EFFICACY OF *Trichoderma* sp. TO CONTROL *Lasiodiplodia theobromae*, CAUSAL AGENT OF SWEETPOTATO ROOT ROT DISEASE

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ABSTRACT

*Microbial control of sweetpotato root rot pathogen (**Lasiodiplodia theobromae** (Pat.) Griff. & Maubi.) was studied with the use of an antagonist (**Trichoderma** sp.) under screenhouse and field conditions. Application of 10-60g **Trichoderma** Fl7c suppressed **L. theobromae** infection and reduced the incidence of root rotting during storage especially if applied two to three months after planting of sweetpotato in pots. Likewise, in field experiments, regardless of inoculum densities of **Trichoderma** Fl7c (50, 75 and 100 g), root infection was reduced during storage when the antagonist was applied two and three months after planting than when applied during planting to one month. Control provided by **Trichoderma** Fl7c was economically sound and even better compared to the effect of Benlate, a systemic fungicide.*

KEY WORDS: Microbial control. Antagonist. *Trichoderma* sp. Sweetpotato root rot. *Lasiodiplodia theobromae*. Economics of control.

INTRODUCTION

Root crops have been identified as good source of energy for people and animals especially in terms of food security. Increasing demand for root crops such as sweetpotato has resulted in farmers cultivating large areas and also in successive cropping seasons. The practice subsequently resulted to serious occurrence of pests and diseases at preharvest and even until storage.

Important contributors to yield loss include a postharvest disease of sweetpotato called root rot. Root rot is considered a major postharvest disease problem and in the Philippines, several fungi have been associated with rapid deterioration of harvested roots. *L. theobromae* has been observed to cause the highest disease incidence (24%) in sweetpotato storage sheds and screenhouse (Palomar *et al.*, 1980).

Several naturally occurring biological agents have been screened against postharvest pathogens of sweetpotato roots (Palomar *et al.*, 1998). *In vitro* experiments from the initial phase of the biological control project at Leyte State University (LSU) found that *Trichoderma* isolate FI7c was the most effective among the fungal isolates collected and tested against *L. theobromae*.

This study, therefore, was conducted to verify the effectivity of *Trichoderma* isolate FI7c against *L. theobromae* under screenhouse and field conditions, to determine the effect of timing of application of the antagonist in its activity as biocontrol agent and to assess the economic potential of such control measure.

METHODOLOGY

Screenhouse Experiment

Sweetpotato cuttings (VSP-5 variety) were planted in sterilized soil inside clay pots. All pots were applied once with 50 g of *L. theobromae* in a substrate. The pots were arranged in a two-factor factorial design in a completely randomized block design (RCBD) replicated three times. Treatments included different inoculum densities (20, 30, 40, 50 and 60 g) of *Trichoderma* applied to the soil in pots planted with sweetpotato at different times (at planting, one, two and 3 months) after planting. Plants in pots not treated with *Trichoderma* but only applied with *L. theobromae* were included as control check. Four months after planting, plants were harvested and newly harvested roots were brought to the laboratory for storage. Root rotting was noted during harvest and after two months of storage.

Field Test

Sweetpotato VSP-5 cuttings were planted using a 0.25 x 0.75-cm planting distance. Plants were inoculated with one-month old culture of *L. theobromae* multiplied on 1:3 of harvested empty rice grain and sawdust. One hundred and fifty grams of organic substrate with *L. theobromae* were applied at the base of the plant according to the treatment used. All plants were applied with *L. theobromae* during planting. Likewise, 50, 75, 100 g of *Trichoderma* FI7c cultured in 1:1 ratio of rice bran and sawdust were placed at the base of the plant following the different time of application as indicated in treatments used. Benlate (Benomyl) as fungicidal check and treated control of *L. theobromae* alone were also included in the experimental set-up. The

trial was laid out in a 3 factorial design replicated 3 times. Plots were separated with 1-meter borders but planted with sweetpotato to prevent inter-plot interference from *L. theobromae* and the antagonist. The observation of root rotting was recorded during harvest and after two months of storage. The number and weight of marketable roots were also determined.

