


The complete mitochondrial genome of *Priotyranus closteroides* Thomson, 1877 (Coleoptera: Cerambycidae)

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ABSTRACT

Priotyranus closteroides Thomson, 1877 (Coleoptera: Cerambycidae) is the trunk borer of orange trees. In this study, we sequenced and annotated the whole mitochondrial genome of *P. closteroides*. The results showed that the length of the complete mitochondrial genome is 15,854 bp with an overall GC content of 32.11%. The genome encodes 13 protein-coding genes (PCGs), 22 transfer RNA genes (tRNAs), and two ribosomal RNA genes (rRNAs). The relevant phylogenetic tree distinctly showed that *P. closteroides* is clustered with *Dorysthenes paradoxus* and *Dorysthenes granulatus*. This study provides a piece of valuable genomic information for the population genetics, evolution, and classification of *P. closteroides*.

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Priotyranus closteroides belongs to the insect subfamily Prioninae, within the Cerambycidae family which comprises more than 25,000 species (Sama et al. 2010). The bodies of *P. closteroides* adults are flat and wide, with russet feet and abdomen. The females are larger than males and the species prefers living in warm and humid environments. *P. closteroides* has been considered a trunk borer in forestry. In general, it attacks the orange trees roots which affects the growth and yield of the trees and causes plant death in the later stage. Therefore, it is important to study the biology and develop tools for control strategies. However, the information available about this species' mitogenome and its taxonomic status is still limited. Here, we describe the complete mitochondrial genome of *P. closteroides* and the relevant phylogenetic tree. These results will support important genetic information for future studies related to the genetic evolution of *P. closteroides*.

The *P. closteroides* samples were obtained by using the sexual trapping method to trap in Hongwei, Fujian Province, China (118°57'12"E, 26°9'15"N). The voucher specimen (code TN-202103) was photographed and deposited at the Key Laboratory of Integrated Pest Management in Ecological Forests, Fujian Agriculture and Forestry University (contact person: Songqing Wu; email: dabinyang@126.com) (Figure 1(A)). Extraction and purification of the total genomic DNA was obtained by using the TruSeq DNA Sample Preparation Kit (Vazyme, Nanjing, China). The remaining part of the DNA was deposited in Dr. Wu's lab with a unique code (DNA-PC1). A sequencing library with genomic DNA randomly broken into fragments using transposase was created and sequenced

libraries with fragment peaks at 300 bp by using Agencourt SPRIselect Beads (Beckman Coulter, Indianapolis, IN) were selected by Shanghai Tianhao Biotechnology Company (Shanghai, China). To sequence the library preparations, we used the Illumina Hiseq 2500 (Illumina, San Diego, CA) with 2 × 150 bp paired-end reads (Yin et al. 2016). A total of 40,028,884 clean reads were assembled by using MitoZ and metaSPAdes (Nurk et al. 2017). Furthermore, the Mitos web-server was used to annotate the assembled sequences (Evans and Ślipiński 2016). To significantly improve the annotation obtained by Mitos was revised by using the closely related genome of *Megopis sinica* (Cerambycidae, GenBank accession number: MN594765) (Su and Wang 2020).

The results showed that the *P. closteroides* complete mitochondrial genome length is 15,854 bp, with 14,583 bp encoding open reading frames, with 13 protein-coding genes (PCGs) in Coleoptera typical order, 22 transfer RNA genes (tRNAs), two ribosomal RNA genes (rRNAs) (*rrn5*, *rrnL*) (Boore et al. 1998; Nie and Yang 2014). The mitochondrial genome map was visualized by Geneious Prime 2022.1.1 (Figure 1(B)). Among these 13 PCGs, three PCGs (*atp6*, *nad3*, *nad4*) start with ATA, four PCGs (*nad2*, *atp8*, *nad5*, *nad1*) start with ATT, two PCGs (*cox2*, *nad6*) start with ATC and three PCGs (*cox3*, *nad4l*, *cob*) start with ATG. However, the *cox1* gene must use a non-conventional start site because no regular start codon is available after the last stop codon upstream to the *cox1* open reading frame. The same phenomenon also has been found in other insects (De Oliveira et al. 2005; Castro et al. 2006; Lee et al. 2006; Sheffield et al. 2008). Also, 10 PCGs (*nad2*, *cox1*, *cox2*, *atp6*, *cox3*, *nad5*, *nad4*, *nad4l*, *nad6*, and

