UNIVERSITY GRANTS COMMISSION

MAJOR RESEARCH PROJECT

BOTANY

EXECUTIVE SUMMARY OF THE WORK DONE ON THE PROJECT FILE NO.41-475/2012(SR)

'ATMOSPHERIC SURVEY OF FUNGAL SPORES AT INTRAMURAL AND EXTRAMURAL ENVIRONMENT OF KAMPTEE WITH RESPECT TO DIFFERENT HEIGHTS'

Submitted by

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Aerosols contain many substances of biological and non-biological origin which is variable according to region and seasons. Presence of different types of chemicals, particulate matter or biological material into the atmosphere that cause harm or discomfort to organism lead to air pollution. Therefore the field of aerobiology has its own importance.

Fungal spores contribute major fraction of airborne particles. Air is most vital component of environment without which nobody can survive. One can survive without food for weeks, for a few days without water, but hardly for few minutes without air. Today it is a well established fact that bacteria, viruses, fungi and pollen grains cause air pollution. These biological agents are called as Biopollutant and presence of biopollutant indicates the air quality. The plants, animals and human beings are affected by air-pollution including bio-pollution which is responsible for causing diseases besides being allergic to them. Among the Biopollutant, in India 90% of counts of air-borne biota belongs to fungi. The main aim and objective of this study were to daily monitoring of airborne fungal spores and make an approximation of airborne content.

Fungi are the main agent for deterioration, pathogenicity and allergy as well. Their diverse effects require more attention in the research field of mycology. Therefore with these objectives' an atmospheric survey of fungal spores in intramural and extramural environment of Kamptee was carried out.

According to geography of Kamptee, six Indoor and six Outdoor sites were selected for the study.

Quantitative and Qualitative sampling had done with the help of two samplers for consecutive two years at various intramural and extramural environment of Kamptee are as follow;

i) Selection of Sampling Sites

SITE I	:- Sub-district Government Hospital Wards(INDOOR 1)
SITE II	:- Sub-district Government Hospital premises(OUTDOOR 1)
SITE III	:- S.K.Porwal college library(INDOOR 2)
SITE IV	:- S. K. Porwal College outdoor premises.(OUTDOOR 2)
SITE V	:- Choudhary hospital wards (INDOOR 3)
SITE VI	:- Kendriya Vidyalaya library.(INDOOR 4)
SITE VII	:- Bhoyar College of Polytechnic library.(INDOOR 5)
SITE VIII	:- Anganwadi (Pre- primary school) (INDOOR 6)

SITE IX :- Market Area.(OUTDOOR 3)

SITE X :- Cantonment Area.(OUTDOOR 4)

SITE XI :- Railway station.(**OUTDOOR 5**)

SITE XII : - Ranala Road side.(**OUTDOOR 6**)

The Sites selected for different height was Indoor of Choudhary Hospital (Ground floor [5 ft] and First floor [15 ft]) and Outdoor of S.K.Porwal College (Ground floor [5ft] and Terries floor [35 ft]).

1) Qualitative Method

Air sampling was conducted using centrifugal impactor type air sampler (Himedia laboratories Ltd, India) by using Czapek's Dox Agar Strips. The sampler was kept at a height 5ft above ground and run for 4 minutes. Exposed strips are incubated for 3 – 4 days at 25-27 degree centigrade temperature. After incubation, the total colony forming unit per cubic meter per minute was counted by the formula as follows:



Sub cultures were maintained and fungal species were identified with the help of available standard literature (Gilman J.C. (1945), Barnett H.L. (1960), Nagmani et. al. (2006), Funder Sigurd (1953) & Tilak S.T. (1989) & (2009)). Some pure culture samples were also submitted to Agharkar Research Institute, Pune for the identification and authentication.

2) Quantitative Method

The 'Volumetric Tilak air sampler' was fixed at the roof of Seth Kesarimal Porwal College, Kamptee at the height of 15 feet from ground and runs continuously from May 2013 to April 2014 and at height of 50 feet for second year May 2014 to April 2015. The glycerin jelly mounted 16 slides were prepared from Vaseline coated cello tape, rotating drum of the sampler at the end of 8th day. The slides were scanned (Tilak, 1989) and fungal spores were observed, counted under Binocular microscope and identified by the standard literature. The Spores per cubic meter were calculated by the following formula;

Spores/ m^3 = No. of same type of spore X 14

(14 is the conversion factor for Tilak Air Sampler)

For Statistical analysis Origin Pro.9 version software is used, which is one of the ideal packages for statistical analysis. Statistical analysis of the data obtained was done by applying different appropriate methods like standard error of mean, ANOVA (analysis of variance 'F' test), student's 't' test and Pearson's correlation coefficient 'r' wherever necessary.

