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Studies on Environmental Monitoring of Microbial Air Flora in the Hospitals

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Indoor air quality is an important determinant of human health and comfort. Airborne bacteria can also contributes to indoor air pollution. The aerobiological survey was carried out in indoor and outdoor environment at 76 hospitals in Amravati. The total 670 air samples were analyzed from indoor and outdoor environment of hospitals by using sedimentation method. The most prominent bacteria isolated were *Staphylococcus aureus* (29.59%), *Pseudomonas aeruginosa* (19.72%). The *Staphylococcus saprophyticus*, *Proteus mirabilis*, *Escherichia coli and Enterobacter aerogenes* were in the range of 2-6%. The rest of bacterial pathogens *Pseudomonas fluorescence*, *Proteus vulgaris*, *Morganella morganii*, *Citrobacter freundii*, *Serratia marcescens* and *Klebsiella pneumoniae* were below 1%. Out of all the hospital examined, maternity and children hospitals showed highest (50.68%) bacterial isolates, which were the highest among all types of hospitals.

Key words: Airborne pathogens, hospital environment, hospital air flora

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INTRODUCTION

Indoor air quality is an important determinant of human health and comfort. There are large evidences on the hazardous nature of indoor air pollutants, on their sources or conditions leading to human exposure. The indoor air quality of hospitals has become an important issue now days. The airborne route of transmission is important for a number of pathogenic microorganisms in hospital buildings (Beggs, 2003). As it is, 5% of all patients who go to hospitals for treatment will develop an infection while they are there. This is because the density of pathogens is greater in hospitals than in most other environments. Indeed, it has been estimated that the airborne route of transmission accounts for between 10 and 20% of endemic nosocomial infections (Brachman, 1970). Unfortunately, hospitals tend to be places where harmful organisms are concentrated.

Airborne transmission is known to be the route of infection for diseases. It has also been implicated in nosocomial outbreaks of methicillin resistant *Staphylococcus aureus* (MRSA) and *Pseudomonas aeruginosa*. Greene *et al.* (1962) reported 42.6% gram-positive cocci and 14% gram-negative rods in hospitals air. Human exposure to these airborne microorganisms may results in adverse health effects, infectious diseases (Sattar and Ijaz, 1987), allergic and irritant responses (Croft *et al*., 1986), respiratory problems (Jacobs, 1989) and hypersensitivity reactions (Woodward *et al*., 1988; Tambekar and Gulhane, 2003).

The contribution made by airborne pathogens towards nosocomial infection and the role played by aerosolized microorganisms is unclear. The fact that many airborne microorganisms are viable even though they are non-culturable (Heidelberg *et al*., 1997) is of importance. Indeed, it might explain why Greene *et al*. (1962) found relatively few gram-negative bacilli when they sampled the air in hospitals. It therefore follows that airborne transmission of infectious agents in hospital buildings is likely to be greater than is currently recognized.

Up till the work on indoor air quality has been conducted in farms, caves, industries, dwelling houses, library buildings, poultry sheds, green houses (Tilak *et al*., 1985), museums and libraries (Manoharachary *et al*., 1997), school building (Razek *et al*., 2000), college field, market area, saw mill area (Basumatary *et al*., 2002). In all the above-mentioned projects, the study has been focused on studies on fungal flora. In comparison, relatively little work has been undertaken on the bacteriological aspects of indoor air quality. The impact of airborne microorganisms on indoor and outdoor air quality of hospital and impact on human health remains poorly understood. Thus, the relative lack of research into the airborne transmission of bacteria tends to conduct the present study for assessment of air contamination by bacterial pathogens in the 76 hospitals of Amravati city.

MATERIALS AND METHODS

Sample collection site: The aero biological survey was carried out in indoor and outdoor environment at 76 hospitals in Amravati which includes 6 general hospitals, 37 maternity and children hospitals, 5 multi-specialty hospitals, 9 cardiac hospitals, 6 each of orthopedic hospitals and eye, nose and throat (ENT) hospitals, 1 each of cancer hospital, dental hospital and mental hospital and 4 clinics.

