

**10-Year Observation of Alien Mysids and Amphipods in Belarus**

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**Aim of the study:** Analysis of own and published data for 10-year monitoring of alien mysids and amphipods and updating of recently published data.

**Material and Methods:** Alien mysids and amphipods have been studying in Belarus since 2006. All the sampling sites were located in public and non-protected areas. No permissions were required for sampling. Samples were taken by hand net (mesh size: 500µm, diameter: 25×25cm) and/or picked by hand from submerged macrophytes, stones, wood and other natural or artificial items floating or submerged in the water. Crustaceans were fixed by 98% ethanol. The specimens were identified based on Cărauşu et al. (1955), Morduchai-Boltovskoy (1969) and Dobson (2013). Laboratory operations were conducted at the Canadian Centre for DNA Barcoding (CCDB), University of Guelph. DNA was extracted from whole legs of 380 crustaceans using an automated silica-based protocol with glass fibre filtration plates (Ivanova et al., 2006). The 658-bp barcode region of the mitochondrial cytochrome c oxidase subunit 1(CO1) gene was amplified by Lepidoptera and Folmer primer cocktail (C\_LepFolF / C\_LepFolR, 1:1) and crustacean primer CrustDF1 / CrustDR1. The polymerase chain reaction (PCR) was performed in 12.5 µL volume containing 6.25µL 10% trehalose, 2µL ddH<sub>2</sub>O, 1.25µL 10× PCR buffer, 0.625µL MgCl<sub>2</sub> (50 mM), 0.125µL of each primer (10µM), 0.0625µL dNTPs (10 mM), 0.06µL Platinum Taq polymerase (5 U/µL) and 2µL DNA template. The thermocycling regime used for all reactions was: initial denaturation at 94°C for 1 min, 5 cycles of 94°C for 40 s, 45°C for 40 s and 72°C for 1 min, followed by 35 cycles of 94°C for 40 s, 51°C for 40 s and 72°C for 1 min, and a final step of 72°C for 5 min. Bidirectional sequencing followed CCDB sequencing protocols using BigDye 3.1 (Ivanova & Grainger, 2007).

**Results:** The revision of publications during 10 years of observation alien mysids and amphipods was done. Some details of records and distribution of alien species were corrected. Nowadays, two species of Ponto-Caspian mysids and nine species of Ponto-Caspian amphipods are known to have established in the water bodies of Belarus. The main hotspot of alien mysids and amphipods in Belarus is the Dnieper River near Niznie Zhary vill., where eleven alien species were recorded. The new records of *Echinogammarus trichiaus*, *Chelicorophium curvispinum*, *Ch. robustum* and *Paramysis lacustris* were pointed out in Belarus. The upper sites of alien mysids and amphipods distribution were revealed in the main waterways in Belarus. DNA sequences were obtained from nine alien amphipods and alien mysid *Limnomysis benedeni*. Identification of *E. trichiatus* was proved by DNA barcoding.

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**Keywords:** Alien mysids and amphipods, monitoring, barcoding, Belarus.