Control of Post-Harvest Disease of Rambutan and Annona by Using a Biocontrol Agent

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I. Introduction

The Industrial Technology Institute (ITI) of Colombo, Sri Lanka, is involved in developing biological control mechanisms for post-harvest diseases of tropical fruits as well as working together with the International Centre for Underutilised Crops (ICUC) on promising underutilised species.

***Botryodiplodia* sp. is causing the stem end rot disease on many fruits and is listed as a major pre- and postharvest disease for rambutan and *Annona* species. Using *Trichoderma* sp. to fight stem end rot disease is an alternative to chemical treatments and would provide a cheaper option for small-holder farmers.**

This study established whether biocontrol (BC) agents are widely available in Sri Lanka and tested the effectiveness of the BC agents against *Botryodiplodia* sp. in a bio-assay. A second experiment tested whether diseased fruits of various species infect each other (cross-inoculation).

II. Material and method

1. Bio-assay:

Petri dishes filled with 15 ml PDA-medium (Potato-Dextrose-Agar) were used for the bio-assay experiment. Four wells were cut in the outer area of the plate by useing a cork-borer (No. 5) and filled with 0.8 ml spore suspension of the BC agents futhermore a mycelial disc was placed in the middle of the plate (Figure 1).
All plates were kept in an incubator at a temperature of 28°C for seven days. Five replications were done for each pathogen and the control, in which the wells were filled with sterile distilled water. (Figure 4)

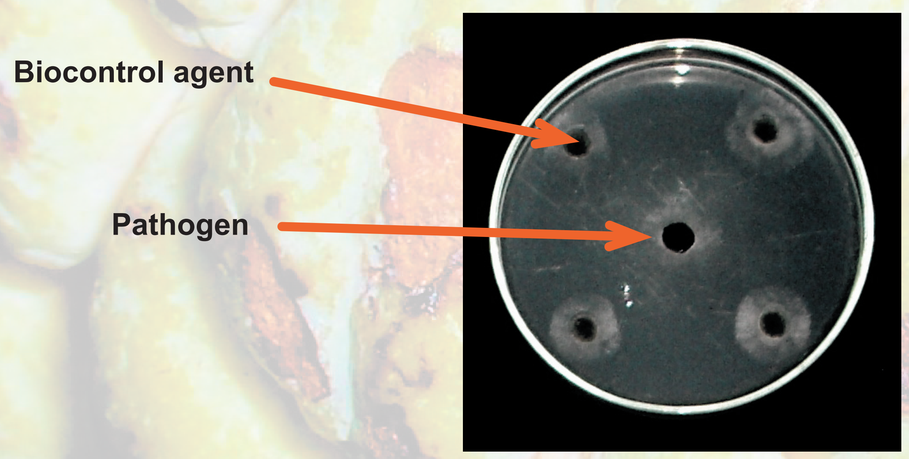

Figure 1: Preparation of a bio-assay plate

2. Cross-inoculation:

After a surface sterilization of the fruits one well per fruit was cut near the stem and a mycelial disc was inserted.
The fruits were kept in humidity chambers at ambient temperature for five days.
Ten fruits were used per pathogen and control. In control 1 the wells were filled with sterile destilled water; in control 2 the fruits were kept without any further treatment.
Per cent severity was assessed daily and the experiment was replicated twice. (Figure 2)

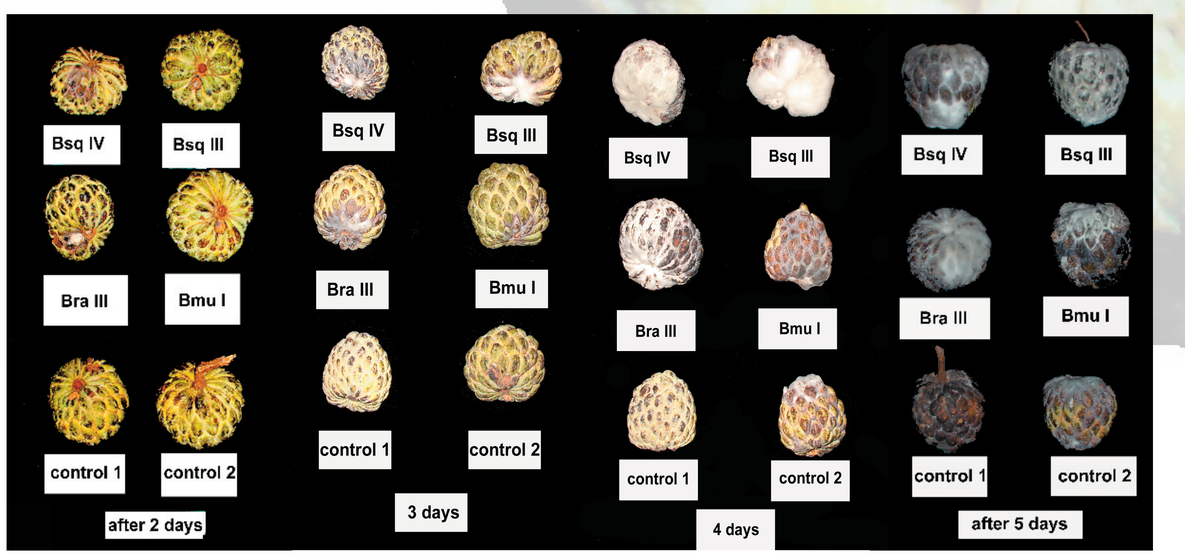

Figure 2: Cross-inoculation experiment day 2 (left) to day 5 (right)

