Int. J. Mendel, Vol. 31 (1-2), 5-9, 2014 AEROPHYCOLOGY ISSN 0970-9649

SIGNIFICANCE OF MICROALGAL AND CYANOBACTERIAL AEROSOLS ISOLATED FROM A SLEEPING ENVIRONMENT - A TWO YEAR STUDY IN MAHARASHTRA

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Key words : Microalgae, Cyanobacteria, aerosal, sleeping environment, Maharashtra.

Experiments were conducted at weekly intervals for a period of two years to isolate microalgal and cyanobacterial aerosols present in the air of a sleeping area of a poorly constructed rural house at Patonde in Chalisgaon, Maharashtra. Duration of the exposure was 8 hours (10 pm to 6 am), which coincided with the sleeping time of the inhabitants. 31 and 37 samples were collected by using agarized BBM as the nutrient medium in first and second year of the study respectively. In the first year, of the 12 genera isolated, 10 represented cyanobacteria and 2 chlorophycean microalgae, while in the second year, of the 20 genera isolated, 15 belonged to cyanobacterial group and 5 to chlorophyceae. Cyanobacteria like *Phormidium, Aphanothece, Scytonema, Nostoc, Lyngbya, Plectonema* and *Myxosarcina* were recorded frequently while, *Chlorella*, a chlorophycean was more frequent among the microalgae. Almost all the genera isolated were allergenically significant.

INTRODUCTION

There are increasing number of instances of airborne algae and cyanobacteria being isolated from diverse habitats (Ramchander Rao and Jadhav, 1997) both from indoor and outdoor environments. Indoor dust besides being a rich source of bacterial and fungal microorganisms entraps a variety of micro algal spores and filaments (Jadhav and Ramchander Rao, 1995). In the earlier investigations at Nagpur (Gadewar and Tarar, 1997) the airborne algae in indoor environment were held primarily responsible for bronchial allergy. Indoor habitats like residential houses (Bernstein and Safferman, 1970 & 1973; Benaim Pinto, 1972; Mittal, 1982; Ramchander Rao and Jadhav, 1997 and Gadewar and Tarar, 1997), bath rooms (Jadhav and Ramchander Rao, 1995), hospital and library (Gadewar and Tarar, 1997) were investigated earlier to isolate microalgae. However, in our knowledge, ours is the first attempt where a sleeping environment is being studied hitherto unexplored.

MATERIALS AND METHODS :

Sleeping environment of a poorly built rural home was selected at Patonde near Chalisgaon in Maharashtra State. Samples were collected at weekly interval by using the petriplates containing agarized BBM (Bold's Basal Medium), composition given by Bold (1942), as the nutrient medium. Petriplates were exposed by keeping the open side facing up on the floor of the sleeping room, since the inhabitants were sleeping on floor. The exposure time coincided with the sleeping time of the inhabitants i.e., from 10 pm to 6 am (8 hours duration). The exposed petriplates were later incubated as per the method described earlier (Jadhav and Ramchander Rao, 1995). Upon visualizing the growth as discrete colony forming units (CFU), those colonies were picked up and identified under the light microscope by referring to the standard literature on algae (Desikachary, 1959; Fritsch, 1935 & 1945).

RESULTS :

A total of 3 l and 37 airsamples were collected in the first and second years of investigations, respectively. In the first year a total of 95 CFUs were detected while in the second year the colony count was 133. During first year 2 microalgal (chlorophyceae) and 10 cyanobacterial genera were isolated whereas in second year 5 microalgal and 15 cyanobacterial genera were isolated. Monthwise occurrence of diverse genera is described in Table 1a and 1b for the first and second years, respectively. *Phormidium* occurred in 8 months followed by *Aphanothece* (5), *Lyngbya* (5), *Plectonema* (5) and *Chlorella* (4) in first year while in second year Phormidium recorded in all 12 months followed by *Aphanothece* (11), *Scytonema* (7), *Nostoc* (6) and *Myxosarcina* (5). The seasonwise occurrence and abundance of various genera (Table 2a & 2b) in first and second year revealed *Phormidium* as the dominant and abundant genus. During first year maximum number of genera were isolated in the monsoon (8) season followed by winter (7), summer (4) and post monsoon (4). In the second year the maximum number of genera were isolated in the monsoon (14) season followed by summer (12), winter (11) and post monsoon (5). Frequency of occurrence indicated Phormidium as the most frequently encountered genus.

DISCUSSION :

In the first year of study 95 CFUs were recorded at an average of 3 CFUs per plate while it was 133 CFUs in second year with an average of 4 CFUs per plate. Average numbers of colonies developed per plate are comparable to our earlier investigations (Ramchander Rao and Jadhav, 1997). Indoor environments and enclosed spaces are increasingly observed as the places of health concern for public health at large, majorly due to the airborne microorganisms and their constituents (Sharma and Rai, 2008).

Of the 21 genera recorded from sleeping environment 16 belonged to cyanobacteria (76%), rest all (5) belonged to chlorophyceae (24%). Evidently, cyanobacterial group was the most predominant group in the present study. This is in conformity with the results of investigations at Aurangabad (Tilak, 1992), Pune (Balakrishnan and Gunale, 1980) and Delhi (Mittal, 1981). Joshi and Mukundan (1997) while working on

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algal disfigurement and degradation of architectural paints in India observed the cyanobacterial group to be dominant on surfaces painted with acrylics, cement based coatings and oil based enamels. In a recent study in Varanasi, Sharma et al. (2006) observed the dominance of cyanobacteria over other diverse groups of aeroalgae attributing it to their broad ecological distribution and tropical climate. Physiologically cyanobacteria have the advantage as many of these genera can tolerate the air dried state for prolonged periods (Malcolm Potts, 1999) and are capable of getting dispersed in viable state either to shorter or longer distances with wind currents. Tropical conditions perhaps, promote the rich and diverse growth of cyanobacteria.

