

Current trends of antibiotic resistance in clinical isolates of *Staphylococcus aureus*

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Abstract *Staphylococcus aureus* (*S. aureus*) is a well known human pathogen known to causes a verity of infections in humans. In recent years *S. aureus* is reported to show drug resistant toward commonly known drugs. Therefore, this study was designed to study the pattern of antibiotic resistance in 50 clinical isolates of *S. aureus* isolated at Dhanwantri Hospital and Research Centre, Jaipur, Rajasthan, India. *S. aureus* cultures were isolated from different clinical samples, pus, throat swabs and urine on Blood agar and MacConkey agar and Chrom agar plats and characterized by an array of microscopic and biochemical tests. Antibiotic sensitivity test was performed by standard disc diffusion method (Kirby bayer's method) on Muller Hinton agar plates. During this study, among 50 *S. aureus* isolates 48 (96%) were found to be resistance toward Aztreonam and Doxycycline followed by Ciprofloxacin ($n = 45, 90\%$), Cefpodoxime and Ceftazidime ($n = 44, 88\%$), Cefuroxime ($n = 40, 80\%$), Piperacillin + Tazobactam ($n = 38, 76\%$), Cefoparazone ($n = 36, 72\%$), Amoxicillin + Clavulanic acid and Ceftriaxone ($n = 33, 66\%$), Levofloxacin ($n = 32, 64\%$), Moxifloxacin ($n = 31, 62\%$), Ofloxacin ($n = 25, 50\%$), Cloxacillin ($n = 22, 44\%$), Azithromycin ($n = 21, 42\%$), Clindamycin ($n = 19, 38\%$), Meropenem ($n = 18, 36\%$), Clarithromycin ($n = 16, 32\%$), Ampicillin + sulbactam ($n = 13, 26\%$), Amikacin ($n = 12, 24\%$), Impipenem ($n = 8, 16\%$), Linezolid and Methicillin ($n = 7, 14\%$) and Teicoplanin ($n = 3, 6\%$). In conclusion, the isolated *S. aureus* found to be resistant toward common antibiotics, however all isolates were found to be susceptible to Vancomycin.

Keywords *Staphylococcus aureus*, infections, antibiotic resistance

Introduction

Microorganisms are one of the major threats to mankind and microbial infections are the leading cause of motility worldwide. According to the World Health Organization (WHO), microbial infections collectively resulted in 25% of death worldwide (WHO, 1999). *S. aureus* is a Gram positive cocci commonly associated with several clinical conditions. Humans are the natural reservoir and the infection is mostly asymptomatic (Chambers, 2001), however *S. aureus* is also reported to cause the symptomatic infection of skin and soft tissues such as abscesses (boils), carbuncles, hidradenitis suppurativa, folliculitis, impetigo, furuncles, and cellulitis, sore throat. Glomerulonephritis, food poisoning, lymphade-

nitis, toxic shock syndrome, osteomyelitis, pneumonia, meningitis, endocarditis and bacteremia are few other complications associated with *S. aureus* infections (Bamberger and Boyd, 2005).

Generally, *S. aureus* infections can be treated by the application of flucloxacillin, dicloxacillin, cephalosporins (cefazolin, cephalothin and cephalexin), clindamycin, lincomycin and erythromycin, vancomycin, teicoplanin, combination of rifampicin and fusidic acid, lincosamides (clindamycin, lincomycin) or cotrimoxazole. New antibiotics such as linezolid and quinupristin/dalfopristin can also be used but are very costly (Rayner and Munckhof, 2005). However in last few decades, reports of microbial drug resistance in *S. aureus* have been documented all around the world (Livermore, 2000; Bal and Gould, 2005; Pantosti et al., 2007). Emergence of the drug resistance could be attributed to the unrestricted and overuse use of antibiotics in a particular environment. Infection with these resistant strains of *S. aureus* is expected to cause more severe disease and may

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require longer treatment period than infection with susceptible strains.