Aerobacterial flora analysis: The total 670 air samples were analyzed from indoor and outdoor environment of hospitals by using sedimentation method (Mathias *et al*., 2000) and air sampler (Hi-media, Mumbai). The petridishes containing mannitol salt agar, MacConkey agar and cetrimide agar were exposed for 5 min in air to sample particles at 1 cubic foot height. The plates were incubated at 37°C for 48 h and examined for types of bacteria. The bacterial isolates were identified using standard procedure (Bergey's Manual of Determinative Bacteriology, 1974).

RESULTS AND DISCUSSION

The total 670 air samples were analyzed from indoor and outdoor environment of hospitals, out of these, 953 strains of 15 bacteria were isolated. Out of them 457 were from indoor and 496 from outdoor hospital environment. The most prominent bacteria isolated were *Staphylococcus aureus* (29.59%), *Pseudomonas aeruginosa* (19.72%), *Micrococcus luteus* (16.05%) and *Staphylococcus epidermidis* (15.84%). The *Staphylococcus saprophyticus*, *Proteus mirabilis*, *Escherichia coli and Enterobacter aerogenes* were in the range of 2-6%. The rest of bacterial pathogens *Pseudomonas fluorescence*, *Proteus vulgaris*, *Morganella morganii*, *Citrobacter freundii*, *Serratia marcescens* and *Klebsiella pneumoniae* were below 1% (Fig. 1).

The indoor environment refers to inside of general wards, private rooms, Operation Theater (OT), labour rooms, Intensive Care Unit (ICU), pathology laboratories, X- ray rooms, Trade Meal Test-Pulmonary Function Test (TMT-PFT) rooms, Electro Cardio Graphy-Electro Encephalo Graphy (ECG-EEG) rooms, sonography rooms, lithotripsy rooms, pschychologist's rooms, dressing rooms, gynecology wards, medicine wards, pediatric

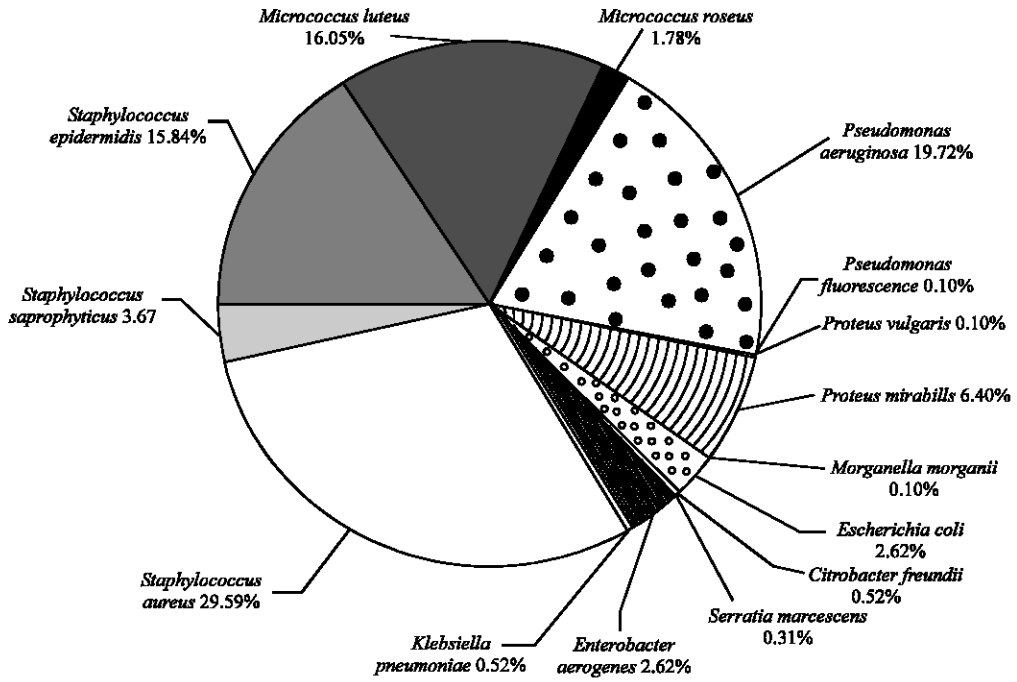

Fig. 1: Total types of airborne pathogens isolated in hospital environment of Amravati

wards, surgery wards, ENT wards and burn wards. The outdoor environment refers to corridors of these wards and sections including OPD's, which are the out patient department or the examination areas.

