Journal of Biological Control, 35(2): 57-67, 2021, DOI: 10.18311/jbc/2021/28324

Research Article

Assessment of incidence of collar rot disease of groundnut in Andhra Pradesh and evaluation of bioefficacy of *Trichoderma* spp. against *Aspergillus* spp.

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ABSTRACT: Roving survey was conducted during *Kharif*, 2018-19 in three major groundnut growing districts of Rayalaseema in Andhra Pradesh to know the status of collar rot (caused by *Aspergillus* spp.) incidence under field conditions. Effect of variety, seed treatment, farming situation, soil type and cropping pattern on disease incidence was studied. Maximum collar rot incidence was observed in Anathapuramu district (17.24%) followed by Chittoor district (15.90 %) and Y.S.R. Kadapa district (12.56%). Even though fungicides were reported to be effective against collar rot of groundnut, there is limited data available on biological control of collar rot. Hence, a study was also conducted to isolate and test *Trichoderma* spp. that is effective against the collar rot pathogen. Eight *Trichoderma* spp. isolates were evaluated against six virulent *Aspergillus* spp. isolates (i.e., two isolates of *A. niger*, two isolates of *A. flavus* and two isolates of *A. terreus*) using dual culture technique. From the results it was revealed that, *Trichoderma* spp. isolate GT2 (88.64 %) exhibited highest overall mean per cent inhibition followed by GT1 (85.17 %) and GT7 (84.20 %). Whereas, the lowest mean per cent inhibition was recorded in GT3 (45.59 %).

KEY WORDS: Collar rot, disease incidence, Dual culture technique, Rayalaseema region, *Trichoderma* spp.

(Article chronicle: Received: 19-05-2021; Revised: 22-06-2021; Accepted: 24-06-2021)

INTRODUCTION

Groundnut (*Arachis hypogaea*) is an important commercial crop in India. It is a good oil yielder with rich protein content. It belongs to the Leguminosae family and originated from South America. It is also known as 'King of oilseeds' (Aycock, 1966). It is cultivated extensively in tropical and subtropical countries during *kharif* and *rabi* seasons as sole or intercrop with an average rainfall requirement of 600 to 1200 mm. Many fungal diseases affect the crop during its growth. Among all the diseases associated with groundnut, collar rot is one of the most severe diseases and causes heavy losses both in haulm and pod yield. Collar rot is widespread in almost all groundnut growing states of India *viz.,* Andhra Pradesh, Tamil Nadu, Gujarat, Punjab, Uttar Pradesh, Maharashtra, Karnataka, Rajasthan and Orissa. In India, 40 to 50 per cent losses may occur in terms of mortality of plants due to *Aspergillus niger* (Ghewande *et al*., 2002). Recent studies revealed that, along with *A. niger* other species like *A. flavus* and *A. terreus* are also capable of causing collar rot to some extent. Collar rot can be managed by seed treatment with both non-systemic and systemic fungicides like mancozeb, tebuconazole, thiram, carbendazim and vitavax (Rakholiya *et al.,* 2012). Even though the disease can be effectively managed by fungicides, usage of fungicides leads to problems such as high capital investment, non-remunerative, poor availability, selectivity, temporary effect, efficacy affected by physicochemical and biological factors, environmental pollution, development of pest resistance, pollution of food and feeds and health hazard. To overcome these problems, biological control can be a safe and alternative method for long time sustainability (Amin *et al*., 2010; Dhotre and Vanmare, 2017). *Trichoderma* species effectively control many soil borne fungi by different modes of action including antibiotic production, secretion of lytic enzymes and direct penetration of the host hyphae (Singh and

Jadon, 2019). The success of *Trichoderma* as a biocontrol agent is due to various antagonistic interactions that can lead to biological control include antibiosis, competition and hyperparasitism (Cook and Baker, 1983). Therefore, in the present investigation different isolates of *Trichoderma* spp. were evaluated to find out the most effective fungal antagonists against *A. niger, A. flavus* and *A. terreus* which were collected during survey.

MATERIAL AND METHODS

Survey for collection of disease samples and rhizosphere soil samples

Roving survey was conducted and Groundnut plants showing typical symptoms of collar rot and composite soil samples from the rhizosphere of healthy plants in collar rot infected Groundnut fields of different mandals in Chittoor, Anantapuramu and Y.S.R. Kadapa districts were collected. In each district three mandals were selected and in each mandal three villages were surveyed for the assessment of disease incidence and data on soil type, cropping pattern, farming situation and variety cultivated were collected in surveyed areas. Per Cent Disease Incidence (PDI) was calculated using formula

PDI = No. of plants infected/Total no. of plants x 100

Isolation of the pathogen by tissue segment method and blotter plate method

Sixteen isolates of *Aspergillus niger*, four isolates of *A. flavus* and five isolates of *A. terreus* were isolated from collar rot affected Groundnut fields from the surveyed areas (3 mandals each in Chittoor, Y.S.R. Kadapa and Anantapuramu districts) by tissue segment method (Devi and Prasad, 2009; Singh and Jadon, 2019). One isolate of each *A. niger*, *A. flavus* and *A. terreus* were isolated from groundnut seed of 'Narayani' variety by blotter plate method (ISTA, 1985; Agarwal and Sinclair, 1987). Axenic culture of the pathogens were obtained by single spore isolation method (Rani, 2015) and single hyphal tip method (Rasheed *et al*. 2004; Gajera *et al.* 2016) and maintained on PDA slants throughout the present investigation.

Identification of *Aspergillus* spp.

Based on the mycological keys provided by Barnett and Hunter, 1972 pathogens were identified.

In case of *A. niger,* conidiophores were simple, upright having globose swelling terminally and bears phialides attached to the globose head which covered entire vesicle. Conidia were one celled, dark brown to light brown in colour and arise basipetally. Mycelium is hyaline, highly branched and septate. Colony is white initially and becomes black after sporulation.

In *A. flavus* conidiophores were pitted, rough, spiny and variable in length and covered 3/4th of vesicle. They were either biseriate or uniseriate. Conidia were globose to subglobose, echinulate to smooth walled and produced sclerotia. Mycelium is hyaline, highly branched and septate. Colony is white initially and becomes green to yellow after sporulation.

Aspergillus terreus have dense columnar head with sub spherical vesicle and biseriate conidiogenous cells. Phialides covered half to 3/4th of the vesicle. Conidia were smooth walled. Colony is white initially and becomes brown after sporulation.

