

# Intensive Care Units and Operating Rooms Bacterial Load and Antibiotic Susceptibility Pattern

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**Abstract:** Introduction. The indoor air environment can potentially place patients at a greater risk because enclosed spaces can confine aerosols and allow them to build up to infectious levels as a result, this study intended to determine the bacterial load and antibiotic susceptibility pattern in operating theater (OR) and intensive care unit (ICU) at Hawassa University Referral Hospital. Methods. A cross sectional study was conducted to measure indoor air microbial quality from Nov 2014 to February 2015 on 120 air samples collected from selected sites in 15 rounds using purposive sampling technique by Settle Plate Method (Passive Air Sampling following 1/1/1 Schedule). Sample processing and antimicrobial susceptibility testing was done using standard microbiological methods. The data was analyzed using SPSS version 16.0 and was inferred based on baseline values recommended by Fisher. Result. The mean bacterial load of ICU 454.2 CFU/dm<sup>2</sup> was recorded. Likewise, at OR during active, 87.27 CFU/dm<sup>2</sup> and 13.12/CFU/dm<sup>2</sup> during passive were recorded. Compared to the standard set by Fisher, the ICU and OR while at passive were higher than the acceptable limit. Among the isolated six bacteria, *S. aureus* 36 (30%) was the predominant species in both OR and ICU were as *P. auriginosa* 16 (26.7%) was the second prevailing isolates at ICU. *S. aureus* was highly resistant to penicillin, tetracycline 86.1%, 72.2% respectively were as *P. auriginosa* showed low level resistance to Ciprofloxacin (22.2%), Cotrimoxazole (27.7%) and Ceftriaxon (16.7%). Conclusion. This finding indicates that resistant isolates for the commonly used drugs and high bacterial load of indoor air judges the risk factor for SSI as well more risking ICU patients. Hence adequate attention should be given to maintenance of proper hygiene in the ICU and OR environments since it is well known those patients are highly susceptible to microbial infection.

**Keywords:** Indoor Air, Intensive Care Unit, OR, Southern Ethiopia

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## 1. Introduction

Microbial contamination of indoor hospital, especially in an operating theatre and other specialized units had continued to increase prevalence of nosocomial infections. With resultant effect of high morbidity and mortality rate among patient admitted for post-operative surgery, patients [1, 2].

Enclosed air environment can potentially place patients at a greater risk because enclosed spaces can confine aerosols and allow them to build up to infectious levels [3]. Therefore Infection prevention and patient safety should be at the great concern of hospital management [4]. Patients admitted to intensive care units, in particular, often become colonized

with resistant organisms and may serve as the focus for hospital-wide bacterial resistance [5]. The high prevalence of resistance in intensive care units (ICUs) has been attributed to the severity of illness of the patients, prolonged hospital stays, and the widespread use of invasive devices and broad spectrum antibiotics. The rate of hospital acquired infection in critically ill patients is about 20%, depending on the type of admission diagnosis and underlying conditions predisposing to microbial colonization and infection [6].

Microorganisms were transmitted to the patients by the contaminated hands of healthcare workers. OR and ICU are the places that need the maximum cleanliness standards, also the same necessities for the health workers [4]. The significance of the estimation of the indoor bacterial load can

be used as an index for the cleanliness of the environment as well as an index they bear in relation to human health and as source of hospital-acquired infections.

## 2. Materials and Methods

### 2.1. Study Area

The study was conducted at Hawassa Referral Hospital, Hawassa, Southern Ethiopia. Hawassa, the capital of SNNPR and it is located 275 km from the capital city of Ethiopia, Addis Ababa. The total population of Hawassa town is 130,579 with one to one male to female ratio. Hawassa Referral Hospital was inaugurated in November, 2005; it has 850 beds and serves about 12 million people. The hospital has about 300 daily outpatient visits. It has one OR with 4 separate space and one ICU with a total of 16 beds.

### 2.2. Study Design and Sampling

A descriptive cross-sectional study was conducted between Nov 2014 and February 2015 in Hawassa Referral Hospital. The air samples collected twice per day in two representative section in each site during active and passive in OR and at morning and afternoon in ICU over a period of 15 weeks. Air samples were collected using Settle Plate or Passive Air Sampling method following 1/1/1 schedule (a nine cm in diameter sterile Petri dish with 5% Sheep's blood agar was left open to the air for an hour, a meter above the floor and a meter from the wall) [7]. During air sampling sterile gloves, mouth masks and protective gown was worn to prevent self contamination of the 5% Sheep's blood agar plate (Oxoid, UK).

