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International Journal of Current Research in Life Sciences
Vol. 07, No. 11, pp.2822-2826, November, 2018

ISSN: 2319-9490

RESEARCH ARTICLE

STUDY ON FUNGAL POPULATION IN THE SOIL SAMPLES OF PULSES FIELD IN JHANSI DISTRICT

***[1]Priyanka Sinha, [2]Gazala Rizvi and [1]Roopam Parashar**

[1]Research Scholar, Department of Botany, Bundelkhand University Jhansi, U.P. India
[2]Associate Professor, Department of Botany, Bundelkhand University Jhansi, U.P. India

Received 14th September, 2018; Accepted 17th October, 2018; Published 30th November, 2018

ABSTRACT

Soil microorganisms play an important role in soil fertility and promoting plant health. Their populations play an important role in plant growth and regulations. Soil harbors maximum biodiversity. This study was undertaken to investigate the diversity among fungal flora in the soil samples from villages in and around Jhansi. Soil samples were collected from10 different agriculture fields of pulses. A total of 14 fungal species belonging to 6 genera, were isolated on potato dextrose agar media (PDA), by using soil dilution technique. Identification and characterization of the mycoflora was done with the help of Introductory Mycology by Alexopolus *et al.* (2017) and was confirmed from NCFT, New Delhi. The overall dominant genera were Trichoderma, Aspergillus, Fusarium and Penicillium.

Key words: Soil mycoflora, Trichoderma, Aspergillus, Fusarium and Penicillium.

Citation: Priyanka Sinha, Gazala Rizvi and Roopam Parashar, 2018. "Study on fungal population in the soil samples of pulses field in Jhansi District" *International Journal of Current Research in Life Sciences*, 7, (11), 2822-2826.

INTRODUCTION

Soil is the major component of earth's ecosystem and regulates global biogeochemical cycles (Kennedy and Smith, 1995; Richards, 1987, Rakesh Sharma and Raju, 2013). Organic matters, minerals, gases and large numbers of macro and microorganisms are the major constituent of soil (Buscot, 2005). Microorganism in the soil and rhizosphere is beneficial in increasing soil fertility and plant growth and play a key role in many essential processes such as organic matter decomposition, elemental release by mineralization, turnover of organic matter, symbiotic and non-symbiotic atmospheric nitrogen fixation, denitrification, aggregation (Chenu and Stotzky, 2002; Chandrashekar *et al*., 2014; Christensen, 1989; Gaddeyya *et al.,* 2012). Among these microorganism's fungi are an important component of soil microbiota depending on soil depth and nutrient conditions. Different soils have specific fungal flora, but the majority of species found in them are cosmopolitan (Ainsworth and Sussman, 1968). Type of cultivation and crop management practices have greater influence on the activity of soil microflora (Mc Gill et. al., 1980). Soil matrix of microbial activity hot spots are important and have drawn the attention of today's researchers (Gaillard *et al.,* 2003, Rakesh Sharma and Raju, 2013). According to Chandrashekar *et al*. (2014), almost 1.5million fungal species are present in natural ecosystems, but only 5-10% has been described formally. Continuous use of chemical fertilizers over a long period may cause imbalance in soil mycoflora and thereby indirectly affect biological properties of soil leading to soil degradation (Manickam *et al.,* 1972; Rakesh Sharma and Raju, 2013). The main focus of the study is to isolate mycoflora from different pulse fields and to estimate the percentage contribution of different fungal species. The study involves isolation, identification and enumeration of fungal species from different pulse growing fields in Jhansi district.

***Corresponding author: Priyanka Sinha**
Research Scholar, Department of Botany, Bundelkhand University Jhansi, U.P. India

MATERIAL AND MATHODS

Study Area and Location

Jhansi is a historic city of Uttar Pradesh and also called gateway of Bundelkhand. It is located between Betwa andPahuj River at 25.4333 N 78.5833 E on the plateau of central India. Being on a rocky plateau, Jhansi experiences extreme temperatures. The temperature varies between 4°Cto 47°C. Average annual rainfall is 900 mm. The nature of the soil is red or black with different soil texture like rocky, gravely, sandy loam to clay loam. The land is suitable for species of citrus fruits, cereals, pulses and oilseeds.

