Indian Journal of Fundamental and Applied Life Sciences ISSN: 2231– 6345 (Online)
An Open Access, Online International Journal Available at http://www.cibtech.org/jls.htm
2015 Vol. 5 (1) January-March, pp. 293-302/Doley et al.
Research Article

EFFECTS OF ARBUSCULAR MYCORRHIZAL FUNGI AGAINST *SCLEROTIUM ROLFSII* IN GROUNDNUT (JL-24)

***Doley K.[1], Dudhane M.[2] and Borde M.[3]**
Department of Botany, Savitribai Phule Pune University, Pune, Maharashtra, India, Pune-411007
**Author for Correspondence*

ABSTRACT
Groundnut is considered to be an important oilseed crop worldwide and is susceptible to various pathogens. Among them *Sclerotium rolfsii* is one the most important pathogens which causes stem-rot in groundnut plants. The present investigation was carried out to suppress harmful effects of *S. rolfsii* by the use of arbuscular mycorrhizal fungi (AMF). In the pot culture experiment AM fungi (*G. fasciculatum*) inoculum was applied before sowing the groundnut seeds or pathogen *S. rolfsii*. The incidences of stem-rot were found to be considerably reduced due to pre-application of AM fungi as compared to control ones. Moreover, the percentage of arbuscule formation also got elevated notwithstanding lesser arbuscule formation in presence of pathogen as compared to healthy mycorrhizal ones. The biochemical and antioxidant enzyme activities of phosphatase, protein, total phenols, polyphenol oxidase and peroxidase showed significant results which were correlated to ability of AM fungi in resistance to disease or pathogen attack. These data indicated the role of AM fungi in bringing up combined defense responses.

Keywords: *Antioxidant Enzyme, Biochemical, Defense Response, Pathogen, Stem-rot*

INTRODUCTION
Sclerotium rolfsii (Sacc.) is a soil borne destructive fungi of worldwide significance which has a host range of over 500 species of plants (Susleendra and Schlosser, 1999). The mycelium of *S. rolfsii* causes stem-rot or wilting which is often difficult to control in groundnut plants even with chemical based fungicides. Annually *S. rolfsii* may account for huge losses to groundnut crop production (Bowen *et al.*, 1996). Moreover, *S. rolfsii* was reported to be one of the most destructive pathogen of tropics and subtropics (Mukherjee and Raghu, 1997).

The arbuscular mycorrhizal fungi (AMF) belonging to phylum Glomeromycota (Schubler *et al.*, 2001) colonizes almost 90 % of land plants (Smith and Read, 2008). There are several beneficial functions of this mutualistic association between fungal and plant partner. When this type of association is established manifestation of growth is visible. Also, the AM fungi associations helps host plants in several ways such as improvement in dealing with water and pest (Smith and Read, 1997; Farahani *et al.*, 2008) tolerance towards abiotic stresses (Feng *et al.*, 2008; Arabi *et al.*, 2013), heavy metal stress (Kramer, 2005), mineral acquisition (Azaizeh *et al.*, 1995; Clark and Zeto, 2000), impact on structure of soil (Rillig, 2004), nitrogen fixation (Haystead and Grove, 1988) and protection against fungal plant pathogens (Wu *et al.*, 2013, (Harrier and Watson, 2004). Hence, the symbiotic associations of AM bring about significant potential quality to life as a whole in our natural and agricultural ecosystems (Smith and Read, 1997).

The present pot culture investigation was undertaken to determine various effects in plant against pathogen infection by inoculating AM fungi in autoclaved soil.

MATERIALS AND METHODS
Biological Materials
The seeds of local susceptible var. Phule Pragati (JL 24) were obtained from Naik Seeds Limited, Pune, Maharashtra, India. The AM fungi *Glomus fasciculatum* (Thaxter Sensu Gerd.) was isolated as per Gerdemann and Nicolson (1963) and identified as per Trappe (1982) manual. Mycorrhizal inoculums were prepared in an open pot culture of *Sorghum vulgare* plants which consisted of spores, colonized root pieces and soil. Twenty grams of mycorrhizal inoculums were placed below groundnut seeds at the time of sowing. The pathogen *S. rolfsii* was isolated from the fields of Pune, Maharashtra, India and identified

through the Division of Mycology, Agharkar Research Institute, Pune, India. Pathogen inoculum was prepared by sterilizing sorghum seeds and inoculating it with pure culture of *S. rolfsii* in conical flasks which were incubated for three weeks. The grains served as pathogen inoculum and were applied after fifteen days of plant growth at the rate of five grams.

Growth Conditions and Experimental Design

Plants were grown in greenhouse condition in pots. Plants were watered at regular interval of times with no addition of any kind of other chemicals. The experiment consisted of randomized complete design block (RCBD) in triplicates and consisted of four treatments as follows: [1] Uninoculated Control (C); [2] Control + *S. rolfsii* (C+Sr); [3] *G. fasiculatum* inoculated (Gf); [4] *G. fasiculatum* + *S. rolfsii* inoculated (Gf+Sr).

