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Research Article

COMPARATIVE ANALYSIS OF PHYLLOSPHERE MYCOFLORA OF CONVENTIONAL AND ORGANIC DIFFERENT COTTON FIELD

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ABSTRACT

The percentage frequency and contribution of leaf surface mycoflora of conventional and organic cotton field was observed fortnightly with the help of gravity petriplate method. For mycofloral study 15common, dominant and pathogenic fungi were taken. The Fungi *viz. Alternaria alternate, Alternaria macrospora, Aspergillus flavus, Aspergillus niger, Cercospora gossypina, Colletotrichum gossypii, Curvularia lunata, Drechslera tetramera, Helminthosporium gossypii, Penicillium notatum, Penicillium chrysogenum, Rhizoctonia solani, Stemphylium solani, Rhizopus oryzae, Trichoderma harzianum, Trichoderma viride*were reported. The result shows that fungal occurrence is higher with the age of cotton plant than that conventional cotton field. If total fungal pathogen population higher in organic cotton with the age of cotton plant fungal pathogen also increase specially in conventional cotton field. It was reported that *Alternaria spp, Penicillium spp* and *Aspergillus* were the most common fungi in all the months on both the epidermises of all the plants.

INTRODUCTION

Cotton is major Fiber yielding crop with global importance, which is cultivated in tropical and sub tropical regions of more than 80 countries of the world. It belongs to genus Gossypium of family Malvaceae. Its cultivation is widely spread due to fibres are spun into yarns. Cotton's strength, absorbency, and capacity to be washed and dyed also make it adaptable to a considerable variety of textile products. India is leader in cotton cultivation and production in world. Since last one decade cotton has became an important cash crop of the region Maharashtra. The cultivation area under cotton has considerably increased in most of the parts of Maharashtra specially Vidarbha, Marathwada and Khandesh belts as well as few parts of western Maharashtra belt are now commonly known as supreme cotton belts due to large number of cotton growers achieving significant cotton yield. Environmental factors, several pests, fungi, bacteria, viruses and nematodes are known to reduce the production of crop yield for better production and productivity of crop researchers and farmers concentrating towards plant protection. The phyllosphere is an important habitat in which to study microbial ecology and affecting the health of plants. Because of the importance of many phyllosphere microbial inhabitants to plant health, there will likely be many practical applications that result from a better understanding of the interactions of microbes with plants and among themselves. Understanding of the ecology of pathogenic phyllospheric fungi provides new insights for the development of prevention or control strategies to manage pre-harvest contamination of crops with enteric pathogens. The presence of fungal species and consequently their concentration in the atmosphere are the result of the action of many biological and environmental factors. Last (1955) introduced term phyllosphere to denote the leaf surface of plants. The report of the intensive investigations on leaf surface mycoflora has been reported by Last and Deighton (1965). The aerial parts of living plants including leaves, stems, buds, flowers and fruits provide a habitat for microorganisms termed the phyllosphere. Bacteria are considered to be the dominant microbial inhabitants of the phyllosphere, although archaea, filamentous fungi and yeasts may also be important. These microbes can be found both as epiphytes on the plant surface and as endophytes within plant tissues (Arnold *et al*. 2000; Inacio *et al*. 2002; Lindow and Brandl 2003; Stapleton and

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Simmons 2006; Whipps 2008). The global surface area of the phyllosphere has been estimated to total over 4 $x10^8$ km^2, supporting bacterial populations in the region of 10^{26} cells (Morris and Kinkel 2002). The effect of OM amendments was found to be suppressive in 45% and non-significant in 35% of the cases. In 20% of the cases, a significant increase of disease incidence was observed by Bonanomi *et.al*. (2007). Considering all these aspect present study is carried out to check phyllospheric mycoflora conventional and organic cotton field.

MATERIAL AND METHODS

Sample collection

Cotton plant leaf sample were collected from the Kultabad location, Marathwada (MS), region in *kharif* (2015) season. Sample brought in the laboratory of plant pathology in Dr B.A M. University, Aurangabad. Samples collected randomly and used for the isolation.

