



MYCORRHIZA NEWS

The Quarterly Newsletter of Mycorrhiza network

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About TERI

The Energy and Resources Institute (TERI) is a dynamic and flexible organization with a global vision and a local focus. TERI's focus is on research, in the fields of energy, environment, and sustainable development, and on documentation and information dissemination. The genesis of these activities lie in TERI's firm belief that the efficient utilization of energy, sustainable use of natural resources, large-scale adoption of renewable energy technologies, and reduction of all forms of waste would move the process of development towards the goal of sustainability.

TERI's Mycorrhiza Network

TERI's Mycorrhiza Network is primarily responsible for establishing the Mycorrhiza Information Centre (MIC), the Centre for Mycorrhiza Culture Collection (CMCC), and publishing Mycorrhiza News. The Network helps scientists carry out research in mycorrhiza and promotes communication among mycorrhiza scientists.

Mycorrhiza News

The Mycorrhiza News provides a forum for dissemination of scientific information on mycorrhiza research and activities; publishes state-of-the-art papers from eminent scientists; notes on important breakthroughs; brief accounts of new approaches and techniques; publishes papers compiled from its RIZA database; provides information on forthcoming events on mycorrhiza and related subjects; lists important research references published during the quarter; and highlights the activities of the CMCC.



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RESEARCH FINDING PAPERS

Activity of AM Fungi on Growth of *Jatropha curcas* L. in Fly Ash and Coal Dump Soil in Nursery

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Introduction

Huge coalmine dumps are formed after the excavation of coal and fly ash ponds emerge from thermal power. These are devoid of supportive and nutritive capacity for biomass development and create several problems to regetate such wastes. Several microbial processes such as nitrogen and carbon cycling, humification, and soil aggregation are practically non-functional in such habitats, posing challenges in the restoration of soil fertility. The role of bioinoculnats in such degraded soils is well understood by several workers (Juwarkar and Jambhulkar, 2008; Maiti, 2007).

Jatropha curcas is an important oil-yielding plant and a source of biodiesel production. The AM fungi and other microbes help this plant in utilizing soil phosphorus, protect them from the detrimental effect of root pathogens and provide resistance against drought. The AM fungi will enable in the restoration of degraded land and help to rejuvenate the area biologically. AM fungi also provide access for substantial carbon dioxide sink, built up top soil and enhanced holding capacity in degraded soil (Juwarkar and Jambhulkar, 2008). These Mycorrhizal fungi accumulate heavy metals from the fly ash and coal dump soil through their mycelial network and retain them in their physical structure or they are converted to some other forms. The occurrence of AM spores in the coal dump and fly ash soil are found to be double than non-mining areas due to higher disturbance,

porosity, available carbon and abiotic stress (Maiti, 2007). Therefore, looking to the enormous potential of these bioinoculants present investigation deals with establishment of *J. curcas* plant using coal dump and fly ash soil as substrates, with the help of bioinoculants in nursery.

Materials and Methods

Collection of Soil

Coal dump soil was collected from the open cast coalfield mines of Nigahi Project of the Singrauli district from the Northern Coal field Limited (NCL). The soil of fly ash was collected from the National Thermal Power Corporation (NTPC), Vindhyanagar, Singrauli, Madhya Pradesh (India).

Isolation and Identification of AM Fungi

The soil for isolation of AM fungi was collected from *Jatropha curcas*, plantation which is well established and is of 5 years old from Singrauli area. The VAM fungi were enumerated using wet-sieving and decanting technique (Gerdemann and Nicolson, 1963). The species were identified on the basis of spore characters (Schenck and Perez, 1990). The AM was consortium in the mother plant soil of *J. curcas* containing *Glomus intraradices*, *Glomus aggregatum*, *Acaulospora* sp., *Gigaspora margarita*, and *Scutellospora* sp. was used for inoculation purposes. These were

