GLOBAL JOURNAL OF BIO-SCIENCE AND BIOTECHNOLOGY

www.scienceandnature.org

RELATIONSHIP BETWEEN MYCORRHIZAL FUNGI AND PLANTS PRESENT ON IRON ORE MINED OVERBURDEN DUMP OF DALLI RAJHARA CHHATTISGARH, INDIA

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ABSTRACT
28 planted and 22 naturally growing tree species was assessed from iron ore mine overburden (OB) dump of Dalli Rajhara Chhattisgarh, India. Mycorrhizal fungi showed variation in root percent colonization and number of spores. Both planted and naturally growing tree species showed maximum number of spores in three year old dump and minimum in nine year. 60% colonization in *Moringa pterygosperma*, 33.33 in *Albizia odoratissima*, 53.33 in *Dodonaea viscosa*, 46.67 in *Gmelina robusta* and 66.67 in *Annona squanrosa* was only studied from iron ore mine OB land. In planted species maximum number of spores was observed in Fabaceae and minimum in Poaceae. Zero percent colonization was observed in Amaranthaceae, Combretaceae, Asteraceae, Sapindaceae and Apocynaceae family.

KEYWORD: Mining, plantation, VAM.

INTRODUCTION

The nature bestowed India with highly valuable forest and rich mineral resources. Throughout the human civilization, human has been exploiting these resources without bothering for replenishment. In mining area exploitation of mineral resources is the stepping stone for development (Ghosh, 1990). Mining is the second largest industry after agriculture and has played a vital role in the growth of civilization from ancient days (Khoshoo, 1984). The potential of soil microorganisms has been recognized widely in development of soil quality, soil formation, aggregation and re-vegetation through their activities in litter decomposition and nutrient cycle (Mishra, 1989). Their activities such as phosphate solubilisation, nitrogen fixation, oxidation of various inorganic components of soil or mineralization of inorganic components and mycorrhizal association are major beneficial activities that play a very important role in soil system functioning (Butler *et al*., 2003). A major component of the soil microbial community is a group of fungi that form a symbiotic association with the roots of most terrestrial plants knows as vesicular arbuscular mycorrhiza (VAM) or arbuscular mycorrhiza (AM) (Brundrett, 1991). AM associations are important in natural and managed ecosystems due to their nutritional and non-nutritional benefits to their symbiotic partners (Verma *et al*., 2016; Verma *et al*., 2017a). They can alter plant productivity because AM fungi acts as biofertilizers, bioprotectant or biodegraders (Xavier and Boyetchko, 2004) and know to improve plant growth and health by improving mineral nutrient or increasing resistance or tolerance to biotic and abiotic stress (Clark and Zeto, 1996; Verma and Verma, 2016). The role of AM in successful rehabilitation of mine areas and other plantation sites is significant as it increase the absorptive capacity of the plant to nutrients and water supply (Verma *et al*., 2017). They provide plant a powerful mycelial network (Jasper *et al*., 1987) which enable them to extract essential minerals especially phosphorus from the soil beyond root zone (Jakobson, 1995). They also stimulate growth hormones (Barea and Azcon-Anguilar, 1982), protect root from various stresses and pathogens (Wood *et al*., 1986), and compensate pH, salt, toxic metals and biotic factors (Sylvia and Williams, 1992).

Studies related to the diversity of AM fungi from iron ore mines are very scarce (Rodrigues, 1999; Sastry and Johri, 1999; Bukhari and Rodrigues, 2005; Kullu and Behera, 2016; Mohan *et al*., 2016; Verma and Verma, 2017). Hence, the present investigation was carried out with an aim to study the colonization and diversity of native AM fungal species in the rhizosphere soils from iron ore mine OB land at Chhattisgarh.

MATERIALS & METHODS

Study Site

Dalli is located on a hill range bounded by 20° 33' 0" and 20° 34' 30" N latitude and 81° 1' 0" and 81° 4' 30" E longitude and Rajhara mines are bounded by 20° 33' 0" and 20° 35' 0" N latitude and 81° 0' 45" and 81° 07' 0" E longitude under Balod (District) in the state of Chhattisgarh. Name of planted tree species was followed: *Ailanthus excelsa* Roxbs., *Albizia lebbeck* Benth., *Annona squamosa* L., *Artocarpus heterophyllus* Lam., *Azadirachta indica* A. Juss, Bamboo sp., *Butea monosperma* (Lam.) Taub., *Cassia siamea* Lam., *Cassia fistula* L., *Dalbergia sissoo* DC., *Delonix regia* (Hook.) Raf., *Eucalyptus* hybrid, *Eugenia jambolana* Lamk., *Ficus benghalensis* L., *Ficus religiosa* L., *Gmelina arborea* Linn., *Gmelina robusta* A. Cunn., *Leucaena leucocephala* (Lam.) de Wit, *Mangifera indica* L., *Moringa pterygosperma* Gaertn., *Peltrophorum ptercocarpus* (DC.) K. Heyne, *Phyllanthus officinalis* L., *Polyalthia longifolia* (Sonn.) Thwaites,