A great number of fungal spore diversity was seen in Kamptee environment, these may due to large number of area sampled. Indoor and outdoor comparison showed little variation although some sites shows greater variation. High count and diversity was seen in outdoors of Market area than indoor of Choudhary Hospital while indoor of Kendriya Vidyalaya shows high count than Outdoor of Cantonment area. The sites were dense vegetation present showed least diversity and count of aero fungal flora. This may due to a less soil/ dust was blown-up by air. Sites such as Colleges, Hospital and Market shows huge diversity and count this may be possible due to high number of visitors, less trees, improper ventilation and cleanliness. Although the presence of *Yeast* spores is unique feature to Kamptee environment.

A difference was seen according to seasons, weather conditions and according to sampling heights. A good diversity and count was seen in winter. Moderate rainfall blooms spores in air while heavy rainfall washout fungal spores. Temperature between 25 to 30°C and relative humidity between 50% – 80 % favored the release of spores in atmosphere. The most common species of *Aspergillus, Penicillium, Cladosporium* and *Rhizopus* were termed as non seasonal perhaps their presence throughout the year while species such as *Chaetomium, Geotrichum, Helminthsporium and Fusarium* are truly seasonal. The ground level had more concentration and diversity of fungal species than the first floor. Results of breathing level showed the dominance of allergenic, pathogenic and toxic fungal species such as *Aspergillus, Penicillium, Cladosporium, and Rhizopus* on first floor indicates that height is most important variable for variation in count.

Most of the fungal types isolated in this study were similar as studied by other workers (Bhattacharjee, 2010 and Hazarika, 2008). Perhaps *Candida, Saccharomyces, Stachybotrys, Syncephalastrum* was first time isolated in library environment, may be due to many small scale industries in Kamptee and surrounding areas. Whereas *Oospora* and *Phoma* was observed in outdoor environment. *Aspergillus, Cladosporium, Curvularia, Penicillium, Fusarium* were the most predominant fungi in all types of environment(Grinn-Gofroń et al., 2011 and Hasnain et al., 2012). Including all above fungal types *Candida* and *Yeast* species were dominant in all sampling environment. Intramural fungal diversity, variation and count was mostly depend on extramural environment (Chakraborty et al., 2000), population, number of visitors, proper ventilation, cleanliness and hygiene (Wei et al., 2015). The presence of all above fungal types was highly insignificant in health point of concern (Giri &Sawane, 2010). To avoid fungal related diseases frequent cleanliness and proper ventilation systems are mandatory.

The study showed that the indoor environments were more polluted than of outdoor sites. An outdoor site with high vegetation shows lower aeromycoflora biodiversity, which proves plants value for human life. The study gives essential information for diversity of Culturable atmospheric fungal spores, concentration of *Aspergillus, Penicillium*, *Cladosporium, Alternaria, Curvularia, Candida, Yeast* were showed the higher biopollutant in dwelling places of Kamptee(Thaware and Jawade,2013).

Higher numbers of mould fungi are associated with high shade and high levels of organic debris near the home and poor landscaping. The aeromycoflora of the three schools that were taken in to consideration for study had a distinct structural and location differences. In case of Seth Kesarimal Porwal college building completed its 50 years while Government and Choudhary Hospital completed more than four decades without proper ventilation and maintenance. Thus in the process library and both hospital intramural environment remain humid for several months in the year, representing typical sick building syndrome. Further it has a congested surroundings with few commercial markets, busy traffic flow and slum dwellers. This has probably made the indoor environment quite inhospitable for staff, students and patients.

A good number of forms produced abundant spores (*Aspergillus, Penicillium, Cladosporium, Geotrichum, Rhizopus, Candida, Yeast*, etc.) that easily become airborne and they frequency were well in excess (i.e. 10^8 m^{-3}) of the concentration needed for sensitization in allergic diseases (Lacey 1996). The indoor flora of Bhoyar College of Polytechnic Library and Kendriya Vidyalaya Classrooms was interestingly less than S.K.P. College, C. Hospital and Sub-district Government Hospital sites. This low concentration could definitely be attributed to its better and well maintained infrastructure, age of the building; moreover the surroundings are free from any commercial activities.

