

Original Research Article

<https://doi.org/10.20546/ijcmas.2020.901.038>

Integration of Seed Biopriming, Soil and Foliar Application of Formulations of *Trichoderma* Species for management of Anthracnose of Sorghum under Field Condition

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ABSTRACT

The different mode of application like seed biopriming, soil application and integration of seed biopriming+soil application enriched FYM + foliar spraying with T3,T4,T15 and T19 isolates of *Trichoderma asperellum* and T6 isolate of *Trichoderma harzianum* were evaluated for management of anthracnose of sorghum. In additive effect of biopriming of seed with different isolates of *Trichoderma* + soil application of *Trichoderma* isolates enriched FYM + foliar spraying with different isolates of *Trichoderma* was found highly effective for reducing the percent disease index(PDI) and increased the grain yield as compared to individual application of seed biopriming and soil application of different isolates of *Trichoderma*. Minimum PDI was recorded in T19 isolate (45.82%) at 75DAS, minimum AUDPC (659.08) and lowest mean infection rate (0.08 unit- days) was recorded in T19 isolate and maximum green fodder yield (68.64 tonnes/ha, 34.70% higher over check) was observed in T3 isolate.

Keywords

Areca nut, UHPLC,
Redox titration,
Vitamin B₆,
Vitamin C

Article Info

Accepted:
15 December 2019
Available Online:
20 January 2020

Introduction

Sorghum crop (*Sorghum bicolor* L. Moench) is originated from North East Africa (Pedersen *et al.*, 2003), it belongs to the family Poaceae and grown in hot and dry agro

ecosystem with low moisture content (Prasad, 2012). It can be easily grown in drought prone condition. More than 35% of the sorghum production can be directly used for human consumption and remaining percentage is used for production of alcohol, animal feed and

industrial products (Awika and Rooney, 2004). In India, mostly prevalent foliar diseases of sorghum are anthracnose, tar spot, downy mildew and rust (Sharma *et al.*, 1978). One of the most important foliar diseases is anthracnose caused by *Colletotrichum graminicola* (Ces.) Wilson, that causes huge loss in productivity.

Due to foliar disease, the losses in yield upto 32-45% (Cheeser, 1959). Cultural and chemical management were used for managing the anthracnose of sorghum but for long term management it is avoided as sorghum crop used as fodder crop and due to chances of environment pollution, considered as hazardous in nature.

For sustainable management of this disease, the application of biocontrol agent is alternative method.

Due to lack of information on judicious management of anthracnose of sorghum through *Trichoderma* isolates, the present study was carried out under field condition to check the efficacy of seed biopriming, soil application and combined application of seed biopriming, soil application and foliar spray of *Trichoderma* isolates for reducing the percent disease index and increase in grain yield of sorghum.

Materials and Methods

Isolates of T3, T4, T15 and T19 of *Trichoderma asperillum* and T6 isolate of *Trichoderma harzianum* isolated from sorghum rhizosphere of Uttarakhand were used for field evaluation.

Field Trials

The field experiment was conducted in Randomized block design with five replications in Kharif Season 2015.

Susceptible Sorghum cultivar PC-23 was planted at sorghum pathology block, Livestock Research Centre, GBPUA&T for evaluation of effectiveness of growth promoting activity of *Trichoderma* isolates.

Seed biopriming

Seed were presoaked in water for 24 hrs. Presoaked seeds were treated with talc based product of *Trichoderma* having 108cfu/g @ 10g/kg of seeds in 2% gum arabic solution.

The seed were incubated for 48h at 25-28°C. *Trichoderma* isolates adhered on the seed; grow on the seed surface to form a protective layer all around the seed coat under moist condition. The observation regarding root length, shoot length and stem diameter were recorded after 90DAS.

Soil application

For the soil application the 20kg of talc based formulation of *T. asperillum* and *T. harzianum* isolates was mixed with 200kg of well-rotted Farm yard manure in a pit and covered for 10 days by polythene sheet.

The mixture was turned every 3 days regularly. When *Trichoderma* mycelium proliferated throughout the FYM then on 10th of day pit was opened and mixture was turned well and spread in the field plots one week before sowing.

Foliar spray

Foliar spray of talc based formulation *Trichoderma* isolates @ 10g/lit of water was given after 30 DAS.

Statistical analysis

Data obtained on various traits under field experiments were analyzed by one-way

analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) using Statistical Product and Service Solution (SPSS) version 16.0 software Developed by SPSS Inc., now IBM SPSS. All results were expressed at $P < 0.05$ to compare difference among the treatment means. All the treatment replicated five times.