Pongamia pinnata (L.) Pierre, *Psidium guajava* L., *Tamarindus indica* L., *Tectona grandis* L.f., *Terminalia arjuna* (Roxb. ex DC.) Wight & Arn. Name of plant, shrub and herb which grow naturally as followed: Name of tree species *Acacia auriculiformis* A. Cunn. ex Benth., *Acacia nilotica* (L.) Delile, *Ailanthus excelsa* Roxbs., *Albizia lebbeck* (L.) Benth., *Albizia odoratissima* (L.f.) Benth., *Alternanthera sessilis* (L.) R. Br. ex DC., *Anogeissum latifolia* Wall, *Argemone maxicana* L., *Azadirachta indica* A. Juss, *Blumea alata* (D. Don) DC., *Butea monosperma* (Lam.) Taub., *Calotropis procera* R. Br., *Cassia alata* L., *Dalbergia sissoo* DC., *Dodonaea viscosa* Linn, *Hyptis suaveolens* (L.) Poit, *Lantana camara* L., *Nerium indicum* Mill, *Tecomas stans* (L.) Juss. ex Kunth, *Tridax procumbens* L., *Woodfodiya fruticosa* (L.) Kurz, *Ziziphus jujuba* Lam.

In the present study, following plantation of different age group raised on OB dump and plain areas (adjoining plantation outside the OB areas) of Dalli Rajhara were studied in detail for edaphic and microbial attributes.

1. 2011-12 OB plantation (3 year dump)
2. 2005-06 OB plantation (7 year dump)
3. 2006-07 OB plantation (8 year dump)
4. 2007-08 OB plantation (9 year dump)
5. Plain plantation or undisturbed soil plantation (Natural soil)

Soil sampling

Soil sample were collected from planted trees in iron ore mined OB dump of different age groups including 9, 8, 7, 3 and natural vegetation. Sample uses for microbial quantification were taken from rhizosphere zone by removing one cm soil from surface. A soil auger used which as washed thoroughly before starting of sampling procedure. Sampling done in 10-20 cm depth in soil horizon and fine root of planted tree were also carefully collected in polyethylene bags and their mouth were tied with rubber bands. In lab sample were homogenized and spread on paper to remove plant material, they are air dried, sifted with 2mm mesh sieve and stored at 4ºC used for experiment (Parkinson, 1979).

Isolation of AM fungal spore

Isolation and identification of arbuscular mycorrhizal fungi

Isolation Collected soil samples were processed for isolation of AM spore. To extract AM spores, wet sieving and decanting technique (Sylvia, 1994) was followed.

Cone method Filter paper cone was kept in a funnel after all the water trickles down transfer the filter paper cone after unfolding in similar sized Petri plate. The spore and extrametrical hyphae were collected from filter paper and counted either wet or after air drying under a stereo-zoom microscope (Mukerji *et al*., 2002).

Identification of arbuscular mycorrhizal fungi Identification of AM fungi on the basis of morphology of their resting spores by referring taxonomic manual (Schenck and Perez, 1990); and matter available on net (http://www.zor.zut.edu.pl/Glomeromycota/Classification.html).

Determination of root colonization (RC) Development of mycorrhizal fungi in term of RC was detected by staining the root. The method prescribed by Phillips and Hayman (1970) was applied. Hundred root bits were examined and the presence of RC was calculated by following the formula mentioned below:

$$\% \text{ Root Colonization} = \frac{\text{Number of root bits infected with AM fungi}}{\text{Total number of root bits examined}}$$

RESULTS

Status of AM fungi in OB and NS (natural, undisturbed soil)

Status of AM fungi in rhizosphere region of both kinds (planted and naturally growing) on 3, 7, 8 and 9 year old OB dump and natural soil was analysed (Table 1 and 2).

Status of AM fungi in planted and naturally growing tree species in OB dump

Status of AM fungi was observed in rhizosphere region of planted tree species in 3 year old planted OB dump. Maximum number of AM fungi/100g of spoil soil was observed in *Annona squamosa* and *Artocarpus heterophyllus* followed by *Psidium guajava*, *Mangifera indica*. *Phyllanthus officinalis* have lower number of spore followed by *Moringa pterygosperma*. The mean spore number in the samples was 146.6. Similarly in 7 years old planted OB dump maximum number of AM fungi was observed in *Cassia fistula* followed by *Ficus benghalensis*, and minimum number was observed in *Eucalyptus hybrid* and *Ficus religiosa* followed by *Leucaena leucocephala*, *Polyalthia longifolia* and *Pongamia pinnata*. The mean spore number in samples was 126.3. In 8 years old planted OB dump maximum number of spore was observed in *Cassia siamea* followed by *Dalbergia sissoo*, *Butea monosperma* and minimum was observed in *Cassia fistula* followed by *Gmelina robusta*. The mean spore number in the samples was 119.7. In 9 years old planted OB dump *Dalbergia sissoo* have highest number of spore followed by *Peltophorum pterocarpum*, *Gmelina robusta* and lower number of spore in *Tectona grandis* followed by *Delonix regia*, *Cassia siamea*. The mean spore number in the samples was 90.8. Data was statistically significant at $P = 0.05$ (Table 1).

