

Studies on Am Fungi Associated With *Andrographis Paniculata* (Burm.F.) Nees From Tapowan, Amravati.

Dongare S.B¹. and Maggirwar R.C^{2*}

PG, Department of Botany, Shri Shivaji Science College, Amravati-MS

Abstract

The current day emphasis is on sustainable agriculture, which uses less of chemical inputs like fertilizers and pesticides having adverse effect on soil health, fertility and environment. Thus, use of microbial inoculants plays an important role in sustainable agriculture. AM fungi are known to improve the nutritional status, growth and development of plants.

Asian countries are very rich in medicinal plant species and are the major exporters of these plants and their products. These medicinal plants can be popularized and used to improve the economy of low-income countries of Asia and create livelihoods for its people.

Kalmegh (*Andrographis paniculata* Nees.) is such a wonder plant which has numerous medicinal properties. Hence, the purpose of this research was to isolate and identify Arbuscular Mycorrhizal Fungi (AMF) associated with the rhizosphere soils of *Andrographis paniculata* and mass propagate the native dominant species for further studies. The soil samples were collected from Tapowan area of Amravati, Maharashtra. These samples from rhizosphere area of *Andrographis paniculata* were wet sieved, and AMF spores were isolated and identified morphotaxonomically. In all five species of *Glomus* were observed e.g., *Glomus leptotichum*, *Glomus aggregatum*, *Glomus fasciculatum*, *Glomus maculosum*. The most dominant were of *Glomus aggregatum*. This dominant species will be mass multiplied and applied in further studies to study growth parameters of Kalmegh.

The authors believe that, as this technology being simple can easily be adapted by farmers cultivating this medicinal crop.

Key Words: *Andrographis paniculate*, AMF, *Glomus*

Introduction

About 80–90% of higher plants on earth and in all of the planet's environments have roots that are in mutualistic symbiotic partnerships with arbuscular mycorrhizal fungus (van der Heijden et al., 2015). The plant gives the fungus products of photosynthesis, and the fungus gives the plant nutrients, particularly immobile nutrients like phosphorus (Cardoso et al., 2017; Ma et al., 2019).

In recent years, due to their enormous potential in both traditional and modern medicine, the cultivation of medicinal and herbal plants has taken on greater significance. In the pharmaceutical, cosmetic, and fragrance industries, they are also employed as raw ingredients. ISM (Indian System of Medicine) employs 25,000 species from more than 1000 genera. Industries employ about 25% of species (Jaleel et al 2009). Inoculating AM fungus at the beginning of the acclimatisation phase has evolved into a different tactic for better establishment by enhancing plant growth. Microorganisms, particularly AM fungus, have been found on therapeutic plants, as has been previously documented (Karthikeyan et al., 2008). Along with promoting the growth of medicinal plants, the AM fungal relationship also increased the yield of medicinal chemicals.

Traditional medicine can be accessed through age-old, all-natural health care methods like folk medicine, home herbal remedies, and the Baidhya, Ayurveda, and Amchi (traditional healing systems of Tibet and the mountain regions of Nepal) systems (Kunwar *et al.*, 2010).

According to estimates, over two thirds of the population in many poor nations primarily rely on traditional healers and medicinal herbs to meet their basic healthcare needs. Although there may be access to modern medicine in these nations, traditional herbal medicine is frequently employed for historical, cultural, and ecological reasons. In particular, this is owing to ongoing availability, superior compatibility, and high acceptance (Kunwar *et al.*, 2010).

One of the significant plants is *Andrographis paniculata*, which is used traditionally in Ayurvedic (Indian), Thai, and Chinese medicine due to its geographic spread. The plant's aerial parts contain andrographolide, an active ingredient with an extremely unpleasant taste. It has numerous pharmacological uses as a bicyclic diterpenoid lactone (Maiti *et al.*, 2006).

This plant can be found growing in a number of settings, including roadside vegetation, wetlands, seashores, mountain slopes, and rangelands (Rajpar *et al.*, 2011).

This plant's bitterness is associated to its many medicinal characteristics, including antibiotic, antiviral, antimicrobial, anti-inflammatory, antivenom, immunostimulatory, anti-cancer, anti-HIV, anti-allergic, and hypoglycemic activity (Jegathambigai *et al.*, 2010; Sivananthan and Elamaran, 2013). It has been also used to treat pneumonia, TB, pyelonephritis, tonsillitis, pharyngitis, laryngitis, upper respiratory tract infection, and hepatic impairment (Maiti *et al.*, 2006).

