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Management of Banana Anthracnose Caused by *Colletotrichum musae* with Commercial Botanical-Formulations

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ABSTRACT

Plant based pesticides which are relatively economical, safe and non hazardous can be used successfully against the pathogenic fungi. The present investigations were carried out during 2015-16 in department of plant pathology, UAS, Dharwad which is aimed to study the efficacy of five commercially available botanical formulations viz; Soldier, nimbicidine, Neemgold, agroneem and Discheck for the management of banana anthracnose, a predominant postharvest disease of banana caused by Colletotrichum musae. Among those botanicals, Neemgold and Discheck were most effective in the in vitro as well as in vivo studies. Maximum per cent disease reduction (92.11 %) has been exhibited in fruits treated with Neemgold, Discheck and nimbicidin at 2.0 per cent concentration which were statistically at par with same botanicals at 1.0 per cent (89.47 %) as well as Neemgold at 0.5 per cent (86.84 %). Therefore, commercial botanicals formulations have the potential to manage the postharvest diseases in banana.

Key words: *Banana anthracnose, Botanical formulations, Colletotrichum musae, Disease management*

INTRODUCTION

Anthracnose of banana is caused by *Colletotrichum musae* is most important postharvest disease, it has been a major constraint in the marketing of fruit intended for local as well as distant markets. Control of postharvest diseases has been traditionally achieved by pre and postharvest applications of fungicides. However, in the recent years there has been major thrust on residue free, organic fruit production. In view of this, different eco-friendly approaches need to be explored for their effectiveness and to fit-in with the management schedule of the diseases suitably such as use of botanicals, plant oils and other natural products. Natural plants products and their analogues are an important source of agricultural pesticides[3]. These are used in the control of plant diseases as well[1]. Recently the exploitation of natural products to control postharvest decay and to enhance shelf life of fruits had received significant attention.

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Contrary to the problems associated with the use of synthetic chemicals, botanicals are environmentally non pollutive, non-phytotoxic, systemic ephemeral, readily biodegradable, relatively cost effective and hence constitute a suitable plant protection in the strategy of biological management of diseases. Hence, screening of plant products for its effective antifungal activity against the pathogen is essentially required to minimize the use of fungicides and to consider as one of the components in the integrated disease management. Neem based commercial products *viz*., Neemgold, achook, biotas, tricure and neemazal have shown their effectiveness against *C. capsici*[12]. With this background, present investigation was undertaken to study the efficacy of commercially available botanicals for the management of anthracnose disease, a predominant postharvest disease of banana.

MATERIAL AND METHODS

Present experiments were carried out during 2015-16 in Department of Plant Pathology, College of Agriculture Sciences, UAS, Dharwad. Five commercially available botanical formulations *viz;* Soldier (*Aegle marmelos* (20%)+ *Ricinus communis* (20%)+ *Hygrophila spinosa* (20%)+ *Laminaria* sp. (20%) + *Lantana camara* (20%), Nimbicidine (Azadirachtin 0.03%), Neemgold (Azadirachtin 1.00%), Agroneem (Neem oil based herbal product) and Discheck (*Ficus bengalensis* 0.0001% +*Ficus religiosa* 0.0001% +*Ficus retusa* 0.0001% +Aqua solvent 99.99%) have been evaluated for their efficacy in managing banana anthracnose disease.

Inhibition of mycelial growth: To study the antifungal effect of botanicals, poisoned food technique was employed[6]. Poisoned medium was prepared for the test pathogen to get 0.5, 1.0 and 2.0 per cent concentration; mycelium of five mm size plugs was placed on the centre of each potato dextrose agar plate topsy-turvy. Controls were maintained by growing the pathogen on PDA plates. Incubated plates were incubated at 27±1°C temperature. The efficacy of botanical pesticides was expressed as per cent inhibition of redial growth over the control which was calculated by using the formula suggested by Vincent[13].

Inhibition of spore germination: Same botanicals were evaluated for inhibition of spore germination of *C. musae.* Conidial suspension of *C. musae* was prepared separately in a test tube containing sterile water and stirred well for uniformity in spore spread. Botanicals were diluted in sterile water to get the required concentrations, both spore suspension and botanicals were mixed together in a cavity slide in equal proportions to achieve concentration of requirement in the assay. Slides were incubated under room temperature for 24 hr in moist chambers. The observations on number of spores germinated and total number of spores in each microscopic field was recorded. Sterile water served as control. Percent inhibition of spore germination was calculated by formula given by Vincent[13] and data was later analysed statistically.

