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Prevalence of *Staphylococcus aureus* and Methicillin-resistant *S. aureus* in Adult Hospitalized Patients in "Mother Theresa" Hospital Center in Albania

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Abstract

AIM: The aim of this study was to evaluate the prevalence of *Staphylococcus aureus* and methicillin-resistant *S. aureus* (MRSA) in clinical specimens hospitalized to "Mother Theresa" Hospital Center for 2 years.

METHODS: We isolated and identified *S. aureus* on 356 clinical specimens using standard tests. Furthermore, for further accurate microbial identification, we have to use the VITEK® 2 system. The samples were tested to detect the presence of MRSA by a slide latex agglutination kit for the rapid detection of PBP2.

RESULTS: The overall prevalence of *S. aureus* in patients was 34.2%. The prevalence of MRSA was 20.5% of cases. Of the MRSA isolates identified in this study, 28% were susceptible to antibiotics, 24% demonstrated intermediate resistance, and 48% were multi-drug resistant with resistance to nineteen antibiotics involved in the examination. In addition, seven of the 25 MRSA cases showed 100% resistance to norfloxacin, imipenem, meropenem, levofloxacin, etc.

CONCLUSIONS: The rate of *S. aureus* in hospitalized patients on this study was 34.2% and the MRSA 20.5%. These results indicated that this type of infection is a significant concern for health services and patients included. A screening of all hospitalized cases can lead to reduce the incidence of this infection in the hospital environment.

Introduction

Staphylococcus aureus, a member of the family Micrococcaceae, is a Gram-positive coccus whose cells tend to occur either singly or if dividing cells do not separate, form pairs, tetrads, and distinctive irregular "grape-like" structures [1], [2]. S. *aureus* is a common human opportunistic pathogen often carried asymptomatically on the human body. However, in some circumstances, this bacteria can cause more serious infections in a wide range which affecting the superficial skin and soft tissue [3], [4], as well as invasive infections including pneumonia, bloodstream infections, bone, and joint infections [2], [4]. Infections caused by S. aureus, above all, by antibiotic-resistant strains, have reached epidemic proportions globally [5]. Methicillin-resistant *S. aureus* (MRSA) was first reported in 1961 and has since become a major nosocomial pathogen worldwide [2], [5], [6], [7], [8]. MRSA includes those strains that have acquired a gene giving them resistance to methicillin and essentially all other beta-lactam antibiotics [9]. Since that time, MRSA has emerged as a significant problem worldwide [10], [11], [12], [13], [14], and the term has evolved to include resistance to additional ß-lactam antimicrobials. At present, the term MRSA is often used to describe multi-drug resistant S. aureus [15]. In nowadays, the incidence of MRSA is growing and unfortunately efforts to develop new, more effective antibiotics are dwindling. The continued emergence of resistant strains of bacteria such as MRSA demands an urgent revival of the search for new antibiotics [16]. MRSA in a different group of patients (especially in immunocompromised and operated patients) causes different clinical syndromes and different antimicrobial susceptibility patterns. For this reason, the aim of this study was to evaluate the prevalence of *S. aureus* MRSA cases who were hospitalized patients at the "Mother Theresa," Hospital Center, during the period January 2014–December 2015.

Materials and Methods

This study was carried out from January 2014 to December 31, 2015, at "Mother Theresa" Hospital,

a tertiary care university hospital in Tirana, Albania. UHC "Mother Theresa" is the only reference center in Albania for acute and critical patients. About 356 clinical specimens were collected by hospitalized patients aged 18 years or older.

Patients enrolled in this study were hospitalized in different wards such as infection diseases, cardiology, dialyses, surgery, dermatology, nephrology, cardiology, neurology, intensive care, etc.

A questionnaire was used to collect socioeconomic and demographic data from medical personalized files of each patient. The demographic data include age, sex, school education, residence (urban/rural), marital status, and number of children. In the medical records, we have viewed information about regarding the reason why he or she was been hospitalized, if they were used antibiotic during the hospitalized, there have chronic diseases (such as hypertension, cardiac diseases, diabetes mellitus (type 2 and 1), and liver and kidney dysfunction). Wound, abscess, pus/exudates, skin and soft tissue swab, blood, urine, sputum, throat swab, and indwelling medical device samples were collected from suspected patients. All specimens were sent to the microbiology laboratory within 1 h. Immediately, the samples were streaked on to Sheep Blood agar plate and other media for Gram-negative bacteria and incubated in 37°C for 24 h. We isolated and identified *S. aureus* on the basis of colony morphology and also we used catalase test, coagulase test, mannitol fermentation on mannitol salt agar, production of deoxyribonuclease (DNase) on DNase agar, Staphyloslide Latex Test, and CHROMagar Orientation.

Furthermore, for further accurate microbial identification, we have to use the VITEK® 2 system. For susceptibility tests, we use both automated Microbiology System (VITEK® 2 system) and Kirby–Bauer Disk Diffusion Susceptibility Test. The main antibiotics used for susceptibility tests were cefoxitin, moxifloxacin, cefixime, norfloxacin, imipenem, doxycycline, meropenem, vancomycin, rifampicin, clindamycin, amoxicillin, levofloxacin, ceftriaxone, gentamycin, nitrofurantoin, cefaclor, ceftazidime, cefotaxime, cefuroxime, and amikacin.

The samples were tested to detect the presence of MRSA (S. aureus methicillin resistance) by a slide latex agglutination kit for the rapid detection of PBP2 (the enzyme encoded by mecA), the cefoxitin disk screen test and sometime with CHROMagar™ MRSA.