Phyllosphere Mycoflora of Cotton

Study of Phyllosphere Mycoflora by Leaf Impression Method

This method was useful for superficial fungal spores. At the time of isolation fresh leaf were taken and pressed from its dorsal surface momentarily against the agar surface of petriplates at three places. Some leaf was placed from ventral surface against the agar surface in the same way like the first. Same procedure was repeated for other leaf samples. Inoculations of plates were carried out at 26 °C in an inverted position for 7 days.

Study of Phyllosphere Mycoflora by Serial Dilution Plate Method

This method was established by Dickinson (1945) and described as standard washing method for isolating microflora of leaf. Serial dilution plate method was one of the common methods adopted for study of phyllosphere microflora. Fresh, healthy leaves of all ages were collected from **Conventional field and Organic** cotton fields in fresh sterile polythene bags and brought into the laboratory. The 5 discs each of 6 mm diameter from every leaf were cut with the help of sterile cork borer. These discs were transferred in 100 ml water blank and stirred for near about 20 minutes using magnetic stirrer. After that 10 ml of the suspension was added to 90 ml sterile water blank (labeled 2) with the help of pipette diluting the original sample to 10 times. Contents were shaken for 2 to 3 minutes 10 ml suspension while in motion to another 90 ml sterile water blank (labeled 3) with the help of another fresh pipette. Contents of the test tube were shaken for the uniform distribution of spores / cells of aliquots from 10^{-1} and 10^{-2} dilutions were transferred to sterile petriplates (three replicates). Melted and cooled (45- 50°C) agar medium were poured in three plates. The plates were incubated at 26 °C in an inverted position for 7 days.

Table No.1 Percent frequency of fungal population in phyllosphere of conventional and organic cotton fields

Name of fungi	Khultabad Conventional cotton field	Khultabad Organic cotton field				
	30 Days	60 Days	90 Days	30 Days	60 Days	90 Days
Alternaria alternata	5.1	3.4	5.1	3.4	0.0	1.7
Alternaria macrospora	5.1	5.1	3.4	1.7	1.7	3.4
Aspergillus flavus	3.4	0.0	3.4	5.1	5.1	6.8
Aspergillus niger	0.0	1.7	1.7	5.1	8.5	5.1
Cercospora gossypina	0.0	1.5	3.8	0.0	0.0	1.7
Colletotrichum gossypii	5.1	5.1	6.8	0.0	1.7	0.0
Curvularia lunata	3.4	3.4	1.7	1.7	0.0	3.4
Drechslera tetramera	5.1	5.1	0.0	3.4	6.8	5.1
Helminthosporium gossypii	6.8	0.0	1.7	5.1	1.7	1.7
Penicillium notatum	3.4	3.4	0.0	1.7	5.1	3.4
Penicillium chrysogenum	0.0	1.7	0.0	0.0	3.4	6.8
Rhizoctonia solani.	3.4	1.7	3.4	1.7	0.0	0.0
Rhizopus oryzae	1.7	1.7	3.4	1.7	3.4	6.8
Trichoderma harzianum	1.7	0.0	1.7	8.5	8.5	8.5

Table No. 2 Percent Phyllospheric fungal population **per cm^2** of Conventional and organic cotton fields

Name of fungi	Khultabad Conventional cotton field	Khultabad Organic cotton field				
	30 Days	60 Days	90 Days	30 Days	60 Days	90 Days
Alternaria alternata	4.5	3.0	4.5	3.0	0.0	1.5
Alternaria macrospora	4.5	4.5	3.0	1.5	1.5	3.0
Aspergillus flavus	3.0	0.0	3.0	4.5	4.5	6.0
Aspergillus niger	0.0	1.5	1.5	4.5	7.5	4.5
Cercospora gossypina	0.0	0.0	3.0	0.0	0.0	1.5
Colletotrichum gossypii	4.5	4.5	6.0	0.0	1.5	0.0
Curvularia lunata	3.0	3.0	1.5	1.5	0.0	3.0
Drechslera tetramera	4.5	4.5	0.0	3.0	6.0	4.5
Helminthosporium gossypii	6.0	0.0	1.5	4.5	1.5	1.5
Penicillium notatum	3.0	3.0	0.0	1.5	4.5	3.0
Penicillium chrysogenum	0.0	1.5	0.0	0.0	3.0	6.0
Rhizoctonia solani.	3.0	1.5	3.0	1.5	0.0	0.0
Rhizopus oryzae	1.5	1.5	3.0	1.5	3.0	6.0
Trichoderma harzianum	1.5	0.0	1.5	7.5	7.5	7.5

