

ANTAGONISTIC POTENTIALS OF SOME SOIL FUNGI AGAINST THREE POST-HARVEST PATHOGENIC FUNGI OF *CARICA PAPAYA* L.

REZUANA BINTE HELAL¹ AND SHAMIM SHAMSI*

Department of Botany, University of Dhaka, Dhaka-1000, Bangladesh

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Abstract

Four antagonistic fungi were isolated from the papaya field soil by serial dilution and were identified as *Aspergillus flavus* Link., *A. fumigatus* Fresenius, *A. niger* van Tiegh. and *Trichoderma viride* Pers. The soil fungi were selected to evaluate their antagonistic potentiality against the three postharvest pathogenic fungi of *Carica papaya* L. following "dual culture colony interaction" and volatile and non-volatile metabolites. In dual culture colony interaction, out of four soil fungi, *T. viride* showed the highest growth inhibition on *C. gloeosporioides* (84.28%), *F. nivale* (95.23%) and *Fusarium* sp. (87.15%). On the other hand *A. niger* showed the highest growth inhibition on *C. gloeosporioides* (77.39%), *F. nivale* (98.63%) and *Fusarium* sp. (35.05%). The maximum inhibition of radial growth of *C. gloeosporioides* (77.64%), *F. nivale* (58.76%) and *Fusarium* sp. (79.37%) were observed in case of *T. viride* owing to the volatile metabolites. Whereas the maximum inhibition of radial growth of *C. gloeosporioides* (58.23%), *F. nivale* (37.43%) and *Fusarium* sp. (82.31%) were observed in case of *A. niger* owing to the volatile metabolites. The maximum inhibition of radial growth of *C. gloeosporioides* (90.90%), *F. nivale* (89.13%) and *Fusarium* sp. (76.84%) were observed in case of *T. viride* owing to the effect of non-volatile metabolites. The maximum inhibition of radial growth of *C. gloeosporioides* (92.42%), *F. nivale* (73.01%) and *Fusarium* sp. (68.67%) were observed in case of *A. niger* owing to the effect of non-volatile metabolites. *Trichoderma viride* and *A. niger* may be exploited commercially as a biocontrol agent against anthracnose and fruit rot pathogens of papaya.

Introduction

Papaya (*Carica papaya* L.) is one of the most popular fruits of the world belongs to the family "Caricaceae"⁽¹⁾. In Bangladesh, total estimated production of papaya is 133370 million tons and cultivated area is 3126 acres⁽²⁾. Papayas are attacked by a number of pathogens from bloom to harvest and in storage which cause considerably deteriorate the fruit quality⁽³⁾. It is estimated that about 20 - 25% of the harvested papaya fruits are decayed by pathogens during postharvest handling even in developed countries⁽⁴⁻⁵⁾. Approximately 39.9% post-harvest losses of papaya fruits were estimated in Bangladesh and the value stands for that is Tk. 132.91 crore⁽⁶⁾.

*Author for correspondence: <prof.shamsi@gmail.com>. ¹A part of MS thesis of first author (RBH).

Over the past few decades, farmers have increasingly relied on chemical pesticides for protecting plants against pathogens. However, the increasing use of chemical pesticides negatively affects the environment and human health. Biological control of plant diseases including fungal pathogens has been considered a viable alternative method to chemical control. Biological control presents a better alternative with relative low cost, without any side effects⁽⁷⁻⁸⁾.

Study of antagonist as biological control agent has now become one of the most exciting and rapidly developing areas in plant pathology because it has great potential to solve many agricultural and environmental problems. At present, *Trichoderma*-based products are considered as relatively novel biological control agents which can help farmers to reduce plant diseases and increase plant growth⁽⁹⁾.

In Bangladesh, research on biological control of postharvest diseases of papaya fruits is very limited and considering the importance of this popular fruits present investigation was undertaken to find out the biological efficacy as antagonist of the selected soil fungi against the pathogens of postharvest diseases of papaya.

Materials and Methods

Nineteen species of fungi were isolated from infected Shahi papaya during April to November, 2016. Among the isolated fungi three were found to be pathogenic to papaya fruits. The pathogenic fungi were *C. gloeosporioides*, *F. nivale* and *Fusarium* sp. These three fungi were selected as test pathogen against selected four antagonistic fungi.

