



Complete Genome Sequence of a *Blochmannia* Endosymbiont of *Colobopsis nipponica*

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ABSTRACT *Blochmannia* endosymbionts (*Gammaproteobacteria*) live in bacteriocytes, which are specialized cells found in the genus *Camponotus* and its neighbor genera. In this announcement, we describe the complete genome sequence of the *Blochmannia* endosymbiont of *Colobopsis nipponica*, which originated from a colony collected in the Republic of Korea.

Endosymbionts are very commonly identified among at least 15% to 20% of insect species (1), including ants (2) and planthoppers (3). The genus *Camponotus* and its neighbor genera have developed a strong relationship with the *Gammaproteobacteria* genus *Blochmannia*, especially in the early life stage (4). Their roles have been predicted to enrich the nourishment and immunity of the host ants in the bacteriocytes of the ants' midgut (2, 4, 5). Six *Blochmannia* complete genomes are available (6–9), which can be used to understand species-specific adaptations at the genomic level.

Although large numbers of ant mitochondrial and whole genomes are available because of the rapid development of next-generation sequencing technologies (10–29), one *Colobopsis* complete mitochondrial genome (30) and one endosymbiont bacterial genome (8) are available. More genomes of endosymbionts in this genus can provide their detailed characteristics.

A colony of *Colobopsis nipponica* was collected in a coastal forest of Geoje Island, Republic of Korea (34°51'59.6"N, 128°44'22.0"E) (number KFDS00186; InfoBoss Cyber Herbarium). Total DNA was extracted from multiple *C. nipponica* workers by using DNeasy blood and tissue kits (Qiagen, Hilden, Germany). The sequencing library was constructed using the TruSeq Nano DNA library preparation kit (Illumina, San Diego, CA) following the manufacturer's recommendations, with ~350-bp DNA fragments. Genome sequencing was performed using a HiSeq X system at Macrogen, Inc. (Republic of Korea), with extracted DNA from *Colobopsis nipponica*, yielding 38.41 million 151-bp reads. Bioinformatic analyses were performed with default parameters except where otherwise noted. *De novo* assembly was conducted by Velvet v1.2.10 (31) with a minimum depth of 30×, after raw read filtering with Trimmomatic v0.33 (32). Contig sequences were selected based on BLASTN results against the nonredundant data set; matched sequences were from endosymbiont bacteria with >95% query coverage and >80% identity. GapCloser v1.12 (33), BWA v0.7.17 (34), SAMtools v1.9 (35), and Geneious R11 v11.1.5 (Biomatters Ltd., Auckland, New Zealand) were used for filling gaps and conforming assembled sequences. Assembled genome sequences were confirmed as a circular genome via overlapping 2-kb sequences on both ends. After completion of the genome, all bases were confirmed using BWA v0.7.17 (34) and SAMtools v1.9 (35). Genome annotation was conducted by the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (36).

The complete genome of the *Blochmannia* endosymbiont of *Colobopsis nipponica* was 728,116 bp long, its coverage was 232.44×, and its GC content was 28.7%. It is the

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second shortest genome, after that of “*Candidatus Blochmannia vafer*” (GenBank accession number [CP002189](#)), which was isolated from *Camponotus vafer* (8). The genome of the *Blochmannia* endosymbiont of *Colobopsis obliquus* (GenBank accession number [CP010049](#)) (773,940 bp) (8) presents no inversion against our genome. A total of 609 protein-coding genes (PCGs), 40 tRNAs, 3 rRNAs, 2 noncoding RNAs, and 1 transfer-messenger RNA were predicted. The number of PCGs is the third highest among all available *Blochmannia* complete genomes, after those of the *Blochmannia* endosymbionts of *Camponotus nipponensis* (9) and *Camponotus pennsylvanicus* (6). The number of tRNAs is the same as those of the aforementioned two genomes, the highest among seven genomes.

We expect that our genome sequence will be used for comparative genomic analyses of ant endosymbionts to clarify their genomic characteristics, as well as their coevolutionary histories with host species.

Data availability. The whole-genome sequencing project for the *Blochmannia* endosymbiont of *Colobopsis nipponica* was deposited in DDBJ/ENA/GenBank under accession number [CP046533](#), BioProject number [PRJNA592762](#), and BioSample number [SAMN13439370](#). Raw sequences were deposited under SRA accession number [SRR12768939](#).

