



**First records of *Paracladopelma nigrītula* (Goetghebuer, 1942)
and *Thienemanniella vittata* (Edwards, 1924) (Diptera: Chironomidae)
in Poland**

**Pierwsze stwierdzenia *Paracladopelma nigrītula* (Goetghebuer, 1942)
i *Thienemanniella vittata* (Edwards, 1924) (Diptera: Chironomidae)
w Polsce**

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ABSTRACT. The paper presents two newly recorded Chironomidae species for Poland, sampled in the Rawka River in the Łódź Voivodeship: *Paracladopelma nigrītula* (Goetghebuer, 1942), collected from 4 sampling points (5 specimens) and *Thienemanniella vittata* (Edwards, 1924), collected from 24 sampling points (48 specimens). Both species were recorded during the studies on aquatic macroinvertebrates inhabiting common water moss – *Fontinalis antipyretica* Hedw. In the discussion, *P. nigrītula* and *T. vittata* are treated in terms of their occurrence in such habitats.

KEY WORDS: Chironomidae, mime midges, larvae, faunistics, Poland

INTRODUCTION

Mime midges (Chironomidae) are a species-rich family of Nematocera that consists of approximately 7500 species (Zakrzewska *et al.* 2023) and are widely distributed in all types of freshwater habitats (Andersen *et al.* 2013). The immature stages of Chironomidae are commonly used in water quality assessment, paleolimnological and ecological research (*e.g.* Tarkowska-Kukuryk 2014, Woźniak *et al.* 2018, Plóciennik *et al.* 2020, Baranov *et al.* 2022). Despite this, larvae of Chironomidae are not often used as a subject of faunistic studies. Since the larvae exhibit fewer distinguishing features compared to adults, it is frequently impossible to identify them to the species level. Thus, molecular identification *via* DNA barcoding has become an important

tool in studies on immature stages of chironomids (Gdawski *et al.* 2022). Until now, 432 species of Chironomidae have been recorded in Poland (Szadziński *et al.* 2023), yet only 60 of them are known to occur in the Rawka River and its oxbow system (Nijboer *et al.* 2006, Słomczyński *et al.* 2023). Further research on the Chironomidae larvae composition of the Rawka River revealed two new species to the Polish fauna – *Paracladopelma nigrītula* (Goetghebuer, 1942) and *Thienemanniella vittata* (Edwards, 1924).

MATERIAL AND METHODS

The sampling sites were located in the middle and upper section of the Rawka, a lowland river in central Poland (Fig. 1). Rawka has been protected by a nature reserve since 1983 and is situated within the Bolimów Landscape Park. The studied river section is characterized by a swift current, sandy or sandy-stony bottom and a large amount of submerged wood (Fig. 2).