Serial dilution method was used to isolate antagonistic fungi from rhizosphere soil of the host varieties. Among the isolated soil fungi, *Aspergillus flavus*, *A. fumigatus*, *A. niger* and *Trichoderma viride* were selected to test their antagonistic potential against the pathogens following dual culture technique⁽¹⁰⁾. Aforesaid fungi were previously tested on fungal isolates obtained from *Musa* sp. and found effective against the test fungi. The parameter used for the assessment of the colony interaction and per cent inhibition of radial growth was calculated⁽¹¹⁾. Effects of volatile and non-volatile metabolites of the selected soil fungi against the test pathogens were also studied⁽¹²⁾. The results were statistically analyzed by t test⁽¹³⁾.

Results and Discussion

The results of colony interactions have been summarized in Table 1 and Fig. 1. In this study, antagonistic relationships (Grade) among the soil fungi and test pathogens were 2 and 4. However, grade 2 was found to be the most commonly encountered type of colony interaction as 10 interactions were incorporated in this grade which was followed by grade 4 (2 out of 12) (Table 1).

In dual culture colony interaction *Trichoderma viride* showed the highest growth inhibition on *Colletotrichum gloeosporioides* (84.28%) which was followed by *Aspergillus niger* (77.39%), *A. flavus* (43.71%) and *A. fumigatus* (29.32%) (Fig. 1). *Aspergillus niger* (98.63%) showed the highest growth inhibition on *Fusarium nivale* which was followed by *Trichoderma viride* (95.23%), *A. flavus* (74.70%), and *A. fumigatus* (32.94%). *Trichoderma viride* showed the highest (87.15%) growth inhibition on *Fusarium* sp. which was followed by *A. fumigatus* (55.27%), *A. flavus* (48.66%) and *A. niger* (35.05%).

Table 1. Grades of colony interaction between the test pathogens and antagonists.

Name of antagonists	Name of test pathogens		
	<i>Colletotrichum gloeosporioides</i>	<i>Fusarium nivale</i>	<i>Fusarium</i> sp.
<i>Aspergillus flavus</i>	2	2	2
<i>A. fumigatus</i>	4	2	2
<i>A. niger</i>	2	2	2
<i>Trichoderma viride</i>	2	2	4

Grade 2 = Mutual intermingling growth where the growth of the fungus is ceased and being over growth by the opposed fungus. Grade 4 = Slight inhibition of both the interacting fungi with narrow demarcation line (1 - 2 mm) based on Skidmore and Dickinson (1976)⁽¹¹⁾.

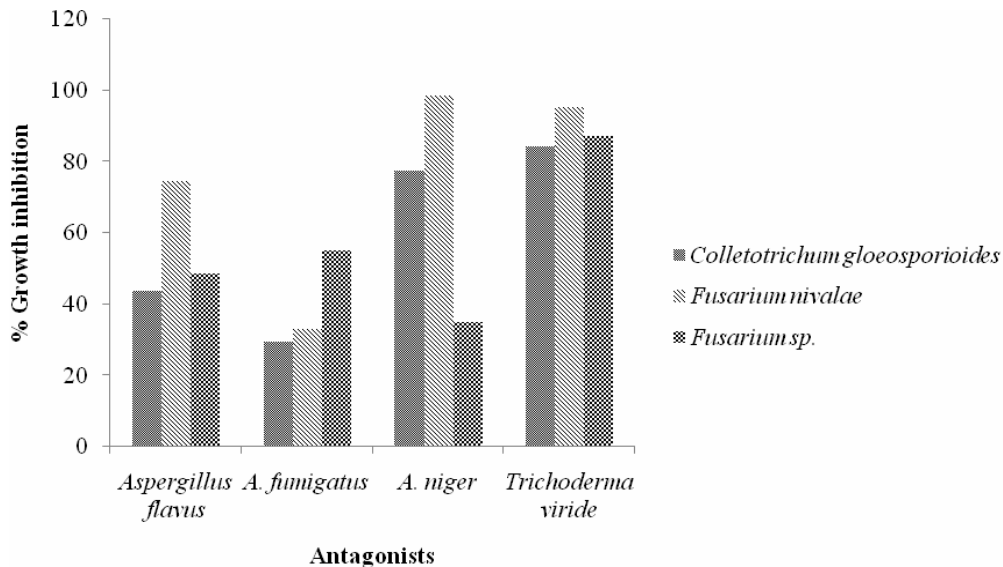


Fig.1. Per cent inhibition of *Colletotrichum gloeosporioides*, *Fusarium nivale* and *Fusarium* sp. owing to fungal antagonists (in dual culture).

