

New species records of flower flies (Diptera, Syrphidae) for Iran

Nuevos registros de especies de sírfidos (Diptera, Syrphidae) de Irán

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ABSTRACT

New records of flower flies (Diptera, Syrphidae) from Iran are reported. The species *Criorhina talyshensis* (Stackelberg) is reported for the first time from Iran, and a female of *Caliprobola aurea* Sack from Mazandaran Province represents the second record of the species from this country. DNA barcodes are provided for both species, together with images and an identification key to *Caliprobola* species.

Key words: Syrphidae; hover flies; flower flies; *Caliprobola*; *Criorhina*; Iran; new record; DNA barcode.

RESUMEN

En este estudio se presentan nuevos registros de moscas de las flores (Diptera, Syrphidae) de Irán. Se cita la especie *Criorhina talyshensis* (Stackelberg) por primera vez en Irán y se presenta una hembra de *Caliprobola aurea* Sack, capturada en la provincia de Mazandaran, como el segundo registro de esta especie en este país. Se proporcionan códigos de barras de ADN para ambas especies, junto con imágenes y una clave de identificación para las especies de *Caliprobola*.

Palabras clave: Syrphidae; hover flies; flower flies; *Caliprobola*; *Criorhina*; Irán; nuevo registro; código de barras de ADN.

INTRODUCTION

Syrphidae (Insecta, Diptera) is a speciose family of true flies with more than 6,300 described species worldwide (SKEVINGTON *et al.*, 2019). Commonly known as hover or flower flies, adults feed on pollen and nectar (LARSON *et al.*, 2011; ROTHERAY & GILBERT, 2011) while larvae have a large array of different natural histories, such as phytophagy (including fungivory), saprophagy, and predation including parasitoidism (ROTHERAY & GILBERT, 1999, 2011; ROJO *et al.*, 2003; PÉREZ-LACHAUD *et al.*, 2014). Flower flies provide important ecosystem services, like pollination in natural and agricultural systems (SSYMANK & KEARNS, 2009; INOUE *et al.*, 2015), biological control of pests (TENHUMBERG, 1995; ARCAYA *et al.*, 2017) and decomposition of organic matter (LARDÉ, 1989; MORALES & WOLFF, 2010). Moreover, some species are of economical and medical significance (MAGNI *et al.*, 2013; HEO *et al.*, 2020; PÉREZ-BAÑÓN *et al.*, 2020).

Iran has a rich fauna of flower flies with more than 226 recorded species and growing (VAN STEENIS & GHARALI, 2016; GILASIAN *et al.*, 2017; KAZERANI *et al.*, 2017; VUJIĆ *et al.*, 2017; VAN STEENIS *et al.*, 2018). Many new records and new species have been reported during the last two decades (see KAZERANI *et al.*, 2017) as a result of active Iranian researchers, extensive field campaigns and international collaboration.

In the present work, we report new flower fly records from Iran, including a new *Criorhina* species record.

MATERIAL AND METHODS

Study area, terminology and imaging

Studied flower flies were collected in northern Iran (Mazandaran and Gilan Provinces) using Malaise traps during 2018, within the project “Biodiversity of insects in Hyrcanian Forest”. The Hyrcanian Forest (also known as Caspian Forest), dating back some 50 million years, is

composed mostly of deciduous forest dominated by *Fagus orientalis* Lipsky and ranges from the Talysh Mountains in the Republic of Azerbaijan to the northern slopes of the Alborz Mountains in North Iran (in the provinces of Gilan, Mazandaran and Golestan). This remarkably intact ecosystem is highly valued by the high number of endemic species, with 3,234 species belonging to 856 genera and 148 families of vascular plants until now (AKHANI *et al.*, 2010; JALILI & JAMZAD, 1999).

Morphological terminology follows THOMPSON (1999). We used the following published references for identification: SPEIGHT (2020a) for the genus, VAN STEENIS & GHARALI (2016) for *Criorhina* Meigen, 1822, and SACK (1910) for *Caliprobola* Rondani, 1845.

