



Spectrum of antimicrobial sensitivity of *Staphylococcus aureus* isolated from pus in a Tertiary Medical Care Hospital in Kolkata, a 7 years experiences.

Medicine

Dr. Ashis Kumar Saha

M.D.(Cal), D.T.M & H (Cal), FRCP(Edinburgh), FRCP(Glasgow), FACP(USA), FICP(India), MNAMS(India). Associate Professor & Head of the department of Medicine K P C Medical College & Hospital Jadavpur, Kolkata, West Bengal

ABSTRACT

Aims and objectives: *Staphylococcus aureus* is the most common pus forming organism worldwide. It is more commonly present in nasal passage and skin, it is mostly responsible for nosocomial and post-operative infection. Now-a-days this organism developed resistance to many antibiotics and in hospital "hospital strains" is responsible for mortality. **Materials and methods:** Samples were collected in our K P C Medical College & Hospital from 7531 patients having diabetic foot, abscess, postoperative infection, and chronic otitis media during the year 2009 to 2016 December and sent to our bacteriology department for culture-sensitivity testing. The data was collected and analyzed extensively with the help of Statistical Package for the Social Sciences (SPSS) version 17. **Results:** Data demonstrated that 291 were *Staphylococcus aureus* (162 were methicillin sensitive *Staphylococcus aureus* (MSSA), 74 were methicillin resistant *Staphylococcus aureus* (MRSA) and 55 CoNS (Coagulase negative *Staphylococcus aureus*). In all groups of staphylococci males were significantly affected as compared to females ($p=0.00$). MRSA was highly (>90%) sensitive to teicoplanin, linazolid and vancomycin, MSSA to linazolid and teicoplanin and CoNS to vancomycin and teicoplanin. MSSA was > 70% to <90% sensitive to piperacillin-tazobactam, aminoglycoside group except Tobramycin, tetracycline and tigicycline, MRSA to netilmicin, amikacin and clindamycin and CoNS to chloramphenicol, tetracycline and linazolid. All the strains were nearly hundred percent resistance to polymyxin B and colistin, mild sensitive to semi synthetic penicillin and all generations of cephalosporins. **Conclusions:** This high degree of resistance is the result of extensive, illogical and inadvertent use of different antibiotics. So our aim to use the antibiotics very cautiously to prevent the development of resistance as well as to prevent the development of bacteria resistant to many commonly used antibiotics.

KEYWORDS:

Introduction:

In case pyogenic bacterial wound infection pus formation is pretty common – pus is a collection of dead white blood cells, dead bacteria along with necrotic tissue¹. Pus when collected in a closed space it is called abscess, on the other hand pus collection is visible within epidermis – it is called pustule or pimple². Different bacterial species are responsible for this pus formation amongst which the most common is *Staphylococcus* followed by *Klebsiella* species, *Escherichia coli*, *Pseudomonas*, and *Streptococci*. In 1880 in Aberdeen, Scotland, a surgeon, Sir Alexandu Ogston isolated gram positive cocci, *Staphylococcus aureus* from surgical abscess in knee joints. In 1881 he again demonstrated that this organism is responsible for purulent infection in case of any breakage of skin and epithelial linings. It is commonly present in nasal passage and skin; it is responsible for most common nosocomial infection and post-operative infection³. *Staphylococcus aureus* is coagulase positive and is responsible for local purulent infection in the skin (bullous impetigo), ear (otitis media), Breast (mastitis puerperalis), post influenza pneumonia, sepsis etc. Again its toxins are responsible for toxic shock syndrome, food poisoning and exfoliative dermatitis. In case of immunocompromized individual this organism will become very violent⁴. But now-a-days this organism started demonstrating resistance against many antibiotics. Again in most hospitals drug resistant "hospital strain" was responsible for many deaths in surgical units and newborn nurseries⁵. So, this antibiotic resistant strain (MRSA) ultimately increases the hospital stay, cost of hospital stay, cost of treatment. MRSA strain demonstrates resistance to beta lactams, tetracycline, and microlides and even to fluoquinolone⁶. Inappropriate and unnecessary use of antibiotics leads to development of emergence of antibiotic resistant pathogens, as a result new and newer drugs are needed. So, the best way to control drug resistance is to control inadvertent and improper use of drugs⁷. So, the aim of our study to find out the spectrum of antimicrobial sensitivity of *Staphylococcus aureus* isolated from pus in our tertiary care hospital, Kolkata.

