Evaluation of chilli (Capsicum annuum L.) genotypes, fungicides and plant products for management of anthracnose disease

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Thirty-seven genotypes were screened against Colletotrichum capsici C. gloeosporioides, the causal agents of anthracnose disease of chilli under artificial method of inoculation. The surface sterilized chilli (Capsicum annuum L.) fruits were pricked with pin bundles and dipped in spore suspension (1x107 spores/ml) of pathogen and incubated in humid chamber at 27±1°C. The infected fruits were scored using 0-4 scale. Genotypes, PC-1, Co-1, LCA-301 and IHR-3023 showed resistant reaction against both the species of Colletotrichum and H-232 showed resistance against only C. capsici. Under field condition, PC-1, LCA-301, IHR-3023, H-232 and KDSC-110-10 were resistant to anthracnose, while ten genotypes were moderately resistant and remaining were susceptible. Five systemic, four non-systemic fungicides and three plant extracts were evaluated against C. capsici at different concentrations using spore germination and poison food techniques. Mancozeb followed by copper oxychloride and chlorothalonil among nonsystemic while carbendazim and triademelon among systemic fungicides were effective in vitro. Among the plant extracts, neem seed kernal extract (NSKE) and nimbicidine were effective in inhibiting the fungus. Promising fungicides and plant extracts were tried under field condition. Carbendazim recorded least anthracnose disease incidence and higest yield while iprodione recorded highest disease. The yield and subsequent net returns were more in carbendazim treated plots. The carbendazim gave highest cost benefit ratio of 1:10 and 1:7.96 at 0.15 per cent concentrations followed by mancozeb with 1:7.06 cost benefit

Key words: Capsicum annuum, Colletotrichum capsici, C. gloeosporoides fungicides, genotypes, management, plant products and resistance.

INTRODUCTION

The chillies (*Capsicum* spp.), a member of solanaceous family is mainly cultivated as a spice in many countries including India. India is the major and highly erratic producer, consumer and exporter of chilli covering an area of 8.83 lakh ha with a productivity of 1266 kg/ha (Peter *et al.*, 2006) and Indian chilli is exported to over 90 countries. Among the diseases, anthracnose of chilli caused by *Colletotrichum capsici* (Sydow), Butler and Bisby, and *C. gloeosporioides* (Penz) Penz and Sacc. is becoming more serious in chilli growing tracts of India. Host plant resistance has been an apt choice

in all crop improvement programmes and is essential to recommend directly to farming community for cultivation in endemic area or to use as donors of the resistant genes in breeding programme.

Further, the disease affects only the economic part of the plant, the fruits and quite often the use of fungicides become inevitable when the disease is already prevalent in the field. Hence, evaluation of fungicides and plant products have also been undertaken to know their efficacy and subsequently to recommend them for disease management.

MATERIALS AND METHODS

Isolation of the fungus

The isolates of *C. capsici* and *C. gloeosporioides* were obtained from infected chilli fruits by following standard tissue isolation method. The morphological characters were studied, the pathogenicity was proved and the identity was confirmed before its use for experimental purpose.

Screening of chilli genotypes

Thirty seven chilli genotypes collected from Indian Institute of Horticultural Research, Hessarghatta, Bangalore and Regional Agricultural Research Station, Raichur were screened against anthracnose pathogens both in vitro and in vivo. For in vitro screening of chilli germplasm, pinprick method was followed (Naik and Sinha, 1995). Fifteen days old cultures of C. capsici and C. gloeosporiodes were used for artificial inoculation. The chilli fruits harvested at red ripened stage were surface sterilized with 0.1% HgCl2 and then washed in two changes of sterile water. Thereafter, the fruits were pricked with sterile pin bundles specially devised for pricking the fruits. The pinpricked fruits were then dipped in spore suspension (1×107 spores/ml) for five minutes and then kept for incubation on perforated tray under humid chamber. The humid chamber was prepared by keeping water in tray which was placed below the perforated tray kept with inoculated fruits, the wetted cotton were placed on the trays and the trays were covered with polythene sheet to maintain the relative humidity and then incubated at 27. 7± 1°C. Ten fruits from each genotype were used for pinprick inoculation and three replications were maintained for each genotypes. The infected fruits were scored using 0 to 4 scale. The genotypes were screened under field condition by planting them in test rows regularly interspersed with susceptible check for everyfive rows of test genotypes. The infected fruits were scored after every harvest as per the grade given below.

