Arbuscular mycorrhizal association in some ethnobotanical plants of Tripura

ATITHI DEBNATH, AJAY KRISHNA SAHA, AND PANNA DAS

J. Mycopathol, Res, 54(2) : 239-244, 2016;
ISSN 0971-3719

Department of Botany,
University of Calcutta,
Kolkata 700 019, India

J. Mycopathol. Res. ***54(2)*** *: 239-244, 2016;* (ISSN 0971-3719)

Arbuscular mycorrhizal association in some ethnobotanical plants of Tripura

ATITHI DEBNATH[1]*, AJAY KRISHNA SAHA[2], AND PANNA DAS[1]
[1]Microbiology Laboratory, Department of Botany, Tripura University, Suryamaninagar 799 022, Tripura
[2]Mycology and Plant Pathology Laboratory, Department of Botany, Tripura University, Suryamaninagar 799 022, Tripura

Received : 22.01.2016 RMS Accepted : 31.05.2016 Published : 25.07.2016

Arbuscular mycorrhizal fungi (AMF) form association with the roots of higher plants and provide a better pathway to obtain nutrition for their host plants. In this study fifteen ethnobotanical plants were observed for their mycorrhizal structures. All the plants exhibited AM fungal colonization. The hyphal colonization percentage was significantly higher than vesicles percentage and arbuscules percentage in all the species. The root length arbuscule percentage was highest in *Aquilaria malaccensis* and lowest in *Diodella samentosa* whereas absent in *Colocasia* sp. *Mimosa pudica* was observed maximum for root length percentage of vesicle and lowest by *Lindernia crustacea. A. malaccensis* showed the highest percentage of hyphal colonization and *Colocasia* sp. represented the lowest hyphal colonization percentage. The study reveals that ethnobotanical plants are colonized by AM fungi and its association may play an essential role in nutrition of these plants

Key words: AM fungi colonization, ethnobotanical plants

INTRODUCTION

Mycorrhizae are a group of fungi that establishes mutualistic symbiosis with a majority of plant species Among these associations, Arbuscular mycorrhizal fungi (AMF) belonging to the Phylum Glomeromycota, are the most prevalent one and main component of soil microbiota and probably represent the most important terrestrial symbiosis (Fitter, 2005). It is an interaction where both partners benefited primarily from the exchange of nutrients i.e., mycorrhizal fungi gets a carbon substrates from plants and in turn the plants are provided with nutrients. They can improve plant establishment and survival, enhance plant nutrient uptake, reduce the negative effects of various biotic or abiotic stresses, and improve soil structure (Smith and Read, 2008). Mycorrhizal symbiosis is universally distributed among the majority of plants and forms a network of extra-radical mycelium that provides a direct physical link between the plant root and the soil (Smith and Read, 2008). Arbuscular mycorrhizal fungi are thought to be more efficient at scavenging for soil nutrients, owing to their larger surface-to-volume ratios. This

*Corresponding author: atithi16dn@gmail.com

ability is particularly important for acquisition of P, which is relatively immobile in soil.

Ethnobotany is a multidisciplinary science defined as the interaction between plants and people. The relationship between plants and human cultures is not limited to the use of plants for food, clothing and shelter but also includes their use for religious ceremonies, ornamentation and health care. The focus of ethnobotany is on how plants have been or are used, managed and perceived in human societies and includes plants used for food, medicine, divination, cosmetics, dyeing, textiles, for building, tools, currency, clothing, rituals, social life and music (Choudhary *et al,* 2008). Ethnobotanical studies have become increasingly precious in the development of modern health care and conservation programs in different parts of the globe.