Method for Collection of Soil Samples

The soil samples were collected from 10 different villages of Jhansi district. Five samples were collected from different fields of each village.overall 50 samples were taken. Sampling was done thrice during November 2017 to January 2018 at regular intervals. The sample collection was done with the help of sterilized spatula, by removing the top soil and digging the soil up to 15cm depth. Soil was keptin a small sterilized polythene bags andbrought to laboratory for further studies (Table1).

Isolation of Fungi from the Soil Samples: The soil mycoflora were enumerated by Soil dilution method. According to Johnson, (1995), dilution of 10^{-5} was used to isolate fungi on Potato Dextrose Agar. One percent streptomycin solution was added to the medium before pouring onto Petri plates for preventing bacterial growth.Under aseptic condition an aliquot of diluted sample was poured on to agar surface and was uniformly spread with a sterile, bent glass rod. The petriplates were kept for incubation at28 ± 1^{o}C for 7 days.

Identification of the Soil Fungi: The fungal flora was analyzed from different samples of selected sites. The morphology of isolated fungi was studied macroscopically colony features like colourand texture were recorded. The observations revealed the presence of 14 species belonging to 06 genera. The identification and classification of fungi was done by the "Introductory Mycology" by Alexopolus et.al., (2017). It is simple and widely accepted.

Statistical Analysis

The number of colonies per plate in 1g of soil was calculated. The percent contribution of each isolate was calculated by using the following formula:

$$\% \text{ contribution} = \frac{\text{Total no.of CFU of an individual specie}}{\text{Total no.of CFU of all species}} \times 100$$

$$\text{Frequency} = \frac{\text{Total no.of CFU of an individual field}}{\text{Total no.of CFU of all field}} \times 100$$

**CFU= colony forming unit*

Soil is the habitat of a complex microbial community and is the main source of microorganisms. Among these microorganism, fungi are one of the dominant groups present in soil. Different properties of soil play a pivotal role in fungal population. Soil samples collected from pulse fields harbor a large number of fungi. During our investigation of 50 soil samples collected from pulse fields, 168 colony forming units (CFU) were obtained. We have obtained total 14 fungal species belonging to 8 genera (Table2). The maximum fungal species were from Ascomycotina (117 colonies) and Zygomycotina (13 colonies). Results revealed that most dominant genera were of *Trichoderma* and *Aspergillus*, whereas *Rhizopus* sp. And *Curvularia* sp. were least present, 09 unidentified species were also recorded. Jalander and Mamatha (2015) reported species belonging to genera viz., *Aspergillus, Penicillium, Fusarium, Trichoderma, Rhizpous* etc., from different pulse field of leguminous plants. There was no uniform trend visualized from the soil samples of these villages. The most dominant genera were of *Trichoderma viride*(12.5%), *Trichodermaharzianum* (10.71%), *Fusarium oxysporum* (10.71%), *Aspergillus niger* (10.11%), and *Penicillium chrysogenum* (7.73%). *Fusarium* sp. (6.5%) *Aspergillus flavus* (6.5%) *Trichoderma virens* (5.35%), *Curvularia lunata* (5.35%), *Rhizopus stolanifer* (5.35%), *Aspergillusfumigatus* (4.16%) followed by, *Penicillium sp.* (3.57%), *Curvulariasp.* (3.57%), *Rhizopus* sp. (2.38%) and unidentified species (5.35%). Rakesh Sharma and Raju (2013) recorded *Aspergillus* was most dominant in their study with maximum percent contribution followed by *Penecillium.* Same species were dominant in the soil samples of red gram Gaddeyya *et al*, (2012). While Niharika, P.S., (2013), reported the dominance of *Aspergillus* in the soil of Black gram or green gram.

