A new macrolactin derivate, 7-O-2′E-butenoyl macrolactin A (1), together with three known macrolactin compounds, macrolactin A (2), 7-O-malonyl macrolactin A (3) and 7-O-succinyl macrolactin A (4), was isolated from the bacterial strain *Bacillus subtilis* B5, which was isolated from the 3000 metres deep sea sediment of the Southwest Pacific Ocean. The structures of the new compounds were assigned by spectroscopic methods including 1D/2D NMR and MS analysis techniques. Compounds 1 and 2 demonstrated antifungal activities against tea pathogenic fungi *Pestalotiopsis theae* and *Colletotrichum gloeosporioides*.

**Keywords:** *Bacillus subtilis* B5; 7-O-2′E -butenoyl macrolactin A; antifungal activity.
List

Figure S1. The $^1$H-NMR Spectrum of Compound 1 in DMSO-$d_6$

Figure S2. The $^{13}$C-NMR Spectrum of Compound 1 in DMSO-$d_6$

Figure S3. The HSQC Spectrum of Compound 1 in DMSO-$d_6$

Figure S4. The HMBC Spectrum of Compound 1 in DMSO-$d_6$

Figure S5. The 1H-1H COSY Spectrum of Compound 1 in DMSO-$d_6$

Figure S6. The HRESI-MS Spectrum of Compound 1

Figure S7. Detection of inhibitory activity of compound 1 (1) and compound 2 (2) against pathogenic fungi *Colletotrichum gloeosporioides* (A) and *Pestalotiopsis theae* (B)
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