www.sospublication.co.in Journal of Advanced Laboratory Research in Biology

We- together to save yourself society e-ISSN 0976-7614

Volume 3, Issue 2, April 2012 Research Article

Biodiversity and Concentration of Airborne Fungi of Suburban Weekly Market Associated Environment

Apurva K. Pathak*

Dept. of Microbiology, Modern Dental College and Research Centre, Gandhinagar, Indore-453112, M.P., India.

Abstract: It is supposed that the vegetable markets of tropical countries are the significant source of airborne fungal spores, which are generated during transportation, handling and putrefaction. The aim of this work was to monitor the prevalence of the airborne cultivable fungi in the air of weekly market associated environments to evaluate whether the vegetable trading zone of market is a source of airborne cultivable fungal spores of weekly market environment or not. Airborne cultivable fungal spore levels were monitored by using Andersen two-stage viable (microbial) particle sampler. The Spearman correlation coefficients and stepwise linear regression analysis test was used to analyze the influence of meteorological factors on spore concentration and paired Student's t-test was used to compare the bioload of total viable cultivable fungi of vegetable trading area and general item trading area of weekly market extramural environment, the percentage frequency and the percentage contribution of the individual genus was also reported. In both areas, *Aspergillus*, *Cladosporium*, *Alternaria*, and *Penicillium*, were the most abundant fungal types observed. The spp. of *Candida* was reported only at the vegetable trading area. The bioload of fungal spore presented maximum values during the Monsoon and lowest in the season of summer. There is no significant difference in quantity between the mean values of the bioload of total viable cultivable fungi of vegetable trading area and general item trading area of weekly market extramural environment were observed. For present environment, activities of animals and humans were supposed to be the key factor governing aerosolization of microorganism.

Keywords: Aero mycology, Weekly market, Vegetable, Environmental factors, Extramural, Regression model.

1. Introduction

The atmosphere contains only allochthonous microorganisms; Brock (1966) stated that it is unlikely that microorganism grows in the air itself, but he reported that, and some workers suggested the possibility of survival at high altitudes [1, 2]. The aerobiological investigations broadly classified into two categories when it conducted in the building or in a closed atmosphere; it is known as intramural aerobiology and aerobiological survey of open space regarded as extramural aerobiology.

Outdoor or extramural aerobiology has the manifold practicable significance and utility. This study involves the experiments conducted for the detection of aero allergenic biocomponents, microscopic and submicroscopic particles of pollen and dust and bacteria, moulds, yeast and viruses, which have their impact on the health of humankind [3]. Fungi are ubiquitous in nature; it has the ability to grow on all the substance available in the environment. The study of airborne fungal spores is of great importance an interest to understand the dissemination, spread and movement of microbes particularly the pathogenic types in the atmosphere. Fungi have been considered primarily as plant pathogens; however, some plant pathogenic fungi are reported as a causative agent of human disease [4], particularly under immune-compromised condition.

Extramural airborne fungal spore have previously been extensively studied in India by several workers [5, 6, 7, 8, 9, 10, 11]. It is supposed that extramural environments are the significant sources of intramural

*Corresponding author:
E-mail: pathak.apurva@gmail.com; Mobile: +91-9425678621.

airborne fungal spores [12]. This extramural study was therefore undertaken to determine the presence and seasonal distribution of the airborne cultivable fungi in the air of the weekly market or Haat area to evaluate the quantity and quality of airborne cultivable fungi in this environment. The concentration of the fungal spores may vary according to the location, altitude, time of day, season of year, condition of the surrounding area and climatic conditions. Consequently, the effects of environmental factors on the total airborne fungal bioload were also analyzed by using correlation analysis and a regression model is prepared for this environment.

2. Material and Methods

The city of Indore (Latitude: 22°48' North; Longitude: 75°48' East; Altitude: 553.) is the biggest city in terms of population of Madhya Pradesh state of India. Indore is a landlocked city, located on the southern fringes of the Malwa Plateau, with the Vindhyas range to the south. The climate of Indore is overall pleasant and salubrious, has a year-round tropical climate generally characterized by warm days and cooler evenings with a cool breeze which is also known as the *Shab-e-Malwa* during the evening. The year may divide into three main seasons; the summer season (middle of March to middle of June), the Monsoon season (middle of June to the end of September), the winter season (November to middle of March) and a transitional period of seasons which observe during the October characterized by moderate to high humidity and moderate temperature.