***In vivo* evaluation of botanicals:** Postharvest dipping solutions were prepared in sterilized water to achieve required concentrations (0.5, 1.0 and 2.0 % v/v). Fruits selected for *in vivo* studies were at mature green stage, free from blemishes and injuries. Banana fingers with intact crown portion were sterilized in 1.0 per cent sodium hypochlorite and rinsed with sterilized distilled water. These fingers were dipped in postharvest treatment solutions of required concentration which were for ten min and allowed to dry in aseptic chamber. These fruits were dipped in spore suspention of *C. musae* ($1x10^5$) for one min and allowed to dry aseptically. The fingers dipped in sterilized water for ten min followed by spore suspension of *C. musae* ($1x10^5$) served as untreated control. Cotton swabs dipped in sterile water were placed in moist chamber to maintain sufficient humidity during incubation. Observations were recorded on per cent area infected on fruit in 0-5 scale i.e. [0]- No symptoms on fruit surface, [1]- 0.1 – 5 %

area covered by lesions, [2]- 5.1 – 10 % area covered by lesions, [3]- 10.1 – 25 % area covered by lesions, [4]- 25.1 – 50 % area covered by lesions and [5]- 50 % of area covered by lesions[8] (Prasannakumar, 2001) after eight days of inoculation. Per cent disease index was computed by using the formula given by Wheeler[14] and analysed statistically. Per cent disease reduction in different treatments over control was derived by using the formula suggested by Vincent[13].

Statistical analysis: The experiment was laid out in a completely randomized design (CRD) in three replications. The experiment was conducted at room temperature in the laboratory. Statistical analysis was done as per the procedures given by Gomez and Gomez[5]. All comparisons of means were subjected to analysis of variance (ANOVA) and the significant differences among treatments were determined with a least significant difference (lsd) separation test. Arcsine or square root transformation was used wherever required to normalize variance.

RESULTS AND DISCUSSION

***In vitro* evaluation of botanicals:** Results of *In vitro* evaluation of botanicals against *C. musae* are presented in Table 1. Neemgold at 2.0 per cent concentration was very effective in arresting the mycelial growth of *C. musae* (80.38%) which was significantly superior over all other treatments. Next best was Dischecк at 2.0 per cent concentration (75.11%) which was statistically at par with nimbicidin at 2.0 per cent concentration (74.37%), Dischecк (72.11%) and Neemgold (71.92%). Neemgold was consistently showed inhibition of pathogen at all the concentrations tested.

The same botanicals tested for inhibition of spore germination of *C. musae* varied in their efficacy. Dischecк at 2.0 per cent concentration resulted in 76.70 per cent inhibition of spore germination which was statistically at par with Neemgold at 2.0 per cent concentration (73.27%) and superior to all others. Next best was Neemgold at 1.0 per cent which showed 67.95 per cent inhibition. At 0.5 per cent concentration, Neemgold (57.30%) was effective over other botanicals at same concentration (Table 2). Efficacy of these botanicals may be attributed to antifungal activity of azadirechtin, the active ingredient of Neemgold which was well documented. These results are in accordance with findings of Padder *et al*[7]., who reported the efficacy of Neemgold against *Colletotrichum lindemuthianum* (Sacc. & Magnus) Briosi and Cavara. Similarly, Tiwari *et al*[12]., reported that neemazal, tricure and Neemgold inhibited the growth and sporulation of *Colletotrichum capsici* (Sydow) Butler and Bisby.

***In vivo* evaluation of botanicals:** The efficacy of botanicals in terms of disease reduction of banana anthracnose showed that (Table 3), maximum disease reduction (92.11%) was recorded in Neemgold and Dischecк at 2.0 per cent concentration which were found at par with nimbicidin (89.47%) at same concentration. It was evident from the data shown that Neemgold and Dischecк were consistently showing disease reduction in all concentrations tested. Mode of action of these natural products was not investigated in this study. However, according to Subramanian[10] the fungicidal spectrum of *Azadirachta indica* A. Juss has been attributed to the presence of azadirachtin which belongs to C_{25} terpeniodes. Meanwhile, Srivastava *et al*[9]., from their investigations concluded that, the inhibitory action of natural products on fungal cells involves cytoplasm granulation, cytoplasmic membrane rupture and inactivation and/or inhibition of synthesis of intracellular enzymes. Results of this study are supported by findings of Thangavelu *et al*[11]., and Dey *et al*[4]. In another study, Bazie *et al*[2]., gave conclusive evidence that postharvest treatment of banana fruits with 20 per cent extracts of *Acacia albida* (Del.) A. Chev. and *Prosopis juliflora* (Swartz) De Candolle had reduced anthracnose incidence and severity on banana fruits