Parameters Measured

Disease Incidence

For incidences of disease above ground stem-rots were observed weekly as per Kokalis (1992).

Arbuscule Percentage

Randomly selected root samples were cleared in 10 % KOH at 90°C for 1 hour and stained in 0.01 % trypan blue according to Phillips and Hayman (1970) for 10 minutes. The fungal structures were visualized under a compound microscope and the measurements of arbuscule percentage by *G. fasciculatum* were determined by Trouvelot *et al.*, (1986).

Biochemical Parameters

Acid and alkaline activities in roots were determined by p-nitro phenol released at 405 nm in spectrophotometer described by Lowry *et al.*, (1954). Protein content in roots were determined by Folin-Ciocalteau reagent which was read at 660 nm described by Lowry *et al.*, (1951). Total phenols in roots were determined as per Malick and Singh, (1980) by using Folin reagent and reading optical density at 650 nm. Polyphenol oxidase activities in root were assayed by using catechol and optical density were read at 495 nm as per Mahadevan and Shridhar (1982). Peroxidase activities in roots were determined by monitoring guiacol and hydrogen peroxidase at 436 nm as described by Putter (1974).

Statistical Analysis

Data's from the greenhouse experiments were subjected to analysis of variance (ANOVA) followed by Duncan's multiple range test (DMRT). Differences at P=0.05 were considered significant. The values are expressed as mean ± SD. All the calculations were made by using a Statistical Package for Social Sciences (SPSS Inc. 1999) for windows version and Microsoft Excel 2007 to analyze the data.

RESULTS AND DISCUSSION

Disease Incidence

The AM fungi associations have been believed to increase the resistance in plants against pathogen attack by induced systemic resistance as compared to improved nutritional requirements of plants (Fritz *et al.*, 2006). The data in Figure 1 showed that the AM fungi (*G. fasciculatum*) inoculation significantly reduced the number of incidences of diseases caused by the *S. rolfsii*. As the non-mycorrhizal groundnut plants in presence of pathogen (C+Sr) demonstrated higher incidences as compared to mycorrhizal diseased ones (Gf+Sr). There were no visible symptoms observed in healthy-mycorrhizal or control ones. Here, the results demonstrated that the AM fungi association must have employed the mechanism of competition with pathogen for access of root zones which led to prevention of pathogen development (Singh and Mukherji, 2006). The data obtained here may be correlated to the presence of arbuscules in diseased mycorrhizal plants (Gf+Sr) even though it demonstrated lower percentage of arbuscules when compared to healthy mycorrhizal ones (Gf).

Arbuscule Formation

The obtained results showed that there was formation of arbuscules in the groundnut roots which confirms colonization by AM fungi (*G. fasiculatum*) was successfully established. The percentage of arbuscules were found to be highest in only healthy mycorrhiza (Gf) inoculated groundnut plant as compared to diseased mycorrhizal ones (Gf+Sr) (Figure 2). The observation of lower arbuscules may be a

result of competition between AM fungi and pathogen. Similar observations of lowering in AM fungi colonization were reported by Aysan and Demir (2009) in bean plants.

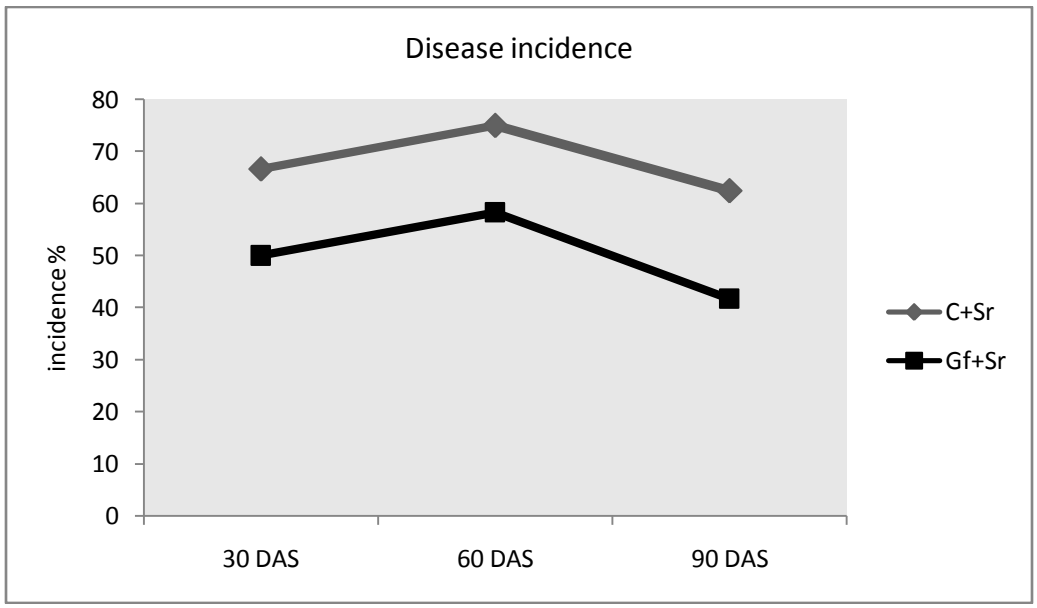

Figure 1: Disease incidences (%) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants
C+Sr: Control + *S. rolfsii*; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated, *n*=3. DAS=Days after sowing

