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Prevalence and Antibiotic Resistance Pattern of *Staphylococcus aureus*Isolated from Clinical Samples: A Cross-sectional Study

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Abstract

Background: Staphylococcus aureus, mainly a hospital acquired infection is responsible for many suppurative lesions and has demonstrated the ability of developing resistance to many antimicrobial agents leading to life threatening infections and long hospital stay. Objective: This study was aimed to determine the prevalence and antibiotic susceptibility profiles of Staphylococcus aureus in different clinical specimens isolated from hospital admitted patients. Methodology: This cross-sectional study was conducted from 1 January 2019 to 31 December 2019 in the Microbiology Department of Dhaka Medical College, Dhaka, Bangladesh. Pus, wound swab and blood samples were included in this study. Isolation and identification of Staphylococcus aureus was based on culture, microscopy and different biochemical tests. The antibiotic susceptibility test was done by disc diffusion method using Kirby-Bauer technique. Results: Among 275 clinical specimens 21.63% Staphylococcus aureus were isolated among which 21.93% were isolated from pus and wound swabs and 19.05% from blood samples. Most of the Staphylococcus aureus showed resistance to azithromycin and erythromycin (68.89%) followed by clindamycin (66.67%), ampicillin (40%), linezolid (31.11%), amikacin (26.67%) and teicoplanin (24.44%). Conclusion: In conclusion there is a high prevalence of Staphylococcus aureus in clinical samples of hospitalized patients and it has shown alarmingly resistance to many of common antimicrobials.

Keywords: Antibiotic; prevalence; resistance; susceptibility; *Staphylococcus aureus Bangladesh Journal of Medical Microbiology, January 2023;17 (1):29-33*

Introduction

Staphylococcus aureus is an aerobic gram positive coccus and ubiquitous commensal that periodically lives on the skin and anterior nares of about one third of the healthy human population without causing illness^{1,2}. Most *Staphylococcus aureus* infections occur in persons who are colonized with the organism.

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Staphylococcus aureus carriage has long been known to be one of the most strongly associated risk factors for subsequent infection^{3,4}. The organism also elaborates toxins that can cause specific diseases and likely participate in the pathogenesis of *staphylococcal* infection⁵. Staphylococcus aureus causes a wide variety of diseases that ranges from skin and soft tissue infections, pneumonia, bloodstream infections, osteomyelitis and endocarditis, as well as toxin-mediated syndromes like toxic shock and food poisoning^{6,7}.

Varying degree of antibiotics resistance towards Staphylococcus aureus may be due to production of β -lactamase enzyme responsible for inactivation of

 β -lactam drugs, gene mutations and horizontal gene transfer and exposure to high selection pressure of antibiotics⁸.

The rise of drug-resistant is a serious problem in the treatment and control of *Staphylococcal* infections⁹. The knowledge of antimicrobial susceptibility profile of *Staphylococcus aureus* in a particular area is important as this can contribute to rational choice and use of antimicrobial agents¹⁰. The aim of the present study was to determine the prevalence and antibiotic susceptibility profiles of *Staphylococcus aureus* in different clinical samples isolated from hospital admitted patients.

Methodology

Study Settings and Population: This cross-sectional study was conducted from 1 January 2019 to 31 December 2019 in the Microbiology Department of Dhaka Medical College, Dhaka, Bangladesh.

Sample Collection Procedure: Samples were collected from pus, wound swab and blood of clinically suspected infected patients of inpatient departments of Dhaka Medical College Hospital or were received in the microbiology department for culture and sensitivity after taking informed written consent irrespective of age, sex and antibiotic intake. Patient who did not give consent were excluded from this study.

Isolation and Identification: Wound swab and pus samples were inoculated in blood agar and MacConkey agar media and incubated at 37°C aerobically for 24 hours. Incubated plates were then examined for the presence of colonies of bacteria. Primary blood culture was done in Trypticase soya broth (TSB) for 24 hours and then subculture was done in blood agar and MacConkey agar media and incubated at 37°C aerobically for 24 hours.

Microscopic Examination: Smears were prepared from culture plates and stained by Gram's stain as per standard procedure. Then were examined under microscope for presence of gram positive or gram negative organisms¹¹.

Biochemical test: Among isolated gram positive cocci, *Staphylococcus aureus* was identified by catalase test, coagulase test (slide and tube method), colony morphology, hemolytic property, pigment production and mannitol fermentation test in mannitol salt agar media as per standard methods¹².

