RESEARCH PAPER

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Effect of plant growth promoting fungal inoculant on the growth of *Arachis hypogea* (L.) and it's role on the induction of systemic resistance against *Rhizoctonia solani*

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Beneficial plant microbe interactions in the rhizosphere are primary determinants of plant health and soil fertility. Plant growth promoting fungi have great effect towards the growth of plant crops. Soil borne pathogenic fungi cause heavy crop losses all over the world. As the use of chemicals for disease control and fertilization causes environmental problems, there is a need for alternative control measures. The most important and economically cultivated plant pea nut was selected to test the growth promotion by antagonistic microorganisms with or without pathogen *Rhizoctonia solani* . This necessitates a study on plant growth promoting fungi (PGPF) as adequate work has gone on rhizobacteria. The present investigation was made to study the effect of PGPF on the growth of *Arachis hypogea* (L.) and its role of induction of systemic resistance against *Rhizoctonia solani.* Forty five rhizosphere fungal isolates were obtained from 12 different cultivated field crops and were screened for their potential to promote growth in *Arachis hypogea* (L.). The isolate (Cc_2) obtained from *Cucumis sotivus* (L.). Duch.ex. poir was identified as the potential plant growth promoting fungus. The effect of soil inoculation of the selected isolate Cc_2 on the growth of healthy plants of *Arachis hypogea* (L.) and those challenged with *Rhizoctonia solani* was studied by pot culture experiment. The effect was studied in terms of morphological and biochemical parameters. The overall vegetative growth of plant (root and shoot development, dry matter accumulation) and Reproductive growth (pod and seed development). The fungal inoculants improved the growth very effectively both in plants challenged and unchallenged with *Rhizoctonia solani.* The soil inoculation of Cc_2 has improved the chlorophyll content, caroteniod content, anthocyanins content, total soluble sugar content, protein content compare to the untreated plants (T_0) and plants infected by *Rhizoctonia solani* (T_1). The phenol and proline contents were found to be more in plants challenged with *Rhizoctonia solani.* The ability of the selected isolate to produce growth hormones was determined. Results revealed that the selected isolate could produce indole acetic acid and gibberellic acid. The *in vitro* study by dual culture method revealed that there was a negative interaction (Antibiosis) between the plant growth promoting fungal inoculant (Cc_2) and the pathogen *Rhizoctonia solani.* It could be concluded that the selected isolate Cc_2 proved to be a potential fungus in promoting plant growth and yield in *Arachis hypogea* (L.) and in inducing systemic resistance in *Arachis hypogea* (L.) against *Rhizoctonia solani.* Finally Cc_2 was identified as *Rhizopus* sp. in generic level.

Key words : Plant growth promoting fungi (PGPF), Phytopathogens, Caroteniod, Antagonistic, Rhizobacteria.

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INTRODUCTION

Environmental concerns have led to the need for sustainable use of natural resources. The conventional agriculture has caused considerable impacts on soils and water. It is important to change certain agricultural managements to environmental cleaner techniques. The sustainable agriculture has pointed many approaches and techniques to reduce environmental impact. One of these strategies is the utilization of soil micro biota for the promotion of plant growth and control of plant diseases..