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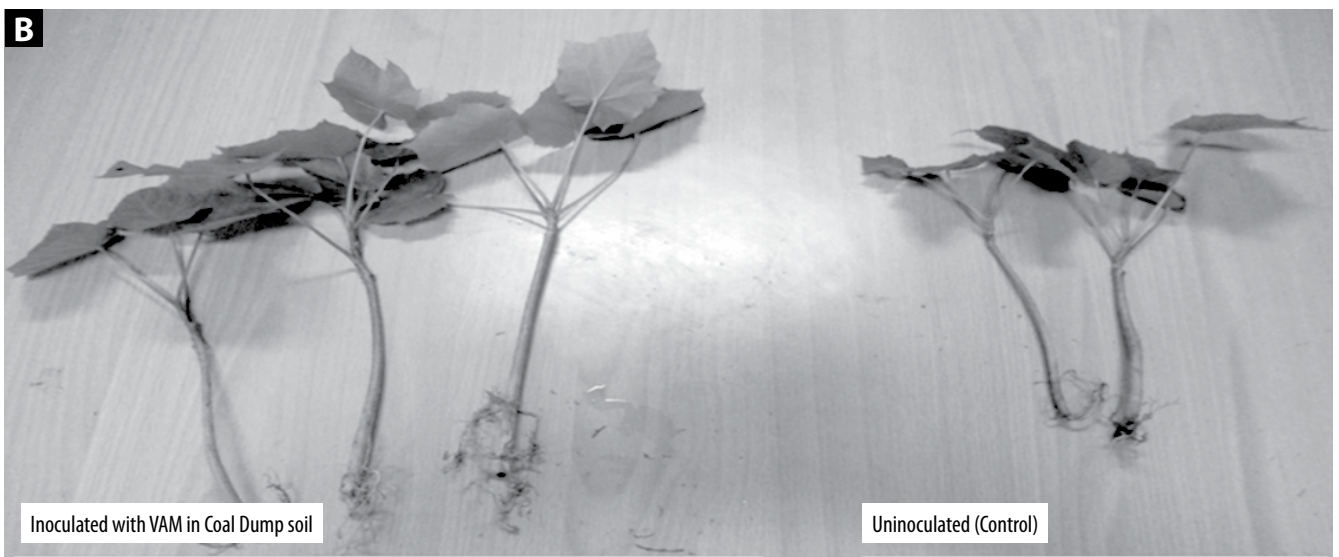
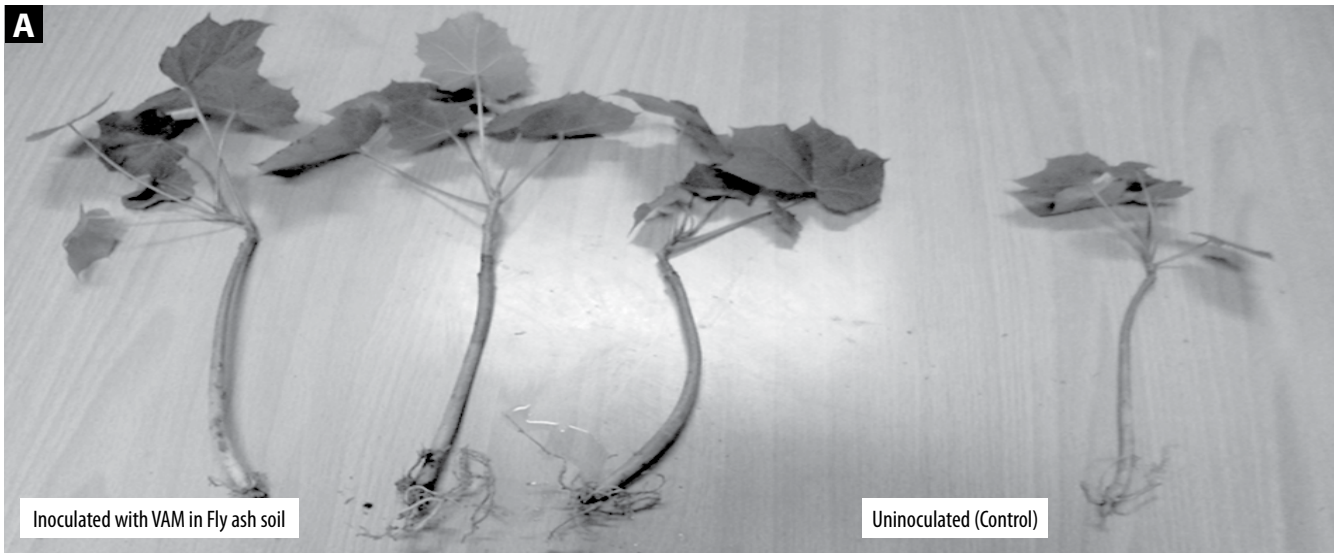


Figure. A, B, and C showing the effect of VAM inoculation in the soil of Fly ash, Coal dump soil, and Nursery soil

cultured in trap plant using maize. The mix cultures of AM fungi so developed in maize were used as inoculum containing mix spores and root bits. About 20 g inoculum in each polypot.

Preparation of Pots

Three types of materials were used to fill up the polypots (21x9 cm sizes). Pots were filled up with fly ash, coal mines soil, and soil mix (sand and soil, 1:1). Thirty polybags were filled up with each potting material, which was previously sterilized with 20 per cent formaldehyde. After the removal of traces of formaldehyde, the polybags were filled up. Each polybag was inoculated with surface sterilized seeds (2 per cent NaCl) of *J. curcas*. Out of 30 polybags of each potting material, 20 were used for AM inoculation while remaining 10 were kept as un-inoculated control. After sprouting of *J. curcas* seeds, the 20 g of mixed inoculum of AM fungi was put in the top of polybags and gently mixed. Single seedling was maintained in each pot. The height, collar diameter, root length, shoot length, average leaf area, and average number of leaves were recorded after two months of treatment. The data was analysed statistically.

Result and Discussion

The results of three treatments containing potting materials of fly ash, coal mines soil, and soil mix are recorded in tables 1, 2, and 3. It is evident from the tables that the VAM inoculation comparatively showed good performance in fly ash and coal mine dump soil on growth of *J. curcas* in nursery. The soil mix showed little more effect on AM performance as most of the growth parameters observed were comparatively significant. The different parameters like collar diameter, root length, shoot length, average leaf area, and number of leaves were recorded which have little variation. The number of leaves of *J. curcas* in fly ash and soil mix were equal and were more than coal mine dump soil. The leaf area in soil mix is more than in fly ash and coal mine soil. This may be possibly due to more nitrogen in soil mix as fly ash contain very less nitrogen (Juwarkar and Jambhulkar, 2008). Similarly root length and collar diameter of *J. curcas* seedlings was little more in fly ash which may also be due to phosphorus and mineral contents of fly ash (Prem Kishor, *et al.*, 2009). The VAM inoculation exhibited better growth performance of *J. curcas*. The un-inoculated control in all the three cases showed less

growth of *J. curcas* except shoot length being more in fly ash control. The experimental findings confirmed that AM inoculation plays a significant role in growth and development of *J. curcas* plants. Probably the nutrient and carbon contents in fly ash and coal soil are significantly mobilized by AM fungi. Effect of AM fungi on growth of *J. curcas* in nursery using fly ash, coal mine dump soil and nursery soil, mix are recorded.