The air samples in ORs were collected during both active and passive whereas samples from ICU was collected in the morning and afternoon in each round per week. Then, blood agar plates was transported to the microbiology laboratory and incubated aerobically for 24 hours at 37°C. The total number of colony forming units (CFU) was enumerated

using colony counter and results were expressed in CFU/dm<sup>2</sup> [8]. The colonies were assessed further using the biochemical tests following standard bacteriological techniques to isolate species [9].

The antimicrobial susceptibility testing was done on Mueller-Hinton agar (Oxoid, UK) for every potential pathogenic bacteria isolates with 12 antibiotics each by Kirby-Bauer disk diffusion method matching the test organism to 0.5 McFarland turbidity standards. Then, the susceptibility result was interpreted according to the principles established by Clinical and Laboratory Standards Institute (CLSI) by measuring the zone diameter of inhibition [10].

Reference strains *S. aureus* (ATCC 25923); *E. coli* (ATCC 25922) and *P. aeruginosa* (ATCC 27853) used as a quality control for culture and susceptibility testing throughout the study.

All data was analyzed using Statistical package for Social Sciences (SPSS) version 16.0 and Microsoft Office Excel. Results were interpreted according to baseline values suggested by Fisher [7, 11] and values of  $p < 0.05$  was considered as statistically significant. The official permission and approval was obtained from the Department of Medical Laboratory Sciences, Hawassa University.

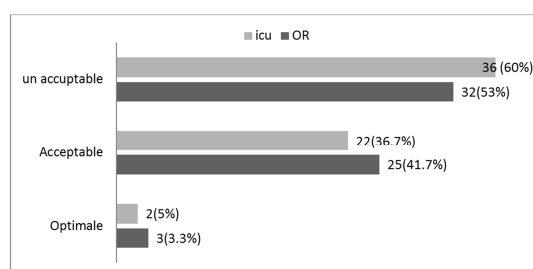
## 3. Results

A total of 120 air samples were collected, twice per day in two representative sections in each site during active and passive in OR also at morning and afternoon in ICU over a period of 15 weeks. The finding showed that ICU had the highest bacterial counts 654 CFU/dm<sup>2</sup> and the OR had the lowest bacterial count 6 CFU/dm<sup>2</sup> during active period. The mean bacterial load of OR during active, 87.27 CFU/dm<sup>2</sup> and during passive 13.12/CFU/dm<sup>2</sup> were recorded. According to Fisher's index of microbial air contamination [7], air microbial count of OR at rest and in activity must not exceed 9.0 and 91.0 CFU/dm<sup>2</sup> respectively (Table 1).

**Table 1.** Mean bacterial counts of air samples against the standard in HURH.

SITE		Aerobic CFU/dm <sup>2</sup> ; No. (mean value)	Standard (CFU/dm <sup>2</sup> ) [5]*		
			OPTIMAL	ACCEPTABLE	UNACCEPTABLE
OR	Active	30 (87.27)	0-60	61-90	>91
	passive	30 (13.12)	0-4	5-8	>9
	Morning	30 (474.26)			
ICU	Afternoon	30 (434.2)			
	Total	60 (454.23)	0-250	251-450	>450

\*According to Fisher's index of microbial air contamination



**Fig. 1.** Proportion of indoor air samples in ICU and OR based on the standard.

Mean aerobic colony count in ICU during morning time of sample collection was 474.3 CFU/dm<sup>2</sup> where as in afternoon 434.2 CFU/dm<sup>2</sup>. The overall mean colony counts in ICU 454.2 CFU/dm<sup>2</sup> was slightly higher than the sated acceptable limit as shown in the Table-1. On the other hand among the collected samples 60% from OR and 53% of ICU were beyond the tolerable limit compared to the standard (Fig. 1).

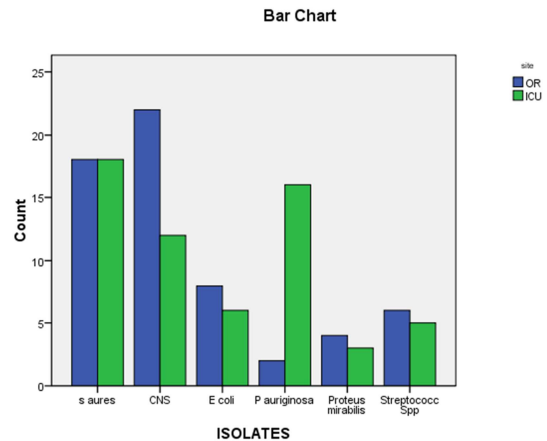
In regard to timing 45% of the afternoon samples result compared to 68.3% of the morning samples were unacceptable which was sign ificantly higher in morning,  $p=0.016$  (Table 3).

