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## Two flower fly species (Diptera: Syrphidae) new to India

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### Abstract

Based on adult morphology and DNA barcoding, two flower fly species are reported for the first time from India: *Helophilus trivittatus* (Fabricius, 1805) and *Lejogaster tarsata* (Megerle in Meigen, 1822). These species were collected from the Kashmir Valley, in the northern fringe of the Western Himalaya of the Indian subcontinent.

**Keywords:** Syrphidae, DNA barcoding, new records, distribution, India

### Introduction

The family Syrphidae, commonly known as flower flies or hoverflies, comprises over 6,300 described species classified into circa 200 genera and it occurs nearly worldwide (Skevington *et al.* 2019). Flower flies are considered important pollinators in both agricultural and natural ecosystems (Larson *et al.* 2011; Inouye *et al.* 2015). The predatory larvae of the subfamilies Pipizinae and Syrphinae are natural biological control agents, reducing aphid and scale populations in both field and laboratory conditions (Tenhumberg & Poehling 1995; Nelson *et al.* 2012; Dunn *et al.* 2020). The syrphid fauna of India is very diverse and is currently made up of 358 species in 71 genera (Ghorpadé 2014a; Sengupta *et al.* 2016, 2020; Mengual & Barkalov 2019; Wachkoo *et al.* 2019); however, flower flies have not been the subject of a dedicated study and remain poorly understood (Shah *et al.* 2014).

Here we present the first records from India of two widespread Palearctic flower fly species: *Helophilus trivittatus* (Fabricius, 1805), and *Lejogaster tarsata* (Megerle in Meigen, 1822). A brief diagnosis and color images with diagnostic characters are given for both species to facilitate their morphological identification, and also to validate these new faunal species records to the region.

## Material and methods

### Collecting sites

The studied specimens were collected by hand netting in the Kashmir Valley (Jammu and Kashmir), the Palaearctic portion of India, located in the northwestern part of the Indian subcontinent between 33°22' and 34°50'N latitude and 73°55' and 73°33'E longitude (Maqbool *et al.* 2018). The specimens were collected in natural vegetation from two localities, Shopian and Srinagar. Shopian (33.710°N 74.844°E) is situated in Pir Panjal Range at an altitude of 2,146 m with an average annual precipitation of 740.5 mm and 14 °C average temperature (Wachkoo *et al.* 2018a), whereas Srinagar (34.131° N 74.835° E) is at mid altitude (1,600 m) with an average annual precipitation of 660 mm and 13 °C average temperature (Wachkoo *et al.* 2018b).

### Morphological studies

The taxonomic study was conducted using a Leica Wild M10 stereomicroscope (Leica, Wetzlar, Germany). Adult identifications were made using Van Veen (2004) and verified with collection material from different collections stated below. Terminology follows the glossary in Skevington *et al.* (2019).

### Institutional abbreviations

CNC	Canadian National Collection of Insects, Arachnids, and Nematodes, Ontario, Canada.
GCSI	Government Degree College, Shopian, Jammu and Kashmir, India.
JSA	Private collection of Jeroen van Steenis, Amersfoort, the Netherlands.
ZFMK	Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany.

### Photography

For photographs, adult specimens were placed inside a light box using cool day light LED lamps (3W) and images were captured using a DSLR camera (Nikon D5300) with a macro lens (Tokina 100 mm f 2.8) attached to microscope objectives. Multiple images were generated using an auto stacking-rail (Stackrail rs90) and merged using Combine ZP software (<https://alan-hadley.software.informer.com/>). Final plates were assembled with Adobe Photoshop® CS4. To prepare genitalia slides for microphotography, male genitalia were removed by cutting between tergites 7 and 8 and then cleared overnight in a 10% KOH solution. The genitalia were then placed into acetic acid for a few minutes followed by rinsing with ethanol before placing in glycerine. Male genitalia were then slide-mounted and examined with an LED lamp (3W) to produce the transmission light beam. Microphotographs were captured with the same set-up as above, with the addition of infinity-corrected microscope objectives (Maqbool *et al.* 2021).