Antimicrobial susceptibility tests: All the Staphylococcus aureus isolates were tested for antimicrobial susceptibility testing by disc diffusion

method on Mueller Hinton agar using Kirby-Bauer technique¹³. Antibiotic discs used for sensitivity tests were Azithromycin, Clindamycin, Erythromycin, Linezolid, Ampicillin, Novobiocin, Teicoplanin, Amikacin.

Statistical Analysis: Statistical analyses performed with SPSS software, versions 22.0 (IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.). Continuous data that were normally distributed were summarized in terms of the mean, standard deviation, median, minimum, maximum and number of observations. Categorical or discrete data were summarized in terms of frequency counts and percentages. When values are missing, denominator was stated. Chi-square test was used for comparison of categorical variables. Every effort was made to obtain missing data. A two-sided P value of less than 0.05 was considered to indicate statistical significance.

Ethical Clearance: All procedures of the present study were carried out in accordance with the principles for human investigations (i.e., Helsinki Declaration) and also with the ethical guidelines of the Institutional research ethics. Formal ethics approval was granted by the Research Review Committee (RRC) of Department of Microbiology and Ethical Review Committee (ERC) of Dhaka Medical College, Dhaka, Bangladesh (Reference number: MEU-DMC/ ECC/2019/171). Participants in the study were informed about the procedure and purpose of the study and confidentiality of information provided. All participants consented willingly to be a part of the study during the data collection periods. All data were collected anonymously and analyzed using the coding system.

Results

A total 275 samples were included in the present study. Among them 230 pus and wound swabs and 45 blood samples. From the 275 samples 208 (75.6%) were culture positive. Culture yielded growth in 187 (81.30%) pus and wound swabs and 21 (46.67%) blood samples (Table 1).

Table 1: Culture Positivity Among Various Clinical Samples (n= 275)

Samples	Culture	Culture	Total
	Positive	Negative	
Pus and wound swabs	187 (81.3%)	43 (18.7%)	230 (100.0%)
Blood	21 (46.7%)	24 (53.3%)	45 (100.0%)
Total	208 (75.6%)	67 (24.4%)	275 (100.0%)

Out of 187 pus and wound swabs samples 41 (21.93%) Staphylococcus aureus was isolated followed by 4(19.05%) from 21 blood samples (Table 2).

Table 2: Distribution of *Staphylococcus aureus* Isolated from Culture Positive Samples (n=208)

Samples	Culture	Culture	Total
	Positive	Negative	
Pus and wound			
swabs	41 (21.93%)	146 (78.07%)	187 (100.0%)
Blood	4 (19.05%)	17 (80.95%)	21 (100.0%)
Total	45 (21.63%)	163 (78.37%)	208 (100.0%)

Staphylococcus aureus showed higher resistance to azithromycin and erythromycin (68.89%) followed by clindamycin (66.67%), ampicillin (40%), linezolid (31.11%), amikacin (26.67%) and teicoplanin (24.44%). All (100%) isolated Staphylococcus aureus were sensitive to novobiocin (Table 3).

Table 3: Antimicrobial Resistance Pattern of Isolated Staphylococcus aureus from Clinical Samples (n=45)

Antimicrobial drugs	Frequency	Percent	
Azithromycin	31	68.89	
Erythromycin	31	68.89	
Clindamycin	30	66.67	
Ampicillin	18	40.0	
Linezolid	14	31.11	
Amikacin	12	26.67	
Teicoplanin	11	24.44	
Novobiocin	0	0.00	

Discussion

Staphylococcus aureus is the most frequently occurring bacterial pathogen among clinical samples from hospital inpatients in the United States and is the second most prevalent bacterial pathogen among outpatients¹⁴. Staphylococcus aureus bacteremia was associated with a long duration of hospital stay, high treatment cost and increased risk of mortality, compared with bacteremia by any other pathogen and a significant economic burden on health care systems¹⁵.