Table 1. Soil samples collected from different pulse fields

S. No.	Sites	Sample Location
1	F1	Bangra
2	F2	Babina
3	F3	Baruasagar
4	F4	Bhojla
5	F5	Chirgaon
6	F6	Digara
7	F7	Karguwan
8	F8	Moth
9	F9	Parichha
10	F10	Simrawari

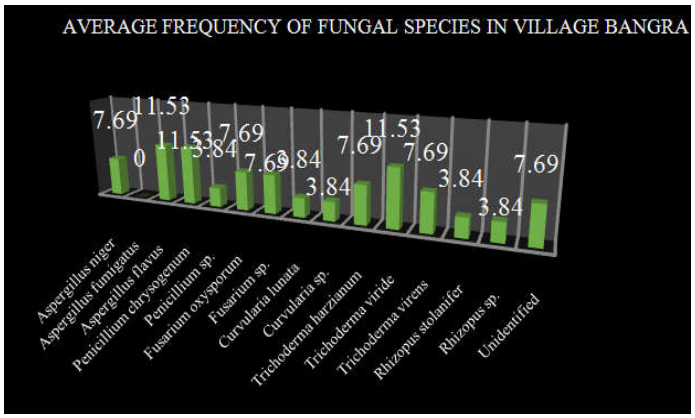
Table 2. Occurrence of fungal species in different pulse fields

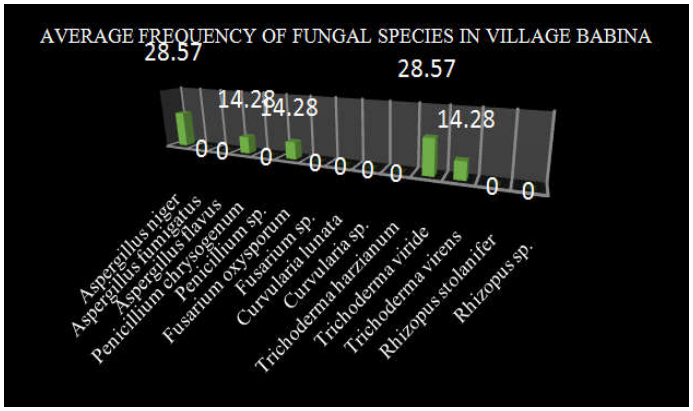
S. No	Sites	Average no of total colonies	Averageno of individual colonies														
			Aspergillus			Penicillium		Fusarium		Curvularia		Trichoderma			Rhizopus		Unidentified
			A	B	C	D	E	F	G	H	I	J	K	L	M	N	
1	F1	26	2	-	3	3	1	2	2	1	1	2	3	2	1	1	2
2	F2	7	2	-	-	1	-	1	-	-	-	-	2	1	-	-	-
3	F3	9	-	1	1	2	1	-	1	-	1	1	-	-	1	-	-
4	F4	15	2	1	1	1	-	3	-	1	-	2	2	-	1	-	1
5	F5	25	3	2	2	1	1	2	3	1	2	3	4	-	-	1	-
6	F6	21	2	-	-	2	-	3	4	2	1	1	3	1	-	-	2
7	F7	2	-	-	-	-	-	-	-	-	-	1	-	-	1	-	-
8	F8	18	2	1	3	-	1	1	-	1	-	3	2	-	2	1	1
9	F9	21	1	1	1	2	-	3	1	2	-	2	4	3	-	-	1
10	F10	24	3	1	-	1	2	3	-	1	1	3	1	2	3	1	2
Total		168	17	7	11	13	6	18	11	9	6	18	21	9	9	4	9
% Conrtibution			10.11	4.16	6.5	7.73	3.57	10.71	6.5	5.35	3.57	10.71	12.5	5.35	5.35	2.38	5.35

