



# **Prevalence and Antimicrobial Resistance Pattern of Community Acquired *Staphylococcus aureus* in Patients Received at the Traumatology Unit of a Secondary Referral Health Setting in the Western Cameroon**

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## **Authors' contributions**

*This work was carried out in collaboration among all authors. Authors TDT and VNM conceived and designed the study, Authors VNM, ACNS and TDT were responsible for data collection and biological analyses. Authors TDT, ILSN, VRN and MOK analyzed the data. Authors TDT, ILSN, ACNS and wrote the first draft of the manuscript. Authors VRN, MJPC, MOK: Provided critical reading of the first draft. All authors read and approved the final manuscript.*

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## ABSTRACT

**Background:** *Staphylococcus aureus* is responsible of a wide range of both community and Hospital acquired infections. Several genomic variability underlie the diversity of *S. aureus* strains responsible for the emergence of antibiotic resistance.

**Objective:** To assess the prevalence and antibiotic resistance pattern of community acquired *S. aureus* isolated from pus samples in patients received at the traumatology unit of Our Lady of Health Centre of Batseng'la in Dschang, Western Cameroon.

**Study Design:** This was a descriptive cross-sectional study, carried out over a period of five months (from January to May 2021) involving a total of 52 participants received at the traumatology unit of Our Lady of Health Centre of Batseng'la in Dschang, Western Cameroon.

**Methods:** Pus samples collected from participants were seeded on Chapman Agar at 37°C for 24 hours. The colonies identification was based on catalase, coagulase and DNase tests. The antibiotic susceptibility test was performed using the Kirby Bauer disk diffusion method on Mueller-Hinton agar. Data were analysed using SPSS version 25 Software.

**Results:** *S. aureus* was isolated in 22 of the 54 pus samples analysed, giving a positivity rate of 42.3%. The majority of strains (63.6%) were isolated from subjects less than 21 years old. From the strains isolated, 40.9% were MRSA and 86.4% presented multi-resistance patterns to the antibiotics tested. All strains of MRSA were found to be cross-resistant with one or more other antibiotics.

**Conclusion:** The multi-resistance of community acquired *S. aureus* to antibiotics is a reality. Adequate care should be taken while handling suppurating wounds and abscesses. Especially in younger age individuals, as this may help in timely setting up proper care and treatment protocols necessary to overcome drug resistance of such extremely flexible pathogens.

**Keywords:** *Staphylococcus aureus*; MRSA; antimicrobial resistance; MDR; traumatology.

## 1. INTRODUCTION

Improvements in global health have been hampered in recent decades due to increased resistance to a wide range of antimicrobials continuously observed with microorganisms responsible for the majority of human infectious diseases [1]. Today we speak of ultra and pan drug resistance to signify the magnitude of this ever growing threat [2]. Despite the development of new antimicrobial molecules, *Staphylococcus aureus*, a Gram-positive Coccus, remains within the range of bacteria exhibiting most of these resistances in various medical conditions [3–6]. This bacterium is responsible for both community [7] and hospital acquired [8,9] infections. Studies have shown a predominance of methicillin-resistant *S. aureus* (MRSA) strains in infected wounds [10,11], nosocomial [12,13], nasal [9,13] infections, Osteomyelitis in children and premature infants [14,15] and pyomyositis [16].

Numerous genomic variations underlie this diversity of *S. aureus* responsible for the emergence of multi-drug resistance [17]. The

resistance of staphylococci to oxacillin/methicillin is due to the binding of the modified protein PBP2a, encoded by the mutant *mecA* gene to  $\beta$ -lactams [18]. Conjugative transfer of the *vanA* gene from vancomycin-resistant *Enterococcus faecalis* to MRSA [19] as well as the thickening of the bacterial cell wall due to a large accumulation of peptidoglycan [20] are major contributors to the resistance of *S. aureus* to vancomycin. A range of existing diagnostic methods and others undergoing development are available for a point of care and accurate detection of pathogenic bacteria and their resistance profile [21].

MRSA infections are more common and devastating in people with co-morbidities such as human immunodeficiency virus (HIV) infection [22] and diabetes [23].

Abdoulaye et al. [24] observed an increased resistance of *S. aureus* to antibiotics during surgical site infections in the surgical wards. Other studies also reported considerable resistance rates in hospital acquired infections including nosocomial infection [12] and nasal

carriage in intensive care units [9]. Data on community acquired *S aureus* infections are still very limited and completely inexistent in the locality. The objective of the study was to highlight the prevalence and antibiotic resistance pattern of community acquired *S. aureus* in patients received at the traumatology unit of Our Lady of Health Centre of Batseng'la (OLHCB) in Dschang, Western Cameroon.

