

The complete mitochondrial genome of *Dolichopus galeatus* Loew, 1871 (Diptera: Dolichopodidae)

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ABSTRACT

The long-legged fly *Dolichopus galeatus* Loew, 1871 belongs to the subfamily Dolichopodinae (Diptera: Dolichopodidae). This work presents the mitochondrial genome (mitogenome) of *D. galeatus* for the first time. The mitogenome has a total length of 15,268 bp, 13 protein-coding genes, two ribosomal RNA (rRNA) genes, and 22 transfer RNA genes (tRNAs) that make up the genome. All genes have comparable positions and strands to those found in earlier reported fly mitogenomes. The nucleotide composition is tilted toward A and T, which account for 75.1% of the total. Phylogenetic approach shows that *Lichtwardtia* is the sister group of *Dolichopus* and that the Dolichopodidae are monophyletic.

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1. Introduction

Dolichopus galeatus Loew, 1871 belongs to the family Dolichopodidae. The family Dolichopodidae belongs to the order Diptera, suborder Brachycera, and superfamily Empidoidea. Six thousand eight hundred and seventy species have been identified in the world, 1026 of which are found in China. The species *D. galeatus* is distributed in Russia and China (Heilongjiang and Inner Mongolia). It is a large breed (Yang et al. 2006). Its diagnosis includes the following; hind femora with short and dense ventral hairs in middle; fore tarsomere 1 slightly thickened with short and dense bristle-like hairs ventrally and dorsally; tarsomere 4 very short; tarsomere 5 distinctly flattened; male cercus nearly knife-like, distinctly longer than wide. Dolichopodidae is a group of important natural enemy insects, some of which prey on agricultural pests, such as aphids, woodlice, bark beetles, thrips, and so on, while others prey on flies, mosquitoes, mites, and other health pests. Using Illumina sequencing technology, the entire mitochondrial genome of *D. galeatus* was examined for the first time in this study. These findings will give a molecular biological method to the identification of *D. galeatus*, as well as a new reference for future research on the population genetics and phylogeny of the Dolichopodidae family.

2. Materials and methods

2.1. Sample collection and preservation



The adult specimens of *D. galeatus* used for this study were collected from Arxan Wuli Spring (47°19'52"N, 119°93'23"E,

1035 m), Inner Mongolia, China in 2014 and deposited in the Entomological Museum (accession number: D-DGL-1) of China Agricultural University (Liang Wang, 1352659341@qq.com). Specimens were stored in 95% alcohol and a –20 °C refrigerator.

Statement: The techniques and processes for trapping small animals followed the standards and regulations set by the Chinese Academy of Agricultural Sciences' Animal Ethics Committee.

2.2. DNA extraction and sequencing

Genomic DNA was extracted from the whole body of a single adult using the TrueLib DNA Library Rapid Prep Kit (Nanjing, China). The DNA sample was sent to Genepioneer Biotechnologies, Ltd. (Nanjing, China) for library construction, and sequenced on the Illumina Novaseq 6000 platform. The final filtered reads (4.43 GB) were assembled with IDBA-UD (Peng et al. 2012), clean data with a read coverage depth over 1250X (Figure 1). A complete, circular mitochondrial sequence was contained in the assembly. The initial annotation was conducted by the MITOS web server (Bernt et al. 2013) for determining the location and direction of genes. The annotation was manually validated by multiple sequence alignment based on other published Meloidae mitogenomes in MEGA v7.0 (Kumar et al. 2016). Finally, the mitochondrial genome cycle graph of *Dolichopus galeatus* was mapped using CGView (<https://cgview.ca>).

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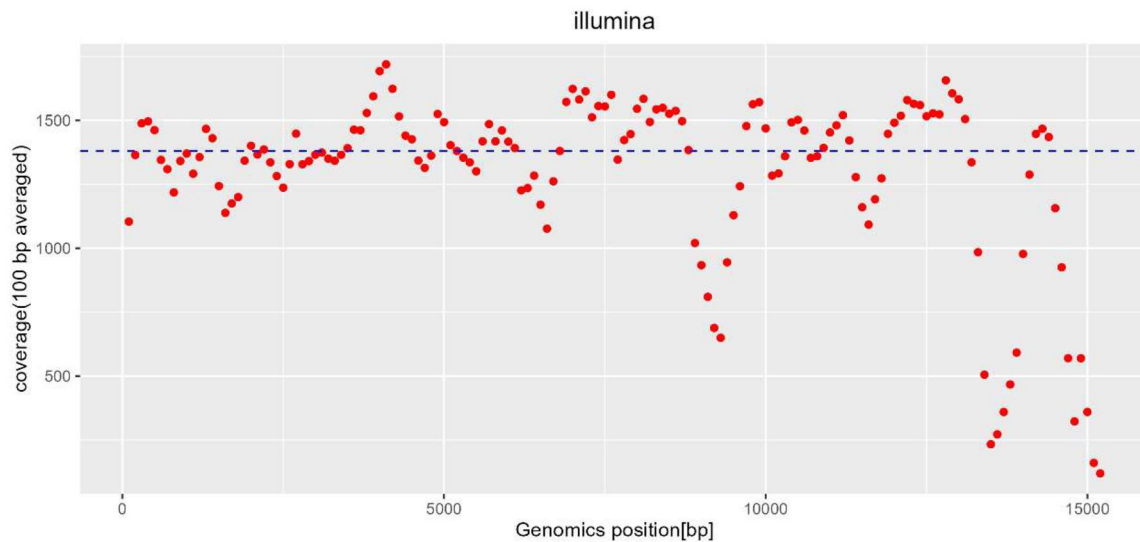


Figure 1. The read coverage depth map of *Dolichopus galeatus*.

