

## The complete mitochondrial genome sequence of *Diaphorina citri* (Hemiptera: Psyllidae)

Fengnian Wu<sup>a,b</sup>, Yijing Cen<sup>a</sup>, Xiaoling Deng<sup>a</sup>, Zheng Zheng<sup>a,b</sup>, Jianchi Chen<sup>b</sup> and Guangwen Liang<sup>a</sup>

<sup>a</sup>Laboratory of Insect Ecology/Guangdong Province Key Laboratory of Microbial Signals and Disease Control, South China Agricultural University, Guangzhou, Guangdong, China; <sup>b</sup>USDA-ARS, San Joaquin Valley Agricultural Sciences Center, Parlier, CA, USA

### ABSTRACT

The first complete mitochondrial genome (mitogenome) sequence of Asian citrus psyllid, *Diaphorina citri* (Hemiptera: Psyllidae) from Guangzhou, China, is presented. The circular mitogenome is 14,996 bp in length with an A+T content of 74.5%, and contains 13 protein-coding genes (PCGs), 22 tRNA genes (tRNAs), two rRNA genes and an AT-rich control region. Synteny was identical to the ancestral insect mitogenomes. All PCGs start with 'ATN' codons. Eleven PCGs use the complete termination codons ('TAA' or 'TAG'), while the remaining two (*cox2* and *nad5*) use a single 'T' as the stop codon. All tRNAs have the typical cloverleaf structure, except for *trnTrp* which lacks the variable arm and *trnSer* (AGN) which lacks the dihydrouridine arm. A phylogenetic tree, including 12 members of Sternorrhyncha, was constructed based on the 13 PCGs sequences. The resulting phylogenetic tree matched the established taxonomic scheme based on morphology.

### ARTICLE HISTORY

Received 11 February 2016  
Revised 16 February 2016  
Accepted 17 February 2016

### KEYWORDS

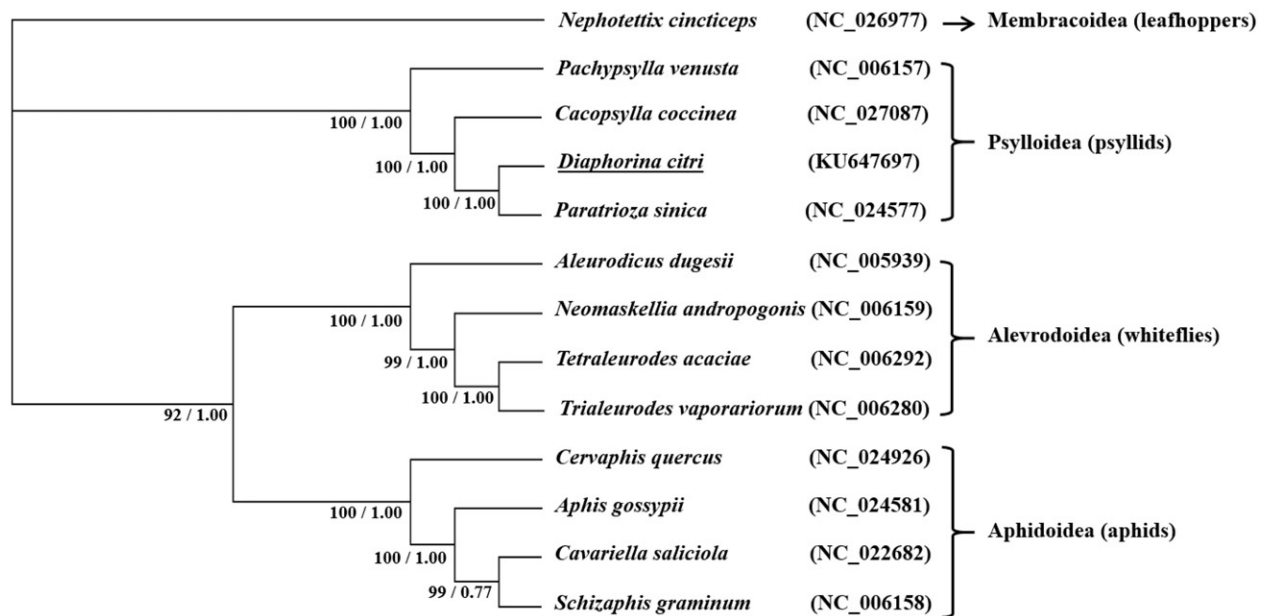
*Diaphorina citri*;  
mitogenome; phylogenetic  
analysis; psyllidae;  
sternorrhyncha

