

ORIGINAL RESEARCH

STAPHYLOCOCCUS AUREUS ANTIMICROBIAL RESISTANCE
PROFILE ISOLATED FROM HOSPITAL KARBALA, IRAQ

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ABSTRACT

Staphylococcus aureus is an acute pathogen; therefore, increasing antibiotic resistance within S. aureus strains is becoming a global issue. And serious nosocomial infections. The current study aims to define resistance collective prevalence to patterns for 21 different agents of antibiotics antimicrobial by S. aureus in hospital Karbala-Iraq. Among 150 different clinical specimens, from sputum swabs wound swabs, and throat swabs (45,55 & 50), respectively, total isolation (n=100) of S. aureus were cultured for isolation by the usual techniques from September 2020 to January 2021. Using the Kirby Bauer disc diffusion method, isolates were tested for sensitivity to a panel of (21) antimicrobials. The isolates appeared (100 %)resistant to ampicillin, clavulanic acid (95%) azithromycin (90%) oxacillin (85%), clindamycin(80%),cefoxitin(78%),tetracycline (75%), bacitracin (70%), cephalothin (68%), vancomycin (66%), methicillin (65%),sulfamethoxazole (64%),erythromycin (60%),trimethoprim (58%),Nitrofurantoin(55%),teicoplanin (53%), gentamycin(52%), Doxycycline (50%), Kanamycin (45%), levofloxacin (40 %) and norfloxacin (35 %). S. aureus isolates from patients in demonstrated resistance to most antimicrobials frequently used to treat staphylococcal infections, highlighting the critical necessity of antimicrobial prudence and the implementation of genuine infection control methods to prevent the spread of S. aureus.

Keywords:*Antimicrobial,isolated, Iraq,resistance, profile, Staphylococcus aureus*

INTRODUCTION

According to Brooks (2001), Staphylococcus aureus is a gram-positive bacterium that produces a range of extracellular enzymes and toxins and thus is coagulase positive. S. aureus is a natural flora associated with the skin, mucous membranes, and skin glands of practically all warm-blooded species, and it colonises around 30% of the human population. It is also a primary source of S. aureus infection, which can result in suppuration, development of abscesses, a range of pyogenic infections, and potentially deadly septicaemia (Tong *et al.*2015). Furthermore, S. aureus is among the most infamous gramme-positive bacterial pathogens due to its capacity to infiltrate the human immune system via numerous virulence factors and its rapid development of a multi-drug resistant phenotype (Davies *et al.*2010).

Likewise, in patients suffering from otitis media in Western Erbil-Iraq Emergency and Rizgary Medical Centres, S. aureus is the most isolated pathogens (Aziz *et al.*2021). S. multidrug-resistant aureus strains, primarily methicillin-resistant. Unless there is a specific infection control policy, S. aureus can spread rapidly in a specific health facility through colonised or sick patients or healthcare providers and contaminated settings (Kesah *et al.* 2003). Resistance to antibiotics, which affects over two million people yearly, is one of the world's most serious problems.

Resistance to antibiotics among *S. aureus* species to beta-lactam antibiotics has caused significant challenges in managing their associated illnesses. Despite significant efforts to limit antibiotic resistance, methicillin-resistant *S. aureus* methicillin-resistant is increasing globally, and regional and local variables impact its dynam catastrophe (Medina et al.2018).

The widespread use of antimicrobials in hospitals for infection prevention or treatment resulted in a proliferation of resistant bacteria. Around 10–90% of antibiotics are excreted in patients' faeces and urine as unaltered or metabolites (Berkner et al. 2014). As a result, antimicrobial resistance among microorganisms is becoming a global health concern. Antibiotic overuse has resulted in 23,000 deaths because of antibiotic-resistant bacterial infections (CDC, 2013). Antibiotic-resistant infections can occur anywhere in the population, but nosocomial infections are significantly more common. Bacterial resistance is a critical issue because when bacteria develop and find ways to nullify antibiotics, the antibiotics become ineffective, allowing these harmful organisms to persist in various conditions (Margonis, 2018).

