The Pharma Innovation Journal 2023; 12(7): 2330-2334

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(7): 2330-2334

www.thepharmajournal.com
Received: 18-05-2023
Accepted: 21-06-2023

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Biosuppression of sugarcane sett rot disease (*Ceratocystis paradoxa*) with *Chaetomium globosum*

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Abstract
The bio-suppression efficiency of various isolates of *Chaetomium* against *C. paradoxa* was tested. In the dual culture technique, the *Chaetomium globosum* isolate cg 6 was found to possess maximum inhibition of 60.37% inhibition on *Ceratocystis paradoxa* mycelium. Sugarcane bud treated with *C. globosum* isolate cg 6 had higher germination, vigour index in both the tested varieties CoC 25 and Co 86032 compared to other isolates tested. Field trial conducted for two years depict that sett treatment @ 4 g/l + soil application of talc formulated *Chaetomium globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare recorded higher germination 80.87% with higher tiller and shoot population with lower sett rot disease incidence of 8.78% which is 33.52 reduction in disease compared to control and recorded higher yield of 117 t/ha.

Keywords: Sugarcane, sett rot, *Ceratocystis paradoxa*, *Chaetomium globosum*

Introduction
Ceratocystis paradoxa incites sett rot disease, is one of the principal causes of germination failure in fields, which leads to patchy crop stand of sugarcane. The problem exists to varied degrees in all cane-growing countries across the world. This disease is responsible for germination loss that requires additional efforts in gap filling. Thick soils with limited drainage capabilities are particularly favouring the disease and more severe where water stagnation in cane fields during and after planting is widespread. The pathogen enters through cut ends when setts are planted without any pre-plant fungicidal sett treatment (Wismer and Bailey, 1989) [14]. The infected setts smell like mature pineapple fruit (Went, 1896) [13]. The pathogen's metabolic activity produces ethyle acetate, which causes the odour (Kuo *et al.*1969) [3]. Setts affected by this disease may decay before buds germinate or settlings may die back shortly after emergence. Sett root production will be sparse and all these conditions impose a poor cane population. Inhibitions of the pathogen growth by systemic fungicides was high compared to non-systemic fungicides (Vijaya *et al*., 2007) [11].
Different fungal (*Chaetomium*, *Trichoderma*) and bacterial antagonist alone or in combination was found promising in the management of diseases (Viswanathan and Malathi, 2019) [12]. A comprehensive review about the usefulness of *Chaetomium* in the biological control of plant disease was discussed (Ashwini, 2019; Madbouly K and Abdel-Wareth 2020) [2, 4]. *Chaetomium globosum* which grows as saprophyte in rhizosphere and phyllosphere in the cellulose substrate was reported as potential bioagents against *Fusarium* and *Helminthosporium.* (Tveit and Moore, 1954) [10]. Hence attempt was made to manage the sugarcane sett rot disease with *Chaetomium globosum.*

2. Materials and Methods
2.1. Antagonistic effects of *Chaetomium* isolates to *Ceratocystis paradoxa in vitro*
Chaetomium isolates 16 Nos. *viz*., cg1, cg5, cg6, cg111, cg15, cg16, cg19, cg20, cg24, cg26, cg29, cg30, cg35, cg37, cg42 and cg44) obtained from the Dept. of Plant Pathology, TNAU, Coimbatore. The above isolates were evaluated for their antagonistic potential in inhibiting the growth of C. *paradoxa* mycelium by dual culture technique (Dennis and Webster, 1971). Observations was taken after seven days of pathogen inoculation on formation of inhibition zone over pathogen growth. Efficacy of the *Chaetomium* isolates to the growth of *Ceratocystis paradoxa* was arrived on the basis of inhibition zone and growth inhibition was expressed as percent inhibition over control.

The effective *Chaetomium* isolate having higher percent inhibition of the pathogen was used for talc and liquid based bioformulation.

2.2. Growth enhancement effect by different isolates of Chaetomium on sugarcane

Growth enhancement in sugarcane seedlings by nine *Chaetomium* isolates effective in dual plat techniques along with the control (water alone) was assessed based on seedling vigour index of sugarcane seedlings (CoC 25 and Co 86032) maintained in pro trays. The single bud chips of sugarcane were treated for 30 minutes with different isolates of *Chaetomium* sp. before planting in portrays. The treated bud chips were placed in wells of portray and filled with sterilized mixture containing 25 parts vermicompost and decomposed coir pith. For every treatment three replications were kept to observe the growth. After one month of planting, germinated buds were counted and expressed as percentage. The length of the root and shoot length of every seedling was measured 30 and 45 days of planting. Sugarcane seedlings vigour index of each treatment were quantified as per Abdul- Baki and Anderson (1973) [1].

