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Prevalence of *Staphylococcus aureus* and *Pseudomonas aeruginosa* in indoor air flora of a district hospital, Mandya, Karnataka

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Abstract: *Two year (1998 and 1999) extensive survey was carried out in Mandya district hospital, Karnataka for a period of three years to monitor the prevalence of Staphylococcus aureus, Pseudomonas aeruginosa (S. aureus and Ps. aeruginosa) from the indoor air samples of operation theater, labor room, children's ward, male and female general wards postoperative wards etc. A rotary air sampler loaded with manital salt and Pseudomonas selection agar media strips were used to collect the samples from various sites. The survivallence results revealed the prevalence of S. aureus and Ps. aeruginosa in almost all sampling sites, irrespective of season indicating their long term survival and consequent threat to hospitalized patients as well the working employees.*

Key words: *Nosocomial infections, Aerosol, Colony forming unit (CFU), Transient flora*

Introduction

The microbial flora of hospital air is variable and transient. In hospitals coughing and sneezing from human respiratory tract besides surface of the skin spread air microorganisms. The larger particles of dust settle within a few minutes on to exposed horizontal surfaces such as floor, furnitures and equipments. Tiny particles may be inhaled into the respiratory tract or settle on to wounds. Although outbreak of air borne nosocomial infections have been uncommon, air borne transmission appears to account for about 10% of all nosocomial infections. The present study was therefore undertaken to assess the prevalence of *Staphylococcus aurens* and *Pseudomonas aeruginosa* in the indoor air of hospital environment.

Materials and Methods

The present study was carried out at the General hospital in Mandya situated on Bangalore Mysore State highway. This campus includes a T.B. center, Cancer center, Isolation ward (IW), Childrens ward (CW), Male postoperative ward (POW male), Female postoperative ward (POW female), Burns ward, Operation theatre (OT), Labor ward (LW) Post partem centre (PPC) *etc.* The sampling was done in all the wards (except T.B. ward) to assess the prevalence of *S. aureus* and *Ps. aeruginosa*.

A handhold Reuter sampler loaded with Manital salt agar/ *Staphylococcus* selection agar, *Pseudomonas* selection agar supplied by M/s Hi-Media, Mumbai, was used to collect samples in all the wards of the hospital as well as control site located 100 m away from the hospital premises. The collected samples were transported to laboratory in ice boxes within 2 hr of collection. After 24 hr of incubation total number of colonies on each strip were counted. Depending on colony morphology, organisms were subcultured on to supportive media using standard procedures. (Bailey and Scott, 1990) *S. aureus* was confirmed by Staph Latex Slide Agglutination test.

***Ps. aeruginosa*:** For the identification of *Ps. aeruginosa* a battery of biochemical tests starting with gram staining, oxidase test, catalase test, growth at 42°C, pigment production on king A and B medium, hydrolysis of arginine, tween80, tyrosine, utilization of carbohydrates (lactose, maltose, arabinose) by oxidative fermentative test were performed. All the above tests were carried out according to the procedures outlined by Cowan and Steel (1974), Bergeys (1984), Bailey and Scot (1990) and Maclado and Mcartneny (1990).

Total density of pathogens as CFU/m^3 was estimated using the formula.

$$CFU/m^3 = \frac{\text{Total number of colonies on agar strips}}{\text{Time of exposure in minutes}} \times 25$$

Bacterial density of different wards was monitored between 1997 to 1999 as shown below.

I samplings - Sept 1997	V sampling - Sept 1998
II sampling - Dec1997	VI sampling - Dec 1998
III Sampling - March 1998	VII sampling - March 1999
IV Sampling - June 1998	VIII Sampling - June 1999

Results and Discussion

The indoor air quality with respect to *S. aureus* and *Ps. aeruginosa* is shown in Fig. 1 and 2. The air quality at OT is expected to be contamination free and that empty operation theater should contain less than 35 Colony Forming Unit (CFU) of bacteria/m^3 and less than one CFU/m^3 of *Clostridium* or *S.*

