Study of the Culex pipiens species complex distributed in Georgia using the DNA barcoding method

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Annotation

In tropical, temperate, and Holarctic regions of the world, mosquitoes of the genus *Culex* serve as vectors of arboviral diseases affecting both humans and animals. These mosquitoes possess the capacity to trigger epidemiological outbreaks, underscoring the importance of accurate species identification for assessing disease transmission risks. However, the presence of cryptic species and overlapping morphological traits renders traditional identification methods unreliable. In this context, DNA barcoding has emerged as a promising tool for precise and efficient species discrimination.

Among individuals identified as *Culex pipiens* and collected annually across various regions of Georgia, distinct morphological groups are periodically observed. This recurring phenotypic variation raises the question of whether these differences are rooted in underlying genetic divergence. Therefore, it is essential to investigate whether the observed morphological variation corresponds to genetic differentiation among these populations.

The aim of this study is to determine the species affiliation of morphologically distinct groups within the *Culex pipiens*species complex found in Georgia. Additionally, the study seeks to validate the morphological identification criteria currently used in the region by employing molecular techniques.

To achieve this objective, four morphologically distinct groups of *Culex pipiens* Linnaeus, 1758 were selected for analysis. DNA was extracted from the samples, and DNA barcoding was performed using next-generation sequencing. The mitochondrial cytochrome c oxidase subunit I (COI) gene region was targeted for this study, as it is commonly used for species identification. To ensure reliable amplification, “universal” primers LCO1490 and HCO2198 were employed, as these primers are known for their ability to effectively distinguish closely related species.

The molecular genetic analysis revealed the presence of *Culex torrentium*, a species within the *Culex pipiens* complex, in Georgia for the first time. This discovery suggests a potentially higher level of species diversity in the region than previously recognized and underscores the critical importance of molecular approaches in identifying species that may be overlooked through morphological methods alone.