Isolation of native antagonistic *Trichoderma* spp. and identification

The collected composite soil samples from the rhizosphere of healthy plants in collar rot infected Groundnut fields were shade dried (Table 1). Serial dilution technique (Johnson and Curl, 1972) was used to isolate *Trichoderma* spp. on Trichoderma Selective Medium (TSM). *Trichoderma* isolates were identified based on the colour of the colony, presence of chlamydospores, branching of conidiophores, shape and disposition of phialides and shape of conidia (Gams and Bisset, 1998).

Antagonistic potential of *Trichoderma* spp. isolates against *Aspergillus* spp. *in vitro* by Dual culture technique:

Individual *Trichoderma* isolates were dual cultured with *A. niger*, *A. flavus* and *A. terreus* isolates *in vitro* (Dennis and Webster, 1971). Twenty ml of melted and cooled Potato Dextrose Agar (PDA) medium was poured into Petri plates and allowed to solidify. Culture disks (5mm) of *Trichoderma* was placed 1 cm away at one end of Petri plates. A 5 mm test pathogen culture disk was placed 1 cm away at the opposite end (with a gap of 7 cm between the two culture discs). Plates monocultured with either of the test fungi served as check. Three replications were maintained for each treatment. The radial growth of the pathogen in dual culture and control plates was measured after seven days of incubation at 28±1^{0}C and the inhibition per cent of pathogen was calculated by using the following formula

$$I = \frac{C - T}{C} \times 100$$

Wherein,

I = Per cent reduction in growth of pathogen C= Radial growth (mm) in control

T = Radial growth (mm) in treatments

Table 1. List of *Aspergillus* spp. and *Trichoderma* spp. isolated from different mandals in Chittoor, Y.S.R. Kadapa and Anantapuramu districts of Andhra Pradesh

District	Mandal	Village	Isolate names			
			A. niger	*A. flavus*	*A. terreus*	*Trichoderma* spp.
Chittoor	Kalahasthi	Panagallu	An1	-	At1	-
		Inagalur	An2	-	-	-
		Chukkalanidi-gallu	-	-	-	GT1
	Yerpedu	Vekatapuram	An3	Af1	-	-
		Madibaka	An4	-	At2	-
		Bandarupalli	-	-	-	GT5
	Somala	Nanjampeta	An5	-	-	-
		Peddaupparapalli	An6	-	-	-
	Tirupati urban		An7	Af2	-	-
Y.S.R. Kadapa	Chintakomma dinne	Chinnakampalle	An8	-	At3	GT6
		Agraharam	An9	-	-	-
		Venkatarampalle	An10	-	-	-
	Siddavatam	Lingampalli	-	-	-	-
		Ponnavolu	An11	-	-	GT7
		S. Rajampeta	-	Af3	-	-
	Valluru	Kotluru	An12	-	At4	GT8
		Tappetla	-	-	-	-
		Koppolu	An13	-	-	-
Anantapuramu	Kadiri	Patnam	An14	-	-	GT2
		Kaalsamudram	-	-	-	-
		Kadiri	-	Af4	-	-
	Obula devara cheruvu	Inagalur	-	-	At4	-
		Narappagaripalle	-	-	-	GT3
		Nallaguntlapalle	An15	-	At5	-
	Nallamada	Donnikota	-	-	-	GT4
		Goppepalle	An16	-	-	-
		Nallamada	-	-	-	-

*An: *Aspergillus niger*, Af: *Aspergillus flavus*, At: *Aspergillus terreus,* GT: Groundnut *Trichoderma*; Number of samples from each village was 3.

RESULTS AND DISCUSSION

From the survey data in Table 2, it is evident that highest collar rot incidence was observed in Anathapuramu district (17.24 %) followed by Chittoor district (15.90 %) and Y.S.R. Kadapa district (12.56 %). The crop was cultivated in sandy loam and clay loam soils in all the locations surveyed.

Among all three districts surveyed, highest disease incidence (28.60 %) was observed in Peddaupparapalli of Somala mandal, Chittoor district followed by Donnikota (24.27 %) of Nallmada mandal, Anathapuramu district, which was at par with Nallaguntlapalle (23.47 %) of Obula devara cheruvu mandal, Anathapuramu district and Narappagaripalle (23.40 %) of Obula devara cheruvu mandal, Anathapuramu district. While lowest disease incidence was observed in Inagalur (4.47 %) of Srikalahasti mandal, Chittoor district which was at par with Kotluru (4.60 %) of Vallur mandal, Y.S.R. Kadapa district.

In Chittoor district, highest disease incidence was recorded in Peddaupparapalli (28.60 %) of Somala mandal

Table 2. Survey for the occurrence and distribution of collar rot of groundnut in major growing districts of Rayalaseema in A.P. during *Kharif,* 2018-19

District	Mandal	Village	Latitude, Longitude	No. of samples	Soil type	Cropping pattern	Farming situation	Variety	Per cent disease incidence
Chittoor	Srikalahasthi	Panagallu	13.726270, 79.671007	3	Clay loam	Groundnut + Paddy	Irrigated	Dharani	10.07 (18.39) stuvwxy
		Inagalur	13.891666, 79.566461	3	Clay loam	Groundnut + Paddy	Irrigated	Dharani	4.47 (12.20)z
		Chukkalanidi-gallu	13.734908, 79.682735	3	Clay loam	Groundnut + Paddy	Rainfed	Kadiri-6	15.53 (23.20) ghijk
	Yerpedu	Vekatapuram	13.685322, 79.643910	3	Sandy loam	Groundnut + Redgram	Rainfed	Narayani	20.37 (26.82) de
		Madibaka	13.648487, 79.604338	3	Sandy loam	Groundnut + Redgram	Rainfed	Dharani	13.43 (21.47) ijklmnop
		Bandarupalli	13.676539, 79.636989	3	Clay loam	Groundnut + Paddy	Rainfed	Kadiri-6	16.07 (23.62) ghi
	Somala	Nanjampeta	13.467388, 78.853995	3	Clay loam	Groundnut + Paddy	Irrigated	Dharani	15.03 (22.79) ghijklmn
		Peddauppara-palli	13.447350, 78.847890	3	Sandy loam	Groundnut + Redgram	Rainfed	Narayani	28.60 (32.32) a
	Tirupati urban	Tirupati urban	13.624919, 79.373939	3	Sandy loam	Groundnut + Redgram	Rainfed	Dharani	19.57 (26.21) ef
								Mean	15.90
Y.S.R. Kadapa	Chintakomma dinne	Chinnakam-palle	14.382420, 78.807375	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	15.70 (23.32) ghij
		Agraharam	14.379888, 78.821473	3	Clay loam	Groundnut + Paddy	Irrigated	Kadiri-6	11.10 (19.45) pqrstuvwx
		Venkataram-palle	14.374079, 78.815984	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	15.00 (22.77) ghijklmno
	Siddavatam	Lingampalli	14.517681, 78.873676	3	Sandy loam	Groundnut + Redgram	Rainfed	Dharani	12.70 (20.87) jklmnopqrstu
		Ponnavolu	14.502616, 78.871973	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	16.73 (24.13) fgh
		S. Rajampeta	14.469755, 78.950539	3	Clay loam	Groundnut + Paddy	Rainfed	Kadiri-6	8.80 (16.66) wxy
	Valluru	Kotluru	14.611932, 78.737076	3	Sandy loam	Groundnut + Redgram	Irrigated	Kadiri-6	4.60 (12.35)z
		Tappetla	14.577757, 78.702000	3	Sandy loam	Groundnut + Redgram	Irrigated	Dharani	15.13 (22.88) ghijklm
		Koppolu	14.560750, 78.705125	3	Clay loam	Groundnut + Paddy	Rainfed	Kadiri-6	13.23 (21.31) ijklmnopqr
								Mean	12.56
Anantapuramu	Kadiri	Patnam	14.223592, 78.093555	3	Sandy loam	Groundnut + Redgram	Rainfed	Dharani	12.43 (20.57) lmnopqrstuv
		Kaalsam-udram	14.194984, 78.123161	3	Sandy loam	Groundnut + Castor	Rainfed	Kadiri-6	13.30 (21.38) ijklmnopq
		Kadiri	14.109304, 78.146132	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	13.07 (21.6) ijklmnopqrs
	Obula devara cheruvu	Inagalur	14.057817, 78.036520	3	Sandy loam	Groundnut + Castor	Rainfed	Kadiri-6	17.33 (24.59) efg
		Narappagari-palle	14.028273, 78.017518	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	23.40 (28.88) bcd