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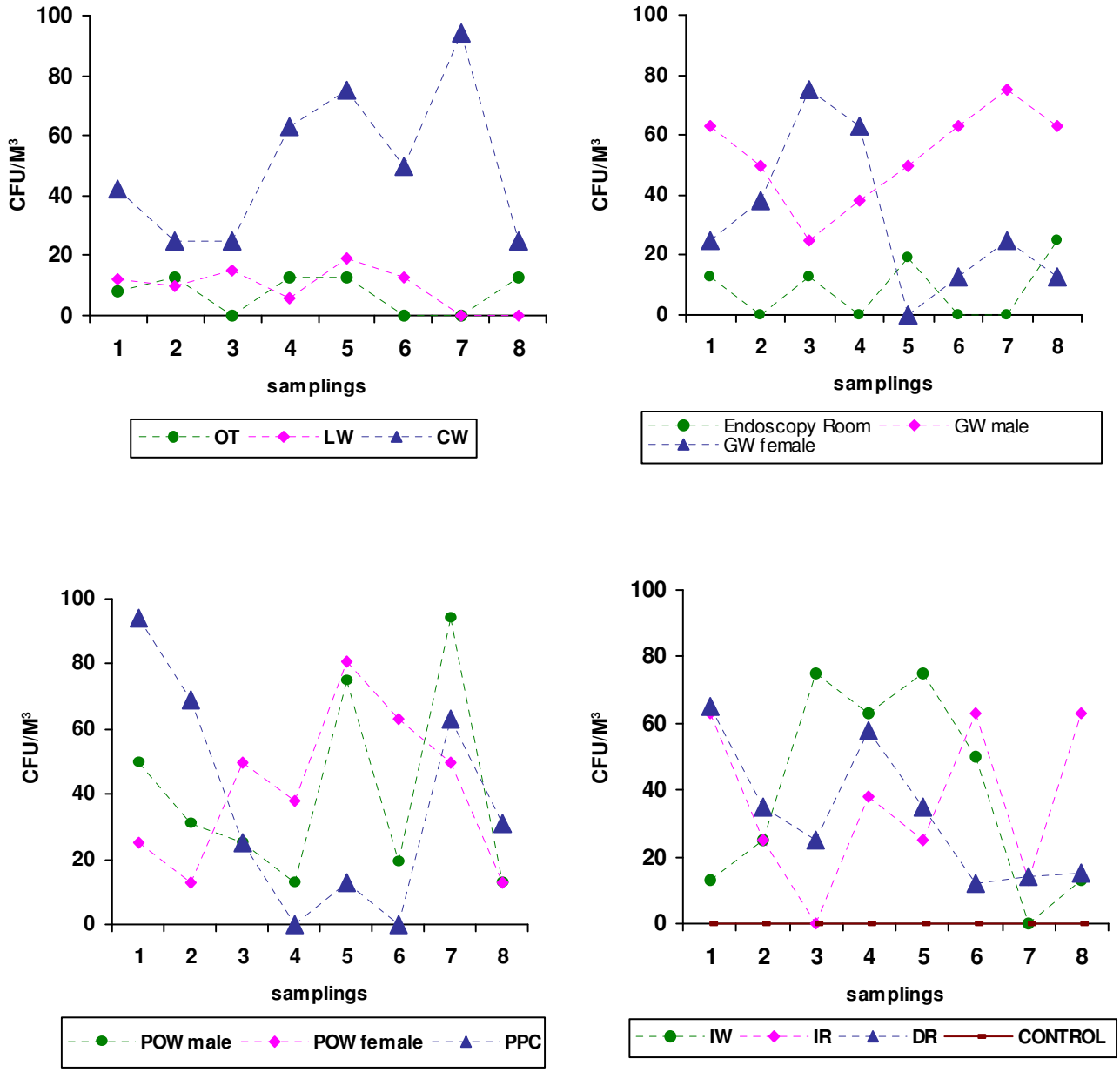

Fig. 1: Prevalence of *S. aureus* in hospital air

aureus in 30 cm^3 (Holten *et al.,* 1990). The samples from operation theater showed an highest 13 CFU/m^3 of *S. aureus* in most of the samplings. Whereas labor ward (LW) harbored an highest of 75 CFU/m^3 in 3rd sampling while children's ward (CW) showed 95 CFU/m^3 in 7th sampling (Fig. 1).

The endoscopy room air contained 25 CFU/ m^3 of *S. aureus* in 7th sampling (Fig. 1) and 50 CFU/m^3 of *Ps. aeruginosa* in 3rd and 4th samplings, that might be transient flora of professionalists and patients of the hospital (Fig. 2).

In the general ward for male and female where patients with all kinds of diseases are housed, the CFU/m^3 count was 63, 75 and 50 CFU/m^3 of *S. aureus* during 1st, 4th, 5th, 6th, 7th and 8th sampling (Fig. 1). The *Ps. aeruginosa* density was 38 CFU/m^3 in 3rd sampling (Fig. 2).