Plant diseases play a direct role in the destruction of natural resources in agriculture. In particular, soil borne pathogens cause greater losses, fungi being more aggressive. The distribution of several pathogenic fungi such as *Pythium, Phytophthora, Botrytis*, *Rhizoctonia* and *Fusarium* has spread during the last few years due to changes introduced in farming with determined effects on crops of economic importance. (Chet *et al*., 1997). *Rhizoctonia solani* can cause significant plant damage, impaired plant growth and crop failure. It can affect plant emergence and cause stem canker symptoms characterized by brown and black sunken lesions on the stems. Misshapen tubers with an uneven size distribution also result .It can cause diseases such as root rot, stem rot, seed decay, damping off and foliar blight in crops. Groundnut is a rich source of protein and it is one of the principle oil seed crops of India. Peanut was grown mainly for its edible oil. *Arachis hypogea* (L.) has a mutually beneficial symbiotic relationship with nitrogen fixing bacteria. Efforts have been continually been concentrated towards increasing its yield. The potential of *Trichoderma* against plant diseases was recognized in the control of *Corticum solani* (Aluko and Hering , 1970), Damping of pea (Lifshitz *et al.*, 1986) *Pythium ultimum* (Migheli *et al*., 1998), Fusarium wilt (Zhang *et al.*, 1996) and *Botrytis cinerea* (Elad and Kapat, 1999). Simon (1989) said that plant growth promoting microbial inoculants play an important role in maintaining sustainable agricultural production. Hormones are naturally synthesized naturally organic compounds which influence the growth and development. Auxins are naturally occurring substances with indole possessing growth promoting activity. Auxin accumulation may be attributed to rapid synthesis by the infected plant, synthesis by the pathogen and operation of new pathway of IAA synthesis such as release of IAA from IAA protein complex (Mahadevan, 1984). Gibberellins induce elongation of internodes and the growth of meristems (or) buds. They usually inhibit adventitious root as well as shoot formation. The fungus produces many types of gibberellins in the culture media. As the exploitation of microbial metabolites has gained status of a new trust area in plant protection and growth promotion, the present investigation was undertaken to study the effect of plant growth promoting fungi on the growth of *Arachis hypogea* (L.) and its role on the induction of systemic resistance against *Rhizoctonia solani.* Aim of the present study to evaluate plant growth promotion using beneficial microorganism for their ability to promote growth of ground nut plants.

RESEARCH METHODOLOGY

Isolation and maintenance of the pathogen:

The present study was carried out with *Rhizoctonia solani* isolated from the infected plants of ground nut collected from in and around Madurai. 20 ml of sterile and warmed PDA media were poured into sterile Petri plate and allowed to solidify. The fungi culture were inoculated in the centre of the Petri plate by placing a 10 mm disc of 7 days old PDA culture. The plate were incubated at room temperature (28^0C) for four days.

Mass multiplication of pathogen:

The inoculum of *R. solani* was developed by inoculating the pathogens on PDB medium and incubated at room temperature (28^0C) for 10 days. The pathogen was thoroughly mixed.

Isolation of beneficial fungi:

Soil samples were collected from the rhizosphere of twelve different cultivated field crops the plants are given below:

Crops selected for sample collection
- *Abelmoschus esculentus* (L.) Moeneh.
- *Allium cepa* (L.)
- *Cucumis sotivus* (L.) Dueh.ex.poir
- *Cyamposis tetragonoloba* (L.) Taub.
- *Eleusine coracana* (L.) Gaertn.
- *Gossypuim* sp.
- *Helianthus annuus* (L.)
- *Murraya koenigii* (L.)
- *Musa paradisiacal* (L.)
- *Pennisetum amerieanum* (L.) Schum
- *Polianthes tuberose* (L.)
- *Saccharum officinarum*
- *Solanum melongena* (L.)

Isolation of fungi from the collected soil samples was done by serial dilution technique. 1g of soil sample was suspended in 9ml of sterile distilled water and 10-fold dilutions were prepared up to 10^{-3}. 1ml of diluted suspension was inoculated into each sterile Petri plate and 20ml of Czepekdox agar was poured. The plates were rotated for uniform distribution and were incubated at room temperature for 4 days. The fungi developed on the medium were then isolated, pure cultured and maintained in Czepekdox agar slants. 45 fungal isolates were obtained and were designed by numbers as follows:

Source of Rhizosphere fungi	Isolates
Abelmoschus esculentus (L.) Moeneh.	Ca_1,Ca_2,Ca_3
Allium cepa (L.)	Cb_1,Cb_2,Cb_3,Cb_4,Cb_5
Cucumis sotivus (L.) Dueh.ex.poir	Cc_1,Cc_2,Cc_3,Cc_4
Cyamposis tetragonoloba (L.) Taub.	Cd_1,Cd_2,Cd_3,Cd_4,Cd_5
Eleusine coracana (L.) Gaertn.	Ce_1,Ce_2
Gossypuim sp.,	Cf_1,Cf_2,Cf_3,Cf_4
Helianthus annuus (L.)	Cg_1,Cg_2,Cg_3
Murraya koenigii (L.)	Ch_1,Ch_2,Ch_3,Ch_4
Musa paradisiacal (L.)	Ci_1,Ci_2,Ci_3
Pennisetum amerieanum (L.) *Schum*	Cj_1,Cj_2,Cj_3
Polianthes tuberose (L.)	Ck_1,Ck_2,Ck_3
Saccharum officinarum (L.)	C_{l1},C_{l2},C_{l3}
Solanum melongena (L.)	Cm_1,Cm_2,Cm_3