Also, can be used as antityphoid and antimalarial since the whole plant is having medicinal value especially leaves which are in fresh and dried condition which are extensively included in the Asian traditional medicine for treating illness (Rajpar *et al.*, 2011).

Currently, there is a need to standardise ayurvedic medications and plant-based ingredients. The physico-chemical parameter is the only one that is described in several pharmacopoeias that have monographs on plant material. Indian traditional medicine routinely treats hepatitis using *Andrographis paniculata* Nees (Acanthaceae), also known as kalmegh. The diterpenoid andrographolide and associated chemical are the primary reasons for using the medicine (Jadhao, 2010). The present study aims to explore AM Fungi associated with this plant from Tapovan, Amravati and mass multiplication of native dominant strains to study the inoculation effect.

MATERIALS AND METHODS

Selection of Study Area

Tapowan, Amravati is a well-known place situated in Amravati, Maharashtra. Amravati is the second largest city in the vidrabha region and ninth largest city in Maharashtra, India. Amravati is located at 20.93°N 77.75°E.

Collection of soil samples

The samples were collected from different sites of Tapowan area. The soil samples were shade dried and kept in labelled polyethylene bags. The rhizospheric soils of each sample was collected from the depth of 2 to 20 cm along with roots with the help of soil auger and other instruments.

Isolation of AMF spores from Rhizospheric Soil

AMF spores were isolated from the rhizospheric soil samples by the method given by Gredemann and Nicolson (1963), with some modification.

Estimation of AMF Spore Count

AMF spore number was estimated by the method given by Gaur and Adholeya (1994), with some modification in this method. The sievates of rhizospheric soil (which is described in the method of Gerdemann and Nicolson, 1963) were filtered from filter paper having rectangles formed by vertical and horizontal lines. The filter paper with spore and other debris was spread on a large petri-plate and observed under Carl Zeiss Stereozoom microscope. The spore number was then counted from each rectangle of columns in upward direction by moving the petriplate. Intact spores were picked up using a wet needle and mounted in polyvinyl alcohol lactoglycerol (PVLG) on a glass slide for identification and photography.

Identification of AMF Spores: -

Identification of AMF spores was done by using 'Manual for the Identification of VA Mycorrhizal Fungi' – by N. C. Schenck and Yvonne Pérez (1990) and using the link in AM. www.edu/the-fungi/species-description.

Photography: -

Photography of AMF spores isolated from rhizospheric soil was done by using Carl Zeiss inverted compound microscope with Tucsen Camera (0.5 MP).

Results and Discussion: -

The present investigation shows the occurrence of AMF species associated with *Andrographis paniculata* Nees species from Tapowan of Amravati (MS). The AMF species along with their morphology and AMF characterization were noted. The number of spores were isolated from the collected soil samples out of which *Glomus* was most dominant. In all four AM fungal species were identified as *Glomus maculosum*, *Glomus aggregatum*, *Glomus leptotichum* and *Glomus fasciculatum*.

Glomus maculosum Miller and Walker

- Colour :- Pale straw colour
- Shape :- Globose to subglobose
- Size :- (95-)135-178(-220) x (95-)130 -187(-220) μm
- Composite spore wall :- 4.4- 15 μm
- The inner wall in many older spores bearing domed, scalloped in growths.

Glomus aggregatum Schenck and Smith emend. Koske

- Colour :- Pale yellow to yellow brown
- Shape :- Globose to sub- globose
- Size :- (20-) 40-85 (-120) μm
- Composite spore wall :- 2-6 (-10) μm
- Subtending hypha: - Straight, constricted, swollen or irregular and (6.4 -) 14.3(-21.6) μm wide at spore base.
- Spores produced in sporocarp

Glomus leptotichum Schenck and Smith

- Colour:- Light yellow
- Shape :- Globose
- Size :- (48-)175(-262) μ m
- Composite spore wall :- 1.5-7.4(-10.5) μ m
- Subtending hypha :- 9-27 μ m wide at the point of attachment
- Spore wall with adhering debris on the outer surface especially at the hyphal attachment

