior parts of the scutum covered with rather short and white pile, in the medial part white pile intermixed with short and semireclined black pile; scutum with two medial white pollinose vittae extending over the anterior two thirds of the scutum and with a small circular grey pollinose spot posterior to transverse suture; lateral margins of the scutum with green metallic reflections. Notopleural sulcus absent. Disc of scutellum with white pile slightly longer than those on the scutum; posterior margin of scutellum with teeth-like protuberances, each bearing a long black pilis apically. Posterior anepisternum, anterior anepimeron and katepisternum on its postero-lateral area densely long white pilose. Katatergum with discrete bunch of yellow setae. Pleuron grey pollinose except the posterior anepisternum postero-laterally, anterior anepimeron centrally, ventral margin of posterior anepimeron and the dorsal margin of katepisternum. Femora black, basis and apices light brown. Basal half of pro- and mesotibia light brown and apical half black. Basal third of metatibia light brown and apical two thirds black. Dorsal surface of all tarsi black, ventrally yellowish brown. Posterior side of pro- and mesofemora white pilose. Metafemur densely white pilose, ventrally with pile of different lengths including some which are nearly twice as long as the reminder. Metafemur with an anterior row of 6 spinae and a posterior row of 14 spinae curved towards the posterior side of the metafemur (Fig. 5A). Metatibia flattened, without a baso-ventral ridge (Fig. 5B). Wing membrane extensively microtrichose; posterior margin of the wing with dense, short and brown pile; margin of ventral calypter with rather long and yellow pile; margin of dorsal calypter with pile shorter than those of ventral calypter; halter yellow.

ABDOMEN. Terga I–IV black. Lateral margin of tergum I grey pollinose and white pilose. Terga II–IV densely white pilose on their lateral margins (pile on anterior corners of tergum II rather long); terga II–IV each with a pair of slightly curved, white pollinose maculae, terga covered with rather short, reclined



**Fig. 7.** *Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov., holotype,  $\mathcal{S}$  (CEUA-CIBIO), genitalia. A. Hypandrium, lateral view. **B**. Epandrium, lateral view. **C**. Epandrium, right side, ventral view. Abbreviations: asl = anterior surstylar lobe; cc = cercus; hl = hyaline lobes; pr = prominence; psl = posterior surstylar lobe. Scale bars: 250 µm.

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Fig. 8. Tree inferred using the Maximum Likelihood method. Bootstrap values > 50 are shown near nodes. Branch lengths measured in number of substitutions per site.

and white pile. Anterior margins of terga III–IV black pilose. Posterior margin of tergum IV light brown with a marginal line of black pile along it. Sternum IV with two quadrangular, straight projections on the posterior margin; central area of the sternum IV black pilose and protruding in a rounded, nose-like expansion; sternum IV with two black spinose valves under the medial area (Fig. 6).

MALE GENITALIA. Epandrium with a simple posterior surstylar lobe convex anteriorly and straight posteriorly (Fig. 7B). Cercus with long pile. Anterior surstylar lobe with two large hyaline lobes and a small third accessory lobe in between (Fig. 7B–C). Hypandrium with a prominence at the base which shows a small indentation (Fig. 7A).

#### Female

Same as the male, besides the sexual dimorphism, except for the following characters: eye pilosity more abundant; basoflagellomere  $1.0-1.2 \times$  wider than long and larger than in male (Fig. 3D, F); black macula at the distal margin of the basoflagellomere more noticeable (Fig. 3F); white pollinose spots behind the posterior ocelli larger than in male; frons white pollinose along eye margins but grey on its central area (Fig. 3B).

## Variation

Based on the examined material, *E. colladoi* Ricarte & Aguado-Aranda sp. nov. shows intraspecific variation in body length ( $\mathcal{F} = 6.43-8.91 \text{ mm}$ , n = 21;  $\mathcal{Q} = 7.69-9.58 \text{ mm}$ , n = 15), length of frontal triangle in males (0.28–0.38 mm), length of eye contiguity in males (0.24–0.41 mm), the presence or absence of two reduced white pollinose spots behind posterior ocelli (in males), the inclination of the spinae on the ventral side of the metafemur (from almost not inclined at all to an inclination of 90°) and the extent of the coloration on the metatibia (from light brown to black).