Results and Discussion

The present experiment was conducted at livestock research centre, sorghum pathology block, G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand. The results obtained are presented systematically and discussed accordingly.

Effect of biopriming of seed with different isolates of *Trichoderma* on growth promotion and disease reduction under field condition

Percent disease index

The effect of seed biopriming with *Trichoderma* isolates on disease severity and crop yield of sorghum was recorded during 2015 under field experiment.

Observation of PDI were recorded on 30DAS, 45 DAS, 60DAS and 75DAS after 15 days of interval to check the effect of different treatments on the progress of anthracnose of sorghum (Table 1 and Fig 1a and Fig b). During 2015, minimum PDI was recorded in T19 isolate (52.69%) followed by T3 isolate (53.88%) which was statistically at par with T19 isolate where as maximum PDI was recorded in untreated control (71.27%) at 75 DAS.

AUDPC

During 2015, all the treatments were effective in reducing AUDPC over check. Minimum

AUDPC were recorded in T19 isolate (715.04) followed by T3 isolate (725.68) where as maximum AUDPC was found in untreated control (983.36).

Infection rate

Apparent infection rate was calculated by Vanderplank's formulae. During 2015, lowest mean infection rate was recorded in T19 isolate (0.09 unit-days) followed by T15 isolate (0.09 unit-days) and T3 isolate (0.09 unit-days) where as maximum infection rate was recorded in untreated control (0.11 unit-days).

Green fodder Yield

During 2015, seed biopriming with different *Trichoderma* isolates significantly increased the yield as compared to control.

T3 isolate recorded the highest green fodder yield being 57.45 tonnes/ha (21.97% higher over check) followed by T19 isolate (56.00 tonnes/ha) (19.95% higher over check) however T4 isolate (53.95 tonnes/ha) (16.91% higher over check) was statistically at par with T19 isolate where as minimum yield was recorded in untreated control.

Our results are similar to that reported by Yadav *et al.*, 2013 who concluded that seed biopriming enhanced germination percentage and increased plant growth as compared to non primed control plants.

Seed biopriming reduces the amount of biocontrol agent that is used for biopriming than seed coating or seed treatment. Seed biopriming also enhanced the uniform and rapid seedling emergence.

The bioprimed seeds tolerate the adverse soil conditions and promote the plant growth (Mathre *et al.*, 1999). Biopriming of seed

enhanced the seedling growth and reduced the incidence of seed and soil borne diseases (Zaidi *et al.*, 2004).

Effect of soil application of *Trichoderma* isolates enriched FYM on disease reduction under field condition:

Percent disease index

The effect of soil application enriched FYM with *Trichoderma* isolates on disease severity and crop yield of sorghum was recorded during 2015 under field experiment.

Observation of PDI were recorded on 30DAS,45 DAS,60DAS and 75DAS after 15 days of interval to check the effect of different treatments on the progress of anthracnose of sorghum(Table 2, Fig 1a and Fig 1b).

During 2015, minimum PDI was recorded in T3 isolate (57.80%) followed by T19 isolate (58.06%) which was statistically at par with T3 isolate where as maximum PDI was recorded in untreated control (71.27%) at 75 DAS.

AUDPC

During 2015, all the treatments were effective in reducing AUDPC over check. Minimum AUDPC were recorded in T19 isolate (772.74) followed by T3 isolate (777.51) where as maximum AUDPC was found in untreated control (983.36).

Infection rate

Apparent infection rate was calculated by Vanderplanks formulae. During 2015, lowest mean infection rate was recorded in T19 isolate (0.10 unit- days) followed by T4 isolate (0.10 unit-days) and T3 isolate (0.10 unit-days) where as maximum infection rate was recorded in untreated control (0.11 unit-days).

Green fodder Yield

During 2015, soil application enriched FYM with different *Trichoderma* isolates significantly increased the yield as compared to control.

T3 isolate recorded the highest green fodder yield being 53.94tonnes/ha (16.87% higher over check) followed by T19 isolate (52.90tonnes/ha) (15.26% higher over check) however T4 isolate (50.26tonnes/ha) (10.82% higher over check) was statistically at par with T19 isolate where as minimum yield was recorded in untreated control (44.82tonnes/ha).

Soil application of *Trichoderma viride* and *Trichoderma harzianum* enriched FYM one week before sowing of seed was more effective in reducing the wilt and root rot of chickpea and enhanced the plant growth.

