The complete mitochondrial genome of *Melanostoma mellinum* (Linnaeus, 1758) (Diptera: Syrphidae) and phylogenetic analysis

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**ABSTRACT**

In this study, the complete mitochondrial genome (mitogenome) of *Melanostoma mellinum* (Linnaeus, 1758) was sequenced using next-generation sequencing technology. The assembled mitogenome of *M. mellinum* has a total length of 16,055bp and contains 13 protein-coding genes (PCGs), 22 transfer RNA genes (tRNAs), and 2 ribosomal RNA genes (rRNAs). The results of phylogenetic reconstruction based on the combined mitochondrial gene dataset indicated that *M. mellinum* belongs to *Melanostoma* genus with a close relationship to *Melanostoma orientale*, but the monophyly of the tribe Bacchini is not well supported.

Syrphidae (Insecta, Diptera) comprise almost 6200 described species worldwide (Evenhuis and Pape 2021) and are well-known by nature enthusiasts and researchers because they provide several crucial ecosystem services (Dunn et al. 2020), such as pollination, biological control of pests and organic matter recycling (Inouye et al. 2015; Moerkens et al. 2021). Among the subfamily Syrphinae, *Melanostoma mellinum* (Linnaeus, 1758) is a small hoverfly with yellow and black markings, which can be well distinguished from other *Melanostoma* species by the following morphological features: face, scutellum, and mesonotum shining black; male adults have nearly square yellow markings on abdominal segments 3 and 4; female adults have inverted yellow triangle markings on abdominal segments 3 and 4 (Huo et al. 2007). The systematic status of the genus *Melanostoma* is still controversial, as this genus has been placed into the subfamily Melanostominae (Williston 1885), tribe Melanostomini (Hull and Riley 1949; Vockeroth 1969, 1990), or classified into the tribe Stenosyrphini (Goffe 1952), or Bacchini (Rotheray and Gilbert 1989). The monophyletic status of the tribe Bacchini is also challenged by the molecular phylogenetic works (Thompson and Skevington 2014; Stähls et al. 2003). In this study, we obtained the complete mitogenome data of *M. mellinum* and build the phylogenetic tree to reconstruct its evolutionary relationships.

The specimen of *M. mellinum* was collected from the Changqing National Nature Reserve (107°17′E, 33°19′N) in 2019, and stored in the Museum of Zoology and Botany, Shaanxi University of Technology, Hanzhong, China (SUHC) (Accession number: R81, https://www.snut.edu.cn/, Le Zhao, email: Lezhao@snut.edu.cn). The genomic DNA of *M. mellinum* was extracted using the DNeasy kit (Qiagen, Hilden, Germany), then paired-end libraries (2 × 150 bp) with 400 bp insert sizes were constructed and sequenced by an Illumina HiSeq 4000 platform. We used the software MITOZ (Meng et al., 2019) to assemble and annotate the complete mitogenome, with the putative control region being delineated by tRNA boundaries.

The complete mitogenome of *M. mellinum* was 16,055 bp in length, including 37 typical mitochondrial genes (13 PCGs, 22 tRNAs, 2 rRNAs) and a putative AT-rich control region (D-loop), which has been deposited in GenBank (accession number: OK032510). The base composition of *M. mellinum* mitogenome was 41.2% A, 40% T, 10.6% C, and 8.2% G, with a positive AT-skew (0.014) and a negative GC-skew (−0.123), and all genes were arranged in the same order like other syrphids (Le and Gang, 2020; Zhou et al. 2021). Eight overlaps and 8 intergenic spacers were found in the mitogenome of *M. mellinum*, with the longest intergenic spacer of 23 bp located between tRNA1 and rRNA. All 13 PCGs used ATN as the start codon (cox1, atp6, and nad1 used ATA, nad2, nad3, nad5 and nad6 used ATT, cox2, cox3, nad4, nad4L, and cytB used ATG, atp8 used ATC), a total of 12 PCGs used TAA as the stop codon except ND3, which stopped with TAG.

To check the phylogenetic status of *M. mellinum*, we reconstructed a phylogenetic tree using all available mitogenome sequences of subfamily Syrphinae species in the NCBI database, and two Eristalinae species were used as an outgroup. Sequence alignments of 13 PCGs were generated by software MAFFT v7.313 with the E-INS-I strategy (Katoh and Standley 2013), and the best fit model of the partition scheme was determined by program PartitionFinder2 v2.1.1 with AICc scoring criteria (Lanfear et al. 2017). Phylogenetic trees were inferred with Bayesian inference (BI) and maximum likelihood (ML) by...
MrBayes v3.2.6 (Ronquist et al. 2012) and IQ-tree v1.6.8 (Nguyen et al. 2015), respectively. The tree topologies reconstructed by ML and BI methods were consistent (Figure 1) and supported the close relationships of *M. mellinum* with *M. orientale* within Melanostoma genus, but the monophyly of tribe Bacchini is not supported.

