

Toruń, 08/09/2025

city, date

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DOCTORAL DISSERTATION ABSTRACT

Scientific discipline: **Biological Sciences**

Title of the doctoral dissertation: **Reconstructing Evolutionary Histories of Calliphoridae (Diptera: Cyclorrhapha: Oestroidea) at Different Taxonomic Levels Using Phylogenomic Methods**

Doctoral dissertation abstract:

Blowflies (Diptera: Calliphoridae) represent a diverse, globally distributed family of calyptrate flies comprising ~2,075 species across 154 genera, representing 9% of calyptrate diversity and 1.3% of all described Diptera. Despite their ecological, medical, veterinary and economic significance as decomposers, pollinators, parasites, and vectors, blowfly diversity remains underestimated, and genomic resources are strikingly limited. Only ~1.6% of species have sequenced genomes, lagging behind other dipteran families, such as mosquitoes (Culicidae, 4%) and tsetse flies (Glossinidae, ~26%). These gaps limit our understanding of the evolutionary history of this group, explaining their ecological adaptations, and elucidating their roles in the transmission of disease-causing pathogens. This dissertation synthesizes multifaceted genomic, phylogenetic, and taxonomic studies aimed at filling significant knowledge gaps across the most important blowfly subfamilies. Leveraging large-scale mitochondrial datasets, it addresses systematic uncertainties and traces biogeographic and evolutionary patterns in key Calliphoridae lineages, with particular emphasis on the subfamilies Calliphorinae and Luciliinae.

A critical review of existing genomic resources for Calliphoridae in public databases reveals a clear tendency for researchers to focus on species of medical, veterinary and forensic importance. Other groups—often with diverse and highly interesting life strategies, including various forms of

parasitism—are often overlooked. This highlights the urgent need to expand genomic data acquisition to a more representative range of taxa. Despite the availability of data for many key species of medical, veterinary, and forensic importance, the subfamilies Calliphorinae and Luciliinae continue to suffer from instability in the current taxonomy, uneven sampling of important genera, and a lack of genomic data even for common and widespread species. This limits the ability to obtain highly supported phylogenies and, consequently, infer the biogeography or trait evolution of these insects (Parmar et al. 2025a).

In response to the challenges outlined above, analyses of mitochondrial genomes were conducted for the broadest set of species from the Calliphorinae and Luciliinae subfamilies to date. One of the objectives was a comparative analysis of the mitochondrial genomes of 78 blowfly species, including 63 species, representing Calliphorinae subfamily. This analysis revealed extensive structural and functional diversity in the mitogenome of individual species, such as gene rearrangements (e.g., in *Calliphora varifrons* Malloch) and base composition biases. The Phylogenomic reconstructions strongly support the monophyly of Calliphoridae. Crucially, it also resolves the long-debated relationships between the numerous blowfly subfamilies. An important result is the demonstrated paraphyly of some large Calliphorinae genera (e.g., *Calliphora* Robineau-Desvoidy and *Onesia* Robineau-Desvoidy), indicating an urgent need for taxonomic revisions (Parmar et al. 2025b).

The species-rich genus *Calliphora* Robineau-Desvoidy is of particular interest to researchers from various fields, including medical, veterinary, and forensic entomology. This research expanded the reference database of COI barcode sequences for Palearctic and Afrotropical species of this genus, adding 33 new sequences to the publicly available databases. Using this data, species delimitation for the taxa included in the analysis was tested. Using integrative taxonomy methods, two new *Calliphora* species were described from the highlands of Ethiopia (*Calliphora teraramma* and *Calliphora mesay*), a groundbreaking finding considering the previous presence of only one species of this genus in the Afrotropical Region. These results highlight the usefulness of molecular tools in revealing cryptic diversity and represent a significant contribution to the knowledge of blowfly diversity in the Afrotropical Region (Parmar et al. 2025c).

Leveraging the most comprehensive mitogenomic dataset of Luciliinae to date (32 species), the phylogenetic relationships of these flies were reconstructed. The resulting phylogenetic tree allowed us to trace the evolutionary history of the Luciliinae, determine the timing of divergence of the main evolutionary lineages, and identify ancestral bionomics. Analysis of the mitochondrial genome of Luciliinae, a group with remarkable trophic plasticity, revealed a surprising conservatism of the mitogenome. Results indicated a paraphyletic nature of the globally distributed genus *Lucilia* Robineau-Desvoidy. The resulting phylogenetic tree topology shows that all species of the other genera included in the analysis remain nested within this genus: *Blepharicnema* Macquart (1 species), *Hemipyrellia* Townsend (2 species), and *Hypopygiopsis* Townsend (4 species). This result challenges the current classification and indicates an urgent need for taxonomic revision. Dating the divergence of the major evolutionary lineages indicates that subfamily-level divergences occurred in the Oligocene–Miocene, and the subsequent radiation of Luciliinae was likely associated with global



climatic changes and new ecological niches created by the expansion of herbaceous vegetation and associated fauna. The Afrotropical region likely formed part of the ancestral range of the Luciliinae (~15–20 Ma), and subsequent diversification of its sister lineage gave rise to all the remaining Luciliinae in other geographic regions. Parasitism arose independently and repeatedly during Luciliinae evolution. Reconstructions of the ancestral life strategy indicate saprophagy as the ancestral habit, while obligate parasitism evolved convergently in the Late Pliocene (Parmar et al. 2025d, submitted).

Collectively, this dissertation significantly expands our understanding of blowfly phylogeny, systematics, evolution and biogeography. It also significantly fills the gap in our knowledge of the variability of the mitochondrial genome of Calliphoridae. The resulting phylogenetic reconstructions will provide a sound basis for future studies on trait evolution and necessary taxonomic revisions. The barcoding libraries enriched by this dissertation's contributions will significantly facilitate species identification in practice and research in medical, veterinary, and forensic entomology. The presented research also provides data for models tracking potential range shifts of parasitic species caused by climate change. This work highlights the importance of foundational taxonomic and genomic research on this globally important family of flies in addressing practical challenges related to biodiversity conservation and public health.

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