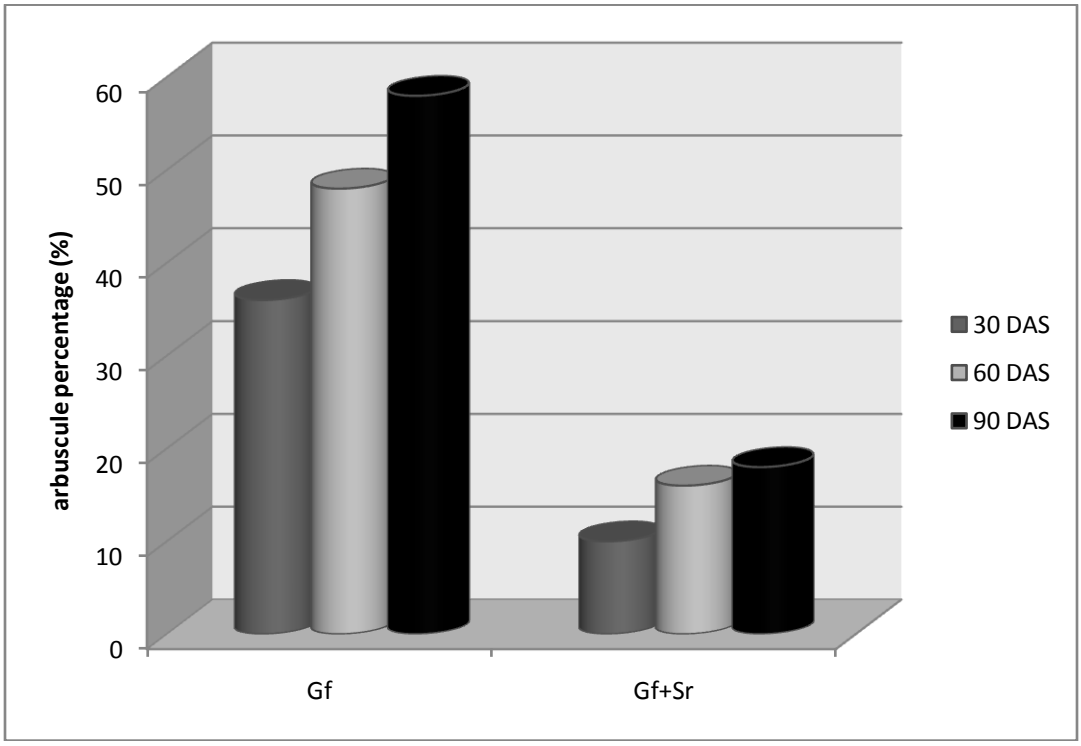

Figure 2: Arbuscule percentage in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants
Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated, *n*=3. DAS=Days after sowing

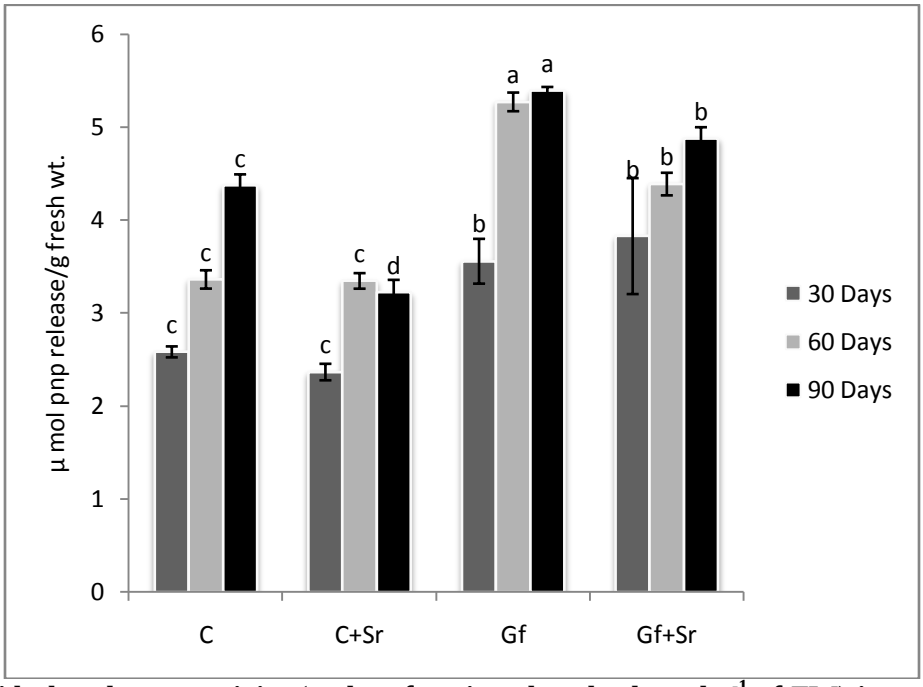

Figure 3: Acid phosphatase activity (moles of p-nitrophenol released g^{-1} of FW) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants
C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

