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ISSN: 2380-2359 (Online) Journal homepage: www.tandfonline.com/journals/tmdn20

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To cite this article: Li Liu, Yi Wu, Wei Wei, Xiao-Yu Duan & Zeng-Qiang Qian (2019) The complete mitochondrial genome of the African malaria mosquito *Anopheles funestus* and its phylogenetic implication, Mitochondrial DNA Part B, 4:1, 1065-1067, DOI: [10.1080/23802359.2019.1586464](https://doi.org/10.1080/23802359.2019.1586464)

To link to this article: <https://doi.org/10.1080/23802359.2019.1586464>



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Published online: 04 Mar 2019.



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MITOGENOME ANNOUNCEMENT

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The complete mitochondrial genome of the African malaria mosquito *Anopheles funestus* and its phylogenetic implication

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ABSTRACT

Anopheles funestus is a major malaria vector in Africa. In this study, its complete mitochondrial genome was assembled from high-throughput sequencing reads. The circular genome is 15,403 bp long with an A + T-biased base composition and harbors 37 genes (including 13 protein-coding genes/PCGs, 22 tRNAs, and two rRNAs) and one control region. The PCGs are initiated with ATN, GTG, TTG, or CGA codons and are terminated with either TAA or TA/T codons. Phylogenetic analysis added support to the current taxonomic framework of the genus *Anopheles* and suggested that *A. funestus* is closely related to *A. culicifacies* and *A. minimus*.

ARTICLE HISTORY

Received 14 December 2018
Accepted 10 February 2019

KEYWORDS

Anopheles funestus; data mining; malaria mosquito; malaria vector; mitochondrial genome

Malaria remains a major public health issue in Africa, with *Anopheles funestus* and *Anopheles gambiae* being the predominant vectors (Djouaka et al. 2016). *Anopheles funestus* is regarded as being among the first species that have adapted to human hosts (Charlwood et al. 1995) and far outstrips *A. gambiae* in its ability to transmit *Plasmodium falciparum* in some cases (Coetzee and Fontenille 2004). This mosquito is morphologically identical at all life stages to three other members of the *funestus* subgroup (*A. arabiensis*, *A. parensis*, and *A. vaneedeni*) of the *funestus* group (series *Myzomyia* within subgenus *Cellia*) (Coetzee and Fontenille 2004). To facilitate the accurate identification of *A. funestus*, we assembled its mitochondrial genome in this study. The annotated sequence has been deposited into GenBank under the accession number MF775371.

Totally, 91.4 M 125-bp-long raw reads (SRA accession: ERR1358729) were retrieved from a previously published study (Barnes et al. 2017). These reads were originally produced from individual females collected from Chikwawa, Malawi (16°3'S, 34°50'E; NCBI BioSample: SAMEA3928332). Following the trimming with Trimmomatic v0.35 (Bolger et al. 2014), they were employed to assemble the mitochondrial genome using the ARC program (Hunter et al. 2015), with that of *A. gambiae* (NC_002084) (Beard et al. 1993) as the seed reference. The genome was annotated using the MITOS web server (Bernt et al. 2013), followed by delicate adjustment as described by Cameron (2014).

The circular genome of *A. funestus* is 15,403 bp in size and has a highly asymmetric base composition (40.3%A, 12.6%C,

9.1%G & 38.0%T; light strand). It encodes 13 protein-coding genes/PCGs, 22 tRNAs, and two rRNAs and harbors a non-coding control region. Its genomic organization is identical to those of its congeners (Peng et al. 2016). Most genes are located on the heavy strand except for four PCGs, nine tRNAs, and the two rRNAs.

All PCGs are initiated with ATN codons except for *COX1* (CGA), *ND1* (TTG), and *ND5* (GTG) and are terminated with either TAA (*ATP6*, *ATP8*, *ND1*, *ND4L*, and *ND6*) or TA/T (the remaining eight PCGs) codon. The 22 tRNAs range in size from 64 to 72 bp with a total length of 1470 bp. The two rRNAs are separated by *tRNA-Val* and are 791 bp (12S rRNA) and 1363 bp (16S rRNA) in length, respectively. The control region is 570 bp long with a remarkably high A + T content (94.0%) and is located between 12S rRNA and *tRNA-Ile*. Seven intergenic spacer regions are present across the genome, ranging in size from 1 to 34 bp with a total length of 76 bp. There are 10 intergenic overlapping regions ranging in size from 1 and 7 bp with a total length of 26 bp.

To investigate its taxonomic placement, a maximum-likelihood (ML) phylogenetic tree was reconstructed using the concatenated sequences of all 13 PCGs with MEGA7 (Kumar et al. 2016) (Figure 1). The phylogeny recovered here corroborates the validity of the series- and subgenus-level taxonomy of *Anopheles* mosquitoes and suggests that *A. funestus* is phylogenetically close to *A. culicifacies* and *A. minimus*.