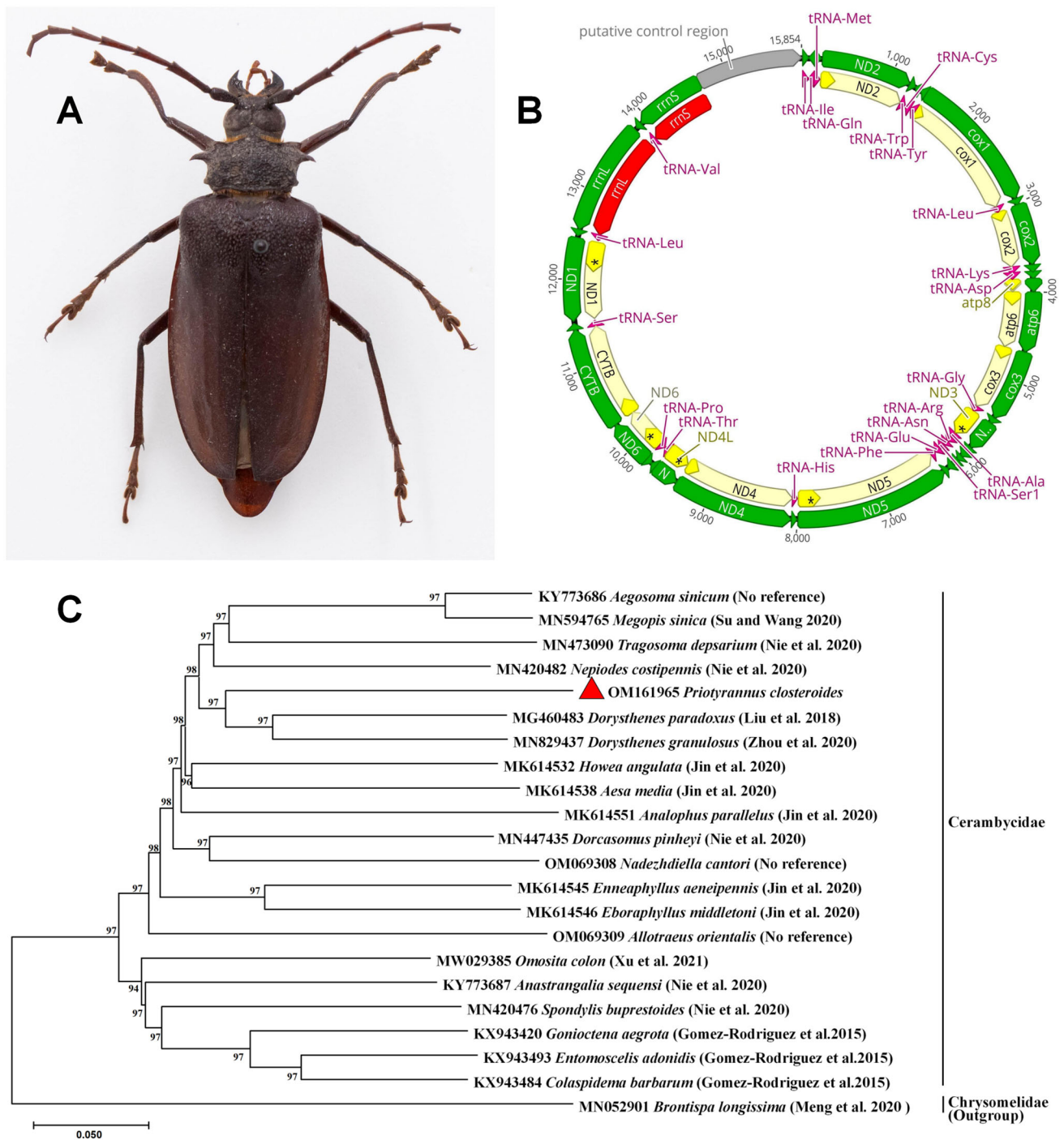


Figure 1. The phylogenetic tree and the picture of *Priotyrannus closteroides* Thomson, 1877. (A) Female of *Priotyrannus closteroides* Thomson, 1877, dorsal view. (B) Gene map of mitochondrial genome of *Priotyrannus closteroides*. (C) Maximum-likelihood tree of *Priotyrannus closteroides* Thomson and related 21 different insect species based on the complete mitochondrial genome. The numbers at each node represent bootstrap percentages of 1000 pseudoreplicates by ML analysis. The scale bar indicates the number of substitutions per site.

cob) stop with TAA, three PCGs (*atp8*, *nad3*, and *nad1*) stop with TAG. The overall GC content is 32.11%. The annotated mitogenome sequence has been deposited in NCBI GenBank under accession number OM161965.

The mitochondrial sequences (including the nucleotide sequences of 13 PCGs) of *P. closteroides* and other mitochondrial genome of 20 Cerambycidae species selected from NCBI-BLAST (<http://blast.ncbi.nlm.nih.gov>) were used to construct phylogenetic tree with *Brontispa longissima* (Chrysomelidae; GenBank accession number: MN052901)

(Meng et al. 2020) as an outgroup (Zhang et al. 2018). The 13 PCGs sequences were aligned using ClustalW (Thompson et al. 2002) in MEGA X (Kumar et al. 2018). Poorly aligned regions were excluded in Gblocks v.0.91b (Castresana 2000). PartitionFinder v2.1.1 (Lanfear et al. 2017) was run to select best-fit partitioning schemes and evolutionary models (GTR + G+I). We used RAxML v8.2.10 (Stamatakis 2014) based on the rapid bootstrap (BS) algorithm with 1000 BS replicates to construct maximum-likelihood (ML) tree. The phylogenetic analysis suggests that the mitochondrial genome of