Psylloidea has more than 3000 described species around the world (Hollis 2004; Forero 2008), many of which are economically important pests. Among them, Asian citrus psyllid, *Diaphorina citri* (Hemiptera: Psyllidae) is an important pest because it is the vector of 'Candidatus Liberibacter asiaticus', associated with citrus Huanglongbing (HLB), a highly destructive disease in citrus production worldwide (Bové 2006). Up to date, only three Psylloidea mitochondrial genomes (mitogenomes) are completely sequenced: *Pachypsylla venusta* (Thao et al. 2004), *Cacopsylla coccinea* (Que et al. 2015) and *Paratrioza sinica* (Zhang et al. 2016).

*Diaphorina citri* adults were collected and specimens were maintained in Citrus Huanglongbing Research Laboratory, South China Agricultural University in Guangzhou, China, with an accession number 150317GZ. Specimen psyllids are available for research purpose at request. For mitogenome sequencing, DNA of an individual *D. citri*, was extracted, sequenced using Illumina MiSeq format (Illumina, San Diego, CA) and assembled *de novo* as described previously (Wu et al. 2015a,b). A single contig of 14,910 bp was identified by standalone BLASTn (version 2.2.30) (Camacho et al. 2009) referenced to the complete mitogenome of three Psylloidea species, NC\_024577 (*P. sinica*), NC\_027087 (*C. coccinea*) and NC\_006157 (*P. venusta*). PCR primer set (MT-F: 5'-TAACAGGGTATCTAATCCTG-3'/MT-R: 5'-ATTATGAGTCCAATAGCTT-3') was designed from both terminal ends, amplified a fragment of 1418 bp determined by Sanger's sequencing method and assembled into a circular genome. The mitogenome of *D. citri* is 14,996 bp in length with coverage of 4001 × (GenBank accession no. KU647697).

Protein-coding genes (PCGs), ribosomal RNAs (rRNAs) and transfer RNA genes (tRNAs) were predicted by MITOs and tRNAscan-SE 1.21 (Lowe & Eddy 1997; Bernt et al. 2013). The *D. citri* mitogenome contains 13 PCGs, 22 tRNAs, two rRNAs and a non-coding region of 902 bp. Twenty-three genes were on the majority strand (J-strand), and the other 14 genes on the minority strand (N-strand). The overall gene order of the *D. citri* mitogenome is identical to three other published Psylloidea mitogenomes (Thao et al. 2004; Que et al. 2015; Zhang et al. 2016). All PCGs start with 'ATN' codons, and 11 PCGs stop with the complete termination codons 'TAA' or 'TAG', while the remaining two (*cox2* and *nad5*) have incomplete termination codons 'T'. All the 22 tRNAs, ranging from 56 to 70 bp, have a typical cloverleaf structure, except for *trnTrp* which lacks the variable arm and *trnSer* (AGN) which lacks the dihydrouridine (DHU) arm. The loss of the DHU arm in *trnSer* is a typical feature of metazoan mitogenomes (Wolstenholme 1992).