#### Distribution

The new species has been recorded from different Spanish and Portuguese localities, all in the Mediterranean region of the Iberian Peninsula (Fig. 1).

#### **Biology**

The flight period of *E. colladoi* Ricarte & Aguado-Aranda sp. nov. runs from late May to October. Adults can be found in a wide altitudinal range (80–1200 m) mainly associated with mountain areas. The predominant vegetation in these areas are woodlands of pine (*Pinus* L.) or oak (*Quercus* L.) where some adults have been sampled feeding on inflorescences of *Thapsia villosa* L. (Apiaceae).

#### Systematic position of E. colladoi Ricarte & Aguado-Aranda sp. nov. based on DNA evidence

Both the NJ and ML trees had virtually identical topologies. In the ML-tree (Fig. 8), high node support values (> 70) were obtained for every species group but for the *E. pulchellus* group (see Introduction). All barcodes of *E. colladoi* Ricarte & Aguado-Aranda sp. nov. were similar to each other with just 1 base pair difference. *Eumerus nudus* is similar to *E. colladoi* sp. nov. in the general shape of the basoflagellomere, the pilosity of the scutum, the coloration of the posterior margin of tergum IV and shape of male genitalia (see Discussion). The morphological similarities between *E. nudus* and *E. colladoi* sp. nov. justified the incorporation of the *E. nudus* sequence in the molecular analyses. A COI-5' sequence of the *E. nudus* was generated as it had no representation in the consulted BOLD and GenBank databases. The barcode sequence of *E. nudus* grouped, as expected, with that of *Eumerus alpinus* Rondani, 1857 but they did not form a clade with *E. colladoi* sp. nov. In addition, the new species did not cluster with sequences of species of the *E. clavatus* group, which have even more similar male genitalia in comparison with *E. colladoi* sp. nov. together with a medium-sized body, a dark body coloration and an elongated and parallel-sided abdomen. The proportion of invariant sites for the first

and third codon positions were 0.29 and 0.07, respectively. Level of variation is usually higher in the third codon position as it tends to have faster rate of evolution (Felsenstein 1978).

# Discussion

In the recent key to the species of *Eumerus* from Switzerland and surrounding parts of central Europe (Speight et al. 2021), E. colladoi Ricarte & Aguado-Aranda sp. nov. would key out as Eumerus amoenus Loew, 1848. However, the new species differs from *E. amoenus* in the eye pilosity, which is slightly longer and denser in E. colladoi sp. nov.; pollinosity found on the anterior apex of vertical triangle, which is sparser in *E. amoenus*; shape of basoflagellomere, which is slightly longer in *E. amoenus*; vertical triangle wholly covered with black pile in E. colladoi sp. nov. (except in the pollinose area of the anterior corner where a few yellow pile are present), black only on the ocellar triangle in *E. amoenus*; absence of a ridge on the baso-ventral side of the metatibia in E. colladoi sp. nov., a row of short reclined black spinae that is present in E. amoenus (Fig. 5C); shape of the sternum IV (Fig. 6), which is simple in *E. amoenus*; and the general morphology of the male genitalia (Fig. 7) (for *E. amoenus* genitalia, see Vujić & Šimić 1998). The fact that E. colladoi sp. nov. lacks a baso-ventral ridge in the metatibia is particularly interesting as this character state appears to be rare in the Iberian Eumerus (Martin Hauser in litt.). In addition, the new species was also run through the Stackelberg's (1961) key to the Palaearctic Eumerus and it keyed out to Eumerus chrysopygus Sack, 1941, which can easily be separated from E. colladoi sp. nov. by the eve pilosity, which is virtually absent in E. chrysopygus; pilosity of the scutum, which is longer and dark brown in E. chrysopygus but shorter and with white and black pile intermixed in E. colladoi sp. nov.; a ridge with a row of black setae on the baso-ventral side of metatibia, which is not present in E colladoi sp. nov.; length of metabasitarsus, which is shorter and much broader in *E chrysopygus* than in *E. colladoi* sp. nov.; pilosity of the posterior half of tergum IV, which is goldenyellow in E. chrysopygus and black in E. colladoi sp. nov.; and the sternum IV morphology, which in E. chrysopygus lacks the two spinose valves typical of E. colladoi sp. nov. (Fig. 6).