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REFERENCES

- Buchner P. 1965. Endosymbiosis of animals with plant microorganisms. Interscience Publishers, New York, NY.
- de Souza DJ, Bézier A, Depoix D, Drezen J-M, Lenoir A. 2009. *Blochmannia* endosymbionts improve colony growth and immune defence in the ant *Camponotus fellah*. *BMC Microbiol* 9:29. <https://doi.org/10.1186/1471-2180-9-29>.
- Park J, Xi H, Park J, Lee W. 2020. The complete mitochondrial genome of fungal endosymbiont, Ophiocordycepsitaceae sp., isolated from *Ricania speculum* (Hemiptera: Ricanidae). *Mitochondrial DNA B Resour* 5:1888–1889. <https://doi.org/10.1080/23802359.2020.1754141>.
- Sauer C, Dudaczek D, Hölldobler B, Gross R. 2002. Tissue localization of the endosymbiotic bacterium “*Candidatus Blochmannia floridanus*” in adults and larvae of the carpenter ant *Camponotus floridanus*. *Appl Environ Microbiol* 68:4187–4193. <https://doi.org/10.1128/aem.68.9.4187-4193.2002>.
- Degnan PH, Lazarus AB, Brock CD, Wernegreen JJ. 2004. Host-symbiont stability and fast evolutionary rates in an ant-bacterium association: cospeciation of *Camponotus* species and their endosymbionts, *Candidatus Blochmannia*. *Syst Biol* 53:95–110. <https://doi.org/10.1080/10635150490264842>.
- Degnan PH, Lazarus AB, Wernegreen JJ. 2005. Genome sequence of *Blochmannia pennsylvanicus* indicates parallel evolutionary trends among bacterial mutualists of insects. *Genome Res* 15:1023–1033. <https://doi.org/10.1101/gr.3771305>.
- Williams LE, Wernegreen JJ. 2010. Unprecedented loss of ammonia assimilation capability in a urease-encoding bacterial mutualist. *BMC Genomics* 11:687. <https://doi.org/10.1186/1471-2164-11-687>.
- Williams LE, Wernegreen JJ. 2015. Genome evolution in an ancient bacteria-ant symbiosis: parallel gene loss among *Blochmannia* spanning the origin of the ant tribe Camponotini. *PeerJ* 3:e881. <https://doi.org/10.7717/peerj.881>.
- Park J, Xi H, Park J, Nam SJ, Lee Y-D. 2020. Complete genome sequence of the *Blochmannia* endosymbiont of *Camponotus nipponensis*. *Microbiol Resour Announc* 9:e00703-20. <https://doi.org/10.1128/MRA.00703-20>.
- Park J, Park J, Xi H. 2019. Ant genome database: current status of ant genomes and their characteristics. *Proc 2019 Fall Int Conf Korean Soc Appl Entomol*. <https://doi.org/10.13140/RG.2.2.25518.13128>.
- Babbucci M, Bassi A, Scupola A, Patarnello T, Negrisolo E. 2014. Is it an ant or a butterfly? Convergent evolution in the mitochondrial gene order of Hymenoptera and Lepidoptera. *Genome Biol Evol* 6:3326–3343. <https://doi.org/10.1093/gbe/evu265>.
- Park J, Xi H, Oh S-H. 2020. Comparative chloroplast genomics and phylogenetic analysis of the *Viburnum dilatatum* complex (Adoxaceae) in Korea. *Korean J Plant Taxon* 50:8–16. <https://doi.org/10.11110/kjpt.2020.50.1.8>.
- Hasegawa E, Kobayashi K, Yagi N, Tsuji K. 2011. Complete mitochondrial genomes of normal and cheater morphs in the parthenogenetic ant *Pristomyrmex punctatus* (Hymenoptera: Formicidae). *Myrmecol News* 15:85–90.
- Liu N, Duan X-Y, Qian Z-Q, Wang X-Y, Li X-L, Ding M-Y. 2016. Characterization of the complete mitochondrial genome of the myrmicine ant *Vollenhovia emeryi* (Insecta: Hymenoptera: Formicidae). *Conserv Genet Resour* 8:211–214. <https://doi.org/10.1007/s12686-016-0535-x>.
- Rodovalho CM, Lyra ML, Ferro M, Bacci JM, Jr. 2014. The mitochondrial genome of the leaf-cutter ant *Atta laevigata*: a mitogenome with a large number of intergenic spacers. *PLoS One* 9:e97117. <https://doi.org/10.1371/journal.pone.0097117>.
- Duan X-Y, Peng X-Y, Qian Z-Q. 2016. The complete mitochondrial genomes of two globally invasive ants, the Argentine ant *Linepithema humile* and the little fire ant *Wasmannia auropunctata*. *Conserv Genet Resour* 8:275–277. <https://doi.org/10.1007/s12686-016-0555-6>.
- Gotzek D, Clarke J, Shoemaker D. 2010. Mitochondrial genome evolution in fire ants (Hymenoptera: Formicidae). *BMC Evol Biol* 10:300. <https://doi.org/10.1186/1471-2148-10-300>.
- Meza-Lázaro RN, Poteaux C, Bayona-Vásquez NJ, Branstetter MG, Zaldivar-Riverón A. 2018. Extensive mitochondrial heteroplasmy in the neotropical ants of the *Ectatomma ruidum* complex (Formicidae: Ectatomminae). *Mitochondrial DNA A DNA Mapp Seq Anal* 29:1203–1214. <https://doi.org/10.1080/24701394.2018.1431228>.
- Kim MJ, Hong EJ, Kim I. 2016. Complete mitochondrial genome of *Camponotus atrox* (Hymenoptera: Formicidae): a new tRNA arrangement in Hymenoptera. *Genome* 59:59–74. <https://doi.org/10.1139/gen-2015-0080>.
- Lee C-C, Wang J, Matsuura K, Yang C-CS. 2018. The complete mitochondrial genome of yellow crazy ant, *Anoplolepis gracilipes* (Hymenoptera: Formicidae). *Mitochondrial DNA B Resour* 3:622–623. <https://doi.org/10.1080/23802359.2018.1467739>.
- Park J, Xi H, Park J. 2020. The complete mitochondrial genome of *Nylanderia flavipes* (Smith, 1874) (Hymenoptera: Formicidae). *Mitochondrial DNA B Resour* 5:420–421. <https://doi.org/10.1080/23802359.2019.1703572>.
- Vieira GA, Prosdocimi F. 2019. Accessible molecular phylogenomics at no cost: obtaining 14 new mitogenomes for the ant subfamily Pseudomyrmecinae from public data. *PeerJ* 7:e6271. <https://doi.org/10.7717/peerj.6271>.
- Berman M, Austin CM, Miller AD. 2014. Characterisation of the complete mitochondrial genome and 13 microsatellite loci through next-generation sequencing for the New Caledonian spider-ant *Leptomyrmex pallens*. *Mol Biol Rep* 41:1179–1187. <https://doi.org/10.1007/s11033-013-2657-5>.
- Park J, Xi H, Park J. 2019. The complete mitochondrial genome of *Ochetellus glaber* (Mayr, 1862) (Hymenoptera: Formicidae). *Mitochondrial DNA B Resour* 5:147–149. <https://doi.org/10.1080/23802359.2019.1698356>.