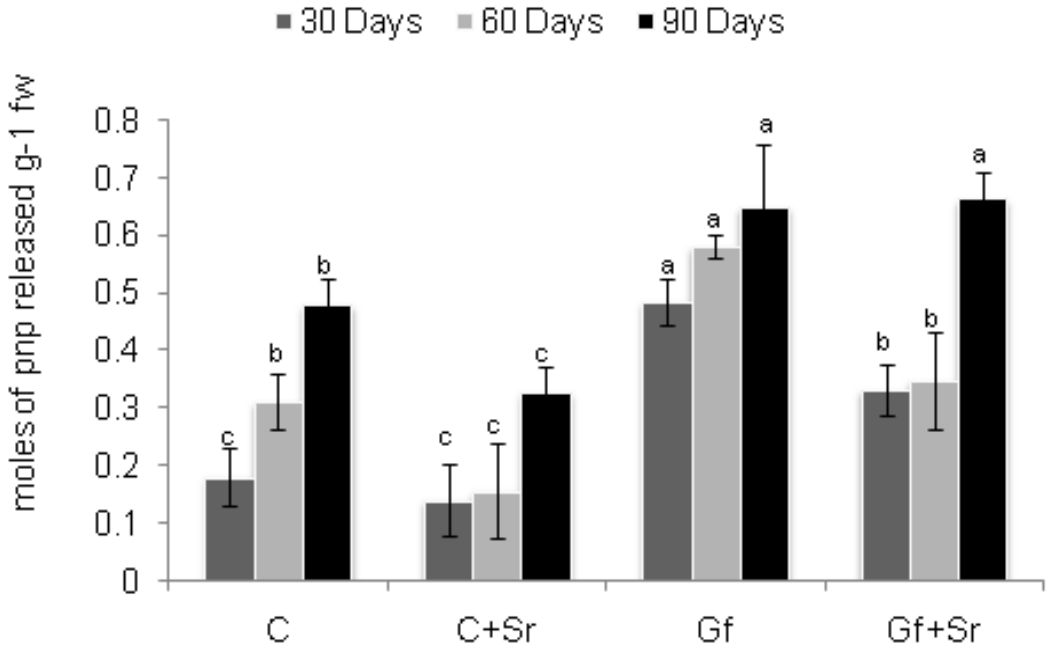

Figure 4: Alkaline phosphatase activity (moles of p-nitrophenol released g^{-1} of FW) in roots of AM or non-AM or S. rolfsii inoculated groundnut plants
C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

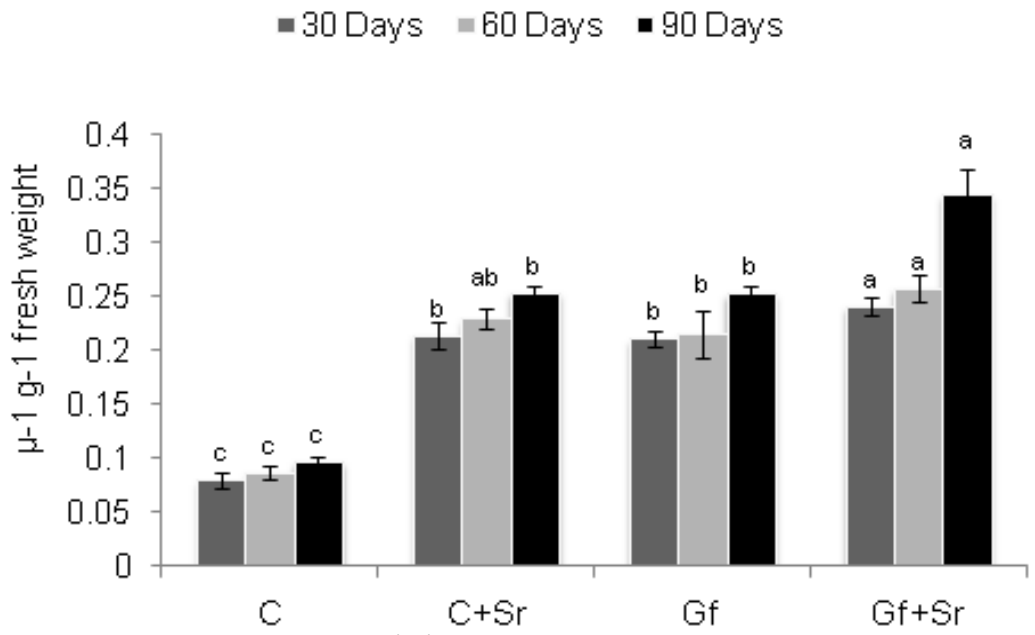

Figure 5: Protein content (protein in $\mu^{-1}g^{-1}$ FW) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants

