





## Draft Genome Sequence of *Burkholderia* contaminans 293K04B, an Endosymbiont of the Sponge-Derived Fungus Stachylidium bicolor

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**ABSTRACT** Here, we present the draft genome of the endofungal symbiotic bacterium *Burkholderia contaminans* 293K04B, isolated from *Stachylidium bicolor* 293K04 (Ascomycota). The fungus was originally isolated from the sponge *Callyspongia* cf. *C. flammea*. *S. bicolor* 293K04 produces the endolides A-B, bioactive cyclic peptides possibly biosynthesized by its endobacterium *B. contaminans* 293K04B.

pecies of the genus Burkholderia are widely distributed in diverse habitats and are known as human and plant pathogens, plant growth promoters, and endosymbionts (1). Some defined bacterial endosymbionts have been recognized as true producers of secondary metabolites that were originally isolated from their hosts (2). Until recently, bacterial endosymbionts have rarely been seen in fungal hosts (3). The first secondary metabolites discovered to be produced by endofungal bacteria were the antimitotic rhizoxins, initially isolated from Rhizopus microsporus, and later found to be produced by its endosymbiont Burkholderia rhizoxinica (4). The heptapeptidic rhizonins contain units of the rare amino acid 3-(3-furyl)-alanine. These compounds were also isolated from cultures of a different R. microsporus strain, yet were later found to be produced by its endosymbiont Burkholderia endofungorum (5). The tetrapeptides endolides A and B, also comprising 3-(3-furyl)-alanine units, have been isolated from the marine-derived fungus S. bicolor 293K04. These compounds showed interaction with vasopressin and serotonin receptors (6). An endosymbiotic bacterial strain was successfully isolated from mycelia exposed to mechanical shearing. This strain belongs to the Burkholderia genus, and accurate phylogenetic placement of the strain was performed by multilocus sequence analysis (MLSA), further corroborated by in silico DNA-DNA hybridization experiments (Almeida C, Silva Pereira C, Gonzalez-Menendez V, Bills G, Pascual J, Sánchez-Hidalgo M, Kehraus S, and Genilloud O, unpublished data), which revealed it to be a member of the species B. contaminans. Hence, the bacterium was named B. contaminans 293K04B. Disclosing its genome may provide valuable insight for understanding the biology of its symbiotic relationship with S. bicolor 293K04, especially its role in the biosynthesis of endolides as a putative symbiotic function of the endobacteria.

DNA of *B. contaminans* 293K04B was isolated from a 2-day-old culture grown in liquid trypticase soy broth (TSB) medium. Genomic DNA was extracted according to the cetyltrimethylammonium bromide/polyvinylpyrrolidone (CTAB/PVP) protocol (7). Purification was achieved using the Purelink Genomic DNA minikit (Invitrogen) and yielded 100 ng/ $\mu$ L concentration with an OD<sub>260/280</sub> ratio of 1.86. Two sets of Illumina pairedend data were assembled using SPAdes, resulting in a draft genome sequence con-

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sisting of 100 contigs with a minimum size of 541 bp. The genome size of *Burkholderia* species is very variable from 2.4 Mb ("Candidatus Burkholderia schumannianae" UZH-bot8) (8) to more than 10 Mb, e.g., 11.5 Mb in the case of *Burkholderia terrae* BS001 (9). The genome of *B. contaminans* 293K04B measures 8.8 Mb. AntiSMASH (10) analysis revealed 16 biosynthetic gene clusters (BGCs) for the production of specialized metabolites. One BGC contains a polyketide synthase (PKS), 2 BGCs contain nonribosomal peptide synthetase (NRPS), and 1 BGC shows a hybrid PKS-NRPS.

**Accession number(s).** This draft genome has been deposited at DDBJ/EMBL/ GenBank under the accession number NQOD00000000. The version described in this paper is the first version, NQOD01000000.

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## **REFERENCES**

- Eberl L, Vandamme P. 2016. Members of the genus *Burkholderia*: good and bad guys. F1000Res 5:Rev-1007. https://doi.org/10.12688/f1000research .8221.1.
- Piel J. 2009. Metabolites from symbiotic bacteria. Nat Prod Rep 26: 338–362. https://doi.org/10.1039/b703499g.
- Lackner G, Partida-Martinez LP, Hertweck C. 2009. Endofungal bacteria as producers of mycotoxins. Trends Microbiol 17:570–576. https://doi.org/10.1016/j.tim.2009.09.003.
- Partida-Martinez LP, Hertweck C. 2005. Pathogenic fungus harbours endosymbiotic bacteria for toxin production. Nature 437:884–888. https://doi .org/10.1038/nature03997.
- Partida-Martinez LP, de Looss CF, Ishida K, Ishida M, Roth M, Buder K, Hertweck C. 2007. Rhizonin, the first mycotoxin isolated from the zygomycota, is not a fungal metabolite but is produced by bacterial endosymbionts. Appl Environ Microbiol 73:793–797. https://doi.org/10.1128/ Aem.01784-06.
- Almeida C, El Maddah F, Kehraus S, Schnakenburg G, König GM. 2016. Endolides A and B, vasopressin and serotonin-receptor Interacting N-methylated peptides from the sponge-derived fungus Stachylidium sp. Org Lett 18:528–531. https://doi.org/10.1021/acs.orglett.5b03553.

- Healey A, Furtado A, Cooper T, Henry RJ. 2014. Protocol: a simple method for extracting next-generation sequencing quality genomic DNA from recalcitrant plant species. Plant Methods 10:21. https://doi. org/10.1186/1746-4811-10-21.
- Pinto-Carbó M, Sieber S, Dessein S, Wicker T, Verstraete B, Gademann K, Eberl L, Carlier A. 2016. Evidence of horizontal gene transfer between obligate leaf nodule symbionts. ISME J 10:2092–2105. https://doi.org/10 .1038/ismej.2016.27.
- Nazir R, Hansen MA, Sørensen S, van Elsas JD. 2012. Draft genome sequence of the soil bacterium *Burkholderia terrae* strain BS001, which interacts with fungal surface structures. J Bacteriol 194:4480–4481. https://doi.org/10.1128/JB.00725-12.
- Weber T, Blin K, Duddela S, Krug D, Kim HU, Bruccoleri R, Lee SY, Fischbach MA, Müller R, Wohlleben W, Breitling R, Takano E, Medema MH. 2015. antiSMASH 3.0—A comprehensive resource for the genome mining of biosynthetic gene clusters. Nucleic Acids Res 43:W237–W243. https://doi.org/10.1093/nar/gkv437.