The occurrence of *Staphylococcus aureus* (13.22%) and *Micrococcus luteus* (6.71%) was less in indoor than in outdoors i.e., 16.36 and 9.33% respectively. At any one time approximately 30% of healthy people are carriers of *Staphylococcus aureus*. It is an opportunistic pathogen, which causes infection at sites of lowered host resistance, such as damaged skin or mucous membranes (Arbuthnott, 1992). The micrococci are parasitic on mammalian skin (Ananthanarayan and Paniker, 2000).

The occurrence of more percentage in the outdoor air of these organisms suggested that the source was the shedders. Shedders can disperse large numbers of cocci into the environment, resulting in high concentrations of airborne staphylococci, which may remain viable for long periods of time. If the visitors, auto rickshaw drivers, healthcare personnel and other people are heavy shedders then, the outdoor air becomes occupied with *Staphylococcus aureus* and *Micrococcus luteus*. It is generally the case that gram-positive microorganisms survive much longer in the aerosolized state than gram-negative bacteria (Sands and Goldmann, 1998). Thus their presence was more in outdoor air.

Coagulase negative staphylococci (CNSs) (19.51%) were found to be very less as compare to *Staphylococcus aureus*. They are commonly found on the skin of healthy persons and rarely cause infections, except in immuno-compromised patients (Arbuthnott, 1992). The transmission route for coagulase negative staphylococci is airborne, which has been observed from staff in an operating room during implant surgery (Lidwell *et al*., 1982).

Pseudomonas aeruginosa concentration was high in indoor air (10.38%) than outdoor air (9.33%) while other members of enterobacteriaceae were found less in number in the outdoor air of the hospitals. *Pseudomonas aeruginosa* is difficult to eradicate from hospital wards as it is resistant to and may multiply in, many of the disinfectants and antiseptics commonly used in hospitals. This is the main reason why *Pseudomonas aeruginosa* is more in indoors than outdoor. The few studies suggested that airborne transmission played an important role in *Pseudomonas* sp. infection as it was isolated in burns units via the airborne route (Govan, 1992). Blessing-Moore *et al*. (1979) recovered *Pseudomonas aeruginosa* from settle plates near patients with cystic fibrosis. *Pseudomonas* sp. along with other gram-negative bacilli can be recovered from hospital air. However, the few studies indicated that *Pseudomonas* sp. play an important part in airborne transmission (Zimakoff *et al*., 1983).

The concentration of gram-negative bacteria (18.99%) was more in indoors than outdoors. Although it is generally true that gram-positive bacteria survive longer in the aerosolized state than gram-negative bacteria, there

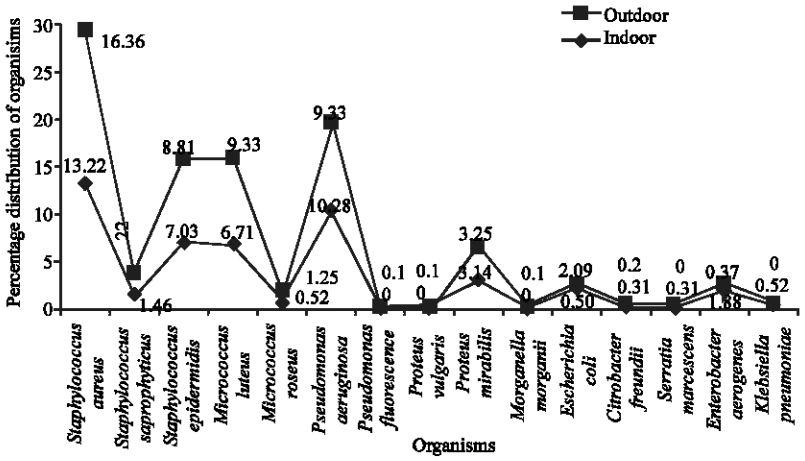

Fig. 2: Bacterial pathogens isolated from indoor and outdoor hospital environment in Amravati

is growing evidence that gram-negatives can survive in the aerosolized state (Jawad *et al*., 1996). According to Beggs (2003) Tambekar *et al*. (2005) the enterobacteriaceae members such as *Proteus vulgaris*, *Morganella morganii*, *Citrobacter freundii*, *Serratia marcescens* and *Klebsiella pneumoniae* were least air contaminated, as the source of contamination may be water droplets and they may not survive for long period in the aerosolized state (Fig. 2).