### DNA barcoding

One or two legs of selected specimens were used for DNA extraction. Extractions were carried out using the DNeasy Blood and Tissue Kit (Qiagen Inc., Santa Clara, CA, USA) following the manufacturer's protocol. Entire specimens or remnants of specimens were preserved and labelled as DNA voucher specimens for the purpose of morphological studies and deposited in the above-mentioned collections as listed in the 'Material examined' sections.

The standard DNA barcode of the 5' region of the mitochondrial cytochrome *c* oxidase subunit I gene (COI) was amplified using the forward primer LCO1-1490 (5'-GCTCAACAAATCATAAAGATATTGG-3'; Folmer *et al.* 1994) and the reverse primer COI-Dipt-2183R, also known as COI-780R (5'-CCAAAAAATCARAATARRTGYTG-3'; Gibson *et al.* 2011). PCR amplification, purification, sequencing protocols and contig assembly were carried out as described in Rozo-Lopez & Mengual (2015) for specimens sequenced at ZFMK, or as described in Gibson *et al.* (2010) for specimens sequenced at CNC. All new sequences were submitted to GenBank via BOLD ([www.boldsystems.org](http://www.boldsystems.org)). GenBank accession numbers are listed for each sequenced specimen in the text. Once we had the sequences, we used the BOLD Identification System 'all barcodes' database (IDS; [https://www.boldsystems.org/index.php/IDS\\_OpenIdEngine](https://www.boldsystems.org/index.php/IDS_OpenIdEngine)) to compare them against all the barcode records in BOLD, including private and public records with a minimum sequence length of 500 bp.

## Results

### *Helophilus trivittatus* (Fabricius, 1805)

Figures 1–2

*Eristalis trivittatus* Fabricius, 1805: 235.

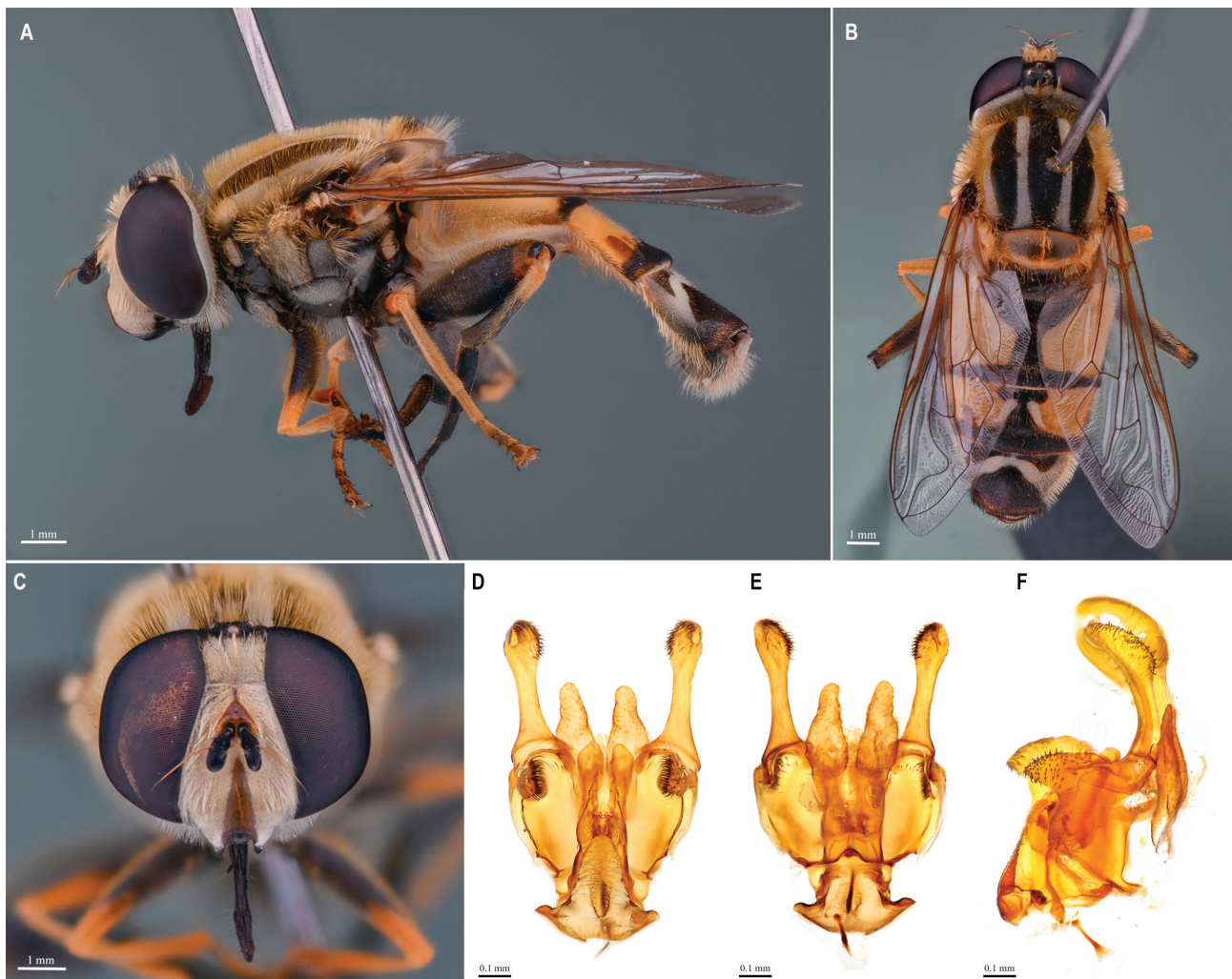
*Musca parallelus* Harris, 1778.

