Isolation and Identification of Soil Fungi from Different Localities of Kolhapur District of Maharashtra, India.

***J. M. Gorule[1], S. A. Bhoi[2], C. M. Dalavi[3], A. R. Kasarkar[4], S. P. Salokhe[5], S. B. Kubal[6], S. A. Patil[7] and S. S. Kamble[8].**

[1,2,3] Research student of Shivaji University Kolhapur.

[4,5,6] Professor in the Department of Botany, Vivekanand College, Kolhapur.

[8] Senior Professor, Mycology and Plant Pathology Laboratory, Department of Botany, Shivaji University, Kolhapur. 416004.

E. Mail: jyotigorule5@gmail.com

Abstract: The present study was conducted to find out the fungal diversity in soil sample collected from District Kolhapur. During investigation 12 samples were studied. Among that 13 isolates of fungi obtained and 9 were identified with standard key and microbial expert.

Keywords: *Soil, fungi, Isolation, Identification.*

Introduction

Soil is a mixture of organic matter, minerals, gases, liquids, and organisms that together support life. Earth's body of soil is the pedosphere, which has four important functions: it is a medium for plant growth; it is a means of water storage, supply and purification; it is a modifier of Earth's atmosphere; it is a habitat for organisms; all of which, in turn, modify the soil.

The soil serves as a reservoir for many microbial communities of plants and herbs which can be producing, CO_2 and nitrogen cycle. The microorganisms plays major role in soil ecosystem. Microbial composition and functioning changes the soil quality through decomposition of organic matter, recycling of nutrients and biological control (Stefanis et al., 2013). For most of the time, fungi are either dormant, or they metabolize and grow very slowly utilizing a range of organic molecules. Fungi are not only beautiful but play a significant role in the daily life of human beings besides their utilization in industry, agriculture, medicine, food industry, textiles, bioremediation, natural cycling, as bio fertilizers and many other ways. Fungus benefits most plants by suppressing plant root diseases and fungi promote healthier plants buy attacking plants pathogens with fungal enzymes. Fungi also use antagonism to reduce competition by producing antibodies, which suppress other microorganisms from growing. They produce many vitamins which promote plant growth. Beneficial fungi also form protective webs and nets around roots and leaves to protect the host plants (Lowenfels and Lewis, 2006; Sylvia et al., 2005). Fungus also protects plants by supplying a protective health to supply both water and phosphorus to the plant roots during droughts (Magdoff and VanEs, 2009). The present study was done for identify fungal community from soil samples collected from different localities of Kolhapur district and it was identified with microbial expert.

Material and Methods

Soil samples: Soil samples (Black and Red) were collected near plant roots (Approximately 5g) with clean dry and sterile polythene bags along with sterile spatula. The collected samples were brought to the laboratory and preserved for further studies. The soil samples were collected in the month of March (2018) from different localities of Kolhapur district of Maharashtra (Chokak, Bawada, Kerli, Kagal, Mhalunge, Kapshi).

Isolation and characterization of fungi: Serial dilution agar plating (Apinis, 1963) and Warcup's soil plate method (Warcup, 1950) were employed for the isolation of soil microbes.

Fungal medium: The aliquots were cultured for fungus on Czapek Dox Agar (NaNO3 2.0 g, KCl 0.5 g, K2HPO4 1.0 g, MgSO4.7H2O 0.5 g, FeSO4.7H2O 0.01 g,); and Potato Dextrose Agar (Peeled potato200.0 g, Dextrose 20.0g) amended with streptomycin sulphate (Patil et al., 2012; Mali et al., 2015). Three plates from each soil samples were incubated for 24-96 h at 28+2^{0}C, and each morphologically unique fungal colony was sub-cultured and purified using standard techniques. On the basis of morphological, microscopic characters and following relevant mycological literature the fungal isolate was identified.