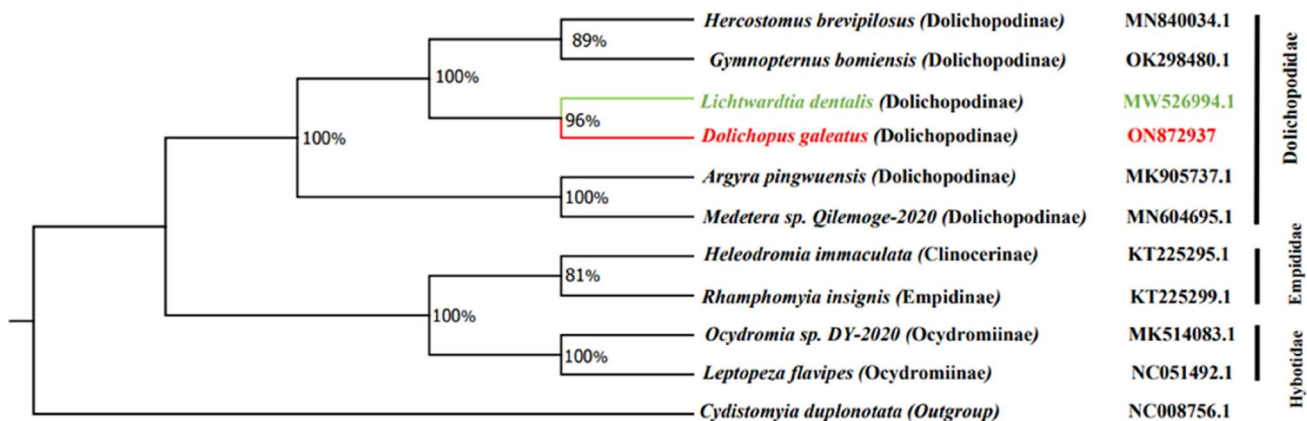


Figure 2. Maximum-likelihood analysis of 13 PCGs was used to estimate the evolutionary connections of *Dolichopus galeatus* and 10 species. GenBank accession numbers of all sequences used in the phylogenetic tree have been included in the figure and corresponding to the names of all species. Newly sequenced data in this study are in red color.

2.3. Phylogenetic analysis method

The molecular phylogenetic analysis was carried out using MEGA 7, and the phylogenetic analysis was completed using the maximum-likelihood (ML) method with 1000 bootstrap repeats. The phylogenetic tree is then enhanced using itol (<https://itol.embl.de>).

3. Results and discussion

3.1. Characteristics of *D. galeatus* mitochondrial genome

D. galeatus' complete mitogenome (GenBank accession number: ON872937) is 15,268 bp long. It encodes 13 protein-coding genes (PCGs), two ribosomal RNA (rRNA) genes, 22 transfer RNA genes (tRNAs), and a control region that has not been completely sequenced, which is similar to previous reports (Li et al. 2016, 2017; Wang et al. 2016; Zhang et al. 2016, 2022; Zhou et al. 2017; Qilemoge, Gao, et al. 2018; Qilemoge, Zhang, et al. 2018; Hou et al. 2019; Qilemoge et al. 2019; Lin and Yang 2021; Qilemoge and Yang 2022). The mitogenome's nucleotide

composition is skewed toward A and T, with 75.1% A + T content (A = 40%, T = 35.1%, C = 14.9%, G = 10%). Among the PCGs, four genes (NAD2, NAD3, NAD6, and ATP8) take the start codon of ATA, two genes (COII and ATP6) use ATG as the start codon, two genes (NAD1 and NAD4) use TTG as start codon, while COI, COIII, NAD5, NAD4L, and CYTB genes used CAG, ACC, GAG, TAC, and TTA, respectively. All the PCGs used the conventional stop codons (TAG for NAD1, NAD3, NAD4, and CYTB, TAA for the rest). The length of tRNA, which has a clover-leaf structure, ranges from 63 to 72 bp.

3.2. Phylogenetic analysis

All PCGs in phylogenetic analysis were performed based on the nucleotide sequences of 13 PCGs and two rRNAs from 11 Diptera species, of which six species are from Dolichopodinae, one species is from Clinocerinae, one species is from Empidinae, two species are from Ocydromiinae, and one species is from Cydistomyia (outgroup). The Phylogenetic tree analysis (Figure 2) shows that the monophyly of the subfamily Dolichopodinae and the sister relationship between Dolichopus



Figure 3. Picture of *Dolichopus galeatus* species (this picture was taken by the corresponding author, Ning Wang).

and *Lichtwardtia* are strongly supported. Further phylogenetic studies of the Dolichopodidae could benefit from the genomic information in the mitogenome of *D. galeatus* (Figures 3 and 4).