Enhancement in the *Trichoderma* population, played a positive role in increasing the uptake of nutrients from the rhizospheric zone that ultimately enhanced the plant growth promotion after the soil application of *Trichoderma* enriched FYM has been reported in Maize (Bjurkman *et al.*,1994), tomato (Ozbay *et al.*,2004), pea (Naseby *et al.*, 2000) and cucumber(Kleifield and Chet, 1992).

Effect of seed bioprimering + soil application + foliar spraying with *Trichoderma* isolates on disease reduction under field condition

Percent disease index

Additive effect of bioprimering of seed with different isolates of *Trichoderma* + soil application of *Trichoderma* isolates enriched FYM + foliar spraying with different isolates of *Trichoderma* on disease severity and crop yield of sorghum was recorded during 2015 and 2018 under field experiment.

Table.1 Effect of seed bioprimering with *Trichoderma* isolates on growth promotion and disease reduction under field condition

2015								
Treatments	30days	45days	60days	75days	AUDPC	r-Value	Green fodder yield(tonnes/ha)	Increase in Yield%
T3	14.36±0.09 ^c (22.27)	25.74±0.64 ^c (30.48)	43.81±0.73 ^d (41.45)	53.88±0.52 ^d (47.23)	725.68	0.09	57.45±1.71 ^a	21.97
T4	15.08±0.05 ^{bc} (22.85)	28.74±0.59 ^b (32.41)	47.30±0.86 ^c (43.45)	55.79±0.58 ^c (48.33)	780.34	0.09	53.95±1.37 ^{ab}	16.91
T6	15.66±0.40 ^{bc} (23.31)	28.09±0.65 ^b (32.00)	49.40±0.37 ^b (44.66)	58.37±0.84 ^b (49.82)	801.54	0.10	52.40±1.43 ^{bc}	14.45
T15(A)	16.21±0.69 ^b (23.73)	28.25±0.88 ^b (32.10)	48.63±0.61 ^{bc} (44.21)	56.04±1.03 ^c (48.47)	791.02	0.09	49.50±1.90 ^c	9.44
T19(B)	14.61±0.53 ^c (22.468)	25.91±0.55 ^c (30.60)	42.58±0.55 ^d (40.73)	52.69±0.37 ^c (46.54)	715.04	0.09	56.00±1.79 ^{ab}	19.95
Control	19.57±0.77 ^a (26.25)	32.40±0.37 ^a (34.69)	62.66±0.46 ^a (52.33)	71.27±0.62 ^a (57.59)	983.36	0.11	44.82±1.16 ^d	-
CD at 5%	1.64 (1.23)	1.86 (1.17)	2.13 (1.21)	1.08 (0.62)			1.99	
C.V.	5.60 (2.87)	3.57 (2.01)	2.35 (1.50)	1.01 (0.69)			2.89	
SE(m)	0.52 (0.45)	0.58 (0.42)	0.67 (0.44)	0.34 (0.12)			0.67	

Table.2 Effect of soil application of *Trichoderma* isolates enriched FYM on disease reduction under field condition

2015								
Treatments	30days	45days	60days	75days	AUDPC	r-Value	Green fodder yield(tonnes/ha)	Increase in Yield%
T3	14.55±0.88 ^b (22.45)	26.39±0.39 ^b (30.91)	48.51±0.44 ^d (44.14)	57.80±0.65 ^d (49.49)	777.51	0.10	53.94±1.12 ^a	16.87
T4	16.16±0.94 ^b (23.68)	28.06±1.59 ^b (31.97)	52.67±0.91 ^c (46.53)	59.99±0.66 ^{cd} (50.76)	831.61	0.10	50.26±0.79 ^a	10.82
T6	14.41±0.98 ^b (22.29)	28.67±1.51 ^b (32.36)	54.56±0.60 ^b (47.62)	61.32±0.79 ^{bc} (51.54)	847.68	0.11	50.04±2.09 ^a	10.35
T15(A)	15.89±1.03 ^b (23.47)	27.38±0.87 ^b (31.54)	55.58±0.52 ^b (48.21)	62.32±0.83 ^b (52.13)	854.46	0.10	51.88±3.54 ^a	13.59
T19(B)	15.79±0.96 ^b (23.39)	26.77±0.64 ^b (31.16)	46.69±0.46 ^c (43.10)	58.06±0.25 ^d (49.64)	772.74	0.10	52.90±1.77 ^a	15.26
Control	19.57±0.77 ^a (26.25)	32.40±0.37 ^a (34.69)	62.66±0.46 ^a (52.33)	71.27±0.62 ^a (57.59)	983.36	0.11	44.82±1.16 ^b	
CD at 5%	3.02 (2.34)	2.71 (1.71)	1.53 (0.87)	2.23 (1.31)			4.29	
C.V.	10.21 (5.45)	5.20 (2.93)	1.55 (1.01)	1.96 (1.39)			6.42	
SE(m)	0.95 (1.65)	0.85 (0.88)	0.48 (0.22)	0.70 (.52)			1.45	

