

**Title** Biological active products from the antagonist *Bacillus subtilis* for the control of fruit-rot diseases in citrus

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### Abstract

The present research aims to screen a suitable medium for cultivation of *Bacillus subtilis* strain ABS-S14, with respect to obtain high production of antifungal substances. Crude extract of its cell free culture was partially purified and the biological efficacy of both bio active substances and formulated bacterial endospores against green mold rot on citrus fruits at post-harvest stage was investigated. *B. subtilis* ABS-S14 showed potent inhibition in control of *Penicillium digitatum* Sacc. pathogen, a cause fruit rot disease of citrus. Highest growth in PDB culture and strongest activity (87% inhibition) against growth of the fungus were detected at 72 hours. An ethanol extract from *B. subtilis* culture supernatant produced inhibitory effect on mycelial growth with EC50 at 20.54 µg/ml. Inhibitory compounds were obtained from TLC of the ethanol extract. Further analysis on HPLC chromatography revealed peaks of some potent antibiotics of *B. subtilis* ABS-S14. The efficacy of *B. subtilis* in controlling citrus fruit rot was shown when inoculation with *B. subtilis* endospores 24 h prior to fungal spore inoculation while addition of 10 mg/ml of the ethanol extract, together with the fungus, produced highest disease suppression in the same manner as imazalil treatment. The remarkable fungal growth inhibition on citrus fruits was demonstrated when a mixture of crude extracts and endospores of *B. subtilis* ABS-S 14 were applied before pathogen challenge. The invented formulation of bioactive products of *B. subtilis* ABS-S 14 (oil-in-water) was stable after freeze & thaw. Bio-assay tests on infect wounded citrus fruits were carried out and discussed.