Materials and Methods:

This study was conducted only after getting permission from local ethical committee. In this study 7531 pus samples were collected from skin ear, foot in patients with sepsis, diabetic foot etc. of K P C Medical College & Hospital, Jadavpur, Kolkata. from the year 2009 to 2016. Firstly the organism *Staphylococcus aureus* was isolated from the sample by gram staining. Then the pus samples were inoculated into blood agar as well as MacConkey agar plates and kept it in the

incubator at 37o C for 24 hours. When growth developed in the media isolates were identified by Gram stain, catalase test, coagulase test both slide and tube method. This was followed by inoculation of the sample into the plates by four flame method and was kept in the incubator for further 24 hours at 37oC⁸. By this method 291 samples were positive for *Staphylococcus aureus*. Then antimicrobial sensitivity of *Staphylococcus aureus* was done by Disc Diffusion Method of Bauer et al⁹. Then with the help of cotton swab the growth of the isolates was streaked on the surface of Mueller Hinton agar plates. These plates were allowed to dry before application of antibiotic disc¹⁰. Then commercially available antibiotic disc containing designated amount of antibiotics were placed firmly over the plates and left it for one hour at room temperature so that the antibiotics were diffused into agar medium. Here antibiotic disc used were from Himedia Labs, Mumbai, India. Now the plates were kept in the incubator for 24 hours at 37o C. Here antimicrobial activity was demonstrated as the zone of inhibition. After 24 hours the zone were measured by scale. Here it was recorded that more than 19 mm diameter as highly susceptible one, 15 – 18 mm as intermediate and less than 15 mm as resistant one. In this study zone of high sensitivity and intermediate sensitivity were considered.

Statistics:

In the three groups of staphylococci, incidence of positive occurrences compared between males and females as well as their probability value was demonstrated. In case of antimicrobial sensitivity testing percentage values were deducted. Software used was Statistical Package for the Social Sciences (SPSS) version 17.

Results:

Amongst 291 staphylococci positive samples 162 were MSSA, 74 MRSA and 55 CoNS. In all the three types of staphylococci males were predominantly affected than females ($p=0.00$) [Table 1]. In case of MSSA, Vancomycin, teicoplanin and linazolid, in case of MRSA teicoplanin and linazolid and in case of CoNS vancomycin, linazolid and tigicycline demonstrated more than 90% sensitivity. Again, MSSA demonstrated sensitivity to tigicycline, tetracycline, gentamicin, netilmicin, amikacin, piperacillin-tazobactam, MRSA netilmicin, amikacin and clindamycin and CoNS linazolid and tetracycline to more than 70%. Lastly, mild 50% to less than 70% sensitivity was found in case of fluoroquinolones and chloramphenicol in MRSA and MSSA, clindamycin and oxacillin in MSSA, tetracycline and tigicycline and gentamicin in MRSA, gentamicin, netilmicin, amikacin and levofloxacin in CoNS.

Table 1: Total cases of staphylococcus aureus isolated from pus:

Bacterial isolates	Total cases	Males	Percentage	Females	Percentage	P value
MSSA	162	101	62.345	61	37.354	0.00
MRSA	74	48	64.864	26	35.135	0.00
CoNS	55	36	65.454	19	34.545	0.00

Table 2: Antimicrobial sensitivity of MSSA, MRSA and CNS:

Antibiotics	MSSA (162)	Percentage	MRSA (74)	Percentage	CNS (55)	Percentage
Amoxicillin	37	22.84	0	0	15	27.27
Oxacillin	100	61.72	8	10.81	16	29.09
Ampicillin	88	54.32	9	12.16	15	27.27
PIPT	125	77.16	23	31.08	21	38.18
CES	7	4.32	5	6.76	0	0
Cefuroxime	78	48.15	4	5.40	12	21.81
Cefotaxime	4	2.47	2	2.70	5	9.09
Cefoxitin	15	9.26	1	1.35	6	10.90
Ceftazedime	5	3.08	0	0	2	3.63
Ceftriaxone	79	48.77	9	12.16	12	21.81
Cefepime	18	11.11	7	9.45	14	25.45
Azithromycin	69	42.59	11	14.86	8	14.54
Erythromycin	77	47.53	20	27.02	4	7.27
Aztreonam	23	14.19	9	12.16	5	9.09
Ertapenem	20	12.34	5	6.76	16	29.09
Imipenem	27	16.66	5	6.76	26	47.22
Meropenem	15	9.26	6	8.10	13	23.63
Gentamicin	130	80.24	43	58.10	31	56.36
Tobramycin	60	37.03	28	37.83	8	14.54
Netilmicin	125	77.16	52	70.27	32	58.18
Amikacin	127	78.39	54	72.97	29	52.72
Ciprofloxacin	92	56.79	44	59.45	20	36.36
Ofloxacin	96	59.26	40	54.05	22	40
Livofloxacin	113	69.75	42	56.75	33	60
Cotrimoxazole	78	48.15	20	27.02	27	49.09
Chloramphenicol	112	69.13	47	63.51	45	81.81
Tetracycline	128	79.01	47	63.51	44	80
Tigicycline	130	80.24	37	50	51	92.72
Clindamycin	99	61.11	53	71.61	22	40
Vancomycin	151	93.20	36	48.64	50	90.90
Teicoplanin	152	93.82	72	97.29	50	90.90
Linezolid	150	92.59	70	94.59	49	89.09
Polymyxin B	3	1.85	0	0	0	0
Colistin	0	0	0	0	0	0