**Table 2.** Time variation of samples against the standard, HURH.

	Number of sample N=60(%)			p-value
	morning	afternoon	Total%	
OPTIMAL	3[5]	2[3.3]	5[4.2]	P=0.016
ACCEPTABLE	16[26.7]	31[51.7]	47[39.2]	
UNACCEPTABLE	41[68.3]	27[45]	68[56.7]	
Total	60[100]	60[100]	120[100]	

Six bacterial isolates were identified. Among these *S. aureus* 36 (30%) and *CNS* 34 (28.3%) were the predominant species. Isolates in respect to site, *CNS* recorded 22 (36.7%) in OR and 12(20%) in ICU. On the other hand the second prevailing isolates at ICU was *p. auriginosa* 16 (26.7%) (Figure 2).

The antimicrobial susceptibility patterns of isolates illustrates that *S. aureus* was highly resistant to penicillin and tetracycline at 86.1% and 72.2% respectively. The highest resistance was recorded against Penicillin (80.51%) followed by Tetracycline (67.23%) and Erythromycin (60.59%). However, they were sensitive to Vancomycin (91.7%) and Clindamycin (75%) (Table 4). On the other hand the least resistance drugs were Vancomycin (29.05%), Clindamycin (29.88%), Ciprofloxacin (30.71%) and Ceftriaxon (31.54%).

**Fig. 2.** Bacterial isolate of indoor air by site at HURH.**Table 3.** Antimicrobial resistance of bacterial isolates from indoor air of study site.

Antibiotics tested	Number of resistant Isolates (%)						Total (%) (n=120)
	<i>S. aureus</i> (n=36)	<i>CNS</i> (n=34)	<i>E. coli</i> (n=14)	<i>P. auriginosa</i> (n=18)	<i>Streptococcus spp</i> (n=11)	<i>Klebsiellasp</i> (n=7)	
Amoxy clavulinic	20[55.6]	12[35.3]	2[14.2]	5[27.7]	5[45.5]	1[14.3]	45[37.35]
Ampicillin	13[36]	14[41.2]	2[14.2]	4[22.2]	3[27.7]	2[28.6]	38[31.54]
Chloramphenicol	18[50]	13[38.2]	3[21.4]	5[27.7]	5[45.5]	2[28.6]	46[38.18]
Clindamycin	9[25]	14[41.2]	4[28.4]	5[27.7]	3[27.7]	1[14.3]	36[29.88]
Ciprofloxacin	16[44.4]	12[35.3]	2[14.2]	4[22.2]	2[18.2]	1[14.3]	37[30.71]
Erythromycin	33[47.2]	19[55.8]	5[35.7]	7[38.9]	7[63.6]	2[28.6]	73[60.59]
Cotrimoxazole	16[44.4]	14[41.2]	2[14.2]	5[27.7]	4[36.4]	2[28.6]	41[34.17]
Ceftriaxon	14[38.9]	15[44.1]	2[14.2]	3[16.7]	4[36.4]	0[0]	38[31.54]
Norfloxacin	16[44.4]	11[32.4]	2[14.2]	5[27.7]	3[27.7]	2[28.6]	39[32.37]
Tetracycline	26[72.2]	22[64.7]	7[50]	15[83.3]	8[72.2]	3[42.8]	81[67.23]
Penicillin	31[86.1]	27[79.5]	10[71.4]	16[88.9]	8[72.2]	5[71.4]	97[80.51]
Vancomycin	3[8.3]	1[3]	9[64.3]	16[88.9]	1[9]	5[71.4]	35[29.05]

## 4. Discussion

The microbial load in hospital indoor air is highly influenced by the number of factors beside with ventilation [4]. Ventilation causes dilution thus reducing the microbial load. So, indoor bacterial load analysis is very important to know the stat of ventilation and cleanness of critical areas within the hospital. As already Saied, microbiological quality of air considered as mirror of the hygienic condition of the ICU and operation theatres.