Out of a total of 68 samples, the genus *Phormidium* was found in 42 samples (62%) which showed its dominance in the atmosphere. Recently, widely distributed cyanobacterium *Phormidium* has been added to the list of genera capable of producing cyanotoxins, viz., anatoxin-a (Gugger *et al.*, 2005). Incidentally, *Phormidium* happens to be one of the most predominant cynobacterial genera distributed in the atmosphere, particularly in the tropical countries. This genus was also studied for its allergenicity and categorized now as a listed potential aeroallergen. However, the toxins present in the atmosphere and the probable entry of these toxins by inhalation into the human subjects need to be thoroughly investigated, in view of increasing number of reports on human exposure to cyanobacterial and HAB (Harmful Algal Bloom) toxins (Kirkpatrick *et al.*, 2006). Of the probable routes of algal sensitization, Bernstein and Safferman (1966) observed inhalation of the organisms as the primary consideration. People exposed to aerosolized brevetoxins experience upper and lower respiratory irritation and some inflammatory response (Backer *et al.*, 2003). Bernstein and Safferman (1973) observed that the patients with symptoms of perennial allergic rhinitis have a high incidence of skin reactivity to various green algae, particularly to a *Chlorella* species isolated from unprocessed dust used in the commercial production of house dust extracts, indicating the heterogeneity and complexity of the dust.

A correlationship was observed between the outdoor and indoor dust borne algae by Ramchander Rao and Jadhav (1997), who concluded that the outdoor dust is the chief contributor to the indoor one. The house selected in the present investigation being a rural one and as there was continuous movement of the people in and out of the house, the foot and footwear dust might have brought enough dust carrying wet or dry forms of the algal and cyanobacterial cells or filaments into the sleeping area as well. Human activity in and around the sampling site in the day time, during indoor air investigations, resulted in increased concentration of airborne microorganisms (Buttner and Stetzenbach, 1992). Investigations conducted earlier (Green *et al.*, 1962) showed significant increase in the airborne concentration of bacteria due to foot traffic. In a recent and interesting study conducted in Kuala Lumpur city, Malaysia, Chu *et al.* (2013) recorded 14 taxa of AAC (Airborne Algae and Cyanobacteria) in an occupational environment (office building). The taxa like *Phormidium* and *Chroococcus* (cyanobacteria) and *Chlorella* and *Chlorococcus* were predominant. The highest occurrence of AAC within the occupational environment was recorded in the ground floor, an area exposed to the outdoor environment, suggesting that the ventilation and human carriers bring in lot of dust into the occupational environments.

While working on the algae over Antarctica Kol and Flint (1968) recorded several species of green algae (*Chlamydomonas* and *Ankistrodesmus*), blue green algae (*Phormidium*) and diatoms (*Nitzschia*) from the green ice. It surprised the investigators as such algae could be isolated from a rigorous and isolated habitat and raised several questions about the biology and ecology of algae. This significantly underlines the capacity of the first colonizers of the earth to grow in any kind of environment virtually possible on this planet.

CONCLUSION :

Indoor habitats and occupational environments are no exception for the presence of the microorganisms. However, the quality of the air and dust components is important and matters the most in terms of human sensitization. All known varieties of microorganisms and insects are reported either from the indoor air or dust. Sleeping area of the inhabitants is very important as it is the place where people take rest at least for 8 hours in the night time. It is also pertinent to know what they inhale into their respiratory tract during their sleep. This helps in understanding the components what the asthma and allergy sufferers inhale during night time when they are asleep.

ACKNOWLEDGEMENTS :

We gratefully acknowledge ICMR for a research grant for conducting the present investigation. We also thank C.E.S. College of Science, Chalisgaon (M.S.) for providing facilities.

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TABLE-1a : Monthwise occurrence of different genera in the first year of Investigation.

Sl. No.	Name of the Genus	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct.	Nov.	Dec.
01.	*Aphanothece*						+		+		+	+	+
02.	*Chroococcus*			+		+							
03.	*Gloeocapsa*	+	+										
04.	*Gloeotrichia*		+										
05.	*Lyngbya*	+	+	+	+								
06.	*Myxosarcina*								+				
07.	*Nostoc*		+										
08.	*Phormidium*	+	+		+			+	+		+	+	+
09.	*Plectonema*	+					+	+		+	+		
10.	*Scytonema*								+			+	+
11.	*Chlorella*		+		+	+	+						
12.	*Gloeocystis*						+						

+ denote presence

TABLE-1b : Monthwise occurrence of different genera in the second year of Investigation.

Sl. No.	Name of the Genus	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct.	Nov.	Dec.
01.	*Aphanocapsa*		+				+						
02.	*Aphanothece*	+	+	+	+	+		+	+	+	+	+	+
03.	*Aulosira*							+					
04.	*Calothrix*			+			+						
05.	*Chroococcus*						+						
06.	*Glococapsa*						+	+					
07.	*Gloeothece*												+
08.	*Hapalosiphon*			+									
09.	*Lyngbya*	+			+	+	+						
10.	*Myxosarcina*	+			+		+	+					+
11.	*Nostoc*		+	+	+	+						+	+
12.	*Phormidium*	+	+	+	+	+	+	+	+	+	+	+	+
13.	*Plectonema*		+	+		+	+	+	+	+	+	+	+
14.	*Scytonema*	+		+	+	+	+	+		+			
15.	*Symploca*			+			+						
16.	*Ankistrodesmus*			+									
17.	*Chlorella*						+						
18.	*Chlorococcum*		+			+							
19.	*Gloeocystis*							+		+			
20.	*Trebouxia*		+										

+ denote presence