Screening for potential plant growth promoting fungus:

Small plastic containers (12×8cm) provided with drainage hole were filled with steam sterilized soil mixture containing garden soil and sand (3:1). Soils in each container was inoculated separately with three 1mm agar disc containing the mycelium of all the 45 isolates and sprinkled with sterile water and then covered with plastic sheet which was punctured with a pin to promote aeration. The inoculants were allowed to establish for a week. The plastic sheet was removed and the soil was thoroughly mixed with the inoculum. Healthy seeds of *Arachis hypogea* (L.) were soaked in water for 2hours and sown (3seeds/container). The soil was watered regularly and the effect of the fungal inoculants on the growth and the seedling was determined after 15days in terms of seedling height and number of roots.

Identification of potential fungal isolate:

The colony characteristics on Czepeckdox agar were observed. The hyphae were stained with lacto phenol cotton blue stain and microscopically analyzed for their characteristics. The isolate was identified to the generic level using the Manual of soil fungi .

Mass multiplication of beneficial microorganism:

The inoculum of *Rhizoctonia solani* was developed by inoculating the on beneficial microorganism PDB medium and incubated at room temperature (28°C) for 15 days. The beneficial microorganism was thoroughly mixed and the inoculum was used for the seed treatment experiments.

In vivo study on the effect of plant growth promoting fungal inoculant on the growth of healthy seeds of *Arachis hypogea* (L.) and that chanagelled with pathogen *Rhizoctonia solani*

A pot culture study was made to determine the effect of selected plant growth promoting fungal inoculant (Cc_2) on the growth of healthy and infested plants of *Arachis hypogea* (L.) and those challenged with *Rhizoctonia solani.* Earthen pots (21×20cm) were filled with steam sterilized soil mixture containing garden soil and sand in the ratio 3:1.

Treatment schedule:

T_0 - Control (sterile soil only)

T_1 - Pathogen (*Rhizoctonia solani*)

T_2 - Plant growth promoting fungal inoculant (Cc_2)

T_3 - Pathogen (*Rhizoctonia solani*) + Plant growth promoting fungal inoculant (Cc_2)

The pots of T_1 and T_3 were inoculated each of three 5mm agar discs containing mycelium of *Rhizoctonia solani* and allowed to established for a week followed by inoculation of pots of T_2,T_3 treatments with plant growth promoting fungal inoculant Cc_2 (three 5mm discs of inoculum/pot). Sterile water was sprinkled after the inoculation and the pots were covered with polyethylene sheets to avoid the entry of air borne pathogens. The inoculated organisms were allowed to infest the soil for a week. The polyethylene sheets were removed and the thoroughly mixed with the inoculum. Certified seeds of *Arachis hypogea* (L.) were soaked overnight in sterile distilled water and seeds were sown in the pots (6 seeds / pot). Regular watering was done throughout the investigation. Duplicates were maintained for all the treatments. The plants were uprooted 60 days after germination and the following parameters were analyzed.

Morphometric analysis:

- Number of roots
- Root length
- Shoot length
- Number of leaves
- Number of branches
- Number of pods/plant
- Leaf area
- Fresh weight of the pods/plant
- Dry weight of the plant

Determination of dry matter yield:

The plants from each treatment were cut into bits and kept in an oven at 100°C for 2 days. The dried sample was weighed and the dry matter yield was determined.

Biochemical analysis

Estimation of chlorophyll:

The chlorophyll content was estimated by Arnon's (1949) method. 100mg leaf sample was homogenized with 10 ml of 80% prechilled acetone. The extract was centrifuged at 3000rpm for 10 minutes. The supernatant was collected and absorbance was read at 645 nm and 663 nm. Total chlorophyll content was measured by using the formula, total chl = 20.2 (A645) + 8.02 (A 663) x V/ 1000x W x10 (where, A - OD at specific nm, V- Final volume of plant extract in 80% acetone, W - Fresh weight of leaf tissue used).