C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

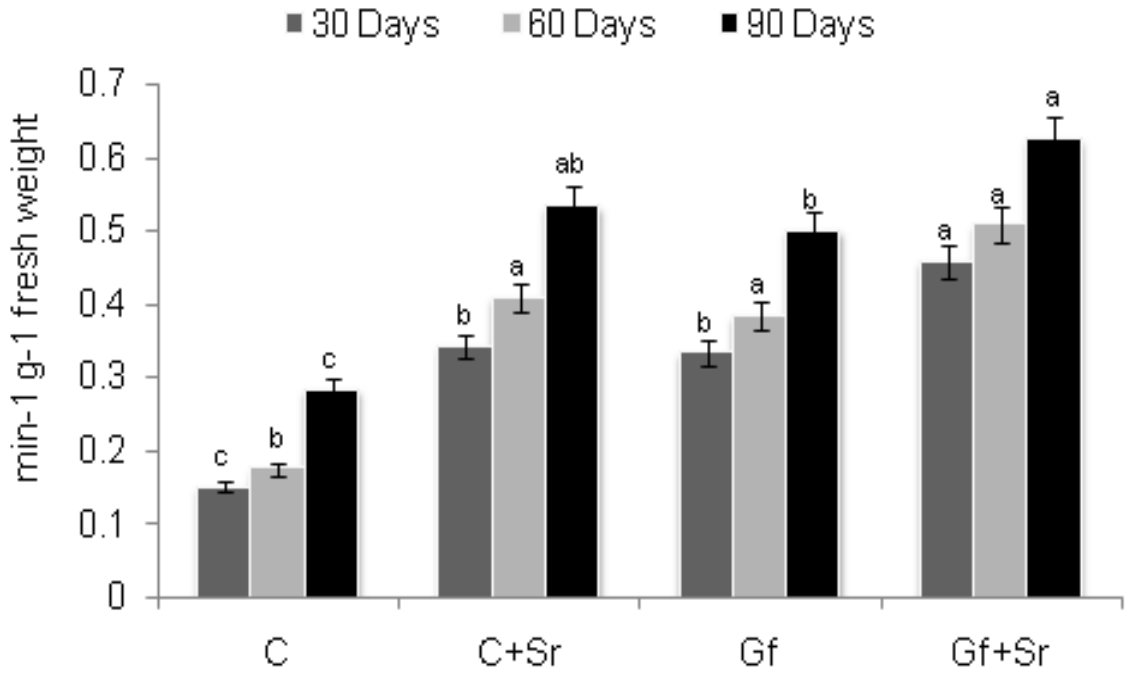

Figure 6: Polyphenol oxidase enzyme activity ($min^{-1}g^{-1}$ FW) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants

C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

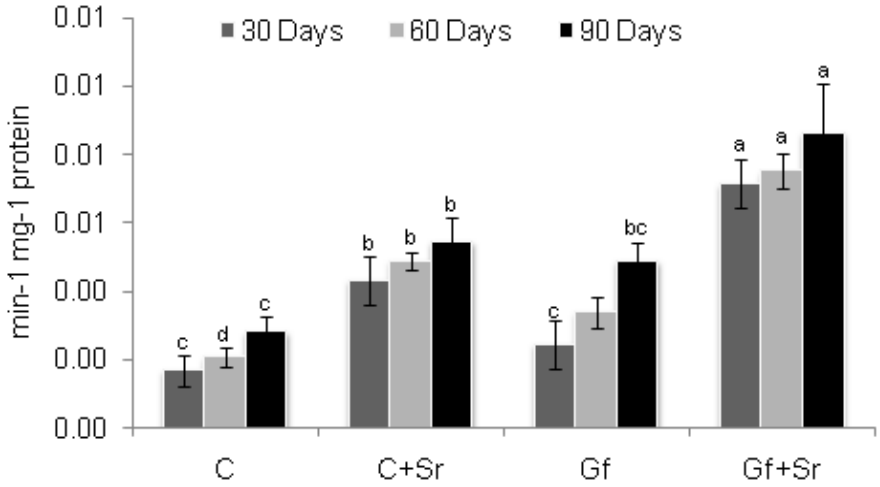

Figure 7: Peroxidase enzyme activity ($min^{-1}mg^{-1}$protein) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants
C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