District	Mandal	Village	Latitude, Longitude	No. of samples	Soil type	Cropping pattern	Farming situation	Variety	Per cent disease incidence
		Nallagunt-lapalle	14.009384, 78.123161	3	Sandy loam	Groundnut + Castor	Rainfed	Dharani	23.47 (28.96) bc
	Nallamada	Donnikota	14.121205, 78.071680	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	24.27 (29.49) b
		Goppepalle	14.137470, 78.010421	3	Sandy loam	Groundnut + Redgram	Rainfed	Kadiri-6	15.17 (22.91) ghijkl
		Nallamada	14.142269, 77.984036	3	Sandy loam	Groundnut + Castor	Rainfed	Kadiri-6	12.77 (20.89) jklmnopqrst
								Mean	17.24
								C.D.	3.06 (2.68)
								SE(m)	1.08 (0.94)
								SE(d)	1.52 (1.33)
								C.V.	12.23 (7.21)

and lowest in Inagalur (4.47 %) of Srikalahasti mandal. While in Y.S.R. Kadapa district, highest incidence of disease was recorded in Ponnavolu (16.73 %) of Siddavatam mandal and lowest was recorded in Kotluru (4.60 %) in Valluru mandal; whereas in Anathapuramu district, maximum disease incidence was recorded in Donnikota (24.27 %) of Nallamada mandal and minimum in Patnam (12.43 %) of Kadiri mandal.

Depending upon the place of cultivation, per cent disease incidence varied with each variety. Among all the villages surveyed, variety Kadiri-6 (K-6) was predominantly cultivated in all the areas. Out of 27 villages surveyed, K-6 was cultivated in 16 villages and per cent disease incidence was ranging from 4.60 per cent in Kotluru of Valluru mandal, Y.S.R. Kadapa district to 24.27 per cent in Donnikota of Nallamada mandal, Anathapuramu district.

In Chittoor district, K-6 variety of Groundnut was cultivated in only two villages of Chukkalanidigallu of Srikalahasti mandal and Bandarupalli of Yerpedu mandal with 15.53 per cent and 16.07 per cent disease incidence respectively which are at par with each other. While in Y.S.R. Kadapa district, it was ranged from 4.60 per cent (Kotluru, Valluru mandal) to 16.73 per cent (Ponnavolu, Siddavatam mandal) with significant difference between maximum and minimum disease incidence. In Anathapuramu district, K-6 was cultivated in seven out of nine villages surveyed with 12.43 per cent (Patnam, Kadiri mandal) to 24.27 per cent (Donnikota, Nallamada mandal) where the difference between maximum and minimum disease incidence was significant. Narayani variety was cultivated in two villages i.e., Venkatapuram of Yerpedu mandal and Peddaupparapalli of Somala mandal in Chittoor district with a disease incidence of 20.37 per cent and 28.60 per cent respectively with significant difference between them. Dharani variety was cultivated in nine villages with per cent disease incidence ranging from 4.47 per cent in Inagalur village of Srikalahasti mandal, Chittoor district to 23.47 per cent in Nallaguntlapalle of Obula devara cheruvu mandal, Anathapuramu district where the difference in maximum and minimum disease incidence was significant.

Disease incidence varied from 4.47 per cent in Inagalur of Srikalahasti mandal to 19.57 per cent in Tirupati urban of Chittoor district where the difference was significant. 12.43 per cent and 23.47 per cent disease incidence was observed in Patnam of Kadiri mandal and Nallaguntlapalle of Obula devara cheruvu mandal respectively in Anathapuramu district. In Y.S.R. Kadapa district, per cent disease incidence varied from 12.70 per cent (Lingampalli, Siddavatam mandal) to 15.13 per cent (Tappetla, Valluru mandal) which are at par with each other.

Among all the varieties cultivated in survey area, Dharani variety had recorded minimum disease incidence of 4.47 per cent in Inagalur of Srikalahasti mandal in Chittoor district and maximum of 28.60 per cent was recorded with Narayani variety at Peddaupparapalli of Somala mandal in Chittoor district.

Seed treatment

Collar rot of groundnut is a soil borne disease which occurs within 30 days after sowing. So, seed treatment is the effective means of control in the initial stages of the crop. Seed treatment was followed only in five villages out of 27 villages investigated. The PDI varied from 4.47 per cent in Inagalur of Srikalahasti mandal, Chittoor district to 12.27 per cent in Nallamada village and mandal of Ananthapuramu

district where seed treatment was practiced, while it was from 11.10 per cent (Agraharam of Chintakomma dinne mandal, Y.S.R. Kadapa district) to 28.60 per cent (Peddaupparapalli of Somala mandal, Chittoor district) where seed treatment was not practiced.