Values given in column are the average of five replications. Values with different alphabetical (a–d) superscripts within a column are significantly different ($P \leq 0.05$) using Duncan's multiple range tests (DMRT). Values in parenthesis are arcsine transformed value

Fig.1a Sorghum anthracnose disease progress under seed bioprimering with different *Trichoderma* isolates in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

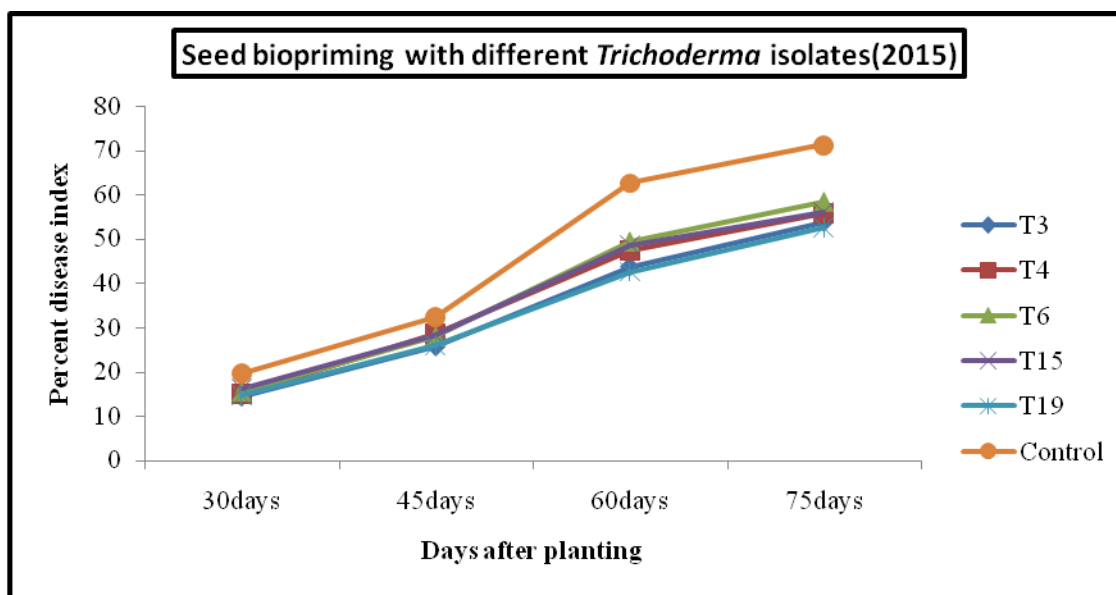


Fig.1b 'A' and 'r' values of Sorghum anthracnose disease under seed bioprimering with different *Trichoderma* isolates in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