Table 1 The effect of AM fungi on growth of *J. curcas* plants in Fly ash

Parameters	Observations	
	Treated seedlings (VAM inoculated)	Untreated seedlings (Control)
Root length(cm)	10.4 ± 1.8	7.6 ± 1.3
Shoot length(cm)	20.5 ± 2.4	17.2 ± 0.9
No. of leaves	6 ± 1.0	4 ± 0.8
Collar diameter (cm)	3.1 ± 0.3	1.6 ± 0.3
Average leaf area (sq. cm)	52.25 ± 0.64	43.5 ± 2.44

Values given in the table is mean ± SEM of all the five replicates; Amount of inoculums used in In vivo studies.

Table 2 The effect of AM fungi on growth of *J. curcas* plants in coal mine dump soil

Parameters	Observations	
	Treated seedlings (VAM inoculated)	Untreated seedlings (Control)
Root length(cm)	9 ± 1.5	7.3 ± 1.1
Shoot length(cm)	18.3 ± 2.6	14.6 ± 1.7
No. of leaves	5 ± 0.9	4 ± 0.5
Collar diameter (cm)	2 ± 0.1	1.5 ± 0.2
Average leaf area (sq. cm)	54.8 ± 1.0	42.6 ± 1.7

Values given in the table is mean ± SEM of all the five replicates; Amount of inoculums used in In vivo studies.

Table 3 The effect of AM fungi on growth of *J.curcas* plants in nursery soil mix (1:1)

Parameters	Observations	
	Treated seedlings (VAM inoculated)	Untreated seedlings (Control)
Root length(cm)	8.9 ± 0.9	7.1 ± 0.9
Shoot length(cm)	21 ± 1.4	16.4 ± 0.8
No. of leaves	6 ± 0.6	4 ± 0.5
Collar diameter (cm)	2.8 ± 0.2	1.7 ± 0.3
Average leaf area (sq. cm)	65.7 ± 2.3	48.08 ± 1.8

Values given in the table is mean ± SEM of all the five replicates; Amount of inoculums used in In vivo studies.

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Maize and Groundnut Prefer Different Form of Phosphorus on P-fixing Soil

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Materials and Method

Experiments were carried out using a factorial design with and without benomyl on the P fertilized plots of the previous year. The P fertilization levels were: P-0, P-50, and P-400 mg kg⁻¹ soil. A fresh application of phosphate was made only on the P-400 plots during 2000-2001, to assure maximum growth and high influx. Fifteen days before sowing of crops, benomyl was incorporated at the rate of 500 kg ha⁻¹. Nitrogen and potassium were applied to all the plots by broadcasting at the rate of 120 kg ha⁻¹ and 50 kg ha⁻¹ for maize, and 25 kg ha⁻¹ and 50 kg ha⁻¹ for groundnut in the form of urea and muriate of potash. Micro nutrients Zn, B, Mo, and Cu were applied at the recommended doses. After levelling the field plots, seeds of two crops were sown in rows keeping the row to row spacing at 50 cm for maize (cv.DDH103) and 30 cm for groundnut (cv.AK 12/24). Plant to plant distance was 25 cm for maize and 20 cm for groundnut. Plot size for both treated and untreated benomyl treatment was 1.25 x 2 m for the first three harvests and 2 x 2 m for the final harvest (at maturity). Four harvests were made for each crop; at each harvest, soil and plant parameters such as soil solution concentration, shoot yield, P content in shoot, available P, root length, root radius, and root hair were determined.

The fractionation procedures (Chang and Jackson, 1957) are based on the differential solubility of various inorganic P forms in various extracts. Ammonium chloride (NH₄Cl) is used first to remove soluble and loosely bound P, followed by separating Al-P from Fe-P with NH₄F, and then removing Fe-P with NaOH. The reductant-soluble P is removed with CDB.

Root infection was assessed on a representative root sample taken from each plot at each harvest. At harvest, roots to a depth of 15 cm were taken from plants in fixed positions evenly distributed over each plot. The roots from each plot sample were separated, washed free of soil, and cut into 1 cm –1.5 lengths. Root samples were stained with trypan blue

(Philips and Hayman, 1970). AM infection of each plant was determined by estimating the percent root colonization as described by Bierman and Linderman (1981). Alkaline hydrolysis of root samples with 10 per cent potassium hydroxide was done at 90°C in an oven for 8 to 10 minutes to clear the plant cytoplasm depending on the stiffness of the root. The roots were then washed in several changes of water and then treated with 1N hydrochloric acid for 10 minutes and ultimately stained with 0.05% trypan blue (made in lactophenol) for about 24 hours. A minimum of 50 root fragments were examined at each time.

Per cent root infection was obtained as follows :

$$\text{Root infection (\%)} = \frac{100 \times \text{number of intersections with AM infection}}{\text{Total number of intersections counted}}$$

Result and Discussion

The results pertaining to P uptake by maize and groundnut have been presented in Table 1. Phosphorus uptake increased with P fertilization for both species. At limiting P supply i.e., P-0, maize absorbed almost twice the amount of P compared to groundnut which suggests that maize had greater P uptake efficiency. In maize, at the start of the growing seasons P uptake was found to be only one seventh to one eighth of groundnut in both the years; but subsequently, P uptake in maize got accelerated right upto the final harvest. In groundnut, P uptake rate increased consistently throughout the growth period and upto maturity.

It was further observed that in maize (Table 2), Fe-P was reduced to 20.4 mg kg⁻¹ soil at final harvest, where the initial value was 62 mg kg⁻¹ soil at the start of the experiment. This indicates that maize took high amount of P from Fe-P. The uptake was different in groundnut. In groundnut (Table 2) most of the P was obtained from Al-P fraction and there was a reduction of 35.2 mg kg⁻¹ soil from an initial value of 55 mg kg⁻¹ soil as observed after the harvest of groundnut. It can be stated that maize and groundnut can take more P from Fe and Al-P due to its additional ability to desorb P from sparingly available P sources. Both

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these crops might have increased the concentration of P in soil solution and therefore its uptake efficiency through chelation, dissolution, and occupation of P-sorption sites, and/or by ligand exchange (Jungk *et al.*, 1993). Gahoonia and Nielsen (1996), reported that depletion of P fractions in the rhizosphere varies with plant species and soil type.