Grade	Per cent infection	Reaction		
0	No disease	Immune		
1	1-10% disease	Resistant		
2	11-25% disease	Moderately resistant		
3	26-50% disease	Susceptible		
4	51-100% disease	Highly susceptible		

Observations on number of fruits falling in each grades was recorded. Per cent disease index was calculated by using the formula given by Wheeler (1969). All the genotypes were grouped into different categories based on scale given by Bansal and Grover (1969) as above.

In vitro evaluation of fungicide and plant products

Poison food technique

The fungus, *C. capsici* was predominant among the two *Colletotrichum* spp. Hence, *C. capsici* was used for all further studies. The fungus was grown on potato dextrose agar (PDA) medium in Petri plates for 10 days prior to setting up of the experiment. The PDA media was prepared and melted. The fungicidal suspension and plant extracts were added to the melted media on the basis of active ingredient present in the chemical to obtain the required concentration. The fungal disc was placed and incubated at $27\pm1^{\circ}\text{C}$ and the radial growth was recorded (Sharvelle, 1961)

Spore germination technique

The efficacy of the fungicides and plant products were tested against C. capsici by assessing per cent inhibition of germination of conidia. A single drop of condidial suspension was added to the wells of a series of cleaned cavity slides, to which a single drop of different fungicides (double concentration) and required quantity of plant product was also added to get the desired concentrations. Later, coverslips were placed on the cavity slide. The periphery of the cavity was smeared with vaseline to prevent contamination and evaporation of water. Each concentration was replicated thrice on a separate cavity slide. A control treatment was maintained with distilled water. These cavity slides were kept in the Petri dishes lined with moist blotting paper and were incubated at room temperature (27±1°C). Observations were collected from ten microscopic fields for each slide and the total number of conidia germinated in each microscopic field was recorded and per cent germination was calculated. The average of three cavity slides was worked out. The per cent inhibition was calculated as per Naik (1985) for each fungicide.

Per cent inhibition (I) = $C-T/C \times 100$, where C-Number of spores germinated in control; and T-Number of spores germinated in treated cavity slides.

Field evaluation of fungicides and plant products

A field experiment was conducted during rainy season at Horticultural Farm, Regional Agricultural Research Station, Raichur under irrigated condition in order to find out suitable fungicides and plant products among the promsing ones selected during in vitro studies.

The efficacy of four non-systemic fungicides (mancozeb, chlorothalonil, iprodione and copper oxychloride), two systemic fungicides (carbendazim and triademeton) and two plant extracts (neem seed kernal extracts and nimbicidine), which were found effective in vitro conditions, were tried under field condition. Two concentrations of selected fungicides and plant extracts were administered along with untreated control in field experiment. The chemicals and plants extracts were measured accurately just before spraying and mixed thoroughly with water. The first spray was given at green to red turning stage of the chilli fruit and the second after 15 days. The observations were taken for number of fruits infected and the extent of severity of disease. The cost-benefit ratio and net returns were also worked out for finding out its economic feasibility to the farming community.

RESULTS AND DISCUSSION

Screening of chilli genotypes

Thirty seven chilli genotypes when screened against C. gloeosporioides and C. capsici in the laboratory after harvest gave different reactions to the disease. Among the genotypes S-32 showed maximum PDI (81.00%) where as PC-1, Co-1, LCA-301 and IHR-3023 showed least PDI of 2.0, 6.0, 7.0 and 6.0 per cent respectively. These genotypes were further classified into five classes based on reaction and genotypes falling in particular classes are presented in Table 1. Among the genotypes, none of them were found immune against C. gloeosporioides. However, four genotypes viz., PC-1, Co-1, LCA-301 and IHR-3023 were found resistant. Ten genotypes were moderately resistant and twenty genotypes were susceptible and three lines were found highly susceptible.

The data regarding the reaction of genotypes screened against *C. capsici* are presented in Table 1. The genotypes Co-1, PC-1, LCA-301, IHR-3023 and H-232, showed less than 10.0 per cent incidence, as against S-32 which recorded maximum PDI (83.00%). Based on the reaction, these

Table 1 : Reaction of chilli genotypes against C. gloeosporioides and C. capsici in vitro