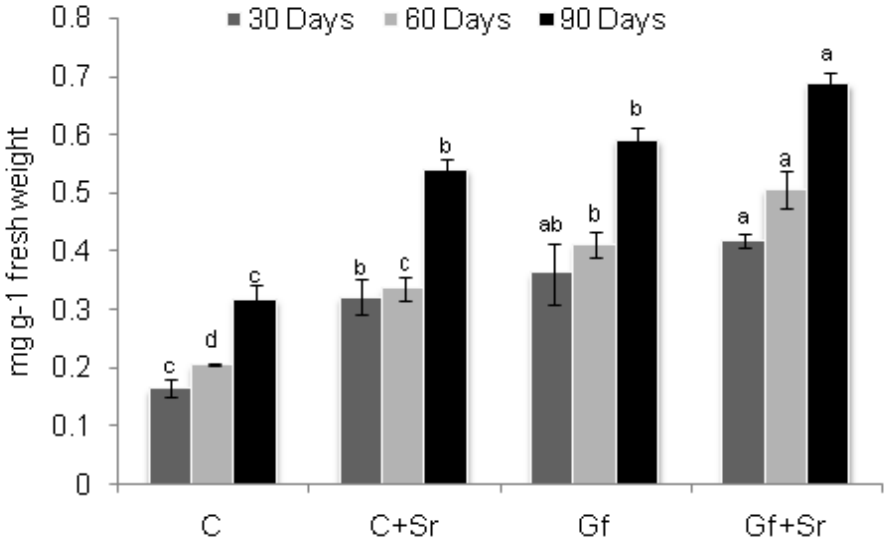

Figure 8: Total phenol content (mg g^{-1} FW) in roots of AM or non-AM or *S. rolfsii* inoculated groundnut plants
C: Uninoculated Control; C+Sr: Control + *S. rolfsii*; Gf: *G. fasiculatum* inoculated; Gf+Sr: *G. fasiculatum* + *S. rolfsii* inoculated. Means with the same letter are not significantly different from each as per Duncan's Multiple Range Test (P<0.05), *n*=3

Phosphatase Activity
The occurrences of phosphatase are ubiquitous in plants, animals and microorganisms. The secretion of phosphatase into rhizosphere under P-deficient condition is believed to be involved in organic P mineralization (Duff *et al.*, 1994). The present data demonstrated in Figure 3 and 4 revealed that the activities of phosphatase in the roots of groundnut plants showed higher activity in mycorrhiza inoculated groundnut plants as compared to non-mycorrhizal or control ones. The elevated responses of acid and alkaline phosphatase were higher in only mycorrhiza (Gf) treated groundnut plants as compared to

mycorrhizal diseased (Gf+Sr) or control ones. The reason for fluctuation in phosphatase activity may be obstruction in acquisition of phosphates in the groundnut plants. That is why lower acid or alkaline phosphatase activities were observed in diseased ones but their activities increased due to mycorrhizal colonization. The phosphatase activities have been suggested to be more in mycorrhizal association than non-mycorrhizal plants (Krishna and Bagyaraj, 1985; Tarafdar and Claassen, 1988).

Biochemical Activities

The content of total proteins as showed in Figure 5 revealed higher levels in roots of groundnut plants in presence of pathogen (C+Sr). However, the mycorrhiza inoculated diseased ones (Gf+Sr) showed highest total protein level followed by only mycorrhiza (Gf) treatments or controls ones. The elicitation of host protein synthesis is considered to be brought about by pathogen penetration in host plants which it leads to the restriction of pathogens (Adrienne and Barbara, 2006). The specific defense mechanisms that are involved in reducing the pathogen attack by way of mycorrhizal symbiosis are compound production of pathogenesis-related (PR) proteins (Conrath *et al.,* 2006) which were reported as one of the major defense mechanism involved in the inhibition of disease development in plants (Van *et al.,* 2006).

Generally plants express peroxidase activity which is involved in lignification of host cell wall during pathogen interaction (Maksimov *et al.,* 2014; Saikia *et al.,* 2006). Hence, the levels of peroxidase activity were found to be more in diseased non-mycorrhizal groundnut plants (C+Sr) as compared to healthy mycorrhizal (Gf) or control ones. But, the peroxidase activity elevated to highest extent in mycorrhizal diseased groundnut plants (Gf+Sr) than any other treatments. In the present experiment the peroxidase activity must have increased the mechanism of cell-wall reinforcement due to pathogen attack and the role of AM fungi in bringing peroxidase activity may be correlated to the observations of Goicoechea *et al.,* (2010) in which peroxidase specific activities were elevated due to inoculation of mycorrhizal fungus against *Verticillium dahlia* Kleb. of pepper plants.

In the development of the plants, low molecular compounds such as phenolics plays significant role and its release and synthesis may be induced by biotic as well as abiotic factors (Joachim *et al.,* 2007). The role of phenols has been studied extensively in suppression of pathogen attack as it is responsible for providing barriers to pathogen attack and helps in building mechanical strength to cell wall (Conceica *et al.,* 2006). The results of our investigation showed that the total phenols were increased due to presence of pathogen (C+Sr) in non-mycorrhizal groundnut plant as compared to control ones. However, in presence of pathogen the mycorrhizal groundnut plants (Gf+Sr) showed highest total phenol activity which shows role of AM fungi in induction of total phenol activity. In our results, the activities of PPO was higher in diseased groundnut plants (C+Sr) as compared to healthy mycorrhizal (Gf) or control ones which signifies their role in reducing harmful effects of pathogen *S. rolfsii*. As PPO activity is attributed to their possible involvement in oxidation of polyphenols into antimicrobial compounds such as quinones which plays significant role during pathogen attack (Ahl *et al.,* 1992). The results demonstrated that the highest PPO was observed upon mycorrhizal inoculation in diseased groundnut plants (Gf+Sr). Thus, it supports the role of AM fungi in getting higher PPO activity. Moreover, the results can be correlated with Raj *et al.,* (2006) who showed higher levels of total phenol and PPO in resistant varieties. From the results of present experiment, we may conclude that AM fungi colonization ensued into beneficial role in groundnut plants by reducing the incidences of diseases caused by pathogen *S. rolfsii*. Moreover, the association or establishment of AM fungi resulted into induction of several defense related activities such as phosphatase, protein, total phenols, polyphenol oxidase and peroxidase in the roots of groundnut plants.