A routine check of antibiotic sensitivity pattern could help to understand the emergence of drug resistance in microorganisms and may help in deciding the drug of choice for the treatment of infection. Therefore, the aim of the present study is to record the antibiotic sensitivity pattern of various antibiotics toward the *S. aureus* isolated from the clinical samples. This study also highlights the need for continuous surveillance of antibiotic sensitivity pattern of *S. aureus* with a view to selecting appropriate therapy.

Material and methods

Chemicals and media

Blood agar base, MacConkey agar, Chrom agar, Muller Hinton agar, Amikacin (AK), Amoxicillin + clavulanic acid (AMC), Ampicillin + sulbactam (A/S), Azithromycin (AZM), Aztreonam (AT), Cefoparazone (CPZ), Cefpodoxime (CPD), Ceftazidime (CAZ), Ceftriaxone (CTR), Cefuroxime (CXM), Ciprofloxacin (CIP), Clarithromycin (CLR), Clindamycin (CD), Cloxacillin (Cox), Doxycycline (DO), Impipenem (IPM), Levofloxacin (LE), Linezolid (LZ), Meropenem (MRP), Methicillin (MC), Moxifloxacin (MO), Ofloxacin (OF), Pipracillin + Tazobactam, (PIT), Teicoplanin (TEI), Vancomycin (VA) discs were purchased from Himedia Pvt Ltd, Mumbai, India. All other chemical used during the study were of analytical grade.

Antibiotic sensitivity test

Samples

A total of 50 isolates of *S. aureus* were isolated from different clinical samples, pus, throat swabs, urine and sputum on Blood agar and MacConkey agar and Chrom agar plates at Dhanwantri Hospital and Research Centre, Jaipur, Rajasthan, India during year 2013. The cultures were characterized microscopically by Gram's staining and biochemically by catalase and coagulase test.

Assay

The 50 *S. aureus* cultures were screened for their sensitivity toward 25 standard antibiotics. Antibiotics included Amikacin (30 µg/disc), Amoxicillin + clavulanic acid (AMC), Ampicillin + sulbactam (A/S), Azithromycin (AZM), Aztreonam (AT), Cefoparazone (CPZ), Cefpodoxime (CPD), Ceftazidime (CAZ), Ceftriaxone (CTR), Cefuroxime (CXM), Ciprofloxacin (CIP), Clarithromycin (CLR), Clindamycin (CD), Cloxacillin (Cox), Doxycycline (DO), Impipenem (IPM), Levofloxacin (LE), Linezolid (LZ),

Meropenem (MRP), Methicillin (MC), Moxifloxacin (MO), Ofloxacin (OF), Pipracillin + Tazobactam, (PIT), Teicoplanin (TEI), Vancomycin (VA). Antibiotic sensitivity test was performed by disc diffusion method on Muller Hinton agar (MHA) plates. *S. aureus* isolates were swabbed on Mueller Hinton agar plates by using sterilize cotton swabs. The antibiotic discs were placed on the agar surface using a sterilize forceps. Plates were incubated at 37°C for 24 h. Plates were observed for zone of inhibition (Iqbal et al., 2004).