Photographs were taken with a Canon EOS 7D® mounted on a P-51 Cam-Lift (Dun Inc., VA, USA) and imported with the help of Adobe Lightroom® (version 5.6). Later, images were stacked with Zerene Stacker® 1.04 (Richland, Washington, USA) and edited with Adobe Photoshop® CS5.1. The studied material is deposited in the Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany (ZFMK).

Molecular methods

We sequenced the Folmer region (5' end) of the mitochondrial cytochrome c oxidase subunit I (COI), the so-called DNA barcode (HEBERT *et al.*, 2003), of the studied specimens. One leg of ethanol-preserved specimens was used for DNA extraction. The extraction protocol follows MENGUAL *et al.* (2018). Entire specimens were preserved and labelled as DNA voucher specimens for the purpose of morphological studies and deposited at the ZFMK.

The COI sequence fragment was amplified using the forward primer LCO-1490 (5'-GGTCA-ACAAATCATAAAGATATTG-3') and the reverse primer 780R (5'-CCAAAAATCARAATARRT-GYTG-3'). PCR amplification protocol follows ROZO-LOPEZ & MENGUAL (2015). All new sequences were submitted to GenBank using BOLD (www.boldsystems.org) and GenBank Accession Numbers are indicated for each specimen.

A Neighbour-Joining analysis with the Jukes-Cantor model was carried out with the newly obtained COI sequences we used together with other published COI sequences from GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>) and in BOLD systems (<http://www.boldsystems.org/index.php>). Sequences were aligned using Geneious R7 (version 7.1.9, Biomatters Ltd.). Then, we run the Neighbour-Joining analysis using the same software and 1,000 bootstrapping replicates were performed (Figures S1 and S2 in Supplementary material).

RESULTS

Caliprobola aurea Sack, 1910

Examined material. IRAN: Mazandaran Province: Noor, 36.56331 N 52.07111 E, 0 m, 15 May 2018, F. Kazerani leg. (1♀, ZFMK-DIP-00067273).

Differential diagnosis (Figs. 1A, 1B). Easy to identify as there is no other flower fly like this large, shiny golden-metallic green genus. This species can be distinguished from *Caliprobola speciosa* (Rossi, 1790) by the entirely yellow legs (femora partly black in *C. speciosa*), longer abdominal pilosity, and wing entirely yellow tinted (with a dark macula on apical cell r_{2+3} in *C. speciosa*).

Distribution. Azerbaijan and Iran.

Genetics. A female specimen was sequenced (GenBank Accession Number MW128362). The obtained COI sequence is similar (between 97.853% and 98.006%) to the COI sequences of *Caliprobola speciosa* from Germany and Georgia (see Table SI and Fig. S1 in Supplementary material). This is the first published DNA barcode for *C. aurea*.

Remarks. This is the second specimen record of *C. aurea* from Iran and the fourth time that this species is reported in the literature. SACK (1910) described this species based on an unknown number of males and females from the Talysh Mountains (south-eastern Azerbaijan and north-western Iran within Ardabil and Gilan Provinces). PECK (1988) mentioned the Azerbaijani Lankaran District for the distribution of this species.

HAUSER (1998) collected a male specimen from Lankaran District and a female specimen from Astara District, both in south-eastern Azerbaijan. Specimens reported by HAUSER (1998) were collected in June and the newly reported female from Mazandaran Province was collected on May. More recently, GILASIAN (2004) reported a male specimen from Mazandaran Province. The record from GILASIAN (2004) and our new female specimen were collected outside the Talysh Mountains.

SACK (1910) provided an identification key for the two species of *Caliprobola*, but he also included in the key *Hadromyia cimbiciformis* (Portschinsky, 1879) as *Caliprobola cimbiciformis*. Since then, no identification key has been published. Here we provide an updated identification key for the only two *Caliprobola* species in the world.

