DATA NOTE





First complete genome sequence of a Bacterial Panicle Blight causing pathogen, Burkholderia glumae, isolated from symptomatic rice grains from Bangladesh

Ismam Ahmed Protic^{1,2}, Md. Nasir Uddin², Abu Sina Md. Tushar², Shah Tasdika Auyon³, David Alvarez-Ponce^{1*} and Md. Rashidul Islam^{2*}

Abstract

Objectives Rice (*Oryza sativa*) is the most important food for more than two thirds of the world's population. Bangladesh is the third largest producer and consumer of rice globally. Recently, several symptoms of Bacterial Panicle Blight (BPB) in rice, including seedling blight, sheath rot, floret sterility, and spotted grains, have been detected in the country. In addition, the presence of the most prevalent and virulent causative agent of BPB, Burkholderia alumae, has been confirmed in rice displaying symptoms of the disease. BPB could become one of the next emerging diseases of rice in Bangladesh, and a complete genome of a *B. glumae* strain from the country will help clarify its origin and devise proper management systems to continue sustainable rice production.

Data description We report the first complete genome sequence of a B. glumae strain (BD_21g) isolated from symptomatic rice grains in Bangladesh (Natore District). The genome contains 2 chromosomes (1 and 2, with 3,417,499 and 3,855,283 bp, respectively) and 4 plasmids (1-4, with 123,248, 46,628, 88,744 and 53,064 bp, respectively).

Keywords Bacterial Panicle Blight, Rice, Burkholderia glumae

Objective

Burkholderia glumae is a Gram-negative, aerobic bacterium that accounts for most of the cases of Bacterial Panicle Blight (BPB) in rice worldwide [1, 2]. BPB has become a threat to rice production globally. It can cause losses in grain yield and milling quality in epidemic years. The

*Correspondence: David Alvarez-Ponce dap@unr.edu Md. Rashidul Islam rashidul.islam@bau.edu.bd

² Plant Bacteriology and Biotechnology Laboratory, Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

disease causes several types of damage, including seedling blight, sheath rot, floret sterility, spotted grains, dark brown chaffy grains, and milling quality reduction, resulting in yield reductions of up to 75% [3, 4]. BPB-infected seeds act as the primary inoculum for BPB infection [5-8]. The symptoms of BPB often appear during the rice heading stage and are pronounced when rice is grown under high night temperatures and frequent rainfalls, predisposing rice to disease outbreaks [2, 9]. Occurrences of BPB are heavily dependent on weather conditions such as prolonged high daily minimum temperatures and frequent rainfall during the panicle emergence and flowering periods [9-11]. The disease was first noticed in Japan (1955) [5], then in most rice producing countries, and more recently in Bangladesh (2023) [3]. BPB has produced significant yield and economic losses worldwide; for



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¹ Department of Biology, University of Nevada, Reno, USA

³ Department of Environmental Science, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

instance, in the Mid-South US, BPB caused an estimated \$61 million in losses from 2003 to 2013 [12]. Bangladesh's humid, precipitous and warm climate is favorable for the spread of the disease. In addition, Bangladesh is the third largest rice producer globally, with 48% of rural employment, one sixth of the national income, and two thirds of the consumed calories depending on rice production [13]. Therefore, spread of the disease throughout the country would have severe effects.

The main virulence factor of *B. glumae* is toxoflavin, which generates hydrogen peroxide and causes tissue damage [14, 15]. Other virulence factors have also been identified; however, those are not completely characterized. Genome resources are essential for a better understanding of the mechanisms of action of BPB and to develop an effective management system to control the disease in Bangladesh.

Data description

Field surveys were conducted in 20 districts cultivating eight different rice varieties from mid-October to November 2022 in Bangladesh. Three locations in each district, and five fields in each location, were surveyed. All 300 fields contained some plants displaying the typical symptoms of BPB. From each field, a sample was collected and bacteria were isolated following Uddin et al. (Uddin, Protic, Tushar, Hasan, Saha, Singha, Sultana, Akter, Jinnah, Islam: First report of Burkholderia glumae causing Bacterial Panicle Blight in rice grains from Bangladesh, submitted revised version). From each sample, a bacterial suspension was streaked onto S-PG medium [16] and purple-colored colonies were purified as candidates. For those colonies showing the typical symptoms of toxoflavin production [6], genomic DNA was extracted using the Wizard® Genomic DNA Purification Kit (Promega). The 16S and gyrB genes were amplified using the 16SF (5'-AGAGTTTGATCC TGGCTCAG-3'), 16SR (5'-GGCTACCTTGTTACGACT T-3') [6], glu-FV (5'-GAAGTGTCGCCGATGGAG-3'), and glu-RV (5'-CCTTCACCGACAGCACGCAT-3') primers [17]. One of the samples—BD_21g, obtained from a farmer's field in the Natore district cultivating the Swarna variety, 24°21'0.00"N, 89°04'59.88"E—produced PCR products of the expected sizes for *B. glumae* [17]. After sequencing the PCR products from both genes, the obtained partial sequences were used for species identification via BLAST searches and confirmed as *B. glumae*. The partial sequences of 16S rDNA and *gyrB* were deposited in Genbank under accession numbers OR573691 and PP332812, respectively.