PHYML 3.0 (Guindon & Gascuel 2003) for the maximum likelihood (ML) method and MrBayes 3.2.5 (Ronquist & Huelsenbeck 2003) for the Bayesian inference (BI) method were utilized to construct phylogenetic trees. Thirteen PCGs in the mitogenome of *D. citri* with those of the Sternorrhyncha members were used following the method of Jeyaprakash and Hoy (2009) with *Nephotettix cincticeps* (Hemiptera: Cicadellidae) as an outgroup (Figure 1). Including *D. citri*, all members in Psylloidea formed a monophyletic clade matching the existing taxonomy scheme that was derived on morphology.



**Figure 1.** Phylogenetic relationship of representative members of Sternorrhyncha based on 13 protein coding gene sequence in their mitogenomes. Numbers at the nodes are bootstrap values of maximum likelihood method/posterior probabilities of Bayesian inference method. The mitogenome of *Nephrotettix cincticeps* was used as outgroup. All sequence accession numbers are in parentheses and downloaded from GenBank sequence database.

## Disclosure statement

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper. Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture. USDA is an equal opportunity provider and employer.

## Funding information

This research was supported by Chinese Modern Agricultural Technology Systems (CARS-27) and California Citrus Research Board.

## References

- Bernt M, Donath A, Jühling F, Externbrink F, Florentz C, Fritzsche G, Pütz J, Middendorf M, Stadler PF. 2013. MITOS: improved *de novo* metazoan mitochondrial genome annotation. *Mol Biol Evol.* 69:313–319.
- Bové JM. 2006. Huanglongbing: a destructive, newly-emerging, century-old disease of citrus. *J Plant Pathol.* 88:7–37.
- Camacho C, Coulouris G, Avagyan V, Ma N, Papadopoulos J, Bealer K, Madden TL. 2009. BLAST+: architecture and applications. *BMC Bioinform.* 10:421.
- Forero D. 2008. The systematics of the Hemiptera. *Rev Colomb Entomol.* 34:1–21.
- Guindon S, Gascuel O. 2003. A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Syst Biol.* 52:696–704.
- Hollis D. 2004. Australian Psylloidea: jumping plantlice and lerp insects. Canberra, Australia: Australian Biological Resources Study (ABRS). p. 232.
- Jeyaprakash A, Hoy MA. 2009. First divergence time estimate of spiders, scorpions, mites and ticks (subphylum: Chelicerata) inferred from mitochondrial phylogeny. *Exp Appl Acarol.* 47:1–18.
- Lowe TM, Eddy SR. 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. *Nucleic Acids Res.* 25:955–964.
- Que S, Yu L, Xin T, Zou Z, Hu L, Xia B. 2015. Complete mitochondrial genome of *Cacopsylla coccinea* (Hemiptera: Psyllidae). *Mitochondrial DNA.* [Epub ahead of print]. DOI: 10.3109/19401736.2015.1007319.
- Ronquist F, Huelsenbeck JP. 2003. MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics.* 19:1572–1574.
- Thao ML, Baumann L, Baumann P. 2004. Organization of the mitochondrial genomes of whiteflies, aphids, and psyllids (Hemiptera, Sternorrhyncha). *BMC Evol Biol.* 4:25.
- Wolstenholme DR. 1992. Animal mitochondrial DNA: structure and evolution. *Int Rev Cytol.* 141:173–216.
- Wu F, Deng X, Liang G, Huang J, Cen Y, Chen J. 2015a. Whole-genome sequence of “*Candidatus Proffttella armatura*” from *Diaphorina citri* in Guangdong, China. *Genome Announc.* 3:e01282–15.
- Wu F, Zheng Z, Deng X, Cen Y, Liang G, Chen J. 2015b. Draft genome sequence of “*Candidatus Liberibacter asiaticus*” from *Diaphorina citri* in Guangdong, China. *Genome Announc.* 3:e01316–15.
- Zhang QL, Guo ZL, Yuan ML. 2016. The complete mitochondrial genome of *Paratrioza sinica* (Insecta: Hemiptera: Psyllidae). *Mitochondrial DNA.* 27:734–735.