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Biting midges (Diptera: Ceratopogonidae) in tree hole habitats in Slovakia

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#### Abstract

The water-filled tree hole fauna of biting midges (Diptera: Ceratopogonidae) in Slovakia is documented for the first time. Three *Culicoides* Latreille and two *Dasybelea* Kieffer species were reared from these habitats. Epidemiological significance of *C. obsoletus* (Meigen, 1818) is briefly discussed.

#### INTRODUCTION

Small cavities in trees, very often filled with stagnant rainwater (so-called dendrotelmata), are ecosystems with a frequent occurrence of immature stages of various Diptera (e.g. Röhnert 1950, Kitching 2004). The species living in tree holes are Received: Accepted:

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also known to include biting midges (Diptera: Ceratopogonidae), a family of small nematocerous flies. These tiny midges, which are morphologically very similar to chironomids, are represented by c. 140 species in Slovakia (Tóthová & Knoz 2009, Szadziewski et al. 2013). Despite the fact that dendrotelmata are ephemeral and unstable habitats, they are suitable breeding sites for some species of *Culicoides* Latreille, *Dasyhelea* Kieffer and *Forcipomyia* Meigen. Their larvae are common inhabitants of rotting wood, tree sap, sediments and water accumulated at the bottom of tree holes (Mullen & Hribar 1988).

During studies on the fauna of tree holes in Slovakia, five biting midge species were reported in these microhabitats, including *Dasyhelea flavifrons* (Guérin, 1833) that has been based on a doubtful record from the country so far.

#### MATERIALS AND METHODS

The tree hole sampling was carried out at two sites in the vicinity of the village Diviacka Nová Ves (district Prievidza). Geomorphological units and grid mapping codes of DFS (Databank of the fauna of Slovakia) are cited according to Stloukal & Grujbárová (2013). All of the sampled tree holes were at heights up to 0.5 m measured from the ground, with a water capacity of 5–30 liters. More than 70% of the sampled tree holes completely lost water at least once a year.

Sampling sites:

Site 1: Slovakia, Diviacka Nová Ves, beech forest (beech monoculture, with old trees along the forest edges), west of the village, with 7 sampled beech tree holes and 1 sampled elm tree hole, 340 m a.s.l., 48°44'51.89"N, 18°28'57.5"E, 7276 (DFS).



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**Site 2:** Slovakia, Vrbany env., Diviacka Nová Ves, oak forest (old forest dominated by oak), northeast of the village, with 8 sampled oak tree holes, and 1 sampled ash tree hole, 320 m a.s.l., 48°45'25.1"N, 18°30'43.5"E, 7277 (DFS).

For a reliable identification of specimens of many Diptera inhabiting dendrotelmata, it is recommended to rear the larvae to the adult stage or to collect some adults in the field (e.g. Oboňa et al. 2012, Oboňa & Ježek 2013, Oboňa & Starý 2013). Consequently, the studies presented in this paper were based only on determinations of adults. The method of rearing the biting midge larvae follows Oboňa & Ježek (2013). Sediments obtained from the tree holes were transported to the laboratory, and the rearing containers were checked daily. Adults were preserved in 75% ethanol. All specimens examined were dissected, mounted on microscope slides in phenol-Canada balsam as described by Wirth & Marston (1968), and then studied with the light microscope. The materials are deposited at the Department of Invertebrate Zoology and Parasitology, University of Gdańsk (Poland).

## RESULTS

# Subfamily Dasyheleinae

Dasyhelea bilineata Goetghebuer, 1920 (Fig. 1)

Material examined:

Slovakia occ., Prievidza district, Diviacka Nová Ves, Strážovské vrchy Mts., 7276 (DFS), site 1: VI.2012, *Fagus*, 2  $\Im$ , 3  $\Im$ , VII.2012, *Fagus*, 1  $\Im$ ; all J. Oboňa leg., P. Dominiak det.

# Comments:

Larvae of this species are known as inhabitants of phytotelmata, and were reported from small water reservoirs in leaf axils of teasel (*Dipsacus* sp.) (Disney & Wirth 1982). However, they usually develop in small, temporary rock pools and their artificial equivalents (e.g. rain gutters). Preimaginal stages of *Dasyhelea bilineata* are well adapted to periodical desiccation and wide fluctuations in temperature (Zilahi-Sebess 1931, Valkanov 1941, Disney 1975).

