

Production of Bacillus Lipopeptides for Biocontrol of Postharvest Crops

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Abstract : With overpopulation threatening the world's ability to feed itself, food production and protection has become a major issue, especially in developing countries. Almost one-third of the food produced for human consumption, around 1.3 billion tonnes, is either wasted or lost annually. Postharvest decay in particular constitutes a major cause of crop loss with about 20% of fruits and vegetables produced lost during postharvest storage, mainly due to fungal disease. Some of the major phytopathogenic fungi affecting postharvest fruit crops in South Africa include *Aspergillus*, *Botrytis*, *Penicillium*, *Alternaria* and *Sclerotinia* spp. To date control of fungal phytopathogens has primarily been dependent on synthetic chemical fungicides, but these chemicals pose a significant threat to the environment, mainly due to their xenobiotic properties and tendency to generate resistance in the phytopathogens. Here, an environmentally benign alternative approach to control postharvest fungal phytopathogens in perishable fruit crops has been presented, namely the application of a bio-fungicide in the form of lipopeptide molecules. Lipopeptides are biosurfactants produced by *Bacillus* spp. which have been established as green, nontoxic and biodegradable molecules with antimicrobial properties. However, since the *Bacillus* are capable of producing a large number of lipopeptide homologues with differing efficacies against distinct target organisms, the lipopeptide production conditions and strategy are critical to produce the maximum lipopeptide concentration with homologue ratios to specification for optimum bio-fungicide efficacy. Process conditions, and their impact on *Bacillus* lipopeptide production, were evaluated in fully instrumented laboratory scale bioreactors under well-regulated controlled and defined environments. Factors such as the oxygen availability and trace element and nitrate concentrations had profound influences on lipopeptide yield, productivity and selectivity. Lipopeptide yield and homologue selectivity were enhanced in cultures where the oxygen in the sparge gas was increased from 21 to 30 mole%. The addition of trace elements, particularly Fe^{2+} , increased the total concentration of lipopeptides and a nitrate concentration equivalent to 8 g/L ammonium nitrate resulted in optimum lipopeptide yield and homologue selectivity. Efficacy studies of the culture supernatant containing the crude lipopeptide mixture were conducted using phytopathogens isolated from fruit in the field, identified using genetic sequencing. The supernatant exhibited antifungal activity against all the test-isolates, namely *Lewia*, *Botrytis*, *Penicillium*, *Alternaria* and *Sclerotinia* spp., even in this crude form. Thus the lipopeptide product efficacy has been confirmed to control the main diseases, even in the basic crude form. Future studies will be directed towards purification of the lipopeptide product and enhancement of efficacy.

Keywords : antifungal efficacy, biocontrol, lipopeptide production, perishable crops

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