e-ISSN: 2248-9126
Vol 6 | Issue 1| 2016 |27-30. Print ISSN: 2248-9118

Indian Journal of Pharmaceutical Science & Research

www.ijpsrjournal.com

MICROBIAL DIVERSITY IN AND AROUND NEW BUS STAND, THANJVUR DISTRICT, TAMIL NADU, INDIA

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ABSTRACT

Microbial diversity near public rest rooms of new bus stand, Thanjavur district, Tamil Nadu, India was carried out in the present study. The study reveals that, PDA represented higher colony number (7.33±1.69) while LB and KB revealed only 5.67±0.94. The isolated colonies were identified as *Escherchia coli*, *Streptococcus* sp. *Klepsiellapneumoniae*and *Staphyloccus*sp. Among the bacteria, the antibacterial resistance level was high in *Staphylococcus* sp. against Ciprofloxacin while *Streptococcus* sp. reveal only 19mm zone of inhibition against Carbenicillin and Ampicillin/Sulbactam. Thus, the present study reveals that, new bus stand would be projected as possible health risk prone area and are to be studied with people who are travelling in and around them.

Keywords: New Bus stand, Bacteria, Microbial diversity, Antimicrobial resistance.

INTRODUCTION

Earth's atmosphere is known to team with airborne microorganisms, make the atmosphere as unsuitable environment habitat for microbial growth [1]. Biological material may contribute about 20%, 22% and 10% to the total airborne particulate by volume in remote continental, populated continental and remote maritime environments respectively [2]. Most of them originate from natural sources such as soil, lakes, animals and humans [3]. Moreover, agricultural practices, healthcare units and industrial operations such as sewage treatment, animal rendering, fermentation processes, and food processing plants also emit viable microorganisms into the air [4]. Among the microorganisms present in the atmosphere, bacteria are often the highest in number, despite their high death rate due to environmental factors producing stress of various kinds, of the major being dehydration stress [5]. Though, most of the bacteria, or bacterial agents are not very potent allergens with the exception of spore forming actinomycetes; bacterial cell wall components, such as endotoxin (most prevalent in gram negative bacteria) and peptidoglycens (most prevalent in grampositive bacteria) are crucial agents with important pro-inflamatory properties that may induce respiratory symptoms [6]. In this context, the present study, aim tostudy the airbornemicrobial diversity in and around publicrest rooms near
New Bus standThanjvur District, Tami Nadu, India.

MATERIALS AND METHODS

Study Area

The study was carried out in New Bus stand, Thanjavur District during 2014 - 2015.

ISOLATION OF MICROORGANISMS

Air sampling and microbiological examination

The microbiological samples were collected from New Bus stand, ThanjavurDt, and Old Bus stand, Thanjavur Dt. by exposing the prepared petridishes containing NA, LB, KB and Potato dextrose agar (PDA) for the period of 15 minutes. Upon exposure, the plates were transported to the laboratory for examination. The bacterial culture plates were incubated at 37°C for 24 hrs while the fungal culture plates were incubated at 27°C temperature for 48-72 hrs.

The total number of colony forming units (cfu) was enumerated and converted to organisms per cubic meter air. Bacterial colonies were initially characterized by

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morphology and microscopic examination and identified further by biochemical tests. The fungal colonies were identified the test were based mainly on gross colonial appearance, microscopic examination of the spore and hyphal characteristics of lactophenol cotton blue preparations.

IDENTIFICATION OF BACTERIA

The bacteria was identified by using gram staining and biochemical analysis by using the method followed by Bailey and Scott (1966).

Antimicrobial Activity

The antibiotic sensitivity of isolated bacterial species to the commercial antibiotic tests was analyzed by disc diffusion method. Antimicrobial activity test was carried out following the modification of the method originally described by Bauer *et al.,* (1996).

Statistical Analysis

The results obtained in the present investigation were subject to statistical analysis like Mean $(\bar{x})$ and Standard Deviation (SD) by Zar (1984).

Results

The present study investigated the airborne microbial diversity around public rest rooms (toilet) near New bus stand, Thanjavur District, Tamilnadu, India. The study site was exposed with different nutrient medium such as Nutrient Agar (NA), Luria Bertani Agar (LB), King's-B medium (KB) and Potato Dextrose Agar (PDA) for 15 minutes and the bacterial and fungal colonies assemblages were identified after 24 and 72 hr incubation respectively . Table 1 reveals that the average number of colonies obtained on the different nutrient media, in which PDA represented higher colony number (7.33±1.69) while LB and KB revealed only 5.67±0.94. The isolated colonies were identified as *Escherchia coli*, *Streptococcus* sp. *Klepsiella pneumoniae* and *Staphyloccus* sp. and their morphological characters of the isolated bacteria were presented in table 2. The biochemical characterization of these bacterial species were depicted in table 3.Those of the isolated bacteria species were subjected to different antibiotic disc and identified their level of antibacterial resistance. Table 4 reveals that, the gram positive bacteria, *Staphylococcus* sp. had shown highest level of zone of inhibition (20mm) against Ciprofloxacin while *Streptococcus* sp. reveal only 19mm zone of inhibition against Carbenicillin and Ampicillin/Sulbactam. A similar resistance level was also observed on *Klepsiella pneumoniae* and *Escherchia coli* against Carbenicillin and Ampicillin/Sulbactam and are 19 and 18mm zone of inhibition respectively.