Vigour-index = (Germination%) x (Sum of root and shoot length)

2.3. Field testing the bioformulation of *Chaetomium globosum* cg 6 against *Ceratocystis paradoxa*

Field experiments was carried out in Sugarcane Research Station Farm located at Cuddalore (11º 46' N 79º.46' E, MSL 4.60 m) to identify the effect of bioformulation of *C. globosum* cg6 based on talc and liquid on sett rot disease (*C. Paradoxa*) *during* 2018-19 and 2019-20 using the sugarcane variety Co 86032. The liquid and talc formulated *C. globosum* cg 6 was applied in sugarcane sett and in the soil either individually or in sequence. Carbendazim (0.1 percent) was included as check for comparison. Both formulations are administered to the soil after 30 and 90 days after planting (DAP). The design of the experiment was RBD with three replications. Treatments are as follows.

T_1- Sett treatment with talc formulated *C. globosum* cg 6 @ 4 g/l for 10 minutes
T_2- Soil application of with talc formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare
T_3- Sett treatment + Soil application with talc formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare
T_4- Sett treatment with liquid formulated *C. globosum* cg 6 @ 10 ml/l for 10 minutes
T_5- Soil application with liquid formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare
T_6- Sett treatment + Soil application with liquid formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare
T_7-Sett treatment with carbendazim (0.1 percent) for 10 minutes
T_8- Control
The disease incidence was calculated as follow

Percent disease incidence = (Total number of infected sett / Total number of setts) x 100

Table 1: *In vitro* antagonistic activity of *Chaetomium* isolates against *C. paradoxa* by dual plate technique.

S. No.	Isolates	Mycelial growth of *C. paradoxa* (cm)	Percent inhibition over control
1.	*Chaetomium globosum* cg 1	4.87	45.93 (42.66)
2.	*C. globosum* cg 5	5.00	44.44 (41.81)
3.	*C. globosum* cg 6	3.57	60.37 (50.99)
4.	*C. globosum* cg 11	4.53	49.63 (44.79)
5.	*C. globosum* cg 15	4.00	55.56 (48.19)
6.	*C. globosum* cg 16	4.57	49.26 (44.58)
7.	*C. globosum* cg 17	4.67	48.15 (43.94)
8.	*C. globosum* cg 20	4.03	55.19 (47.98)
9.	*C. globosum* cg 24	4.13	54.07 (47.34)
10.	*C. globosum* cg 26	4.93	45.19 (42.24)
11.	*C. globosum* cg 29	5.20	42.22 (40.53)
12.	*C. globosum* cg 30	4.80	46.67 (43.09)
13.	*C. globosum* cg 35	3.87	57.04 (49.05)
14.	*C. globosum* cg 37	4.53	49.63 (44.79)
15.	*C. globosum* cg 42	3.93	56.30 (48.62)
16.	*C. globosum* cg 44	5.13	42.96 (40.95)
17.	Control (*C. paradoxa alone*)	9.00	--
	SEm±	0.15	0.96
	CD (p=0.05)	0.42	2.77

Figures in parenthesis are arcsine transformed values

Table 2: Effect of *Chaetomium* bud chip treatment on the growth of sugarcane seedling var. Co 86032

