



ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF STAPHYLOCOCCUS AUREUS ISOLATES FROM VARIOUS CLINICAL SPECIMENS AT TERTIARY CARE TEACHING HOSPITAL IN SOLAPUR, MAHARASHTRA.

Microbiology

Dr Anu Sharma*	Assistant Professor, Microbiology department, Dr VMGMC Solapur, *Corresponding Author
Dr Sonal Agarwal	Post Graduate student, Microbiology department, Dr VMGMC, Solapur,
Dr K.V. Ingole	Professor & Head, Microbiology department, Dr VMGMC, Solapur.

ABSTRACT

Staphylococci are normal inhabitants of human skin and mucous membrane. They play important role as human pathogens causing both nosocomial as well as community acquired infection. Over the time there has been increased prevalence of MRSA among hospitalized patients resulting in increased morbidity and mortality among patients.

AIM & OBJECTIVE of the study is to detect the distribution of *Staphylococcus* in various clinical specimens and to detect antibiotic susceptibility pattern of these strains against various antibiotics used for the treatment of patients.

MATERIAL & METHOD- *Staphylococcus* strains were identified using standard procedures and antibiotic susceptibility testing was done on Mueller Hinton agar by Kirby Bauer disc diffusion method.

RESULT- Total sample received for culture & sensitivity over 6 months from May to October 2018 were 3452 of which 1054 (36.32%) were sterile and 2198 (63.67%) showed growth. From 2198 (63.67%) samples 350 (15.92%) showed growth of *Staphylococcus* species. From these *Staphylococcus* strain MRSA were 190 (54.28%) followed by MRCONS 86 (24.57%), MSSA 50 (14.28%) and MCONS 24 (6.85%) Most of the *Staphylococcus aureus* strains isolated were from pus and wound swab 236 (67.42%) followed by blood 64 (18.28%), urine 16 (4.57%), vaginal swab 14 (4%), peritoneal fluid 8 (2.28%), sputum 6 (1.71%) and others 6 (1.71%). Male 194 (55.42%) were more commonly infected with *Staphylococcus aureus* then females 156 (44.57%). Most common age group affected was between 20 to 30 years accounted to 90 (25.71%) cases followed by 30 to 40 years 75 (21.42%), 40 to 50 years 60 (17.14%), 10 to 20 years 52 (14.85%), more than 50 years 40 (11.42%) cases, least number of cases were reported from age group less than 10 years 33 (9.42%). Majority of *Staphylococcus aureus* strains isolated were from hospitalized patients of Surgery department 132 (37.71%) followed by Obstetrics and Gynecology 60 (17.14%), OPD 56 (16%), Pediatrics 42 (12%), ENT 20 (5.71%), Medicine 20 (5.71%), TICU 14 (4%), Orthopedics 06 (1.71%). Antibiotic susceptibility pattern of *Staphylococcus* strains showed 100% sensitivity to Vancomycin, followed by Gentamicin 81.80%, Ciprofloxacin 50.28%, Erythromycin 35.42%, Clindamycin 34.28%. Maximum resistance was noted for Cefoxitin 78.85%.

KEYWORDS

MRSA (Methicillin resistant *Staphylococcus aureus*), MRCONS (Methicillin resistant coagulase negative *Staphylococcus aureus*) MSSA (Methicillin sensitive *Staphylococcus aureus*), MCONS (Methicillin sensitive coagulase negative *Staphylococcus aureus*).

INTRODUCTION

Staphylococcus genus is heterogeneous group of bacteria consisting of 30 species from which *Staphylococcus aureus* is most important clinical entity¹. It is part of normal flora of human body and is common inhabitant of skin and nose of healthy individuals, which leads to easy transmission by air or fomite from patient or carrier to another person^{2,3}.