Simplifying more suitable treatment selections, minimalizing the illness and humanity due to the infection resistance and preserving the efficiency of antimicrobials require synthesis and indication concerning antimicrobial resistance in a country. Evidence that has been adequately summarised and synthesised is required for revising global clinical guidelines. To our knowledge, no prior meta-analysis or systematic analysis of *S. aureus* AMR to all antimicrobials routinely used in Ethiopia has been undertaken. Assumed from the best-known data, this investigation aimed to establish the combined prevalence of *S. aureus* resistance to major antimicrobial drugs in Iraq.

MATERIALS AND METHOD

Isolates Collected: among 150 different clinical specimens from sputum swabs, wound swabs and throat swabs (45,55 & 50), respectively. Total isolation (n=100) *S. aureus* isolation was cultured according to standard procedures from September 2020 to January 2021. In the laboratory testing, all demographic information such as infections, location of specimen isolation, ages, and gender of patients were recorded.

Diagnosis:

Isolation of Bacteria: the techniques suggested by Cheesbrough (2006) suggested that all isolates were confirmed as *S. aureus* after being put onto mannitol salts agar and brood for twenty-four to forty-eight hours at 37°C. The colonies' size, form, colour, texture, and borders were used to identify them. Gram stain was used to detect colonies with similar morphological aspects as *S. aureus*. Specimens were subjected to biochemical analysis such as (Coagulase, Urease, Catalase, Nitrate reduction, Vogas -Proskaurtests and Citrate utilization). Following the agreement, the samples were placed in medium Tryptic soy broth (TSB), which included thirty per cent glycerol at -20o C till further use.

Antibiotic sensitivity test

The antibiotics such as ampicillin, oxacillin, azithromycin, clindamycin, cefoxitin, tetracycline, bacitracin, cephalothin, vancomycin, methicillin, sulfamethoxazole, erythromycin, trimethoprim, Nitrofurantoin, teicoplanin, levofloxacin, gentamycin, Doxycycline, Kanamycin and norfloxacin were applied strain of bacteria sensitive *S. aureus* then cultivated in nutritional broth surroundings over eighteen hours at $2 \pm 37^{\circ}\text{C}$. A UV-vis spectrophotometer was used to test the detection limit of the medium containing, which was corrected to 1.0 OD on 600 nm and employed as

inoculation. Antibacterial test using Mueller-Hinton agar, the antibacterial activities of essential oils, was investigated using the diffusion technique (India, Mumbai, Himedia).

In nutritive broth media, freshly cultivated *S. aureus* strains were used in the antibacterial experiment to achieve consistent microbial increase; a millilitre of culture was introduced to Mueller-Hinton agar. Its oil extracted was poured on an MHA plate after being deposited onto a sterile unfilled antimicrobial disc (20 mL). The growth plates were then incubated at 4 degrees Celsius for around 30 minutes before being moved to an incubator at 37 degrees Celsius. Throughout 24 hours of incubation, an inhibition zone was assessed (mm) (Bindhu et al.2020). The isolates were divided into resistant, intermediate, and susceptible groups. For quality control, *S. aureus* ATCC strain 34662 was employed. If such an isolate was sensitive and intermediate to the antibiotic, it was classified as non-susceptible. MDR was identified by a lack of sensitivity to at least three types of antibiotics.

RESULTS AND DISCUSSION

S. aureus Identification and Isolation

One hundred isolates were found in 150 different clinical samples, counting swabs, throat swabs and wound swabs (45,55 and 50), and total isolation (n=100) of *S. aureus*. The isolates were identified using a traditional approach based on cell morphology, culture features, the VITEK 2 compact systems, and Gram stain response biochemical properties. The isolates could develop on mannitol salt agar. *S. aureus* does have the potential to alter the colouring of the medium between pink, orange to yellow by fermenting the mannitol contained in the medium (Morello et al.2003) round, smooth, yellow to golden colony formed on blood agar with different degrees of haemolysis (beta haemolysis mostly). Under a microscopic examination, smears of *S. aureus* isolates were purple singles, Diplo, with grape-like gramme positive cocci. Catalase, DNase, and coagulase were all found in all the isolates. The VITEK2 condensed system recognised all isolates as *S. aureus* with a likelihood of more than 95%. Twenty isolates were found amongst Fifty distinct clinical specimens in research by (Aziz et al.2021): Twenty burn swabs, Ten wound swabs, ten urines, and ten from dental caries). When all participants with *S. aureus* colonisation were examined, 34 were contaminated with MRSA and 124 with methicillin-sensitive *S. aureus* (MSSA) (Al-Talib et al., 2015).