TABLE 1: Status of AM fungi (spore/100g soil) in rhizosphere of planted tree species in different year old OB

S.No.	Name of tree species	Family	D_3	D_7	D_8	D_9	NS
1	*A. excelsa*	Simaroubaceae	-	72	-	-	90
2	*A. lebbeck*	Leguminosae	-	-	-	52	59
3	*A. squamosa*	Annonaceae	187	-	-	-	71
4	*A. heterophyllus*	Moraceae	187	-	-	-	84
5	*A. indica*	Meliaceae	-	-	-	98	38
6	Bamboo sp.	Poaceae	-	57	-	42	14
7	*B. monosperma*	Leguminosae	-	112	178	-	189
8	*C. siamea*	Caesalpiniaceae	-	-	268	48	108
9	*C. fistula*	Caesalpiniaceae	-	-	36	24	33
10	*D. sissoo*	Fabaceae	-	-	238	287	148
11	*D. regia*	Caesalpiniaceae	-	-	-	46	79
12	*Eucalyptus* hybrid	Myrtaceae	-	46	56	-	201
13	*E. jambolana*	Myrtaceae	-	98	-	-	43
14	*F. benghalensis*	Moraceae	-	36	-	-	84
15	*F. religiosa*	Moraceae	-	398	-	-	123
16	*G. arborea*	Lamiaceae	-	-	-	102	65
17	*G. robusta*	Proteaceae	-	-	38	112	141
18	*L. leucocephala*	Leguminosae	-	239	-	-	93
19	*M. indica*	Anacardiaceae	130	-	-	-	103
20	*M. pterygosperma*	Moringaceae	98	-	-	-	76
21	*P. ptercocarpus*	Leguminosae	-	-	-	148	54
22	*P. officinalis*	Phyllanthaceae	90	-	-	94	123
23	*P. longifolia*	Annonaceae	-	146	-	-	81
24	*P. pinnata*	Leguminosae	-	146	24	-	162
25	*P. guajava*	Myrtaceae	178	58	-	-	85
26	*T. indica*	Leguminosae	156	-	-	-	56
27	*T. grandis*	Lamiaceae	-	-	-	36	73
28	*T. arjuna*	Combretaceae	-	107	-	-	74
Mean			146.6±41.2*	126.3± 102.6*	119.7± 105.1*	90.8± 72.4*	91.1± 45.3*
Z value			0.402	0.188	0.606	0.528	0.134
SE			15.57	29.61	39.73	20.88	8.55

D_3, D_7, D_8, and D_9 = 3-9 year old dumps respectively, NS= natural soil, *Significant at *P=0.05*

TABLE 2: Status of AM fungi (spore/100g soil) in rhizosphere of naturally growing species in different year old OB

S.No.	Name of tree species	Family	D_3	D_7	D_8	D_9	NS
1	*A. auriculiformis*	Leguminosae	197	-	-	6	98
2	*A. nilotica*	Leguminosae	146	97	-	-	128
3	*A. excelsa*	Simaroubaceae	-	-	54	-	90
4	*A. lebbeck*	Leguminosae	202	-	35	-	59
5	*A. odoratissima*	Mimosaceae	-	62	-	-	84
6	*A. sessilis*	Amaranthaceae	15	-	-	-	30
7	*A. latifolia*	Combretaceae	-	108	-	-	165
8	*A. maxicana*	Papaveraceae	-	-	34	-	48
9	*A. indica*	Meliaceae	-	-	64	-	38
10	*B. alata*	Asteraceae	115	-	-	-	100
11	*B. monosperma*	Leguminosae	284	-	-	-	189
12	*C. procera*	Asclepiadaceae	145	178	-	-	63
13	*C. alata*	Fabaceae	-	-	-	86	49
14	*D. sissoo*	Fabaceae	348	-	-	-	148
15	*D. viscosa*	Sapindaceae	-	-	72	-	84
16	*H. suaveolens*	Lamiaceae	24	92	-	-	102
17	*L. camara*	Verbenaceae	-	120	48	6	168
18	*N. indicum*	Apocynaceae	-	-	-	78	184
19	*T. stans*	Bignoniaceae	-	-	-	13	28
20	*T. procumbens*	Leguminosae	96	-	-	-	79
21	*W. fruticosa*	Lythraceae	-	-	35	40	149
22	*Z. jujuba*	Rhamnaceae	110	12	-	-	92
Mean			152.9 ± 100.9*	95.6 ± 51.1*	48.9 ± 15.3*	38.2 ± 36.3*	98.9 ± 50.1*
Z value			0.49	0.07	0.91	0.69	0.29
SE			30.43	19.32	5.77	14.81	10.68

D_3, D_7, D_8, and D_9 = 3-9 year old dumps respectively, NS= natural soil, *Significant at (*P=0.05*)

AM fungi in rhizosphere region of naturally growing species were observed in 3 year old planted OB dump, *Dalbergia sissoo* have highest spore number in spoil soil followed by *Butea monosperma*, *Albizia lebbeck* and minimum number in *Alternanthera sessilis* followed by *Hyptis suaveolens*, *Tridax procumbens.* The mean spore

number in the samples was 152.9. In 7 year old OB dump highest number of spore was recorded in *Calotropis procera* followed by *Lantana camara, Anogeissus latifolia* and lower number of spore was observed in *Ziziphus jujuba* followed by *Albizia odoratissima*. The mean spore number in the samples was 95.6. In 8 year old OB dump lowest number of spore was observed in *Argemone maxicana* followed by *Woodfordia fruticosa* and *Albizia lebbeck*. Highest number of spore was recorded in *Dodonaea viscosa* followed by *Azadirachta indica*. The mean spore number in the samples was 48.9. In 9 year old OB dump highest number of spore was recorded in *Cassia alata* followed by *Nerium indicum* and lowest number was observed in *Acacia auriculiformis* and *Lantana camara* followed by *Tecomas stans*. The mean spore number in the samples was 38.2. Data was statistically significant at P = 0.05 (Table 2).