2.1 Sampling Site

Weekly market of suburban area or Haat is a place where various items like vegetables, food items, groceries, clothes etc. are sold on a weekly basis. Vegetables are being sold in one area of market and other areas are specifically for groceries and general items of daily lives. The weekly market is the backbone of the rural economy, traders from different regions come to sell their products on a weekly basis and people from all walks of life come at this place for fresh vegetables and variety of things of everyday use. The aero- Mycological sampling had been done in two places of the weekly market area, the first site of sampling was selected for this study was the area where vegetables were sold, and another one devoted for general items of daily uses. The distance between the two areas was only 500 Mt., both the areas were characterized as a human activity-enriched site and also a highly trafficked site. The meteorological data collected from weather station Indore. Apart from temperature and humidity five other meteorological parameters (Mean sea level pressure (millibar); Mean wind speeds (Km/h); Maximum sustained wind speed (Km/h); Maximum wind gust (Km/h); Indicator for occurrence of rain or drizzle) were also recorded to analyze their effect on airborne fungal load.

2.2 Isolation from the Air

The Andersen two-stage viable (microbial) particle sampler (2-STG) has been developed for monitoring bioaerosols. It is a multi-orifice, cascade impactor with 400 holes per stage, drawing air at a flow rate of 28.3 L min^{-1}. The different stages separate the airborne particles in size fractions [13, 14]. For this study, air sampling was done on Sabouraud Dextrose Agar (SDA) Medium (HiMedia, Mumbai, India) and HiChrome Candida agar medium (HiMedia, Mumbai, India) at one-meter height from the ground. The sampler was operated for ten minutes at the site in duplicate and the average was taken. For enumeration and medium plate kept on the upper stage of the sampler. The level of airborne fungal spores is usually expressed in term colony forming unit per cubic meter (cfu/m^3). In this atmosphere, cfu/m^3 were calculated by using equation given by Senior (2006) with some modification [15]. Colony forming unit per cubic meter (cfu/m^3) is calculated from the following equation:

$$\text{Colony forming unit} = \frac{1000P}{RT} cfum^{-3} \qquad (1)$$

Where, P is the number of colonies counted on the sample plate after correction by using the positive hole conversion table provided by Andersen (1958) [13], T is the duration of the test in min (10min.), and R is the air-sampling rate in Liters/min (28.3 L min^{-1}) [15].

Processing of samples was done by incubating SDA plates into B.O.D. Incubator at 27 ± 2°C for 2 to 7 days and HiChrome Candida agar medium plate 37 ± 2°C for 24 to 48 hrs. The colonies developed were isolated and identified up to generic level for fungal colonies with the help of standard literature [15, 16, 17, 18, 19, and 20].

2.3 Statistical analysis

The number of samples collected will influence the precision of the exposure estimate and the associated confidence limits [21]. In order to analyze the influence of various environmental factors on the prevalence of airborne fungal population with other environmental factors, correlation coefficients and stepwise linear regression analysis (Criteria: Probability-of-F-to-enter <= .050, Probability-of-F-to-remove > = .100) was done [22, 23, 24].

The bioload of total viable cultivable fungi of weekly market environment obtained by sampling were compared by the paired Student's t-test by using the SPSS Win 12.0 program (SPSS Inc, Chicago, U.S.A.). Differences in the bioload of total viable cultivable fungi between vegetable and general items trading area of weekly market extramural environment were considered to be significant for P of 0.05. The null hypothesis (HO) rejected in favor of the alternative hypothesis (H1) at significance level α (0.05) if; T >tn-1, α/2 (value of the Student table with n -1 degrees of freedom). The null hypothesis was H0: δ = 0 (there is no difference in bioload of total viable cultivable fungi between vegetable and general items trading area of weekly market extramural environment) and the alternative hypothesis was H1: δ = 0 (there is a difference in bioload of total viable cultivable fungi between vegetable and general items trading area of weekly market extramural environment). The percentage frequency (% F) and the percentage contribution (% Con.) of the individual genus were calculated by using the following equation:

$$\%F = \frac{\text{No.of observations in which colony appear}}{\text{Total no of observations recorded}} \times 100 \quad (2)$$

$$\%Con. = \frac{\text{Total number of colonies of one genus}}{\text{Total number of colonies of all genus}} \times 100 \quad (3)$$

All the data were presented in the form of table and figures.