In contrast to the present study, Akter *et al.*⁽¹⁴⁾ reported that in dual culture colony interaction *Aspergillus niger*, *Trichoderma viride*, *A. flavus* and *A. fumigatus* showed 68.66,

57.24, 54.19 and 50.25% growth inhibition on *Colletotrichum* sp., respectively. Again *Aspergillus niger*, *Trichoderma viride*, *A. flavus* and *A. fumigatus* showed 75.87, 75.5, 51.78 and 45.52% growth inhibition on *Curvularia lunata*, respectively. Further *Trichoderma viride*, *Aspergillus niger*, *A. flavus* and *A. fumigatus* showed 56.52, 50.70, 47.36 and 46.15% growth inhibition on *Fusarium semitectum*, respectively. Bashar and Chakma⁽¹⁵⁾ reported that in dual culture colony interaction *A. niger*, *T. viride*, *A. flavus* and *A. fumigatus* showed 65.21, 64.24, 57.14 and 34.78% growth inhibition on *F. oxysporum*, respectively. Tapwal *et al* ⁽¹⁶⁾ reported that in dual culture colony interaction *T. viride* showed 12.50% growth inhibition on *C. gloeosporioides*. This variation might be due to selection of different test pathogens. In dual culture technique, significantly maximum inhibition was recorded in *T. viride* (66.40%) according to Patel and Joshi⁽¹⁷⁾.

The results of effect of volatile metabolites of antagonistic fungi against papaya pathogens are presented in Table 2. The maximum inhibition of radial growth of *Colletotrichum gloeosporioides* was observed in *Trichoderma viride* (77.64%) which was followed by *Aspergillus flavus* (75.58%), *A. fumigatus* (60.88%) and *A. niger* (58.23%) due to the volatile metabolites after 6 days of incubation at 25 ± 2°C. The maximum inhibition of radial growth of *Fusarium nivale* was also observed in *Trichoderma viride* (58.76%) followed by *Aspergillus fumigatus* (48.34%), *A. niger* (37.43%) and *A. flavus* (34.40%) owing to the volatile metabolites after 6 days of incubation at the same temperature. The maximum inhibition of radial growth of *Fusarium* sp. was observed in *Aspergillus niger* (82.31%) which was followed by *Trichoderma viride* (79.37%), *A. flavus* (46.56%) and *A. fumigatus* (34.37%) owing to the volatile metabolites after 6 days of incubation at 25 ± 2°C.

Table 2. Per cent inhibition of radial growth of the test pathogens by volatile metabolites of antagonistic fungi.

Name of antagonist	% inhibition of radial growth of the test pathogens		
	<i>Colletotrichum gloeosporioides</i> .	<i>Fusarium nivale</i>	<i>Fusarium</i> sp.
<i>Aspergillus flavus</i>	75.58 ^a	34.40 ^c	46.56 ^b
<i>A. fumigatus</i>	60.88 ^b	48.34 ^c	34.37 ^c
<i>A. niger</i>	58.23 ^b	37.43 ^c	82.31 ^a
<i>Trichoderma viride</i>	77.64 ^a	58.76 ^b	79.37 ^a

a and b indicate significance of 't' value at p = 0.001 and 0.01, respectively.

In contrast to the present study, Aktar *et al.*⁽¹⁴⁾ reported that volatile metabolites produced by an isolate of *Aspergillus niger*, *A. flavus*, *A. fumigatus* and *Trichoderma viride* inhibited the mycelial growth of *Colletotrichum* sp. by 14.68, 11.78, 11 and 11%, respectively. Further the volatile metabolites produced by an isolate of *Trichoderma viride*, *Aspergillus niger*, *A. flavus* and *A. fumigatus* inhibited the mycelial growth of *Fusarium*

semitectum by 13.5, 9.5, 8 and 7.75%, respectively. Differences in per cent inhibition with the present study might be due to the difference in organism involved in the interaction. Bashar and Chakma⁽¹⁵⁾ reported that volatile substances produced by *T. viride*, *A. niger*, *A. flavus* and *A. fumigatus* showed 29.75, 20.15, 15.78 and 12.25% growth inhibition on *F. oxysporum*, respectively. Thakur and Harsh⁽¹⁸⁾ reported that volatile metabolites produced from the culture of *Aspergillus niger* showed 42.43% inhibition of mycelial growth of *C. gloeosporioides*.