In the present study, 45 (21.63 %) *Staphylococcus aureus* were isolated from clinical samples. This finding was almost similar to the related study in India who reported that 18.70% *Staphylococcus aureus* were isolated in their studies¹⁶. Another study showed that 33.3% *Staphylococcus aureus* were isolated in their studies¹⁷. This divergence might be due to the difference in types of samples, difference in sample size and number of hospital surveyed, season of

collecting samples and medication taken before sampling¹⁸. In our study, maximum (21.93%) *Staphylococcus aureus* were isolated from pus and wound swabs which concurs with a previous study (29.4%)¹⁹. Antimicrobial agents have been used extensively to combat *Staphylococcus aureus* infections but the increasing level of resistance of *Staphylococcus aureus* to many antibiotics is complicating the treatment of serious infections caused by this pathogen²⁰.

In the present study, 68.89% *Staphylococcus aureus* showed resistance to erythromycin. This result was similar to the related previous study²¹. Another study showed that 46.3% erythromycin resistance for *Staphylococcus aureus*²². The rate of erythromycin resistance varies greatly in different countries which may reflect on the difference in selective pressure of antibiotics and infection control policies²³.

In this current study, 68.89 % *Staphylococcus aureus* showed resistance to azithromycin. A previous study found 49.2% resistance²⁴. In our study, 66.67% *Staphylococcus aureus* showed resistance to clindamycin. Two previous studies found 77.3% and 40.7% clindamycin resistance respectively^{25,26}.

In the present study, 40% *Staphylococcus aureus* showed resistance to ampicillin. This result was similar to the study of Turutoglu et al²⁷. Another study observed that 100% *Staphylococcus aureus* showed resistance to ampicillin. The higher resistance of *Staphylococcus aureus* to ampicillin may be attributed to the fact that it is the frequently used antibiotic for the treatment of skin and nasal infections²⁸.

In this current study, 31.11% *Staphylococcus aureus* showed resistance to linezolid. This result was in agreement with the study of Amr et al²⁹. Garcia et al³⁰ showed that linezolid resistance probably develops on exposure.

Many factors are responsible for the differences in the antibiotic resistance profile such as various geographic regions, local infection preventive measures implemented, antibiotic prescribing guidelines and epidemiology of the studied strains themselves³¹.

Antibiotic resistance usually occurs in Bangladesh and other developing countries as a result of some incidences including the use of antibiotics without performing AST and the inappropriate prescribing of antibiotics, unethical practices of health professionals, unqualified drug sellers offering alternative drugs when the prescribed drugs are unavailable and the uncontrolled use of antibiotic in agriculture and livestock^{24,25}.

There are some limitations of the study. The incidence of Staphylococcus aureus infection and complications has increased sharply in recent years because of the increased frequency of invasive procedures, increased numbers immunocompromised patients and increased resistance of Staphylococcus aureus strains to available antibiotics. epidemiology This changing Staphylococcus aureus infection, in combination with the inherent virulence of the pathogen, is driving an urgent need for improved strategies and better antibiotics to prevent and treat Staphylococcus aureus infection and its complications.

Conclusion

Staphylococcus aureus is a particularly complex, virulent and successful pathogen. There is a higher risk of acquiring multidrug resistant Staphylococcus aureus infection in hospital admitted patients. The chronic misuse and overuse of antibiotics in community level have deteriorated the drug-resistant problem. We should cautious of prescribing antibiotics with accurate dose and duration after culture and sensitivity reports are available. Policies need to be adopted in order to stop an increasing resistance of Staphylococcus aureus to different antibiotics due to high cost and time in developing new antibiotics. The clinical isolates of hospitalized patients exhibit a relatively high prevalence of Staphylococcus aureus which is an alarming issue and significant economic burden on health care systems.

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Conflict of Interest

There is no conflict of interest.

Financial Disclosure

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Authors' contributions

Noor-E-Jannat Tania conceived and designed the study, analyzed the data, interpreted the results, and wrote up the draft manuscript. SM Shamsuzzaman contributed to the analysis of the data, interpretation of the results and critically reviewing the manuscript. Aminul Islam contributed to the data analysis and manuscript writing. Maminur Rahman involved in the manuscript review and editing. Khadijatul Kubra involved in the manuscript review and editing. All authors read and approved the final manuscript.

Data Availability

Any inquiries regarding supporting data availability of this study should be directed to the corresponding author and are available from the corresponding author on reasonable request.

Ethics Approval and Consent to Participate

Ethical approval for the study was obtained from the Institutional Review Board. As this was a retrospective study the written informed consent was not obtained from all study participants. All methods were performed in accordance with the relevant guidelines and regulations.

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