ACKNOWLEDGEMENT

This research was supported by an operating grant from the authorities of University Grants Commission, New Delhi, for sanctioning fellowship under RGNF (No. F. 14-2(SC)/2009(SA-III).

REFERENCES

Adrienne CS and Barbara JH (2006). Parallels in Fungal Pathogenesis on Plant and Animal Hosts. *Eukaryotic Cell* **5**(12) 1941-1949.

Ahl Goy P, Felix G, Metraux JP and Meinz Jr F (1992). Resistance to disease in the hybrid *Nicotiana glutinosa x Nicotiana debneyi* is associated with high constitutive levels of â-1, 3-glucanase, chitinase, peroxidase and polyphenoloxidase. *Physiological and Molecular Plant Pathology* **41**(1) 11-21.

Arabi MIE, Kanacri S, Ayoubi Z and Jawhar M (2013). Mycorrhizal Application as a Biocontrol Agent against Common Root Rot of Barley. *Research in Biotechnology* **4**(4) 07-12.

Aysan E and Demir S (2009). Using arbuscular mycorrhizal fungi and *Rhizobium leguminosarum*, *Biovar phaseoli* Against *Sclerotinia sclerotiorum* (Lib.) de bary in the common bean *Phaseolus vulgaris* L.). *Plant Pathology Journal* **8**(2) 74-48.

Azaizeh HA, Marschner H, Romheld V and Wittenmayer L (1995). Effects of a vesicular-arbuscular mycorrhizal fungus and other soil microorganisms on growth, mineral nutrient acquisition and root exudation of soil grown maize plants. *Mycorrhiza* **5** 321-327.

Bowen KL, Hagan AK and Weeks JR (1996). Soil-borne pests of peanut in growers field with different cropping histories in Alabama. *Peanut Science* **23**(1) 36-42.

Clark RB and Zeto SK (2000). Mineral acquisition by arbuscular mycorrhizal plants. *Journal of Plant Nutrition* **23**(7) 867-902.

Conceica LF, Ferreres F, Tavores RM and Dios AC (2006). Induction of phenolic compounds in *Hypericum pertoralum* L. cells by *Colletotrichum gloeosprioides* elicitation. *Phytochemistry* **67**(2) 149-155.

Conrath U, Beckers GJM, Flors V, García-Agustín P, Jakab G, Mauch F, Newman MA, Pieterse CMJ, Poinssot B, Pozo MJ, Pugin A, Schaffrath U, Ton J, Wendehenne D, Zimmerli L and Mauch-Mani B (2006). Priming: Getting Ready for Battle. *Molecular Plant-Microbe Interaction* **19**(10) 1062-1071.

Duff SMG, Sarath G and Plaxton WC (1994). The role of acid phosphatase in plant phosphorus metabolism. *Physiologia Plantarum* **90**(4) 791-800.

Farahani A, Lebaschi H, Hussein M, Hussein SA, Reza VA and Jahanfar D (2008). Effects of arbuscular mycorrhizal fungi, different levels of phosphorus and drought stress on water use efficiency, relative water content and praline accumulation rate of Coriander (*Coriandrum sativum* L.). *Journal of Medicinal Plants Research* **2**(6) 125-131.

Feng G, Zhang FS, Li XL, Tian CY and Rengel Z (2002). Improved tolerance of plants to salt stress by arbuscular mycorrhizal is related to higher accumulation of soluble sugars in roots. *Mycorrhiza* **12** 185-190.

Fritz M, Jacobsen I, Lynqkjaer MF, Thordal-Christensen H, Pons-Kühnemann J (2006). Arbuscular mycorrhiza reduces susceptibility of tomato to *Alternaria solani*. *Mycorrhiza* **16** 413-419.

Gerdemann JW and Nicolson TH (1963). Spores of mycorrhizal *Endogene* species extracted from soil by wet sieving and decanting. *Transactions of British Mycological Society* **46**(2) 235-244.

Goicoechea N, Garmendia I, Sanchez-Diaz M and Aguirreolea J (2010). Review. Arbuscular mycorrhizal fungi (AMF) as bioprotector agents against wilt induced by *Verticillium* spp. in pepper. *Spanish Journal of Agricultural Research* **8**(S1) S25-S42.

Harrier LA and Watson CA (2004). The potential role of arbuscular mycorrhizal (AM) fungi in the bioprotection of plants against soilborne pathogens in organic and/or other sustainable farming systems. *Pest Management Science* **60**(2) 149-157.

Haystead A, Malajczuk N and Grove TS (1988). Underground transfer of nitrogen between pasture plants infected with vesicular-arbuscular mycorrhizal fungi. *New Phytologist* **108**(4) 417-423.

Joachim HJR, Ndakidemi M and Ndakidemi PA (2007). Biological ecological and agronomic significance of plant phenolic compounds in rhizosphere of the symbiotic legumes. *African Journal of Biotechnology* **6**(12) 1358-1368.

