THE FIRST PILOT STUDY OF BLACK FLY (DIPTERA: SIMULIIDAE) BREEDING SITES IN MOLDOVA

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Abstract: *The data on black fly fauna in Moldova are absent in the old and modern literature. Overall, 45 black fly species from the genera Prosimulium (four species) and Simulium (41 species) are common both for Romania and Ukraine and expected to be present in Moldova. For the first time cross-sectional entomological survey was conducted in 2018 to detect the breeding sites of Simuliidae in Moldova. Productive breeding sites of black flies were identified in the creek close to the Dniester River section located in Vascauti village, Racovat River sections located in Branzeni and Burlanesti villages. Immature stages of black flies also have been found in the small creeks in Chisinau and Duruitoarea Veche.*

Key words: *black fly, Simuliidae, Republic of Moldova*

INTRODUCTION

Black flies (Diptera: Simuliidae) are found almost everywhere with running water suitable as a habitat for their aquatic stages and distributed in all zoogeographical regions. According to the last version of the Inventory of the world's Simuliidae, the family includes 26 genera and 2351 species (2335 living and 16 fossils) [1]. The females of most black fly species feeding on blood of humans, mammals and birds being the main vectors of some human and animal pathogens. Among the blood-borne parasites most commonly transmitted by black flies are the blood protozoan of the genera *Leucocytozoon* that cause leucocytozoonosis in birds, the filarioid nematodes of the genus *Onchocerca* that cause human onchocerciasis, mansonellosis and bovine onchocerciasis and viruses like vesicular stomatitis in cattle and horses. Some of these diseases cre-

ate severe economic declines and public health threats [5]. Larvae and pupae are aquatic and may be used as water quality indicators [3].

To the best of our knowledge, the data on black fly fauna in Moldova are absent in the old and modern literature. The closest well known black fly faunas of Romania and Ukraine consist of approximately 102-111 species belonging to six genera such as *Prosimulium*, *Metacnephia*, *Twinnia*, *Cnephia*, *Simulium* and *Stegopterna* [1]. Overall, 71 black fly species from three genera (*Prosimulium*, *Metacnephia* and *Simulium*) were recorded in Romania, including eight species which presence in the country is questionable [1; 2; 4]. In total, 92 black fly species belonging to five genera (*Prosimulium*, *Twinnia*, *Cnephia*, *Simulium* and *Stegopterna*) were recorded in Ukraine (species from Crimean peninsula have not been excluded from the list) [1; 6; 7; 10]. Overall, 45 black fly species from the genera *Prosimulium* (four species) and *Simulium* (41 species) such as *Prosimulium hirtipes* (Fries, 1824), *P. latimucro* (Enderlein, 1925), *P. rufipes* (Meigen, 1830), *P. tomosvaryi* (Enderlein, 1921), *Simulium erythrocephalum* (De Geer, 1776), *S. maculatum* (Meigen, 1804), *S. angustipes* Edwards, 1915, *S. aureum* Fries, 1824, *S. krymense* (Rubtsov, 1956), *S. rubzovianum* (Sherban, 1961), *S. angustitarse* (Lundström, 1911), *S. angustatum* (Rubtsov, 1956), *S. codreanui* (Sherban, 1958), *S. costatum* Friederichs, 1920, *S. crenobium* (Knoz, 1961), *S. verum* Macquart, 1826, *S. nigrum* (Meigen, 1804), *S. argenteostriatum* Strobl, 1898, *S. bezzii* (Corti, 1914), *S. bukowskii* Rubtsov, 1940, *S. noelleri* Friederichs, 1920, *S. baracorne* Smart, 1944, *S. deserticola* Rubtsov, 1940, *S. frigidum* Rubtsov, 1940, *S. intermedium* Roubaud, 1906, *S. kiritshenkoi* Rubtsov, 1940, *S. ornatum* Meigen, 1818, *S. rotundatum* (Rubtsov, 1956), *S. reptans* (Linnaeus, 1758), *S. voilense* Sherban, 1960, *S. vulgare* Dorogostaisky, Rubtsov, Vlasenko, 1935, *S. argyreatum* Meigen, 1838, *S. maximum* (Knoz, 1961), *S. monticola* Friederichs, 1920, *S. monticoloides* (Rubtsov, 1956), *S. variegatum* Meigen, 1818, *S. morsitans* Edwards, 1915, *S. paramorsitans* Rubtsov, 1956, *S. auricoma* Meigen, 1818, *S. balcanicum* (Enderlein, 1924), *S. equinum* (Linnaeus,