Adult males and females of *Dasyhelea bilineata* are very similar to *D. flavifrons* (Guérin, 1833). They differ slightly in the palpal ratio of the third palpal segment



**Fig. 1.** *Dasyhelea bilineata* Goetghebuer, 1920, male (a, b, d), female (c): a – palpus; b – femur and tibia of fore leg; c – subgenital plate and seminal capsule; d – male genitalia, ventral view

(length of palpal segment 3 divided by its greatest width), and in the body coloration. The shape of the female subgenital plate is also characteristic and allows identification of these two species (Dominiak 2012).

# Dasyhelea flavifrons (Guérin, 1833) (Fig. 2)

#### Material examined:

Slovakia occ., Prievidza district, Diviacka Nová Ves, Strážovské vrchy Mts., 7276 (DFS), site 1: 20.IV.2012, *Fagus*, 1  $\Im$ ; IV.2012; *Ulmus*, 7  $\Im$ , 6  $\Im$ , 9 V.2012, *Ulmus*, 8  $\Im$ , 9  $\Im$ , 9  $\Im$ ; VI.2012, *Fagus*, 6  $\Im$ , 9  $\Im$ ; Vrbany env., Diviacka Nová Ves, Hornonitrianska kotlina basin, 7277 (DFS), site 2: 23.III.2012, *Quercus*, 1  $\Im$ ; 28.III.2012, *Quercus*, 4  $\Im$ , 4  $\Im$ , 10. IV.2012, *Quercus*, 1  $\Im$ ; 3.V.2012. *Quercus*, 3  $\Im$ , 4  $\Im$ , 4  $\Im$ ; 21.V.2012, *Fraxinus*, 1  $\Im$ ; all J. Oboňa leg., P. Dominiak det.

# Comments:

Larvae of *Dasyhelea flavifrons* are terrestrial, and they can be found in sap oozing from various species





**Fig. 2.** *Dasyhelea flavifrons* (Guérin, 1833), male (a, b, d), female (c): a – palpus; b – femur and tibia of fore leg; c – subgenital plate and seminal capsule; d – male genitalia, ventral view

of trees and in tree holes. Adults are rather small, with the wing length less than 1.5 mm. The third palpal segment in both sexes is short, and the male palpal ratio is less than 4.0. The female is distinct in having an evenly sclerotized subgenital plate, with a notum in the shape of an equilateral triangle (Dominiak 2012).

Dasyhelea flavifrons is a widely distributed, Holarctic arboreal species (Dominiak & Szadziewski 2010). Although this specific name was listed from Slovakia by Országh (1984) (p. 66, as *D. dufouri*), the record had to be recognized as doubtful and omitted in recent publications (cf. Szadziewski & Dominiak 2006, Dominiak & Szadziewski 2010). Presently, *D. flavifrons* is confirmed in Slovakia

# Subfamily Ceratopogoninae

Culicoides fagineus Edwards, 1939 (Fig. 3a)

# Material examined:

Slovakia occ., Prievidza district, Diviacka Nová Ves, Strážovské vrchy Mts., 7276 (DFS), site 1: 30.V.2012, *Ulmus*, 1  $\Diamond$ ; VI.2012, *Fagus*, 6  $\Diamond \Diamond$ , 4  $\bigcirc \bigcirc$ ; VI.2012, Ulmus,  $3 \Diamond \Diamond$ ; VII.2012, Fagus,  $1 \heartsuit$ ; VIII.2012,  $1 \Diamond$ ,  $2 \heartsuit \heartsuit$ ; XI.2012, Fagus,  $1 \Diamond$ ; all J. Oboňa leg., P. Dominiak det.

#### Comments:

*Culicoides fagineus* breeds in sap oozing from trees and in tree holes (Edwards et al. 1939, Dzhafarov 1964, Glukhova 1979). Females of this species suck blood from various animals, including human (Gutsevich 1973).

Culicoides obsoletus (Meigen, 1818) (Fig. 3b, c)

## Material examined:

Slovakia occ., Prievidza district, Diviacka Nová Ves, Strážovské vrchy Mts., 7276 (DFS), site 1: XI.2012, *Fagus*, 1  $\stackrel{\circ}{\circ}$ ; J. Oboňa leg., P. Dominiak det.