The mean colony count obtained from ICU 454.2 CFU/dm<sup>2</sup> was slightly higher than the acceptable range set by Fisher [11]. This finding goes in line with the study conducted in Gaza [12] which revealed that, the total bacterial count within indoor air in two ICUs ranged from (1170 to 1470) cfu/m<sup>3</sup> (standard is less than 50 cfu/m<sup>3</sup>) our finding was also

similar with Venezuelans finding [13] which reported, bacteria in surgical settings were within the "clean" range, in addition study from India [14] observe a significant increased percent of bacterial count in ICU. However, the intensive care unit depicted bacterial contamination. However this studies result was different compared to the findings of the study done in Czech Republic [15] which reported that ICU were in the acceptable rang.

The total concentrations of airborne bacteria in this study exceed the standard set by Fisher of 450 cfu/dm<sup>2</sup>. The finding shows that the hygienic level of the ICU under investigation is unacceptable. The possible explanation for the increased load of CFU counts might be the human trafficking in these room as HURH is a teaching hospital. Besides, the extent of cleaning and continues follow-up of sterility of critical areas of the hospital might contribute for the undesirable aerobic bacterial load observed in these rooms.

The mean colony count obtained OR in this study is 13.12 cfu/d<sup>2</sup> during active and 87.27 cfu/d<sup>2</sup> at rest time respectively (Table-1). In relation with the standard the OR finding during passive reflect a higher counts above standard. Comparable finding was reported in different area such as study in Northern Ethiopia [16] that reported mean colony count of 91.8, during activity and 17.2 cfu/dm<sup>2</sup> during rest, in addition with study in Jordan [17]. Our finding a beat differ to the findings of the study done in Jimma Ethiopia [18] which reported that ORs were not in the acceptable rang compared to standard set by Fisher both during activity and during passive time.

The highest bacterial populations recorded in the morning samples 38 (63.3%) compared to the 27 (45%) of the afternoon samples were beyond the tolerable limit. which was sign ificantly higher in morning,  $p=0.016$  (Table 2). This result was in line with the finding in Northern Ethiopia [16]. But, disagree with Nigeria [19] in which the highest bacterial population was recorded in the evening between time 5 pm and 6 pm compared to the morning also with Jimma South west Ethiopia finding [18] that reported the colony count sinall ORs except Sterilized cloth store were found to be higher in the afternoon than in the morning.

Six bacteria were isolated from a total of 120 air samples examined. Among these *S. aureus* 36 (30%) and *CNS* 34 (28.3%) were dominant. The finding mainly in OR was almost comparable with most of the studies else where, like the Venezuela [13], Nigeria, [20]; Jordan, [17], Also study conducted in Nigeria [19] where *S. aureus* was predominant. Which indicates the carrier state of health workers at ICU and OR.

On the other hand the second predominant isolates at ICU was *p. auriginosa* the finding is similar with studies conducted in Indonesia [21], Gaza [1], Czech Republic [15] and Taiwan [22] in which *p. auriginosa* were reported as a major isolate in ICU.

The high level of resistance of *S. aures* and *CNS* to penicillin and tetracycline found in our study has been widely reported throughout the world, (16, 18, 19 and 23) this might be due to emergence of penicillin resistant beta-lactamase producing strains. The most effective antimicrobials for gram positives was Vancomycin, with are sistance rate of 6.1%, were as Ceftriaxon (12.8% resistance) was best for gram negative isolates. There least resistance drugs were Vancomycin 35 (29.05%) and Clindamycin 36 (29.88%). The possible explanation for this might be these drugs not intensively used and not easily available outside the hospitals. In the other hand penicillin was the highest resistant drug with 97 (80.51%) of resistance. This study's finding was similar with others related study else where [16, 18, 21].

## 5. Conclusion

In general 60% of OR and 53% samples from ICU were under the unsatisfactory level of bacterial load. The finding imply that the high bacterial load of indoor air judges as a risk factor for SSI as well more risking ICU patients since

there is a linear relationship between bacteria load in air and consequent bacterial infection rate. The acceptable and optimal CFU, in ICU does not guarantee a clean and healthy environment for the reason that, the airborne microbial concentration and the types could be useful in determining the degree of cleanliness of an environment as well as an index they bear in relation to ICU patients health [24]. Hence adequate attention should be given to maintenance of proper hygiene in the ICU environments since it is well known those patients are highly susceptible to microbial infection. This would be greater extent assist to minimize the patient risk as well as for workers who usually spend worth time in the ICU. Mainly effective approach including restrictive entrance of outdoor aerosols, installing suitable filtration devices improving the hospital infection prevention and patient safety (IPPS) should be strenghen.

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## Authors' Contribution

Both authors (MH and EA) participated in proposal development, data collection, statistical analysis and manuscript write up.

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