3. Result and Discussion

Airborne fungal spore levels were monitored in the weekly market extramural environment at vegetable and general items trading area for a period of one year to cover a full cycle of the season. Present study reported a mean ± standard deviation (SD) 88.35 ± 21.20cfu/m^{-3} and 81.58 ± 31.95cfu/m^{-3} of viable cultivable total fungal bioload from the weekly market extramural environment at vegetable and general items trading area respectively much lower than previously recorded by researchers [25, 26]. 300 fungal colonies were obtained from 12 samples from a vegetable trading area of weekly market extramural environment. From this environment, 254 colonies of Deuteromycotina followed by 13 colonies of Zygomycotina and nine colonies of Ascomycotina isolated. From Deuteromycotina, 108 colonies of various species belong to the genera *Aspergillus* were reported; similarly, from the general items trading area, of the total 277 colonies; 247 colonies of Deuteromycotina obtained from 12 samples, *Aspergillus* spp. Accounted 90 colonies. Species of the genus *Aspergillus* (36% and 32%), *Cladosporium* (7.3% and 9%), *Alternaria* (4.6% and 9%), *Penicillium* (2.6 % and 8.7%) and *Fusarium* (6.7% and 2.5%) were predominantly present in the weekly market extramural environment at vegetable and general items trading area respectively.

Verma and Sheore (1994) while studying vegetable market environment also reported that Deuteromycotina contributed 87% of the total air spore in which *Aspergillus* were dominant flora and among *Aspergillus* [25], *A. flavus* were reported dominant, similar to the previous finding [27]. The dominance of *Aspergillus* spp. Spores in the vegetable market environment were also previously reported by the scholars [25, 28, 29, 30] (Table 1, 2).

The average concentration of airborne fungi in the weekly market extramural environment is 79.55cfu/m^{-3} (general items trading area), 86.59cfu/m^{-3} (vegetable trading area) in which the average concentration of mitosporic fungi including *Candida* spp. is 21.17cfu/m^3 (vegetable trading area) and 20.58cfu/m^3 (general items trading area), accounting for 85% and 89% respectively; the average concentration of Zygomycotina 1.1cfu/m^3 (vegetable trading area) and 1.0cfu/m^3 (general items trading area), accounting for 4.3% and 3.3% respectively; the concentration of Ascomycotina reported less than one cfu/m^3 for vegetable trading area and general items trading area, accounting for 3% and 1% respectively; the concentration of unidentified fungi 2.0cfu/m^3 (vegetable trading area) and 1.5cfu/m^3 (general items trading area), accounting for 6% and 5% respectively (Fig. 1).

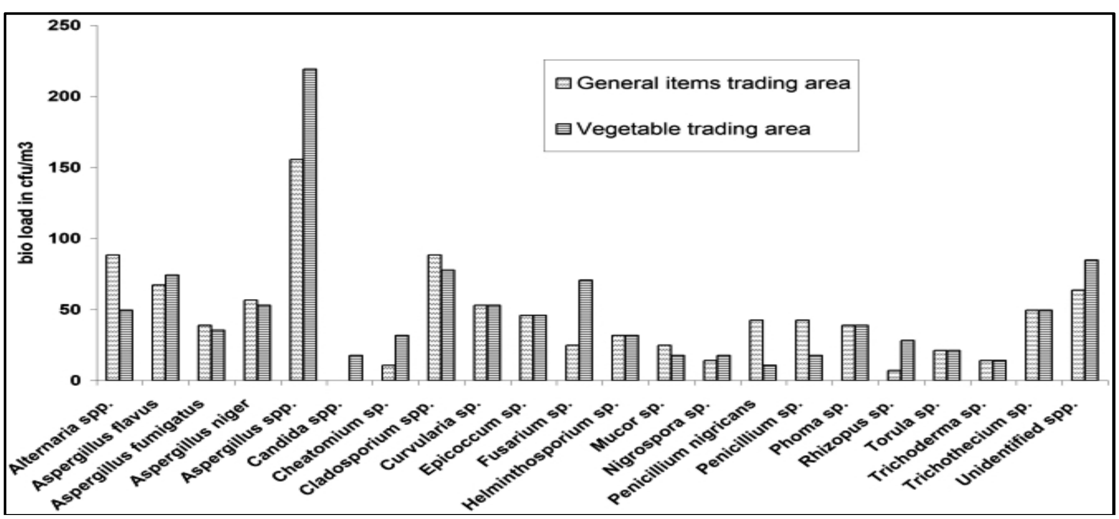

Fig. 1. Bioload of airborne cultivable fungi of weekly market extramural environment during the study period.