Antibiotics susceptibility testing was performed using the disc diffusion technique on 100 *S. aureus* isolates versus 21 routinely used antibiotics ampicillin (100%), clavulanic acid (95%) azithromycin (90%)oxacillin(85%),clindamycin(80%),cefoxitin(78%),

tetracycline (75%), bacitracin (70%), cephalothin (68%),vancomycin (66%), methicillin (65%),sulfamethoxazole (64%),erythromycin (60%),trimethoprim (58%),Nitrofurantoin(55%),teicoplanin (53%), gentamycin(52%), Doxycycline(50%), Kanamycin (45%), levofloxacin (40 %) and norfloxacin (35 %) Figure 1.

As indicated in Table 1, the resistance per cent of *S. aureus* samples differed from the antibiotics employed in this research. Antibiotic-resistant rate for all other antibiotics were 65% for CAZ, 60% for E, 65% for TM, 60% for T, 50% for CRO, S, 45% for L, 45% for CTX, 40% for RA, 35% for CIP, 40% for DA, 35% for C, 10 % for GM and 15% for KF, but all samples were vancomycin-resistant. Our findings are consistent with those of (Al-Jebouri et al. 2013), who discovered that *S. aureus* samples from UTIs were significantly resistant to both amoxicillin and ampicillin. According to Al-Ugaili (2014), *S. aureus* resistance samples were: norfloxacin (16 %),

for levofloxacin (20 %), for ofloxacin (18 %), for ciprofloxacin (16 %), for nalidixic acid (50%), for levofloxacin (14%) the [18] results revealed that: -

- (86.48 %) of isolates were resistant to ampicillin.
- (8.10%) of *S. aureus* isolates were resistant to amikacin.
- (54.05 %) were resistant to cephotaxim, tetracycline and erythromycin.
- (100%) of isolates were resistant to amoxicillin.
- (10.81%) were resistant to vancomycin.
- (21.62%) of isolates were resistant to methecillin (MRSA).

Table 1 shows the percentage of *S. aureus* that is antibiotic resistance

Antimicrobial agent	Antibiotics	% of resistance	No. of resistant isolates
Ampicillin	Amp	100	100
Oxacillin	OX	95	95
Clavulanic Acid	CA	90	90
Azithromycin	AZ	85	85
Clindamycin	CL	80	80
Cefoxitin	CEF	78	78
Tetracycline	T	75	75
Bacitracin	B	70	70
Cephalothin	CE	68	68
Vancomycin	VA	66	66
Methicillin	ME	65	65
Sulfamethoxazole	SUL	64	64
Erythromycin	E	60	60
Trimethoprim	TR	58	58
Nitrofurantoin	NIT	55	55
Teicoplanin	TEI	53	53
Gentamycin	G	52	52
Doxycline,	DO	50	50
Kanamycin	K	45	45
Levofloxacin	LE	40	40
Norfloxacin	NO	35	35

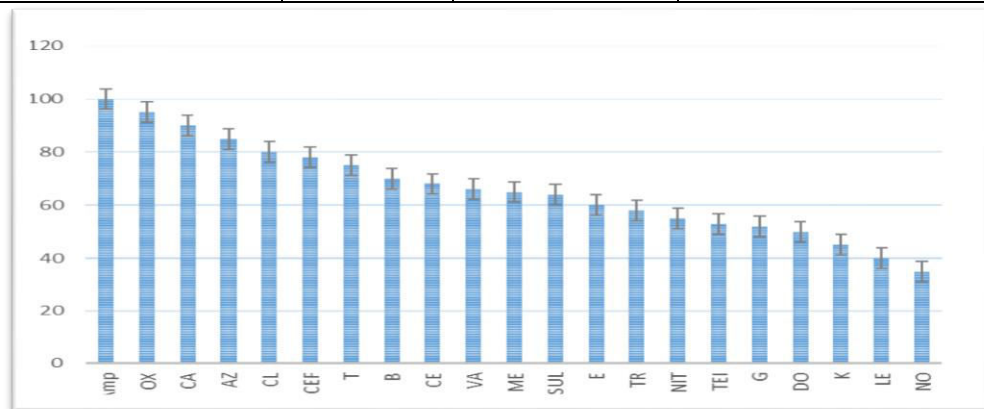


Figure 1. resistance *Staphylococcus aureus* to different antibiotics in Iraq hospitals

CONCLUSION

Because it produces many virulence factors involved in colonisation and the invention of the host, *Staphylococcus aureus* is a significant pathogen that has led to the severity of human infection. In addition, *S. aureus*, we may determine that Amoxicillin resistance was 100 per cent.

