

## A Comparative Study of Two Different Methods of Microbiological Surveillance of Operation Theatres

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**Abstract:** Prevalence of air borne Nosocomial infection is a major attribute to morbidity and mortality in immunocompromised patients. Two methods are commonly used for measurement of bacterial density in hospital environment particularly Operation theater. Air samples from conventionally ventilated operating rooms and intensive care units were taken simultaneously by the sedimentation method (settle plate) and with the HiAir petri air sampler. Bacterial density was calculated in form of colony forming unit (cfu). The isolated bacteria were identified by conventional methods. The investigations showed that the number of colony forming units per cubic metre obtained with the sedimentation method was, on an average, higher than those found with the HiAir petri air sampler from the same area. The most common isolate from settle plate method was *Bacillus subtilis* (54.91%) followed by *Pseudomonas aeruginosa* (38.09 %), Coagulase negative staphylococcus (30.12%), *Klebsiella pneumoniae* (25.85%), *Acinetobacter baumannii* (23.12 %), *Escherichia coli* (10.22 %), *Staphylococcus aureus* (8.32%) and *Proteus mirabilis* (2.72%). Settle plate method was found to be more informative than the air sampling system for bacteriological analysis of air in present study.

**Keywords:** Air sampling; settle plate method, Air sampler.

### INTRODUCTION

Nosocomial infections transmitted by the airborne route are a major source of morbidity and mortality in immunocompromised patients [1]. The prevalence of wound infection by air borne pathogens is more common in two special settings, the operating room and the wards for burned patients.

A linear relationship between air counts of bacteria in operating rooms and surgical site infection or wound contamination rate has been reported by many investigators [2-6]. A 13-fold reduction in bacterial density in the operating room would reduce the wound contamination by about 50%. The microbiological quality of operation theatre air is one of the significant parameter affecting incidence of surgical wound infection [8]. Bacteriological analysis of air by air sampling in hospital environment can be helpful in prevention and control of Nosocomial infections. In an outbreak of airborne Nosocomial infection, it provides good information which can be helpful for formulation of control measures. Although routine air sampling is not indicated, it would be appropriate to monitor hospital air quality in critical areas, such as operating rooms and intensive care units. The aim of the present study was to compare two different methods of microbiological surveillance of hospital air and to assess the utility of the HiAir petri air sampler (HIMEDIA) for monitoring air contamination in the hospital environment.

### MATERIALS & METHODS

Air samples were collected from eight different conventionally ventilated operating rooms and three different intensive care units. Air samples from each of the investigated rooms were taken once in a month over a period of a year; & following two methods were used simultaneously for air sampling.

- The HiAir petri air sampler loaded with blood agar plate. Sampling time: 200 seconds (Air sample volume: 1000 litres).
- The sedimentation method with air exposed plate kept for 1 hour, 1 meter above the floor & 1 meter from the wall [9].

All of the air samples were obtained from an empty Theatre. The plates were transported to the laboratory and incubated at 37 °C for 2 days. After incubation the colonies were counted. The identification of isolated strains was performed by standard staining methods and biochemical reactions. With HiAir petri air sampler method, concentrations of airborne bacteria are calculated as colony forming units

per cubic meter (CFU/m<sup>3</sup>). It is counted according to the following equation:

$$\text{Bacterial load (B)} = 1000N/RT [10]$$

N number of colonies on the plate;  
R rate of air sampling in liters/min  
T - duration in minutes

For settle plate method formula used is number of bacteria settling on 1 square meter of medium per minute = number of such particles per 0.3 cubic meter of air [10]