Table 1. Airborne cultivable fungi of weekly market associated vegetable trading zone during the study period.

FUNGAL COLONIES	JAN	FEB	MAR	APRIL	MAY	JUNE	JULY	AUG	SEPT	OCT	NOV	DEC	Total	Cfu/m^{-3}	%F	%Con.
Alternaria spp.	3	2		1		2	3	2			1		14	49.5	58.3	5.05
Aspergillus flavus		3	1		2	3		4	1	1	4	2	21	74.2	75	7.58
Aspergillus fumigatus		3		1			2		1	3			10	35.3	41.7	3.61
Aspergillus niger	2	1		2	1			4		2		3	15	53	58.3	5.42
Aspergillus spp.	8	3	6	8		2	5	9	5	4	4	8	62	219	91.7	22.4
Candida spp.					3		2						5	17.7	16.7	1.81
Chaetomium sp.			2						3		4		9	31.8	25	3.25
Cladosporium spp.	1	3	4		1	3		1		1	3	5	22	77.7	75	7.94
Curvularia lunata	4			5				1		3		2	15	53	41.7	5.42
Epicoccum nigrum		3	2	3				5					13	45.9	33.3	4.69
Fusarium sp.			4		1	2	4			4	2	3	20	70.7	58.3	7.22
Helminthosporium sp.		4				2				1	2		9	31.8	33.3	3.25
Mucor mucedo	3				1			1					5	17.7	25	1.81
Nigrospora oryzae						1		3				1	5	17.7	25	1.81
Penicillium nigricans					3								3	10.6	8.33	1.08
Penicillium sp.				5									5	17.7	8.33	1.81
Phoma betae				3	1				5		2		11	38.9	33.3	3.97
Rhizopus nigricans			3		2	1			1	1			8	28.3	41.7	2.89
Torula sp.	2					1						3	6	21.2	25	2.17
Trichoderma viride						2						2	4	14.1	16.7	1.44
Trichothecium roseum	2							2		5		5	14	49.5	33.3	5.05
Unidentified sp.	2	3	1	3	2	2	2	1	4	2		2	24	84.8	91.7	8.66

Table 2. Airborne cultivable fungi of weekly market associated general item trading zone during the study period.

FUNGAL COLONIES	JAN	FEB	MAR	APRIL	MAY	JUNE	JULY	AUG	SEPT	OCT	NOV	DEC	Total	Cfu/m^{-3}	%F	%Con.
Alternaria spp.	3	4		2		2	3	5	2		1	3	25	88.4	75	9.03
Aspergillus flavus		2	1			3		6	2		3	2	19	67.1	58.3	6.86
Aspergillus fumigatus		3			1		4			3			11	38.9	33.3	3.97
Aspergillus niger	2			5				4		2		3	16	56.5	41.7	5.78
Aspergillus spp.	1	5	4	8		2	5	6	6		2	5	44	155	83.3	15.9
Chaetomium globosum								1	2				3	10.6	16.7	1.08
Cladosporium sp.		4	5			4	2	1		1	4	4	25	88.4	66.7	9.03
Curvularia sp.	4			5				1		3		2	15	53	41.7	5.42
Epicoccum sp.		3	2	3				5					13	45.9	33.3	4.69
Fusarium moniliforme			4				2				1		7	24.7	25	2.53
Helminthosporium sp.		4								1	4		9	31.8	25	3.25
Mucor mucedo	3				1	2		1					7	24.7	33.3	2.53
Nigrospora oryzae								3				1	4	14.1	16.7	1.44
Penicillium nigricans					3		2		4			3	12	42.4	33.3	4.33
Penicillium spp.		3		5				3		1			12	42.4	33.3	4.33
Phoma spp.				3	1				5		2		11	38.9	33.3	3.97
Rhizopus sp.			1							1			2	7.07	16.7	0.72
Torula sp.	2					1						3	6	21.2	25	2.17
Trichoderma viride						2						2	4	14.1	16.7	1.44
Trichothecium sp.	2							2		5		5	14	49.5	33.3	5.05
Unidentified sp.	2	1	3	1		1	4		3	1		2	18	63.6	75	6.5