Cost and Return Analysis

Cost inputs were recorded to determine whether application of 100 g. *Trichoderma* FI7c grown in organic substrate to control root rot caused by *L. theobromae* was economically feasible compared to the application of Benlate. Percent infection and weight of roots harvested were used as the basis for comparison and the expenses incurred in the production of *Trichoderma* FI7c in organic substrate and Benlate was converted into one hectare.

RESULTS AND DISCUSSION

Control of Sweetpotato Root Rot Using *Trichoderma* FI7c

No rotting was observed in newly harvested roots of sweetpotato in pots. After two months of storage, there was a significant reduction in percent infection and percent disease control of stored roots from plants treated with different inoculum densities of *Trichoderma* FI7c and Benlate compared to the untreated control (Figure 1). Moreover, the lowest percent infected roots (7.14 and 8.33) and highest percent disease control (74.11 and 72.73) were recorded from plants with 60g *Trichoderma* FI7c at two and three months after planting. Both treatments had the same significant

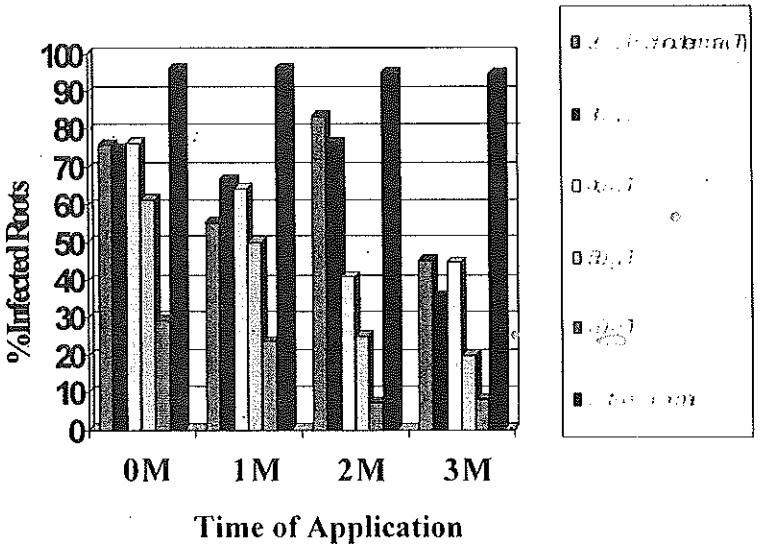

Figure 1. Effect of different *Trichoderma* (F17c) inoculum densities in pot experiments on rotting of sweetpotato roots.

value based on percent infected roots and disease control. The results indicate that in pot experiments *Trichoderma* FI7c when applied at different inoculum levels (10-60g) in each plant planted in infested soil with *L. theobromae* significantly decreased percent infection of roots or reduced the possibility of postharvest disease occurrence. Furthermore, there was a highly significant interaction between time of application and inoculum level of *Trichoderma*. Applying 10-60 g of *Trichoderma* FI7c suppressed *L. theobromae* and reduced the incidence of root rotting after harvest until marketing and up to storage.

There was a significant difference in number of marketable roots (28.70 and 26.35 g) after field harvest, percent infection (66.83 and 79.00) and percent disease control (28.66 and 22.97) after two months of storage from roots of plants inoculated with different levels of *L. theo*bromae at 50 and 100 g, respectively. However, there was no significant difference in weight of marketable roots recorded from plants separately inoculated with the two levels of the pathogen (Table 1). The results suggest that 50-g inoculum level of *L. theobromae* is capable of infection in roots after harvest especially if they are stored. Higher percent infection will also be expected if higher population of *L. theobromae* is found in the soil without affecting the number and weight of harvested marketable root.

Table 1. Effect of different inoculum levels of *L. theobromae* on percent infection, number of roots, weight of marketable roots and disease control.