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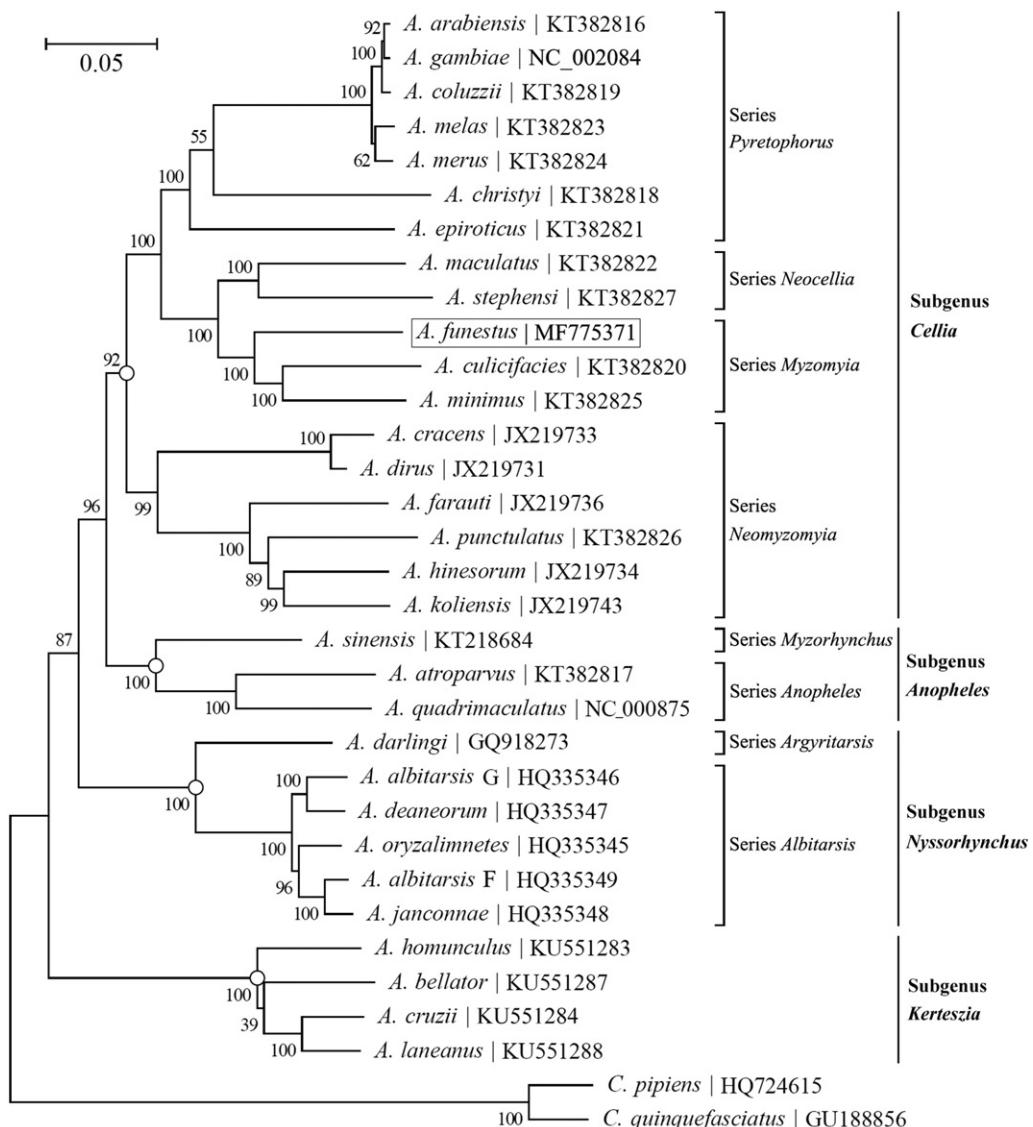


Figure 1. Phylogeny of 31 *Anopheles* mosquitoes based on the maximum likelihood (ML) analysis of the concatenated sequence of 13 PCGs. The 'GTR + G+I' substitution model was employed as suggested by MEGA7 (Kumar et al. 2016). The bootstrap values are based on 500 resamplings. The tree was rooted with *Culex pipiens* and *Culex quinquefasciatus*. Codon positions included are 1st + 2nd + 3rd.

Acknowledgements

The authors thank Dr. Kayla G. Barnes and her colleagues for generating the genomic data used in this study.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This study was financially supported by the Natural Science Basic Research Plan in Shaanxi Province of China [2018JM3001] and the Fundamental Research Funds for the Central Universities in China [GK201703038].

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