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Study of shelf life of *Trichoderma viride* on different carrier materials by using coconut husk, groundnut shell, charcoal powder

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Abstract

Trichoderma viride widely used as a biofertilizer and as a bio-controlling agent for management of soil borne plant pathogen. Though *Trichoderma viride* has very good potential in the management of disease it could not be used as a spore suspension under field condition. Thus culture of *T. viride* should be immobilised in certain carrier material and should be prepared as formulation for easy application, storage, commercialization and field use.

In this present work different carrier materials like coconut husk, groundnut shell, charcoal powder of different pore size were used to store and keep viable the culture of *T. viride*. The carriers which are easily available, free of cost and not harmful to the environment were selected for experiment. Potato dextrose broth formulation was used and the population of *T. viride* assessed for 120 days after 30 days of interval. Charcoal powder of 2 mm pore size was found to be the best carrier material as it shows maximum number of viable *T. viride* after 120 days (58×10^8 cfu g⁻¹) compared to other carrier materials.

Keywords: *Trichoderma viride*, carriers, shelf life

Introduction

In a challenging climate there is growing responsibility to prevent food crops from disease as the human population is increasing continuously. The plant debris generally falls into the field and is decomposed in the soil. Several soil fungi like *Alternaria*, *Helminthosporium*, *Pythium* often survive in soil humus as saprophyte and in favourable conditions of climate cause a disease like root rot, damping off, wilt, blight etc. *Trichoderma* species are friendly and soil inhabiting fungi act as antagonistic to several plant pathogens. *Trichoderma* species are rarely found in living plants so they are commercially utilised for biological treatment of fungus-induced plant disease [11].

Trichoderma species are a filamentous saprophytic fungus which reproduce asexually by conidia and chlamydo-spores and sexually by ascospore. They use carbon and nitrogen compounds as a source for their growth. With that few amounts of salt like magnesium, iron is also important for the growth of *Trichoderma* species. They have discovered the ways or mechanisms to attack fungi and to help the plant in growth [8, 3, 14].

Trichoderma positively influences plant growth and its development. They have capability to produce antibiotics, to parasitize the other fungi, and to kill the detrimental pathogenic micro-organism and also enhance nutrient uptake in plants by increasing root hair growth in plants [2, 3]. Several economically important plant diseases of a variety of crops are now effectively controlled by using *Trichoderma* based fungicide; there are reports that *Trichoderma* can also be used against plant parasitic nematodes in soil [7, 16].

At present it is not very necessary to depend on poisonous chemicals. The fungicide, for the management of plant disease of economically important crops instead *Trichoderma viride* can be successfully employed for this purpose.

Hence this experiment was conducted to check the viability of *Trichoderma viride* on 3 different carrier materials by using 2 different pore sizes.

Materials and Methods

Material

- a) **Fungal culture:** *Trichoderma viride*.
- b) **Carrier material:** 1) Coconut husk, 2) Charcoal powder, 3) Groundnut shell.

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Table 1: Shows *Trichoderma viride* as a potential biocontrol agent (4)

<i>Trichoderma</i> species	Mode of application	Crop	Pathogen	Effect	Reference
<i>T. viride</i>	Culture applied on seeds.	Groundnut	<i>Sclerotium rolfsii</i>	Reduce disease	Manjula <i>et al.</i> ; 2004 [6]
<i>T. viride</i>	Combine with talc material	Vanilla	<i>Fusarium oxysporum</i> Phytophthora meadii	Reduce disease	Radjaccomare <i>et al.</i> ; 2004 [9]
<i>T. viride</i>	Culture applied on	Potato,	<i>R. solani</i>	Reduce disease	Somani and Arora 2010 [15]
<i>T. viride</i>	Combine with talc material	Nut	<i>Lasiodiplodia theobromae</i>	Reduce disease	Latha <i>et al.</i> ; 2011 [5]

Method [10]

1. Coconut husk, charcoal and shell of groundnut were evaluated as carrier material.
2. The carrier material were dried in sun to remove total moisture and powdered and sieve through sieve pore of 2 mm and 0.5 mm
3. This carrier material is sterilised in an autoclave for 30 minutes.
4. And this sterilised carrier material is mixed with the culture of *T. viride* by 2:1 proportion (carrier material 2: liquid culture 1).
5. *T. viride* used were previously cultured using potato dextrose broth.
6. 50 g of this mixture is filled in a polypropylene bag tied and kept at 25-30 °C.
7. Observations on cfu g-1 were made after each 30 days of interval for 4 months and shelf life was carried out.