Tripura, a small hilly state is situated in the southern most part of North-east states. It is a land-locked state and its geographical limits touch both national and international boundaries. It shares its international boundary line with Bangladesh and national boundaries with Assam and Mizoram (Sharma *et al,* 2013). Tripura is rich in its plant wealth and has 379 tree species, 320 shrubs, 581 herbs 165 climbers, 16 climbing shrubs, 35 ferns and 45 epiphytes (Kshirsagar and Upadhaya, 2009). There are about 19 ethnic groups namely Tripuri, Jamatia, Reang, Noatia, Chakma, Bhil, Bhutia, Chaimal, Garo, Halam, Khasia, Kuki, Lepcha, Lushai Mag, Munda, Kaur, Orang, Santhal and Uchai. Among them, Tripuri and Reang are the major groups. Different ethno-medicinal surveys were conducted by different researches (Shil *et al,* 2009; Majumdar *et al,* 2006; Majumdar and Dutta, 2007; Das *et al,* 2009). But no survey was conducted on the mycorrhizal status of the ethnobotanical plants of this state.

MATERIALS AND METHODS

Selection of ethnobotanical plants

Plants were selected on the basis of established studies and also by survey among tribal peoples. Study has been carried out in several time intervals during the period of 2013–2014 in different tribal villages. At first phase, using different established studies, a list of different species was listed. Then ethnobotanical information on different purposes of plants was collected through interviewing local informants The local informants were men and women of tribal villages.

Collection of root and soil samples

For the assessment of association of mycorrhiza root samples from different ethnobotanical plant species were collected by digging depth of 20 cm. Fine roots were collected from the rhizosphere. The soil samples were collected at 0–20 cm depth around each species and approximately 200 g soil per plant was collected. All the soil samples from each location were combined and collected in polythene bags, tagged and were brought to the laboratory for soil and spore analysis. Ethnobotanical plants were collected from Kunjaban, Takhmachara and Suryamaninagar sites.

Determination of soil physico-chemical properties

The pH and electrical conductivity were determined by taking 10 g of soil dissolved in 50 ml distilled water and stirred for 20 mins and kept it for overnight. Measurement of the soil pH and electrical conductivity were determined using a digital pH meter and conductivity meter. The organic carbon was estimated by using Walkley-Black (1934) method. The soil available Nitrogen was estimated following Black (1982) method. Available Phosphorus of soil was determined using Jackson (1978) method.

Assessment of AM fungal association

The collected roots were prepared by thoroughly washing them in tap water several times and cut into approximately 1cm long pieces. Then the roots were cleaned with 10% NaOH at 350°C (Stuart UC210) for 24 hrs depending on the root structures. The cleared roots were washed again with tap water for 4-5 times and bleached in 2 drops of alkaline H_2O_2 for 5 mins. Roots were stained with Black Faber Castell stamp pad ink with 1-3 drops of 1% HCl followed by slight heating (Das and Kayang, 2008). After a while the roots were mounted in lactoglycerol on slide and observed under compound microscope (Olympus C X 21i) for mycorrhizal structures. The estimation of AM fungal colonization was done by the magnified intersection method (McGonigle *et al,* 1990).

Statistical analysis

Standard error of means was calculated by using Origin 0.6.