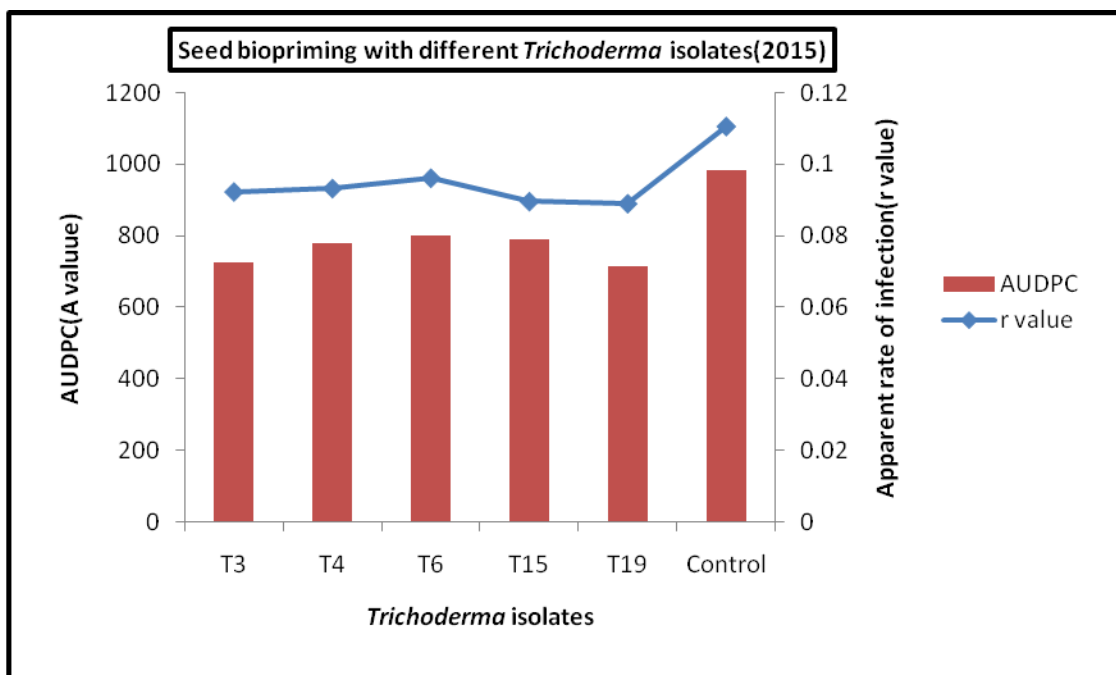


Fig.2a Sorghum anthracnose disease progress under soil application of different *Trichoderma* isolates enrich FYM in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

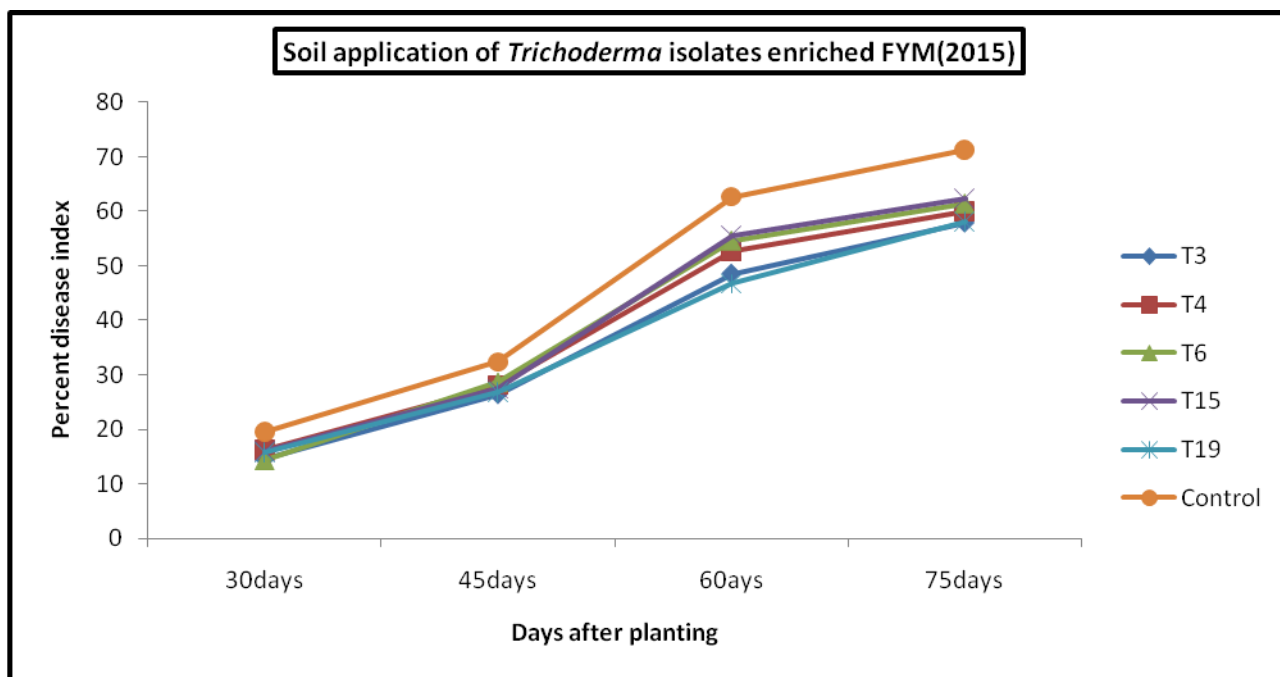


Fig.2b 'A' and 'r' values of Sorghum anthracnose disease under soil application of *Trichoderma* isolates enrich FYM in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

