mccEh252, A NEW MICROCIN PRODUCED BY ERWINIA HERBICOLA

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The biocontrol agent of fire blight, *Erwinia herbicola* Eh252, produces a compound which inhibits the fire blight pathogen and other strains of *Erwinia* in vitro. The compound passed through a dialysis membrane with a cut off point of 3 000 MW and lost its activity in presence of pronase, proteinase K or L-histidine but not in presence of D-histidine or Fe++. It retained activity after exposure to high temperatures or to extreme pH. Eh252 produced this compound only when grown below 28°C. The compound was detected after 8 h growth and continued to accumulate in the supernatant for up to three days. Genes necessary for production of this compound have been cloned and sequenced. Analysis of the DNA sequence revealed that a 62 base pair segment shared 57% homology with the gene *mccA* isolated from *E. coli* which codes for the peptidic structure of microcin C7. We also found homology between the amino acid sequence of Eh252 and MccB which is believed to adenylate the product *mccA*. These results indicate that the compound produced by Eh252 is a new member of the microcin family.

ESTIMATING BIOMASS OF BOTRYTIS CINEREA

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The estimation of biomass of a pathogen at an early stage of disease development could be useful for infection studies and could save considerable time when assessing the efficacy of biocontrol agents (e.g. for control of *Botrytis cinerea* infections of kiwifruit). Glutaraldehyde has been shown to give useful auto-fluorescence of fungal hyphae in wood. We used this technique to study the effect of biological control agents on *Botrytis cinerea* infection of bean and lettuce leaves by Laser Scanning Confocal Microscopy (LSCM). Bacterial isolates of *Enterobacter agglomerans* and *E. aerogenes*, and yeast isolates of *Candida sake* and *Trichosporon pullulans* which had already shown promise against *B. cinerea* infection of kiwifruit and tomato were used in this work. Images of glutaraldehyde-fixed specimens were produced by LSCM and manipulated by an image analysis package to remove background fluorescence from host tissue and thus facilitate measurement of the volume of *B. cinerea* hyphae. Hyphal volume was used as an indicator of fungal biomass. There was less hyphal development in leaf tissue when bacterial and yeast biocontrol agents were applied together with the pathogen than in the *B. cinerea*-only treatment.