Status of AM fungi in natural (undisturbed) soil

In natural soil population of AM fungi varied in different species. *Eucalyptus* hybrid have highest number of spore followed by *Butea monosperma*, *Pongamia pinnata* and lowest number of spore was observed in bamboo sp., followed by *Cassia fistula*, *Azadirachta indica*. The mean spore number in the samples was 91.1/100g of spoil soil (Table 1). Similarly *Nerium indicum* have highest number of spore followed by *Lantana camara, Anogeissus latifolia* and lower number of spore was present in *Tecomas stans* followed by *Alternanthera sessilis*, *Argemone maxicana*. The mean spore number in the samples was 98.9 (Table 2). Data was statistically significant at P = 0.05.

STATUS OF ROOT COLONIZATION (RC)

Status of RC on OB dump planted and naturally growing plants

Status of percent RC of plants by AM fungi in both kind of plants (planted and naturally growing) on 3, 7, 8 and 9 year old OB dump and natural soil were determined and presented in table 3 and 4. In 3 year old planted OB dump *Mangifera indica* have higher RC followed by *Annona squamosa*, *Moringa pterygosperma*, *Tamarindus indica* and *Phyllanthus officinalis*. There were 2 plants namely *Artocarpus heterophyllus* and *Psidium guajava* in which feeder roots were not observed. The mean percent RC in the samples was 56.1. In 7 year old planted OB dump maximum RC was observed in *Terminalia arjuna* followed by *Eucalyptus* hybrid and *Ficus benghalensis* and minimum in *Polyalthia longifolia, Ficus religiosa, Pongamia pinnata, Leucaena leucocephala*. There were five plant species, namely *Butea monosperma*, *Eugenia jambolana, Ailanthus excelsa, Psidium guajava* and bamboo species in which feeder root was not observed. The mean percent RC in the samples was 38.1. In 8 year old planted OB dump maximum RC was observed in *Cassia siamea* followed by *Butea monosperma, Pongamia pinnata*. Minimum RC was observed in *Eucalyptus* hybrid and *Gmelina robusta*. There were 2 plants, *Dalbergia sissoo* and *Cassia fistula* in which feeder roots were not observed. The mean percent RC in the samples was 60. In 9 year old planted OB dump total 6 plant roots were observed of which *Albizia lebbeck* have highest RC followed by *Gmelina arborea, Cassia fistula, Delonix regia* and lowest in *Gmelina robusta* followed by *Phyllanthus officinalis* and *Peltophorum pterocarpum*. There were four plants, *Dalbergia sissoo, Azadirachta indica, Cassia siamea* and *Tectona grandis* in which feeder root were not collected. The mean percent RC in the samples was 49.5. Data was statistically significant at P = 0.05 (Table 3).

The status of percent RC in naturally growing tree species on OB dump was observed. In 3 year old dump RC of *Acacia nilotica* was maximum followed by *Hyptis suaveolens*, *Acacia auriculiformis* and minimum was observed in *Calotropis procera* followed by *Alternanthera sessilis, Albizia lebbeck* and *Butea monosperma*. There were 3 species, *Dalbergia sissoo, Tridax procumbens* and *Ziziphus jujuba* where feeder roots were not collected. The mean percent RC in the samples was 58.8. In 7 year old OB dump *Calotropis procera* and *Albizia odoratissima* were shown RC. There were five plants, *Lantana camara, Anogeissus latifolia, Acacia nilotica, Hyptis suaveolens* and *Ziziphus jujuba* in which feeder roots were not collected. The mean percent RC in the samples was 40.0. In 8 year old OB dump tree, *Albizia lebbeck* have highest RC followed by *Woodfordia fruticosa, Azadirachta indica* and minimum colonization was observed in *Dodonaea viscosa* and *Ailanthus excelsa*. Two plants feeder root were not collected (*Lantana camara* and *Argemone maxicana*). The mean percent RC in the samples was 66.7. In 9 year old OB dump *Nerium indicum, Woodfordia fruticosa, Acacia auriculiformis* and *Lantana camara* feeder root were not observed. Maximum RC was observed in *Tecomas stans* followed by *Cassia alata* and minimum RC was observed in *Woodfordia fruticosa*. The mean percent RC in the samples was 42.2. Data was statistically significant at P = 0.05 (Table 4).