Estimation of anthocyanins:

The estimation of anthocyanins content was determined by Swain and Hillis (1959). 100 mg fresh leaves were taken and ground with 10 ml of ethanol and filtered through what man No.1 filter paper. One ml of extract along with methanolic HCl was added to 1 ml of peroxide reagent and kept in dark for 15 minutes and the absorbance was read at 525nm.Anthocyanin content was represented by O.D value (A525)/gram of leaf tissue.

Estimation of protein:

The protein content was estimated by the procedure described by Lowry *et al*. (1951). One gram of leaf sample was ground with 5 ml of phosphate buffer (pH 7) and centrifuged at 3000 rpm for 20 minutes. 3 ml of the extract was taken with 3 ml of 20%TCA and kept in water bath for 20 minutes and again it was centrifuged at 3000rpm for 20 minutes. Pellet was collected and washed with 6ml of acetone and centrifuged. Pellet was dissolved in 5 ml of 0.1 NaOH and mixed well. It was kept for 10 minutes and 0.5 ml of folin was added. It was incubated for 30 minutes in dark and the absorbance was read at 660nm. The amount of protein was estimated using Bovine serum albumin as the standard.

Estimation of total soluble sugars:

The amount of total soluble sugar present in the leaf extract was estimated by Anthrone method (Dubois *et al.,* 1951). Leaf sample (100 mg) was ground with 10 ml of distilled water and added with 2 ml of 10%Trichloro acetic acid and centrifuged at 3000rpm for 5 minutes. Four ml of Anthrone reagent was added to 1 ml of supernatant .The test tubes were kept in boiling water bath after covering their mouth with glass marbles for 10 minutes. The content was then cooled and the absorbance was read at 625nm. The amount of sugar was estimated using glucose as the standard.

Estimation of proline:

Free proline from plant tissues may be selectively extracted in aqueous Sulphosalicylic acid and its concentration was measured using ninhydrin method. 200mg of leaf sample was taken and ground with 10 ml sulphosalicylic acid and filtered with What man 1 filter paper. 2ml of the extract along with 2ml acid ninhydrin and 2 ml of glacial acetic acid was taken, mixed well and kept in boiling water bath (100°c) for one hour. It was cooled in an ice for 5 minutes and added with 4 ml of toluene .The tubes were agitated vigorously. The upper pink chromophore layer was separated and the absorbance was read at 520 nm. The amount of proline was estimated using proline as the standard.

Estimation of phenol:

The phenolic content was estimated by folin-ciocalteu method .100 mg fresh leaves were taken and ground with 10 ml of ethanol and filtered through what man 1 filter paper. One ml of extract was added with 2 ml of 20% sodium carbonate. It was shaken well and kept in boiling water bath for 1 minute and cooled. The blue solution obtained was diluted to 25 ml with water and the absorbance was read at 650nm. The amount of phenol was estimated using catachol as the standard.

Estimation of plant hormones:

The healthy seeds of *Arachis hypogea* (L.) was soaked overnight in 1week, 2weeks, and 3weeks old culture filtrate (C_1, C_2, and C_3) of selected fungal inoculant (Cc_2) and sown in the small containers containing sterile soil. The water soaked seeds served as the control. Regular watering was done and the growth of seedling was determined. The culture filtrate that revealed the enhanced growth of seedling was selected to determine the production of auxins and gibberellins.

Estimation of indole acetic acid:

The amount of IAA produced in the culture medium was estimated by the method described by Mandal *et al.* (2007). Two weeks old culture of Cc_2 was centrifuged at 1000g for 10 minutes. To 1ml of cell free culture filtrate, 2ml of Salknowsky reagent was mixed and incubated at room temperature for 30minutes. The absorbance was read at 530nm. The commercial indole acetic acid was used as the standard.

Estimation of gibberllic acid:

The amount of GA produced by Cc_2 was estimated by the method described by Mahadevan (1984). 15ml of

cell free culture filtrate was taken in a test tube and mixed with 2ml of zinc acetate solution. After 2 minutes, 2ml of potassium ferro cyanide was added and centrifuged at 2000g for 15 minutes. To 5 ml of supernatant, 5ml of 30% HCl was added and incubated at 20^{0Z}C for 75 minutes. The absorbance was read at 254nm. The commercial gibberllic acid was used as the standard.