It was observed that in both the crops, P uptake was significantly more in the untreated plot than in benomyl-treated plot in both years (Table 1). Benomyl application had almost no effect on P concentration in the shoot and P uptake was closely related to dry matter production. The difference in P uptake between treated benomyl and untreated plot can be due to the effects of infection (Table 3) of the host plant of untreated plots by AM fungi which may increase P uptake (Koide, 1991) due to its capacity to absorb phosphate from soil and transfer it to the host roots. The survival and proliferation of indigenous AM in improving P uptake have been suggested (Wilcox, 1996) to be dependent on their

association with cultivated plant species. It can be observed from P fractionation data (Table 2) that in maize crop, Fe-P fraction in benomyl treated plot was more than in untreated benomyl plot indicating less acquisition of Fe-P fractions by the maize plant. In case of groundnut, the Al-P fraction was considerably higher in benomyl treated plot as compared to that in untreated benomyl plot. The other P fractions were more or less same in benomyl treated and untreated benomyl plot. A higher P fraction value was always observed in benomyl treated plots of both the crops.

There was no significant interaction effect in relation to benomyl and phosphorus application on the uptake of P except at final harvest of maize in both the years. The effect of benomyl at high level of phosphorus application (P-400) showed negligible influence on P uptake in both the crops. With increasing P level, the effect of benomyl was masked which implies that mycorrhiza might have functioned effectively upto some intermediate P levels in the present experiment.

Table 1 Effect of benomyl on P uptake of maize and groundnut at no P (P-0), 50 mg P kg⁻¹ (P-50), and 400mg P kg⁻¹ (P-400) application to the soil

Maize (Mg m ⁻²)														
P levels (mg kg ⁻¹ soil)	25 DAS			47 DAS			81 DAS			124 DAS				
	Benomyl													
	-	+	Avg.	-	+	Avg.	-	+	Avg.	-	+	Avg.		
0	3	1.4	2.2	25.1	13.1	19.1	849	586	718	1376	1038	1207		
50	4.7	3.4	4.05	36	22.1	29.1	1233	953	1093	1611	1407	1509		
400	34	32.4	33.2	445	414	429.5	3581	3022	3302	5010	4663	4735		
Average	13.9	12.4		168.7	149.7		1888	1520		2667	2369	1706		
SEm LSD(0.05)														
P		0.5		1.5		7.5		22		106		320	52.9	159
B		0.4		1.2		6.2		19		87		261	43.5	130
PxB		0.8		NS		9.9		NS		147		NS	74.8	225
Groundnut														
	-	+	Avg.	-	+	Avg.	-	+	Avg.	-	+	Avg.		
0	21	10.5	15.8	116	87	102	399	320	360	857	756			
50	28	17.2	23	129	99	114	543	476	510	1171	1130			
400	83	73	78	144	119	132	959	855	907	2074	1965			
Average	44	33.6		130	102		634	550		1367	1284			
SEm LSD(0.05)														
P	1.9	5.8		5.7		17.4		16.8		51		37	112	
B	1.5	4.7		4.6		13.4		13.8		42		30	90	
PxB	2.7	NS		8.0		NS		23.8		NS		54	NS	

Table 2 Phosphorus fractionation (mg kg⁻¹) soil at harvest of Maize and Groundnut at no P (P-0), 50 mg P kg⁻¹ (P-50), and 400 mg P kg⁻¹ (P-400) application to the soil

P levels (mg kg ⁻¹ soil)	Maize				Groundnut			
	Initial		Final		Initial		Final	
	-	+	-	+	-	+	-	+
	Benomyl							
	Saloid-P							
0	3.1		2.9	2.7	4.6		3.4	3.4
50	3.6		3.0	2.8	5.2		4.6	4.2
400	6.3		21.4	21	8.2		21.3	20.9
	Al-P							
0	48.4		45.3	45.9	27.5		19.8	25.9
50	97		113	116	90		96.8	114.5
400	229		259	268	234		284	289
	Fe-P							
0	31.6		20.4	30.2	48.2		41.6	42.3
50	85		69	79	95		87.5	91.2
400	213		241	245	247		267	270
	Ca-P							
0	25.5		24.9	25	25.7		25.1	25.0
50	27.5		28.1	28	28.5		29.2	28.5
400	30.4		31.2	30.8	30.4		31.2	31.0

Table 3 Effect of benomyl on root infection (%) of maize and groundnut at no P (P-0), 50 mg P kg⁻¹ (P-50), and 400 mg P kg⁻¹ (P-400) application to the soil

Days after sowing	Crop	Benomyl (B)					
		Without			With		
		P-0	P-50	P-400	P-0	P-50	P-400
25	Maize	18	14	3	5	2	0
		LSD(0.05) B = 1.87, P = 2.29, Interactions = 3.25					
47		32	20	9	11	7	0
		LSD(0.05) B = 2.71, P = 3.35, Interactions = 4.73					
81		50	44	13	20	13	8
		LSD(0.05) B = 3.44, P = 4.19, Interactions = 5.94					
At maturity		34	32	14	14	10	7
		LSD(0.05) B = 3.71, P = 4.52, Interactions = 6.42					
30		56	43	0	0	0	0
		LSD(0.05) B = 2.42, P = 2.95, Interactions = 4.20					
50	Groundnut	43	40	6	6	0	0
		LSD(0.05) B = 3.04, P = 3.74, Interactions = 5.29					
68		36	30	10	8	6	0
		LSD(0.05) B = 3.88, P = 4.70, Interactions = 6.67					
At maturity		34	29	11	24	12	12
	LSD(0.05) B = 4.31, P = 5.45, Interactions = 7.78						

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SEVENTH INTERNATIONAL CONFERENCE ON MYCORRHIZA: A REPORT

The Seventh International Conference on Mycorrhiza (ICOM 7) was held from 6 to 11 January, 2013 at the India Habitat Centre, New Delhi. Under the auspices of the International Mycorrhiza Society and in collaboration with Mycorrhiza Network, the conference was successfully organized by The Energy and Resources Institute (TERI). This was the first time that a conference of this stature had taken place in Asia.