Staphylococcus aureus causes wide range of diseases which includes septicemia, meningitis, endocarditis, osteomyelitis, septic arthritis, toxic shock syndrome and food poisoning^{4,5,6}. Previous studies suggests that *Staphylococcus aureus* has become resistant to most of the available antibiotics^{4,7}. In early 1950 acquisition and spread of beta lactamase producing plasmids led to rapid development of resistance to penicillin. In 1950 methicillin which is semi synthetic penicillin was introduced but in 1960 methicillin resistant *Staphylococcus aureus* strains were identified. The mechanism by which methicillin resistance occur was mostly by alteration of penicillin binding protein PBP to PBP2a which is encoded by mec-A gene. Increase number of isolated MRSA strains has led to complication in treatment of *Staphylococcus infection*^{8,9,10}.

Present study aims at finding the prevalence of *Staphylococcus aureus* in various clinical specimens received in Microbiology department for culture & sensitivity at a tertiary care teaching hospital in Solapur, Maharashtra and to know its antibiotic susceptibility pattern.

MATERIAL & METHOD

Present study is retrospective study carried out in Microbiology department for a period of 6 months from May to October 2018 at tertiary care hospital in Solapur. All samples received for culture & sensitivity from different departments were inoculated on blood agar, Mac conkey agar and CLED medium (in case of urine), these plates were incubated overnight at 37°C. Next day growth was noted, gram staining was done, further identification was done using standard procedures like catalase test, slide and tube coagulase test, DNase and Phosphatase test. Coagulase negative *Staphylococcus aureus* was considered pathogenic only when it was isolated on repeat sample.

Antibiotic susceptibility testing was done on Mueller Hinton agar using Kirby Bauer disk diffusion method as per CLSI guidelines¹¹

RESULTS

Total sample received for culture & sensitivity over 6 months from May to October 2018 were 3452 of which 1054 (36.32%) were sterile and 2198 (63.67%) showed growth. From 2198 (63.67%) samples 350 (15.92%) showed growth of *Staphylococcus* species.

TABLE-1 Age wise distribution of *Staphylococcus aureus* in various clinical specimens

AGE GROUP (years)	TOTAL NUMBER	PERCENTAGE (%)
Less than 10 year	33	9.42
10-20	52	14.85
20-30	90	25.71
30-40	75	21.42
40-50	60	17.14
More than 50 year	40	11.42

Graph-1 Gender wise distribution of *Staphylococcus aureus* in various clinical specimens

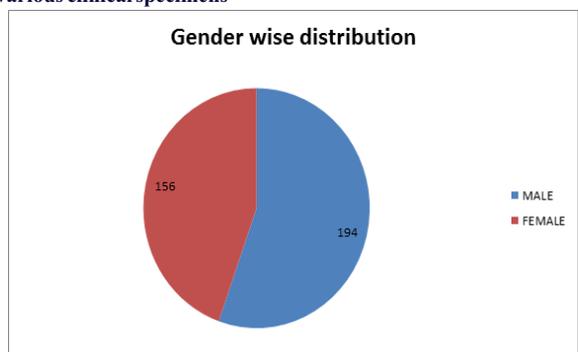


TABLE-2 Sample wise distribution of *Staphylococcus aureus* in various clinical specimens

SPECIMEN	TOTAL NUMBER OF ISOLATES	PERCENTAGE (%)
BLOOD	64	18.28
SPUTUM	06	1.71
CSF	0	0
VAGINAL SWAB	14	4
URINE	16	4.57
PUS & WOUND SWAB	236	67.42
PERITONEAL FLUID	8	2.28
OTHERS	6	1.71
TOTAL	350	100

TABLE-3 Department wise di

DEPARTMENT	TOTAL NUMBER OF ISOLATES	PERCENTAGE (%)
MEDICINE	20	5.71
SURGERY	132	37.71
OBSTETRICS & GYNECOLOGY	60	17.14
PAEDIATRICS	42	12
OPD	56	16

TABLE-6 Overall antibiotic susceptibility pattern of *Staphylococcus aureus*

ANTIMICROBIAL AGENT	TOTAL SENSITIVE ISOLATES	PERCENTAGE (%)	TOTAL RESISTANT ISOLATES	PERCENTAGE (%)
Cefoxitin 30µg	74	21.15	276	78.85
Erythromycin 15µg	124	35.42	226	64.58
Vancomycin 30µg	350	100	0	0
Clindamycin 2µg	190	34.28	160	45.72
Ciprofloxacin 5µg	176	50.28	174	49.71
Gentamicin 10µg	286	81.80	64	18.20