#### Comments:

Immatures of *Culicoides obsoletus* live i.a. in mushrooms, tree holes, dung, compost heaps, wet



**Fig. 3.** *Culicoides fagineus* Edwards, 1939 (a), *C. obsoletus* (Meigen, 1818) (b, c), *C. truncorum* Edwards, 1939 (d, e); male (b–e), female (a): a, b – wing; c, d – male genitalia, ventral view; e – male genitalia, ventral root

soil (Szadziewski 1985) and, as suggested by Mellor & Pitzolis (1979), they can also dwell in banana plantations. In general, however, its development sites still remain poorly documented (Szadziewski 1985). Females of *C. obsoletus* are known to attack livestock and humans (Szadziewski 1985), but they can also readily feed on birds (Lassen et al. 2011). This very common and widely distributed species is susceptible to infection with BTV-8 and BTV-9, therefore it is considered to be a probable vector of the bluetongue virus in Europe (Carpenter et al. 2008). Females of *C. obsoletus* can also act as vectors of the Schmallenberg virus (Elbers et al. 2013).

#### Culicoides truncorum Edwards, 1939 (Fig. 3d, e)

## Material examined:

Slovakia occ., Prievidza district, Diviacka Nová Ves, Strážovské vrchy Mts., 7276 (DFS), site 1: IV.2012, Ulmus, 1  $\Diamond$ , 1  $\heartsuit$ ; VI.2012, *Fagus*, 1  $\heartsuit$ ; VI.2012, *Ulmus*, 2  $\Diamond \Diamond$ ; all J. Oboňa leg., P. Dominiak det.

## Comments:

Culicoides truncorum is known to develop in damp rotten wood and in tree holes (Edwards et al. 1939, Callot & Kremer 1961). In literature, it is usually quoted as C. sylvarum Callot & Kremer, 1961 (e.g. Gutsevich 1973), and all records of this species from peat bogs refer to C. clintoni Boorman, 1984 (cf. Callot & Kremer 1961: p. 394, Gutsevich 1973: p. 150). Adults of both species are similar, but differ i.a. in the distribution of sensilla coeloconica on female antennal flagellomeres, in the shape of maxillae and mandibles (both bearing teeth in C. truncorum), distribution of wing patterns and details of the structure of the male genitalia. Females of C. truncorum are ornithophilic, and they can attack their hosts also in the bird nest boxes (Votýpka et al. 2009).

#### DISCUSSION

As a result of the one-year study on biting midges associated with dendrotelmata, three species of the genus *Culicoides* and two species of the genus *Dasyhelea* were reported from these habitats. All three *Culicoides* species recorded in this study are known to develop in tree holes. *Culicoides fagineus* was the most abundant and dominated in summer and autumn. In contrast to that, *C. truncorum* was characterized by earlier occurrence, whereas *C. obsoletus* was obtained from the sediments only in autumn. *Dasyhelea flavifrons* was most abundant in spring and early summer. Another species of this genus, *D. bilineata*, was collected only during summer months. Larvae of this midge usually occur in phytotelmata or litothelmata, and are probably not a permanent inhabitants of tree holes.

The tree holes are important development sites for many insect larvae, including potential vectors of various diseases. It is worthwhile to take a note, that such habitats are often colonized by Culicoides, which are known to spread a number of pathogens of medical and veterinary importance (Linley 1985, Braverman et al. 1996). Haematophagous females of this genus can play a role as vectors of orbiviruses, which cause i.a. the African horse sickness (African horse sickness virus AHSV), bluetongue disease (BTV) and epizootic haemorrhagic disease (EHDV) (Mellor et al. 2000), as well as orthobunyaviruses, e.g. the Oropouche virus which infects humans, the Akabane virus (Mellor et al. 2000) and the novel Schmallenberg virus associated with a disease in ruminants (Elbers et al. 2013). One of the probable vectors of the bluetongue virus and the Schmallenberg virus in Europe is C. obsoletus, which was also recorded in the presented study. Further research on small lentic habitats such as tree holes, still poorly studied in Slovakia, are thus greatly justifiable.

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