ICOM 7 received an overwhelming response with participants pouring in from across 48 countries. Shri Jaipal Reddy, Union Minister for Science and Technology and Earth Sciences, inaugurated the event by lighting the traditional lamp along with Dr Ashok Gulati, Chairperson of the Commission for Agriculture Costs and Prices; Dr R R Sinha, Advisor/Scientist G with Department of Biotechnology; Dr Melanie Jones, President of the International Mycorrhiza Society; Dr R K Pachauri, Director-General, TERI; and Dr Alok Adholeya, Director of the Biotechnology and Bioresources Division, TERI. The inaugural ceremony witnessed the release of TERI's field manual, a database compiling 18 years of the Centre for Mycorrhizal Research's (CMR) field trials of AMF inoculation in different crops across various agro climatic zones in India and other countries. The launch of the Centre for Mycorrhizal Culture Collection's (CMCC) website — an online databank housing more than 600 different isolates of ecto and arbuscular mycorrhizal fungi along with seed coating technology developed by TERI — were major highlights of the conference.

With the theme "Mycorrhiza for All: An Under-Earth Revolution", the conference covered various aspects of mycorrhizal symbiosis and their role in a wide range of areas covering ecosystem dynamics, ecology, nutrient cycling, cellular interactions, gene expression, metabolic regulation, multi-trophic interactions, agriculture applications, and reclamation technologies. Highlights of the conference included keynote speeches, conference symposia, poster presentations, workshops, trade exhibitions, an industrial special session, and visits to fields that had received mycorrhiza product in real-time field demonstration mode.

Plenary Sessions

The Four Plenary Sessions Covered the Following Themes:
Session 1: Developmental, functional, and environmental genomics

Session 2: Population, community, and physiological ecology

Session 3: Physiology, including carbon and nutrient exchange between symbionts and the saprotrophic/biotrophic continuum

Session 4: Mycorrhizae in agriculture and horticulture; including inoculation of seedlings, inoculum production, and policy development

The keynote speakers, Dr Francis Martin, INRA France; Dr Uwe Nehls, Bremen University; Dr Ian Dickie, Landcare Research; Dr J André Fortin, Université Laval; along with various other plenary and workshop speakers gave an enriching talk to around 400 audiences, who had gathered from various corners of the world to discuss the most prevalent form of symbiosis on Earth. The 13 workshops further gave an insight into various aspects of Mycorrhizal research.

The major highlights of the summit included:

- Organization of ICOM by India for the first time
- Participation from approximately 400 participants from all over the world
- Presentations by 120 speakers (on diverse topics) in plenary sessions and workshops
- Display of 210 posters from across the world. One-day field trip to showcase farmers' field demonstration trials
- Proceedings of the conference
- Trade exhibitions

The cultural evening on 7 January 2013 began with the Hall of Fame Awards ceremony, felicitating early pioneers of mycorrhizal research. Following this was the 'Confluence', which gave the participants glimpses of India's cultural diversity through the folk dances organized for the evening. ICOM has a tradition of holding the 'Wines of the World' event, in which each of the delegates present wines made in their country that are tasted and rated by co-delegates.

The valedictory ceremony was graced by Mr J K Daddo, Joint Secretary with the Ministry of Commerce and Industry. The eight awards for various Poster and Oral presentations were also distributed during the session. Concluding remarks from Dr R K Pachauri and Dr Alok Adholeya marked a successful end of ICOM 7.



CENTRE FOR MYCORRHIZAL CULTURE COLLECTION

Centre for Mycorrhizal Culture Collection: "A Visualization"

Chaitali Bhattacharya* and Alok Adholeya#

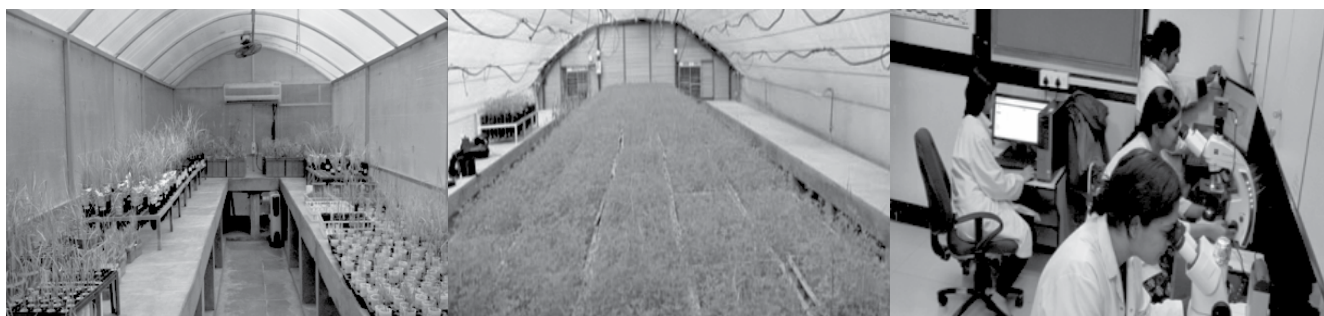
The official website for Centre for Mycorrhiza Culture Collection (CMCC) was inaugurated during the 7th International Conference on Mycorrhiza (ICOM 7), which was held from 6 to 11 January 2013 at New Delhi. The website was inaugurated by Shri Jaipal Reddy, Hon'ble Union Minister for Science, Technology, and Earth Sciences. This website opens opportunities to all researchers and industrialists occupied in the field of arbuscular mycorrhiza fungi or ectomycorrhiza fungi to access the rich culture collection that the bank houses with it. This article is an overview to the website so that it could be used proficiently by all interested mycorrhizologists. As any conventional website, CMCC has also been divided into a home page that provides an overview of the organization, the services it delivers, and finally the list of cultures that it maintains, which are available to our esteemed patrons.