4. Conclusions

The first complete mitochondrial genome sequencing of *D. galeatus* was revealed in this work. The mitochondrial genome sequence will be a significant resource for future genetic investigations with *D. galeatus*, as well as a fresh reference for future research on the Dolichopodidae family.

Author contributions

YW: data analysis, conceptualization, methodology, and writing original draft preparation. GL: review and editing, visualization. JW: resources and supervision. ZS: conceptualization and writing review. NW: conceptualization and design, project administration, and funding acquisition.

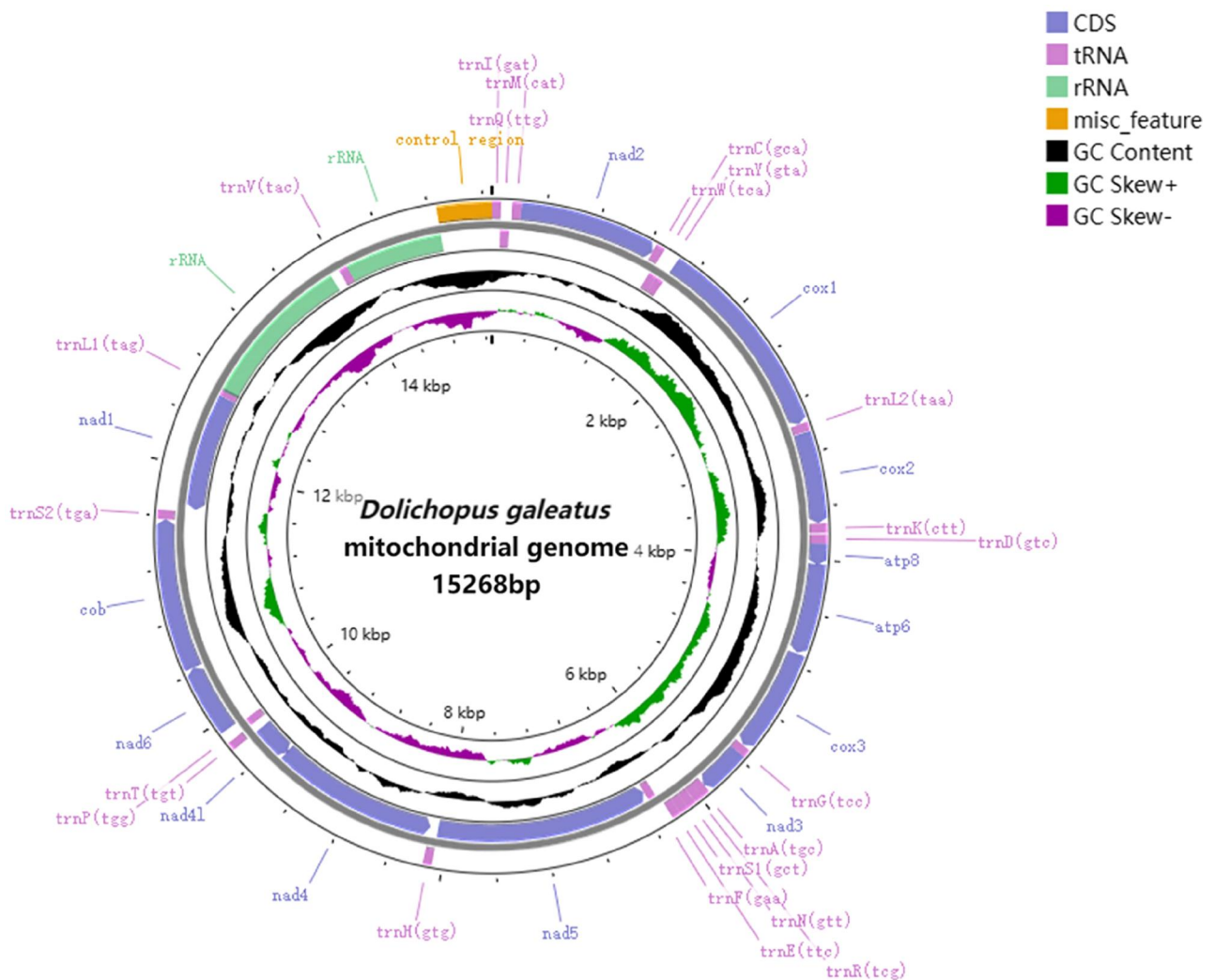


Figure 4. Genome map of *Dolichopus galeatus*.

Disclosure statement

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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Data availability statement

The genome sequence data that support the findings of this study are openly available in GenBank of NCBI (<https://www.ncbi.nlm.nih.gov/>) under the accession no. ON872937. The associated BioProject, Sequence Read Archive (SRA), and Bio-sample accession numbers are PRJNA894116, SRR22097498, and SAMN31438012, respectively.

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