Results and discussion

S. aureus is a common reason of infectious disease in hospitals and is most liable to infect new born babies, surgical patients, old and malnourished persons and patients with diabetes and other chronic diseases. The patients admitted in hospitals with impaired immunity are at highest risk of getting infection with *S. aureus*. Even though several antibiotics are in use to control *S. aureus* infection, they failed to control the associated morbidity and mortality. This failure of antibiotics to efficiently control *S. aureus* could be attributed to the emergence of drug resistance in microorganisms. It had been observed that the excessive use of antibiotics can trigger the development of drug resistance in microorganisms. During this study, 50 samples of *S. aureus* were isolated from the pus, throat swabs and urine specimens collected from admitted patients. Among 50 *S. aureus* isolates, 48 (96%) were found to be resistance toward Aztreonam and Doxycycline followed by Ciprofloxacin ($n = 45$, 90%), Cefpodoxime and Ceftazidime ($n = 44$, 88%), Cefuroxime ($n = 40$, 80%), Pipracillin + Tazobactam ($n = 38$, 76%), Cefoparazone ($n = 36$, 72%), Amoxicillin + Clavulanic acid and Ceftriaxone ($n = 33$, 66%), Levofloxacin ($n = 32$, 64%), Moxifloxacin ($n = 31$, 62%), Ofloxacin ($n = 25$, 50%), Cloxacillin ($n = 22$, 44%), Azithromycin ($n = 21$, 42%), Clindamycin ($n = 19$, 38%), Meropenem ($n = 18$, 36%), Clarithromycin ($n = 16$, 32%), Ampicillin + sulbactam ($n = 13$, 26%), Amikacin ($n = 12$, 24%), Impipenem ($n = 8$, 16%), Linezolid and Methicillin ($n = 7$, 14%) and Teicoplanin ($n = 3$, 6%). All 50 isolates of *S. aureus* were found to be sensitive toward vancomycin. Results are summarized in Table 1 and graphically represented in Fig. 1.

Results of this study are representing the increase in incidents of drug resistance in the clinical isolates of *S. aureus* which is an issue of concern. Among 50 *S. aureus* isolates, 7 isolates were found to be resistance toward Methicillin which is an alarming issue in healthcare industry. A routine screening of microorganism for their antibiotic sensitivity pattern could significantly help to establish the suitable antibiotic and its therapeutic dose, which eventually reduces the chances of getting drug resistance in microorganisms.