***In vitro* study on the interaction between plant growth promoting fungal inoculant (cc_2) and *Rhizoctonia solani*:**

The interaction between the selected plant growth promoting fungal inoculant and *Rhizoctonia solani* was tested by dual culture method suggested by Morten and Stroube (1995) using Czepeckdox agar medium. 1mm agar discs of the 2 fungi cut separately using a sterile cork borer from the edges of 5days old culture were kept apart on Czepeckdox agar medium and the plates were incubated at $35^0\pm 2^0$ C for 5days. The plate inoculated with *Rhizoctonia solani* alone served as the control. The growth of *Rhizoctonia solani* in dual culture was compared with that of the control plate.

RESULTS AND ANALYSIS

The effect of plant growth promoting fungal (PGPF) inoculant and its role on the induction of systemic resistance against *Rhizoctonia solani* was studied by *in vivo* studies using *Arachis hypogea* (L.).

Isolation and screening of rhizosphere fungi for the plant growth promotion in *Arachis hypogea* (L.).

About 45 fungal isolates from soil were collected from the rhizosphere of 12 different cultivated field crops. A preliminary study was made to evaluate their effect on seedling growth of *Arachis hypogea* (L.). The result presented in (Table 1 A and B) revealed that 10 among the 45 isolates tested showed their ability to promote plant growth (Cb_2, Cc_2, Cd_2, Cf_1, Cf_2, Cg_1, Ch_1, Ch_3, and Ck_1). The soil inoculation of 16 isolates resulted in reduced plant growth (Ca_2,Cb_1,Cb_4,Cb_5,Cc_1,Cc_4,Cd_3,Cd_4,Cg_5, Cf_4, Cg_2,Cg_3,Ch_4,Ch_5,Cj_1 and Ck_3) and 19 of the isolates inhibited seed germination. Among the 10 plant growth promoting fungal isolates, maximum growth was observed in *Arachis hypogea* (L.) grown in soil inoculated with the isolate Cc_2 isolated from rhizosphere soil of *Cucumis stavis*. Hence, Cc_2 was selected as the potential fungus for further study.

Identification of selected isolate Cc_2:

The colony of isolate Cc_2 showed pre fused cottony growth of white mycelium which filled the entire culture plate. Using the above mentioned characteristics the fungal isolate Cc_2 was identified to the generic level as *Rhizopus*. The vegetative hyphae are aseptate and branched with an abundant production of sporangiophores bearing sporangia at their ends. Sporangiospores are spherical, unicellular and hyaline.

***In vivo* study on the effect of plant growth promoting fungal inoculant on the growth of healthy seeds of *Arachis hypogea* (L.) and that chanagelled with pathogen *Rhizoctonia solani*:**

The efficiency of the selected growth promoting fungal inoculant (Cc_2) reflected on the growth and biochemical characteristics of *Arachis hypogea* (L.) was studied in healthy plants and those challenged with *Rhizoctonia solani* by pot experiment. The growth parameters were determined in terms of number of roots, root length, shoot length, number of leaves, number of

Table 1(A): Effect on seedling growth of *Arachis hypogea* (L.)

Treatments	No. of roots	Root length (cm)	Fresh weight of the root (g)	Dry weight of the root (g)
T_0	12± 2	22.33± 0.76	0.72± 0.1	0.37± 0.1
T_1	7.33± 2	14.33 ± 0.57	0.14± 0.02	0.06± 0.03
T_2	25.66± 2	41.66± 0.4	1.56 ±0.37	0.24± 0.14
T_3	14.33 ±2	21.33 ±1.55	1.01 ±0.17	0.46± 0.03

Table 1(B): Effect on seedling growth of *Arachis hypogea* (L.)

Treatments	No. of leaves	Shoot length (cm)	Fresh weight of the shoot (g)	Dry weight of the shoot (g)	No. of branches
T_0	8.33 ±1.15	5.83 ±0.57	1.89 ±0.03	0.06± 0.1	1.33± 0.57
T_1	4.33± 0.57	3.33± 0.57	0.72± 0.27	0.31 ±0.04	0.33± 0.57
T_2	26.33± 3.51	13.66± 1.52	11.79 ±1.4	3.06± 0.04	5.66± 0.57
T_3	13.66± 3.1	8 ±1	3.69 ±0.97	1.19± 0.11	3.66 ±0.57