DISCUSSION

Staphylococcus aureus is recognized as major human pathogen, pathogenicity and virulence are associated with capacity to produce several virulence factor⁴. In the present study sample received for culture & sensitivity over 6 months from May to October 2018 were 3452 of which 1054 (36.32%) were sterile and 2198 (63.67%) showed growth. From 2198 (63.67%) samples 350 (15.92%) showed growth of *Staphylococcus* species. The overall prevalence of staphylococcus aureus carriage in health care workers and microbiology laboratories is about 20.1%¹². In the present study males 194(55.42%) are more commonly affected by infections caused by *Staphylococcus aureus* than females 156 (44.57%) similar results were reported by Laupland KB et al¹³ and Roder BL et al¹⁴. This may be due to the fact that males are more exposed to outside environment for employment purposes especially in rural areas, as our hospital is tertiary care hospital most of the patients belong to rural area. In the present study most common age group affected was between 20 to 40 years of age (47.14%) similar results were reported by Jessica et al¹⁵ where most common age group was between 20 to 39 years this may be attributed to the fact that this age group most commonly is involved in managing livelihood so more prone to infections.. In the present study Methicillin resistant staphylococcus aureus 190 (54.28%) was most commonly isolated from various clinical specimens and among them majority of *Staphylococcus aureus* were isolated from pus and wound swab 236 (67.42%) followed by blood 64 (18.28%) and urine 16 (4.57%). Study done by Mazhar Salim Al Zoubi et al¹ shows similar results, where MRSA were most common and majority of *Staphylococcus aureus* were isolated from pus and wound swab followed by blood. In the present study Vancomycin is only antibiotic effective against all isolates in study. Overall maximum sensitivity was observed for Vancomycin (100%) followed by Gentamicin (81.80%) and Ciprofloxacin (50.28%). Maximum resistance was noted for Cefoxitin 276 (78.85%). Studies done by various workers have shown variation in antibiotic susceptibility pattern which may be attributed to study population, geographical conditions, socioeconomic status of patient and adherence to hospital infection control practices among health care workers and awareness among general population and health care workers about importance of hand washing.

TRAUMA ICU	14	4
ENT	20	5.71
ORTHOPEDICS	06	1.71
TOTAL	350	100

TABLE-4 Distribution of *Staphylococcus aureus* based on sensitivity to Cefoxitin

MRSA	MSSA	MRCNS	MSCNS
190 (54.28%)	50 (14.28%)	86 (24.57%)	24 (6.85%)

TABLE-5 Antibiotic susceptibility pattern of *Staphylococcus aureus*

ANTIMICROBIAL AGENT	MRSA n=190, (sensitive isolates)	MSSA n=50, (sensitive isolates)	MRCNS n=86, (sensitive isolates)	MSCNS n=28, (sensitive isolates)
Erythromycin 15µg	56 (29%)	34 (68%)	28 (32%)	06 (25%)
Clindamycin 2µg	94 (49%)	44 (88%)	44 (51%)	08 (33%)
Vancomycin 30µg	190 (100%)	50 (100%)	86 (100%)	28 (100%)
Gentamicin 10µg	164 (86%)	50 (100%)	70 (81%)	02 (8%)
Ciprofloxacin 5µg	90 (47%)	36 (72%)	38 (44%)	14 (50%)

CONCLUSION

Staphylococcus aureus infection commonly showed response to beta lactams and other related group of antibiotics in the past but due to emergence of MRSA at very rapid pace especially in hospitals, it has exerted serious therapeutic challenge which has led to increased mortality and morbidity among hospitalized patients So it is important that we should spread awareness among general population as well as health care workers about importance of infection control practices, hand hygiene practices, and all health care workers should adhere to infection control practices strictly and follow bundling protocols to prevent device related infections . it is important duty as doctors that we must report if there is any negligence in following hospital infection control policies, or irregularities in biomedical waste disposal to the concerned authority as these strains spread through infected and colonized patients in hospital and main source of transmission is hospital staff. Judicious use of antibiotics will also prevent spread of resistance