*Helophilus camporum* Meigen, 1822.

*Helophilus solitarius* Rondani, 1857.

**Diagnosis.** The genus *Helophilus* Meigen, 1822 has the wing vein  $R_{4+5}$  strongly sinuate into cell  $r_{4+5}$  and the wing cell  $r_1$  open; thorax pollinosity velvet black with off-white fasciae along the lateral margin and two additional ones medio-laterally; protibia very short pilose; metafemur with a posterobasal patch of densely set, black setulae; bare eyes; male eyes widely dichoptic; face with medial bare vitta from central knob downwards; and face concave with clearly protruding ventral part.

*Mesembrius* Rondani, 1857 and *Parhelophilus* Girschner, 1897 are both very similar to *Helophilus*, but they can be distinguished as follows: face entirely pollinose in *Mesembrius* or with a short and very narrow medial bare vitta in *Parhelophilus*; eyes in male holoptic or slightly dichoptic in *Mesembrius* (or males of *Parhelophilus* also have eyes widely dichoptic like *Helophilus*); katepimeron pilose in *Mesembrius* (katepimeron bare in *Helophilus* and *Parhelophilus*); metabasitarsomere basoventrally with globuliferous pile in *Mesembrius* (*Helophilus* and *Parhelophilus* without such modified pile); and pterostigma very short, simulating a crossvein in *Mesembrius* and *Parhelophilus* (pterostigma elongate, longer than broad, in the form of a pigmented patch of wing membrane in *Helophilus*).



**Figure 1.** *Helophilus trivittatus* (Fabricius, 1805), male. **A**, Lateral habitus; **B**, Dorsal habitus; **C**, Head, frontal view; **D–F**, Male genitalia: **D**, Dorsal view; **E**, Ventral view; **F**, Lateral view.





**Figure 2.** *Helophilus trivittatus* (Fabricius, 1805), female. **A**, Lateral habitus; **B**, Dorsal habitus; **C**, Head, frontal view.