DISCUSSION

The present study isolate bacterial species from air samplings near public restrooms at New Bus stand, Thanjavur. Public rest rooms in addition to help people, had been greatly contaminated with microbes from human secretions as saliva skin, urine and faecal origin [7] The most implicated probable sources of infections is door handles of toilets and bathroom [6]. Bacteria seeded into toilets remain in the toilet for a long time after multiple flushing and cleaning with antimicrobial fluids [8]. The increasing incidence of epidemic outbreaks of certain diseases and its rate of spread from one community to the other has become a major public health concern [9]. In the present investigation also there are few bacterial species were isolated from air samplings such as *Escherchia coli*, *Streptococcus* sp. *Klepsiellapneumoniae*and *Staphyloccus*sp. Restroom surfaces host relatively diverse microbial communities dominated by human-associated bacteria with clear linkages between communities on or in different body sites and those communities found on restroom surfaces, relevant to the public health field that human-associated microbes are commonly found on restroom surfaces suggesting that bacterial pathogens could readily be transmitted between individuals by the touching of surfaces. Bacteria sampling of public restrooms which enable the people to develop an understanding of the restroom sites that pose the greatest risk of contamination to the public. Studies of hostel restroom, toilet seat have lower number of *Staphylococcus aureus* and *Pseudomonas* sp. than sinks and floor. This study supported the present investigation that, there was dispersal of *Staphylococcus* sp. was identified in the air samplings. Thus, the present investigation had shown a light into the views of people about public restroom and the human health risk factors were identified in the present study.

Table 1. Number of Colonies of bacteria and fungi observed in different nutrient media after 15 minutes exposure near public rest room of New Bus stand, Thanjavur District, Tamil Nadu, India.

S.No.	Nutrient Medium	Number of Colonies
1	Nutrient Agar (NA)	6±0.8
2	Luria Bertani Agar (LB)	5.67±0.94
3	King's B (KB)	5.67±0.94
4	Potato Dextrose Agar (PDA)	7.33±1.69

Table 2. Name and Morphological characters of bacteria isolated from public rest room of New Bus stand, Thanjavur District, Tamil Nadu, India.

S. No	Morphological Character	*Escherchia Coli*	*Streptococcus sp.*	*Staphylococcus sp.*	*Klepsiella sp.*
1	Size	Large	Moderate	Moderate	Small
2	Shape	Filamentous	Circular	Irregular	Circular
3	Pigmentation	Pink	Colorless	Pink	Pink
4	Margin	Rhizoid	Entire	Curled	Undualated
5	Elevation	Pulvinate	Embonate	Convex	Pulvinate
6	Texture	Smooth	Rough	Rough	Smooth
7	Appearance	Glistering	Dull	Dull	Glistering
8	Optical Property	Tansluscent	Translucent	Translucent	Translucent

Table 3. Biochemical characterization of airborne bacterial species isolated from public rest room near New Bus stand, Thanjavur district, Tamil Nadu, India.

S. No.	Morphological and Biochemical Characterization	Isolated Bacterial Colony			
		Staphylococcus sp.	*Pseudomonas sp.*	*Klepsiella pnuemoniae*	*E.coli*
1.	Gram staining	'+'	'+'	'–'	'–'
2.	Shape	Coccus	Coccus	Rod	Rod
3.	Motility Test	'–'	'–'	'–'	'+'
4.	Indole Test	'–'	'–'	'–'	'+'
5.	Methyl Red Test	'–'	'+'	'±'	'+'
6.	VogesProskauer Test	'+'	'+'	'+'	'–'
7.	Citrate Utilization Test	'–'	'±'	'+'	'–'
8.	Oxidase Test	'+'	'–'	'–'	'–'
9.	Triple Sugar Iron Test	'+'	AG	AG	AG
10.	Catalase Test	'–'	'+'	'+'	'+'
11.	Carbohydrate fermentation Test				
	Glucose	'+'	'+'	'+'	'+'
	Lactose	'+'	'+'	'–'	'+'
12.	Sucrose	'+'	'+'	'+'	'–'
13.	Ureas Hydrolysis Test	'–'	'+'	'+'	'–'
14.	Starch Hydrolysis Test	'–'	'–'	'+'	'–'
15.	Hydrogen Sulfide Production	'–'	'–''	'+	'+'

Table 4. Antibacterial resistance of bacteria isolated from public rest rooms at New Bus stand, Thanjavur against selected antibacterial discs

Antibiotics	Zone of Inhibition			
	Gram Positive		Gram Negative	
	Staphyloccus sp.	*Streptococcus sp.*	*Klepsiella pneumoniae*	*Escherchia coli*
Cephalothin	0	0	18	0
Carbenicillin	18	19	19	18
Ampiciiln/Sulbactam	12	19	17	18
Cefixime	0	0	0	17
Ciprofloxacin	20	0	0	0
Nalidixic Acid	19	0	0	0

ACKNOWLEDGEMENT: None

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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