Key to Caliprobola species

1. Femora and tibiae entirely yellow (at most the basal 1/20 of the femora dark) (Figs. 1A, 1B). Wing remarkably yellow tinted anteriorly (Fig. 1A). Abdominal pilosity longer, usually covering the posterior half of the tergite so the background colour is not visible (Fig. 1A).....*C. aurea* Sack
- Femora black at least on basal half, usually black on basal 2/3; tibiae yellow (Figs. 1C, 1D). Wing remarkably yellow tinted anteriorly with a dark maculae at apex, between cells r_1 and r_{4+5} (Figs. 1C, 1D). Abdominal pilosity shorter, with longer pile at posterior margin only (Figs. 1C, 1D).....*C. speciosa* (Rossi)

Criorhina talyshensis (Stackelberg, 1960)

Examined material. IRAN (new record): Gilan Province: Shafarood, 37.47167 N 48.82306 E, 1114 m, 22 July 2018, F. Kazerani leg. (1♂, ZFMK-DIP-00067345).

Differential diagnosis (Figs. 1E, 1F). Members of this genus are large bumble bee and bee mimics with the face produced ventrally and the basoflagellomere broader than long. As indicated by VAN STEENIS & GHARALI (2016),

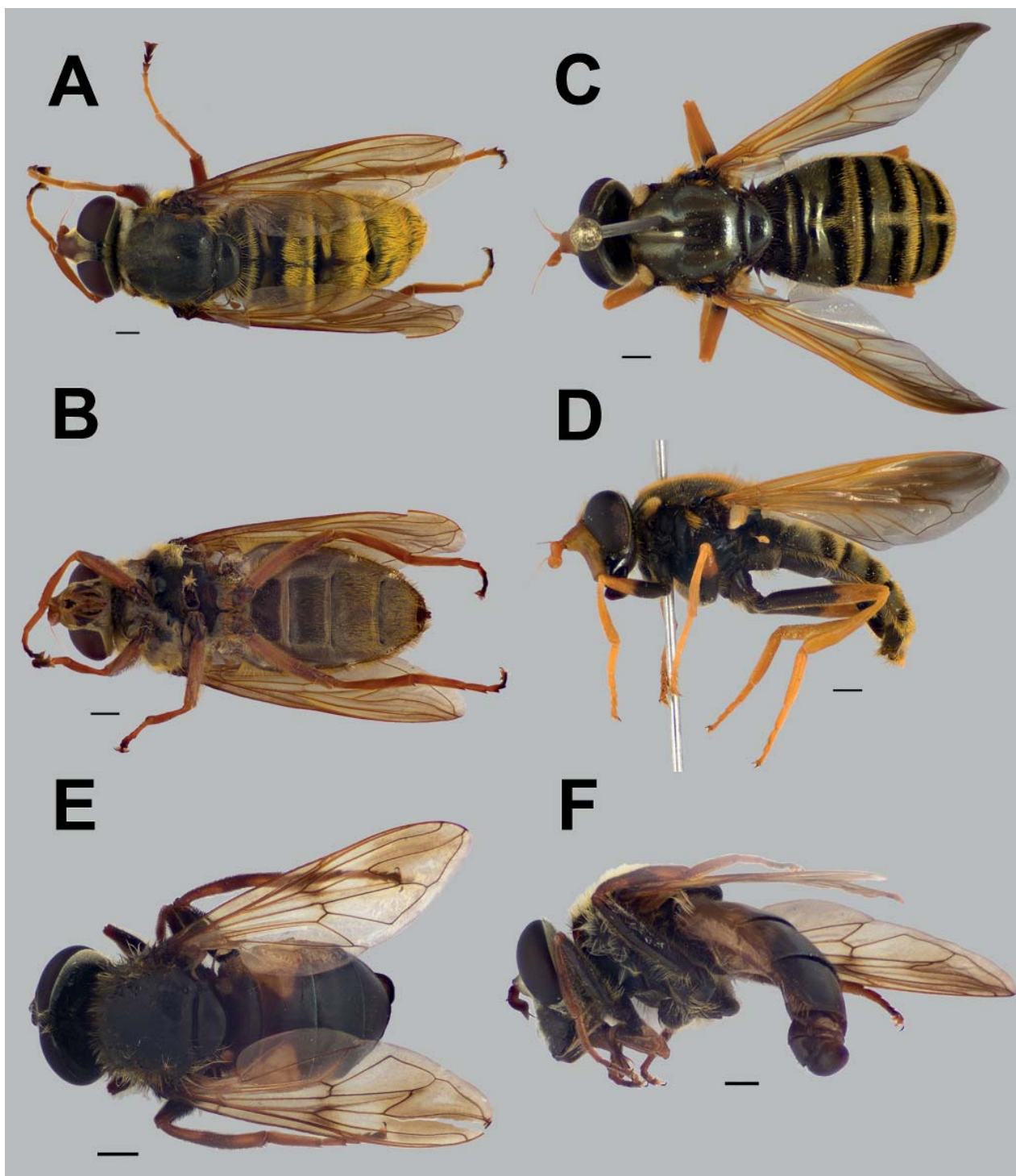


Fig. 1. *Caliprobola aurea* female, Iran (ZFMK-DIP-00067273). A) dorsal view; B) ventral view. *Caliprobola speciosa* female, Georgia (ZFMK-DIP-00063082). C) dorsal view; D) lateral view. *Criorhina talyshensis* male, Iran (ZFMK-DIP-00067345). E) dorsal view; F) lateral view. Scale bars = 1.0 mm.

Fig. 1. *Caliprobola aurea* hembra, Irán (ZFMK-DIP-00067273). A) vista dorsal; B) vista ventral. *Caliprobola speciosa* hembra, Georgia (ZFMK-DIP-00063082). C) vista dorsal; D) vista lateral. *Criorhina talyshensis* macho, Irán (ZFMK-DIP-00067345). E) vista dorsal; F) vista lateral. Barras de escala = 1.0 mm.



Fig. 2. *Criorhina talyshensis*, male genitalia (ZFMK-DIP-00067345). Scale bar = 0.5 mm.