CMCC is a mycorrhizal bioresources centre that aims at the conservation of mycorrhizal biodiversity by means of collection, propagation, isolation, characterization, and maintenance of cultures under in-situ conditions. These mycorrhizal cultures are further sterilized and successfully brought into in-vitro condition for mass propagation and preservation. Our endeavour is to preserve mycorrhizal biodiversity that shows up in morphology, physiology, genetics,

and functionality. The objective is to study the rich germplasm that exists among nature and perpetuates them under in-situ as well as in-vitro conditions so that we are capable of supplying them to researchers and industry for its apposite application. The Centre for Mycorrhizal Culture Collection (CMCC) was established in 1993 with seed support from the Department of Biotechnology, Government of India. Since then, the bank has a glorious collection of above 700 different isolates of which 257 are Ectomycorrhizal Fungi (EMF) and over 350 are Arbuscular Mycorrhizal Fungi (AMF) isolates collected from different parts of the globe.

The Culture Collection Bank is the largest, and to date only, collection bank for mycorrhiza in India. The facility includes three temperature-controlled greenhouses dedicated solely towards culture development and maintenance. The cultures are given special attention and examined on a daily basis by manual watering, fertilization, and de-weeding as required. Our facility embraces the state-of-the-art laboratories with contemporary equipment which include work areas to accommodate three major ranges of activities:

- **Classical identification/Morphotaxonomic identification lab:** This lab is engaged in conduction-specific activities pertaining to



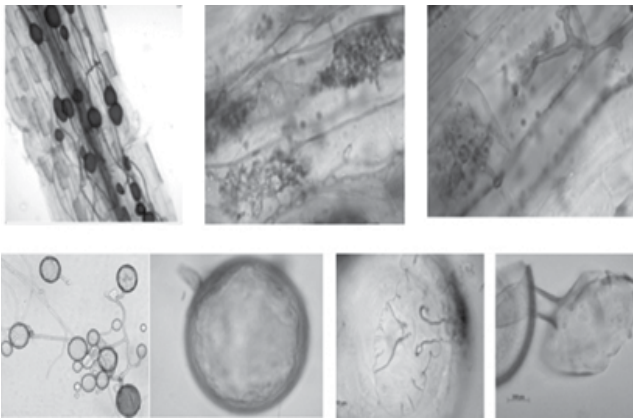
Infrastructure

* Research Associate, Biotechnology and Bioresources, Centre for Mycorrhizal Research, The Energy and Resources Institute, Darbari Seth Block, IHC Complex, Lodhi Road, New Delhi-110 003, India

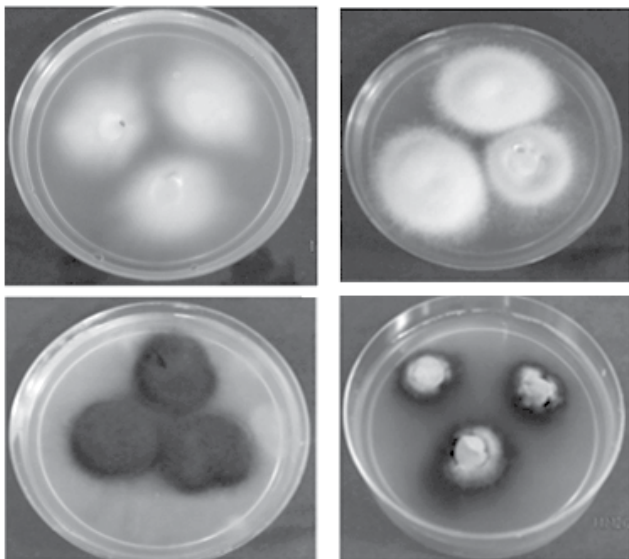
Director, Biotechnology and Bioresources, Centre for Mycorrhizal Research, The Energy and Resources Institute, Darbari Seth Block, IHC Complex, Lodhi Road, New Delhi-110 003, India

the collection, identification and isolation of monosporal establishment. The lab also has a microscopy-photography area in it with software installed for doing wall layer analysis and measurements.

- **Molecular identification lab:** Once the monosporal have been identified on morphotaxonomic basis they are counter asserted using molecular tools. The lab has modern equipment like Real Time PCR, Hybrid Multi Reader, and Gel Documentation Machine, etc.



Arbuscular Mycorrhizal Fungi (AMF)



Ectomycorrhizal Fungi (EMF)

- **Biochemical identification:** Lipid profiling of the spores of Arbuscular Mycorrhiza Fungi is done using Gas Liquid Chromatography. The Fatty Acid Methyl Ester (FAME) database is created for all the pure cultures that are maintained in the lab.