The primary focus of bio-control research is the direct coating of seeds with *Trichoderma* spores. When research discoveries are applied from the lab to the field, only then will

the technology be beneficial. Despite having excellent potential for disease management, *Trichoderma* cannot be used as a spore suspension in the field. As a result, the *Trichoderma* culture should be immobilised in a specific carrier material and prepared as a formulation for simple application, storage, commercialization, and field use.

Characteristic of ideal formulation [12]

1. Should have a longer shelf life.
2. Should not be inhibitory to growth of the crops plant and poisonous to them.
3. Should withstand unfavourable environmental conditions.
4. Should be affordable and should give effective control of plant diseases.
5. Carrier material must be cheap and readily available for formulation'.
6. Should be compatible with chemicals used in agriculture.
7. Should be water soluble.

Observation table**Table 2:** Viable count of colonies

Carrier material	Pore size (0.5 mm)	Viable count of colonies of <i>T. viride</i> x 10 ⁶ cfu g-1)				
		Initial	After 30 days	After 60 days	After 90 days	After 120 days
Charcoal	0.5 mm	152.2	134.6	94.2	71.9	55.9
Coconut husk	0.5 mm	153.6	127.2	81.0	65.3	49.7
Groundnut shell	0.5 mm	152.8	121.6	72.1	58.1	38.5

Table 3: Viable count of colonies of *T. viride*

Carrier material	Pore size (2 mm)	Viable count of colonies of <i>T. viride</i> x 10 ⁶ cfu g-1)				
		Initial	After 30 days	After 60 days	After 90 days	After 120 days
Charcoal	2 mm	154.2	139.6	96.8	81.8	58.6
Coconut husk	2 mm	152.8	125.4	84.2	70.9	51.2
Groundnut shell	2 mm	154.5	110.1	75.7	61.6	46.2

Results and Discussion

Data represented in table 2.1 and table 2.2 shows the variation in the viability of *T. viride* on different carrier materials. The viability of spores is more in the 2 mm pore size material compared to 0.5 mm pore size of the carrier in all of the carrier material. Groundnut shell shows less viability on pore size 0.5 mm which is 38.5 x 10⁸ cfu g-1 and on 2mm pore size it is 46.2 x 10⁸ cfu g-1. Charcoal powder was found to be the best carrier material. It shows the viability of spores is 58.5 x 10⁸ cfu g-1 for 2 mm pore size and 55.9 x 10⁸ cfu g-1 for 2 mm pore size respectively. The coconut husk shows the viability in between the other two carrier materials

All the carrier material shows viability of *Trichoderma viride* spores. With the increasing months the viability goes on decreases but still it shows a good quantity of viability after 4 months so that the formulation can be stored and can be used in the field.

Conclusion

Dried powder of groundnut shell, coconut husk, charcoal powder all can work as carries material for *Trichoderma* species culture. Charcoal is the best out of 3 having the highest viability of *Trichoderma viride* spores. Thus, this formulation can be used by farmers and horticulturists as a bio-controlling agent to prevent common plant parasitic disease.

Method of application [1]

- a) **Seed treatment:** Coating of the seed by *Trichoderma* formulation just before sowing.
- b) **Seed biopriming:** Coating the seed by *Trichoderma* and incubating under moist and warm temperature before radical growth.
- c) **Soil treatment:** Adding *Trichoderma* formulation directly to the soil.
- d) **Aerial spraying:** *Trichoderma* applied to the plant's part like stem, leaves, flower, fruit etc.

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