In Chittoor district, maximum PDI was recorded in Panagallu (4.47 %) of Srikalahasti mandal and minimum was Inagalur (4.47 %) of Srikalahasti mandal with significant difference. In Y.S.R. Kadapa district, maximum disease incidence was observed in S. Rajampet (8.80 %) Siddavatam mandal and significantly minimum was in Kotluru (4.60 %), Valluru mandal, while 12.77 per cent disease incidence was recorded in Nallamada mandal of Ananthapuramu district.

Farming situation

Per Cent Disease Incidence (PDI) varied depending on the farming situation, groundnut is widely cultivated as a rainfed crop so, it depends on rainfall. Collar rot disease is predisposed by high temperature and dry conditions. Hence, soil moisture plays a crucial role in disease development. In the villages surveyed, crop was cultivated under both rainfed and irrigated situations. Out of 27 villages surveyed, 21 villages were under rainfed and 6 villages were under irrigated situation. PDI varied from 8.80 per cent (S. Rajampeta of Siddavatam mandal in Y.S.R. Kadapa district) to 28.60 per cent (Peddaupparapalli of Somala mandal, Chittoor district) under rainfed and 4.47 per cent (Inagalur of Srikalahasti mandal, Chittoor district) to 15.13 per cent (Tappetla, Valluru mandal of Y.S.R. Kadapa district). The disease severity was more under rainfed than the irrigated condition. In Chittoor district, PDI varies between 4.47 (Inagalur, Srikalahasti mandal) to 15.03 per cent (Nanjampeta, Somala mandal) under irrigated and 13.43 per cent (Madibaka, Yerpedu mandal) to 28.60 per cent (Peddaupparapalli, Somala mandal) under rainfed where, the difference in PDI was significant. In Y.S.R. Kadapa district, PDI ranges from 4.60 per cent (Kotluru, Valluru mandal) to 15.13 per cent (Tappetla, Siddavatam mandal) under irrigated and 8.80 per cent (S. Rajampeta, Siddavatam mandal) to 16.73 per cent (Ponnavolu, Siddavatam mandal) under rainfed with significant difference in PDI. In Ananthapuramu district, groundnut is cultivated under rainfed in all the villages surveyed with significantly minimum PDI (12.43 %) was observed Patnam of Kadiri mandal and maximum PDI 24.27 per cent in Donnikota of Nallamada mandal.

Soil type

In the areas surveyed, groundnut is cultivated in sandy loam soil type in 19 villages and clay loam in eight villages. In clay loam, higher PDI i.e., 16.07 per cent was reported in Bandarupalli, Yerpedu mandal of Chittoor district and lowest 4.47 per cent was in Inagalur of Srikalahasti mandal, Chittoor district. In Chittoor district, PDI ranges from 4.47 per cent Inagalur, Srikalahasti mandal to 16.67 per cent in Bandarupalli, Yerpedu mandal, whereas in Y.S.R. Kadapa district 8.80 per cent to 13.23 per cent [DI in S. Rajampeta, Siddavatam mandal and Koppolu, Valluru mandal respectively. Whereas in Ananthapuramu district, groundnut is not cultivated in clay loam in the areas surveyed. In sandy loams, Peddaupparapalli, Somala mandal in Chittoor district observed highest PDI (28.60 %) Kotluru, whereas Valluru mandal of Y.S.R. Kadapa district recorded lowest PDI (4.60 %). In Chittoor district, PDI ranged from 13.43 per cent Madibaka, Yerpedu mandal to 28.60 per cent in Peddaupparapalli, Somala mandal, In Y.S.R. Kadapa district, maximum PDI (16.73 %) was recorded in Ponnavolu, Siddavatam mandal and minimum PDI (4.60 %) was found in Kotluru of Valluru mandal. In Ananthapuramu district, highest PDI (24.27 %) was recorded in Donnikota of Nallamada mandal and lowest (13.07 %) in Kadiri mandal.

Cropping pattern

Per cent disease incidence varied based on the cropping system followed. Groundnut- Paddy cropping system was followed in eight villages where PDI varied from 4.47 per cent in Inagalur of Srikalahasti mandal, Chittoor district to 16.07 per cent in Bandarupalli of Yerpedu mandal, Chittoor district. Groundnut-Castor cropping system was followed only in four villages of Ananthapuramu district where the PDI varied from 12.77 per cent in Nallamada village and mandal to 23.47 per cent in Nallaguntlapalle of Obula devara cheruvu mandal. Groundnut-Redgram cropping system was observed in 15 villages of the areas surveyed and PDI ranged from 4.60 per cent in Kotluru, Valluru mandal of Y.S.R. Kadapa district to 24.27 per cent in Donnikota of Nallamada mandal, Ananthapuramu district.

Nandeesha *et al.* (2013) carried out a survey on the occurrence of collar rot disease in groundnut in four major groundnut growing villages of Tirupati and its surrounding areas *viz.,* Srikalahasti, Renigunta, Ramachandrapuram and Chandragiri and the highest mean per cent disease incidence (11.21 %) was recorded in Srikalahasthi mandal and least mean per cent disease incidence (6.47 %) was noticed in Chandragiri mandal. Rani *et al.* (2016) conducted a survey in major groundnut growing areas of Andhra Pradesh during *Kharif,* 2012-13 and Telangana during *Rabi,* 2012-13 to assess the distribution and the incidence of collar rot and stem rot diseases. They recorded groundnut cultivar Kadiri-6 (K-6) as the prominent cultivar in all the districts surveyed and highest incidences of stem rot and collar rot was observed in Chittoor district of Andhra Pradesh. Similar studies were also conducted by Rao *et al.* (1997), Mohammed and Chala (2014) and Kumari and Singh (2016).

Bioefficacy of *Trichoderma* spp. against *Aspergillus* spp.

Among the 28 *Aspergillus* spp. isolates (i.e., 17 isolates of *A. niger*, five isolates of *A. flavus* and six isolates of *A.*

terreus), two isolates of *A. niger* (An1 and An8), two isolates of *A. flavus* (Af1 and Af5) and two isolates of *A. terreus* (At1 and At4) were proved to be more virulent in pathogenicity test. So, they were used to screen eight isolates of *Trichoderma* spp. by dual culture technique. The data noted was presented in Table 3; Plate 1, 2 and 3.

Screening of *Trichoderma* spp. isolates against *Aspergillus niger*

When An1 isolate of *A. niger* was tested against all eight isolates of *Trichoderma* spp. maximum disease inhibition (91.25 %) over control was recorded with GT2 followed by GT1 (88.75 %), GT7 (88.33 %), GT4 (86.25 %), GT5 (65.25 %), GT6 (65.00 %) and GT8 (61.25 %). GT2 was on par with GT1 and GT7 and significantly differed with remaining isolates. Minimum disease inhibition was recorded with GT3 (54.58 %) which was significantly differed with all other isolates.