*Helophilus trivittatus* is easily recognized by the pale (usually yellow, sometimes brownish) medial bare vitta on the face in contrast to most other *Helophilus* species in which it is black. The only other species with this facial pattern are two Nearctic species (*H. fasciatus* Walker, 1849 and *H. latifrons* Loew, 1863), but these have the protarsus black instead of predominantly brown-yellow as in *H. trivittatus*. The abdominal markings are very characteristic with large lime-yellow maculae with a straight medial margin on tergum 2; tergum 3 with a pair of squarish maculae along the anterior margin and medially with strongly oblique greyish pollinose maculae; tergum 4 with strongly curved off-white pollinose fascia.

**Material examined.** India: Jammu and Kashmir, Srinagar, 34.131° N 74.835° E, 1,750 m a.s.l., 23.iv.2016, Aijaz A. Wachkoo leg. (2♂, 2♀, GCSI – A\_Wachkoo00001 to A\_Wachkoo00004, 1♀, CNC – CNC\_Diptera254129); same data as previous, except: 26.iv.2016 (1♂, 1♀, GCSI – A\_Wachkoo00005 to A\_Wachkoo00006, 1♂, CNC – CNC\_Diptera254130); with same data as previous, except: 22.ix.2016 (1♀, GCSI – A\_Wachkoo00007).

**Geographical distribution.** West Palearctic eastwards through Eurasia to the Pacific, including Iran and Afghanistan (Speight 2020), and India (first records presented here). This species was previously reported from



northern Afghanistan (Bala Murghab; see Bańkowska 1969; Ghorpadé 2014b), but this genus has not been recorded from Pakistan (Shehzad *et al.* 2017). The new locality in India (Srinagar) is more than 1,000 km away from Bala Murghab to the East.

**DNA barcodes.** The GenBank numbers for CNC\_Diptera254129 and CNC\_Diptera254130 are MZ995225 and MZ995226, respectively. Both DNA barcodes are 659 base pairs long and are a 100% match for over 20 other *H. trivittatus* specimens on BOLD.

**Remarks.** The specimens collected in the present study are the first valid records of the genus *Helophilus* and the species *H. trivittatus* from India. The report of *H. trivittatus* from India by Khan (2017) is questionable as he did not provide literature used for his identifications, and did not mention if any experts verified his identifications. Moreover, vouchers were not lodged in a museum and consequently unavailable for examination. The fact that most of the species cited from India previously identified as *Helophilus* belong to *Mesembrius* Rondani, 1857 or *Mallota* Meigen, 1822 (Ghorpadé 2014a, 2019), and that Khan (2017) cited the New World genus *Palpada* Macquart, 1834 from Kashmir make the re-evaluation of the material studied by Khan (2017) necessary.

### ***Lejogaster tarsata* (Megerle in Meigen, 1822)**

Figures 3–4

*Chrysogaster tarsata* Megerle in Meigen, 1822: 271.

*Chrysogaster splendida* Megerle in Meigen, 1822.

*Chrysogaster bicolor* Macquart, 1829.