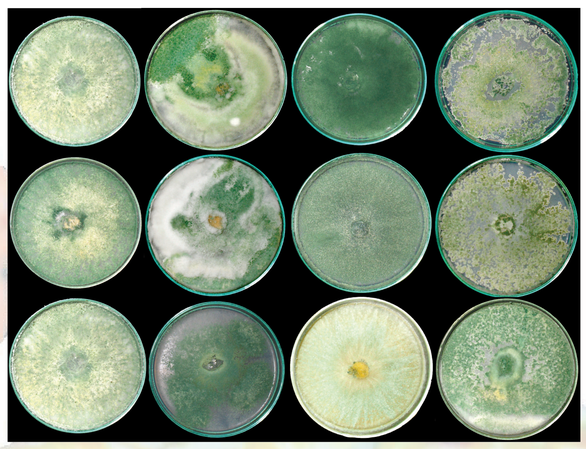

Figure 3: The 12 isolated *Trichoderma* strains

Code	Source	BC activity
Tra I	Urapola	++
Tra II	Warakapola	+++
Tra III	Warakapola	+++
Tsq I	Matale	0
Tsq II	Matale	+
Tsq III	Wijayapura	++
Tsq IV	Medawachchiya	0
Tmu I	Galagedera	++
Tmu II	Galagedera	++
Tmu III	Galagedera	0
Tmu IV	Matale	+
Tmu V	Wijayapura	+++

Table 1: Overview of the *Trichoderma* strains and their biocontrol (BC) activity (+++=high BC activity, ++=moderate BC activity, +=low BC activity, 0=non BC activity)

III. Results

Isolates:

Twelve different *Trichoderma* strains were found in six different soil-sources (Figure 3) and eight pathogens isolated from diseased fruits.

Bio-assay:

Three of the *Trichoderma* isolates have a high, four have a moderate and three have a low BC activity. Three isolates were not effective. (Table 1)

Cross-inoculation experiment:

The pathogens which were cross-inoculated into the rambutan fruits showed similar severity after the third day.
The cross-inoculation experiment on *Annona muricata* fruits did not lead to clear results.
The variability in the cross-inoculation experiment with *Annona squamosa* fruits was high therefore did the experiment not lead to clear results as well. Nevertheless we can conclude that all pathogens of the various fruits do infect the others.

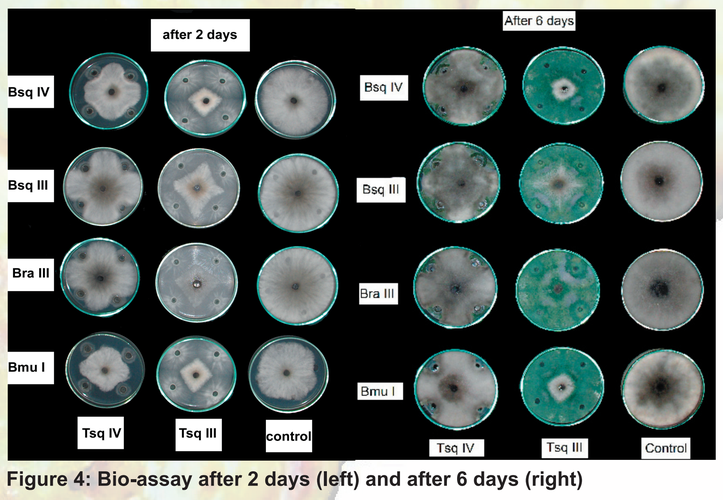

Figure 4: Bio-assay after 2 days (left) and after 6 days (right)

IV. Conclusion

The study shows that *Thrichoderma* strains are widely available in Sri Lanka and that some of these strains are effective as BC agents.

The *Trichoderma* strains isolated from soil-samples taken from rambutan sources have a rather good BC activity compared to the others strains. But only one *Trichoderma* strain which was isolated from soil surrounding a *Annona muricata* tree could completely control the pathogen isolated from a red *Annona squamosa* fruit.

All pathogens which were isolated from various fruits are able to infect all fruits. This result shows that rambutan and annona should not be stored or transported together. To establish a plantation of these fruits next to each other can lead to difficulties due to the cross-infection.

In further studies the BC effectiveness of the *Trichoderma* strains should be proved in an in vivo study. This could be done by introducing the BC agents into the soil of rambutan or annona plantations or by dipping the fruits into a solution of the BC agents after havesting.
The effect of *Trichoderma* sp. against other microorganisms especially against those who benefits the crops should be proved in futher studies as well.

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