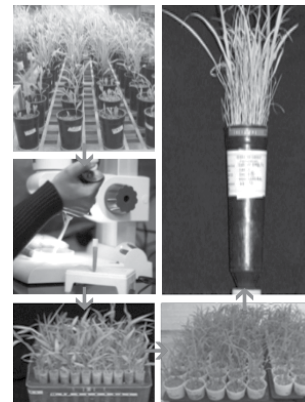
Services

We offer a number of services pertaining to both Arbuscular Mycorrhizal Fungi (AMF) as well as Ectomycorrhizal Fungi (EMF) for amateur as well as professionals.



Deposit and Safeguarding Services

Donors can deposit cultures and receive accession codes for their cultures. The bank can assist by growing and safeguarding these cultures for individuals who are unable to do the same due to lack of knowledge or lack of facilities. For the collection protocol to be followed, please refer to the October 2011 article of Mycorrhizal News.



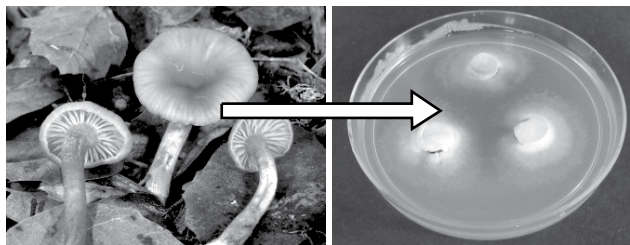
Purification Services

For individuals who are interested in analysing and characterizing the diversity that exists in their deposit, the bank has the expertise to purify their culture.

Monosporal of AMF: Soil samples of the individual are commenced and maintained as trap cultures to enhance the AMF spore population using different bait plants. Once these reference cultures are established and show good health, the spores are screened. Screening is done on the basis of morphotaxonomy, and scrutinizing of the diversity is done in order to generate pure cultures (Monosporal).

Monosporal are initiated by attempting a symbiotic relationship between single spore of AMF with single pre-germinated seed of sorghum vulgare placed in a micropipette tip. A routine check to evaluate the spore count and colonization status is done on monthly intervals. Once the monosporal culture shows high sporulation and colonization, it is designated as a successful culture.

Pure cultures of ectomycorrhiza: Sporocarp or fruiting bodies of Ectomycorrhiza are collected or



Laccaria scaber

received from the depositor. A small portion of the sporocarp is surface sterilized and then placed on a suitable medium for its growth and proliferation. Once the mycelial growth establishes itself, it is constantly subcultured and purified till a pure culture is achieved.

Substantiation Services

■ Identification of Mycorrhizal Reference

Material: The bank nurtures the expertise of identifying the mycorrhizal diversity using different conventional and modern tools as follows:

Morphotaxonomic Characterization: For Arbuscular Mycorrhizal Fungi, conventional methods are adopted that include preparing voucher specimen using Polyvinylalcohol (PVLG) and Melzer Reagent. Microscopic studies of spore characters such as wall layers, colour reaction with Melzer Reagent, mycelia attachment, and presence or absence of septum, etc., are recorded.

For ECM, morphotype analysis of the hyphal sheath is done since they differ in appearance. The hyphal sheath can vary in colour, branching, shape of the hyphae in the soil, appearance of the rhizomorph, and the structure of the inner surface.

Molecular Characterization: Molecular characterization is one of the most promising tools for authentication of both ecto- and endomycorrhiza. The DNA extraction of pure cultures is carried out using total DNA extraction methods. PCR amplification of partial sequences of rDNA, which include partial sequence of SSU, complete sequence of ITS1, 5.8 S rDNA, ITS2, and partial sequence of LSU is done using gene specific primers. Similarly, PCR amplification of partial sequence of the mitochondrial LSU (mtLSU) rDNA is done using gene specific primers. These partial sequences are cloned, sequenced, and analysed with the large sets of sequences available in the database and its phylogenetic analysis is carried out to characterize and identify the culture identity.

Biochemical Characterization: Fatty Acids Methyl Esters (FAME) profiles using lipids present in the

spore of AMF of individual isolates collected from different regions are generated using gas liquid chromatography. The abundance of lipids in spores and vesicles of AMF colonized is a potentially useful biochemical character for taxonomic purposes and quantification of glomalean fungi. The bank, with the wide diversity that it is preserving, has created an extensive fatty acid profile library that has helped in understanding the biochemical biodiversity that exists among different mycorrhiza collected from different regions. The quantitative and qualitative differences in the fatty acid composition in the spores among different isolates can be done by generating Fatty Acid Methyl Esters (FAME) profiles using gas liquid chromatography.

Supply of Culture

The cultures that are purified and authenticated by the bank are maintained under two categories:

- Pure non-sterile spores (In-situ) of Arbuscular Mycorrhizal Fungi.
- Cultures are provided as starter cultures where a specific number of spores are mixed with soil substrate.
- Pure culture (in-vitro) of Ectomycorrhiza.
- Cultures are provided in petri plates as pure mycelial mat (two replicates).

The list of both AMF and EMF, along with the accession codes, is provided on the website http://mycorrhizae.org.in/cmcc/prod_res.php. The list is constantly updated as per the new cultures establishment. Interested individuals can place their requirements by filling the culture request form. On receiving the request, the curator of the bank reverts back to the queries of the individual. The mandate of the Centre for Mycorrhizal Culture Collection is to search for functionally superior mycorrhizal isolates which can provide notable benefits to mankind. It can be achieved only with help from the industry and researchers who would be able to use these Arbuscular Mycorrhiza Fungi and Ectomycorrhiza Fungi germplasm with desired and innovative focus.