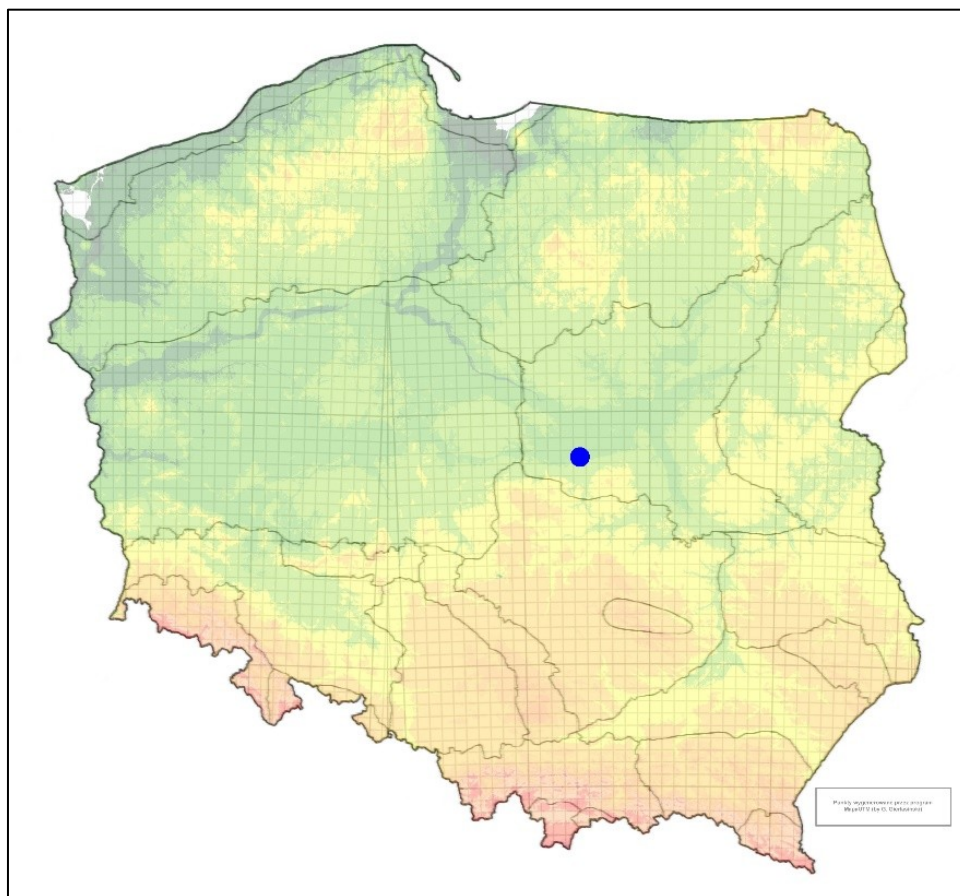


Fig. 1. UTM map with location of the Rawka River, the place of sampling the larvae of *Paracladopelma nigrītula* and *Thienemanniella vittata*.

Ryc. 1. Mapa UTM z lokalizacją rzeki Rawki, miejsca zbioru larw *Paracladopelma nigrītula* i *Thienemanniella vittata*.



Fig. 2. Meander of the Rawka River near Doleck (Łódź Voivodeship).

Ryc. 2. Meandry rzeki Rawki koło Dolecka (województwo łódzkie).

The studied material was collected from the common aquatic moss (*Fontinalis antipyretica* Hedw.) and from the river bottom using a benthic net with a mesh size of 0.5 mm. After the sampling, the benthic and moss samples were put in containers and transported to the laboratory. The collected macroinvertebrates were sorted under stereomicroscope (mag. 1.5–5×10) and preserved in 96% ethanol. The larvae were mounted on permanent slides in Hoyer's medium. Based on the head capsule's characteristics, the larvae were identified to the lowest possible taxonomic level using the keys of Schmidt (1993) and Moller Pillot & Klink (2003). In the case of *Thienemanniella vittata*, molecular identification was also performed to refine morphological identification.

The DNA extraction was performed using the GeneMATRIX Tissue DNA Purification Kit (manufactured by EURx) following the attached protocol. During the PCR, using LCO1490 JJ and HCO2198 JJ primers (Astrin & Stüben 2008), the fragment of cytochrome c oxidase subunit I gene (COI) was amplified and then sequenced using Third Generation Sequencing (Oxford Nanopore Technology) to obtain a sequence referring to the exact species. All the COI sequences obtained were uploaded to the BOLD database in a dataset "DS-RAWKA: DATASET Macroinvertebrates of the Rawka River Nature Reserve" (DOI: dx.doi.org/10.5883/DS-RAWKA). The taxonomic assignment was made by using COI Species Database provided by BOLD. The reference specimens are deposited in the second author's collection. In order to generate the map for this publication, the program *Mapa UTM ver. 6* by G. Gierłasiński was used.