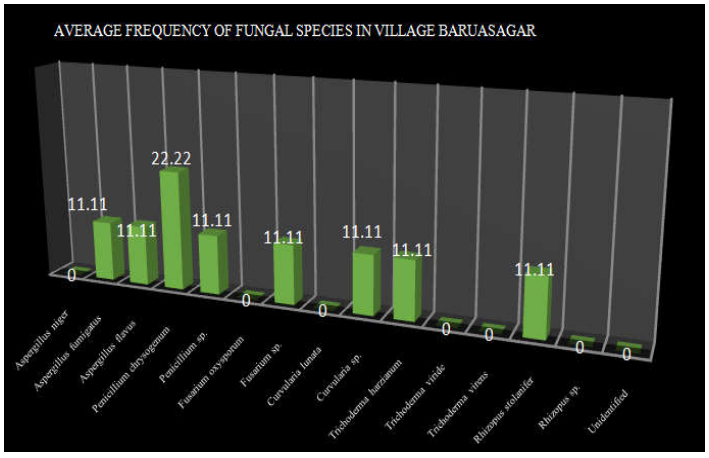
A- *Aspergillus niger* E- *Penicillium* sp. I- *Curvularia* sp.
B- *Aspergillus fumigatus* F- *Fusarium oxysporum* J-*Trichoderma harzianum*
C- *Aspergillus flavus* G- *Fusarium* sp. K- *Trichoderma viride*
D- *Penicillium crysogenum* H- *Curvularia lunata* L- *Trichoderma virens*
M- *Rhizopus stolanifer* N- *Rhizopus* sp.

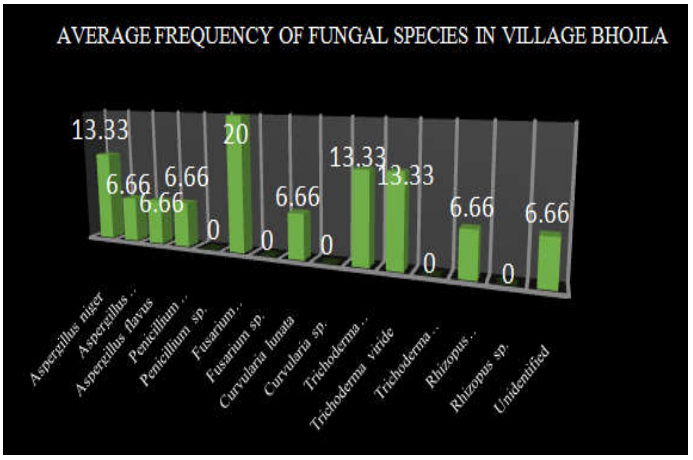
Table 3. Frequency of mycoflora in different sites of Jhansi district

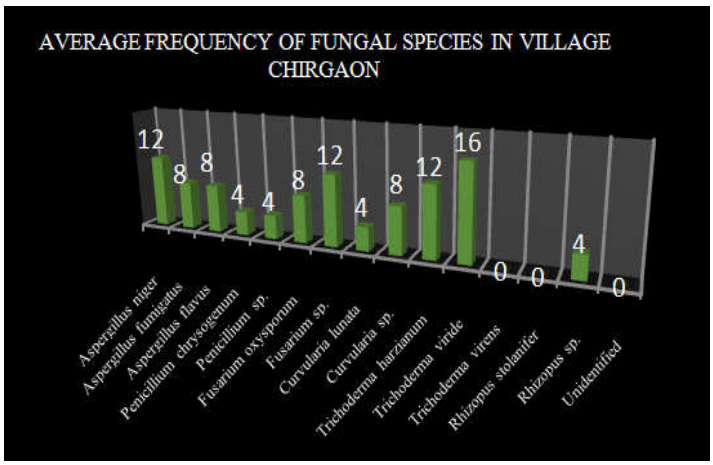
S. No	Fungal species	Frequency									
		F1	F2	F3	F4	F5	F6	F7	F8	F9	F10
1	*Aspergillus niger*	7.69	28.57	-	13.33	12	9.52	-	11.11	4.76	12.5
2	*Aspergillus fumigates*	-	-	11.11	6.66	8	-	-	5.55	4.76	4.16
3	*Aspergillus flavus*	11.53	-	11.11	6.66	8	-	-	16.66	4.76	-
4	*Penicillium chrysogenum*	11.53	14.28	22.22	6.66	4	9.52	-	-	9.52	4.16
5	*Penicillium sp.*	3.84	-	11.11	-	4	-	-	5.55	-	8.33
6	*Fusarium oxysporum*	7.69	14.28	-	20	8	14.28	-	5.55	14.28	12.5
7	*Fusarium sp.*	7.69	-	11.11	-	12	19.04	-	-	4.76	-
8	*Curvularia lunata*	3.84	-	-	6.66	4	9.52	-	5.55	9.52	4.16
9	*Curvularia sp.*	3.84	-	11.11	-	8	4.76	-	-	-	4.16
10	*Trichoderma harzianum*	7.69	-	11.11	13.33	12	4.76	50	16.66	9.52	12.5
11	*Trichoderma viride*	11.53	28.57	-	13.33	16	14.28	-	11.11	19.04	4.16
12	*Trichoderma virens*	7.69	14.28	-	-	-	4.76	-	-	14.28	8.33
13	*Rhizopus stolanifer*	3.84	-	11.11	6.66	-	-	50	11.11	-	12.5
14	*Rhizopus sp.*	3.84	-	-	-	4	-	-	5.55	-	4.16
15	Unidentified	7.69	-	-	6.66	-	9.52	-	5.55	4.76	8.33

