



ISOLATION AND IDENTIFICATION OF BACTERIAL ISOLATES FROM THUMB IMPRESSION

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Abstract: Millions of people around the globe suffer from various disease per annum. Extensive part of these infections is hand acquired infections. World Health Organization suggests hand hygiene as a primitive precaution focus to decrease the infection rates. We evaluated various aspects related to the occupational behavior and prioritization regarding hand hygiene measures among the College students, Department of Microbiology, Dr.N.G.P Arts and Science College, Coimbatore, Tamilnadu. Gram positive cocci were found in abundance followed by Gram positive rods. The antibiotics resistance profile showed that the isolated bacterial strains were mainly sensitive to Streptomycin, whereas, majority of the isolates were resistant to cefixime - a third generation Cephalosporin. Methicillin resistance was also found common among *Staphylococcus spp.* The study indicated the resistance spread among the opportunistic normal flora and the role of unhygienic hands in their transmission.

Index Terms – Hand hygiene, Kirby-bauer test, MRSA, Colony forming unit

I. INTRODUCTION

Infection transmission through contaminated hands is a typical pattern seeing in most healthcare settings. Usually, health care- associated infections are associated with inappropriate hand hygiene practices. The World Health Organisation reported that Multi Drug Resistant Organisms as a significant contributor to outbreaks of infectious diseases . It has been reported that hand hygiene is the single most important measure to prevent cross-transmission of microorganisms from one patient to another and preventing Healthcare-associated infections. It prevents the transmission of pathogens through contact and faecal-oral route. It is one of the essential components of infection control programs. Hand rubbing with alcohol-based rub-in cleanser (waterless) is very common now-a-days in hospitals instead of hand washing. Scientific evidence supports employment of hand rubs for routine hand hygiene. It is microbiologically more efficient in vitro and in vivo and saves time and preliminary data demonstrate better compliance than with hand washing. Hand hygiene has now been recognised as one of the most effective interventions to control the transmission of infections in a hospital. The proper use of hand hygiene techniques also needs continuing education. The display of posters provided by the World Health Organisation (WHO) is an effective tool of education. Also, continuing medical education programs for all level of health care providers are important. Although, the discovery of antibiotics predicted the ultimate control of microbial infections but this goal could not be successfully achieved because of the rapid emergence of resistance among pathogenic micro-bugs. In general, the frequent use of antimicrobial agents causes the selective pressure which led to the resistance. The pathogenic microbes and their antibiotic sensitivity may change from time to time and place to place. Thus, understanding prevalent antibiotic resistance pattern of the common pathogenic bacteria in a particular region is particularly useful in clinical practices. Various epidemiological studies manifested the spread of antibiotic resistance among microbial pathogen. The human microbiota warrants special attention as perhaps being the most accessible reservoir of resistance genes due to the high likelihood of contact and genetic exchange with potential pathogens. Hands are the most important part of the body with respect to the exposure and spread of bacteria. Particularly, in health care settings and clinical lab setups where surfaces are often contaminated, the hands of the staff and lab personnel do serve as a vector for cross contamination, the present study was conducted for the isolation and identification of bacterial and fungal isolates from the thumb of the students, Department of Microbiology, Dr.N.G.P Arts and Science College, Coimbatore, Tamilnadu

II. MATERIALS

AND METHODS

Nutrient Agar

Agar Agar Conical flask Weighing balance Autoclave

Petriplates and Petri can

Hot Air oven

Laminar Air flow

chamber Bunsen

burner

Handwash Incubator

2.1 THUMB IMPRESSION ANALYSIS :

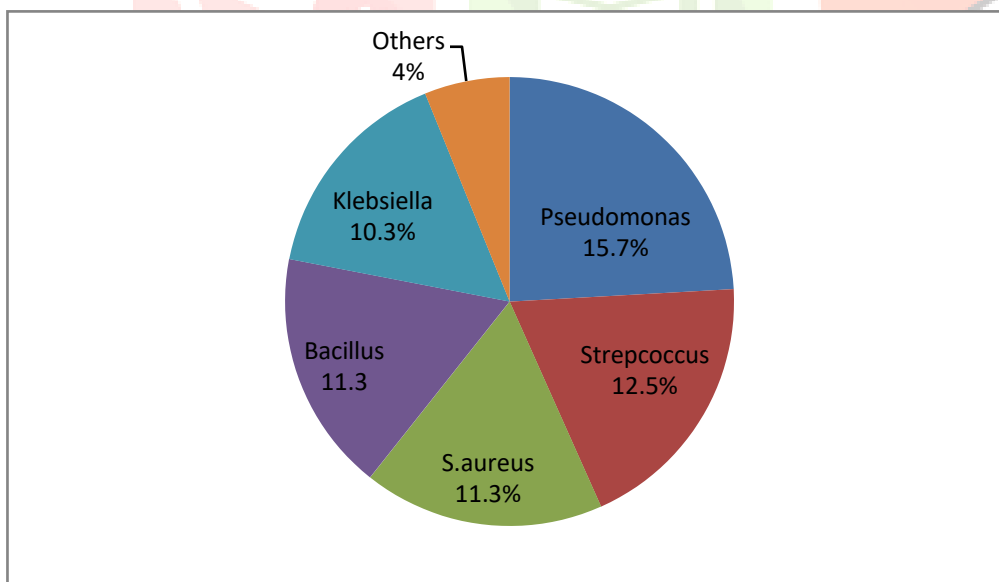
Thumb impressions of students from Department of Microbiology, Dr. N.G.P Arts and science College of Coimbatore, Tamilnadu were taken on nutrient agar plates. After a procedure, three fingertips on the dominant hand of the volunteer were pressed on the surface of agar plate for approximately 10 second. In order to obtain identical conditions for each specimen, the agar was applied on to the fingers by the help of the students to obtain identical pressure. The hand hygiene technique was then performed, and a second imprint of the fingertips was obtained one minute later. Plates were incubated at 37°C under aerobic conditions, and Colony-Forming Units (CFUs) were counted after 48 hours. The maximum count was 300 CFUS; beyond this figure, it was considered too many to count (TMTC). Potential pathogenic bacteria from transient were identified using standard microbiological techniques (Gram staining biochemical tests) and Next day, morphologically distinct colonies were selected and purified by further streaking on nutrient agar slants.