RESULTS AND DISCUSSION

Paracladopelma nigrītula (Goetghebuer, 1942)

MATERIAL EXAMINED (5 larvae). **Rawka River**. UTM DC46, 29 IV 2023 (3 larvae), 5 VII 2023 (2 larvae), leg. K. Słomczyński, det. T. Matera & M. Plóciennik. **First record for Poland.**

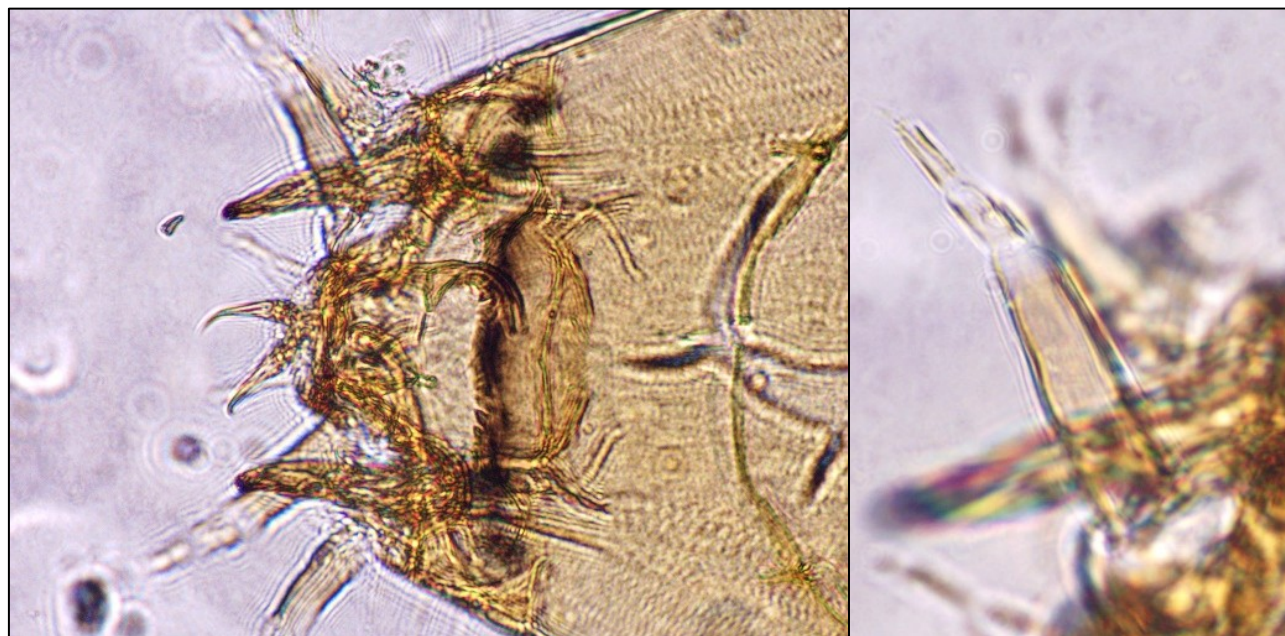


Fig. 3. Larva of *Paracladopelma nigrītula*, mouthparts and antenna.

Ryc. 3. Larwa *Paracladopelma nigrītula*, narządy gębowe i czułek.

The genus *Paracladopelma* Harnisch, 1923 consists of approximately 40 species (Andersen *et al.* 2013, <https://www.gbif.org/species/1455604> accessed 19.07.2024), yet only two of them – *P. camptolabis* (Kieffer, 1913) and *P. laminatum* (Kieffer, 1921) have been recorded in the Polish fauna (Szadziewski *et al.* 2023). Larvae inhabit sandy substrates and can be found in fast flowing streams and lakes. This study, conducted at Rawka River, reports it only from the river bottom. It serves as an indicator species, showing low tolerance to water eutrophication (Lang *et al.* 2016). Some studies consider *P. nigrītula* a complex of species aggregating species with similar larval morphology (Jackson 1977). Nevertheless, Moller Pillot (1978) questions this classification. According to the key of Moller Pillot & Klink (2003) *P. nigrītula* figures as a separate species that can be distinguished from *laminata* species group sensu Moller Pillot (1984) aggregating larvae of the other species in Europe, by mentum with 6th lateral mental tooth normal, 2 median mental teeth and very large SIII seta.

