

## Sequence analysis of mitochondrial genome of the false and phantom crane-fly *Ptychoptera qinggouensis* Kang, Yao and Yang, 2013 (Diptera, Ptychopteridae)

Shuo Ma and Zehui Kang

Key Lab of Integrated Crop Pest Management of Shandong Province, College of Plant Health and Medicine, Qingdao Agricultural University, Qingdao, China

### ABSTRACT

The genus *Ptychoptera* Meigen, 1803 is the largest genus of the family Ptychopteridae with 78 known species. In this study, we report a nearly complete mitochondrial (mt) genome of this genus, which is a circular molecule of more than 15,028 bp. The mt genome contains 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes and a non-coding region. The overall base composition is A (38.1%), T (36.7%), C (14.9%), and G (10.4%), with an AT content of 74.8%. The AT content of N-strand genes (75.7%) is higher than that of the J-strand genes (71.7%). Phylogenetic analysis reveals that the monophyly of Ptychopteridae, Bibiomorpha, Tipulomorpha and Brachycera are strongly supported, and the sister group relationship between Tanyderidae and Ptychopteridae is not supported.

### ARTICLE HISTORY

Received 16 June 2020  
Accepted 20 June 2020

### KEYWORDS

Mitochondrial genome;  
phylogeny; *Ptychoptera*;  
false and phantom crane-fly

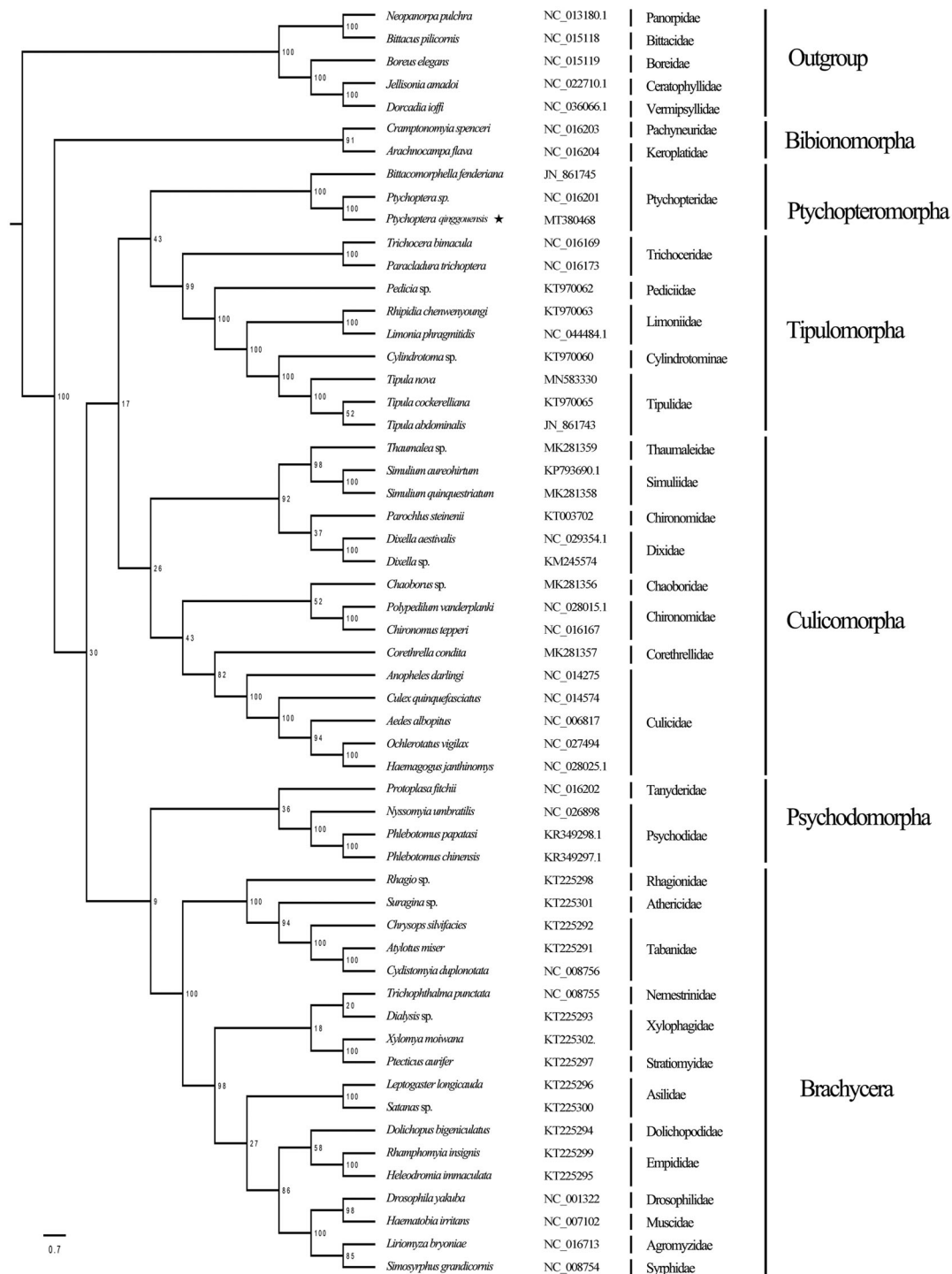
The family Ptychopteridae includes three genera, *Bittacomorpha* Westwood, 1835, *Bittacomorphella* Alexander, 1916, and *Ptychoptera* Meigen, 1803, of which *Ptychoptera* is the largest genus and widely distributed in the world except for the Australasian (Oceanian) region with 78 described species (Kang et al. 2013a, 2013b; Paramonov 2013; Török et al. 2015). More mitochondrial (mt) genomes have been widely used for reconstructing phylogenetic relationships in many insect groups, including the lower Diptera (Beckenbach 2012; Timmermans and Vogler 2012; Wang et al. 2012; Caravas and Friedrich 2013; Li et al. 2013; Li et al. 2015; Wang et al. 2017). There are nearly 700 complete or nearly complete lower dipteran mt genomes available in GenBank, of which only two mt genomes are from Ptychopteridae. Here, we report another nearly complete mt genome of Ptychopteridae.