Table 1: *In vitro* evaluation of botanicals on mycelial growth of *Colletotrichum musae*

Botanical	Per cent inhibition of mycelial growth			Mean
	At concentrations (%)			
	0.5	1.0	2.0	
Discheck	31.86 (34.36)*	72.11(58.12)	75.11 (60.07)	59.69 (50.59)
Multineem	32.97(35.04)	64.21 (53.25)	67.25 (55.09)	54.81 (47.76)
Neemgold	63.02 (52.55)	71.92 (58.00)	80.38 (63.71)	71.77 (57.91)
Nimbicidin	56.49 (48.73)	58.64 (48.98)	74.37 (59.58)	63.12 (52.60)
Soldier	15.50 (23.19)	56.45 (48.71)	63.13 (52.61)	45.03 (42.15)
Mean	39.97 (39.21)	64.64 (53.49)	72.05 (58.08)	
Source	**S.Em ±**		**CD at 1%**	
Treatment (T)	0.32		1.23	
Concentration (C)	0.25		0.96	
T×C	0.55		2.14	

*Arcsine transformed values

Table 2: *In vitro* evaluation of botanicals on spore germination of *Colletotrichum musae*

Botanicals	Per cent inhibition of spore germination			Mean
	At concentrations (%)			
	0.5	1.0	2.0	
Discheck	45.86 (42.62)*	56.44 (48.70)	76.70 (61.15)	59.66 (50.82)
Multineem	50.53 (45.30)	56.38 (48.67)	63.87 (53.06)	56.93 (49.01)
Neemgold	57.30 (49.20)	67.95 (55.53)	73.27 (58.88)	66.17 (54.54)
Nimbicidin	35.75 (36.72)	47.57 (43.61)	61.99 (51.94)	48.44 (44.09)
Soldier	17.89 (25.02)	22.32 (28.19)	33.57 (35.40)	24.59 (29.54)
Mean	41.47 (39.77)	50.13 (44.94)	61.88 (52.09)	
Source	**S.Em ±**		**CD at 1%**	
Treatment (T)	0.36		1.40	
Concentration (C)	0.28		1.09	
T×C	0.63		2.43	

*Arcsine transformed values

Table 3: Bioefficacy of botanicals for the management of banana anthracnose

Botanicals	Per cent disease index				Per cent disease reduction over control			
	At concentrations (%)				At concentrations (%)			
	0.5	1.0	2.0	Mean	0.5	1.0	2.0	Mean
Discheck	13.33 (21.42) *	8.89 (17.35)	6.67 (14.97)	9.63 (18.08)	84.21 (66.59)	89.47 (71.32)	92.11 (73.68)	88.60 (70.53)
Multineem	51.11 (45.64)	24.44 (29.63)	20.00 (26.57)	31.85 (34.36)	39.48 (38.19)	71.05 (57.50)	76.32 (61.09)	62.28 (52.50)
Neemgold	11.11 (19.47)	8.89 (17.35)	6.67 (14.97)	8.89 (17.35)	86.84 (68.95)	89.47 (71.32)	92.11 (73.68)	89.47 (71.32)
Nimbicidin	26.67 (31.09)	17.78 (24.94)	8.89 (17.35)	17.78 (24.94)	68.42 (55.81)	78.95 (62.78)	89.47 (71.32)	78.95 (63.30)
Soldier	75.56 (60.37)	62.22 (52.07)	35.56 (36.60)	57.78 (49.47)	10.53 (18.61)	26.32 (30.81)	57.90 (49.56)	31.58 (32.99)
Untreated control				84.45 (66.77)				
Source	**S.Em ±**		**CD at 5%**		**S.Em ±**		**CD at 5%**	
Treatment (T)	0.72		2.08		1.09		3.15	
Concentration (C)	0.93		2.69		0.84		2.44	
T×C	1.61		4.65		1.89		5.45	