Airborne viable fungal spore concentrations varied according to the season [23, 31]. Highest average recorded bioload from the weekly market extramural environment is during Monsoon was 98.952cfu/m^3 (general items trading area) and in winter was 96.8316cfu/m^3 (vegetable trading area) similar to previous studies [5]. During the Monsoon in August and during the winter in the month of December, the highest average bioload was recorded in the ambient air of weekly market associated extramural environment, followed by April and February. During the summer the mean of the bioload recorded for this environment was lowest (Fig. 2).

According to Adhikari *et al*., (2004) and Kasprzyk (2008) in tropical and subtropical regions airborne viable fungal spore concentrations were reported lowest during the summer reiterates the present findings [27, 32].

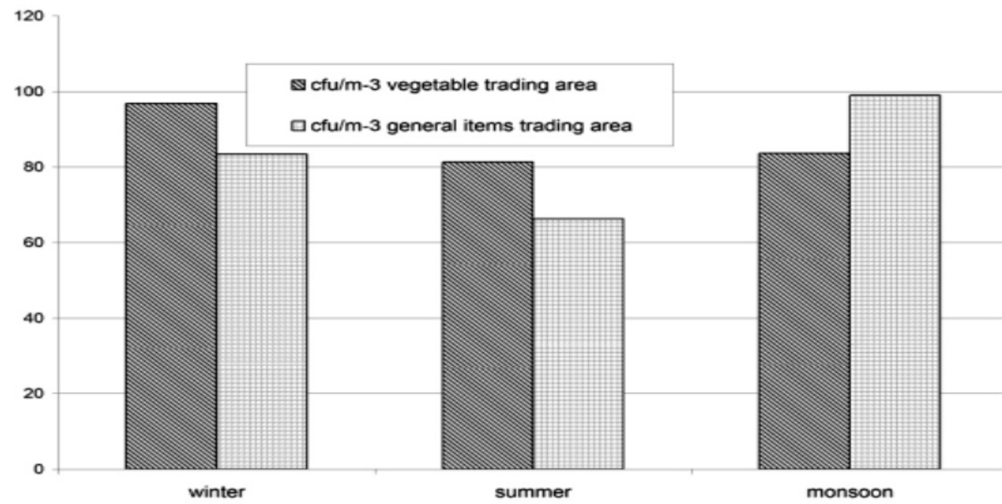

Fig. 2. Seasonal distribution of airborne cultivable fungi of weekly market extramural environment during study period.

There is no correlation (r) established between the environmental factors like dew point (r: -0.010), humidity (r: 0.258), sea level pressure (r: 0.163), wind (r: -0.164) and temperature (r: -0.490, Sig.: 0.053) with the bioload of the general items trading area of weekly market extramural environment, whereas; bioload of a vegetable trading area of weekly market extramural environment (r: .794, Sig.: 0.001) significantly positively correlated with the bioload of the general items trading area of weekly market extramural environment. Previous studies in the vegetable market area reported that humidity and temperature were the major factors for dissemination and distribution of microorganism in the vegetable market area; however, other factors like dwellers, transporters, services, domestic animals, water spray, and putrefaction are major contributors for the generation and aerosolization of airborne microorganisms [33].

The multiple regression models predicted that there were 0.527 unit increases in bacterial bioload accounted for about 63.1% of the variance by increasing one unit of the bioload of vegetable trading area in the bioload of the general items trading area of weekly market extramural environment were obtained. The constant was 45.335 showing any bioload of vegetable trading area. Square Root of Mean Square Error for the model was 11.105; bioload could vary by ± 11.105 about the estimated regression equation for the value of bioload of the vegetable trading area of weekly market extramural environment (± 0.127).