Status of RC by AM fungi in NS (natural soil, undisturbed soil)

In NS total 28 plants were observed. Four plants bamboo species, *Gmelina arborea*, *Leucaena leucocephala* and *Phyllanthus officinalis* have not any colonization by AM fungi. Minimum RC was observed in *Eugenia jambolana* followed by *Azadirachta indica*, *Annona squamosa* and maximum was observed in *Moringa pterygosperma* followed *Cassia fistula*, *Ailanthus excelsa* and *Ficus benghalensis*. 60% RC was observed in *Cassia siamea, Eucalyptus* hybrid, *Ficus religiosa* and *Polyalthia longifolia. Dalbergia sissoo, Mangifera indica, Pongamia pinnata* and *Tamarindus indica* have 33.3% RC. The mean percent RC in the samples was 36.6 (Table 3). *Acacia nilotica, Alternanthera sessilis, Anogeissus latifolia, Blumea alata, Dodonaea viscosa* and *Nerium indicum* have zero percent RC. 6.7% RC was observed in *Azadirachta indica, Argemone maxicana* and *Tridax procumbens*. Five plant *Albizia odoratissima, Calotropis procera, Hyptis suaveolens, Lantana camara* and *Woodfordia fruticosa* showed 20% RC. Maximum RC was observed in *Ailanthus excelsa* (66.7%) followed by *Butea monosperma* (53.3%), *Acacia auriculiformis* and *Cassia alata* (40%). The mean percent RC in the samples was 19.4. Data was statistically significant at P = 0.05 (Table 4).

TABLE 3: Status of root colonization in rhizosphere of planted tree species in different year old OB

S.No.	Name of tree species	D_3				D_7				D_8				D_9				NS			
		RC %	A	V	H	RC %	A	V	H	RC%	A	V	H	RC %	A	V	H	RC %	A	V	H
1	*A. excelsa*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	66.7	√	×	√
2	*A. lebbeck*	-	-	-	-	-	-	-	-	-	-	-	-	86.7	√	√	√	26.7	×	√	√
3	*A. squamosa*	66.7	√	√	×	-	-	-	-	-	-	-	-	-	-	-	-	13.3	√	√	√
4	*A. heterophyllus*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	20.0	√	√	√
5	*A. indica*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	6.7	√	√	√
6	Bamboo *sp.*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0.0	-	-	-
7	*B. monosperma*	-	-	-	-	-	-	-	-	60.0	×	√	√	-	-	-	-	53.3	√	×	√
8	*C. siamea*	-	-	-	-	-	-	-	-	80.0	√	√	√	-	-	-	-	60.0	×	√	×
9	*C. fistula*	-	-	-	-	-	-	-	-	-	-	-	-	66.7	×	√	√	80.0	√	√	√
10	*D. sissoo*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	33.3	√	√	×
11	*D. regia*	-	-	-	-	-	-	-	-	-	-	-	-	53.3	×	√	√	26.7	×	√	×
12	*Eucalyptus* hybrid	-	-	-	-	40.0	×	√	√	53.3	×	√	√	-	-	-	-	60.0	√	√	√
13	*E. jambolana*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	6.7	×	√	√
14	*F. benghalensis*	-	-	-	-	40.0	×	√	√	-	-	-	-	-	-	-	-	66.7	×	√	√
15	*F. religiosa*	-	-	-	-	20.0	√	×	√	-	-	-	-	-	-	-	-	60.0	×	√	√
16	*G. arborea*	-	-	-	-	-	-	-	-	-	-	-	-	73.4	√	√	√	0.0	-	-	-
17	*G. robusta*	-	-	-	-	-	-	-	-	46.6	×	√	√	6.7	√	√	√	53.3	×	√	√
18	*L. leucocephala*	-	-	-	-	33.4	×	√	√	-	-	-	-	-	-	-	-	0.0	×	×	×
19	*M. indica*	93.6	×	√	√	-	-	-	-	-	-	-	-	-	-	-	-	33.3	×	√	√
20	*M. pterygosperma*	60.0	√	×	√	-	-	-	-	-	-	-	-	-	-	-	-	90.0	×	√	√
21	*P. ptercocarpus*	-	-	-	-	-	-	-	-	-	-	-	-	40.0	√	√	√	40.0	√	√	√
22	*P. officinalis*	20.0	√	√	√	-	-	-	-	-	-	-	-	20.0	×	√	×	0	-	-	-
23	*P. longifolia*	-	-	-	-	13.3	×	√	√	-	-	-	-	-	-	-	-	60.0	×	√	√
24	*P. pinnata*	-	-	-	-	26.6	√	√	√	60.0	×	√	√	-	-	-	-	33.3	√	√	×
25	*P. guajava*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	26.6	×	×	√
26	*T. indica*	40.0	×	√	√	-	-	-	-	-	-	-	-	-	-	-	-	33.3	×	√	√
27	*T. grandis*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	46.6	×	×	√
28	*T. arjuna*	-	-	-	-	93.3	√	√	√	-	-	-	-	-	-	-	-	26.6	×	√	×
Mean		56.1±27.8*				38.1±26.3*				60.0±12.5*				49.5±29.0*				36.6±25.7*			
Z value		0.58				0.18				0.54				0.22				0.13			
SE		12.45				9.94				5.578				10.97				4.85			