Table 1: Some selected ethnobotanical plants and their uses

Botanical name/ Family	Local name	Parts used	Uses	References
Aquilaria malaccensis Lamk. Thymelaeaceae	Agar (All)	Stem and branches	Both branches and stem are used as firewood. It is believed that it makes the departed soul pure if it is used for cremation.	Sharma *et al,* 2013
Cassia tora L. Caesalpiniaceae	Chakunda (R)	Leaf and seed	Worm infection, vision problem, liver disease, leprosy. Juice of the leaf is useful in worm infection; leaf and seed paste is applied topically as poultice in skin diseases like leprosy; decoction from full pant is also used in vision disorder and as liver tonic.	Sen *et al,* 2011
Clerodendrum viscosum Vent. Verbenaceae	Killiashak (C) Bhati pataa (K)	Root and leaf	Dried roots are grounded, mixed with water and kept overnight, which is useful in fever; paste of the leaf/root is applied to cure skin infection and reduce inflammation. Extract is used as expectorant. Decoction of the leaves is used to check high blood pressure. Root extract is administered 1 teaspoon thrice daily as febrifuge.	Sen *et al,* 2011; Das and Choudhury. 2012
Colocasia esculenta (L.) Schott, Melet. Araceae	Mwitu, Lati (K)	Leaf, whole and plant	Fever, respiratory disorder. Whole plant is used to make curry.	Deb *et al,* 2013
Diodella sarmentosa (Sw.) Bacigalupo & Cabral ex Bortidi Rubiaceae	Maduka (D)	Plant and leaf	Infertility, menstruation and pregnancy.	By interviewing tribal people
Chromolaena odorata (L.) King & H.E. Robins. Asteraceae	Mychongdi (K)	Tender leaves	Leaves are grinded and the paste is taken to stop bleeding from cuts and wounds.	By interviewing tribal people
Evolvulus nummularius L. Convolvulaceae	Bhui akra (C)	Whole plant	Used as a medicine for hysteria, to cure burns, cuts, wounds and scropion stings.	Jain, 1991
Hevea brasilensis (H. B. & K) Muell.-Arg. Euphorbiaceae	Rubber	Latex	Latex is collected from lower part of tree for marketing. Whole tree is used as fire wood after 30 years.	Sharma *et al,* 2013
Lindernia crustacean (L.) Muell. Scrophulariaceae	Khumsai (R)	Leaf	Leaf paste with lemon juice is given orally to cure excess bile secretion; also applied externally on ringworm and boils. Decoction of herb is given ½ teaspoonful twice a day for 7 to 21 days for the treatment of abdominal ailments. Paste of herb with cow' s urine is applied on cuts and wounds for early healings.	Panda and Mishra, 2011; Dangwal *et al,* 2010
Mimosa pudica L. Mimosaceae	Samsunduru (H) Dugjat lajari (C)	Root, leaf whole plant	Leaf paste applied on acne and pimples. Root extract @ 1 teaspoon twice daily in jaundice. Leaves and roots used in piles and fistula; leaf paste applied to hydrocele; leaf and stem used in scorpion sting. Also used in leprosy, burning sensation, fever.	Sen *et al,* 2011; Das *et al,* 2012; Das and Choudhury, 2012

Contd...

Oplismenus burmannii (Retz.) P. Beauv. Poaceae	Jhabra (K)	Leaf	Leafs are used as antitode of (venomous stings, bites, etc.); eye treatments; genital stimulants/depressants; pain-killer.	By interviewing tribal people
Phaulopsis dorsiflora (Retz.) Santapau Acanthaceae	Shiphaphak (R)	Whole plant	Dried and pulverized, is used as a dressing for wounds. Fresh juice of the plant is applied to sores.	By interviewing tribal people
Urena lobata L. Malvaceae	Wakkhansu buphang(K)	Leaves	Decoction of the leaf is taken twice daily to reduce blood pressure; and also is taken before sleep to relieve rheumatic pain and body ache (69%).	Das *et al,* 2012
Solanum torvum Sw. Prodr. Solanaceae	Khwmkha skam(R)	Fruit	Fruit is used to prepare curry.	Deb *et al,* 2013
Stephania japonica (Thunb.) Miers Menispermaceae	Dufai-u-che-na (K)	Leaf, stem	Juice of the leaf is used to treat urinary disorder; infusion of root is useful in abdominal pain and flatulence.	Sen *et al,* 2011; Das *et al,* 2012; Majumdar *et al,* 2006

C-Chakma; D-Darbang; H-Halam; K-Kakbarak; R-Reang

RESULTS AND DISCUSSION

Selection of ethnobotanical plants

Fifteen plants were so far collected which have ethnobotanical importance through use of their various plant parts. The uses of the plant parts along with their local name and references were presented in Table 1.

Soil physico-chemical properties

All the soil samples showed acidic pH. Of which rubber plantation Takhmachara 1 was highly acidic Electrical conductivity (EC) and available nitrogen was highest in Takhmachara 1 whereas lowest in Suryamaninagar and Kunjaban respectively. Organic carbon was highest observed in Suryamaninagar and lowest observed in Takhmachara 1. The quantity of available phosphorus was maximum in Takhmachara 2 and minimum in Suryamaninagar. The soil physico-chemical properties are given below in Table 2.