Results and discussion

The present study was conducted in month of March (2018) to find out the fungal diversity in soil sample collected from District Kolhapur. During investigation 12 samples were studied. Among that 13 isolates of fungi obtained and 9 were identified with standard key and microbial expert.

Soil is a multi-layered surface complex of mineral and organic constituents present in solid liquid and gaseous states. Broad soil type and, slit or clay is defined as largest to smallest of particle size. These particles pack loosely, and pour spaces, and plant roots are particular habitats for microorganisms, often in bio films (Jadhav and Shinde, 2017). A gram of garden soil can contain around 1 million fungi such as yeasts, and moulds fungi have no chlorophyll and are not able to photosynthesis. They require a chemical source of energy rather than being able to use light as an energy source as well as organic substrates to gate carbon for growth and development. Some fungi are parasitic, causing disease to their living host plant although some have beneficial with plants. Where ever adequate moisture, temperature and organic substrates are available fungi are present. The environmental, moisture, organic carbon an nitrogen play an important role in distribution of mycoflora. (Adams et al, 1999)

Present study was carried out for to understand the soil fungal diversity in Kolhapur district. Some fungi like *Mucor, Alternaria, Aspergillus flavus, Penicillum, Aspergillus niger, Nigrospora, Alternaria, Trichoderma, Penicillium* were isolated and identified. Among that *Nigrospora* was found only in red soil of Chokak. Black and red soil contains near about same fugal diversity.

***Table* 1: The fungi isolated from different localities of Kolhapur district of Maharashtra**

Sr. No.	Place of collected samples	Type of soil	Isolation method	Media	Name of Isolated fungi
1	Chokak	Black	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose Agar	*Alternaria, Fusarium*
			Warcup's soil plate method	Czapek Dox Agar	*Penicillum, Aspergillus niger*
				Potato Dextrose Agar	*Alternaria, Mucor*
		Red	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose Agar	*Nigrospora, Alternaria*
			Warcup's soil plate method	Czapek Dox Agar	*Nigrospora*
				Potato Dextrose Agar	*Nigrospora, Mucor*
2	Bawada	Black	Serial dilution	Czapek Dox Agar	*Mucor, Aspergillus flavus, Aspergillus niger*
				Potato Dextrose Agar	*Trichoderma Alternaria, Aspergillus flavus*
			Warcup's soil plate method	Czapek Dox Agar	*Alternaria, Rhizopus*
				Potato Dextrose Agar	*Trichoderma, Mucor*
		Red	Serial dilution	Czapek Dox Agar	*Trichoderma, Alternaria, Penicillium*
				Potato Dextrose Agar	*Alternaria. Mucor*
			Warcup's soil plate method	Czapek Dox Agar	*Trichoderma*
				Potato Dextrose Agar	*Alternaria, Mucor, Fusarium*
3	Kerli	Black	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose Agar	*Alternaria, Mucor*
			Warcup's soil plate method	Czapek Dox Agar	*Aspergillus niger*

				Potato Dextrose Agar	*Alternaria,Rhizopus Mucor*
		Red	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose Agar	*Alternaria*
			Warcup's soil plate method	Czapek Dox Agar	*Aspergillus flavus*
				Potato Dextrose Agar	*Alternaria, Mucor*
4	Mhalunge	Black	Serial dilution	Czapek Dox Agar	*Aspergillus flavus, Aspergillus niger*
				Potato Dextrose Agar	*Mucor, Aspergillus flavus*
			Warcup's soil plate method	Czapek Dox Agar	*Alternaria*
				Potato Dextrose Agar	*Aspergillus flavus*
		Red	Serial dilution	Czapek Dox Agar	*Alternaria, Mucor*
				Potato Dextrose Agar	*Alternaria, Aspergillus flavus*
			Warcup's soil plate method	Czapek Dox Agar	*Penicillium*
				Potato Dextrose Agar	*Alternaria, Rhizopus*
5	Kagal	Black	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose Agar	*Alternaria, Mucor,*
			Warcup's soil plate method	Czapek Dox Agar	*Mucor*
				Potato Dextrose Agar	*Alternaria, Mucor*
		Red	Serial dilution	Czapek Dox Agar	*Mucor, Aspergillus flavus*
				Potato Dextrose Agar	*Mucor, Rhizopus*
			Warcup's soil plate method	Czapek Dox Agar	*Aspergillus niger*
				Potato Dextrose Agar	*Alternaria, Mucor*
6	Kapshi	Black	Serial dilution	Czapek Dox Agar	*Mucor, Alternaria, Aspergillus flavus*
				Potato Dextrose	*Mucor, Trichoderma*