For whole genome sequencing, library preparation was conducted using the Illumina NovaSeq 6000 Reagent Kit, and 150-bp paired-end reads were obtained using the Illumina NovaSeq 6000 platform (Illumina, CA, USA). Illumina reads (Data set 1) were adapter trimmed using fastP v0.23.0 [18]. Assembly and annotation were conducted on the Galaxy platform [19]. Unicycler v.0.4.8 [20] was used for *de novo* assembly. Scaffolding was performed by mapping the trimmed reads to the *B. glumae* BGR1 genome [21] using BWA-MEM2 v.2.2.1 [22]. The resulting BAM file was used to polish the assembly with Pilon v.1.24 [23]. Genome annotation was performed using Prokka v.1.14.5 [24]. Default parameters were used for all programs.

The genome of *B. glumae* BD_21g is 7,584,466 bp long, exhibits a GC content of 68.5%, and contains 6806 protein-coding genes, 6352 mRNAs, 145 tRNAs, 7 rRNAs and 4 repeat regions (Data file 1, Data set 2). The genome contains 2 chromosomes (1 and 2, with 3,417,499 and 3,855,283 bp, respectively) and 4 plasmids (p1_21g-p4_21g, with 123,248, 46,628, 88,744 and 53,064 bp, respectively) (Data file 2). Chromosome 1 includes the house-keeping genes *gyrB*, *rpoD*, *atpD*, 16S and 23S. BLASTn v2.2.19 searches [25] revealed the presence of all toxoflavin-producing and quorum-sensing genes (*toxA*, *toxB*, *toxC*, *toxD*, *toxE*, *toxF*, *toxG*, *toxH*, *toxI* and *toxJ*) in chromosome 2 (Data file 3). A circular map of the genome was generated using the BV-BRC server [26] (Data file 4; Table 1).

Label	Name of data file/data set	File types (file extension)	Data repository and identifier (DOI or accession number)
Data file 1	Genome features of <i>B. glumae</i> BD_21g	MS Excel file (.xlsx)	Figshare (https://doi.org/10.6084/m9.figshare.25792 578) [29]
Data file 2	Features of specific chromosomes and plasmids	MS Excel file (.xlsx)	Figshare (https://doi.org/10.6084/m9.figshare.25792 578) [29]
Data file 3	List of housekeeping and toxoflavin producing genes with their residing chromosomes	MS Excel file (.xlsx)	Figshare (https://doi.org/10.6084/m9.figshare.25792 578) [29]
Data file 4	Circular map of the <i>Burkholderia glumae</i> BD_21g genome	PDF (.pdf)	Figshare (https://doi.org/10.6084/m9.figshare.25792 578) [29]
Data set 1	Burkholderia glumae BD_21g raw sequence reads	Raw sequence reads (.fastq)	NCBI Sequence Read Archive (http://identifiers.org/ insdc.sra:SRP440706) [30]
Data set 2	Burkholderia glumae BD_21g genome assembly	Standard fasta files (.fasta)	NCBI Assembly (http://identifiers.org/insdc.gca:GCA_030505635) [31]

 Table 1
 Overview of data files/data sets

We compared the *B. glumae* BD_21g genome with all 70 publicly available *B. glumae* genomes using fastANI [27]. The Average Nucleotide Identity (ANI) was 99.43% with the reference genome (257sh-1). The most similar genome to BD_21g was R2, which was obtained from diseased rice panicles from South Korea (ANI = 99.79%) [28].

Limitations

The emergence of BPB in Bangladesh has been reported recently [3]. During the survey, only one of the fields was found to be infested by *B. glumae*. Currently, this is the only genome resource for a *B. glumae* strain from Bangladesh. Therefore, a more comprehensive survey would be required to determine the present status of this pathogen.

Abbreviations

- BLAST Basic Local Alignment Search Tool
- NCBI National Center for Biotechnology Information
- BAM Binary Alignment Map
- ANI Average Nucleotide Identity
- BPB Bacterial Panicle Blight

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Authors' contributions

I.A.P.: PCR confirmation and partial sequence analyses based on 16S and gyrB genes, whole genome sequence data analyses, preparation of the manuscript draft and revision. M.N.U., A.S.M.T., and S.T.A.: sample collection, isolation, DNA preparation, PCR confirmation and partial sequence analyses of 16S and gyrB genes. M.R.I.: experimental planning and designing, draft preparation and partial editing, and supervision. D.A.-P.: technical editing of the manuscript, and supervision.

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Availability of data and materials

The genome assembly described in this Data Note can be freely and openly accessed on NCBI GenBank under accession number GCA_030505635. Raw sequence reads and associated details can be accessed on BioProject PRJNA978594. Associated Data files are available on Figshare (https://doi.org/10.6084/m9.figshare.25792578) [29]. Please see Table 1 and refs. [29–31] for further details on the genome. Rice grains were preserved as dry herbarium specimens both in the Department of Plant Pathology and in the Agricultural University.

Declarations

Ethics approval and consent to participate

Rice grains were obtained from a private field with permission from the owner.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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