Root colonization of AM fungi

Root length colonization of some ethnobotanical plants was depicted in Table 3 and Fig 1. Arbuscules (%) was highest in *Aquilaria malaccensis* and lowest in *Diodella samentosa*. The structure of arbuscule was absent in *Colocasia* sp. The *Mimosa pudica* showed higest vesicle (%) and lowest in *Lindernia crustacea*. In *Aquilaria malaccensis* hyphal (%) was highest and *Colocasia* sp. showed the lowest hyphal (%).

In this investigation all the plant species exhibited AM fungal colonization. The AM fungal colonization in *A. malaccensis* plant was also reported by Turjaman *et al,* (2006). According to Uma *et al,* (2012) the AM fungi colonization was found in *C. viscosum*. *E. odoratum* showed colonization was recorded by Hemavani and Thippeswamy, (2013). The AM fungi colonization in *M. pudica* was also recorded by Sarwade *et al*, (2012); and Gupta *et al,* (2009). According to Deka *et al,* (1998) AM fungal infection in the roots of five-year-old rubber plantation ranged between 68 to 88 per cent on surface layers in different treatments which resembles with this study. In Kerala, Nair and Girija (1988) recorded highest AM fungal colonization in rubber (71 per cent) compared to other tree crops of economic importance. Debnath *et al,* (2014b) reported AM fungal colonization in *H. brasiliensis* which is higher than this study whereas, AM fungal colonization in *D. sarmentosa, O. barmani* and *L. crustacea* was lower than this observation. The colonization in roots of *A. malaccensis, C. viscosum, P. dorsiflora, E. odoratum* and *M. pudica* was higher in this study as compare to Debnath *et al,* (2014a). Edaphic factors or soil nutrient status might be responsible in the patterns and the developement of AM fungi.

Table 2. Soil properties of soils from the rhizosphere of some ethnobotanically important plants

Soil samples	pH	Electrical conductivity (cS cm^{-1})	Organic Carbon (%)	Available Nitrogen (Kg/ha)	Available Phosphorus (Kg/ha)
Kunjaban	5.35±0.49	84±35	9.41±2.09	11.14±2.7	0.67±0.08
Takhmachara 1	4.59±0.07	126±2	8.39±0.41	349.09±5.21	1.32±0.08
Takhmachara 2	5.1±0.02	99.5±0.5	7.39±0.14	344.99±3.11	1.87±0.11
Suryamaninagar	6.85±0.02	8±1.15	14.4±0.17	312.86±25.45	0.35±0.03

Table : 3 Arbuscular mycorrhizal (AM) fungal association of some ethnobotanical plants

Name of Plants	%RLA	%RLV	%RLH
Aquilaria malaccensis	19.95±3.08	14.85±3.01	88.39±2.46
Cassia tora	18.89±1.91	13.74±2.06	71.04±3.22
Clerodendrum viscosum	11.91±2.25	13.34±2.36	71.69±3.83
Colocasia sp.	0.0±0.00	2.44±0.72	18.88±3.54
Diodella samentosa	4.47±1.28	20.43±2.91	65.96±1.28
Eupatorium odoratum	18.23±2.24	15.42±2.78	71.2±3.55
Evolvulus nummularius	13.56±2.72	25.63±3.48	65.97±3.69
Hevea brasilensis	18.34±2.34	26.56±3.54	78.56±2.82
Lindernia crustacean	6.21±1.36	12.54±2.33	38.64±2.64
Mimosa pudica	9.45±2.49	33.60±3.84	60.46±3.23
Oplismenus burmannii	16.25±4.02	14.99±3.38	41.92±4.84
Phaulopsis dorsiflora	8.55±1.70	13.69±3.45	58.43±3.01
Urena lobata	12.27±2.60	30.98±3.50	70.6±4.37
Solanum torvum	13.73±2.05	21.16±3.58	77.46±3.16
Stephania japonica	11.52±2.16	21.37±3.83	71.99±3.61

