# *Culicoides poperinghensis*, a new species of biting midge for the Netherlands (Diptera: Ceratopogonidae)

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**Faunistics, Malaise trap, hematophagy**

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**In May 2013, Wageningen University operated a Malaise trap to collect insects for a first year course in Biology. The Malaise trap was placed about 100 meters from the stream Kortenburgsebeek on the field of a skating rink west of the town of Renkum in the province of Gelderland. Unsorted material not used for the course was stored at Naturalis Biodiversity Center, Leiden. Among the Ceratopogonidae in this material, a species new for the Dutch fauna was discovered, *Culicoides poperinghensis*. Subsequently it appeared that the species was previously collected near Vorden, province of Gelderland, in 2008. *Culicoides poperinghensis* is not often recorded and its biology is poorly known. In this contribution we add *C. poperinghensis* to the fauna of the Netherlands, shortly discuss its recognition and describe what is known of its biology.**

## Introduction

The genus *Culicoides* of the dipteran family Ceratopogonidae includes the smallest hematophagous flies, which rarely exceed three millimeters in length (Mellor *et al.* 2000). Females *Culicoides* feed opportunistically on humans. The biting activities of these midges cause an enormous nuisance and can impact human activities such as agriculture, recreational tourism and forestry (Carpenter *et al.* 2013). However, the greatest economic impact lies in the ability of midges to transmit diseases such as blue-tongue virus (BTV), and African horse sickness virus (AHSV). AHSV can cause mortality levels of over 90% in low resistant equine populations (Mellor *et al.* 2000), while BTV can cause more than 70% mortality in sheep populations (Mullen & Durden 2009). BTV has spread through Europe since 2006 (Lassen *et al.* 2012) and caused great economic damage (Wilson & Mellor 2009).

Recently *Culicoides* biting midges have also been identified as vectors of the novel arboviruses *Orthobunyavirus* and Schmallenberg virus (Harrup *et al.* 2015). A member of the genus *Orthobunyavirus*, the Oropouche virus, causes febrile illness epidemics in humans in Southern and Central America, however, the probability of an epidemic outbreak in Europe is low (Carpenter *et al.* 2013). Symptoms of this nonfatal virus are fever and athralgia (joint pain) (Swanson 2012). Schmallenberg virus was discovered in cattle in Germany in 2011. The affected animals suffer from diarrhoea and reduced milk production (Carpenter *et al.* 2013).

*Culicoides* is the largest genus of the family Ceratopogonidae containing about 1400 species (Harrup *et al.* 2015). Despite the veterinary importance of *Culicoides* biting midges the taxonomy of this genus is far from being resolved. *Culicoides* is divided into 31 subgenera, which represent 63% of the extant species and 38 species groups (24% of the species). About 13% of *Culicoides* species are unplaced (Harrup *et al.* 2015). The subgeneric classification is based mainly on overal similarity and not on sound cladistics (Borkent 2014). Morphological identification is difficult even for specialists (Meiswinkel *et al.* 2015) and molecular tools are increasingly used to aid identification (Ander *et al.* 2012). Identification is also complicated by the lack of keys that cover all biogeographic regions (Swanson 2012). Moreover, the biology of most species of *Culicoides* is poorly known. Studies are mainly focused on species that have a deleterious impact. According to the website of Fauna Europaea (Szadziewski *et al.* 2013), 19 species of *Culicoides* occur in the Netherlands, while the website Nederlands Soortenregister reports 22 species (www.nederlandsesoorten.nl). In this article we add another species to the Dutch list, namely *C. poperinghensis* Goetghebuer, 1953 (figure 1).

## Biology of *Culicoides poperinghensis*

The life cycle of *Culicoides* biting midges consist of four stages: egg, four larval instars, pupa and adult (Carpenter *et al.* 2013). Larvae need moisture rich habitats to develop and moisture seems a key limiting factor. *Culicoides* larvae occur in various microhabitats, ranging from pond and lake shorelines to mud, damp soil, damp leaf litter, tree holes and livestock dung (Gonzàlez de Heredia & Lafuente 2011). The larvae are generalists feeding on diatomes, rotifers, algae, oligochaetes and other arthropods (Swanson 2012). Females feed on mammals and birds. Few species feed on arthropods and there are records of *Culicoides* feeding on turtles and frogs. Feeding behaviour of different species can be predicted by the distribution of sensilla coeloconica on the flagellomeres and the amount of sensilla basiconica on the third palpal segment. Mammalophilic species tend to have lesser sensilla coeloconica and sensilla basiconica.