REFERENCES

- Mazhar Salim Al Zoubi, Ibrahim Ali Al Tayyar, Emad Hussein, Alla Al Jabali, Salih Khudairat. Antimicrobial Susceptibility pattern of *Staphylococcus aureus* isolated from clinical specimens in Northern area of Jordan. Iran J Microbiol. Volume 7, number 5 (October 2015) 265-272.
- Brown DF, Edwards DI, Hawkey PM, Morrison D, Ridgway GL, Towner KJ. Guidelines for the laboratory diagnosis and susceptibility testing of methicillin resistant *Staphylococcus aureus* (MRSA). J Antimicrob Chemother 2005;56:1000-1018.
- Anupurba S, Sen MR, Nath G, Sharma BM, Gulati AK, Mohapatra TM. Prevalence of methicillin resistant *Staphylococcus aureus* in tertiary referral hospital in eastern Uttar Pradesh. Indian J Med Microbiol 2003;21: 49-51.
- Fischetti V.A., Novick R.P., Ferretti J.J., Portnoy D.A. Rood J.I. Gram positive pathogens. Washington, DC: ASM Press: 2000: Pathogenicity factors and their regulation: pp 392-407.
- Arbuthnot J.P., Coleman D.C., de Avazedo J.S. *Staphylococcus* toxins in human disease. Soc Appl Bacteriol Symp Ser. 1990;19:s101-7. [PubMed].
- Lina G, Gillet Y, Vandenesch F, Jones M.E, Floret D, Etienne J. Toxin involvement in *Staphylococcus aureus* scalded skin syndrome. Clin Infect Dis 1997;25:1369-73.
- Novick R.P. Autoinduction and signal transduction in the regulation of *Staphylococcus aureus* virulence. Mol Microbiol. 2003;48: 1429-1449 [PubMed].
- Gomes A.R., Vinga S, Zavolan M, de Lencastre H. Analysis of genetic variability of virulence related loci in epidemic clones of methicillin resistant *Staphylococcus aureus*. Antimicrob Agents Chemother. 2005;49:366-379. [PubMed].
- Hiramatsu K, Chui L, Kuroda M., Ito T. The emergence and evolution of methicillin resistant *Staphylococcus aureus*. Trends. Microbiol. 2000;9:486-493.
- Stefani S, Varaldo P.E. Epidemiology of methicillin resistant *Staphylococci* in Europe.

- Clin. Microbiol. Infect. 2003;9:1179-1186.
11. Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobial Disks Susceptibility Tests; Approved Standard. 25th informational supplement CLSI document M100-S25. Wayne, PA: CLSI; 2015.
 12. Xiaoying Xie, Xinlu Dai, Lijia Ni, Baiji Chen, Zhaofan Luo, Yandan Yao, Xiquan Wu, Hongyu Li, Songyin Huang. Molecular epidemiology and Virulence characteristics of *Staphylococcus aureus* nasal colonization in medical laboratory staff: comparison between Microbiological and non Microbiological laboratories. BMC infectious diseases <http://doi.org/10.1186/s12879-018-3024-x>.
 13. Laupland K.B, Gregson DB, Zygun DA, Doig CJ, Mortis G, Church DL. Severe blood stream infections: a population based assessment. Critical care medicine. 2004;32:992-7.
 14. Roder BL, Wandall DA, Frimodt Moller N, Espersen F, Skinhoj P, Rosdahl VT. Clinical features of *Staphylococcus aureus* endocarditis: a 10 year experience in Denmark. Archives of Internal Medicine. 1999 Mar 8; 159:462-469.
 15. Bauer K.A., West JE, Balada Llasat JM, Pancholi P, Stevenson KB, Goff DA. An antimicrobial stewardship program's impact with rapid polymerase chain reaction methicillin resistant *Staphylococcus aureus* blood culture test in patients with *S. aureus* bacteremia. Clinical infectious disease: an official publication of the infectious diseases society of America. 2010; 51: 1074-80.