## RESULTS

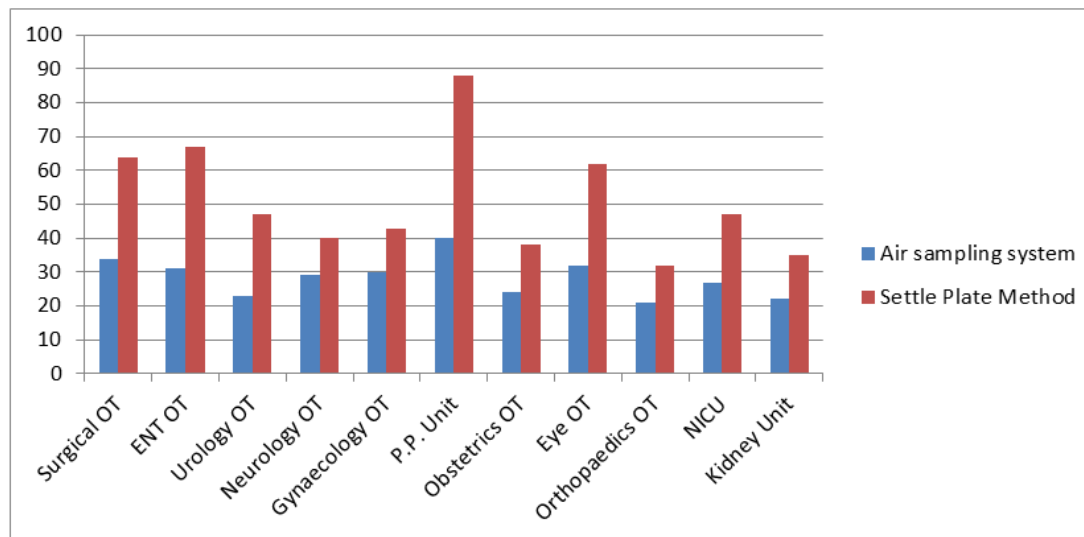
The sedimentation method revealed magnitude of air contamination was varied from Minimum 32 CFU/m<sup>3</sup> in Orthopedic Operation Theater & up to 88 CFU / m<sup>3</sup> in Post partum unit (Graph-1); & magnitude of air contamination detected by Hi air petri air sampler method was as low as 21 CFU / m<sup>3</sup> in Orthopedic OT to 40 CFU/m<sup>3</sup> in Post partum unit (Graph-2).

The air samples collected from eight operation theatres and three different intensive care units at interval of one month from the period of July 2016 to June 2017 for bacteriological analysis. Table-1 shows comparison between Numbers of Colony forming units obtained by air sampling system Vs settles plate

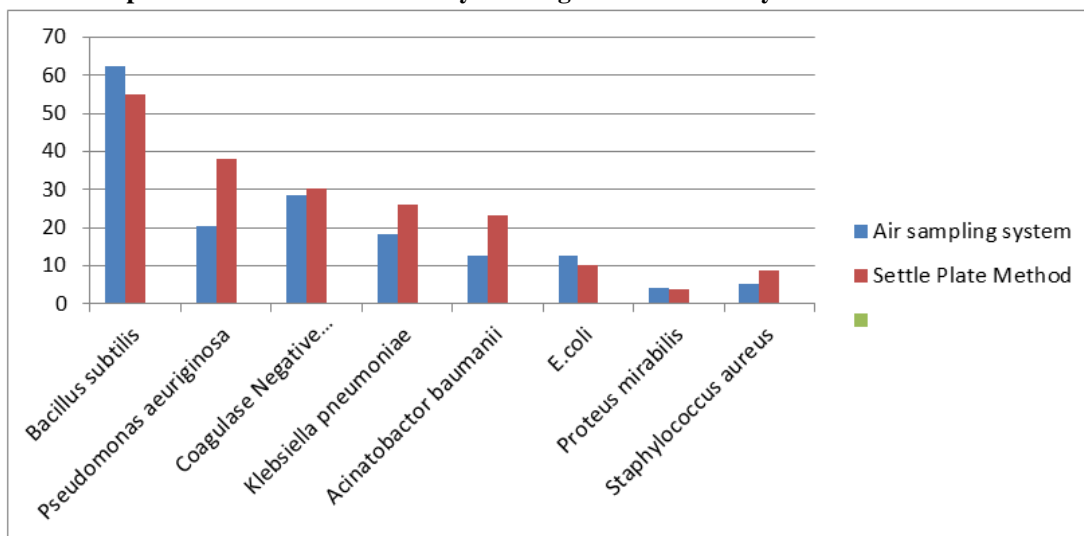
method. On applying independent sample t test (unpaired student t test two tailed) it was found that at almost all locations, the p value was < 0.0001 means statistically highly significant difference was there between two different methods. As it is obvious from the data that settle plate method has high yield, it was found statistically proven better method than air sampling method.

The most common micro-organism isolated from settle plate method was *Bacillus subtilis* (54.91 %) which is considered as a common environmental contaminants. Amongst the pathogenic bacteria, the most common isolate was *Pseudomonas aeruginosa* (38.09 %) followed by *Coagulase negative staphylococcus* (30.12%), *Klebsiella pneumoniae* (25.85%), *Acinetobacter baumannii* (23.12 %), *Escherichia coli* (10.22 %), *Staphylococcus aureus* (8.32%) and *Proteus mirabilis* (2.72%).