Fig. 2. *Criorhina talyshensis*, genitalia del macho (ZFMK-DIP-00067345). Barra de escala = 0.5 mm.

C. talyshensis can be distinguished from other *Criorhina* species by having the katapimeron bare like *C. berberina* (Fabricius, 1805) and *C. floccosa* (Meigen, 1822) and by the presence of two large, triangular yellow maculae on tergite 2 (also in tergite 3 in male) (Figs 1E, 1F), which are absent in *C. berberina* and *C. floccosa*. Male genitalia is figured in Fig. 2.

Distribution. Known from south-eastern Azerbaijan and Iran (new record).

Genetics. The male specimen from Iran was sequenced (GenBank Accession Number MW128352) and its COI sequence is very similar (98.75% similarity) to other published COI sequences of the same species from Azerbaijan (GenBank Accession Number MK155108). In the Neighbour-Joining analysis both COI sequences

of *C. talyshensis* grouped together (see Fig. S2 in Supplemental material). As MENGUAL *et al.* (2020) noted, the nearest neighbour of *C. talyshensis* in BOLD systems is *C. berberina*.

Remarks. Reported for Iran for the first time. Our specimen is the fifth ever published individual after the type series (STACKELBERG, 1960) and the couple collected by HAUSER (1998). All previous records were from Azerbaijan. This is the third *Criorhina* species recorded from Iran, after *C. portschinskyi* (Stackelberg, 1955) (VAN STEENIS & GHARALI, 2016) and *C. ranunculi* (Panzer, 1804) (GILASIAN *et al.*, 2017). The occurrence of this species in Iran was hypothesised by VAN STEENIS & GHARALI (2016) and confirmed with our record. The flight period of *C. talyshensis* based on published records was

from end April to mid June (STACKELBERG, 1960; HAUSER, 1998), but this new male record is from end July.

