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Multiple Antibiotics Resistance of a Soil-Borne Bacterium Burkholderia cepacia

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Several antagonistic bacteria producing chitinase and antifungal component were isolated from forest soil by a chitin-bating method. Among them, *Burkholderia cepacia* CH-67 strain has antagonistic activity against many of fungal plant pathogens. However the bacteria exhibited resistance to many aminoglycoside antibiotics such as kanamycin, streptomycin, and neomycin but not to trimethoprim. To search for the antibiotics resistance gene and its characterization, we constructed genomic library of the strain CH-67 using a fosmid vector. Among aminoglycoside antibiotics, we selected kanamycin for initial antibiotics resistance screening from the genomic library and could select 10 kanamycin-resistance clones. One of them pF67Km confers kanamycin resistance constantly on retransformed *Escherichia coli*. The clone pF67Km conferred resistance to aminoglycoside antibiotics such as gentamycin, streptomycin and spectinomycin on *E. coli*. The antibiotic resistance gene from pF67Km clone carrying 40 kb insert DNA will be identified through subcloning and mutagenesis analysis.

Key words: Aminoglycoside, Burkholderia cepacia, multiple antibioitics resistance

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Field Evaluation of Biocontrol Formulation without live Biocontrol Strain CH-67

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Previously, we showed that a biocontrol bacterium *Burkholderia cepacia* CH-67 is highly effective to control tomato leaf mold caused by *Fulvia fulva*. Various formulations of the CH-67 strain were tested for biocontrol activity against tomato leaf mold in a plastic house and one of formulations CH-67C containing starches and sugars was the most effective in the plastic house. One of features of CH-67C is the elimination of live bacteria CH-67 strain during formulation process indicating that the culture metabolites are effective to exhibit the biocontrol activity and highly stable during the preparation. The biocontrol activity of CH-67C was evaluated in the tomato production condition under natural disease occurrence. The 100-fold diluted application of CH-67C was effective to control the disease to the equivalent level of disease control value of cheimical fungicide triflumizole but it was the more effective than other biocontrol formulations such as A2-MP generated from other biocontrol strain *Bacillus amyloliquefaciens* A-2. This result suggested that the live bacteria free formulation may have a potential as a biocontrol agent.

Key words: Biocontrol, Burkholderia cepacia, live bacteria free