Maternity and children hospitals showed 50.68% bacterial isolates which were the highest among all types of hospitals (Fig. 3). This may be due to unhygienic state of children patient with their parents and more crowds as well as nearby and open defecation. The airborne contamination of fomites, i.e. curtains and furnishings and of floors plays a role in the spread of airborne bacteria (Beggs, 2003). Thus the dust, skin squamae on the surfaces may get airborne and contributes in the highest microbial flora of maternity hospital. Intestinal organisms, through dried particles of feces, from napkins of infants, also get disseminated (Ananthanarayan and Paniker, 2000).

General hospitals had 17.62% pathogenic bacterial flora. The most common contaminants were *Staphylococcus aureus* (4.61%), *Staphylococcus epidermidis* (3.67%), *Micrococcus luteus* (2.51%) and *Pseudomonas aeruginosa* (3.88%). As patients attending these hospitals have lower socio-economic status. The dirty clothes and the skin of those people may contribute in the airborne organisms in the hospitals. The ultimate source of common pathogenic organisms is dust derived from human beings. The more occurrence of *Pseudomonas aeruginosa* suggested that it has minimal growth requirements and has ability to produce a large number of extracellular protective and toxic substances (Whitby and Rampling, 1972). It can survive and replicate within the hospital environment, where it colonizes sinks, hospital distilled water systems (Zimakoff *et al*., 1983), mattresses (Fujita *et al*., 1981), hand wash basins, humidifiers, floor mops, plastic washing bowels, soap dishes, nail brushes, bedrails (Lowbury *et al*., 1970) and even disinfectants (Burden and Whitby, 1967).

The cardiac hospitals indoor and outdoor had 12.69% bacterial pathogens. The patients in the cardiac hospital were usually immunocompromised. Frequent visit of doctors and nurses, visitors and relatives add to the contamination of airborne pathogens. According to Cairns *et al*. (2000) most airborne microorganisms found in hospitals are generated within the building by the staff, patients and visitors. Respiratory droplets produced by patients coughing or sneezing can impact upon the conjunctivae or oro-nasal mucosae of susceptible patients and healthcare personnel resulting in subsequent infection. If these healthcare personnel go to treat patients, the organisms on their uniforms might be expelled into the air in the form of cloth dust (Boyce *et al*., 1997).

The orthopedic hospitals showed 6.71% of airborne pathogens contamination and reported the presence of *Staphylococcus aureus* (1.88%), *Staphylococcus epidermidis* (0.83%) and *Micrococcus luteus* (1.57%) (Table 1). The microbiological studies confirmed that gram-positive bacteria such as *Staphylococcus aureus* and *Staphylococcus epidermidis* are the primary pathogens responsible for wound infection in prosthetic joint surgery (Fig. 3).

All the examined air flora of the hospitals was contaminated with airborne pathogens. The most dominant pathogens within all examined hospitals were *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Table 1: Percent bacterial aerial flora of various hospitals in Amravati