**Ethical approval**

Ethics approval was not required for this study. All procedures performed in this study involving animals followed the Guidelines for Shaanxi University of Technology, Hanzhong, China. The field studies did not involve vertebrates, regulated invertebrates, endangered or protected species.

**Author contribution**

Le Zhao, Keke Huo and Gang Li conceived and designed the experiments; Yicheng He performed the experiments; Le Zhao and Han Yue Liu analyzed the data; Le Zhao and Han Yue Liu wrote the paper.

**Disclosure statement**

No potential conflict of interest was reported by the author(s).

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**Data availability statement**

The mitogenome sequence data that supported the findings in this study are openly available in GenBank of NCBI at [https://www.ncbi.nlm.nih.gov/](https://www.ncbi.nlm.nih.gov/) under the accession no. OK032510. The associated SRA, BioProject and BioSample numbers are SRR19141043, PRJNA836006 and SAMN2812889, respectively. The specimen was deposited at the Museum of Zoology and Botany, Shaanxi University of Technology, Hanzhong, China ([https://www.snut.edu.cn/](https://www.snut.edu.cn/), Le Zhao, email: Lezhao@snut.edu.cn).

**References**

Dunn L, Lequerica M, Reid C, Latty T. 2020. Dual ecosystem services of syrphid flies (Diptera: Syrphidae): pollinators and biological control agents. Pest Manag Sci. 76(6):1973–1979.

Evenhuis NL, Pape T. 2021. Systema Dipterorum. The biosystematics database of world Diptera. Version 3.4. Last updated: 27 October 2021. 

Goffe ER. 1952. An outline of a revised classification of the Syrphidae (Diptera) on phylogenetic lines. Trans Soc Br Entomol. 11:97–124.

Hull FM, Riley ND. 1949. The morphology and inter-relationship of the genera of syrphid flies, recent and fossil. Trans Zool Soc Lond. 26(4):257–408.

Huo K, Ren G, Zheng Z. 2007. Fauna of Syrphidae from Mt. Qinling-Bashan in China (Insecta: Diptera). Beijing: Chinese Agricultural Science and Technology Press, 512 pp.

Inouye DW, Larson BMH, Ssymank A, Kevan PG. 2015. Flies and Flowers III: ecology of foraging and pollination. J Poll Ecol. 16:115–133.

Lanfear R, Frandsen PB, Wright AM, Senfeld T, Calcott B. 2017. PartitionFinder2: new methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. Mol Biol Evol. 34(3):772–773.

Le Z, Gang L. 2020. The first complete mitochondrial genome of the tribe Rhingiini (Diptera: Syrphidae) and phylogenetic analysis. Mitochondrial DNA Part B. 5(3):3489–3509.

Meng G, Li Y, Yang C, Liu S. 2019. MitoZ: a toolkit for animal mitochondrial genome assembly, annotation and visualization. Nucleic Acids Res. 47(11):e63–e63.

Moerkens R, Boonen S, Wäckers FL, Pelas A. 2021. Aphidophagous hoverflies reduce foxglove aphid infestations and improve seed set and fruit yield in sweet pepper. Pest Manag Sci. 77(6):2690–2696.

Nguyen LT, Schmidt HA, Von Haeseler A, Minh BQ. 2015. IQ-TREE: a fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. Mol Biol Evol. 32(1):268–274.

Ronquist F, Teslenko M, Van Der Mark P, Ayres DL, Darling A, Hohna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP. 2012. MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Syst Biol. 61(3):539–542.

Rotheray GE, Gilbert FS. 1989. The phylogeny and systematics of European predacious Syrphidae (Diptera) based on larval and puparial stages. Zool J Linn Soc. 95(1):29–70.

Stählis G, Hippa H, Rotheray G, Muona J, Gilbert F. 2003. Phylogeny of Syrphidae (Diptera) inferred from combined analysis of molecular and morphological characters. Syst Entomol. 28(4):433–450.

Katoh K, Standley DM. 2013. MAFFT multiple sequence alignment software version 7: improvements in performance and usability. Mol Biol Evol. 30(4):772–780.

Thompson FC, Skewington JH. 2014. Afrotropical flower flies (Diptera: Syrphidae). A new genus and species from Kenya, with a review of the Melanostomine group of genera. Zootaxa. 3847(1):97–114.

Vockeroth JR. 1969. A revision of the genera of the Syrphini (Diptera: Syrphidae). Mem Entomol Soc Can. 101(562):5–176.

Vockeroth JR. 1990. Revision of the Nearctic species of *Platychirus* (Diptera, Syrphidae). Can Entomol. 122(4):659–766.

Williston SW. 1885. On classification of North American Diptera. (First paper). Bull Brooklyn Entomol Soc. 7:129–139.

Zhou Z, Liu H, Gang L, Dang L, Zhao L, Huo K. 2021. Characterization and phylogenetic analysis of the complete mitochondrial genome of *Lathyrophthalmus quinquestratius* (Fabricius, 1794)(Diptera, Syrphidae). Mitochondrial DNA B Resour. 6(3):1183–1185.