Judging from the small size of its third palpal segment and the reduced number of sensilla coeloconica on the female

**1.** Habitus of male *Culicoides poperinghensis*. Renkum (province of Gelderland), May 2013 Photo: Pasquale Ciliberti
**1.** Habitus van mannetje *Culicoides poperinghensis*. Renkum (Gelderland), mei 2013.

**2.** Known distribution of *Culicoides poperinghensis* in the Netherlands
**2.** Verspreiding van *Culicoides poperinghensis* in Nederland.

antennae, *C. poperinghensis* is not ornithophilic. Males feed on nectar and their antennae have sensilla coeloconica plus a setal plume. The male third palpal segments is less developed compared to that of females.

*Culicoides* biting midges are generally active at dusk, although diurnal species also occur. *Culicoides poperinghensis* has been found feeding on cattle in Denmark (Lassen *et al.* 2012) and in France (Ninio *et al.* 2011). Santiago-Alarcon *et al.* (2012) report *C. poperinghensis* feeding on humans in Southwestern Germany. So far, *C. poperinghensis* has not been implicated in the transmission of BTV. This species is far too rare to play a major role in the transmission of economically important arboviruses, although its vectorial capacity has not been tested. Foxi & Delrio (2010) reared two larvae from a mud sample taken from a shallow pond shoreline, which was covered by short grass.

## Discovery in the Netherlands

In 2013, Wageningen University operated a Malaise trap near Renkum, in the province of Gelderland. The Malaise trap was placed about 100 meter from the stream Kortenburgsebeek on the field of a skating rink west of the town of Renkum. The insects captured were used for an undergraduate course on biodiversity. After the course, the material was deposited in the national natural history collection of Naturalis Biodiversity Center in Leiden. This material was examined by the first author and some specimens belonging to the Ceratopogonidae were kept separately for further identification. The specimens from this subsample were later identified as *C. poperinghensis*. Furthermore the private collection of the second author contains a specimen that was collected in 2008 with a light trap in Vorden, also in the province of Gelderland. Specimens were cleared in KOH 10% and slide mounted. Keys used for identification were the one produced by Campbell & Pelham-Clinton (1960) and Delécolle (1985).

## Distribution

*Culicoides poperinghensis* is a Palaearctic species, which is known from Europe, Northwest Africa (Algeria), and Eastern Siberia (Remm 1988). According to Fauna Europaea, *C. poperinghensis* is present in the following European countries: Belgium, Great Britain and Ireland, Denmark, mainland France, Germany, Romania, Spain, Sweden and Georgia (Szadziewski *et al.* 2013). Moreover, it is known from the islands of Corsica and Sardinia, while it is apparently absent from the Italian mainland.

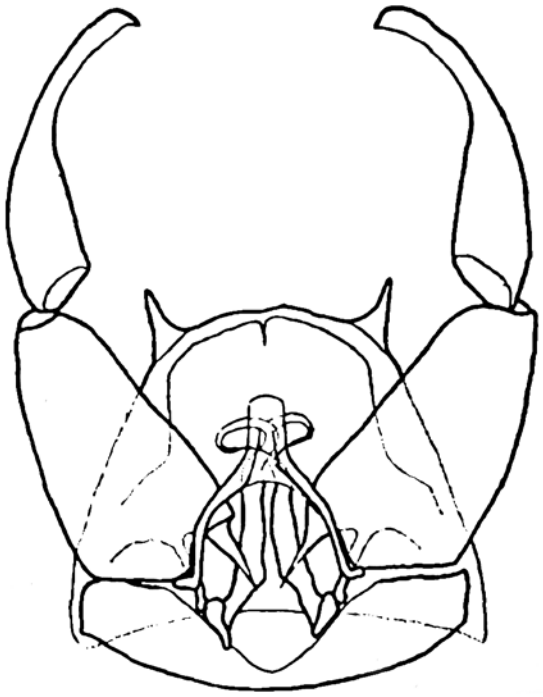
*Culicoides poperinghensis* is not often collected and this probably limits our knowledge of its distribution and explains its patchy distribution in Western Europe. For example, in a study using $CO_2$ baited counterflow traps on population dynamics and phenology of *Culicoides* in the Netherlands, no *C. poperinghensis* were collected (Takken *et al.* 2008). Campbell & Pelham-Clinton (1960) report that the bulk of their specimens were from the salt-marsh area of the Tay estuary in Scotland. They claim that *C. poperinghensis* could be exclusively a salt marsh species. This could explain why the species is rarely collected inland. However, the type locality is Poperinghe, which is 20 miles from the Belgian coast (Goetghebuer 1953). The Dutch localities reported in this article are also far from the coast (figure 2). It could be that optimal development takes place in a salty environment and that occasionally small populations arise from migrants.