Grade	Reaction	C. gloeosporioides	C.capsici
0	Immune (0%)	Nil	Nil
1	Resistant (01-10%)	Co-1, IHR-3023, LCA-301, PC-1	Co-1, H-222, IHR-3023, LCA 301, PC-1
2	Moderately resistant	Byadgi dabbi (Hosalli), Byadgi dabbi	Arka Lohit, Byadgi dabbi (Hosalli),
11-25%)	(Chakalabbi), Byadgi ka	addi (Raichur),	Byadgi dabbi (Sanshi), Byadgi kaddi
,		G-4, H-232, H-20, IHR-3006, IHR- 3018, KOS-110-10, PMR-5	(Raichur), G-4, GPC-80, H-20, IHR-3006, IHR-3018, KOSC-6, KOSC-110-10,
3	Susceptible (26-50%)	Arka Lohit, BC-14-2, Byadgi dabbi (Gudigeri), Byadig dabbi (Sanshi), 0-3-75, GPC-80, GPC-82, GPC-69, HMT-9, H-1, H-3, Hissar Shakta, IHR-3002, KOSC-6, LCA-312, LCA-206, LCA-304, Pusa Jwala, Paprika, RCH-1	PMR-5, RCH-I BC-14-2, Byadgi dabbi (Gudigeri), Byadgi dabbi (Chakalabbi), D-3-75, CGP-82, GPC-69, Hissar Shakta, IHR-3002, LCA- 206, ICA-304, LCA-312, Punjab Guttedar, Paprika, Pusa Jwala
4.	Highly susceptible (51-100%)	D-51, Pubjab Guttedar, S-32	D-51, H-1, H-3, HMT-9, S-32

genotypes were classified in to five classes. Genotypes *viz.*, Co-1, PC-1, LCA-301, IHR-3023 and H-232 were found resistant. Thirteen genotypes were moderately resistant, fourteen genotypes were susceptible and five genotypes were highly susceptible.

In the field screening of genotypes PC-1 scored least per cent disease incidence followed by LCA-301, IHR 3023, H 232 and KDSC 110-10, whereas S-32 and HMT-9 were highly susceptible. Among the genotypes, six were identified as resistant and ten were moderately resistant and remainings were susceptible (Table 2). The genotypes identified as resistant are the sources which can be used in advancing breeding for anthracnose resistance in chilli. Such wide response of the chilli genotypes to anthracnose disease were also observed by different workers from diffrent regions. (Bansal and Grover,

Table 2: Reaction of chilli genotypes against anthracnose under field condition

Grade	Reaction	Genotypes
0	Immune (0%)	Nil
1	Resistant	Co-1, H-232, IHR-3023,
	(01-10%)	KOSC-110-10, LCA-301, PC-1
2	Moderately	Arka Lohit, Bydgi dabbi
	resistant	(Hosalli), Byadgi dabbi (Sanshi)
	(11-25%)	Byadgi kaddi (Raichur), GPC-
		80, G-4, IHR-3006, IHR-3018,
		KOSC-6, PMR-5, RCH-1
3	Susceptible	BC-14-2, Byadgi dabbi (Gudi-
	(26.50%)	geri), Byadgi dabbi (Chakalabbi),
		GPC-82,GPC-69, H-1, H-3, H-20,
		Hissar Shakta, IHR-3002, LCA-
		206, LCA-304, LCA-312, Pusa
		Jwla, Paprika, Punjab guttedar
4.	Highly	D-51, D-3-75, HMT-9, S-32
	susceptible	
	(51-100%)	

1969; Kenchaiah, 1975; Singh et al., 1977; Patil et al., 1993). A diffeential reaction was noticed when the two species of Colletorichum, viz., C. capsici and C. gloeosporioides were inoculated to various chilli genotypes. Differential reaction exhibited by genotypes against two species is attributed to variation in pathogenicity among the two species of Colletotrichum. But it is important to look for resistant reaction particularly against C. capsici since that is the most predominant species among the two anthracnose causing fungal species. Although

immune ones were not available, the resistant will go a long way as a component in integrated disease management, thus may prolong the life of even a moderately resistant cultivar. In addition, it will serve as a donor or sources of resistance while furthering the cause of breeding for anthracnose resistance in chilli.

In vitro evalution of fungicides and plant products

Poison food technique

Efficacy of four different non-systemic fungicides and five systemic fungicides were tested on inhibition of mycelial growth of *C. capsici* and results are presented in Table 3. All the non-systemic fungicides

Table 3: Per cent inhibition of mycelial growth of *C. capsici* by different non-systemic and systemic fungicides.