In post operative ward for male and female, where the patients admitted before and after performing the operation are housed the air flora contained an highest of 94,75 and 50 CFU of *Ps. aeruginosa* in 1st, 5th and 6th samplings in male POW,

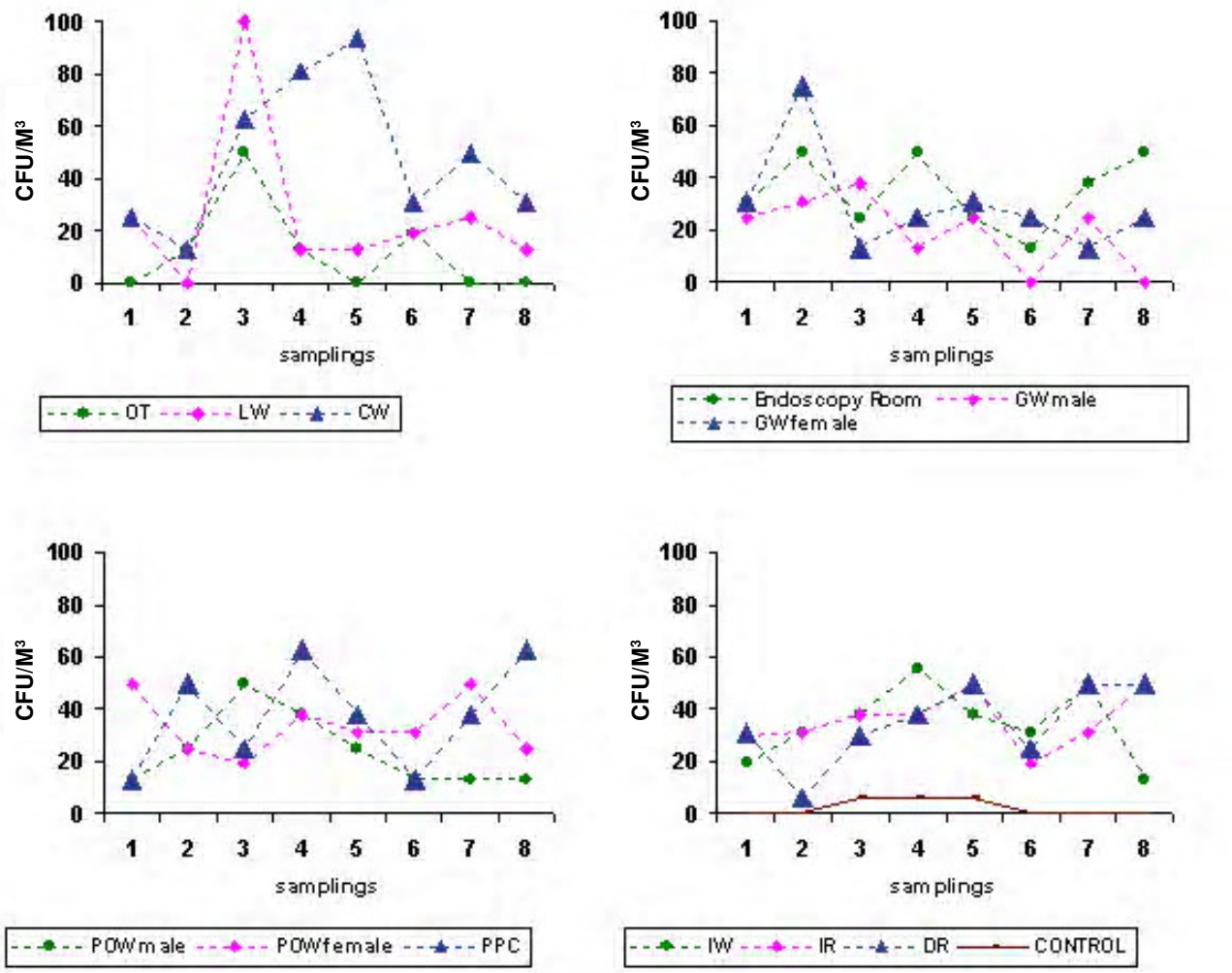

Fig. 2 Prevalence of *Pseudomonas aeruginosa* in hospital air

while female POW showed 81, 63 and 50 CFU of *Ps. aeruginosa* during the 5th, 6th and 8th samplings respectively. *Ps. aeruginosa* showed an highest of 50 CFU/m³ in1st, 3rd and 7th samplings while 13 CFU/m³ was observed during 7th and 8th sampling in female POW indicating the transient flora of the hospital.