Whereas An8 isolate of *A. niger* tested against all the *Trichoderma* isolates, highest inhibition was recorded with GT2 (92.08 %) which is on par with GT1 (89.58 %) and GT7 (89.58 %) and significantly differed with all the remaining isolates. Lowest inhibition over the control was recorded with GT5 (58.83%) which was significantly differed with all other isolates. The descending order of inhibition percentage of all *Trichoderma* isolates against An8 was presented below:

GT2 > GT1 > GT7 > GT6 > GT8 > GT4 > GT5 > GT3

Screening of *Trichoderma* spp. isolates against *A. flavus*

When Af1 isolate of *A. flavus* when screened against eight *Trichoderma* spp. Isolates, GT2 (93.75 %) caused maximum inhibition of radial growth followed by GT7 (92.08 %), GT1 (90.42 %), GT8 (80.00 %), GT4 (65.42 %), GT5 (65.00 %) and GT6 (64.17 %) at seven DAI.

No significant difference was observed with GT2, GT1, GT7 and significantly differed from all other isolates. Least antagonism effect was observed with GT3 (56.25 %), which was also significantly differed from remaining isolates.

Whereas Af5 isolate of *A. flavus* was screened against all eight *Trichoderma* spp. isolates, maximum per cent inhibition of radial growth of test pathogen isolate i.e., Af4 over control obtained with GT2 (92.08 %) followed by GT1 (88.75 %), GT7 (88.75 %), GT8 (87.92 %), GT6 (67.92 %), GT5 (67.50 %) and GT4 (63.75 %) at seven DAI. GT2 inhibition percentage (92.08 %) was at par with GT1 and GT7 and differed significantly by all other isolates. Least inhibition of growth of *A. flavus* Af4 isolate (57.08 %) was noted with GT3 isolate of *Trichoderma* spp.

Screening of *Trichoderma* spp. isolates against *A. terreus*

When At1 isolate of *A. terreus* was screened with eight *Trichoderma* spp. isolates by dual culture. Per cent inhibition of pathogen radial growth ranged between 31.63 to 82.33. GT2 showed maximum inhibition of pathogen growth i.e., 82.33 per cent which was at par with GT1 (78.80 %) and GT7 (77.56 %). Minimum inhibition per cent was observed with GT6 (31.63 %) which was at par with GT8 (36.31 %) and GT3 (36.40 %). It was observed that among the *Trichoderma* isolates tested the maximum to minimum antagonism effect was as follows: GT2 (82.33 %) > GT1 (78.80 %) > GT7 (77.56 %) > GT4 (49.56 %) > GT5 (41.17 %) > GT3 (36.40 %) > GT8 (36.31 %) > GT6 (31.63 %)

Whereas, At4 isolate of *A. terreus* was evaluated against 8 *Trichoderma* spp. *in vitro* condition. It is evident from Table 3 that GT2 isolate of *Trichoderma* showed significantly maximum inhibition percentage (82.22). Minimum inhibition of growth of A. terreus At4 isolate (7.78 %) was noted in GT3 isolate of *Trichoderma* spp. The per cent inhibition of all isolates of *Trichoderma* spp. descending order was as follows: GT2 (82.22 %) > GT1 (76.67 %) > GT7 (70.00 %) > GT6 (41.11 %) > GT4 (32.22 %) > GT8 (30.00 %) > GT5 (28.89 %) > GT3 (7.78 %).

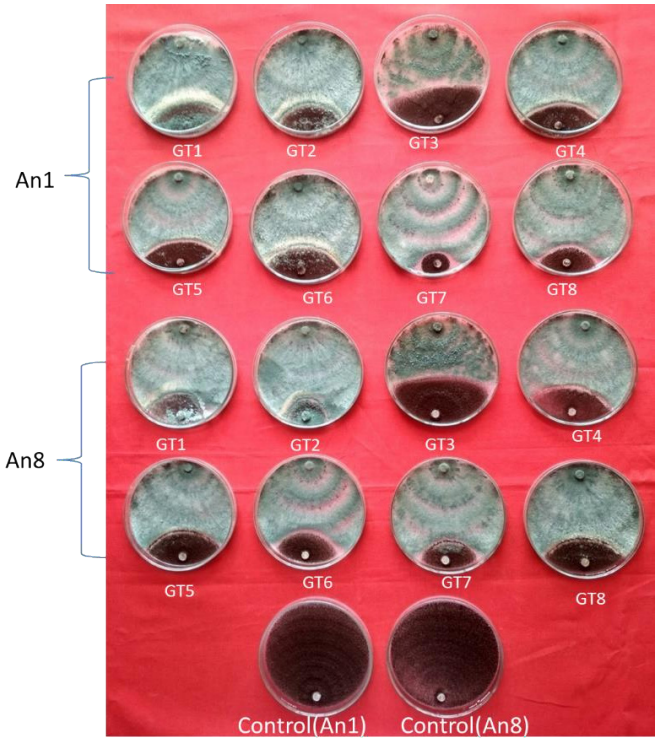

Plate 1. *In vitro* evaluation of *Trichoderma* spp. isolates (GT1 to GT8) against *A. niger* isolates (An1 and An8) by dual culture

Table 3. *In vitro* evaluation of *Trichoderma* spp. isolates against *Aspergillus* spp. isolates by dual culture