Non-systemic fungicides	Concentration (%)					
	0.1	0.2	0.3	Mean**		
Chlorothalonil (Kavach)	48.10	77.50	83.18	69.59		
Copper oxychloride (Blitox)	42.90	59.80	76.01	59.57		
Iprodione (Rovral)	37.64	55.02	64.66	52.44		
Mancozeb (IM-45)	58.76	83.69	91.11	77.85		
Mean	46.85	69.00	78.74	64.86		
Mean	46.85	69.00	78.74	64		

Systemic fungicides	Concentration (%)					
	0.1	0.2	0.3	Mean**		
Carbendazim (Bavistin)	64.00	83.61	90.82	79.48		
Propiconazole (Tilt)	43.18	67.14	85.15	65.16		
Penconazole (Topas)	44.06	65.04	80.24	63.11		
Triademefon (Bayleton)	54.99	88.44	91.86	78.43		
Thiophanate methyl (Roko)	44.25	65.79	73.59	61.21		
Mean	50.10	74.00	84.33	69.48		

	Non-systemic fungicides	Concentration	Fungicides X Concentrations
SEm±	0.486	0.421	0.841
CD @ 1%	1.936	1.676	3.352
	Systemic fungicides	Concentration	Fungicides X Concentrations
SEm±	0.511	0.396	0.886
CD @ 1%	1.999	1.548	3.462

^{**}Mean of three replications

differed significantly with respect to per cent inhibition of radial growth of *C. capsici*. Mancozeb (77.85%) was found to be the best in inhibiting the radial growth of *C. capsici* over rest of the nonsystemic fungicides at 0.3 per cent concentration followed by chlorothalonil (69.59 per cent) and copper oxychloride (59.57 per cent). Efficacy of mancozeb was earlier reported by previous workers (Malaraju and Swamy, 1988; Datar *at al.*, 1990; Mesta, 1996; Shivakumara, 2004)

Among the systemic fungicides, carbendazim inhibited the growth of fungus to a maximum extent (79.48%) and was on par with triademefon(78.43%) followed by propiconazole (65.16%). However, maximum inhibition of mycelial growth was at 0.15 per cent concentration irrespective of fungicides. Amongst the previous workers, Kumar and Mishra (1988) had also reported similar findings.

Among the plant extracts (Table 4), neem seed kernal extract (NSKE) recorded the maximum inhibition followed by nimbicidine at five per cent concentration. The NSKE gave the maximum inhibition (82.66%) followed by nimbicidine at 0.5 per cent concentration (77.14%). Gupta et al., (1981) noticed the efficacy of neem extracts against *C. capsici*. Nimbicidine has fungicidal activity due to the presence of alkaloids and azadirachtin which might have inhibited the growth of *C.capsici*. Antifungal properties of neem has been reported by Singh et al., (1990).

Table 4: Per cent inhibition of mycolial growth of *C. capsici* by different plant extracts.

Plant extracts	Concentration (%)					
	0.1	0.2	0.3	Mean**		
Neem seed kernal extract	65.17	75.34	82.66	74.40		
Nimbicidine***	55.82	67.00	77.14	66.65		
Neem leaf extract	43.43	63.61	73.80	60.28		
Mean	54.81	68.65	77.87	67.11		

	Plant extracts	Concentration	Plant extracts X Concentrations
SEm±	0.427	0.427	0.740
CD @ 1%	1.764	1.764	3.055

^{**}Mean of four replications

Spore germination technique

The efficacy of different non-systemic and systemic fungicides on inhibition of germination of spores of *C. capsici* differed significantly (Table 5). Mancozeb (89.5%) was the best and significantly superior over other chemicals. Copper oxychloride (79.45%) was the next best followed by chlorothalonil (71.47%). Mancozeb inhibited 92.83% gemination of conidia at 0.3 per cent concentration followed by copper oxychloride (84.91%).

Among systemic fungicides, carbendazim (87.77%) was found to be the best and significantly superior over the other fungicides. Traidemeton (83.76%) was next best followed by propiconazole (74.14%). However, the maximum inhibition of conidial

Table 5: Per cent inhibition of germination of conidia of *C. capsici* in different non-systemic and systemic fungicides.

Non-systemi	c fungicides		Concen	tration (%)
		0.1	0.2	0.3	Mean**
Copper oxyo	chloride	68.82	84.61	84.91	79.45
(Blitox)					
Chlorothalor	nil	60.00	72.51	81.91	71.47
(Kavach)					
Mancozeb (IM-45)	85.56	90.10	92.83	98.50
Iprodione (R	lovral)	47.66	59.38	73.92	60.32
Mean		65.51	76.65	83.39	75.19
Systemic fu	ngicides	Concentration (%)			
		0.1	0.2	0.3	Mean**
Carbendazir	n (Bavistin)	86.10	83.48	93.74	87.77
Propiconazo	le (Tilt)	66.43	74.51	81.49	74.14
Penconazole	(Topas)	50.43	67.13	74.02	63.86
Triademefon	(Bayleton)	76.98	83.15	91.16	83.76
Thiophanate (Roko)	methyl	49.89	61.74	72.33	61.32
Mean		65.97	74.00	82.55	74.17
	Non-systemic fungicides	Concentration		Fungicides X Concentrations	
SEm±	0.353	0.3	05	0.6	11
CD @ 1%	1.360	1.1	78	2.356	
	Systemic fungicides	Concentration		Fungic Concen	ides X trations
SEm±	0.398	0.3	808	0.6	89
CD @ 1%	1.554	1.2	204	2.6	92

^{**}Mean of four replications

^{***}Nimbicidine used at (0.25%, 0.3% and 0.5%) concentration

germination was at 0.15 per cent concentration irrespective of fungicides.