				Agar	
			Warcup's soil plate method	Czapek Dox Agar	*Penicillium*
				Potato Dextrose Agar	*Alternaria,*
		Red	Serial dilution	Czapek Dox Agar	*Mucor, Aspergillus flavus*
				Potato Dextrose Agar	*Aspergillus flavus*
			Warcup's soil plate method	Czapek Dox Agar	*Aspergillus flavus*
				Potato Dextrose Agar	*Mucor, Aspergillus flavus*

References:-

1. *Apinis A. E. "Occurrence of thermophilous micro-fungi in certain alluvial soils near Nottingham". Nova Hedwigia 5:57-58. 1963.*
2. *C. Stefanis, A. Alexopoulos, C. Voidarou, S. Vavias, E. Bezirtzoglou. "Principal methods for isolation and identification of soil microbial communities". Folia Microbiol. (Praha), 58 (1) pp. 61-68, 10.1007/s12223-012-0179-5. 2013.*
3. *Jadhav S. Y. and Shinde P. P. "Isolation and Identification of Soil Fungi from Kadegaon Tehsil, Sangli District, Maharashtra, India". International Journal of Scientific and Research Publications, Volume 7, Issue 12, December 2017 616 ISSN 2250-3153.*
4. *Lowenfels, J. and Lewis, W. "Teaming with Microbes: A Gardener's Guide to the Soil Food Web", Chapter 3: Bacteria, Timber Press, Portland, Oregon. 2006.*
5. *Magdoff, F and Van Es, H. Management, "chapted; the Living soil" (3rd ed). sustainable Agricultural network, Handbook series book 10. SARE sustainable Agricuture Research &Education: Beltsville, Maryland.*
6. *Mali, A. M., Patil, V. B., Ade, A. B., Chavan, N. S. And Kamble, S. S. "First Report Of Fusarium Sp. FIESC_17 On Cucumis trigonus In India". Plant Disease. 10.1094/PDIS -09 -14 -0881 -PDN. http://dx.doi.org/10.1094/PDIS -09 -14 -0881 -PDN . 2015.*
7. *Mali, A. M., Patil, V. B., Pise, N. M. and Ade, A. B. "First Report of Leaf Spot Caused by Fusarium sp. NFCCI 2882 on Angiopteris evecta: A King Fern from Western Ghats, India". Plant Disease. 2016.*
8. *Patil, V. B., Mali, A. M., Mahamuni, R. J., Chavan, N. S. And Kamble, S. S. "First Report Of Leaf Spot Caused By Phoma Costarricensis on Delphinium Malabaricum In Western Ghats Of India". Plant Disease. 96:1074. 10.1094/PDIS -12 -11 -1012 -PDN. http://dx.doi.org/10.1094/PDIS -12 -11 -1012 -PDN.*
9. *Sylvia, D.M., Hartel, P.G. Fuhrmann, J.J. and Zuberer, D.A. "Principles and Applications of Soil Microbiology" (2nd ed.). Edited by David M. Sylva, Pearson Prentice Hall, Upper Saddle River, New Jersey. 2005.*
10. *Warcup, J. H. "The Soil-Plate Method for Isolation of Fungi from Soil", Nature volume166, pages117–118 (15 July 1950).*