%RLA= Root length of arbuscule percent %RLV= Root length of vesicle percent

%RLH= Root length of hyphal percent

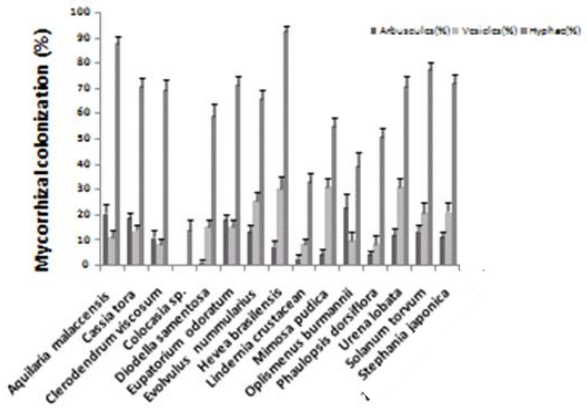

Fig.1 : Arbuscular mycorrhizal colonization in fifeen ethnobotanical plants

ACKNOWLEDGEMENTS

The authors are thankful to Head, Department of Botany, Tripura University for providing facilities. The first author is grateful to UGC for BSR fellowship.

REFERENCES

Black, C.A., 1982. *Methods of Soil Analysis.* Pregmon Press, England.

Choudhary, K., Singh, M. and Pillai, U., 2008. Ethnobotanical Survey of Rajasthan - An Update. *American-Eurasian Journal of Botany.* **1:** 38-45.

Dangwal, L.R., Sharma, A. and Rana, C.S., 2010. Ethno-medicinal Plants of the Garhwal Himalaya Used to Cure Various Diseases: A Case Study. *New York Science Journal.* **3:** 28-31.

Das, H.B., Majumdar, K., Datta, B.K. and Ray, D., 2009. Ethnobo-

tanical uses of some plants by Tripuri and Reang tribes of Tripura. *Natural Product Radiance*. **8:** 172-180.

Das, P. and Kayang, H., 2008. Stamp pad ink, an effective stain for observing arbuscular mycorrhizal structure in roots. *World Journal of Agricultural Science*. **4:** 58–60.

Das, S. and Choudhury, M.D., 2012. Ethnomedicinal uses of some traditional medicinal plants found in Tripura, India. *Journal of Medicinal Plants Research*. **6:** 4908-4914.

Das, S., Choudhury, M.D., Mandal, S.C. and Talukdar, A.D., 2012. Traditional knowledge of ethnomedicinal hepatoprotective plants used by certain ethnic communities of Tripura state. *Indian Journal of Fundamental and Applied Life Sciences*. **2:** 84-97.

Deb, D., Sarkar, A., Deb Barma, B., Datta, B.K. and Majumdar, K., 2013. Wild Edible Plants and Their Utilization in Traditional Recipes of Tripura, Northeast India. *Advances in Biological Research*. **7:** 203-211.

Debnath, A., Sinha, S., Saha, A.K. and Das, P. 2014a. Arbuscular mycorrhizal fungal diversity in the open land adjacent to rubber plantation in Tripura, Northeast India. *Mycorrhiza news*. **25 :** 2-7.

Debnath, A., Saha, A. K. and Das, P. 2014b. Arbuscular mycorrhizal in *Aquilaria malaccensis* Lamk. and its surrounding herbaceous community. *Mycorrhiza news*. **26 :** 4-9.

Deka, H.K., Philip, V., Vinod, K.K. and Krishnakumar, A.K., 1998. Spatial distribution of soil microflora in a five year old rubber plantation in Tripura. *Indian Journal of Natural Rubber Research*. **11**: 88–93.

Fitter, A.H., 2005. Darkness Visible: Reflections on Under- ground Ecology. *Journal of Ecology*. **93:** 231-243.