RESULTS AND DISCUSSION

During the *kharip* season study was carried out. In present study 15pathogenic as well as saprophytic fungal species was observed commonly and dominates the phyllosphere of cotton. Diksha Khare and K. L. Tiwari (2014) *Barleria prionitis* 30 fungal species were observed in winter, 25 in rainy season and 20 in summer season. The percentage frequency of the leaf surface mycoflora was also observed. *Chaetomium aureum*, *Asp. niger, Asp.* flavus, *Penicillium rugulosum, Curvularia lunata, Curvularia clavata, Alternaria citri, Alternaria alternata, Cladosporium oxysporum, Cladosporium cladosporioides,* Mycelia sterile while were most frequent fungi. On the contrary moderate frequent fungi were *Choaenephora cucurbitarum*, *Mucor sp. Rhizopus rhizopodiformis, A.ustus, Asp. awamori, Asp.nidulans, Curvularia eragrostidis, Nigrospora sphaerica, Nigrospora oryzae, Fusarium oxyspourm, Colletotrichum dematium*. Mane and Chavan (2015) 16 dominant fungi was isolated in cotton field. In the present study comparative analysis of Phyllospheric mycoflora of conventional and organic cotton fields were shown from the (Table 1 and 2) fungal population is higher in organic cotton field than the conventional cotton fields; whereas the population of pathogenic fungi were higher in the conventional cotton fields as compare to organic cotton fields. (Zhang *et al*., 2010) shows the comparative analysis of phyllosphere microbial community between various vegetable crop plants. They show the rate of microbial community in spinach and rape is higher than the celery, broccoli and cauliflower. The fungi like *Colletotrichum gossypii* and *Helminthosporium gossypii* shows highestpercentage of population 6.8 % respectively in at per $cm^2$6.0 % of conventional cotton leaf sample. In the organic cotton leaf sample *Aspergillus niger* and *Trichoderma Harzianum* shows highest population at per cm^2area of leaf i.e. 7.5%; whereas percent fungal population 8.5 % *Alternaria alternata, Rhizoctonia solaniand Cercospora gossypina* lower percent fungal population. Percent frequency of occurrence of pathogenic fungi is higherin conventional cotton field as compare to organic cotton field. *Alternaria alternata, Alternaria macrospora, Cercospora gossypina, Colletotrichum gossypii, Curvularia lunata and Rhizoctonia solani s*hows dominates at all ages of conventional cotton field; whereas *Trichoderma harzianum, Penicillium notatum, Penicillium chrysogenum, Rhizopus oryzae, Aspergillus flavus and Aspergillus niger s*hows dominance in organic cotton fields. Organic cotton fields show the dominancy of saprophytic fungi as compare to pathogenic fungi.

CONCLUSION

During the present investigation pathogenic fungi like *Alternaria alternata, Alternaria macrospora, Cercospora gossypina, Colletotrichum gossypii, Curvularia lunata and Rhizoctonia solani s*hows highest percentage of occurrence in conventional cotton field. Where saprophytic fungi like *Trichoderma harzianum, Penicillium notatum, Penicillium chrysogenum, Rhizopus oryzae, Aspergillus flavus and Aspergillus niger* dominated. From the study and results it is concluded that the fungalpopulation were superior in the organic cotton fields as compareto conventional fields. The dominancy of saprophytic fungi inorganic cotton field was observed.

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