25. Du Y, Song X, Yu H, Lu Z. 2019. Complete mitochondrial genome sequence of *Tapinoma melanocephalum* (Hymenoptera: Formicidae). Mitochondrial DNA B Resour 4:3448–3449. <https://doi.org/10.1080/23802359.2019.1674205>.
26. Park J, Choi YG, Yun N, Xi H, Min J, Kim Y, Oh S-H. 2019. The complete chloroplast genome sequence of *Viburnum erosum* (Adoxaceae). Mitochondrial DNA B Resour 4:3278–3279. <https://doi.org/10.1080/23802359.2019.1667919>.
27. McKenzie SK, Kronauer DJ. 2018. The genomic architecture and molecular evolution of ant odorant receptors. Genome Res 28:1757–1765. <https://doi.org/10.1101/gr.237123.118>.
28. Park J, Kwon W, Park J. 2019. The complete mitochondrial genome of *Cryptopone sauteri* Wheeler, WM, 1906 (Hymenoptera: Formicidae). Mitochondrial DNA B Resour 4:614–615. <https://doi.org/10.1080/23802359.2018.1561231>.
29. Park J, Kwon W, Park J. 2019. The complete mitochondrial genome of *Ectomomyrmex javanus* Mayr, 1867 (Hymenoptera: Formicidae). Mitochondrial DNA B Resour 4:1636–1637. <https://doi.org/10.1080/23802359.2019.1604095>.
30. Park J, Park J. 2021. Complete mitochondrial genome of the gate-keeper ant *Colobopsis nipponica* (Wheeler, W.M., 1928) (Formicidae: Hymenoptera). Mitochondrial DNA B Resour 6:86–88. <https://doi.org/10.1080/23802359.2020.1845581>.
31. Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res 18:821–829. <https://doi.org/10.1101/gr.074492.107>.
32. Bolger AM, Lohse M, Usadel B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics 30:2114–2120. <https://doi.org/10.1093/bioinformatics/btu170>.
33. Zhao Q-Y, Wang Y, Kong Y-M, Luo D, Li X, Hao P. 2011. Optimizing *de novo* transcriptome assembly from short-read RNA-Seq data: a comparative study. BMC Bioinformatics 12(Suppl 14):S2. <https://doi.org/10.1186/1471-2105-12-S14-S2>.
34. Li H, Durbin R. 2009. Fast and accurate short read alignment with Burrows–Wheeler transform. Bioinformatics 25:1754–1760. <https://doi.org/10.1093/bioinformatics/btp324>.
35. Li H, Handsaker B, Wysoker A, Fennell T, Ruan J, Homer N, Marth G, Abecasis G, Durbin R, 1000 Genome Project Data Processing Subgroup. 2009. The Sequence Alignment/Map format and SAMtools. Bioinformatics 25:2078–2079. <https://doi.org/10.1093/bioinformatics/btp352>.
36. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI Prokaryotic Genome Annotation Pipeline. Nucleic Acids Res 44:6614–6624. <https://doi.org/10.1093/nar/gkw569>.