The specimen of *Ptychoptera qinggouensis* Kang, Yao and Yang, 2013 used in this study was collected from Daqinggou, Kezuohouqi, Neimenggu, China (42°45'47" N 122°12'14" E, 200 m) and stored in the Entomological Museum of Qingdao Agricultural University, China (No. PTY0001). Qualified DNA samples were pooled for next-generation sequencing library construction following the method proposed by Gillett et al. (2014). The standard PCR reactions were sequenced by primers designed by Simon et al. (2006). BLAST searches were conducted with BioEdit 7.0.5.3 for the bait sequence against mt genome assemblies. The sequence was annotated

following the method proposed by Cameron (2014). Maximum-likelihood analysis was conducted by RAxML v7.0.3 (Stamatakis 2006).

The nearly complete mt genome of *P. qinggouensis* (GenBank accession no. MT380468) is 15,028 bp in length. It contains 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes and a non-coding region. The overall base composition is A (38.1%), T (36.7%), C (14.9%), and G (10.4%), with an AT content of 74.8%. It has a weakly positive AT-skew and a negative GC-skew. For protein-coding genes, the AT content of all PCGs is 81.1%. It has a negative AT-skew and a positive GC-skew. The AT content of N-strand genes (75.7%) is higher than that of the J-strand genes (71.7%). The AT content of PCG third codon positions is much higher than that of the first and second codon positions and the AT content of PCG third codon positions of N-strand genes (87.1%) is the highest. PCGs of J-strand genes have negative AT-skew and GC-skew. PCGs of N-strand genes have negative AT-skew and positive GC-skew. For RNA genes, The AT content of tRNA was 76.9% and the AT-skew and GC-skew are positive. The AT content of the 16S rRNA (80.7%) is slightly higher than that of the 12S rRNA (77%). They both have negative AT-skew and a positive GC-skew.