Thienemanniella vittata (Edwards, 1924)

MATERIAL EXAMINED (48 larvae). **Rawka River.** UTM DC46, 29 IV 2023 (7 larvae), 5 VII 2023 (41 larvae), leg. K. Słomczyński, det. T. Matera & K. Słomczyński (confirmed by molecular identification). **First record for Poland.**

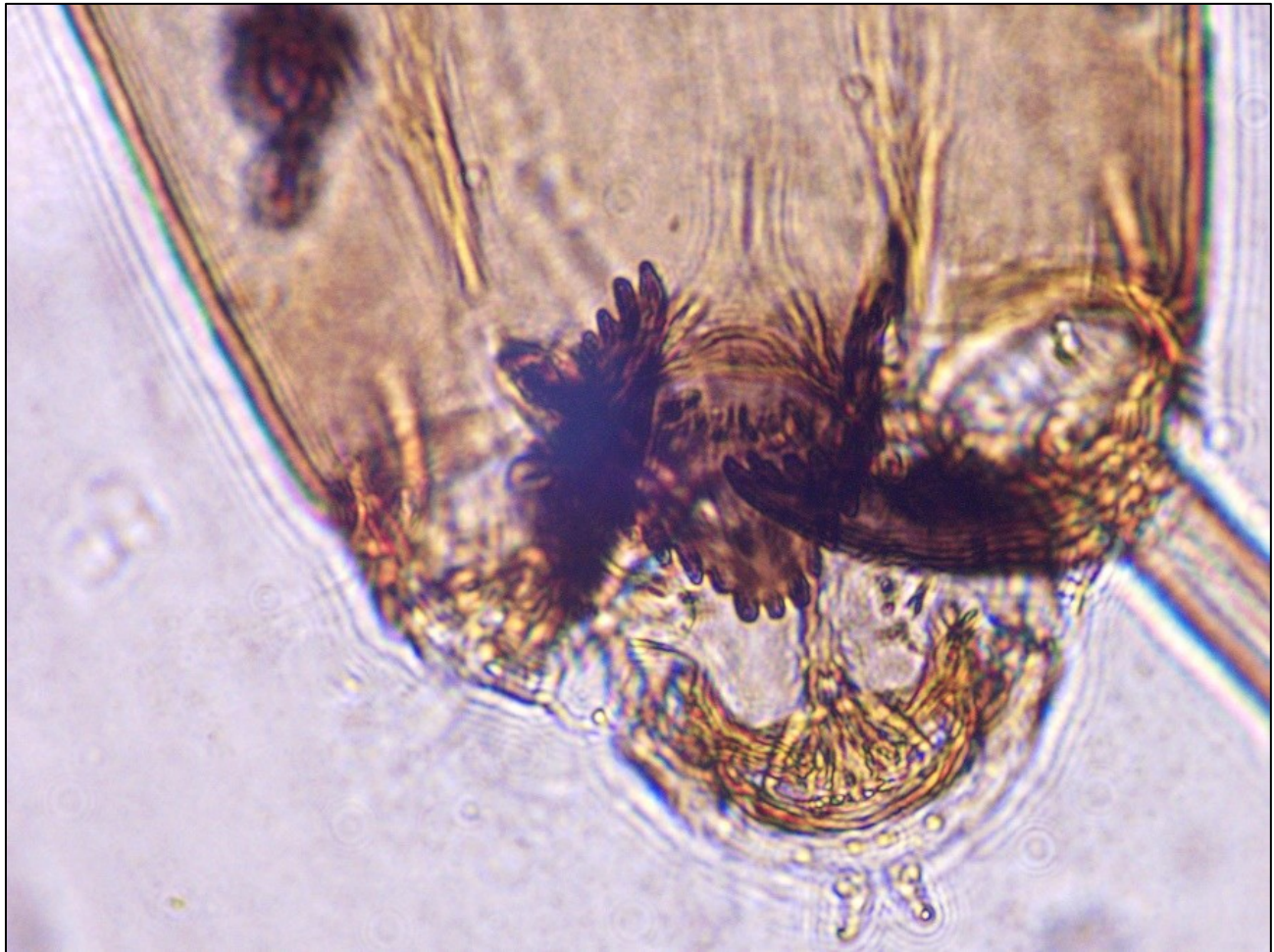


Fig. 4. Larva of *Thienemanniella vittata*, mouthparts.