PIPT: Piperacillin-Tazobactam, CES: Cefoperazone-sulbactam,

Discussion:

In the present study males were predominantly affected than females in all class of staphylococci (MRSA, MSSA, CoNS) ($p=0.00$). It may be due to that males are more exposed to any type of environment, close contact with the patients in the office or hospitals or outdoors of the different hospitals and at the same time low immunity, diabetes of exposed individual.

In the present study MRSA and MSSA were highly sensitive to aminoglycoside group of antibiotics except Tobramycin which was similar to the study found in the study done by Asati Rakesh Kumar in 201311. But CoNs demonstrated no sensitivity to these aminoglycoside antibiotics. This different type of sensitivity as well as resistance may be due to chromosomal mutation modifying different enzymes for producing resistance to staphylococci. Again in the present study all the classes of staphylococci were more than 88% sensitive to linezolid similar to the study done by Rakesh Kumar in 201311.

In the present study all classes of staphylococci demonstrated little sensitivity to semi synthetic penicillin and all generations of cephalosporins, near-resistance to carbapenem group of drugs, and complete resistance to most recent drugs like, polymyxin B and colistin. This was similar to the study of Rakesh Kumar in 201311. This may be due to misuse, inadvertent use and extensive use of these antibiotics. In case of penicillin and cephalosporin this resistance is due to emergence of beta-lactamase producing staphylococcus as well as "selection pressure" due to extensive use of this semi synthetic penicillin. Now-a-days betalactamase inhibitors are added with penicillins and cephalosporins to reduce the resistance as well as toxicity of these drugs^{11, 12, 13}.

The present study also demonstrated very high sensitivity to vancomycin, linezolid and teicoplanin. His was similar to the study done by Qureshi AH et al and Majeed MT et al^{14, 15}. This is contrary to the study done by Wang SH et al and Tentolouris N et al where the resistance to these antibiotics was demonstrated as 15% -- 30%.^{16, 17} This highest sensitivity may be due to very less use of these drugs, which in turn may be due to high cost of them. So the patients are very much less exposed to these drugs as a result sensitivity remains high.

In the present study MRSA was 90% resistance to semi synthetic penicillin and methicillin group of antibiotics. Whereas 34.6% and 65.4% staphylococcus aureus demonstrated resistance to oxacillin and penicillin respectively found in the study of Hailu et al and nearly similar results were found in the study done by Hwang et al, Abera et al, and Seid et al.^{18, 19, 20, 21}.

The present study demonstrated 42% to 87% resistance to chloramphenicol, ciprofloxacin, erythromycin, co-trimoxazole to MRSA, MSSA and CoNS which was contrary to the study done by Hailu et al, Seid et al and Abera et al^{18, 20, 21}.

Conclusion:

MRSA, MSSA and CoNS demonstrated highest sensitivity to vancomycin, teicoplanin, linezolid, MRSA and MSSA were moderately sensitive to aminoglycoside antibiotics, MSSA to vancomycin and tetracycline and tigicycline, MRSA to clindamycin and CoNS to chloramphenicol and tetracycline. All of them demonstrated resistance to cephalosporins, semi synthetic penicillins and polymyxin B and colistin. This resistance is mainly due to extensive inadvertent and illogical use of antibiotics in outdoor or hospital. Again these organisms will become mutant and develop resistant strains against these antibiotics. So to prevent the emergence of extensive resistant strains the internists should be cautious before prescribing antibiotics.

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