Table 6: Per cent inhibition of germination of conidia of *C. capsici* by different plant extracts.

Plant extra	cts	Concentration (%)					
		0.1	0.2	0.3	Mean**		
Neem seed kernal extract		68.37	70.78	85.83	74.99		
Nimbicidine***		51.50	59.10	79.33	63.31		
Neem leaf extract		45.55	52.06	69.10	55.57		
Mean		55.14	60.65	78.09	64.62		
	Plant extracts	Concer	ntration	Plant ex Concen			
SEm±	0.268	0.2	68	0.4	63		
CD @ 1%	1.105	1.1	05	1.9	14		

^{**}Mean of four replications

The results on the effect of different plant extracts on inhibition of germination of conidia of *C.capsici* are presented in Table 6, which revealed that NSKE at 5.0 per cent concentration recorded significantly higher inhibition of conidia germination (85.83%) over nimbicidine at 0.5 per cent concentration.

Field evaluation of fungicides and plant products

Different systemic, non-systemic fungicides and plant extracts which were found promising in vitro studies were imposed for the control of anthracnose of chilli under field condition and results are presented in the Table 7. Carbendazim at 0.1 and 0.15 per cent concentrations were effective in controlling the anthracnose and subsequently resulted in higher yield. This higher yield in carbendazim treated plot is mainly because of the lower disease indidence of 29.80% and 31.66% at 0.15% and 0.1% concentrations respectively. Amongst the previous workers, (Mishra, 1988; Biswas, 1992; and Hegde and Anahosur, 2001) carbendazim was identified as the best fungicide to control the fruit rot of chilli under field condition. In the present study, carbendazim not only reduced the disease incidence but also gave the higher cost benefit ratio (1:10.04). This is due to the higher yield obtained per unit area when imposed with carbendazim spray on chilli fruits infected with Colletotrichum (Table 7). Hence, carbendazim 0.1 and 0.15 per cent concentration is recommended to the farming community.

Table 7: Field evaluation of fungicides and plant extracts against anthracnose of chilli

Treatment	concen- tration (%)	Mean diseases index	Mean yield of dry chillies (q/ha)	Deviation from control Yield (q/ha)	Total returns Rs. (A)	Total cost Rs. (B)	Net income (A-B)	Cost benefit ratio
Mancozeb (IM-45)	0.2	45.70	4.57	2.12	8480	1200	7256	1:7.06
	0.3	41.92	4.68	2.13	8920	1500	7420	1:5.95
Chlorothalonil (Kavach)	0.2	58.45	3.64	1.19	47.60	2400	2360	1:1.98
	0.3	58.51	3.71	1.26	5040	3300	1740	1:1.53
Iprodione (Rovral)	0.2	75.46	3.07	0.62	2480	5350	-2870	1:0.46
	0.3	70.18	3.26	0.81	3240	7725	-4485	1:0.42
Copper oxycloride (Blitox)	0.2	45.64	4.42	1.97	7880	1175	6705	1:6.71
	0.3	42.00	4.63	2.18	8720	1463	7257	1:5.96
Triademefon (Bayleton)	0.1	46.47	4.27	1.82	7280	3100	4180	1:2.35
	0.15	42.90	4.73	2.28	9120	3740	5380	1:2.44
Carbendazim (Bavistin)	0.1	31.66	5.84	3.39	13560	1350	12210	1:10.0
	0.15	29.80	5.88	3.43	13720	1722	11998	1:7.96
NSKE	5	61.00	3.61	1.16	4640	912	3728	1:5.08
	7	59.84	3.82	1.37	5480	1037	4443	1:1.23
Nimbicdine	0.5	64.82	3.40	0.95	3800	1788	2012	1:2.12
This Add Survey Amendodes Automation	0.7	62.23	3.81	1.36	5440	2263	3177	1:2.40
Control	_	84.10	2.45	_	_	_	_	_
SEm±		0.510	0.124					
CD @5%		1.470	0.356					

^{***}Nimbicidine used at (0.25%, 0.3% and 0.5%) concentration

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