S. No	Treatment	Germination (%)	30 DAP			45 DAP		
			Root length (cm)	Shoot length (cm)	Vigour index	Root length (cm)	Shoot length (cm)	Vigour index
1.	*Chaetomium globosum* cg 1	64.2 (53.3)	11.95	27.25	2508	14.35	32.39	2991
2.	*C. globosum* cg 6	80.2 (63.6)	11.55	28.55	3208	17.00	34.70	4136
3.	*C. globosum* cg 11	68.0 (55.5)	12.06	28.94	2788	13.47	31.59	3064
4.	*C. globosum* cg 15	80.1 (63.5)	12.10	26.20	3064	16.95	33.73	4054
5.	*C. globosum* cg 20	68.4 (55.8)	10.25	23.25	2278	13.71	31.66	3085
6.	*C. globosum* cg 24	70.6 (57.2)	11.93	29.03	2867	14.88	34.02	3422
7.	*C. globosum* cg 26	68.6 (55.9)	10.25	23.25	2278	16.20	33.63	3388
8.	*C. globosum* cg 35	70.4 (57.04)	11.93	29.03	2867	14.61	33.78	3580
9.	*C. globosum* cg 42	68.5 (55.8)	12.75	29.75	2890	15.93	33.95	3392
10.	Control	52.4 (46.4)	13.95	29.23	2245	14.96	30.11	2343
	SEm±	1.04	0.31	0.57	84.90	0.36	0.55	130.14
	CD (p=0.05)	3.08	0.92	1.70	252.20	1.05	1.64	386.61

Figures in parenthesis are arcsine transformed values

Table 3: Effect of *Chaetomium* bud chip treatment on the growth of sugarcane seedling var. CoC 25

S. No	Treatment	Germination (%)	30 DAP			45 DAP		
			Root length (cm)	Shoot length (cm)	Vigour index	Root length (cm)	Shoot length (cm)	Vigour index
1.	*Chaetomium globosum* cg 1	64.3 (53.3)	16.57	27.48	2819	18.99	31.43	3227
2.	*C. globosum* cg 6	84.0 (66.4)	14.40	26.05	3397	20.59	34.91	4661
3.	*C. globosum* cg 11	70.2 (56.9)	23.92	14.32	2676	18.70	28.36	3294
4.	*C. globosum* cg 15	84.6 (66.9)	18.20	25.44	3665	19.79	33.14	4661
5.	*C. globosum* cg 20	76.6 (61.1)	18.90	26.50	3450	19.76	32.90	3995
6.	*C. globosum* cg 24	80.4 (63.7)	15.15	27.55	3416	18.74	31.08	3985
7.	*C. globosum* cg 26	72.7 (58.5)	13.00	25.60	2779	16.58	30.83	3413
8.	*C. globosum* cg 35	76.9 (61.3)	15.28	29.33	3390	18.43	33.24	3926
9.	*C. globosum* cg 42	74.2 (59.5)	12.35	26.50	3318	20.76	30.24	3774
10.	Control	56.4 (48.7)	14.55	25.75	2256	16.77	29.15	2571
	SEm±	1.35	0.42	0.52	93.04	0.36	0.73	137.53
	CD (p=0.05)	4.02	1.25	1.54	276.41	1.08	2.18	408.56

Figures in parenthesis are arcsine transformed values

Table 4: Efficacy of talc and liquid based bioformulation of *Chaetomium globosum* cg 6 on the growth and yield of sugarcane (Pooled analysis of 2018-19 and 2019-20)

T. No.	Treatments	Germination (%)	Tiller population ('000/ha)	Shoot population ('000/ha)	Sett rot incidence (%)	Percent reduction over control	Yield (t/ha)
T_1	Sett treatment with talc formulated *C. globosum* cg 6 @ 4 g/l for 10 minutes	69.56 (56.51)	127	109	12.26 (20.50)	7.12	101.86
T_2	Soil application with talc formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare	74.32 (59.55)	132	111	11.16 (19.51)	15.49	105.04
T_3	Sett treatment + Soil application with talc formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare	80.87 (64.06)	145	131	8.78 (17.23)	33.52	117.24
T_4	Sett treatment with liquid formulated *C. globosum* cg 6 @ 10 ml/l for 10 minutes	68.87 (56.09)	124	111	11.71 (20.01)	11.33	98.14
T_5	Soil application with liquid formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare	74.52 (59.68)	135	119	10.55 (18.95)	20.08	107.11
T_6	Sett treatment + Soil application with liquid formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare	71.72 (57.87)	142	130	9.53 (17.98)	27.80	111.37
T_7	Sett treatment of carbendazim (0.1 percent) for 10 minutes	69.96 (56.76)	133	124	10.80 (19.19)	18.18	99.72
T_8	Control	57.17 (49.12)	127	101	13.20 (21.30)	7.12	94.59
SEm±		0.70	3.93	4.41	0.52	-	4.30
CD (5%)		2.05	11.535	12.92	1.52	-	12.61