P. closteroides constitutes a monophyletic group with the mitochondrial genome of other 20 Cerambycidae species, and is in a close relationship with the mitochondrial genome of *Dorysthenes paradoxus* (Liu et al. 2018) and *Dorysthenes granulosis* (Zhou et al. 2020; Figure 1(C)).

Ethics statement

This research does not involve ethical research. The collection of the specimens was carried out following the guidelines provided by the authors' institution (Key Laboratory of Integrated Pest Management in Ecological Forests, Fujian Agriculture and Forestry University) and nation.

Author contributions

Xuanxuan Wu, Yiqi Lin, and Yunzhu Sun did the sample collection and laboratory work. Yiqi Lin photographed the species. Xuanxuan Wu, Songqing Wu, and Mingyong Luo were involved in the conception and design. Xuanxuan Wu, Yiqi Lin, Bowei Zhou, and Yunzhu Sun analyzed and interpreted the data. Xuanxuan Wu drafted the paper. Xuanxuan Wu revised the paper critically for intellectual content. All authors are involved: the final approval of the version to be published. All authors agreed to be accountable for all aspects of the work.

Disclosure statement

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

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Data availability statement

The genome sequence data supporting this study's findings are openly available in GenBank of NCBI at <https://www.ncbi.nlm.nih.gov> under assessment no. OM161965. The associated BioProject, Bio-Sample, and SRA numbers are PRJNA796508, SAMN24891047, and SRR17578369, respectively.

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