## 2. MATERIALS AND METHODS

### 2.1 Study Design and Duration

This was a descriptive cross-sectional study, carried out over a period of five months, from January to May 2021.

### 2.2 Study Frame and Participants

The study was conducted at OLHCB, a confessional Hospital located in the Dschang Health District. This is the only hospital equipped with a traumatology unit and serving as a reference in the locality were consecutively included in the study, all subjects with suppurating wounds or abscesses as primary cause of admission at the traumatology unit of the Hospital who gave their free informed consent. A total of 52 pus samples were collected from the participants and analyzed in the Microbiology laboratory unit.

### 2.3 Specimen Collection and Biological Analyses

#### 2.3.1 Culture Media preparation

Chapmann, DNase and Mueller-Hinton agars (Liofilchem srl, Italy) were prepared according to the manufacturer's instructions. The sterility test was performed by incubating 5% of the prepared culture media at 37°C for 24 hours and examined for any bacterial growth. Plates were stored at 2 – 8°C. Prior to specimen collection, a Chapman Agar plate was placed to dry in an incubator at 37°C for 10 – 15 minutes.

#### 2.3.2 Specimen collection

Pus samples were aseptically collected either by swabbing or needle aspiration by the physician or a trained nurse of the traumatology unit. Soon after collection, samples were labelled appropriately and transported alongside with the participant's useful information slip to the

microbiology unit of the laboratory for immediate processing.

#### 2.3.3 Bacterial culture and identification

Upon reception, each pus sample was immediately seeded on a previously dried Champmann Agar plate and incubated at 37°C for 24 hours. The identification of bacterial isolates was based on their morphology, microscopic appearance after Gram stain, followed by biochemical characterization. As biochemical test, fermentation of mannitol, catalase, coagulase and DNase reactions were performed.

#### 2.3.4 Antibiotics susceptibility testing

Susceptibility tests were done using the Kirby Bauer disk diffusion method on Muller Hinton agar in accordance with the recommendations of the Antibiotics Committee of the French Society of Microbiology 2020 [25]. Briefly, four to five pure bacterial colonies were aseptically taken from Chapmann's medium and placed in a tube containing 5 ml of sterile physiological saline and gently mixed with a vortex until completely homogenization. The opacity of the bacterial suspension was standardized at Mac Farland 0.5 using a densitometer. The inoculation of bacteria on the surface of petri dishes was done with a sterile swab soaked in bacterial suspension. The inoculated plate was then allow to dry at room temperature for 5 minutes. Antibiotic discs were then placed using sterilized forceps and incubated at 37°C for 18-24 hours. Eight antibiotics were used on *Staphylococcus aureus*, namely: Cefoxitin (30 µg), Penicillin (10 µg), Amoxicillin (10 µg), Oxacillin (10 µg), Erythromycin (15 µg), Clindamycin (10 µg), Rifampicin (30 µg), Vanconicin (30 µg), Gentamicin (10 µg). Following incubation, the diameter of the bacterial growth inhibition zone was measured using a caliper. Depending on the diameter of the inhibition zone, the susceptibility was interpreted as sensitive (S), intermediate (I) or resistant (R). *S. aureus* strains with an inhibition diameter ≤ 21 mm around cefoxitin discs were considered resistant to methicillin [18].

## 2.4 Data Analysis

Data collected were inputted in Microsoft Excel 2013 spreadsheet and then transferred to IBM SPSS software version 25.0 (SPSS Inc., Chicago, IL, USA) for cleaning and analysis.

Results are expressed in terms of proportions and frequencies. Fisher's exact test was used to assess the association between demographic and clinical characteristics of the population and *S. aureus* carriage for a significance level of 5%.

### 3. RESULTS

#### 3.1 Distribution of *Staphylococcus aureus* Isolates According to the Socio-demographic and Clinical Data

From Table 1, participants ages ranged from 1 to 92 years old with a median of 31.50 years (IR: 19 - 40). *S. aureus* was isolated from 22 of the 52 pus samples cultured giving a positivity of 42.3%. The vast majority of strains (63.6%) were isolated from subjects younger than 21 years, while none of the participants aged 61 years and above were infected ( $P = .01$ ). The sex ratio for *S. aureus* carriage in males and females was 1.25. Gender was not significantly associated with the bacteria carriage ( $P = .75$ ). Subjects with no medical history were significantly ( $P = .03$ )

more infected with the bacteria (51.2%) than the others. The majority of strains were isolated from infected wounds (17.3%), osteomyelitis (5.8%) and abscesses (5.8%).