Figures in parenthesis are arc signed transformed value

3. Results and discussion

3.1. Antagonistic effects of *Chaetomium* isolates to *Ceratocystis paradoxa in vitro*

Among the 16 isolates of *C. globosum* tested *in vitro* for its efficiency in inhibition of the pathogen growth (*C. paradoxa*) by dual culture technique, the isolate cg 6 has maximum inhibition of 60.37 percent and was on par with isolates cg 35 (57.04%) and cg 42 (56.30%) (Table 1). The antagonistic activity of *Trichoderma harzianum* against *Ceratocystis paradoxa* due to overgrowth and growth inhibition both *in vitro* and *in vivo* conditions and on par with fungicide treatment with Carbendazim was reported by Talukder (2008) [9] was in corroboration with the present findings. The decrease in mycelial development was caused by nutritional competition, mycoparasitism and the formation of antimicrobial metabolites. (Paulina Moya *et al.*, 2016) [5]. The increased activity of both exo and endo glucanase activity of *C. globosum* isolate Cg-6 correlated with retardation of *P. infestans* growth *in vitro* reported by Shanthiyaa *et al*. (2013) [8] confirms the current findings.

3.2. Plant growth promotion activity of *Chaetomium* isolates in sugarcane

Among the treatments, in the sugarcane variety Co 86032 maximum germination of 80.2% was recorded in bud chips treated with *C. Globosum* cg 6 and it was found on par the isolate cg 15 which recorded 80.1% of germination, whereas in untreated control minimum germination of 52.4% was recorded. The sugarcane seedling vigour indicated that the higher vigour index of 6034 was recorded in treatment with the isolate cg 15 and found on par with cg 6 at 30 DAP. The vigour index at 45 DAP recorded high vigour index value of 4136 in treatment with isolate cg 6 which was on par with the isolate cg 15. In the untreated control, the lowest vigour index of 2393 was recorded (Table 2).

The highest germination of 84.6% was recorded in treating the bud chips of sugarcane variety CoC 25 with *C. globosum* isolate cg 15 and found on par with the isolate cg 6 (84.0%). The minimum germination of 56.4% was recorded in untreated control. Among various isolates of *C. globosum* tested, higher vigour index of 3665 was found with isolate cg 15 and found on par with cg 6 (vigour index of 3397), cg 24 (vigour index of 3416) and cg 35 (vigour index of 3390) at 30 DAP. The vigour index at 45 DAP indicated the higher values with the treatment of isolated cg 6 and cg 15 (Table 3). Shanmugam *et al*. (2016) [7] had opined that sugarcane bud chips treated with *Pseudomonas fluorescens* and incorporation of the same in the coco peat resulted in the increased bud germination and reduced sett rot incidence.

3.3. Field testing the bioformulation of *Chaetomium globosum* cg 6 against *Ceratocystis paradoxa*

From the pooled analysis of field trials conducted during 2018-19 and 2019-20 it was evident that sett treatment with talc formulated *C. globosum* cg 6 @ 4 g/l for 10 minutes together with soil application with talc formulated *C. globosum* cg 6 before planting and at 30 and 90 DAP @ 2.5 kg per hectare had higher germination 80.87% with higher tiller and shoot population with lower sett rot disease incidence 8.78% which is 33.52% reduction in disease compared to control and recorded higher cane yield of 117 t/ha (Table 4). The effectiveness of application of various formulations of *Chaetomium* strains against Oomycetes pathogens was reported by Raguchander *et al.* (2014) [6]. Similarly, tuber treatment, soil application and foliar spray of *C. globosum* cg 6 results in the reduction of late blight infestation in potato (Shanthiyaa *et al*., 2013) [8] and sugarcane setts treated with. *Trichoderma harzianum* increased the germination and cane yield (Talukder *et al.*, 2008) [9] supports present findings.

Conclusions

Prevalence of higher moisture in soil leads to failure in the germination of the sugarcane bud due to sett rot disease results in the poor plant population and reduction in the yield. The biocontrol agent *Chaetomium globosum* has growth enhancement of sugarcane and effective against the sett rot pathogen when applied as sett treatment and as soil application in addition *Chaetomium* also survive in the moist soil condition.

Acknowledgment

The authors are acknowledged to the Professor and Head, Sugarcane Research Station, Cuddalore for the conduct of this experiment.

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