No environment is free from fungal spores, factors include the proximity to bioaerosol sources (soil and vegetation) at ground, aerodynamics characteristics, size and shape of sampled bio aerosol, the effect on meteorological conditions on release, dispersal and deposition of fungal spores at the same time the effect of vertical temperature gradient of the air were important for the variation, dominance and diversity. The numbers of people, hygiene, and number of visitors are really affected on fungal count. The results obtained from the study will be helpful to allergologist and clinicians for the treatment of allergic disorders.

CONTRIBUTION TO THE SOCIETY

The results obtained from the project work closely help the medical doctors, plant pathologists, mycologists and meteorologists. Aerobiological observations are helpful and may be used in many other disciplines: Palynology, ecology, botany, phenology, climatology, meteorology and forensics.

Many fungal spores carried in the air are plant pathogens. So farmers can minimize their pesticide use if they are able to track when particular pathogens will pass over their growing crops and when they won't.

Knowledge of the vertical profile of aerospora is important to both plant pathologists and allergologist to treat a vast society of Kamptee.

PUBLICATIONS

1) Thaware, J.S. and Jawade, S. 2013. Aerobiological diversity of fungal spores in Kamptee, Nagpur. Proceedings of National Seminar on Biodiversity Conservation-A Global need organized by J. M. Patel College, Bhandara.:114-122.

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6) Thaware, J., Seema Jawade and A.A. Saoji 2017. *Surveillance of fungal aeroallergens in the vegetable market environment*. The Botanique. Vol. 21(1): 63-70

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Light Microscopic Photographs of Fungal Spores Captured by Hi-Media Air sampler and Tilak Air Sampler during Study

1.	Alternaria alternata	Х	450	
2. 3.	Alternaria brassicicola	Х	450	
3.	Aspergillus niger	Х	450	
4.	Aspergillus fumigatus	Х	450	
5.	Aspergillus tamarii	Х	450	
6.	Allomyces sp.	Х	450	
7.	Botrytis sp.	Х	450	
8.	Candida sp.	Х	1000	
			Plata _ 2	

9.	Chaetomium sp.	Х	100
10.	Chaetomium sp.	Х	450
11.	Chaetomium sp.	Х	1000
12.	Cladosporium herbarum	Х	450
13.	Cladosporium cladosporioides	Х	450
14.	Curvularialunata X	450	
15.	Curvularia brachyspora	Х	450

16.	Fusariummonoliformis	Х	450
17.	Fusarium solani	X	450
18. 19.	Geotrichum sp.	X X	450 450
19. 20.	Helminthosporium sp. Humicola sp.	X	430 450
20. 21.	Eurotium amstelodami	X	100
22.	Eurotium amstelodami	Х	450

23.	Mucor sp.	Х	100
24.	Paecilomyces sp.	Х	450
25.	Penicillium citrinum	Х	450
26.	Nigrospora oryzae	Х	450

23.	Mucor sp.	Х	100
24.	Paecilomyces sp.	Х	450
25.	Penicillium citrinum	Х	450
26.	Nigrospora oryzae	Х	450
20.	Nigrospora oryzae	Λ	4