#### 3.2 Susceptibility Profile of *Staphylococcus aureus* to Antibiotics

Table 2 shows a prevalence of MRSA strains of 40.9%. The greatest sensitivity was observed with Rifampicin (77.3%) while the majority of strains were resistant to Amoxicillin (80%), Erythromycin (77.8%), Penicillin (75%) and Vancomycin (50%) respectively.

As shown in Table 3, 19 (86.4%) strains of *S. aureus* out of the 22 isolated displayed multi-drug resistance (MDR) to the antibiotics tested. MDR with 2 antibiotics was more recurrent (31.7% of all MDRs) followed by that with 4 and 5 antibiotics with the respective rates of 26.4% and 26.5%. All MRSA strains were cross-resistant with one or more other antibiotics.

**Table 1. Distribution of *Staphylococcus aureus* isolates according to the socio-demographic and clinical data**

Variables	Number	Positive Cultures Number (%)	P value
<b>Age Ranges (in Years)</b>			
<b>Median (IR)</b>	31.50 (19 - 40)		
< 21	15	14 (93.3)	.01
[21 ; 40[	25	7 (28.0)	
[41 ; 60]	8	4 (50.0)	
≥ 61	4	0 (0.0)	
<b>Gender</b>			
Female	14	5 (35.7)	.75
Male	38	17 (44.7)	
<b>Medical History</b>			
Hypertensive	2	0 (0.0)	.03
Diabetic	3	1 (33.3)	
Hypertensive and Diabetic	6	0 (0.0)	
None	41	21 (51.2)	
<b>Clinical Diagnosis</b>			
Pyomyositis	2	2 (3.8)	.77
Osteomyelitis	8	3 (5.8)	
Abscess	8	3 (5.8)	
Wound infection	21	9 (17.3)	
Open fracture	2	1 (1.9)	
Ulcer	1	1 (1.9)	
Arthritis	3	1 (1.9)	
Not mentionned	7	2 (3.8)	
<b>Total</b>	<b>52</b>	<b>22 (42.3)</b>	

**Table 2. Susceptibility profiles of *Staphylococcus aureus* isolates to Antibiotics tested**

<b>Susceptibility</b>	<b>FX</b>	<b>P</b>	<b>AM</b>	<b>OX</b>	<b>EM</b>	<b>DA</b>	<b>RIF</b>	<b>VA</b>	<b>GM</b>
	<b>n(%)</b>	<b>n(%)</b>	<b>n (%)</b>						
Sensitive	13(59.1)	2(10)	2(10)	8(44.4)	3(16.6)	14(66.7)	17(77.3)	7(31.8)	15 (71.4)
Intermediate	0(0.0)	3(15)	2(10)	0(0)	1(5.6)	1(4.8)	3(13.6)	4(18.2)	1 (4.8)
Resistant	9(40.9)	15(75)	16(80)	10(55.6)	14(77.8)	6(28.6)	2(9.1)	11(50)	5 (23.8)
Total	22	20	20	18	18	21	22	22	21

*FX=Céfoxitine, P=Pénicilline, AM=Amoxicilline, OX=Oxacilline, EM=Erythromycine, DA=Clindamycine, RIF=Rifampicine, VA=Vancomycine, GM=Gentamicine*

**Table 3. Multi-resistance profiles of *Staphylococcus aureus* isolates**

MDR Profile	Resistance Profile	Nº of isolates	% MDR
Two	FX, AM	01	5.3
	P, GM	02	10.5
	AM, GM	01	5.3
	FX, DA	01	5.3
	FX, EM	01	5.3
Three	FX, P, AM	02	10.5
	AM, RIF, VA	01	5.3
Four	P, AM, EM, VA	02	10.5
	FX, P, AM, OX	01	5.3
	P, OX, EM, VA	01	5.3
Five	FX, EM, VA, GM	01	5.3
	FX, P, AM, EM, VA	01	5.3
	P, AM, OX, EM, DA	01	5.3
	FX, AM, EM, VA, GM	01	5.3
	FX, AM, EM, DA, VA	01	5.3
Global	OX, EM, DA, VA, RIF	01	5.3
		19	86.4

FX=Cefoxitin, P=Penicillin, AM=Amoxicillin, OX=Oxacillin,  
EM=Erythromycin, DA=Clindamycin, RIF=Rifampicin,  
VA=Vancomycin, GM=Gentamicin