Treatments	*A. niger*				*A. flavus*				*A. terreus*			
	An1		An8		Af1		Af5		At1		At4	
	Radial growth (cm)	Per cent inhibition	Radial growth (cm)	Per cent inhibition	Radial growth (cm)	Per cent inhibition	Radial growth (cm)	Per cent inhibition	Radial growth (cm)	Per cent inhibition	Radial growth (cm)	Per cent inhibition
GT1	0.90	88.75 (70.43)^{ab}	0.83	89.58 (71.19)^{ab}	0.77	90.42 (71.95)^{abc}	0.90	88.75 (70.39)^{b}	0.60	78.80 (62.56)^{a}	0.70	76.67 (61.63)^{b}
GT2	0.70	91.25 (72.95)^{a}	0.63	92.08 (73.64)^{a}	0.50	93.75 (75.54)^{a}	0.63	92.08 (73.64)^{a}	0.50	82.33 (65.12)^{a}	0.53	82.22 (65.05)^{a}
GT3	3.63	54.58 (47.62)^{h}	3.93	50.83 (45.46)^{g}	3.50	56.25 (48.58)^{h}	3.43	57.08 (49.06)^{h}	1.80	36.40 (37.08)^{d}	2.77	7.78 (16.12)^{g}
GT4	2.70	66.25 (54.48)^{d}	2.53	68.33 (55.74)^{e}	2.77	65.42 (53.97)^{e}	2.90	63.75 (52.97)^{g}	1.43	49.38 (44.63)^{b}	2.03	32.22 (34.57)^{e}
GT5	2.70	66.25 (54.48)^{de}	2.70	66.25 (54.48)^{ef}	2.80	65.00 (53.71)^{ef}	2.60	67.50 (55.22)^{ef}	1.67	41.17 (39.90)^{c}	2.13	28.89 (32.47)^{ef}
GT6	2.80	65.00 (53.73)^{def}	1.20	85.00 (67.54)^{bc}	2.87	64.17 (53.23)^{efg}	2.57	67.92 (55.48)^{e}	1.93	31.63 (34.16)^{e}	1.77	41.11 (39.86)^{d}
GT7	0.93	88.33 (70.08)^{abc}	0.83	89.58 (71.25)^{abc}	0.63	92.08 (73.67)^{ab}	0.90	88.75 (70.39)^{bc}	0.63	77.56 (61.73)^{a}	0.90	70.00 (56.79)^{c}
GT8	3.10	61.25 (51.48)^{defg}	1.90	76.25 (60.82)^{d}	1.60	80.00 (63.55)^{d}	0.97	87.92 (69.64)^{bcd}	1.80	36.31 (37.00)^{d}	2.10	30.00 (33.20)^{ef}
Control	8.00	0.00 (0.00)	8.00	0.00 (0.00)	8.00	0.00 (0.00)	8.00	0.00 (0.00)	2.83	0.00 (0.00)	3.00	0.00 (0.00)
C.D (P=0.05)	0.41	5.18 (3.70)	0.41	5.13 (4.02)	0.41	5.06 (3.45)	0.19	2.43 (1.66)	0.11	5.24 (3.24)	0.12	4.15 (2.82)
SE(m)	0.14	1.73 (1.24)	0.14	1.71 (1.34)	0.14	1.69 (1.15)	0.07	0.81 (0.55)	0.04	1.75 (1.08)	0.04	1.39 (0.94)
SE(d)	0.20	2.45 (1.75)	0.19	2.42 (1.90)	0.19	2.39 (1.63)	0.09	1.15 (0.78)	0.05	2.47 (1.53)	0.06	1.96 (1.33)
C.V.	8.47	4.63 (4.06)	9.46	4.32 (4.18)	8.99	4.34 (3.64)	4.41	2.06 (1.74)	4.35	6.29 (4.41)	4.07	5.85 (4.33)

*values in parentheses are angular transformed values GT= Groundnut *Trichoderma*

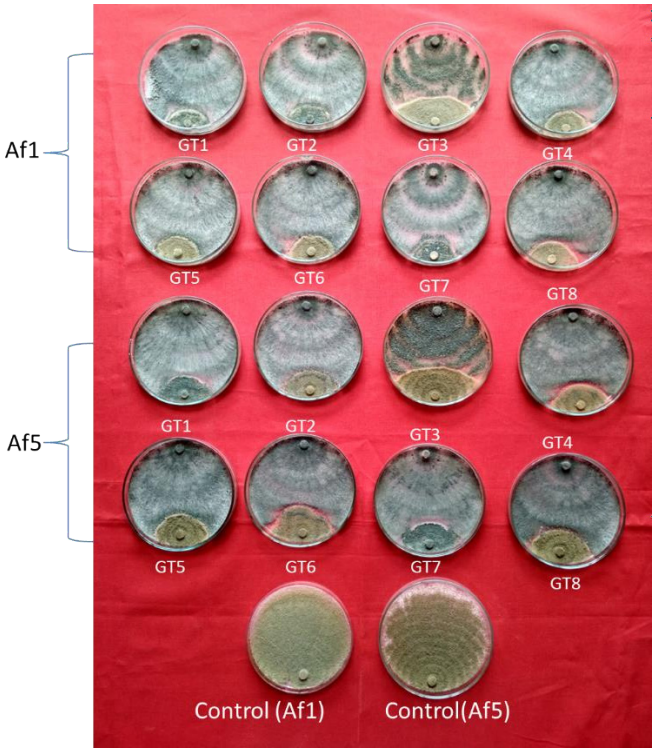

Plate 2. *In vitro* evaluation of *Trichoderma* spp. isolates (GT1 to GT8) against *A. flavus* isolates (Af1 and Af5) by dual culture

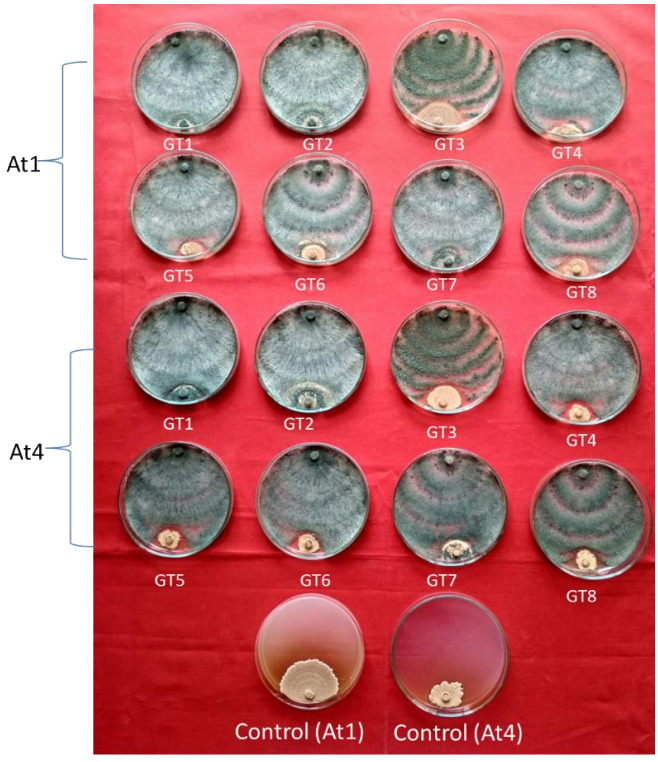

Plate 3. *In vitro* evaluation of *Trichoderma* spp. isolates (GT1 to GT8) against *A. terreus* isolates (At1 and At4) by dual culture