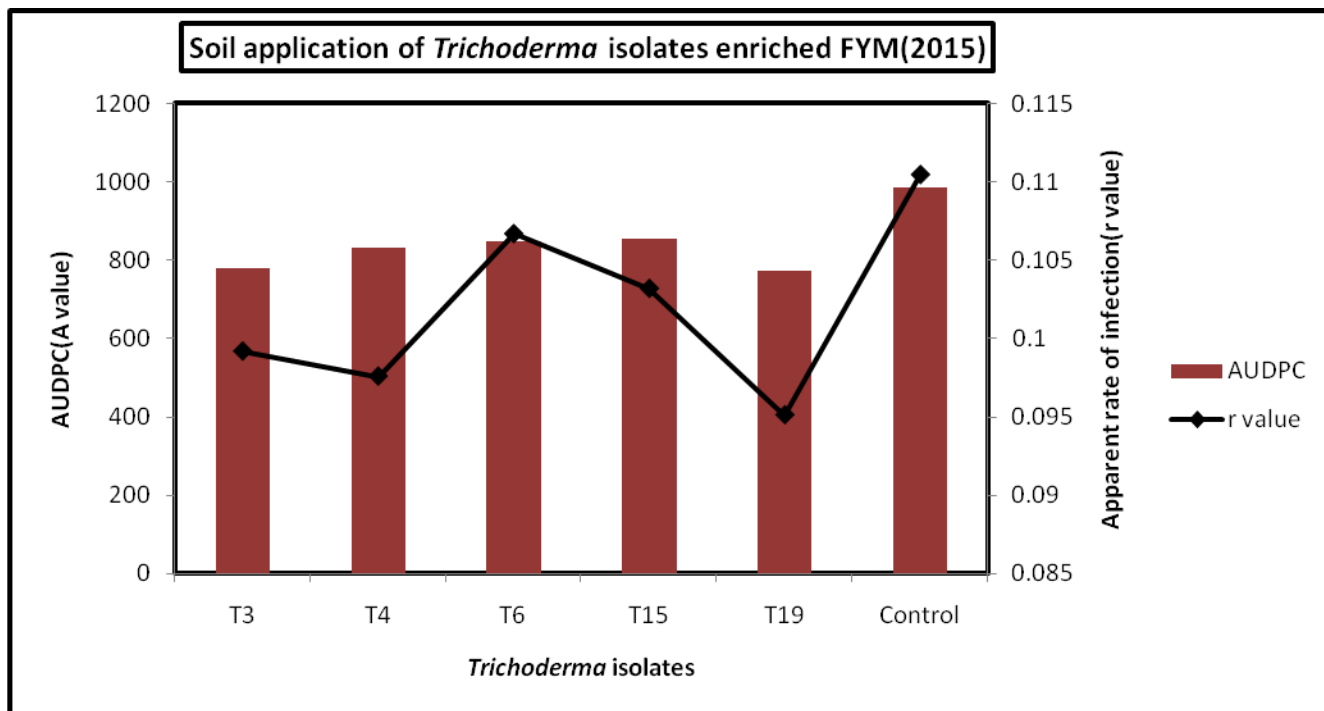


Fig.3a Sorghum anthracnose disease progress under integration seed biopriming + soil application + foliar spraying with *Trichoderma* isolates in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