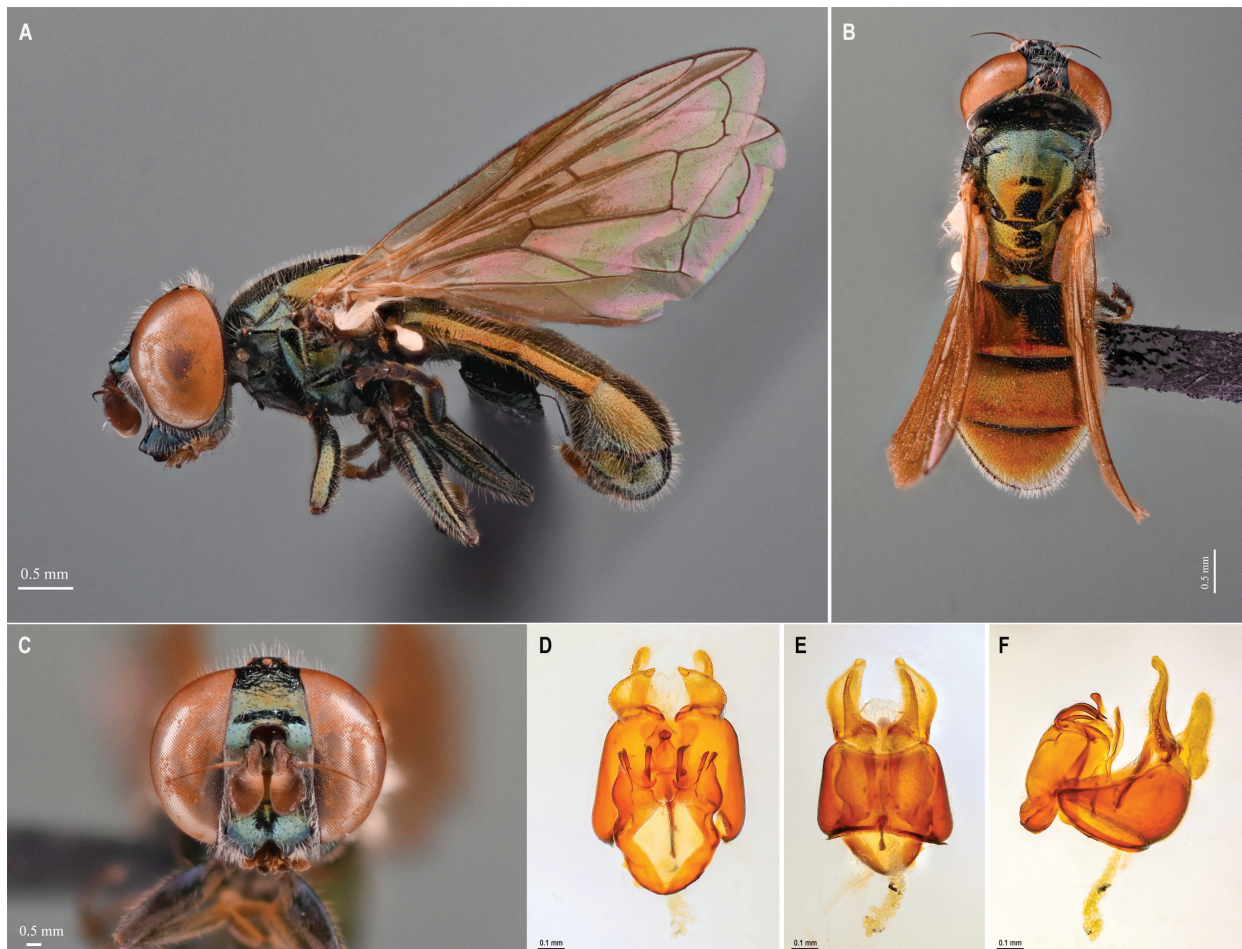
*Chrysogaster amethystina* Macquart, 1834.

*Chrysogaster amethystea* Meigen, 1838 (misspelling).

*Chrysogaster rufitarsis* Loew, 1840.

*Liogaster aurichalcea* Becker in Becker & Stein, 1913.

*Orthoneura longior* Becker, 1921.



**Figure 3.** *Lejogaster tarsata* (Megerle in Meigen, 1822), male. **A**, Lateral habitus; **B**, Dorsal habitus; **C**, Head, frontal view; **D–F**, Male genitalia: **D**, Dorsal view; **E**, Ventral view; **F**, Lateral view.



**Diagnosis.** This species is a member of the tribe Chrysogastrini, which is characterized by: postpronotum pilose; face with inconspicuous or reduced genae; face concave in most species without central knob; females of many species with grooves on the frons. The genera of the tribe Chrysogastrini are heterogeneous in their characters and none of the characters mentioned above are restricted to this tribe, nor are they present in all genera of the tribe. The delimitation of the tribe is still a point of debate (Moran *et al.* 2021).