D_3, D_7, D_8, and D_9 = 3-9 year old dumps respectively, NS= natural soil, (-) = plant feeder root not available/ absent; RC= root colonization by AM fungi; A=arbuscule, V=vesicle, H=hyphae; *Significant at P=0.05

TABLE 4: Status of root colonization in rhizosphere of naturally growing species in different year old OB

S.No.	Name of tree species	D_3				D_7				D_8				D_9				NS			
		RC%	A	V	H	RC%	A	V	H	RC%	A	V	H	RC%	A	V	H	RC%	A	V	H
1	*A. auriculiformis*	86.7	×	×	√	-	-	-	-	-	-	-	-	-	-	-	-	40.0	√	√	√
2	*A. nilotica*	93.7	√	√	√	-	-	-	-	-	-	-	-	-	-	-	-	0.0	×	×	×
3	*A. excelsa*	-	-	-	-	-	-	-	-	40.0	√	√	√	-	-	-	-	66.7	√	×	√
4	*A. lebbeck*	33.3	√	√	√	-	-	-	-	93.7	√	√	√	-	-	-	-	26.7	×	√	√
5	*A. odoratissima*	-	-	-	-	33.3	×	×	√	-	-	-	-	-	-	-	-	20.0	×	√	×
6	*A. sessilis*	33.3	×	√	√	-	-	-	-	-	-	-	-	-	-	-	-	0.0	×	×	×
7	*A. latifolia*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0.0	×	×	×
8	*A. maxicana*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	6.6	×	√	×
9	*A. indica*	-	-	-	-	-	-	-	-	60.0	×	×	×	-	-	-	-	6.7	√	√	√
10	*B. alata*	66.7	√	√	√	-	-	-	-	-	-	-	-	-	-	-	-	0.0	×	×	×
11	*B. monosperma*	40.0	√	√	√	-	-	-	-	-	-	-	-	-	-	-	-	53.3	√	×	√
12	*C. procera*	26.7	×	×	√	46.7	√	√	√	-	-	-	-	-	-	-	-	20.0	×	√	√
13	*C. alata*	-	-	-	-	-	-	-	-	-	-	-	-	46.7	×	√	√	40.0	√	√	√
14	*D. sissoo*	53.3	×	×	√	-	-	-	-	-	-	-	-	-	-	-	-	33.3	√	√	×
15	*D. viscose*	-	-	-	-	-	-	-	-	53.3	√	√	√	-	-	-	-	0.0	×	×	×
16	*H. suaveolens*	93.3	×	√	×	-	-	-	-	-	-	-	-	-	-	-	-	20.0	×	√	×
17	*L. camara*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	20.0	√	√	×
18	*N. indicum*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0.0	×	×	×
19	*T. stans*	-	-	-	-	-	-	-	-	-	-	-	-	60.0	√	√	√	13.3	×	√	×
20	*T. procumbens*	60.0	×	√	×	-	-	-	-	-	-	-	-	-	-	-	-	6.6	×	√	×
21	*W. fruticosa*	-	-	-	-	-	-	-	-	86.7	×	√	×	20.0	√	√	√	20.0	×	√	×
22	*Z. jujuba*	60.0	×	√	√	-	-	-	-	-	-	-	-	-	-	-	-	33.3	×	√	√
Mean		58.8±24.4*				40±9.4*				66.7±22.7*				42.2±20.4*				19.4±18.9*			
Z value		0.22				0.71				0.59				1.09				0.32			
SE		7.36				6.67				10.16				11.76				4.02			

D_3, D_7, D_8, and D_9 = 3-9 year old dumps respectively, NS= natural soil, (-) = plant feeder root not available; RC= root colonization by AM fungi; A=arbuscule, V=vesicle, H=hyphae; *Significant at P=0.05

Number of spores and Root percentage colonization in different family

The planted tree species was belonged to 15 different families like Simaroubaceae, Meliaceae, Poaceae, Fabaceae, Proteaceae, Anacardiaceae, Moringaceae, Phyllanthaceae, Combretaceae, Lamiaceae, Annonaceae, Caesalpiniaceae, Myrtaceae, Moraceae and Leguminosae.

In three year old planted dump, maximum number of spores was observed in Moraceae and Annonaceae family and minimum in Phyllanthaceae. Seven year dump maximum number of spores was observed in Moraceae family and minimum in Poaceae. Eight year dump maximum number of spores was observed in Fabaceae and minimum in Proteaceae. Nine year dump maximum number of spores was observed in Fabaceae and minimum in Caesalpiniaceae. Trees present in natural soil maximum number of spores was observed in Fabaceae and minimum in Poaceae (Table 1). Similarly in natural soil maximum percentage of root colonization was observed in Moringaceae family followed by Caesalpiniaceae and Simaroubaceae, however minimum was observed in Meliaceae followed by Lamiaceae and Combretaceae (Table 3).

