A New Biocontrol Agent *Bacillus velezensis* SF334 Against Fungal Leaf Anthracnose of Rubber Tree and Its Genome Analysis for Versatile Plant Probiotic Traits

Muyuan Wang 1,†, Yikun Zhang 1,†, Xinyang Zhao3,, Haibin Cai2, Zhongfeng Zhu1, Yichao Yan1, Ke Yin1, Guanyun Cheng1, Yinsheng Li1, Gongyou Chen1,5, Lifang Zou1, 5, \*, Min Tu2, 4, \*

**Table S1. The plant growth promotion associated genes in *B*. *velezensis* SF334**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Gene ID** | **KO** | **Gene** | **Function** | **Class** |
| SF334GL002344 | K01695 | *trpA* | tryptophan synthase alpha chain | Phenylalanine, tyrosine and tryptophan biosynthesis |
| SF334GL003147 | K14155 | *patB*, *malY* | cysteine-S-conjugate beta-lyase | Biosynthesis of secondary metabolites |
| SF334GL002056 | K00128 | *aldh* | aldehyde dehydrogenase (NAD+) | Tryptophan metabolism |
| SF334GL002972 | K00128 | *aldh* | aldehyde dehydrogenase (NAD+) | Tryptophan metabolism |
| SF334GL004032 | K00128 | *aldh* | aldehyde dehydrogenase (NAD+) | Tryptophan metabolism |
| SF334GL002345 | K01696 | *trpB* | tryptophan synthase beta chain | Phenylalanine, tyrosine and tryptophan biosynthesis |
| SF334GL002347 | K01609 | *trpC* | indole-3-glycerol phosphate synthase | Phenylalanine, tyrosine and tryptophan biosynthesis |

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **General features** | ***B*. *velezensis***  **SF334** | ***B*. *velezensis***  **FZB42** | ***B*. *velezensis***  **SQR9** | ***B*. *amyloliquefaciens***  **DSM7** | ***B*. *subtilis***  **168** |
| Genome size (bp) | 4,078,641 | 3,918,596 | 4,117,023 | 3,980,199 | 4,227,167 |
| GC content | 46.5% | 46.48% | 46.10% | 46.08% | 43.49% |
| Coding density (%) | 89.33 | 89.026 | 89.68 | 88.54 | 88.33 |
| Protein coding sequences (CDS) | 4,142 | 3,888 | 4,111 | 4,220 | 4,388 |
| tRNA | 86 | 89 | 72 | 94 | 86 |
| 5s rRNA | 9 | 10 | 7 | 10 | 10 |
| 16s rRNA | 9 | 9 | 7 | 10 | 10 |
| 23s rRNA | 9 | 10 | 7 | 10 | 10 |
| Repeat region | 30 | 31 | 20 | 43 | 38 |

**Table S2. General features of genomes of *B*. *velezensis* SF334, *B*. *velezensis* FZB42, *B*. *velezensis* SQR9, *B*. *amyloliquefaciens* DSM7 and *B*. *subtilis 168***



**Figure. S1.** Antagonistic effect of strain SF334 against *C. siamense* and *C. australisinense,* which are major pathogens causing leaf anthracnose of rubber trees in Hainan province of China. The average inhibition rates of SF334 against *C. siamense* and *C. australisinense* were calculated.

**

**Figure S2.** The phylogenetic tree of strain SF334 based on *16S rDNA* sequences constructed through the TYGS platform. The sequence of *16s rDNA* of 39 strains with the lowest E-value in the National Center for Biotechnology Information (NCBI) database were selected as the reference sequences. The results were finally imported into MEGA 11 soft to construct a phylogenetic tree by the NJ (1200 bootstrap) method.

****

**Figure S3.** Observation of the hyphal lysis of *C*. *siamense* and *C*. *australisinense* when interacting with the CS of *B. velezensis* SF334 on PDA plates. The mycelium of *C*. *siamense* and *C*. *australisinense* were inoculated with the indicated bacterial concentration for 0.5 h, 1h, 1.5 h, 2.5 h and 4 h.



**Figure S4.** Observation of mycelium morphology of *C*. *siamense* and *C*. *australisinense* when interacting with the CS of *B. velezensis* SF334 for 1 h, 3 h, 6 h, and 12 h in PDB medium by optical microscope. Scale bar, 10 μm.

****

**Figure S5.** Genomic analysis of *B. velezensis* SF334. The COG analysis (A), GO analysis (B), KEGG pathway analysis (C) and CAzy annotation (D) of *B. velezensis* SF334



**Figure S6.** Comparative genomic analysis of *B. velezensis* SF334 with other related four *Bacilus* species. The COG analysis (A) and KEGG pathway analysis (B) of *B. velezensis* SF334 with *B*. *velezensis* FZB42, *B*. *velezensis* SQR9, *B*. *amyloliquefaciens* DSM7 and *B*. *subtilis* 168. Green, red and blue boxes indicate core genes, accessory and unique genes between five strains.