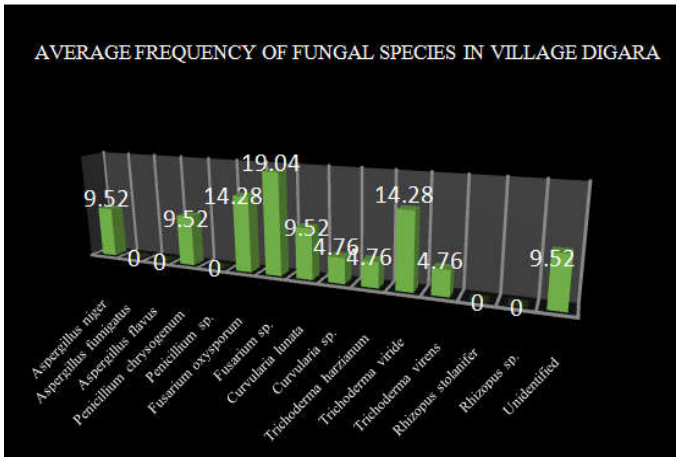


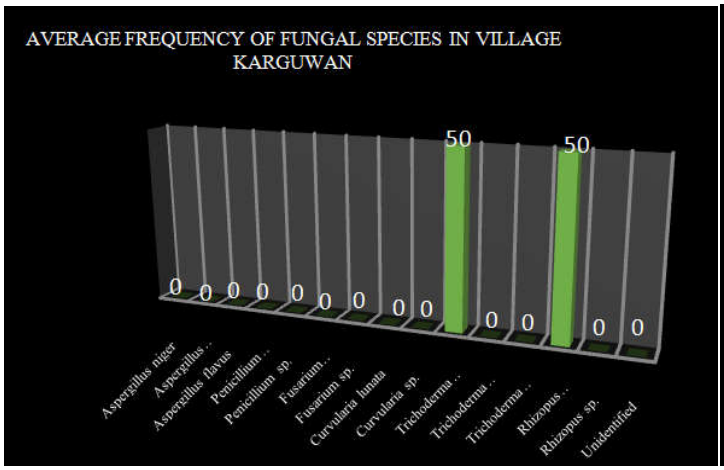


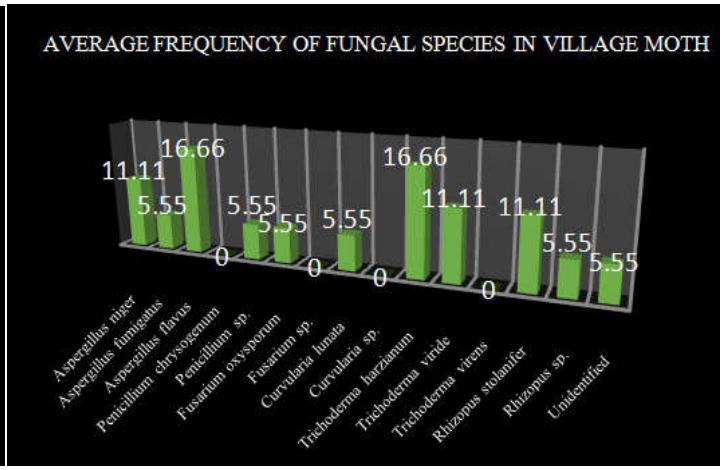


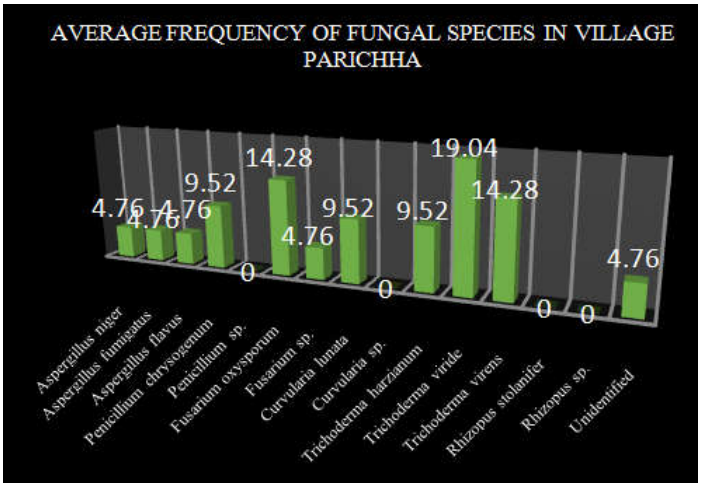


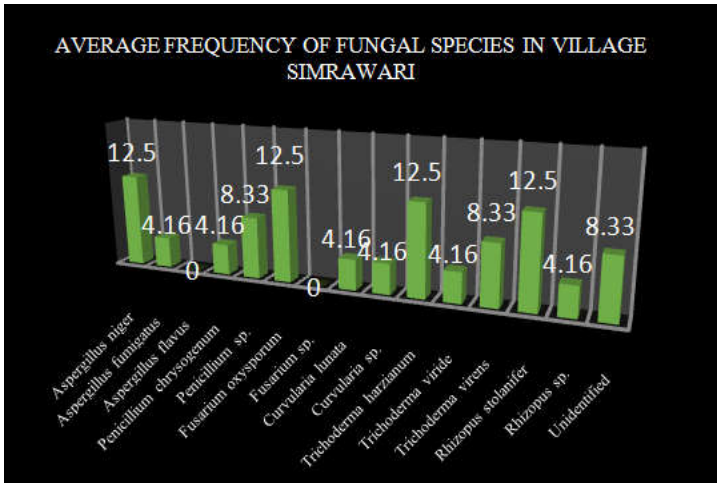












The frequency of isolated fungal flora was estimated and statistically analyzed the recorded results are given in (Table 3). Among the isolated species of fungi *Aspergillus niger, Penicillium crysogenum, Fusarium oxysporum* and *Trichoderma virens* showed significant presence i.e. above 80% present among the tested soil samples followed by *Curvularia lunata*. Another species of *Aspergillus* is approximately 60%. The rest were averagely reported. The maximum fungal flora was recorded from the soil sample of Bangra. *Aspergillus niger* and *Trichoderma viride* showed maximum frequency in the soil of village Babina. In village Bangra the genera penicillium, curvularia and rhizopus were least recorded. Same fungal genera i.e., *Alternaria, Penicillium, Fusarium, Rhizopus, Mucor* and *Aspergillus*, were reported from pulse field by Chandrashekar *et al*. (2014). Their study also revealed the dominance of *Aspergillus* and *Penicillium*, and that *Penicillium* sp. And *Rhizopus* sp. were least encountered.

Conclusion

In the present study screening of soil samples from 10 villages of Jhansi district was conducted to study the fungal diversity in pulse fields. The observations recorded revealed the presence of 168 CFU from 50 soil samples. The isolated mycoflora possessed 8genera and 14 species. Results clearly indicated that *Trichoderma, Aspergillus, Fusarium* and *Penicillium* were of high occurrence in all fields and some species of *Curvularia,* and *Rhizopus* were least encountered. Among the isolates *Trichoderma* and *Aspergillus* were dominant in all pulse fields.

Acknowledgement

The authors are thankful to the Department of Botany, Bundelkhand University for extending all due facilities and support during the work.

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