2.2 ANTIBIOTIC SENSITIVITY TEST:

Identification of the isolates and antibiotic sensitivity tests By Morpho-cultural and biochemical characteristics, the isolated bacterial strains were identified. Gram staining and biochemical tests were done as per guidelines of Clinical and Laboratory Standards Institute (CLSI). Kirby-Bauer disc diffusion method was used for the antibiotic sensitivity tests. A true antibiotic is an antimicrobial chemical produced by microorganisms against other microorganisms. Mankind has made very good use of these antimicrobials in its fight against infectious disease. Many drugs are now completely synthetic or the natural drug is manipulated to change its structure somewhat, the latter called semisynthetics. Bacteria respond in different ways to antibiotics and chemosynthetic drugs, even within the same species. For example, *Staphylococcus aureus* is a common normal flora bacterium found in the body. If one isolated this bacterium from 5 different people, the 5 isolates would likely be different strains, that is, slight genetically different. It is also likely that if antibiotic sensitivity tests were run on these isolates, the results would vary against the different antibiotics used.

III. RESULT AND DISCUSSION:

Fig1, Percentage of isolates from hand



Out of 10 samples, 29 bacterial isolates was found. Of these, 30% (3/10) showed monomicrobial colony however, polymicrobial colonies were observed in 70% (7/10). *Pseudomonas* species was isolated in 15.7% and *Staphylococcus aureus* and *Bacillus* species each was isolated in 12.3% of bacterial specimens. Methicillin resistant *Staphylococcus aureus* was isolated in 11% of the bacterial isolates. *Escherichia coli* was isolated in 12.6% of the bacterial isolates. *Klebsiella* species isolated in 10.3%. The isolation of other bacteria was less than 10%. It was observed that the bacterial count was varied from person to person as per the nature of their job and hygienic conditions. A significantly high bacterial load was found on the hands of canteen workers followed by laboratory staff.

Table 1, organisms found after self-contamination of hands, and handwashing with either soap, water only or no handwashing

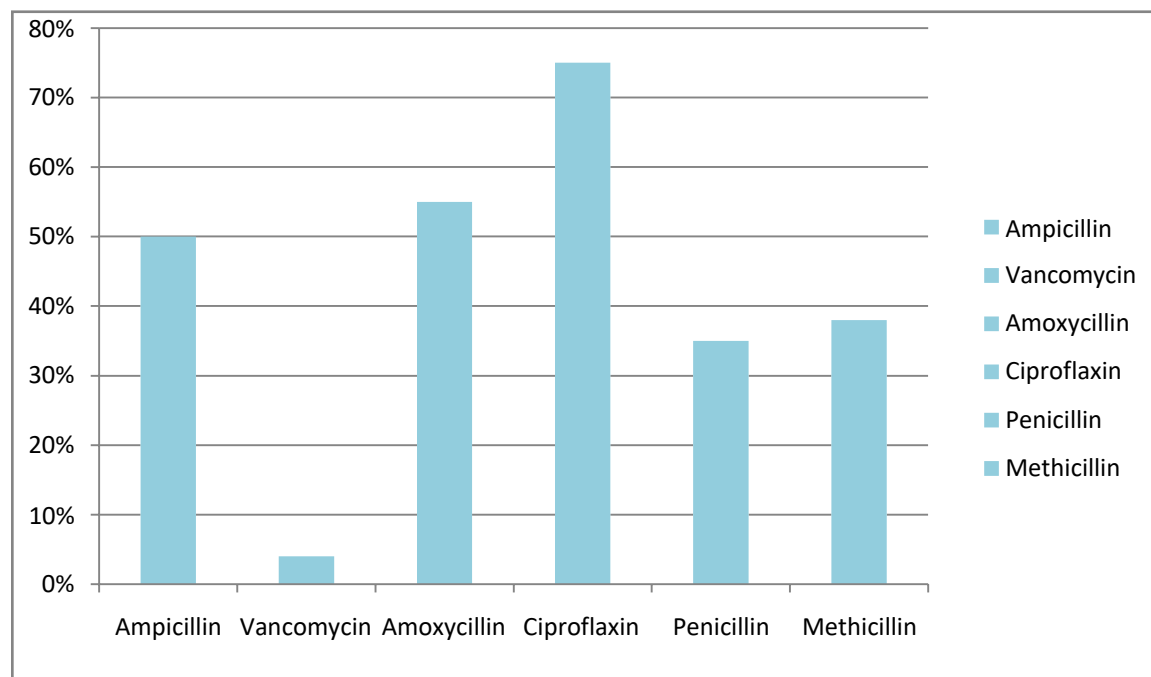
FAECAL BACTERIA	NO HANDWASHING	WATER ONLY	SOAP AND WATER
<i>Enterococcus spp.</i>	46(29%)	24(15%)	4(3%)
<i>Enterobacter amnigenus</i>	14(9%)	4(3%)	4(3%)
<i>Enterobacter cloacae</i>	13(8%)	5(3%)	2(1%)
<i>Shigella spp.</i>	2(1%)	1(1%)	0(0%)
<i>Klebsiella spp</i>	5(3%)	2(1%)	1(1%)
<i>E.coli spp</i>	0(0%)	0(0%)	1(1%)
Multiple isolation	10(6%)	2(1%)	0(0%)
Any bacteria	70(44%)	36(23%)	13(8%)
Total	160(100%)	160(100%)	160(100%)