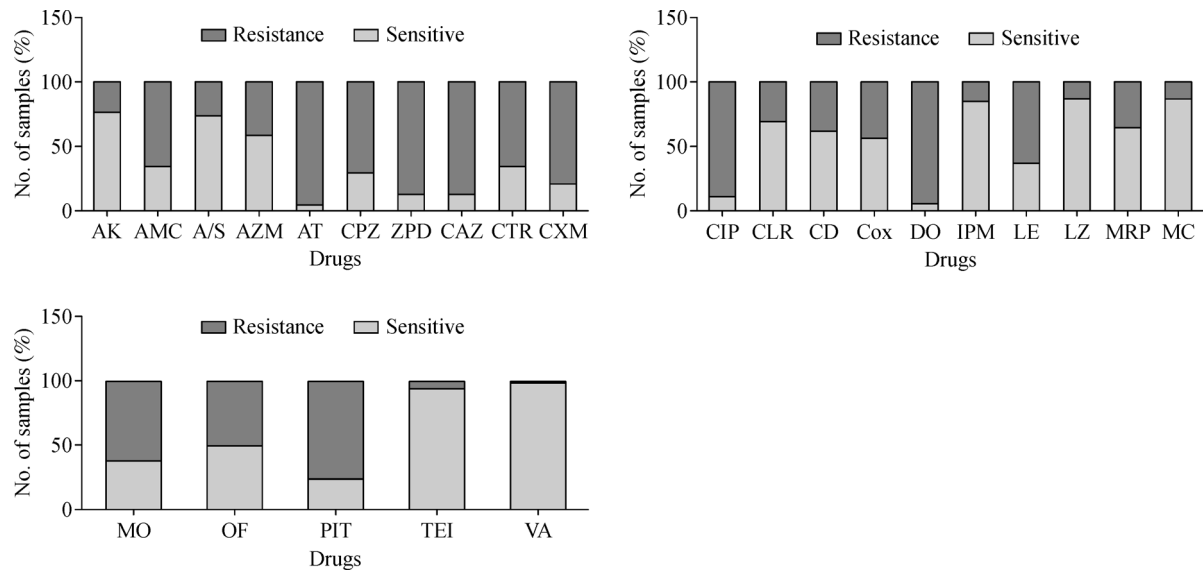


Figure 1 Antimicrobial resistance in *S. aureus*. Here, Total number of samples used in study was 50; Amikacin (AK), Amoxicillin + clavulanic acid (AMC), Ampicillin + sulbactam (A/S), Azithromycin (AZM), Aztreonam (AT), Cefoparazone (CPZ), Cefpodoxime (CPD), Ceftazidime (CAZ), Ceftriaxone (CTR), Cefuroxime (CXM), Ciprofloxacin (CIP), Clarithromycin (CLR), Clindamycin (CD), Cloxacillin (COX), Doxycycline (DO), Impipenem (IPM), Levofloxacin (LE), Linezolid (LZ), Meropenem (MRP), Methicillin (MC), Moxifloxacin (MO), Ofloxacin (OF), Pipracillin + Tazobactam, (PIT), Teicoplanin (TEI), Vancomycin (VA).

Table 1 Antimicrobial resistance in *S. aureus*

Antibiotics	Levels	Dose ($\mu\text{g}/\text{disc}$)	Sensitive		Resistance		Total
			N	%	N	%	
Amikacin	AK	30	38	72	12	24	50
Amoxicillin + clavulanic acid	AMC	10	17	34	33	66	50
Ampicillin + sulbactam	A/S	10	37	74	13	26	50
Azithromycin	AZM	15	29	58	21	42	50
Aztreonam	AT	30	2	4	48	96	50
Cefoparazone	CPZ	75	14	28	36	72	50
Cefpodoxime	CPD	10	6	12	44	88	50
Ceftazidime	CAZ	30	6	12	44	88	50
Ceftriaxone	CTR	30	17	34	33	66	50
Cefuroxime	CXM	30	10	20	40	80	50
Ciprofloxacin	CIP	05	5	10	45	90	50
Clarithromycin	CLR	15	34	68	16	32	50
Clindamycin	CD	02	31	62	19	38	50
Cloxacillin	Cox	30	28	56	22	44	50
Doxycycline	DO	30	2	4	48	96	50
Impipenem	IPM	10	42	84	8	16	50
Levofloxacin	LE	05	18	36	32	64	50
Linezolid	LZ	30	43	86	7	14	50
Meropenem	MRP	10	32	64	18	36	50
Methicillin	MC	05	43	86	7	14	50
Moxifloxacin	MO	30	19	38	31	62	50
Ofloxacin	OF	05	25	50	25	50	50
Pipracillin + Tazobactam	PIT	10	12	24	38	76	50
Teicoplanin	TEI	30	47	94	3	6	50
Vancomycin	VA	30	50	100	0	0	50

Methicillin-resistant *Staphylococcus aureus* (MRSA) is one of the major cause of morbidity and mortality when compare to the other *S. aureus* infections. The resistance pattern of *S. aureus* has underscored the need for new antimicrobial drugs. It is now migrated into the community. This strain shared some characteristics features with nosocomial strains due to this it antimicrobial susceptibility and potential virulence will vary (Appelbaum, 2006).

This study was alarming the resistance of *S. aureus* and also the diminishing efficacy of antimicrobial agents for the treatment of *S. aureus* infections. Still new drug exist, its shelf-life is likely to be increasingly limited. Hence, new approaches to treatment and prevention will become more and more important, especially with the diminishing availability of new “wonder drugs” (Lowy, 2003).

Conclusion

This study draws attention on the need of routine surveillance of antibiotic sensitivity pattern of *S. aureus* with a view to selecting appropriate therapy. The results of this study showed that the Vancomycin is the most effective antibiotic to control *S. aureus* infection. In addition Teicoplanin also demonstrated the promising results.

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Compliance with ethics guidelines

Kapil Dev Sharma, Rajendra Prasad Saini, Loganathan Karthik declare that they have no conflict of interest. This article does not contain any studies with human or animal subjects performed by any of the authors.

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