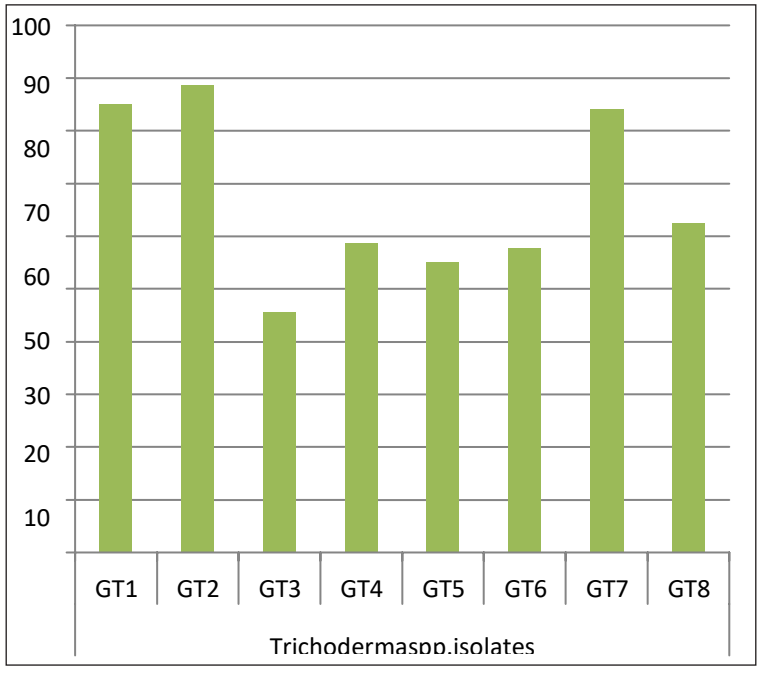

Fig. 3. Mean per cent inhibition by eight *Trichoderma* spp. Isolates

Table 4. Summarized per cent inhibition of collar rot associated pathogens by *Trichoderma* spp.

Name of the pathogen	Name of the isolate	*Trichoderma* spp. isolates								Mean per cent inhibition
		GT1	GT2	GT3	GT4	GT5	GT6	GT7	GT8	
Aspergillus niger	An1	88.75	91.25	54.58	66.25	66.25	65.00	88.33	61.25	72.71
	An8	89.58	92.08	50.83	68.33	66.25	85.00	89.58	76.25	77.24
A. flavus	Af1	90.42	93.75	56.25	65.42	65.00	64.17	92.08	80.00	75.89
	Af5	88.75	92.08	57.08	63.75	67.50	67.92	88.75	87.92	76.72
A. terreus	At1	78.80	82.33	36.40	49.38	41.17	31.63	77.56	36.31	54.20
	At4	76.667	82.22	7.78	32.22	28.89	41.11	70.00	30.00	46.11
Mean per cent inhibition		85.17	88.64	45.59	58.65	55.09	57.66	84.20	62.55	

GT= Groundnut *Trichoderma*

On comparing the overall per cent inhibition of *Trichoderma* spp. isolates on *Aspergillus* spp. isolates, the potential *Trichoderma* spp. isolate from Chittoor district GT1 exhibted 85.17 % inhibition, GT7 from Y.S.R. Kadapa district showed 84.20 % inhibition and GT2 from Anantapuramu district was highly effective (88.64 %) (Table 4) and was selected for further investigation. Similar results were obtained by Kumari and Singh (2017) who studied the *in vitro* antagonistic effect of two *Trichoderma* strains (*T. harzianum* and *T. viride*) against *A. niger* and observed that *T. harzianum* inhibited maximum (81.66 %) growth of test fungus, followed by *T. viride* (66.94 %). While Prajapati *et al.* (2017) evaluated antagonistic potential of *Trichoderma* spp. *viz., T. harzianum, T. asperellum, T. atroviride, T. fasciculatum*, *T. viride* and *T. virens* against *A. niger* by dual culture and recorded that highest mycelial growth inhibition was recorded in *T. asperellum* (78.89 %) followed by *T. harzianum* (74.44 %) and *T. viride* (71.86 %). The findings are also in confirmation with Gaikwad *et al.* (2018) who screened *T. harzianum* against soilborne pathogens of groundnut *viz., A. niger, A. flavus, Sclerotium rolfsii, Fusarium roseum, Macrophomina phaseolina, Pythium myriotylum* and *Rhizoctonia solani and* observed that inhibition of *A. flavus* and *A. niger* was 50.37 and 46.55 per cent respectively. Whereas, Shaheen (2018) studied the antagonistic effect of seven isolates of three *Trichoderma* spp. (*T. virens, T. harzianum* and *T. viride*) against

Aspergillus niger and observed that *T. viride* 60 inhibited maximum (86.20 %) growth of test fungus followed by *T. harzianum* 2A (80.40 %).

CONCLUSION

Prevalence of groundnut collar rot incidence in all the surveyed areas was documented and highest collar rot incidence was recorded in Anathapuramu district. Among all the villages surveyed, Kadiri- 6 (K-6) was predominantly cultivated in all the areas Per cent disease incidence varied based on the soil type, cropping pattern, farming situation and variety cultivated in surveyed areas. Among the eight *Trichoderma* spp. isolates tested in dual culture, the potential *Trichoderma* spp. isolate was from Anantapuramu district GT2, which exhibited highest per cent inhibition of 88.64 % followed by GT1 from Chittoor district (85.17 %) and GT7 from Y.S.R. Kadapa district (84.20 %) on all *Aspergillus* spp. (Fig. 1). Future research in this area should include *in vivo* studies on the effectiveness of the *Trichoderma* spp. as biocontrol agents.

REFERENCES

Agarwal VK, Sinclair JB. 1987. Principles of Seed Pathology. Vol. I (p.1-176) and Vol. II (p.1-168). CRC Press, Inc, Boca, Raton, Florida, U.S.A. Available from: https://www.perlego.com/book/1498547/principles-of-seed-pathology-1987- volume-ii-pdf

Amin VKF, Razdan FA, Bhat MKA, Banday S. 2010. Potential of *Trichoderma* species as biocontrol agents of soilborne fungal propagules. *J. Phytol.* **2**(10): 38-41. Available from: https://www.cabdirect.org/cabdirect/abstract/20113064341

Aycock R. 1966. Stem rots and other disease caused by *Sclerotium rolfsii*, North Carolina, *Agric. Exp. Stn. Techn. Bull. no*. 174 p. 202. Available from: https://library.wur.nl/WebQuery/titel/446069

Barnett HL, Hunter B. 1972. Illustrated genera of imperfect fungi, Burgess publishing company, Minnesota. Available from: https://www.cabdirect.org/cabdirect/abstract/19721103191