REFERENCE

1. Tong SY, Davis JS, Eichenberger E, Holland TL, Fowler VG Jr. *Staphylococcus aureus* infections: epidemiology, pathophysiology, clinical manifestations, and management. *Clin Microbiol Rev.* 2015;28(3):603–61.
2. Davies J, Davies D. Origins and evolution of antibiotic resistance. *Microbiol Mol Biol Rev.* 2010;74(3):417–33
3. Aziz, K. E., & Abdulrahman, Z. F. A. (2021, May). Detection of Tetracycline tet (k) Gene in Clinical *Staphylococcus aureus* Isolates. In *IOP Conference Series: Earth and Environmental Science* (Vol. 761, No. 1, p. 012128). IOP Publishing.
4. Kesah C, Ben Redjeb S, Odugbemi TO, Boye CS, Dosso M, Ndinya Achola JO, Koulla-Shiro S, Benbachir M, Rahal K, Borg M. Prevalence of methicillin-resistant *Staphylococcus aureus* in eight African hospitals and Malta. *Clin Microbiol Infect.* 2003;9(2):153–6
5. Centres for Disease Control and Prevention (2013). Antibiotic resistance threats in the United States, 2013. Centres for Disease Control and Prevention, US Department of Health and Human Services.
6. Margonis GA, Buettner S, Andreatos N, Kim Y, Wagner D and Sasaki K et al. (2018). Association of BRAF in health care setups of Karachi. *Pak. J. Pharm. Sci.*, 30(6): 2417-2421.
7. Medina Cruz D, Mi G, Webster TJ. Synthesis and characterization of biogenic selenium nanoparticles with antimicrobial properties made by *Staphylococcus aureus*, methicillin-resistant *Staphylococcus aureus* (MRSA), *Escherichia coli* and *Pseudomonas aeruginosa*. *J Biomed Mater Res Part A.* 2018;106:1400–12.
8. Berkner S, Konradi S, Schönfeld J. Antibiotic resistance and the environment—there and back again. *EMBO Rep.* 2014;15 (7):740–744. doi:10.15252/embr.2014-38978
9. Cheesbrough M. District laboratory practice in tropical countries. Cambridge: Cambridge University Press. 2006.
10. Bindhu MR, Umadevi M, Esmail GA, Al-Dhabi NA, Arasu MV. Green synthesis and characterization of silver nanoparticles from *Moringa oleifera* flower and assessment of antimicrobial and sensing properties. *J Photochem Photobiol B: Biol* 2020;205:111836, <http://dx.doi.org/10.1016/j.jphotobiol.2020.111836>.
11. Morello, JA, Granato, PA and Mizer, HE 2003, Laboratory Manual and Workbook in Microbiology Applications to Patient Care. 7th ed., McGraw-Hill company. New York, USA.
12. Al-Talib, H., Al-Khateeb, A., & Hassan, H. (2015). Antimicrobial resistance of *Staphylococcus aureus* isolates in Malaysian Tertiary Hospital. *International Medical Journal*, 22(1), 1-3.
13. Al-Jebouri, MM and Mdish, SA 2013, Antibiotic resistance pattern of bacteria isolated from patients of urinary tract infections in Iraq. *Open Journal of Urology*, 3(2), 8-14.
14. Al-Ugaili, D, Fadhil, AMA and Wohaieb, SA 2014, Comparison of Oxacillin disc diffusion test with *mecA* polymerase chain reaction and cefoxitin disc diffusion test for the detection of oxacillin-resistant *Staphylococcus aureus* collected from Baghdad hospitals. *Journal of Al-Nahrain University*, 17(2), 172-180.