Table 3 shows the effect of non-volatile metabolites on the growth of *Colletotrichum gloeosporioides*, *Fusarium nivale* and *Fusarium* sp. The maximum inhibition of radial growth of *Colletotrichum gloeosporioides* was observed with the culture filtrates of *A. flavus* (92.42%), which was followed by *T. viride* (90.90%), *A. niger* (86.13%) and *A. fumigatus*

Table 3. Per cent inhibition of radial growth of test pathogens by non-volatile metabolites of antagonistic fungi.

Name of fungi	Concentration (%)	% inhibition of radial growth of test pathogens by non-volatile metabolites owing to different antagonists			
		<i>Aspergillus flavus</i>	<i>A. fumigatus</i>	<i>A. niger</i>	<i>Trichoderma viride</i>
<i>Colletotrichum gloeosporioides</i>	5	60.20 ^a	49.49 ^a	62.37 ^a	67.81 ^a
	10	70.72 ^a	59.59 ^a	74.25 ^a	74.09 ^a
	15	85.14 ^a	69.69 ^a	78.20 ^a	82.18 ^a
	20	92.42 ^a	73.73 ^a	86.13 ^a	90.90 ^a
<i>Fusarium nivale</i>	5	49.25 ^b	40.25 ^b	53.19 ^b	58.69 ^b
	10	52.22 ^a	44.15 ^b	66.38 ^b	78.26 ^a
	15	67.61 ^a	64.93 ^a	73.19 ^a	80.43 ^a
	20	73.01 ^a	71.42 ^a	77.65 ^a	89.13 ^a
<i>Fusarium</i> sp.	5	51.80 ^a	42.85 ^b	33.82 ^b	58.77 ^b
	10	59.03 ^a	50.01 ^a	35.29 ^b	72.00 ^a
	15	61.44 ^a	61.90 ^a	64.70 ^a	72.63 ^a
	20	68.67 ^a	66.66 ^a	70.78 ^a	76.84 ^a

^a and ^b indicate significance of 't' value at p = 0.001 and 0.01, respectively.

(73.73%) at 20% concentration. The maximum inhibition of radial growth of *Fusarium nivale* was observed with the culture filtrates of *Trichoderma viride* (89.13%) which was followed by *A. niger* (77.64%), *A. flavus* (73.01%), and *A. fumigatus* (71.42%) at 20% concentration. The maximum inhibition of radial growth of *Fusarium* sp. was observed with the culture filtrates of *T. viride* (76.84%) which was followed by *A. niger* (70.58%), *A. flavus* (68.67%) and *A. fumigatus* (66.66%) at 20% concentration.

In contrast to the present study, Aktar *et al.*⁽¹⁴⁾ reported that non-volatile metabolites produced by an isolate of *Aspergillus niger*, *Trichoderma viride*, *A. flavus* and *A. fumigatus* inhibited mycelial growth of *Colletotrichum* sp. by 52.56, 44.72, 40 and 37.2%, respectively. Further, the non-volatile metabolites produced by an isolate of *Trichoderma viride*, *Aspergillus niger*, *A. flavus* and *A. fumigatus* inhibited mycelial growth of *Fusarium semitectum* by 50, 45, 8 and 7.75%, respectively. Differences in per cent inhibition with the present study might be due to the difference in organism strain involved in the interaction. Bashar and Chakma⁽¹⁵⁾ reported that culture filtrates of *T. viride*, *A. fumigatus*, *A. niger* and *A. flavus* showed 82.05, 80.56, 72.22 and 66.66% growth inhibition of *F. oxysporum* at 20% concentration owing to non-volatile metabolites. Madhanraj *et al.*⁽¹⁹⁾ reported that culture filtrates of *T. viride* and *A. niger* inhibited the mycelial growth of *F. solani* by 85 and 70% at 20 per cent concentration, respectively. Tran⁽⁹⁾ used *T. viride* to control *S. rolfsii* and found effective result. Tapwal *et al.*⁽¹⁶⁾ reported that culture filtrates of *T. viride* showed 13.33% growth inhibition on *C. gloeosporioides*.

The present investigation suggests that *Trichoderma viride* and *Aspergillus niger* may be exploited commercially as a biocontrol agent to protect postharvest decay of papaya fruits belongs to *Carica papaya*.

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