The phylogenetic tree in this study (Figure 1) shows a strong support for the monophyly of the Bibiomorpha (100%), Tipulomorpha (99%) and the Brachycera (100%).



**Figure 1.** Phylogenetic tree of Nematocera based on whole mitochondrial genomes using maximum-likelihood analysis. Numbers above the branches are bootstrap percentages. GeneBank accession numbers of each species are listed in the tree.

The phylogenetic relationship in Tipulomorpha is Trichoceridae + (Pediciidae + (Limoniidae + (Cylindrotomidae + Tipulidae))). The monophyly of the family Ptychopteridae is strongly supported, whereas its phylogenetic position in the lower diptera is undefined.

### Acknowledgements

We express our sincere thanks to Ding Yang (Beijing) and Fan Song (Beijing) for great help during the study. We are also very grateful to Xin Li (Beijing) for collecting the specimen.

### Disclosure statement

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

### Funding

This work was funded by the National Natural Science Foundation of China [41901061], the Shandong Provincial Natural Science Foundation, China [ZR2019BC034] and the High-level Talents Funds of Qingdao Agricultural University, China [663-1119008].

## Data availability statement

The data that support the findings of this study are openly available in NCBI at <https://www.ncbi.nlm.nih.gov/>. GeneBank accession numbers are listed as follows: *Aedes albopitus* (NC\_006817), *Anopheles darlingi* (NC\_014275), *Arachnocampa flava* (NC\_016204), *Atylotus miser* (KT225291), *Bittacomorphella fenderiana* (JN\_861745), *Bittacus pilicornis* (NC\_015118), *Boreus elegans* (NC\_015119), *Chaoborus* sp. (MK281356), *Chironomus tepperi* (NC\_016167), *Chrysops silvifacies* (KT225292), *Corethrella condita* (MK281357), *Cramptonomyia spenceri* (NC\_016203), *Culex quinquefasciatus* (NC\_014574), *Cydistomyia duplonotata* (NC\_008756), *Cylindrotoma* sp. (KT970060), *Dialysis* sp. (KT225293), *Dixella aestivalis* (NC\_029354.1), *Dixella* sp. (KM245574), *Dolichopus bigeniculatus* (KT225294), *Dorcadia ioffi* (NC\_036066.1), *Drosophila yakuba* (NC\_001322), *Haemagogus janthinomys* (NC\_028025.1), *Haematobia irritans* (NC\_007102), *Heleodromia immaculata* (KT225295), *Jellisonia amadoi* (NC\_022710.1), *Leptogaster longicauda* (KT225296), *Limonia phragmitidis* (NC\_044484.1), *Liriomyza bryoniae* (NC\_016713), *Neopanorpa pulchra* (NC\_013180.1), *Nyssomyia umbratilis* (NC\_026898), *Ochlerotatus vigilax* (NC\_027494), *Paracladura trichoptera* (NC\_016173), *Parochlus steinenii* (KT003702), *Pedicia* sp. (KT970062), *Phlebotomus chinensis* (KR349297.1), *Phlebotomus papatasi* (KR349298.1), *Polypedilum vanderplanki* (NC\_028015.1), *Protoplasma fitchii* (NC\_016202), *Ptecticus aurifer* (KT225297), *Ptychoptera qinggouensis* (MT380468), *Ptychoptera* sp. (NC\_016201), *Rhagio* sp. (KT225298), *Rhamphomyia insig-nis* (KT225299), *Rhipidia chenwenyungii* (KT970063), *Satanas* sp. (KT225300), *Simosyrphus grandicornis* (NC\_008754), *Simulium aureohirtum* (KP793690.1), *Simulium quinquestriatum* (MK281358), *Suragina* sp. (KT225301), *Thaumalea* sp. (MK281359), *Tipula abdominalis* (JN\_861743), *Tipula cockerelliana* (KT970065), *Tipula nova* (MN583330), *Trichocera bimacula* (NC\_016169), *Trichophthalma punctata* (NC\_008755) and *Xylomya moiwana* (KT225302).