whereas, the lowest bacteria load was noticed on the hands of girls wearing gloves. Majority of the isolates were gram positives including 20.62% Gram positive cocci in bunches (*Staphylococcus spp.*), 15.92% Gram positive rods (*Bacillus spp.*) and 12.5% gram positive chains (*Streptococcus spp.*). However, 6.11% strains were observed as gram negatives (*E. coli*). Antibiotic resistance profiles of the isolates are summarized. According to the results, the gram positive cocci were found more resistant to the (tested) antibiotics when compared with gram positive rods. The isolates were found sensitive to streptomycin while, the majority of the isolates showed resistance to cefixime- a third generation cephalosporin. Methicillin resistance was observed in 20.4% *Staphylococcus spp.* suggesting the presence of MRSA as a member of normal skin flora. The resistance towards penicillin was observed in 58.2% of *Staphylococcus spp.* and 35.7% in *Streptococcus sp.* The *Bacillus sp.* were found comparatively more susceptible to tested antibiotics. found mostly non-pathogenic bacteria. However, these can be significant in the case of an immune compromised host where the contamination with these bacteria may be responsible for invasive infections. *S. aureus* is one of the important pathogens, which is responsible for health care-associated infections (HCAI) all over the world. Rarely, Gram negative bacteria such as *Klebsiella spp.*, *Escherichia coli* or *Acinetobacter* species can be transient flora of the hands, which may be acquired by touching infected patients or contaminated surfaces. In the present study, *Pseudomonas* species (15.7%) was the most common bacterial isolate following *Staphylococcus aureus* and *Bacillus* species each constituted 11.3%. However, in a study the reported that hands of healthcare workers (HCWs) carried pathogens and *S. aureus* was isolated predominantly from 10 (12.3%). The study from Coimbatore, Tamilnadu had also reported the presence of potential pathogens on hands of various persons, studying in educational institutes. The isolates included *Staphylococcus* species (11%), *E. coli* (12.6%), *Klebsiella* species (10.3%).

Table 2, Percentage of isolates resistant to the tested antibiotics

Antibiotics	Percentage of isolate become resistant				
	<i>Staphylococcus</i>	<i>Bacillus</i>	<i>Streptococcus</i>	<i>Pseudomonas</i>	<i>Klebsiella</i>
Ampicillin	2.50	2.50	3.44	9.06	2.10
Vancomycin	55	9.33	50.2	91	20.3
Amoxicillin	40.2	29.9	66.4	55.2	45.7
Ciproflaxin	70.06	33.5	75.7	40.1	60.0
Penicilin	58.2	25.4	35.7	30.7	35.8
Methicillin	41.3	39.4	41.2	34.5	39.5

Fig2, antibiotic tested



IV. CONCLUSION:

The present findings indicate the importance of investigating bacterial isolates of the thumb to ascertain the epidemiology, level of antibiotic susceptibility of infection and thus develop intervention strategies preventive measures as well as treatment possibilities that the hand hygiene plays an important role in transmission of cross infections and it is necessary to generate evidences manifesting the role of good hygienic status in reducing the bacterial load on the hands as a tool to educate the masses and reduce the frequency of community acquired infections. Prevalence of bacterial resistance to the commonly prescribed antibiotics should also be a particular focus and it demands the training of the primary health care practitioners and to discourage the trends of indiscriminate use of antibiotics and self-medication practices in the country. More work is therefore suggested to address the Changing resistance pattern among the medical student on larger sample size.

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