The PPC (post partem centre) and IW (isolation ward) room, with lower number of patients the condition was pathetic. The bacterial quality of these rooms showed higher incidence of nosocomial pathogens in almost all the samplings irrespective of seasons (Fig. 1, 2). The injection room (IR) and dressing room (DR) always crowded with patients showed 63 CFU/m³ of *S. aureus* in 4th, 6th and 8thsampling and *Ps. aeruginosa* 1st, 6th and 8th sampling. This could be due to the shedding of pathogens by infected patients through cough, sneezing *etc.*

The control sample located 100 m away from the hospital (Bangalore - Mysore highway) was almost devoid of *Ps. aeruginosa,* However, an average 6 CFU of *Ps. aeruginosa* were recorded during the 3rd, 4th and 5th sampling (Fig. 2).

Air pollution poses serious threat to human health. Indoor air pollution in general and occupational exposure in particular contributes substantially to human acquaintance to various microbes at concentrations often higher than outdoor. An average person inhales 25 m³ of air/day and indoor environment often carries higher level of air pollutants and bio aerosols than their surroundings depending on the location, type of fuel used, atmospheric humidity *etc.* The hospital indoor air flora in particular arises from patients, medical personnels and visitors via natural processes (Garnier *et al.*, 1996).

Larger aerosol droplets discharged from a patients nasal and throat secretion settle out rapidly on various surfaces and get dried up. Disturbances caused to these dried materials during bedding, handling handkerchiefs *etc.*, sweeping floor in the ward account for the enty of microorganisms to the circulating air. The survival of these microorganisms for relatively longer period by adhering to dust creates significant hazard. Streptococci have been found in the floor dust near patient or carriers. Pandit *et al.* (1993) reported surveillance of *S. aureus* and *Ps. aerurinosa* in

burns wound and blood of burns patients. It is apparent from our study that hospital air harbors bio aerosols. The results obtained also demonstrate that hospital air flora is transient. The composition of operation theatre air with organisms such as *S. aureus and Ps. aeruginosa* (Fig. 1, 2) confirms the contamination of microorganisms contributed from human activities. The OT is sterilized once a day and subsequent operations are carried out without fumigation. Attenders and professionals moving from OT to corridor with sterilized operation dresses enhance chances of contamination through their exposure to outdoor air. Ayeliffe *et al*. (1969) suggested that operation theatre should have a few zones *viz*., aseptic, clean, less clean and should be equipped with mechanical ventilation, however in the present case no such designated zones existed.

Studies on a number of operation theatres have suggested that there is a general relation between total air count and risk of infection. Counts in the range of 700-1800 per m^3 are related to significant risk of infection and even counts as low as, 180/m^3 have been categorized as slightly risk (Parker, 1978). In the present study, air flora of OT highlight significant risk of infection among the patients undergoing surgery.

Air borne particles are the main sources of contamination of the surgical wounds, burns and respiratory disorders. The air borne pathogens survive better under the conditions of high humidity (Marthi *et al.,* 1991). Rosas *et al. (*1996) studied *E. coli* in settled dust and air samples of residential environment of Mexico city. The air borne gram negative bacteria from indoor represented 2.7% of cultivable air borne bacteria while that from outdoor represented 1.2%. In our study the indoor air harbored S. *aureus* with 94 CFU /m^3 and absence of *S. aureus* in out door flora indicates contamination arising from within hospital environment.

Chandrashekar *et al.* (1997) studied air flora of KC general hospital and Ambedkar general hospital, Bangalore. Zembrzuska (1995), isolated *Ps. aeruginosa* from hospital pharmacy and different wards and reported that airborne microbes settle on the surface of the room. Biradi and Leoni (1993), studied hospital indoor air and compared it with the other indoor air samples. *Ps. aeruginosa* counts were more in the hospital wards and surgeries as compared to office and laboratories. The microbial contamination in the present case did not correlate with air conditioning system probably because of high turn over in the hospital population, the number of patients/ people and their attitude.

The crowded hospital ward air confirmed the prevalence of *S. aureus* and *Ps. aeruginosa* in the present case. The outdoor air (control) however was devoid of *S. aureus* and *Ps. aeruginosa.* The CFU density was also lower then the indoor air flora of the hospital. Existence of pathogens in the hospital wards suggests need for constant monitoring *via* epidemiological investigations to evaluate endemic infections, which appear to be apparent during hospital stay. There is also need to undertake corrective measures to eliminate potential pathogens.

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