## References

- Alexander CP. 1916. New or little-known crane-flies from the United States and Canada: Tipulidae, Ptychopteridae, Diptera. Part 3. Proc Acad Nat Sci Philadelphia. 68(3):486–549.
- Beckenbach AT. 2012. Mitochondrial Genome Sequences of Nematocera (Lower Diptera): Evidence of Rearrangement following a Complete Genome Duplication in a Winter Crane Fly. *Genome Biol Evol.* 4(2): 89–101.
- Cameron SL. 2014. How to sequence and annotate insect mitochondrial genomes for systematic and comparative genomics research. *Syst Entomol.* 39(3):400–411.
- Caravas J, Friedrich M. 2013. Shaking the Diptera tree of life: performance analysis of nuclear and mitochondrial sequence data partitions. *Syst Entomol.* 38(1):93–103.
- Gillett C, Alex CP, Timmermans M, Jordal BH, Emerson BC, Vogler AP. 2014. Bulk de novo mitogenome assembly from pooled total DNA elucidates the phylogeny of weevils (Coleoptera: Curculionoidea). *Mol Biol Evol.* 31(8):2223–2237.
- Kang Z, Yao G, Yang D. 2013a. New species and record of *Ptychoptera* Meigen, 1803 (Diptera: Ptychopteridae) from China. *Zootaxa.* 3682(4): 455–472.
- Kang ZH, Yao G, Yang D. 2013b. Five new species of *Ptychoptera* Meigen with a key to species from China (Diptera: Ptychopteridae). *Zootaxa.* 3682(4):541–555.
- Li H, Shao R, Song N, Song F, Jiang P, Li Z, Cai W. 2015. Higher-level phylogeny of paraneopteran insects inferred from mitochondrial genome sequences. *Sci Rep.* 5:8527.
- Li H, Shao R, Song F, Zhou X, Yang Q, Li Z, Cai W. 2013. Mitochondrial genomes of two Barklice, *Psococerastis albimaculata* and *Longivalvus hyalospilus* (Psocoptera: Psocomorpha): contrasting rates in mitochondrial gene rearrangement between major lineages of Psocodea. *PLOS One.* 8(4):e61685.
- Meigen JW. 1803. Versuch einer neuen Gattungs-Eintheilung der europäischen zweiflügligen Insekten. *Magazin Für Insektenkunde* (Illiger). 2: 259–281.
- Paramonov NM. 2013. Ptychopteridae, a family of flies (Diptera) new to the philippine islands with the description of a new species. *Zootaxa.* 3682 (4):584–588.
- Simon C, Buckley TR, Frati F, Stewart JB, Beckenbach AT. 2006. Incorporating molecular evolution into phylogenetic analysis, and a new compilation of conserved polymerase chain reaction primers for animal mitochondrial DNA. *Annu Rev Ecol Syst.* 37(1):545–579.
- Stamatakis A. 2006. RAxML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics.* 22(21):2688–2690.
- Timmermans MJ, Vogler AP. 2012. Phylogenetically informative rearrangements in mitochondrial genomes of Coleoptera, and monophyly of aquatic elateriform beetles (Dryopoidea). *Mol Phylogenet Evol.* 63(2):299–304.
- Török E, Kolcsár LP, Dénes AL, Keresztes L. 2015. Morphologies tells more than molecules in the case of the European widespread *Ptychoptera albimana* (Fabricius, 1787) (Diptera, Ptychopteridae). *North-western. J Zool.* 11 (2):304–315.
- Wang Y, Liu X, Garzón-Orduña IJ, Winterton SL, Yan Y, Aspöck U, Aspöck H, Yang D. 2017. Mitochondrial phylogenomics illuminates the evolutionary history of Neuropterida. *Cladistics.* 33(6):617–636.
- Wang Y, Liu X, Winterton SL, Yang D. 2012. The first mitochondrial genome for the fishfly subfamily Chauliodinae and implications for the higher phylogeny of megaloptera. *PLOS One.* 7(10):e47302.
- Westwood JO. 1835. *Insectorum novorum exoticorum (ex ordine Dipterorum) descriptiones.* London Edinburgh Philos Mag J Sci. 6(34): 280–281.