RECENT REFERENCES

The latest additions to the network's database on Mycorrhiza are published here for the members' information. The list consists of papers from the following journals:

- *Applied Soil Ecology*
- *Basic and Applied Ecology*
- *Environmental and Experimental Botany*
- *Environmental Pollution*
- *European Journal of Soil Biology*
- *Journal of the Saudi Society of Agricultural Sciences*
- *Pedobiologia*
- *Perspectives in Plant Ecology, Evolution, and Systematics*
- *Review of Palaeobotany and Palynology*
- *Science of The Total Environment*
- *Soil and Tillage Research*
- *Soil Biology and Biochemistry*

Name of the author(s) and year of publication	Title of the article, name of the journal, volume number, issue number, page numbers (address of the first author or of the corresponding author is marked with an asterisk)
Brito I*, Goss M J, Carvalho M d, Chatagnier O, and Tuinen D v. 2012	Impact of tillage system on arbuscular mycorrhiza fungal communities in the soil under Mediterranean conditions <i>Soil and Tillage Research</i> 121: 63–67 [*Universidade de Évora – ICAAM, Apartado 94, 7002 – 554 Évora, Portugal]
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Cornejo P*, Pérez-Tienda J, Meier S, Valderas A, Borie F, Azcón-Aguilar C, and Ferrol N. 2013	Copper compartmentalization in spores as a survival strategy of arbuscular mycorrhizal fungi in Cu-polluted environments <i>Soil Biology and Biochemistry</i> 57: 925–928 [*Departamento de Ciencias Químicas Recursos Naturales, Scientific and Technological Bioresource Nucleus, BIOREN-UFRO, Universidad de La Frontera, Casilla 54-D, Temuco, Chile]
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Kończak P*, Zubek S, Błaszczkowski J, Mleczko P, and Margielewski W. 2013	Erosion or plant succession — How to interpret the presence of arbuscular mycorrhizal fungi (Glomeromycota) spores in pollen profiles collected from mires <i>Review of Palaeobotany and Palynology</i> 189: 29–37 [*Department of Biogeography and Palaeoecology, Faculty of Geographical and Geological Science, Adam Mickiewicz University, ul. Dziejniowa 27, 61-680 Poznań, Poland]

Name of the author(s) and year of publication	Title of the article, name of the journal, volume number, issue number, page numbers (address of the first author or of the corresponding author is marked with an asterisk)
Koorem K*, Saks Ü, Sõber V, Uibopuu A, Öpik M, Zobel M, and Moora M. 2012	Effects of arbuscular mycorrhiza on community composition and seedling recruitment in temperate forest understory <i>Basic and Applied Ecology</i> 13(8): 663–672 [Department of Botany, Institute of Ecology and Earth Sciences, University of Tartu, Lai 40, 51005 Tartu, Estonia]
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FORTHCOMING EVENTS

CONFERENCES, CONGRESSES, SEMINARS, SYMPOSIUMS, AND WORKSHOPS

- Asilomar, USA
12-17 March 2013
- 27th Fungal Genetics Conference**
Anne Marie Mahoney, Genetics Society of America, 9650 Rockville Pike, Bethesda, Maryland 20814
Tel. 301-634-7039
E-mail: Mahoney@genetics-gsa.org
Website: <http://www.fungalgenetics.org/2013/>
- Singapore
18-19 March 2013
- 3rd Annual International Conference on Advances in Biotechnology BIOTECH 2013**
Global Science & Technology Forum (GSTF), 10 Anson Road, International Plaza, Singapore 079903
Tel: +65 6327 0166 Fax +65 6327 0162
E-mail: secretariat@advbiotech.org, info@advbiotech.org
Website: <http://www.advbiotech.org>
- Sitges, Spain
13-15 May 2013
- BITE2013 — 3rd International Conference on Bio-Sensing Technology**
Tel. +86 10 8520 8673
E-mail: r.chi@elsevier.com, customerservicebiosensingtech13@elsevier.com
Website: <http://www.biosensingconference.com/>
- Brno, Czech Republic
20-25 May 2013
- Biosecurity in natural forests, stands and plantations, genomics and biotechnology for biosecurity in forestry**
Dpt. of Forest Protection and Wildlife Management, Faculty of Forestry and Wood Technology, Mendel University, Zemedelska 3, 613 00 Brno, Czech Republic
Tel. +420 739 341 961
E-mail: jankov@mendelu.cz
- Asheville, Nc, United States
26 May-1 June 2013
- Tree Biotechnology 2013 Conference - Forest Biotechnology: Meeting the Needs of a Changing World**
E-mail: mkirst@ufl.edu & jeffdean@uga.edu
Website: <http://forestbiotech.org/news/2012/may-26-june-1-2013-tree-biotechnology-2013-conference-asheville-nc>
- Australian National University, Canberra, Australia
1-4 July 2013
- 8th New Phytologist Workshop: Improving representation of leaf respiration in large-scale predictive climate-vegetation models**
E-mail: symposia@lancaster.ac.uk5
Web site: <http://www.newphytologist.org/workshops/view/3>
- La Antigua Guatemala, Guatemala
29 July-3 August 2013
- The 7th International Workshop on Edible Mycorrhizal Mushrooms -IWEMM-7**
Dr. Roberto Flores Arzú, Director, Instituto de Investigaciones Químicas y Biológicas -IIQB-Edificio T-13 Facultad de Ciencias Químicas y Farmacia, Universidad de San Carlos de Guatemala, Código Postal 01012
Tel. 00(502) 53099177
E-mail: iwemm7@gmail.com
Website: <http://sitios.usac.edu.gt/iwemm7/>
- Greece
1-4 September 2013
- 7th EPSO Conference**
E-mail: lisa.jochum@epsomail.org
Website: <http://www.epsoweb.org/7th-epsos-conference-1-4-september-2013-greece>
- Cornell University, Ithaca, NY USA
9-10 September 2013
- 7th New Phytologist Workshop: Frontiers in chemical ecology and coevolution**
E-mail: np-symposia@lancaster.ac.uk
Website: <http://www.newphytologist.org/workshops/view/2>

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