In OB dump 17 family Fabaceae, Leguminosae, Simaroubaceae, Mimosaceae, Amaranthaceae, Combretaceae, Papaveraceae, Meliaceae, Asteraceae, Asclepiadaceae, Sapindaceae, Lamiaceae, Verbenaceae, Apocynaceae, Bignoniaceae, Lythraceae and Rhamnaceae was naturally grow. In three year old planted dump maximum number of spores was observed in Fabaceae family and minimum in Amaranthaceae**.** Seven year dump maximum number of spores was observed in Asclepiadaceae family and minimum in Rhamnaceae. Eight year dump maximum number of spores was observed in Papaveraceae and minimum in Meliaceae. Nine year dump maximum number of spores was observed in Fabaceae and minimum in Leguminosae and Verbenaceae. Trees present in natural soil maximum number of spores was observed in Apocynaceae and minimum in Bignoniaceae (Table 2). Similarly in natural soil maximum percentage of root colonization was observed in Simaroubaceae family followed by Fabaceae and Rhamnaceae, however minimum was observed in Bignoniaceae followed by Papaveraceae, Meliaceae. In Amaranthaceae, Combretaceae, Asteraceae, Sapindaceae and Apocynaceae family zero percentage colonization was observed (Table 4).

DISCUSSION

Population of AM fungi in OB dump of iron ore mine soil

The population of AM fungi was recorded in rhizosphere of planted tree species in OB dump of different ages. The AM fungal population was maximum 146.6/100g soil in 3 year old dump while minimum in 9 year old OB (Table 1). Similarly AM spore population in rhizosphere of naturally growing species in OB dump was maximum 152.91 in 3 year old dump while minimum in 9 year old OB (Table 2). The distribution and occurrence of AM fungi in different ecological conditions is believed to be influenced by a number of factors such as soil-texture, moisture, depth, pH and temperature (Zajicek *et al.,* 1986). Many workers also observed that with the age number of spores also increased in the dump of mine (Chandra and Jamaluddin, 1999; Mukhopadhyay and Maiti, 2010; Singh and Jamaluddin, 2011; Chaubey *et al*., 2012; Kullu and Behera, 2012). However, in present investigation opposite result was found, these may be due to increased concentration of P (Table 1 and 2). Ryan *et al.* (1994) reported that higher levels of soluble P have negative effect on AM colonization but insoluble rock phosphate did not decrease levels of AM spore population. Possibly another reason attributed to this is growth of invasive species, like *Lantana camara* was very high in oldest OB dump which omit many chemical in the vicinity of root hair (Simelane, 2002; Sharma *et al*., 2005) and showed less root colonization (Table 4) present result was supported by Bhale *et al*. (2011) who observed 4% RC in *Lantana camara.* Chandra and Kehri (2006) also recorded lowest number of spore density 8 spores and 16 sporocarps in rhizosphere of *Lantana camara*. One more reason may responsible for less AM spore population is that in initial stage amount of available P is very low so that AM fungi were rapidly colonized but with increasing age many P soluble bacteria and fungi was grown in soil which increased the P level.

Root colonization (RC) with different plants at iron ore mine OB land spoil

AM root colonization in different aged reclaimed coal mine OB dump at Jharia Coalfield has been studied. *Dalbergia sissoo* have high percent of RC (94%), however in present finding *Dalbergia sissoo* have moderate percent of RC (53.3%). Similar result was observed in *Terminalia arjuna* 93.3% in coal mine OB dump, however in present finding 58% colonization was observed in *Terminalia arjuna. Cassia siamea* (90%) and *Acacia auriculiformis* (89%) have high RC in coal mine OB dump and these were at par with the present finding 80% and 86.7%, respectively. *Leucaena leucocephala* (68%) have moderate RC but in present study showed low level of RC (33.4). *Gmelina arborea* (38%) have low level of RC but in present finding has 73.4% (Table 3) (Mukhopadhyay and Maiti, 2010). These variations may be due to different physiochemical properties of soil. *Acacia auriculiformis* have high level of RC (79%) and this was similar with present finding 86.7%. *Polyalthia longifolia* have moderate level of RC (58%) and in present finding very low percent of RC, 13.3% was observed, while *Azadirachta indica* have 21% colonization but in iron ore OB colonization it was 60% (Table 3) (Kumar *et al*., 2011).

A study was conducted in Kumaun Himalayan region of Central Himalaya, India showed *Delonix regia* have 45% RC (Bargali, 2011) while in the present study it was 53.3%. *Albizia lebbeck* have 47% RC, but in iron ore mine land it has different percentage of RC in different age dump, 86.7% in 9 years, 93.7% in 8 years and 33.3% in 3 years (Table 3).

Kumar *et al.* (2003) observed RC in *Ficus benghalensis* 65% and in present study it has 40%. *Calotropis procera* (60%) has moderate level of RC while in iron ore mine land it was 26.7% in 3 year and 46.7% in 7 year old dump respectively. *A. nilotica* has moderate level of RC (55%) and in iron ore mine land it has high level of colonization