Glomus fasciculatum Walker and Koske

- Colour :- Pale yellow to pale yellow brown
- Shape :- Globose
- Size :- 75-149 μ m
- Composite spore wall :- (2.3)7-12(-16.1) μ m
- Subtending hypha straight

Photoplate I- Most dominant *Glomus* species



Glomus maculosum



Glomus leptotichum



Glomus leptotichum



Glomus fasciculatum



Glomus aggregatum



Glomus leptotichum



Glomus leptotichum



Glomus maculosum

Conclusion

- The present study revealed that the rhizosphere soil of *Andrographis paniculata* Nees shows dominance of associated *Glomus* species.
- In all four AM fungal species were most dominant *Glomus maculosum*, *Glomus aggregatum*, *Glomus leptotichum* and *Glomus fasciculatum*.
- This study has a future scope to explore the AM Fungi association with *Andrographis paniculata* Nees and study the effect of dominant species on growth parameters by raising the inoculum and applying nature's own tool as a biofertilizer.

Reference

1. B. Karthikeyan, C. A. Jaleel, G. M. Alagu Lakshmanan, and M. Deiveekasundaram, Colloids Surf. B: Biointerfaces 62, 143 (2008). doi:10.1016/j.colsurfb.2007.09.004
2. Cardoso E.J.B.N.; Nogueira M.A.; Zangaro W. 2017. Importance of Mycorrhizae in tropical soils. In: de Azevedo J.; Quecine M. (eds) Diversity and benefits of microorganisms from the Tropics. Springer, Cham.
3. C.A. Jaleel, R. Gopi, M. Gomathinayagam, and R. PanneerselAM, Process Biochemistry 44, 205 (2009). doi:10.1016/j.procbio.2008.10.012
4. Jadhao M. Estimation of andrographolide in herbal powder and polyherbal asava by HPTLC. International Journal of Pharma and Bio Sciences 1(4), 242- 245.
5. Jeffries, P., Gianinazzi, S., Perotto, S., Turnau, K., Barea, J. -M., 2003. The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. Biology and Fertility of Soils 37, 1–16
6. Jegathambigai R, Devaraj S, Kumar P, Sivaramakrishnan S. 2010. A Study on the hepatoprotective effect of *Andrographis paniculata* (Burm. F) nees on mice. Journal of Phytology 2(11), 25- 30.
7. Kunwar RM, Shrestha KP, Bussmann RW. 2010. Traditional herbal medicine in Far-west Nepal: A pharmacological appraisal. Journal of Ethnobiology and Ethnomedicine 6, 35, <http://dx.doi.org/10.1186/1746-4269-6-35>
8. Maiti K, Gantait A, Mukherjee K, Saha BP, Mukherjee PK. 2006. Therapeutic potentials of Andrographolide from *Andrographis paniculata* : A review. Journal of Natural Remedies 6(1), 1 – 13.
9. McGonigle TP, Miller MH, Evans DG, Fairchild GL, Swan JA (1990) A new method which gives an objective measure of colonization of roots by vesicular arbuscular mycorrhizal fungi. New Phytol 115: 495-501
10. Miller RM (1987) The ecology of vesicular-arbuscular mycorrhizae in grass and shrublands. In: The Ecophysiology of VA mycorrhizal Plants. Ed GR safir. CRC press, Boca Raton, FL, pp 136-163
11. Rajpar I, Khanif YM, Hassan ZU, Shah AN, Arshad M, Galani S. 2011. Growth, herb yield and phytochemical contents in a medicinal herb *Andrographis paniculata* under saline Irrigation. Journal of Medicinal Plants Research 5(23), 5528- 5533.
12. Sivananthan M, Elamaran M. 2013. In vitro evaluation of antibacterial activity of chloroform extract *Andrographis paniculata* leaves and roots, *Durio zibethinus* wood bark and *Psidium guajava* leaves against selected bacterial strains. International Journal of Biomolecule and Biomedicine 3(1), 12- 19.
13. van der Heijden, M.G.A.; Martin, F.M.; Selosse, M.-A.; Sanders, I.R. 2015. Mycorrhizal ecology and evolution: the past, the present and the future. New Phytologist 205: 1406–1423.