Gupta, A.K, Chaturvedi, S. and Sharma, A.K., 2009. Arbuscular mycorrhizal fungal diversity in some medicinal plants. *Mycorrhiza News*. **20**: 10-13.

Hemavani, C. and Thippeswamy, B., 2013. Arbuscular mycorrhizal fungi associated with some plants of asteraceae in Bhadra wildlife sanctuary. *International Journal of Plant, Animal and Environmental Sciences*. **3**: 106-110.

Jackson, M.L., 1978. *Soil Chemical Analysis.* Prentice Hall, New Delhi, India.

Jain, S.K., 1991. Dictionary of Indian Folk Medicine and Ethnobotany, Deep Publication, New Delhi.

Kshirsagar, R. and Upadhaya, S., 2009. Free radical scavenging activity screening of medicinal plants of Tripura, North East India. *Natural Products Radiance.* **8**: 117-122.

Majumdar, K. and Dutta, B.K., 2007. A study on ethnomedicinal usage of plants among the folklore herbalists and Tripuri medical practitioners: Part-II. *Nat. Prod. Rad.* **6:** 66-73.

Majumdar, K., Saha, R., Datta, B.K. and Bhakta, T., 2006. Medicinal plants prescribed by different tribal and non-tribal medicine men of Tripura state. *Indian Journal of Traditional Knowledge*. **5**: 559-562.

McGonigle, T.P., Miller, M.H., Evans, D.G., Fairchild G.L. and Swan, J.A., 1990. A new method which gives an objective measure of colonization of roots by vesicular–arbuscular mycorrhizal fungi. *New Phytologist.* **115**: 495–501.

Nair, S.K. and Girija, V.K., 1988. Incidence of vesicular arbuscular mycorrhiza in certain tree crops of Kerala. *Journal of Plantation Crops*. **16**: 67–68.

Panda, A. and Misra, M.K., 2011. Ethnomedicinal survey of some wetland plants of South Orissa and their conservation. *Indian Journal Of Traditional Knowledge.* **10:** 296-303.

Sarwade, P.P., Kanade, M.B., Ambuse, M.G. and Bhale, U.N., 2012. Association of arbuscular mycorrhizal fungi in some angiospermic plants of Maharashtra, India. *International Multidisciplinary Research Journal*. **2**: 18-19.

Sen, S., Chakraborty, R., De, B. and Devanna, N., 2011. An ethnobotanical survey of medicinal plants used by ethnic people in West and South district of Tripura, India. *Journal of Forestry Research*. **22:** 417-426.

Sharma, M., Sharma, C.L. and Deb Barma, J., 2013. ethnobotanical uses of some tree species in khowai district of Tripura, NE India. *Life Sciences Leaflets.* **4:** 60-80.

Sharma, M., Sharma, C.L. and Deb Barma, J., 2013. ethnobotanical uses of some tree species in khowai district of Tripura, NE India. *Life Sciences Leaflets.* **4:** 60-80.

Shil, S. and Dutta Choudhury, M., 2009. Ethnomedicinal importance of Pteridophytes used by Reang tribe of Tripura, North East India. *Ethnobotanical Leaflets.* **13:** 634"643.

Smith, S.E. and Read, D.J., 2008. Mycorrhizal Symbiosis, 3rd edn. Academic Press, San Diego. 787.

Turjaman, M., Santoso, E. and Sumarna, Y., 2006. Arbuscural mycorrhizal fungi increased early growth of gaharu wood of *Aquilaria malaccensis* and *A. crasna* under greenhouse conditions. *Journal of Forestry Research*. **3**: 139–148.

Uma, E., Sathiyadash, K., Loganathan, J. and Muthukumar, T., 2012. Tree species as hosts for arbuscular mycorrhizal and dark septate endophyte fungi. *Journal of Forestry Research*. **23**: 641-649.

Walkley, A. and Black, I.A., 1934. An examination of the Degtjareff method for determining organic carbon in soils: Effect of variations in digestion conditions and of inorganic soil constituents. *Soil Sci.* **63:** 251–263.