Types of Hospitals	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	Total % isolates
Clinic	0.52	0	0.41	0.52	0.10	0.83	0	0	0.10	0	0.10	0	0	0	0	2.62
Cancer hospital	0.62	0	0	0.10	0.10	0.20	0.10	0	0.10	0	0	0	0	0	0	1.15
Cardiac hospital	4.82	0.73	0.94	2.09	0.20	1.88	0	0	1.46	0	0	0.10	0	0.41	0	12.69
Dental hospital	0.52	0	0.20	0.20	0	0	0	0	0	0	0	0	0	0	0	0.94
ENT hospital	0.31	0	0.52	0.10	0	0.83	0	0	0.10	0	0	0	0	0	0	1.88
General hospital	4.61	0.52	3.67	2.51	0.31	3.88	0	0	0.83	0	0.52	0.10	0.10	0.31	0.20	17.62
Maternity and Children hospital	15.32	1.67	8.28	8.28	0.83	9.86	0	0.10	3.46	0.10	1.57	0.20	0.20	0.62	0.10	50.68
Mental hospital	0.10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.10
Multispeciality hospital	0.83	0.10	0.94	0.62	0.20	1.46	0	0	0.10	0	0.31	0	0	0.73	0.20	5.56
Orthopedic	1.88	0.62	0.83	1.57	0.10	0.73	0	0	0.20	0	0.10	0.10	0	0.52	0	6.71
Total bacterial Pathogens	282	35	151	153	17	188	1	1	61	1	25	5	3	25	5	953

1. *Staphylococcus aureus*, 2. *Staphylococcus saprophyticus*, 3. *Staphylococcus epidermidis*, 4. *Micrococcus luteus*, 5. *Micrococcus roseus*, 6. *Pseudomonas aeruginosa*, 7. *Pseudomonas fluorescence*, 8. *Proteus vulgaris*, 9. *Proteus mirabilis*, 10. *Morganella morganii*, 11. *Escherichia coli*, 12. *Citrobacter freundii*, 13. *Serratia marcescens*, 14. *Enterobacter aerogenes*, 15. *Klebsiella pneumoniae*

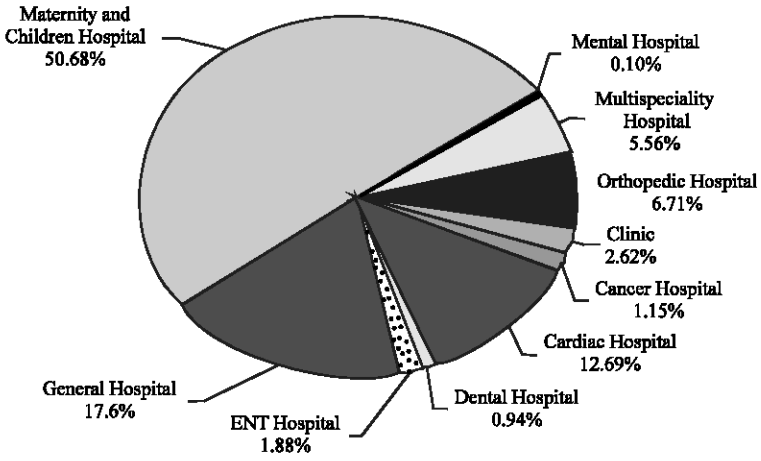

Fig. 3: Total pathogenic bacterial air flora isolated from various hospitals in Amravati

Out of all of the hospitals examined, maternity and children hospitals showed highest bacterial contamination, which may be due to unhygienic state of children patient, parents and more crowds as well as nearby and open defecation of children.

The observations strongly recommend periodical recording of such data to keep the sudden outbreak of airborne infections in hospital patients at minimum. For the pathogens that can spread through the air, there must be proper ventilation and exhaust fans in the hospital wards. Spitting and gargling etc. should be at proper place and not at anywhere which may spread infection in the air. Infectious person should try to avoid sneezing, coughing; talking in the open air or in crowded area and the handkerchief should be used. The bed sheets of the earlier patients should not be reused for the next patients. In OPD, crowd should be avoided or minimised. The signboards should be used indicating the use of napkins etc. and coughing, sneezing and talking by open patients. Maternity and children hospitals should be hygienically clean so that there should be proper disposal of the children pads. During entrance in the hospital, shoes should be kept out side so that the dust cannot enter inside. Highly infectious diseased patients should be hospitalized in quarantine or in isolation. The floor of the wards and hospitals should be swabbed with disinfectants daily. Visitors and relative's visits to the patients should be as low as possible. Limit the movement

and transport of the patient from the room to essential purposes only. If transport or movement is necessary, minimize patient dispersal of droplet nuclei by placing a surgical mask on the patient, if possible. Strategies should be such that prevention is better than cure.

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