## Recognition

Material: Gelderland, Vorden, 52°06'45 N 6°18'57 E, 1 ♀ collected between 22:45 and 23:00 on 31.v.2008 by E.G.M Dijkstra using a light trap; Gelderland, Renkum, Kortenburgsebeek, GPS 51°58'23 N 5°43'13 E, 2 ♂ collected between 15-30.v.2013 by Wageningen University using a Malaise trap.

*Culicoides* biting midges belong to the tribe Culicoidini of the subfamily Ceratopogoninae. In Europe, *Culicoides* is the only


**3.** Wing of a male *Culicoides poperinghensis*. Renkum (province of Gelderland), May 2013. Photo: Jan Muilwijk
**3.** Vleugel van mannetje *Culicoides poperinghensis*. Renkum (Gelderland), mei 2013.



**4.** Genitals of male *Culicoides poperinghensis*. Source: Campbell & Pelham-Clinton 1960
**4.** Mannelijke genitaliën van *Culicoides poperinghensis*. Bron: Campbell & Pelham-Clinton 1960

genus in the tribe Culicoidini. Major characters of this tribe are the presence of two radial cells which are more or less equal in size (figure 3), the presence of macrotrichia on the wings and very often the presence of a colour pattern on the wing membrane.

Species of *Culicoides* can be identified using a set of characters. To distinguish the females, the wing pattern and the distribution of macrotrichia is sometimes used (Rawlings 1996). Female *C. poperinghensis* have two spermathecae, which are oval-shaped and approximately equal in size. The eyes are well separated. Sensilla coeloconica are present on antennal segments 11 to 15 and on the third antennal segment. The third palpal segment is only slightly swollen and the sensilla basiconica are distributed in a few shallow pits. The most distinctive character to recognize female *C. poperinghensis* is however the shape of the atrophied sensilla coeloconica on segments 11 to 15. The pits of the sensilla are surrounded by only two to three macrotrichia compared to more than four macrotrichia of other *Culicoides* species (Gonzàlez de Heredia & Lafuente 2011). Males are identified by the shape of the genitals. Male genitals of *C. poperinghensis* are specific by having a broad and slightly emarginate ninth tergite, two short apicolateral processes and by the form of the aedeagus and parameres (figure 4). There is a discrepancy in the description of the wing in the original publication of Goetghebuer (1953) and in the later revision of the genus by Campbell & Pelham-Clinton (1960). The original description of Goetghebuer (1953) is based on only a male specimen, while the revision of Campbell & Pelham-Clinton (1960) is based on 70 females and 39 males. They give a detailed account of the distribution of macrotrichia on the wing membrane. The latter authors describe the wing as pale grey with hardly distinguishable pale spots. Of these spots, the one on the apical third of the second radial cell is the key diagnostic character that distinguishes *C. poperinghensis* from other British species. In contrast, Goetghebuer (1953) described his specimen as having a pale wing without markings or pale spots except for a dark spot on the second radial cell. In the interpretation of Campbell & Pelham-Clinton (1960) the dark spot described by Goetghebuer (1953) is a mistake.

The female collected at Vorden was characterized by having an indistinct and diffuse pattern on the wing membrane. A pale spot could be distinguished on the second radial cell. In the specimens collected at Renkum (figure 3), the wing pattern was indistinguishable, at least with the microscope used by the first author. In addition, the macrotrichia were lost, probably due to the prolonged time that specimens were stored in alcohol. Identification of these specimens was based on the male genitalia.

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## Samenvatting

***Culicoides poperinghensis*, een nieuw knutje voor Nederland (Diptera: Ceratopogonidae)**
In mei 2013 verzamelde Wageningen Universiteit insecten met een Malaiseval voor een eerstejaars cursus Biologie. De val stond op ongeveer 100 meter van de Kortenburgsebeek op het terrein van een schaatsbaan ten westen van Renkum, provincie Gelderland. Ongesorteerd materiaal dat niet voor de cursus werd gebruikt, werd opgeslagen in Naturalis Biodiversity Center in Leiden. Tussen de Ceratopogonidae in dit materiaal werd een voor de Nederlandse fauna nieuwe soort aangetroffen: *Culicoides poperinghensis* Goetghebuer, 1953. Het bleek dat de soort in 2008 eerder was verzameld bij Vorden, eveneens in Gelderland. *Culicoides poperinghensis* wordt niet vaak aangetroffen en over de biologie is weinig bekend. In deze bijdrage bespreken we hoe de soort kan worden herkend en beschrijven we wat er bekend is over de biologie.

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