The genus *Lejogaster* Rondani, 1857 is, however, clearly differentiated from the other genera within this tribe and other Syrphidae by the overall greenish shiny thorax and abdomen. Other diagnostic characters are: male eyes dichoptic; wing vein  $M_1$  perpendicular to wing vein  $R_{4+5}$ ; base of wing vein R with some long setae on dorsal surface; metafemur with normal pile, apico-ventrally without setae; postpedicel round and large.

*Lejogaster tarsata* is differentiated from *L. metallina* (Fabricius, 1777) by the pilose postero-dorsal corner of the posterior anepisternum (bare in *L. metallina*); the smaller postpedicel, more rounded in the male and more elongate in the female (large and oval in *L. metallina*); the usually yellow ventral part of the basoflagellomere (entirely black or brownish in *L. metallina*); and the medial tarsomeres of all tarsi yellow (tarsi black in *L. metallina*). In West-Mediterranean and Asian specimens of *L. tarsata*, the legs tend to be entirely black.



**Figure 4.** *Lejogaster tarsata* (Megerle in Meigen, 1822), female. **A**, Lateral habitus; **B**, Dorsal habitus; **C**, Head, frontal view.



**Material examined.** India: Jammu and Kashmir, Shopian, 33.710°N 74.844°E, 2,146 m a.s.l., 17.iv.2014, Aijaz A. Wachkoo leg. (3♂, 7♀, GCSI – A\_Wachkoo00008 to A\_Wachkoo00017, 1♂, CNC–Jeff\_Skevington\_Specimen45204, 1♂, 1♀, JSA – JV\_Steenis00001 to JV\_Steenis00002); same data as previous, except: 07.vi.2018 (7♂, 12♀, GCSI – A\_Wachkoo00018 to A\_Wachkoo00038); same data as previous, except: 02.x.2018 (4♂, 4♀, ZFMK– ZFMK-DIP-00082381, ZFMK-DIP-00082382, ZFMK-DIP-00082450, ZFMK-DIP-00082451, ZFMK-DIP-00082383, ZFMK-DIP-00082384, ZFMK-DIP-00082452, ZFMK-DIP-00082453); Srinagar, 34.131N° 74.835E°, 1750 m. a.s.l., 23.iv.2016 (2♂, 3♀, GCSI – A\_Wachkoo00039 to A\_Wachkoo00043, 2♀, CNC – CNC1078224-5), same data as previous, except 12.x.2014 (1♀, CNC – CNC\_Diptera263860).

**Geographical distribution.** West Palaearctic into European parts of Russia; Central Asia (Iran, Afghanistan, Uzbekistan, Tajikistan, Kirghizia, Turkmenistan and Kazakhstan to Mongolia), south-eastern Siberia and the Pacific coast (Speight 2020), and India (first records presented here). This species was previously reported from north and east Afghanistan (Bala Murghab and Jalalabad; Bańkowska 1969). The genus has not been recorded from Pakistan (Shehzad *et al.* 2017). The two new localities in India are more than 400 km eastwards from Jalalabad.

**DNA barcodes.** The COI barcode was sequenced for four specimens (ZFMK-DIP-00082381, ZFMK-DIP-00082382, ZFMK-DIP-00082383, ZFMK-DIP-00082384) and their GenBank accession numbers are OK415797, OK415796, OK415795 and OK415794, respectively. DNA barcodes are 666 base pairs long and all four are identical. The new sequences have a >99% similarity with over 20 specimens of *L. tarsata* on BOLD from different countries, namely Finland, France, Germany, the Netherlands, Pakistan and Russia.

**Comments.** The genus *Lejogaster* is recorded for the first time from India. *Lejogaster tarsata* is a species found in large numbers in the Kashmir Valley and it seems it has two generations per year, one in spring and one in autumn.

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