Ryc. 4. Larwa *Thienemanniella vittata*, narządy gębowe.

The genus *Thienemanniella* Kieffer, 1911 groups ca. 70 species (<https://www.gbif.org/species/1455279> accessed 19.07.2024) and two of them – *T. clavicornis* (Kieffer, 1911) and *T. majuscula* (Edwards, 1924) – were recorded in Poland to date (Szadziwski *et al.* 2023). It closely resembles *Corynoneura* Winnertz, 1846 from which it can be distinguished by shorter antennae. *Thienemanniella* species are rheophiles, inhabiting aquatic plants and submerged wood (Moller Pillot 2014). *Thienemanniella vittata* and *Thienemanniella majuscula* are common species in European streams, however they differ slightly in the habitat preferences. Moller Pillot (2014) indicates that *T. majuscula* has not been found at the river bottom so far, except

for stones, while *T. vittata* also occupies sandy bottom. Both species in this study, conducted in the Rawka River, were collected only from aquatic moss twigs. Since it is difficult to precisely separate *T. vittata* from *T. majuscula* with morphological features (Moller Pillot 2014), the molecular identification was applied in this case. In BOLD there are 21 public sequences referring to *Thienemanniella vittata*, which form 4 BINs (Barcode Index Numbers). The obtained sequences are placed in the species *Thienemanniella vittata* (BIN: BOLD:AAV3048) with 99.84% probability. In Europe, sequences of the species with >98% match to obtained ones are from Germany, Finland and Norway. Nevertheless, in the database there are 25 similar sequences referring to *Thienemanniella majuscula* (p-distance: 3.53%) with the highest similarity to *T. vittata* of 96.26%. The confirmation of the identification was made based on the public record with the following ID: LEFIJ3608-16.COI-5P (BIN: BOLD:AAV3048).

There is wide research on Chironomidae fauna in large and medium rivers, but most of them focus on variously degraded streams (*i.e.* Grzybkowska 1992, Schöll & Haybach 2004, Leszczyńska *et al.* 2019). The study, conducted in one of the last pristine lowland rivers in central Poland, indicates that new species for the country can still be recorded in a relatively well investigated area. The same aquatic moss is rarely investigated as a habitat for mime midges. This study indicates that *F. antipyretica* is also a shelter for rare species that are restricted to macrophyte habitats. The Rawka River with its complex, natural environment and rich aquatic vegetation differs from the other one at Łódź Voivodship but is still neglected in the case of biodiversity research compared to the Grabia River, which is far more transformed (Słomczyński *et al.* 2023). Future studies on pristine streams like Rawka, including adults collection and extensive barcoding may identify many new species from Poland, documenting potentially high biodiversity.

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STRESZCZENIE

Ochotkowate (Chironomidae) to wyjątkowo liczna w gatunki rodzina muchówek o dużym znaczeniu ekologicznym. Ich larwy zasiedlają wszelkie typy środowisk wodnych, natomiast badania faunistyczne dotyczące ochotkowatych, szczególnie oparte na postaciach larwalnych, są nad wyraz rzadkie.

Eksploatując różnorodność gatunkową larw ochotkowatych rzeki Rawki (województwo łódzkie), wykazano dwa nowe dla Polski gatunki Chironomidae – *Paracladopelma nigracula* (Goetghebuer, 1942) i *Thienemanniella vittata* (Edwards, 1924). Larwy tych gatunków stwierdzono w zgrupowaniach bentosowych (*Paracladopelma nigracula*) oraz peryfitonowych, na mchach wodnych z rodzaju *Fontinalis* (*Thienemanniella vittata*).

Uwzględniając fakt, że badania faunistyczne nad larwami Chironomidae w Polsce nie cieszą się dużą popularnością, można spodziewać się, że prowadzenie takich badań przyczyni się do wykazania kolejnych nowych dla fauny Polski gatunków muchówek z rodziny ochotkowatych.



Editorial remarks:

* Papers of the 40th volume of *Dipteron* are dedicated to the late Maria Grzybkowska

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