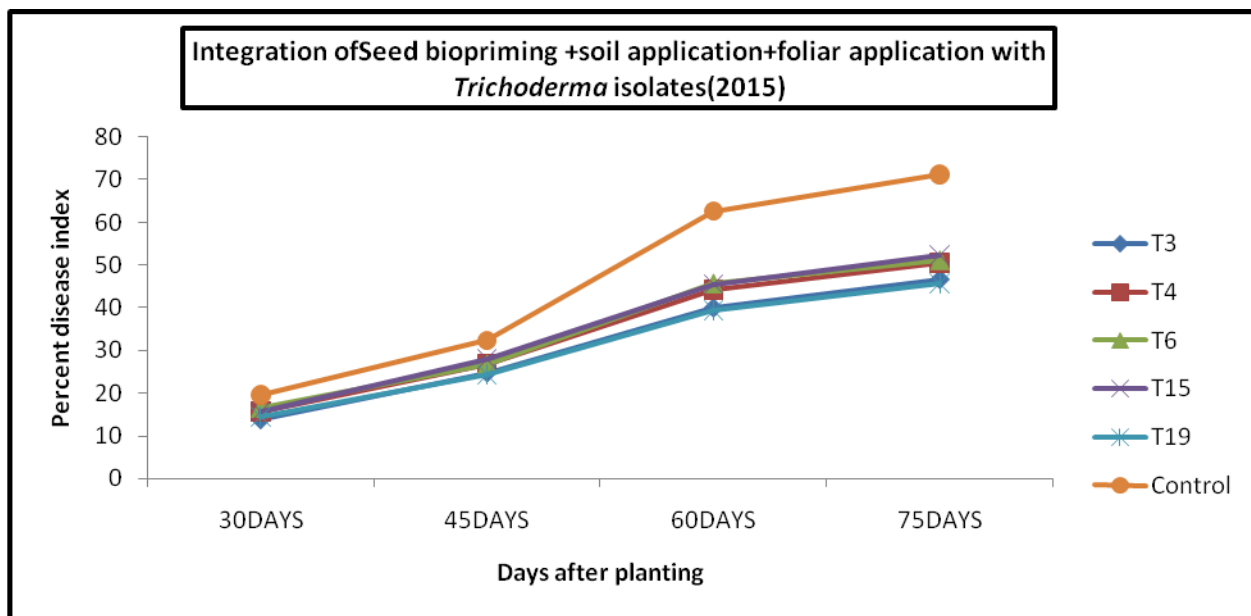


Fig.3b 'A' and 'r' values of Sorghum anthracnose disease under integration seed biopriming + soil application + foliar spraying with *Trichoderma* isolates in the field trial at sorghum pathology block at Pantnagar during *kharif* 2015-16

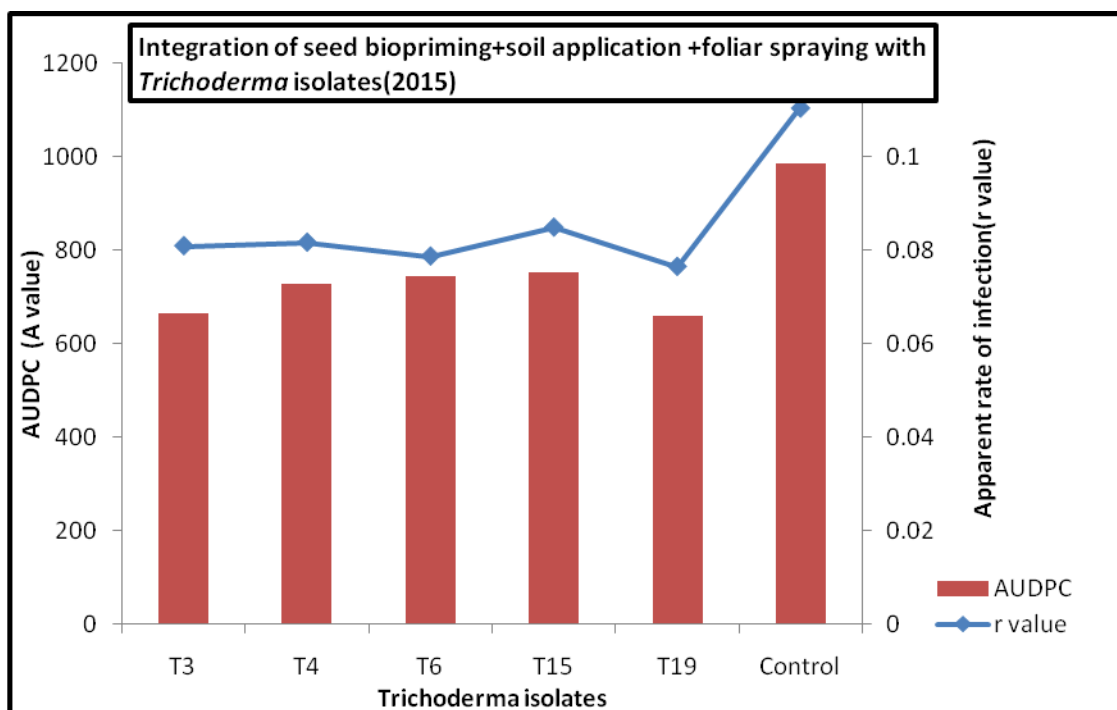


Table.3 Effect of seed bioprimering + soil application + foliar spraying with *Trichoderma* isolates on disease reduction under field condition