*Arcsine transformed values

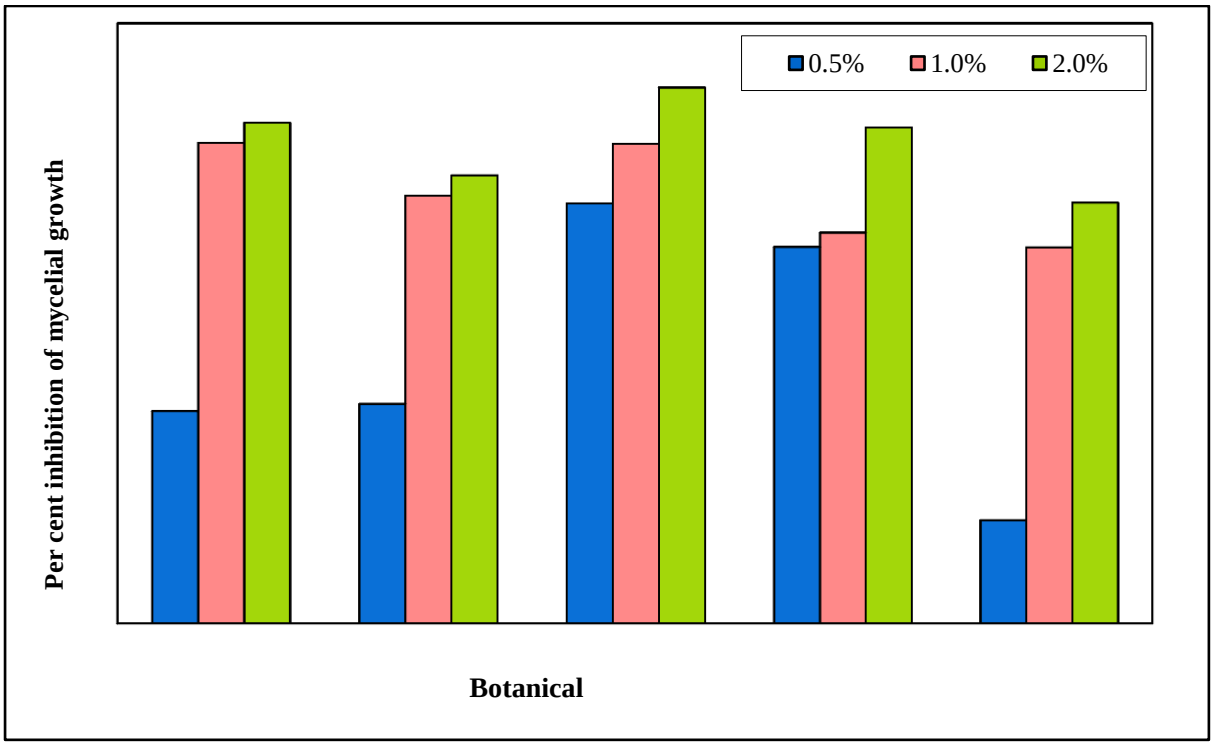

Fig. 1: *In vitro* evaluation of botanicals against *Colletotrichum musae*

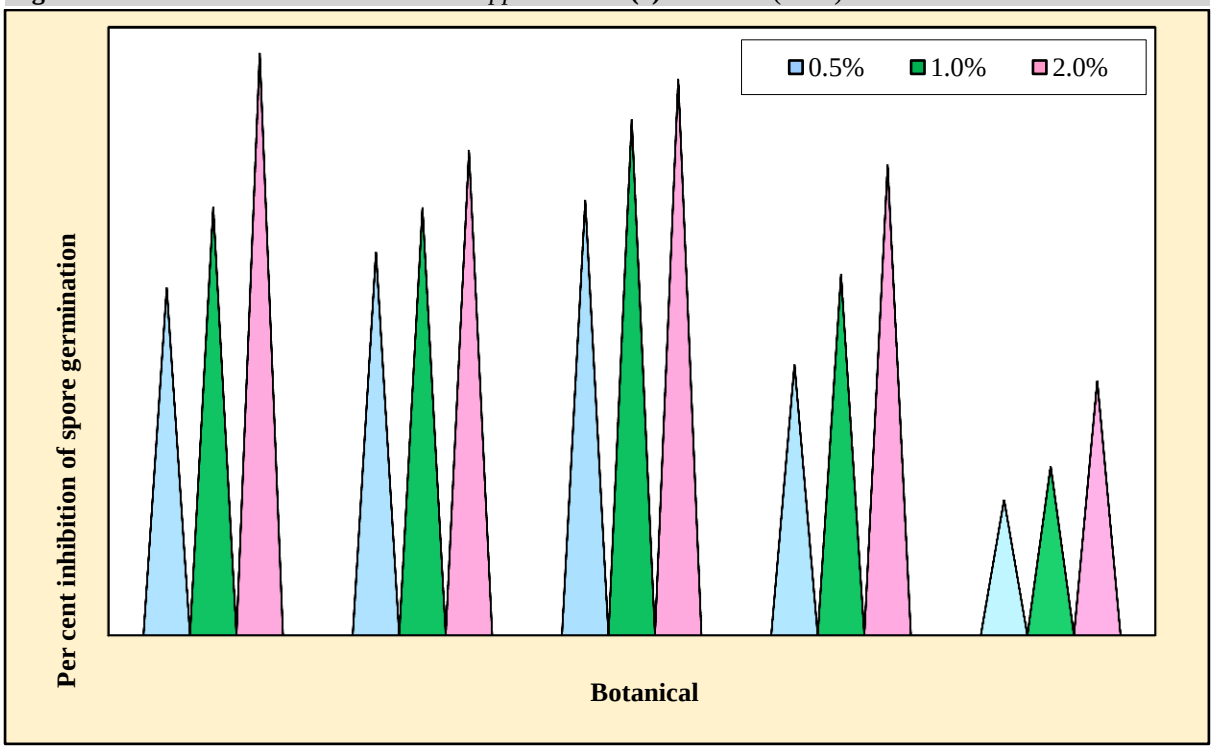

Fig. 2: Effect of botanicals on conidial germination of *Colletotrichum musae*

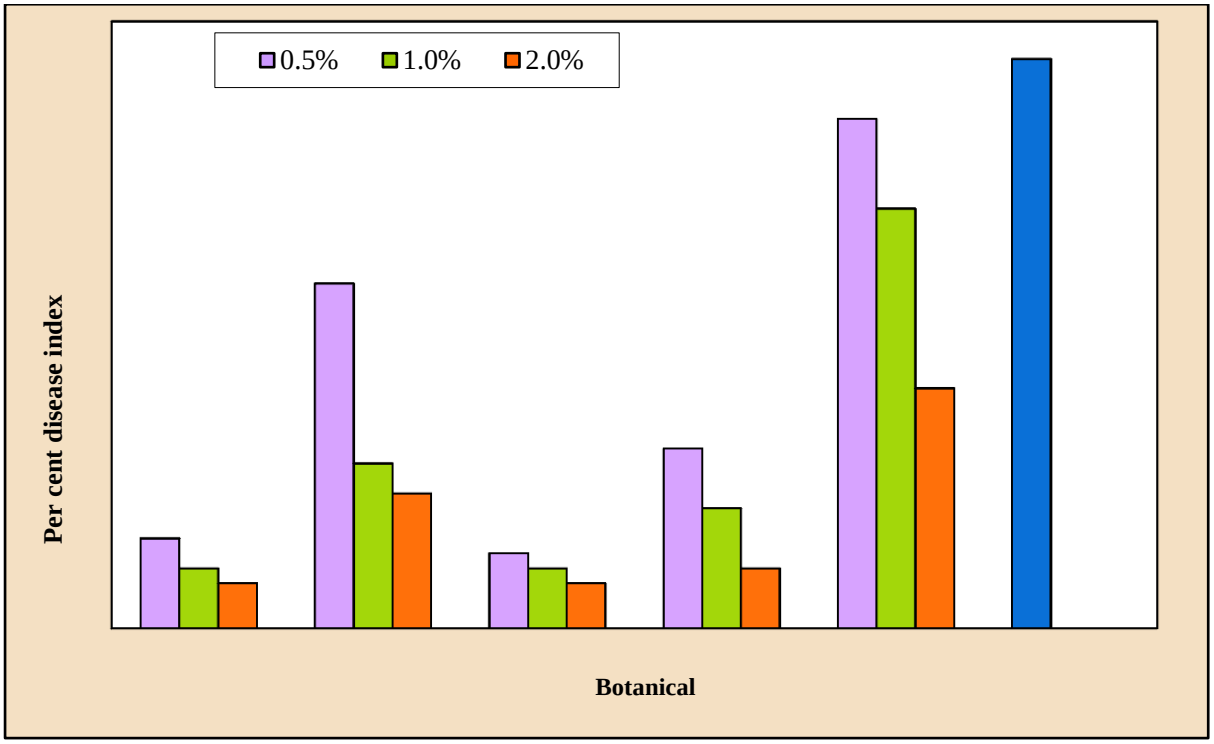

Fig. 3: Bioefficacy of botanicals for the management of banana anthracnose

a. Symptoms of banana anthracnose

b. Culture of *Colletotrichum musae*

c. Conidia of *Colletotrichum musae*

d. Fruits treated with Neemgold

e. Fruits treated with Dischеck

CONCLUSION

Among the botanicals evaluated *in vitro* and *in vivo*, Neemgold and Dischеck were most effective. Maximum disease reduction (92.11%) has been shown in fruits treated with Neemgold and Dischеck at 2.0 per cent concentration which were found at par with nimbicidin at same concentration, Neemgold and Dischеck at 1.0 per cent (89.47%) as well as Neemgold at 0.5 per cent (86.84%).

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