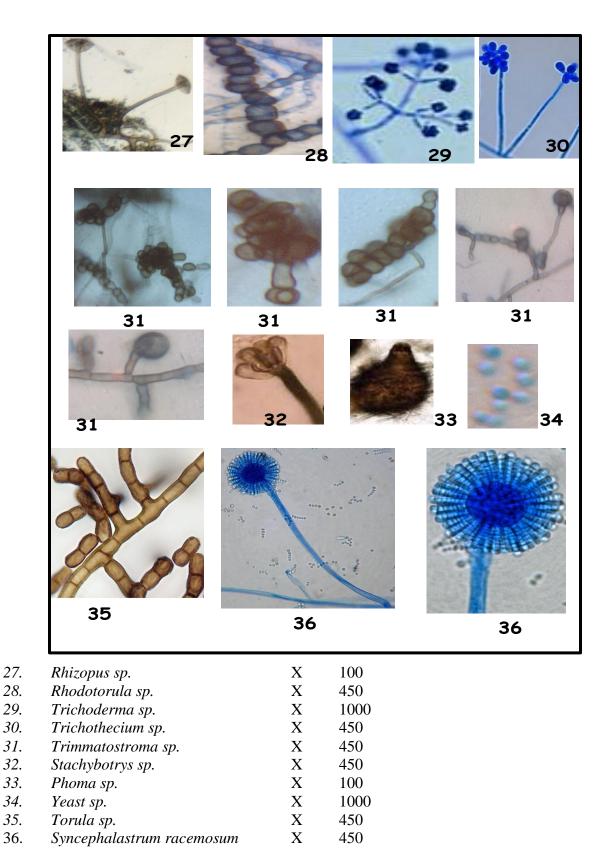
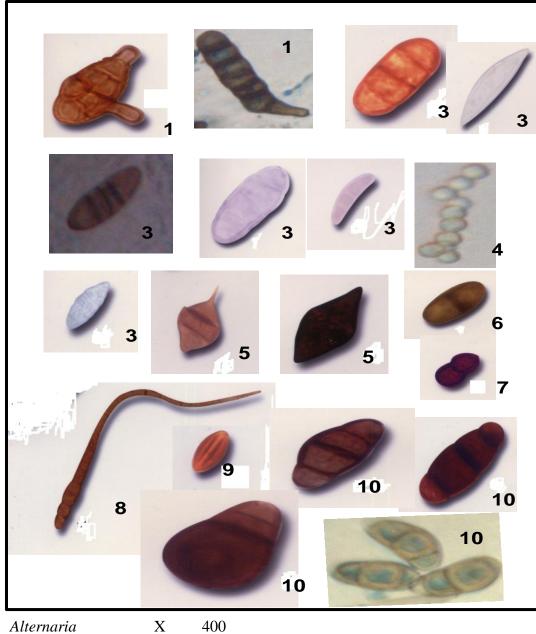
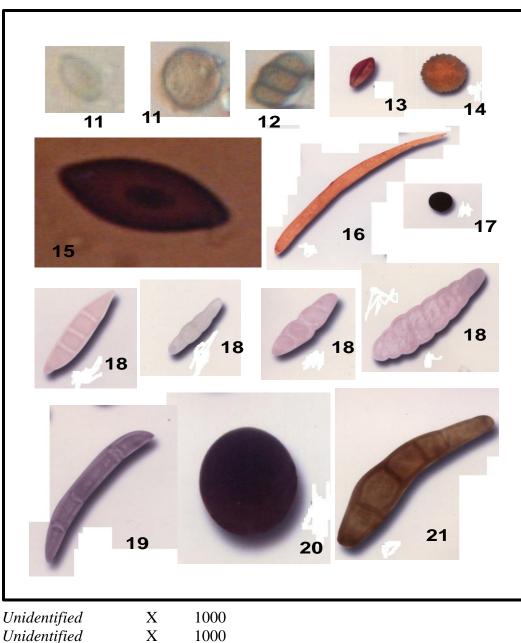


Plate – 1



1.	Alternaria	Х	400
2.	Alternaria	Х	400
3.	Ascospores X	1000	
4.	Mold Spores X	1000	
5.	Beltrania	Х	1000
6.	Bispora	Х	1000
7.	Didymosporium	Х	1000
8.	Cercospora X	400	
9.	Artrinium	Х	1000
10.	Curvularia	Х	1000





- 11.
- 12. Х Unidentified 13.
- Pithomyces Х 1000 Х 1000
- 14. Chaetomium
- 15. Х 1000 Mold Spore
- Cercospora Х 400 16. Х
- 17. Unidentified 1000
- 1000 18. Leptosphaeria Х 19. Fusariella Х 1000
- Х 20. Nigrospora 1000
- Unidentified Х 21. 400

				Plate 3
			22	23
	0		24	25
	8	26		27
			28	29 30
22. 23. 24. 25. 26. 27.	Unidentified Hirudinaria Didymosporium Helminthosporium Smut spores Unidentified	X X X X X X	1000 400 X 400 1000 1000	1000
27. 28. 29.	Unidentified Epicoccum Spegazzinia	X X	1000 X 1000	1000

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