Kokalis-Burelle N, Backman PA, Rodriguez-Kabana R and Ploper LD (1992). Potential for biological control of early leafspot of peanut using *Bacillus cereus* and chitin as foliar amendments. *Biological Control* **2**(4) 321-328.

Kramer U (2005). Phytoremediation: novel approaches to cleaning up polluted soils. *Current Opinion in Biotechnology* **16**(2) 133-141.
Krishna KR and Bagyaraj DJ (1985). Phosphatases in the rhozospheres of mycorrhizal and non-mycorrhizal groundnut. *Journal of Soil Biology and Ecology* **5**(2) 81-85.
Lowry OH, Rosenbrough NJ, Farr AL and Randall RJ (1951). Protein measurement with the Folin-Phenol reagent. *Journal of Biological Chemistry* **193**(1) 265-275.
Lowry OH, Roberts NR, Mei-Ling WS and Crawford (1954). The quantitative histochemistry of brain II. Enzyme measurement. *Journal of Biological Chemistry* **207**(1) 19-37.
Mahadevan A and Shridhar R (1982). *Methods in Physiological Plant Pathology*, 2nd edition (Sivakami publication, Madras) 153-155.
Maksimov I, Troshina N, Surina O and Cherepanova E (2014). Salicylic acid increases the defense reaction against bunt and smut pathogens in wheat calli. *Journal of Plant Interactions* **9**(1) 306-314.
Malick CP and Singh MB (1980). *Plant Enzymology and Histo Enzymology* (Kalyani publishers) New Delhi 286.
Mukherjee PK and Raghu K (1997). Effect of temperature on antagonistic and biocontrol potential of *Trichoderma* sp. On *Sclerotium rolfsii*. *Mycopathologia* **139**(3) 151-155.
Phillips JM and Hayman DS (1970). Improved procedure for cleaning roots and staining parasitic and vesicular arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions of British Mycological Society* **55**(1) 158-160.
Putter J (1974). Peroxidase. In: *Methods of Enzymatic Analysis*, edited by Bergmeyer HU (Academic Press) New York, USA 567-1124.
Raj SN, Sarosh BR and Shetty HS (2006). Induction and accumulation of polyphenol oxidase activities as implicated in development of resistance against pearl millet downy mildew disease. *Functional Plant Biology* **33**(6) 563-571.
Rillig MC (2004). Arbuscular mycorrhizae, glomalin, and soil aggregation. *Canadian Journal of Soil Science* **84**(4) 355-363.
Saikia R, Yadav M, Varghese S, Singh BP, Gogoi DK, Kumar R and Arora DK (2006). Role of riboflavin in induced resistance against *Fusarium* wilt and Charcoal rot diseases of chickpea. *Plant Pathology Journal* **22**(4) 339-347.
Schubler A, Schwarzott D and Walker C (2001). A new fungal phylum, the Glomeromycota: phylogeny and evolution. *Mycological Research* **105**(12) 1413-1421.
Singh G and Mukherji KG (2006). Root exudates as determinant of rhizospheric microbial diversity. In: *Microbial activity in the rhizosphere*, edited by Mukerji KG, Manoharachary C, Singh J (Springer Verlag, Berlin, Heidelberg) 39-55.
Smith SE and Read DJ (1997). *Mycorrhizal Symbiosis*, 2nd edition (Academic Press: London, UK).
Smith SE and Read DJ (2008). *Mycorrhizal Symbiosis*, 3rd edition (Academic Press: London, UK).
SPSS for Windows User's manual, version 10.0. 1999. SPSS Inc. Chicago, IL.
Susleendra D and Schlosser E (1999). Parasitism of *Sclerotium rolfsii* by *Trichoderma*. *Indian Phytopathology* **52**(I) 47-50.
Tarafdar JC and Claassen N (1988). Organic phosphorus compounds as a phosphorus source for higher plants through the activity of phosphatases produced by plant roots and microorganisms. *Biology and Fertility of Soils* **5**(4) 308-312.
Trappe (1982). Synoptic Keys to Genera and Species of Zygomycetous Mycorrhizal Fungi. *Phytopathology* **72**(8) 1102-1108.
Trouvelot A, Kough JL and Gianinazi-Pearson V (1986). Mesure du Taux de Mycorhization V A d'un Systeme Radiculaire Recherche de Methods D'estimation Ayant Une Signification Fonctionnelle. In: *Physiological and Genetical Aspects of Mycorrhizae*, edited by Gianinazzi-Pearso V and Gianinazzi S (INRA Publications, Paris) 217-221.
van Loon LC, Rep M and Pieterse CMJ (2006). Significance of inducible defence-related proteins in infected plants. *Annual Review of Phytopathology* **44** 135-162.

Wu F, Wang W, Ma Y, Liu Y, Ma X, An L and Feng H (2013). Prospect of beneficial microorganisms applied in potato cultivation for sustainable agriculture. *African Journal of Microbiology Research* **7**(20) 2150-2158.