branches, number of pods/plant, leaf area, dry matter yield and fresh weight of the pod. Soil inoculation of plant growth promoting fungal (PGPF) inoculant (T_2) was found to enhance root development as well as shoot growth. There was an increase in the number of roots (108.3%), root length (86.57%), shoot height (134.3%) in T_2 over the control (T_0). There was a triple fold increase in the number of leaves in T_2. There was greater reduction in number of roots (41.6%), root length (36.4%) and shoot height (42.9%) in pathogen infested plants (T1) grown in the soil without PGPF inoculant. There was triple fold increase in the number of leaves in T_3 then T_1. The soil inoculation of PGPF inoculant enornomously increased the shoot growth which made the plant more bushy than the other plants (Plate 1). There was a double fold increase in the leaf area due to soil inoculation with Cc_2 (Plate 2). The development of pods and seeds was found to be improved in T_2 than in the untreated plants (Plate: 3, Table 2). There was a greater inhibition of pod development in plants infected by *Rhizoctonia solani* (T_1). This might be due to unhealthy roots affected by root rot. The overall growth of the plant was also found to be greatly affected (Plate: 4). The pod size was found to be improved in T_3 compared to infected plants (T_1) which might be due to the suppression of *Rhizoctonia solani* by the selected fungal isolate. The dry matter accumulation was found to be greater in plants grown in soil inoculated with Cc_2. The increase was found to be 3 fold over the control (T_0). The infested plants in T_1 showed poor dry matter yield compared to all other treatments. Soil inoculation with Cc2 had improved the dry matter yield in infected plants (T_3).

Effect of soil inoculation with PGPF inoculant (Cc2) on the selected biochemical parameters:

The data collected for the selected biochemical parameters are presented in (Table 3).

Chlorophyll content:

The plants of T_2 recorded maximum chlorophyll content than plants in other treatments (T_0, T_1, and T_3). The chlorophyll content was found to be minimum in *Rhizoctonia solani* infected plants (T_1) and the reduction was found to be 31.5% over the control (T_0). The presence of fungal inoculant in soil was found to improve the chlorophyll content in plants infected by *Rhizoctonia solani* (T_3) with an increase of 36.4% over T_1.

Carotenoids content:

The fungal inoculant was found to increase

Plate 1: Macroscopic comparison of the result

Plate 2: Leaf area

Plate 3: Pods

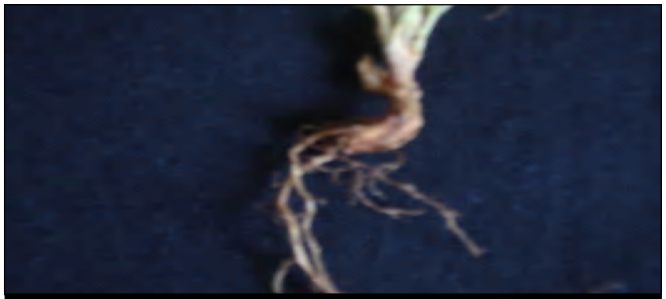

Plate 4: *Rhizoctonia solani* infected root

Table 2: Development of pods and seeds

Treatments	Leaf area (cm^2)	No. of pods / plant	Fresh weight of the pod (g)	Dry weight of the pod (g)
T_0	6.5 ±0.5	4.66 ±0.57	0.29 ± 0.25	0.2 ± 0.11
T_1	3.5 ±0.5	-	-	-
T_2	20.5 ±0.5	8.33 ± 1.52	2.83 ± 0.25	1.2 ±0.61
T_3	11 ± 1	5.66 ±0.57	2.05 ± 0.03	0.39 ± 0.25

Table 3: Effect of soil inoculation with PGPF inoculant (Cc2) on the selected biochemical parameters