Cook RJ, Baker KF. 1983. The nature and practice of biological control of plant pathogens. St. Paul Minnesota. *Am. Phytopathology. Soc.* 539. Available from: https://www.scirp.org/(S(czeh2tfqyw2orz553k1w0r45))/reference/ReferencesPa pers.aspx?ReferenceID=1426342

Dennis C, Webster J. 1971. Antagonistic properties of species group of *Trichoderma*. I. Production of non-volatile antibiotics. *Trans. Brit. Mycol. Soc.* **57**: 25-39. Available from: https://www.sciencedirect.com/science/article/abs/pii/S0007153671800773 https://doi.org/10.1016/S0007-1536(71)80077-3

Devi MC, Prasad RD. 2009. Biointensive management of collar rot of groundnut caused by *Aspergillus niger. J Biol. Control,* **23**(1): 21-24. Available from: http://www.informaticsjournals.com/index.php/jbc/article/view/3610/2696

Dhotre, Vanmare. 2017. Antagonistic activity of *Trichoderma* against root rot causing fungi of soybean. *Int. J Adv. Sci. Res.,* **2**(2): 57-58. Available from: http://www.allscientificjournal.com/archives/2017/vol2/issue2/2-2-30

Gaikwad SN, Salve SN, Rajurkar SK. 2018. In vitro antagonistic activity of *Trichoderma harzianum* against soilborne fungal pathogens. *Int. J Biol. Res.* **3**(2): 87-89. Available from: http://www.biologyjournal.in/archives/2018/vol3/issue2/3-2-20

Gajera HP, Katakpara ZA, Patel SV, Golakiya BA. 2016. Antioxidant defense response induced by *Trichoderma viride* against *Aspergillus niger* Van Tieghem causing collar rot in groundnut (*Arachis hypogaea* L.). *Microb. Pathog.* 26-34. Available from: https://pubmed.ncbi.nlm.nih.gov/26620080/ https://doi.org/10.1016/j.micpath.2015.11.010 PMid:26620080

Gams W, Bissett J. 1998. Morphology and identification of *Trichoderma*. *Appl. Environ. Microbiol*. **65**: 3-34.

Ghewande MP, Desai S, Basu MS. 2002. Diagnosis and management of major diseases of groundnut. *NRCG Bulletin*. 8-9.

ISTA. 1985. International rules for seed testing. *Seed Sci. Technol.,* **13**: 307-500. Available from: https://www.cabdirect.org/cabdirect/abstract/19850778874

Johnson LE, Curl EA. 1972. Methods for research on the ecology of soil borne plant pathogens. Burgess Publishing Company. Minneapolis. 27-35. Available from: https://www.cabdirect.org/cabdirect/abstract/19721103173

Kumari M, Singh M. 2017. Management of collar rot disease of groundnut (*Arachis hypogaea* L.) caused by *Aspergillus niger* through bio-agents. *Int. J Chem. Stud.* **5**(4): 73-76. Available from: https://www.semanticscholar.org/paper/Management-of-collar-rot-disease-of- groundnut-L.)-Kumari-Singh/53ba57f503c53c6779919e83e399a18f382d76ca

Mohammed A, Chala A. 2014. Incidence of Aspergillus contamination of groundnut (*Arachis hypogaea* L.) in Eastern Ethiopia. *African J Clin. Exp. Microbiol.*

Res. **8**(8): 759-765. Available from: https://www.researchgate.net/publication/269675807_Incidence_of_Aspergillus_contamination_of_groundnut_Arachis_hypogaea_L_in_Eastern_Ethiopia https://doi.org/10.5897/AJMR12.2078

Nandeesha BS, Kumar R, Reddy NPE. 2013. Evaluation of different fungicides and their compatibility with Trichoderma spp. for the management of *Aspergillus niger*, incitant of collar rot of groundnut. *Asian J Biol. Life Sci.* **2**(1): 59-63. Available from: https://www.ajbls.com/article/2013/2/1/59-63

Prajapati BK, Patil RK. 2017. Bio-efficacy of *Trichoderma spp*. and its liquid culture filtrate on mycelial growth and management of onion black mould rot (*Aspergillus niger*) in vitro and in vivo. *Indian Phytopathol.* **70**(1): 58-62. Available from: https://www.researchgate.net/publication/315972547_Bio_Efficacy_of_Trichod erma_Spp_And_its_Liquid_Culture_Filtrate_on_Mycelial_Growth_and_Management_of_Onion_Black_Mould_Rot_Aspergillus_niger_in_Vitro_and_in_Vivo https://doi.org/10.24838/ip.2017.v70.i1.48989

Rakholiya KB, Jadeja KB, Parakhia AM. 2012. Management of collar rot of groundnut through seed treatment. *Int. J Life Sci. Pharma Res.,* **2**(1): 62-66. Available from: https://www.ijlpr.com/admin/php/uploads/35_pdf.pdf

Rani AK. 2015. Characterization and rhizosphere competence of *Trichoderma spp*. from different cropping systems in Chittoor dist. M.Sc (Ag.) Thesis submitted to Acharya N G Ranga Agricultural University, Rajendranagar, Hyderabad.

Rani VD, Sudini H, Reddy PN, Devi GU, Kumar KVK. 2016. Survey for the assessment of incidence of stem rot and collar rot diseases of groundnut in major groundnut growing areas of Andhra Pradesh and Telangana states. *Ann. Biol. Res.* **7**(7): 6-8. Available from: https://www.scholarsresearchlibrary.com/articles/survey-for-the-assessment-of-incidence-of-stem-rot-and-collar-rot-diseases-ofgroundnut-in-major-groundnut- growing-areas-.pdf

Rao VVR, Reddy SM, Shantaram MV, Rao CK. 1997. Survey on the occurrence of seedling disease of groundnut (*Arachis hypogaea* L.). *Int. J Plant Prot.* **25**(1): 9-11.

Rasheed S, Dawar S, Ghaffar A, Shaukat SS. 2004. Seed borne mycoflora of groundnut. *Pak. J Bot.* **36**(1): 199-202. Available from: https://www.cabdirect.org/cabdirect/abstract/20043151575

Shaheen SR. 2018. Management of collar rot of groundnut (*Arachis hypogaea* L.) by biocontrol agents. *Int. Conf. Front. Life Earth Sci.,***5**(1): 173-176.

Singh SK, Jadon KS. 2019. Biocontrol efficacy of Trichoderma viride against fungal pathogens of cumin, groundnut and castor. *Indian Phytopathol.* **72**(3): 537-543. Available from: https://www.cabdirect.org/cabdirect/abstract/20203407989 https://doi.org/10.1007/s42360-019-00156-3