The most common micro-organism isolated from Hi air petri air sampler was *Bacillus subtilis* (62.29 %). Other isolates were *Coagulase negative staphylococcus* (28.34 %), *Pseudomonas aeruginosa* (20.23 %), *Klebsiella pneumoniae* (18.12%), *Acinetobacter baumannii* (12.56 %), *Escherichia coli* (12.6 %), *Staphylococcus aureus* (4.9%) and *Proteus mirabilis* (4.09%).



Graph-1: Mean number of Colony forming units obtained by two different methods



Graph-2: Percentage of isolates obtained by two different methods

**Table-1: Comparison between Numbers of Colony forming units obtained by air sampling system Vs settles plate method**

Name	Method	July 16	Aug 16	Sep 16	Oct 16	Nov 16	Dec 16	Jan 17	Feb 17	Mar 17	Apr 17	May 17	June 17	Mean Value	t-test	P value
Surgical OT	air sampling system	30	29	37	33	39	28	44	48	30	32	25	31	34	8.177	<0.0001
	settles plate method	45	58	67	54	51	60	70	74	65	83	72	68	64		
ENT OT	air sampling system	28	30	33	29	35	37	39	35	26	22	32	29	31	8.41	<0.0001
	settles plate method	50	55	53	49	69	64	58	73	18	89	80	82	67		
Urology OT	air sampling system	21	28	30	28	19	17	25	22	23	23	20	17	23	9.882	<0.0001
	settles plate method	63	48	42	51	44	45	50	39	58	37	44	47	47		
Neurology OT	air sampling system	29	28	28	36	30	31	29	33	26	26	21	28	29	-4.3999	<0.0001
	settles plate method	32	35	39	35	48	54	32	50	30	37	46	40	40		
Gynecology OT	air sampling system	27	35	30	27	30	37	32	28	29	30	24	29	30	-5.209	<0.0001
	settles plate method	40	28	51	55	34	39	45	53	42	42	48	38	43		
P.P Unit	air sampling system	30	56	50	44	35	34	38	35	33	40	46	38	40	-17.67	<0.0001
	settles plate method	92	85	78	83	90	89	96	81	87	93	85	90	88		
Obstetrics OT	air sampling system	18	16	28	20	26	20	30	17	32	22	35	24	24	-4.837	<0.0001
	settles plate method	26	39	35	48	47	50	32	38	30	29	42	40	38		
Eye OT	air sampling system	29	53	56	33	30	21	25	28	30	31	22	29	32	-7.626	<0.0001
	settles plate method	56	59	68	60	63	46	70	68	65	57	55	72	62		
Orthopedics OT	air sampling system	10	25	28	25	21	14	18	31	21	23	15	19	21	-5.134	<0.0001

	settles plate method	35	39	31	26	25	29	30	28	33	38	37	37	32		
NICU	air sampling system	28	25	21	40	19	32	27	23	25	38	25	21	27	-7.188	<0.0001
	settles plate method	53	48	44	38	57	60	41	37	45	45	48	48	47		
Kidney Unit	air sampling system	15	18	16	25	31	16	20	22	29	27	17	21	22	-5.504	<0.0001
	settles plate method	34	35	38	23	27	35	40	44	37	32	28	41	35		

## DISCUSSION

The quality of indoor air in terms of bacterial density and bacteriological profile depends on external and internal sources, such as ventilation, cleaning procedures, the surgical team and their movements in and out of Operating room. So, bacteriological profile reflects that which control measures are lacking in the present area.

The investigations show that the CFU/m<sup>3</sup> values obtained with the sedimentation method were on average higher than those measured with the HiAir petri air sampler. Whyte *et al.*, [2] also suggested that settle plates showing bacterial surface contamination represents a more relevant indicator of the wound contamination rate than air counts. There are some advantages of settle plate method like it is cheap, easily available, multiple samples can be taken at the same time and it does not disturb air flow. Settle plates are sterile, economical and readily available. Many places in an environment can be checked at the same time. The natural trend of the microbial population in the air is not disturbed during the sampling time nor is the laminar air flow interrupted in any way.

Settle plates give the measurement of the harmful part of the airborne population which falls on to a critical surface in a given time. Settle plates allow the evaluation of surface contamination settling from the air [4].

## CONCLUSION

The bacteriological quality of air in the Operation Theater and intensive care units reflects the effectiveness of infection control measures in the hospital. The settle plate method was found statistically proven better method than air sampling method for bacteriological analysis of air in these areas.

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