Treatments	Chlorophyll (mg/g) 663 & 645 nm	Carotenoid (mg/g) 480nm	Anthocyanin (mg/g) 525 nm	Protein (mg/g) 660 nm	Proline (mg/g) 520 nm	Phenol (mg/g) 650 nm	Soluble Sugars (mg/g) 625 nm
T_0	0.85± 0.006	0.21 ±0.007	6.65 ± 0.35	76.89± 0.42	23.87± 1.08	22.1 ±0.31	35.94 ±0.03
T_1	0.634± 0.199	0.175 ±0.003	5.3 ± 0.28	104.16	46.77± 0.41	24.08 ±0.13	32.57± 0.07
T_2	1.515± 0.038	0.381 ±0.007	7.3 ± 0.45	205.34 ±0.43	13.84 ± 3.79	22.65± 0.13	39.41 ±0.01
T_3	0.834 ±0.031	0.342 ±0.001	5.95± 0.07	153.26 ±0.63	29.83± 0.40	22.25± 0.09	37.83± 0.24

carotenoids content to 90% over the control in T_2. The carotenoids content was enhanced to 41.2% in infected plants due to the inoculation of PGPF inoculant (T_3).

Anthocyanin content:

The results revealed that the anthocyanin content in infected plants (T_1) was found to be minimum compared to all other treatments. Soil inoculation with the selected isolate Cc_2 had improved the anthocyanin content (22.7% in T_2 and 11.8% in T_3 increase over the control (T_0)).

Sugar content:

The plants infected by *Rhizoctonia solani* (T_1) showed reduction in sugar level compared to all the other treatments. There was a slight increase in the T_3 which contain both the fungal inoculant and the pathogen *Rhizoctonia solani* in the soil. Reduction in the level of sugar in the infected plants might be due to reduction in chlorophyll content which would have reduced the rate of photosynthesis.

Protein content:

The infected plants (T_1) recorded the minimum and the plants grown in soil contained the fungal inoculant showed the maximum protein content, the increase was very significant (300% increase) over the control.

Proline content:

The results recorded for proline content of plants in various treatments is presented in Table 3. It is evident that proline accumulation was higher in plants grown in soil infested by *Rhizoctonia solani* (T_1). The increase was found to be 96.2% over the control (T_0). The soil inoculant Cc_2 reduced proline accumulation to 56.7% though the plants were challenged with *Rhizoctonia solani* (T_3). Proline has been reported as biochemical indicates of resistance. The proline accumulation in *Rhizoctonia solani* infected plants in T_1 might be due to the induction of resistance against infection. Reduction of proline content in T_3 compared to T_1 might be due to minimization of the effect of *Rhizoctonia solani* by the added PGPF inoculant.

Phenol content:

The plants not exposed to the soil inoculant and the pathogen recorded the minimum phenol accumulation. Plants infected by *Rhizoctonia solani* recorded maximum phenol content than all other plants. More accumulation of proline and phenol in infected plants (T_1) might be due to induction of defense mechanisms in plants due to infection. Reduction in these parameters in T_3 might be due to suppression of pathogen due to PGPF inoculant.

Production of hormones by the selected fungal inoculant (Cc_2):

The ability of the selected isolate Cc_2 to produce growth hormones such as auxin and gibberellins in the culture broth was determined. The preliminary study made on the presence of hormones in different culture filtrate revealed that there was adequate production of hormones

by selected fungal isolate grown in Czepeckdox broth for 2weeks which was reflected on the promotion of seedling growth (seedling height) in *Arachis hypogea* (L.). It was found that the isolate could produce indole acetic acid (IAA) and gibberellic acid (GA) in the culture medium at the rate of 2.5mg/l and 4mg/l, respectively. This ability could be attributed to the promotion of growth (vegetative and reproductive) in *Arachis hypogea* (L.). The GA produced by the fungus might be the reason for the enhancement of plant height which might be due to the promotion of longitudinal growth by GA. The growth promotion of rhizobacteria due to auxins and gibberellins had been already recorded. (Basu and Ghosh, 2001, Ghosh and Basu, 2002; Roy and Basu, 2004; Ahmed *et al*., 2005). Gibberellins play an essential role in many aspects of plant growth and development mainly stems elongation and flower development. Greater stem growth in *Fuschia hibrida* and *Phasbitis nil* has also been reported (King *et al.,* 2000).

Interaction between PGPF inoculant and *Rhizoctonia* solani:

The interaction between Cc_2 and *Rhizoctonia solani* was studied by *in vitro* studies by dual culture method. The result clearly revealed that Cc_2 could inhibit (or) suppress the growth of pathogen, *Rhizoctonia solani* very effectively. The antibiosis might be due to antagonistic activity against *Rhizoctonia solani,* production of inhibitory substances by Cc_2 are the direct hyphal interaction.

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