Inoculum Level (g)	% Infection	No. of Roots	Wt. of Marketable Roots(kg)	% Disease Control
50	66.83b	28.70a	2.89	28.66
100	79.00a	26.35b	2.78	22.97
LSD	5.29	2.06	0.24	3.92
C. V. (%)	19.77	20.58	22.99	41.74
Level of significance	**	*	ns	**

Time of Application

The effect of time of application regardless of level of L. *theobromae and* Tri*choderma Fl7*c used (Table 2) revealed no significant difference in the number and weight of marketable roots and percent disease control; however, there was a significant reduction of roots infected (66.33%) on stored roots from plants applied with Tri*choderma Fl7*c at three months after planting followed by two months after planting (71.67%). Root infection from sweetpotato plants applied with Tri*choderma Fl7*c one month after planting was comparable to two months after planting. There was also a decrease in disease control, number and weight of roots after harvest up to storage but they were insignificant. The results show that regardless of L. *theobromae* inoculum level and different level of *Trichoderma* Fl7c (50, 75, and 100 g), it is more advantageous to apply the antagonist at the later part of the vegetative stage of the sweetpotato plants to prevent rotting by *L. theobromae* at harvest or during storage.

High infection of roots on treatments applied at zero and one month after planting maybe due to the activity of L. *theobromae* propagules that survived from the attack of *Trichoderma.* The infectious propagules of the pathogen must have colonized the tissues during harvest by entering at the root attachment from the stem or wounds due to cut from a knife or other tools for harvesting. This corroborates the findings of Ogundana (1982) that *L. theobromae* infects through the entrance by wounds and natural openings right after harvest.

Inoculum Level

There was a significant reduction of percent infection of root and disease control using different levels of *Trichoderma* Fl7c compared to the

the control (*L. theobromae* alone) (Table 3). One hundred grams of inoculum had the lowest percent infection and highest disease control value (60.00 and 37.70%) followed by 75 (65.83 and 32.75%) and 50 (70.83 and 29.35%) grams of *Trichoderma* FI7c. Fifty and 75 g inoculum densities of the antagonist were comparable to the effect of Benlate (72.08 and 28.45%) in terms of infection and disease control. The results demonstrate that the use of 100 g of *Trichoderma* FI7c is advantageous compared to other inoculum levels in terms of percent infected roots and disease control at harvest up to storage. No significant interaction was recorded between the levels of *L. theobromae* in weight of marketable roots, different inoculum densities of *Trichoderma* FI7c in number and weight of roots, and time of application in number and weight of marketable roots and disease control.

Table 3. Effect of different inoculum levels of *Trichoderma* FI7c on percent infection, number of roots, weight of marketable roots and disease control.

Treatment	% Infection	No. of Roots	Wt. of Marketable Roots (kg)	% Disease Control
50g *Trichoderma*	70.83ab	27.00	2.72	29.35c
75g *Trichoderma*	65.83ab	28.38	3.03	32.75b
100g *Trichoderma*	60.00a	26.75	2.61	37.70a
Benlate (RR)	72.08b	27.79	2.86	28.45c
L. theobromae alone	95.83c	27.71	2.95	0.81d
LSD	8.37	3.26	0.37	2.20
C. V. (%)	19.77	20.58	22.99	41.74
Level of Significance	**	ns	ns	**

Economic Analysis

The comparative cost of inputs between application of Benlate (fungicides) and 100 g *Trichoderma* FI7c (Table 4) considering all other inputs were the same but the expenses using Benlate was lower compared to *Trichoderma*. Benlate treatment was better in terms of

Table 4. Cost of inputs of applying Benlate and *Trichoderma* FI7c per hectare.

A. Benlate (Fungicide)			
1. Recommended Rate 0.33 kg /ha	₱3.40/g	₱1,122.00	
2. Labor Cost of (2 man-days) of application	₱200.00/day	400.00	
	Total		₱1,522.00
B. 100g *Trichoderma* FI7c			
1. Cost of Preparation of Pure Cultures in Potato Dextrose Agar			₱ 205.45
200g potato		8.00	
15g		39.00	
15g dextrose agar		19.50	
1500 ml distilled water		3.95	
Fuel expenses		35.00	
Labor cost		100.00	
		₱205.45	
2. Cost of Mass Production of Organic Substrate			₱3,140.00
rice bran 550kg @P3.00/kg		1,650.00	
saw dust			
plastic bags, 3300 pcs. @ 15cts.		990.00	
labor cost (2 man-days)		400.00	
fuel for cooking/sterilization		100.00	
		P3,140.00	
3. Labor Cost of Application (2 man-days)			400.00
4. Transportation for hauling			100.00
		Total	**₱3,845.45**

net sales after harvest (Table 5a). However, when expenses were equated to the amount of root left after deducting the percentage infection after storage, the *Trichoderma* treatment had higher return of ₱41,944.55 compared to Benlate treatment. (₱33,600.80) with a difference of ₱8,343.75 (Table 5b). Thus, using 100g *Trichoderma* F17c is better than Benlate especially if the harvest cannot be sold immediately or if it is being used for commercial purposes and cannot be processed immediately.