An estimated model for total viable cultivable fungi of the general items trading area of weekly market extramural environment (R 63.1%) = 45.335 + 0.527× bioload of a vegetable trading area of weekly market extramural environment. (4)

To test the hypothesis of no difference or no relationship between the bioload of total viable cultivable fungi of vegetable trading area and general item trading area of weekly market extramural environment paired t-test was performed (Table 3). The significance value is high (0.262) and the confidence interval for the mean difference contains zero, it means that there is no significant difference in quantity between the mean values of the bioload of total viable cultivable fungi of vegetable trading area and general item trading area of weekly market extramural environment.

Table 3. Paired Samples t-Test: Airborne cultivable fungi of vegetable trading zone and general item trading zone of weekly market extramural environment during the study period.

			Pair
			cfu/m-^{3} vegetable trading zone - cfu/m-^{3} general item trading zone
Paired Differences of upwind and downwind bioload	**Mean**		-7.068000
	Std. Deviation		21.940834
	Std. Error Mean		6.333773
	95% Confidence Interval of the Difference	**Lower**	-21.008541
		Upper	6.872541
t			-1.116
df			11
Sig. (2-tailed)			0.288

4. Conclusion

The recovery of *Candida* spp. at vegetable trading area, the high significant value of paired t-test, highest average recorded bio load during Monsoon season and the week correlation with humidity and temperature with the bioload of weekly market extramural environment shows, though the temperature and humidity were not a key factor for the generation, dissemination and distribution of airborne fungal spore for this environment, yet it was responsible for the tenacity of the microorganism. Moreover, the weekly market environment was characterized as a human activity-enriched site and also a highly trafficked site and hence, the bioloads of the airborne fungal spores of environment under study were might be due to the mechanical disturbances.

References

[1]. Brock, T.D. (1966). Principles of microbial ecology. Prentitute of Biology, London.

[2]. Shivaji, S., Chaturvedi, P., Begum, Z., Pindi, P.K., Manorama, R., Padmanaban, D.A., Shouche, Y.S., Pawar, S., Vaishampayan, P., Dutt, C.B., Datta, G.N., Manchanda, R.K., Rao, U.R., Bhargava, P.M., Narlikar, J.V. (2009). *Janibacter hoylei* sp. nov., *Bacillus isronensis* sp. nov. & *Bacillus aryabhattai* sp. nov., isolated from cryotubes used for collecting air from the upper atmosphere. *Int. J. Syst. Evol. Microbiol.*, 59:2977-2986.

[3]. al-Dagal, M. & Fung D.Y. (1990). Aeromicrobiology—a review. *Crit. Rev. Food Sci. Nutr.*, 29:333–340.

[4]. Deighton, F.C. & Mulder J.L. (1977). *Mycocentrospora acerina* as a human pathogen. *Trans. Br. Mycol. Soc.*, 69(2):326- 327.

[5]. Verma, K.S. (1989). Atmospheric biopollutants in rural and urban sites of Jabalpur city. *Frontr. Bot.* II & III (1, 2): 57-62.

[6]. Gopi, T.V., Kumar, R.P., Ravindran, P., Nair, P.K.K. (1990). Comparative analysis of airspora of two urban localities in Kerala. *Ind. J. Aerobiol.*, 3:39–44.

[7]. Datta, T.R. & Jain, A.K. (1990). Aerobiological studies at Gwalior-fungal spores in a textile mill area. *Bionature*, 11: 77-81.

[8]. Bhat, M.M. & Rajasab, A.H. (1991). Incidence of airborne fungal spores at two different sites in Gulbarga during 1986. *Indian J. Aerobiol.*, 4: 1-6.

[9]. Singh, A.B., Singh, A. & Pandit, T. (1999). Respiratory diseases among agricultural industry workers in India: cross-sectional epidemiological study. *Ann. Agric. Environ. Med.*, 6:115-126.

[10]. Chauhan, S.V.S. & Kulshrestha, A. (2004). Comparative incidence of fungal spores of two gardens of Agra city. *Indian J. Aerobiol.*, 17:12-16.

[11]. Kumar, P., Mahor, P., Goel, A.K., Kamboj, D.V. & Kumar, O. (2011). Aero-microbiological study on distribution pattern of bacteria and fungi during weekdays at two different locations in urban atmosphere of Gwalior, Central India. *Sci. Res. Essays*, 6(25):5435-5441.