*Eumerus colladoi* Ricarte & Aguado-Aranda sp. nov. is also similar to the species of the *E. clavatus* group, *Eumerus clavatus* and *Eumerus uncipes* Rondani, 1850 (see Results) but the new species differs in the mesonotum pilosity (not golden-yellow but white intermixed with black pile posteriorly and medially), length of tergum IV (not twice the length of tergum III), external morphology of sternum III (lacking the prominence with long pile in its medial part) and the shape of sternum IV which is not plier-like shaped (see Speight *et al.* 2021: fig. 15). The male of *E. colladoi* sp. nov. can be easily distinguished from those of *E. clavatus* and *E. uncipes* by the shape of the metatibia, which is modified in these two species (see Speight *et al.* 2021 figs 16d, 17b), but simple in *E. colladoi* sp. nov.

The new species is easily differentiated from *E. nudus* (see Results) by the body size, which is normally larger in *E. nudus*; pile on the ventral side of the metafemur, which is clearly longer in *E. nudus*; the anterior row of spinae on the ventral side of the metafemur, which is on a gentle elevation in *E. nudus* and the general shape of sternum IV. Finally, females were also run through Stackelberg's key and keyed out to *Eumerus kondarensis* Stackelberg, 1952. Nevertheless, males of *E. kondaresis* have different genitalia (Stackelberg 1952) and the barcode analyses confirmed molecularly that the examined females belonged to the same species as the males of *E. colladoi* Ricarte & Aguado-Aranda sp. nov. These results prove that barcode sequence analysis is useful to match males and females in hoverfly species as sexual dimorphism and morphological similarity between females of different species make sometimes identification difficult.

Nevertheless, the obtained molecular trees must be cautiously taken when assessing species affiliations with groups or species relationships, as the species sampling is still too limited. Barcodes are only available for 43 species confirmed of the 250+ known valid species of *Eumerus* worldwide (Souba-Dols *et al.* 2020). Therefore, it is likely that the current systematic position of *E. colladoi* sp. nov. can change when barcodes of additional species of *Eumerus* are obtained. At the moment, the new species cannot be assigned to any of the defined groups of species and it would conform a monospecific group. Thus, a wider taxon sampling and the use of other informative markers are necessary to disentangle the actual phylogenetic relationships of *E. colladoi* sp. nov. with other species.

Eumerus colladoi Ricarte & Aguado-Aranda sp. nov. is distributed across the Iberian Peninsula (Fig. 1) but it has not been collected in its Eurosiberian Region, to the north (Rivas-Martínez et al. 2014). The fact that E. colladoi sp. nov. has never been reported from the Eurosiberian Region of the Iberian Peninsula (e.g., Marcos-García 1985a, 1985b, 1990; Ricarte et al. 2014) may indicate that it is absent from there. However, the new species may occur in parts of this Region as some records from close-by localities, for example Rodiezmo and Sena de Luna in the province of León show. On the other hand, the ubiquity of the new species suggests that the larvae depend on a widespread plant species in the Iberian Peninsula or they are polyphagous. Despite the availability of large amounts of data on the Syrphidae in Europe (Speight et al. 2020) and the number of recent publications on the taxonomy of Eumerus (e.g., Chroni et al. 2018; Grković et al. 2021; Speight et al. 2021), E. colladoi sp. nov. had not been unrevealed so far. So, it might be really absent from other parts of Europe. However, the generally poor knowledge of the hoverflies of North Africa (El-Hawagry & Gilbert 2019; Mebarkia et al. 2021) leads to a reasonable doubt about the occurrence of this new species in this part of the African continent. since other hoverfly species are present both in the Iberian Peninsula and North Africa, for example Mallota dusmeti Andréu, 1926 (Pennards et al. 2021) and Merodon luteihumerus Marcos-García, Vujić & Mengual, 2007 (Likov & Radenković 2021). At the moment, Eumerus colladoi sp. nov. must be considered as the sixth Iberian-endemic species of Eumerus, which now has 37 species recorded in the Iberian Peninsula. This region of Europe is home to a high diversity of *Eumerus* similar to that in other regions such as the Balkan Peninsula (Grković 2018).

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