2015								
Treatments	30days	45days	60days	75days	AUDPC	r-Value	Green fodder yield(tonnes/ha)	Increase in Yield%
T3	13.74±0.45 ^c	24.73±0.49 ^c	(29.82) 39.90±0.61 ^c	(39.17) 46.59±0.38 ^c	663.63	0.08	68.64±1.69 ^a	34.70
T4	(21.75)	(43.04)	26.73±0.70 ^b	(31.13) 44.24±0.42 ^b	728.23	0.09	59.59±1.32 ^b	24.78
T6	15.54±0.57 ^{bc}	(41.69) 50.58±0.55 ^b	(45.33)	26.79±0.55 ^b	743.55	0.08	55.44±1.66 ^c	19.15
T15(A)	(23.21)	(31.17) 45.59±0.32 ^b	(42.47) 51.04±0.89 ^b	(45.60)	750.68	0.08	57.27±2.06 ^{bc}	21.73
T19(B)	16.63±0.45 ^b	27.87±0.58 ^b	(31.86) 45.48±0.58 ^b	(42.41) 52.26±0.14 ^b	659.08	0.08	66.62±1.86 ^a	32.72
Control	(24.06)	(46.29)	24.50±0.63 ^c	(29.67) 39.49±0.72 ^c	983.36	0.11	44.82±1.16 ^d	
CD at 5%	15.52±0.68 ^{bc}	(38.93) 45.82±0.45 ^c	(42.60)	32.40±0.37 ^a			3.97	
C.V.	(23.19)	(34.67) 62.66±0.46 ^a	(52.33) 71.27±0.62 ^a	(57.59)			5.12	
SE(m)	14.50±0.47 ^c	1.53	(0.97) 1.83	(1.05) 1.85			1.34	

Values given in column are the average of five replications. Values with different alphabetical (a–d) superscripts within a column are significantly different ($P \leq 0.05$) using Duncan’s multiple range tests (DMRT). Values in parenthesis are arcsine transformed value

Observation of PDI were recorded on 30DAS,45 DAS,60DAS and 75DAS after 15 days of interval to check the effect of different treatments on the progress of anthracnose of sorghum(Table 3, Fig 3a and Fig 3b). During 2015, minimum PDI was recorded in T19 isolate (45.82%) followed by T3 isolate (46.59%) which was statistically at par with T3 isolate where as maximum PDI was recorded in untreated control (71.27%) at 75 DAS.

AUDPC

During 2015, all the treatments were effective in reducing AUDPC over check. Minimum AUDPC were recorded in T19 isolate (659.08) followed by T3 isolate (663.63) where as maximum AUDPC was found in untreated control (983.36).

Infection rate

Apparent infection rate was calculated by

Vanderplanks formulae. During 2015, lowest mean infection rate was recorded in T19 isolate (0.08 unit- days), T6 isolate (0.08unit-days) and T3 isolate (0.08 unit-days) where as maximum infection rate was recorded in untreated control (0.11 unit-days).

Green fodder Yield

During 2015, additive effect of bioprimering of seed with different isolates of *Trichoderma* + soil application of *Trichoderma* isolates enriched FYM + foliar spraying with different isolates of *Trichoderma* significantly increased the yield as compared to control.T3 isolate recorded the highest green fodder yield being 68.64 tonnes/ha (34.70% higher over check) followed by T19 isolate (66.62tonnes/ha) (32.72% higher over check) however T4 isolate (59.59tonnes/ha) (24.78% higher over check) was statistically at par with T19 isolate where as minimum yield was recorded in untreated control (44.82tonnes/ha).

Our results are similar to that reported by Meena *et al.*, 2008 on effect of foliar spray and biopriming of seed with *Pseudomonas* and *Trichoderma* on disease severity and plant growth against anthracnose of sorghum. Maximum increase in root length, shoot length and stem diameter and significant reduction in disease severity was observed. Singh and Singh (2008) also reported that biopriming of seed and foliar spraying of *Trichoderma harzianum* isolates decreased the disease severity and increased overall plant growth and yield of sorghum. The combined effect of seed treatment and soil application with *Trichoderma* enriched FYM increased the root and shoot length and significantly reduced the wet root rot incidence in Chick pea as compared to control (Jambhulkar, 2015).

T3 isolate (*Trichoderma asperellum*) was found effective in growth promotion and disease reduction in glass house and field condition. Among the mode of application, the integration of seed biopriming with *Trichoderma* isolates +soil application with *Trichoderma* enrich FYM and foliar spraying with *Trichoderma* isolates was found effective.

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How to cite this article:

Nazia Manzar and Yogendra Singh. 2020. Integration of Seed Biopriming, Soil and Foliar Application of Formulations of *Trichoderma* Species for management of Anthracnose of Sorghum under Field Condition. *Int.J.Curr.Microbiol.App.Sci*. 9(01): 334-344.
doi: <https://doi.org/10.20546/ijcmas.2020.901.038>