(93.7). *Cassia fistula* has low level of RC (45%) but in iron ore mine OB it has moderate RC (66.7%). Moderate RC was observed in *Butea monosperma* (50%), *Eucalyptus hybrid* (40), *Tridax procumbens* (45%), *Tamarindus indica* (50%) and in present finding it has 60% in natural soil and 40% in 7 year old dump, *Eucalyptus* hybrid 40% in 7 year old dump and 53.3% in 8 years dump. *Tridax procumbens* has 60%, *Tamarindus indica* 40% RC in planted and naturally growing plants respectively. Similar RC was observed in *Alternanthera sessilis* 33.3% in 3 years dump, *F. religiosa* 20% in 7 years dump, *P. pinnata* 60% in 8 years dump. *Phyllanthus officinalis* have 40% colonization and in iron ore mine OB 40%. No RC was observed in *Blumea* sp. and *Woodfordia fruticosa* but in present finding *Blumea alata* has 66.7% in 3 years dump, *Woodfordia furticosa* 86.7% in 8 years and 20% in 9 years and natural soil (Table 3). Hazarika *et al*. (2014) observed 47.5% RC in *Mangifera indica* from coal mine OB dump of Margherita, Assam. However, 93.67% RC was observed in iron ore mine OB. In present study *Ailanthus excelsa* and *Peltophorum pterocarpum* show 40% root colonization. Similarly Chandra and Jamaluddin (1999) and Jamaluddin *et al*. (2002) also observed infection in these plants on coal mine OB dump and Ballarpur paper Mills gardens. In present study *Cassia alata* has 46.67% RC but Rani and Bhaduria (2001) were observed 51±01.67% colonization. Chandra (2014) observed 59.44% RC in *Hyptis suaveolens* in different coal mine OB of Chhattisgarh. When in present finding *Hyptis suaveolens* have 93.33% RC and 23.4% RC in *Ziziphus jujuba* on coal mine OB and in present study 60% colonization was observed in iron ore mine OB. In present finding *Tecomas stans* have 60% RC in iron ore mine OB and similar colonization was observed by Logaprabha and Tamilselvi (2015) in a limestone mine spoil from ACC, Coimbatore. Raghupathy and Mahadevan (1993) observed the infection of AM in *Gmelina arborea.* However in present study observed 73.36% high RC.

Root colonization in *Moringa pterygosperma* 60, *Albizia odoratissima* 33.33, *Dodonaea viscosa* 53.33, *Gmelina robusta* 46.67, *Annona squanrosa* 66.67% was only studied from iron ore mine OB land.

Root colonization with different plants in natural soil

Verma (2009) observed variation in percent of RC in different tree species in central India forest including Madhya Pradesh, Chhattisgarh, Orissa and Maharashtra. *Ailanthus excelsa* have 60% RC but in undisturbed area of Dalli and Rajhara 66.7%. Similarly *Azadirachta indica* have 55.67% colonization and in study area 6.67%, *Acacia auriculiformis* have 00.0 and in study area 40%, *Acacia nilotica* have 51.67 and in study area no colonization was observed, *Albizia lebbeck* have 10% colonization and in study area 26.67%, *Butea monosperma* have 40 and in study area 53.33%,, *Cassia siamea* have 35% colonization and in study area 60%, *Dalbergia sissoo* have 28.17 and in study area 33.33%, *Delonix regia* have 00.0 and in study area 26.7%, *Leucaena leucocephala* have 62 and in study area no colonization was observed, *Pongamia pinnata* have 46.75% colonization and in study area 33.33% colonization and Bamboo have 35.6 and in study area no colonization was observed. *Gmelina arborea* have 70% RC (Verma, 2009) and 33% colonization (Chauhan *et al*., 2013) and in study area no colonization was observed. *Mangifera indica* have 00.0 colonization (Verma, 2009) and 70% colonization was observed by Pindi (2011) and in study area 33.33% was observed. Root percent colonization in *Annona squamosa* was 63% (Sarwade *et al*., 2011) but in present study 13.33% colonization was observed. *Peltophorum pterocarpum* showed 19% colonization (Jamaluddin *et al*., 2002) and in present study 40% colonization was observed. *Cassia alata* have 69.5% colonization (Chatterjee *et al*., 2010) but in present study 40% colonization was observed. Singh *et al*. (2012) was observed 52.7% colonization in *Ziziphus jujuba* but in present study 33.33% colonization was observed. *Hyptis suaveolens* have 80.35% colonization (Chandra, 2014) but in present study 20% colonization was observed. This indicates that during the initial stage of vegetation development on iron mine spoil, there is an obligatory requirement of mycorrhizal association for the plant. As noted in the present study, percentage of mycorrhizal infection varied with species. This is in agreement with the findings of Van der Heijden *et al*. (1998).

Variation in root infection among different plant species were might be due to the physiology and/or morphology of the roots. It has been reported that AM association may be reduced due to anthropogenic disturbance (Brundrett, 1991**;** Gould *et al*., 1996). The fluctuation in spore population and quantum of RC might be due to the influence of different environmental factors on AM sporulation and infection (Mohankumar, 1985). In general RC and number of AM spores in the rhizosphere of the plant species grown in mine soil was found higher to those in natural soil (Rao and Tarafdar, 1998; Mukhopadhyay and Maiti, 2010).

The variation in the amount of AM fungal spore and colonization is due to edaphic factors and environmental conditions. The highest percentage of root colonization was observed in a plant which belongs to Poaceae family. Minimum value for AM colonization was recorded in plants belonging to family Cyperaceae (Channasava and Lakshman, 2013).

ACKNOWLEDGEMENT

Authors are thankful to the Director, Tropical Forest Research Institute, Jabalpur for providing laboratory facilities.

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