Table 5a. Cost and return analysis of the application of *Trichoderma* Fl7c and Benlate in controlling root rot caused by *L. theobromae* at harvest.

Treatment	Total Cost of Application/ha (₱)	Wt. of Roots (kg)	Sales of Roots (₱10.00/kg)	Net Return (₱)
Benlate	₱1,522.00	12,543.86	₱125,438.60	₱123, 916.60
Trichoderma Fl7c 100g	₱3,845.45	11,447.37	₱114,473.70	₱110,628.25

Table 5b. Cost and return analysis of the application of *Trichoderma* Fl7c and Benlate in controlling root rot caused by *L. theobromae* after storage.

Treatment	Total Cost of Application/ha (₱)	Wt. of Roots Less % Infection (kg)	Sales of Roots (₱10.00/kg)	Net Return (₱)
Benlate	₱1,522.00	3,512.28	₱35,122.80	₱33,600.80
Trichoderma Fl7c 100g	₱3,845.45	4,578.95	₱45, 789.50	₱41,944.55

CONCLUSION AND RECOMMENDATION

Highly significant interaction was observed between time of application and different levels of *Trichoderma* FI7c grown in organic substrate against infection of *L. theobromae* at harvest in pot experiments. Significant interaction was identified by application of 60g *Trichoderma* at two and three months after planting. Both treatment combinations had the same significant value based on percent infected roots and percent disease control.

Field experiments showed no interaction between inoculum level of *L. theobromae*, different inoculum densities of *Trichoderma* FI7c, and time of application. When test of means of the two levels of *L. theobromae* used were compared, there was a significant difference observed in percent infected roots and percent disease control between 50 and 100 g of *L. theobromae* cultured in 1:3 ratio of empty rice grain the greater the percent infected roots and the lower the percent disease control.

Furthermore, regardless of inoculum density of *Trichoderma* FI7c (50, 75, and 100 g) root infection was reduced during storage when applied two or three months after planting than when applied at planting to one month after planting. The higher percentage infection observed in stored roots from plants applied earlier with the antagonist was attributed to its limited period of exposure. The effect of different inoculum levels of *Trichoderma* FI7c isolate on percent infection of roots and disease control indicates lowest percent root infection and highest disease control in 100 g followed by 75 and 50 g. One hundred-gram antagonist inoculum was better compared to the effect of Benlate that demonstrated that the higher inoculum density of the antagonist was more effective in relation to percent infected roots and disease control at harvest up to storage.

Economic analysis showed the possibility of using *Trichoderma* FI7c as biological control agent against sweetpotato root rot. Application of *Trichoderma* FI7c would be advantageous if done at the later part of the sweetpotato plants' vegetative stage to ensure higher antagonistic activity especially. Results of the study indicate that the organic substrate supports microbial activity only for a limited period.

The results of the study provide an opportunity for biological control and could be used instead of chemicals because the antagonist is soilborne and at present already in the field. The antagonist has no toxic effect on root crops and people. However, further study is highly recommended with emphasis on the frequency of *Trichoderma* FI7c inoculation to increase its antagonistic activity and to develop a practical method for on-farm use of the antagonists.

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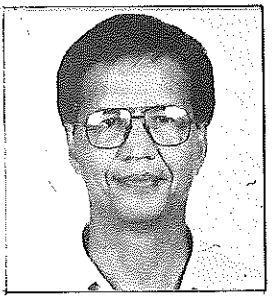

The senior author obtained his Doctor of Philosophy (PhD) degree in the University of Nebraska – Lincoln (USA) with major/minor in Plant Pathology (Biochemistry). His work experience includes stints as researcher at the International Rice Research Institute and as research associate and later visiting professor at the University of Nebraska.

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