[12]. Streifel, A.J., Stevens, P.P., Rhame, F.S. (1987). In-hospital source of airborne *Penicillium* species spores. *J. Clin. Microbiol.*, 25(1): 1-4.

[13]. Andersen, A.A. (1958). New sampler for collection, sizing, and enumeration of viable airborne particles. *J. Bacterial.*, 76: 471 – 484.

[14]. Andersen, A.A. (1966). A sampler for respiratory health hazards assessment. *Am. Ind. Hyg. Assoc. J.*, 27: 160 - 165.

[15]. Senior, B.W. (1996). Examination of water, milk, food and air. In: Collee JG, Fraser AG, Marmion BP, Simmons A, editors. Mackie & McCartney practical medical microbiology, 14th ed. London: Churchill Livingstone; p. 883-921.

[16]. Clements, F.E. & Shear, C.L. (1954). *The genera of fungi*. Hafner Publishing Co., New York, USA, p. 496.

[17]. Barnett, H.L. & Hunter, B.B. (1972). Illustrated genera of imperfect fungi. 3rd Edition, Burgess Publishing Company, Minneapolis, Minnesota.

[18]. Dennis, R.W.G. (1978). British Ascomycetes. J. Cramer, Vaduz, Germany, p. 585.

[19]. Alexopoulos, C.J. & Mims, C.W. (1979). Introductory Mycology (3rd edn.). John Wiley and Sons, Inc., USA.

[20]. Forbes, B.A., Sahm D.F. & Weissfeld, A.S. (2002). Bailey and Scott's Diagnostic Microbiology, 11th Ed., St. Louis: Mosby Publication, USA.

[21]. Grantham, D. (2001). Simplified Monitoring Strategies. Tullamarine, Australian Institute of Occupational Hygienists (AIOH).

[22]. Quinn, G.P. & Keough, M.J. (2002). Experimental Design and Data Analysis for Biologists. Cambridge University Press, Cambridge, UK, pp. 550

[23]. Verma, K.S. & Pathak, A.K. (2009). A Comparative Analysis of Forecasting Methods for Aerobiological Studies. *Asian J. Exp. Sci.*, 23(1):193-198.

[24]. Pathak, A.K. & Verma, K.S. (2010). Extramural Aero-bacteriological Quality of Hospital Environment. *Asian J. Exp. Biol. Sci.*, 1(1):128-135.

[25]. Verma, K.S. & Sheore, L. (1994). Aerobiota of a vegetable market at Jabalpur. *J. Environ. Biol.*, 15(4):325-329.

[26]. Sahney, M. & Purwar, A. (2002). Incidence of fungal airspora in the market area of Allahabad. *Indian J. Aerobiol.*, 15: 32-45.

[27]. Adhikari, A., Sen, M.M., Gupta-Bhattacharya, S. & Chanda, S. (2004). Volumetric assessment of airborne fungi in two sections of rural indoor dairy cattle shed. *Environ. Int.*, 29:1071– 1078.

[28]. Sandhu, R.S., Khan, Z.U. & Randhawa, H.S. (1976). Natural occurrence of *Aspergillus fumigatus* in cane sugar mills. *Sabouraudia*, 15:263-272.

[29]. Chauhan, S.V.S., Kulshreshta, A. & Goyal, R. (2004). Airborne fungal spora of Agra city with special reference to allergic significance. *Indian J. Aerobiol.*, 17: 17-22.

[30]. Bagwan, N.B. (2010). Seasonal variations in aeromycoflora of vegetable market at Udgir, Maharashtra, India. *J. Mycol. Pl. Pathol.*, 40(3):360-364.

[31]. Oliveira, M., Ribeiro, H. & Abreu, I. (2005). Annual variation of fungal spores in atmosphere of Porto: 2003. *Ann. Agric. Environ. Med.*, 12:309–315.

[32]. Kasprzyk, I. (2008). Aeromycology – main research fields of interest during the last 25 years. *Ann. Agric. Environ. Med.*, 15:1–7.

[33]. Pathak, A.K. & Verma, K.S. (2009). Aero-bacteriological study of vegetables market at Jabalpur. *Iran J. Environ. Health Sci. Eng.*, 6(3):187-194.