1758), *S. lineatum* (Meigen, 1804), *S. paraequinum* Puri, 1933, *S. pseudoequinum* Séguy, 1921 and *S. veltistshevi* Rubtsov, 1940 are common both for Romania and Ukraine and expected to be present in Moldova [1]. Moreover, 44 valid species were reported from Ukraine, which have not been registered in Romania and approximately 18-25 species recorded in Romania have not been found in Ukraine. Due to the absence of any old and recent data on black fly species diversity, spatial distribution and public health importance in Moldova, new surveys are particularly needed in the country.

MATERIALS AND METHODS

For the first time cross-sectional entomological survey was conducted between May and August 2018 to detect the aquatic habitats of Simuliidae at 15 localities (Plaiul Fagului, Zberoaia, Grozesti, Vascauti, Saharna, Rezina, Ciorna, Costesti, Duritoarea Noua, Branzeni, Burlanesti, Sturzovca, Tomai, Ceadir-Lunga, Corten) from nine regions (Calarasi, Nisporeni, Orhei, Rezina, Rascani, Glodeni, Edinet, Ceadie-Lunga, Taraclia) in Moldova. Each creek and river section (30-40 m) was sampled once from downstream to upstream by two people during 1-2 hours. For each breeding site the following characteristics were recorded: depth (m), width (m), water temperature (°C), water salinity (PSU) and pH. Each collected site was numbered and mapped using global positioning system (GPS). Larvae attached to the vegetation and stones were collected by pincette and transferred to 96% and then to 70% ethanol solution. Pupae attached to the same substrates were collected together with a piece of substrate (leaf, stem) and transferred to the plastic containers. In laboratory, the mature pupae still attached to the substrate were individually kept alive in vials with highly wet atmosphere until the emerging of the adults from their cocoons. In total, more than 200 adult specimens have been obtained from about 500 pupae. The adults together with their pupal

exuviae and cocoons were kept in 96% ethanol for further species identification. Permanent slides have been prepared from male genitalia, legs, wings and the heads. At present the black fly adults are subjected to the species identification according to taxonomic keys of Rubtsov (1956), Rubtsov and Yankovsky (1984) and Yankovsky (2001) [8; 9; 11]

RESULTS AND DISCUSSIONS

Productive breeding sites of black flies were identified in the creek close to the Dniester River section located in Vascauti village, Racovat River sections located in Branzeni and Burlanesti villages. Immature stages of black flies also have been found in the small creeks in Chisinau and Duriutoarea Veche. In the creek and river habitats with running water where the bottom was sandy/muddy the immature stages were found attached only to the leaves and branches of aquatic plants like *Potamogeton* spp. In the river sections with stony bottom structure immature stages were also found attached to the submerged stones. The high densities of immature stages were found on vegetation substrates not deeper than 10 cm below the water surface at water temperature 24-28°C in Racovat River and the creeks in Chisinau. Due to a high plastic pollution of streams in Chisinau, Simuliidae immature stages were also found attached to the plastic cups. Immature stages of black flies were absent in the creeks with cold running water (about 18°C) located in Saharna and Ciorna villages, where *Anopheles claviger* Meig. larvae have been collected. In our surveys black flies also have not been found in the rivers Prut and Dniester most probably because of the low speed of water flow, but further surveys are needed. The Simuliidae generally inhabit unpolluted running water, being the important components of the stream ecosystem and having the ability to filter dissolved organic matter. Simuliidae larvae and pupae are susceptible to organic and inorganic pollution and may play an important role in the monitoring of freshwater contamination [3].

CONCLUSIONS

Preliminary results revealed the presence of black flies in some aquatic habitats in Moldova, which physicochemical parameters were suitable for immature Simuliidae. Further surveys in the region and the results of species identification will help to understand the Simuliidae species diversity in the Republic of Moldova, breeding habitat preferences, spatial distribution and public health and veterinary importance of black flies as vectors of vector-borne diseases.

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