#### 4. DISCUSSION

*Staphylococcus aureus* is frequently implicated as causative agent of various infections in hospital care units [26]. The present study reports a prevalence of *S. aureus* of 42.3% in suppurating wounds in a traumatology unit in Dschang, Cameroon. Sanjana et al. [5] obtained a prevalence of 43.2% in pus samples from patients at outpatient consultation of a hospital in Nepal. Other studies have also revealed prevalences of 22.8% and 39.31% respectively in nasal swabs in intensive care units in Yaoundé [13] and in samples from surgical sites in Niamey [24]. The difference observed with other studies may come from the nature of the sample analyzed. Moreover, Gitau et al. [27] found an even higher occurrence of *S. aureus* (66%) in pus samples analyzed in their study. These observations suggest that the majority of hospital suppurations are caused by *S. aureus*.

Of the *S. aureus* strains isolated in this study, 40.9% demonstrated resistance to cefoxitin. Kamga et al. [9] and Mohamadou et al. [28] obtained greater MRSA prevalence of 58.62% and 51.5% respectively in Yaoundé and in the Adamawa and Far North regions of Cameroon. This difference could be justified by the sample size, which was larger in their studies. The sex ratio for *S. aureus* carriage in males and females was 1.25. Gender was not significantly associated with the bacteria carriage ( $P = 0.753$ ).

While Kamga et al. [9] and Ouidri et al. [29] obtained similar results, other studies have shown that men are predominant carriers of *S. aureus* compared to women [30,31].

The majority of *S. aureus* strains were isolated from infected wounds (17.3%), osteomyelitis (5.8%) and abscesses (5.8%). Tigist et al. [11] also found a high occurrence (69.5%) of wound infections caused by *S. aureus* in Ethiopia. Although not frequently reported as wound infections, studies also incriminate *S. aureus* as the causative agent of certain osteomyelitis in France [15] and Tunisia [32].

The majority of bacterial strains were resistant to most anti-*Staphylococcal* agents such as Amoxicillin (80%), Erythromycin (77.8%) and Penicillin (75%) respectively. However, Tigist et al. [11] and Mendem et al. [4] observed increased resistance of *S. aureus* to penicillin of 95.5% and 97.5% respectively. Other studies have reported considerable resistance to Erythromycin and Ampicillin [33] or Rifampicin and Cotrimoxazole [28]. This variability in resistance to a wide range of antibiotics reveals the limited possibilities and therapeutic choices applicable in emergency circumstances or in a syndromic approach in facilities with limited resources.

The MDR rate of *S. aureus* strains to the antibiotics tested was 86.4%. Studies show that

this resistance is due to the high genetic plasticity of *S. aureus*, which facilitates the acquisition of antibiotic resistance genes and the development of new regulatory mechanisms of adaptation to increasing concentrations of antibiotics [17,34].

All strains of MRSA have been cross-resistant with one or more other antibiotics. Several studies have shown that methicillin-resistant *S. aureus* strains (MRSA) exhibit cross-resistance to other antibiotics including  $\beta$ -lactams compared to methicillin sensitive ones [5,9,35]. Kaur et al. [2] found no resistance when testing the susceptibility of MRSA strains to new antibiotic molecules such as ceftarolin, phosphonic acid, glycopeptides, glycyclines, fucidans.

## 5. CONCLUSION

The objective of the study was to highlight the occurrence and the antibiotic resistance of community acquired *S. aureus* strains isolated from suppurations at the traumatology unit of OLHCB in Dschang, West Cameroon. This bacterium was more prevalent in suppurations from infected wounds. A considerable proportion of the strains were multidrug resistant and mainly isolated in subjects under the age of 21 years. MRSA strains displayed cross-resistance with one or more other antibiotics. Adequate care should be taken while handling suppurating wounds and abscesses. Especially in younger age individuals, as this may be the application of point of care precision methods in the diagnosis of extremely flexible bacterial pathogens such as *S. aureus* as well as their susceptibility profiles may help in timely setting up adequate proper care and treatment protocols necessary to overcome drug resistance of such extremely flexible pathogens.

## DISCLAIMER

The products used for this research are commonly and predominantly used products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

## CONSENT AND ETHICAL APPROVAL

This study was approved by the administrative authorities of OLHCB. The free and informed consent of each participant was obtained before their inclusion in the study. The study was conducted in compliance with the Declaration of Helsinki on research involving humans [36].

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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