DISCUSSION

The faunistic and floristic biodiversity of the Hyrcanian Region is on a global level significant. Approximately 44% (over 3,200 species) of known vascular plants in Iran occurs in the Hyrcanian Region, which covers only 7% of Iran's territory (AKHANI *et al.*, 2010). About 280 taxa are endemic and sub-endemic for the Hyrcanian Region and almost 500 plant species are Iranian endemics as well as a total of 80 native tree species have been registered from Hyrcanian forest (TOHIDIFAR *et al.*, 2016). The invertebrate diversity in the Hyrcanian Forests is still under research, but nobody would be surprised with similar numbers of endemic taxa. In a study about diversity of saproxylic insects in Hyrcanian forest, 361 beetle and 7 true bug species were identified; of these, 56 species were endemic (MÜLLER *et al.*, 2018). Recently, a new endemic syrphid was described from the Hyrcanian Forests (GILASIAN *et al.*, 2017).

The biology of the two flower fly species here reported is unknown. Known larvae of the genera *Caliprobola* and *Criorhina* are saproxylic and are found in coniferous and deciduous forests with overmature trees (ROTHERAY & GILBERT, 2011; SPEIGHT, 2020b). PROKHOROV & SHPARYK (2019) indicated that larvae of *C. speciosa* develop in soft, moist, rotting wood of old oak and beech trees. HAUSER (1998) stated that the two collected specimens of *C. aurea* flew from hollows in oak (*Quercus*) logs. VAN STEENIS & GHARALI (2016) noted that *Criorhina talyshensis* "has been collected in open, heavily grazed mixed oak forest (*Quercus petraea* (Matt.) Liebl. and *Fagus orientalis*) with small streams and south-facing dry slopes." In the present study, *C. aurea* and *C. talyshensis* were collected in temperate broadleaf and mixed forests (Hyrcanian Forests) covered mostly by the Oriental Beech (*Fagus orientalis*) followed by oak, alder, hornbeam,

maple, elm, etc. (MARVIE MOHADJER, 2005). Published records of both species are very low in numbers, a fact that may indicate that these species have a limited distribution and/or a very specific ecological niche.

Overmature and senescent trees are vital for saproxylic species (MORADI *et al.*, 2012; MICÓ *et al.*, 2013). Saproxylic species, including flower flies, have been used as bioindicators in conservation studies (SPEIGHT, 1989; REEMER, 2005; FAYT *et al.*, 2006; RICARTE *et al.*, 2009; RADENKOVIĆ *et al.*, 2013; RAMÍREZ-HERNÁNDEZ *et al.*, 2014; VUJIĆ *et al.*, 2016). We do not have biological data for *C. aurea*, but we can assume that its biology and conservation status may reflect those of *C. speciosa*, with the aggravating of a more restricted distributional range. PROKHOROV & SHPARYK (2019) considered *C. speciosa* as rare throughout Europe and its range is extremely fragmented. Moreover, these authors said that this species requires constant monitoring and is a typical and generally accepted bioindicator species of ancient oak and beech forests (SPEIGHT, 1989, 2020b), the area of which is declining. In Great Britain, this species is considered as Near Threatened (BALL & MORRIS, 2014) and in Ukraine, it is proposed as Vulnerable (PROKHOROV & SHPARYK, 2019). In Spain *C. speciosa* is regarded as Endangered (MARCOS-GARCÍA, 2006; MARCOS GARCÍA & RICARTE, 2012), and one of the main threats is the removal and logging of old trees (MARCOS-GARCÍA, 2006, 2009). In Europe, isolated populations may disappear due to forest degradation due to anthropogenic factors (MARCOS-GARCÍA, 2006; BARTSCH *et al.*, 2009; BALL & MORRIS, 2014; PROKHOROV *et al.*, 2018). Thus, the new flower fly records of these saproxylic genera and the first record of *C. talyshensis* from Iran, together with endemic new species (GILASIAN *et al.*, 2017), point out the relevance of the Hyrcanian Forests, declared UNESCO World Heritage Site in 2019 (UNESCO, 2019), for saproxylic Syrphidae and emphasize the exceptional importance of the overmature trees for the protection and conservation of biodiversity.

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Supplementary material at

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