For statistical analyses, all the data were expressed in frequencies and percentages. Chi-square test and Fisher's exact test were used to determine relationships between categorical variables. In the univariate analyses of risk factors, age, number of household, and income were categorized. $p < 0.05$ indicated a statistically significant difference in all tests.

All data collected from the medical files of each patient were analyzed by SPSS version 20.0 (SPSS Inc., Chicago, IL, USA).

Results

Overall, 356 samples tested for the presence of *S. aureus*, only 34.2% (122/356) of them, resulted positive with *S. aureus*. The prevalence of MRSA was 20.5% (25/122) cases (Figures 1 and 2).

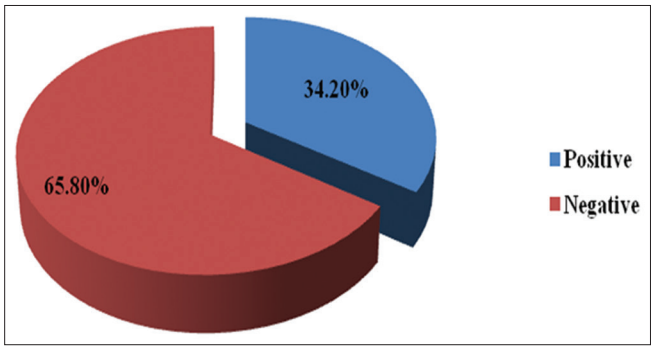

Figure 1: Prevalence of Staphylococcus aureus

In Table 1, we have presented the demographic characteristics data of patients admitted to the "Mother Theresa" Hospital Centre. Male (59.8%) was the most predominant sex in this study compared to female (40.2%) without a significance level between them. The min age was 18 and the max 83 years old and the average was 54.6 ± 17.3.

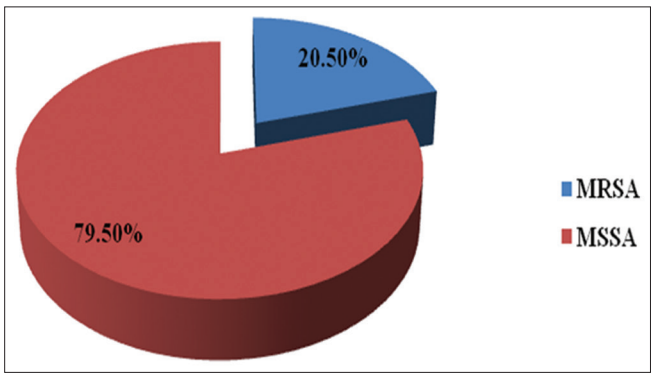

Figure 2: Prevalence of methicillin-resistant Staphylococcus aureus

Patients that living in rural area were more predominant compared to them that living in an urban area 64.9% and 35.1%, respectively. A strong association has found for residence area and presence of *S. aureus*. Based on age groups, patients more than 66-year-old presented the highest number of patients (32.9%) and the prevalence of *S. aureus* in this group was 31.1%. We have not found any association between age groups and the presence of *S. aureus*. For demographic characteristics of patients such as marital status, level of school education, and social level, married patients presented 75.3% of total cases but was not been found any association between the presence of *S. aureus* and marital status. On the other hand, we have found a strong association between the presence of this infection and the school education of patients as well as their social level.

Table 1: Demographic characteristics of patients admitted to "Mother Theresa" Hospital Center

Demographic data	Frequency	Percentage	Prevalence of *S. aureus*	p value	Prevalence of MRSA
Sex				>0.05	
Female	143	40.2	37.7		36
Male	213	59.8	62.3		64
Residence area				=0.003	
Urban	125	35.1	33.6		44
Rural	231	64.9	66.4		56
Age groups (years)				>0.05	
18–25	32	9	5.7		4
26–35	28	7.8	7.4		4
36–45	40	11.2	12.3		8
46–55	73	20.5	20.5		20
56–65	66	18.5	23		36
>66+	117	32.9	31.1		28
Marital status				>0.05	
Married	268	75.3	69		52
Single/separated	37	10.4	10.5		20
Widowed	51	14.3	20.5		28
School education (level)				<0.0001	
Elementary	4	1.1	3.3		4
8-years	132	37.1	41.8		40
High school	120	33.7	33.6		32
University	100	28.1	21.3		24
Social level				=0.004	
School/Student	32	9	6.5		8
Invalid	18	5	7.4		12
Without work	80	22.5	26.2		28
Employed	130	36.5	36.9		28
Retiree	96	27	23		24

MRSA: Methicillin-resistant *S. aureus*, *S. aureus: Staphylococcus aureus*.

Table 2 presents the positivity of S. aureus and MRSA divided by the wards were the samples of patients are collected.

Table 2: Positivity of *S. aureus* and MRSA based toward collected samples

Wards	Total number	S. aureus		MRSA	
		Positive	Negative	MSSA	MRSA
Cardiology	59	16	43	11	5
Dialysis	19	7	12	2	5
Infection diseases	53	20	33	17	3
Nephrology	41	14	27	11	3
Neurology	12	7	5	10	2
Surgical	68	22	46	19	3
Dermatology	104	36	68	32	4
Total	356	122	234	97	25

MRSA: Methicillin-resistant S. aureus, S. aureus: Staphylococcus aureus.

In Figure 3, we have presented the distribution of *S. aureus* and MRSA positive cases.

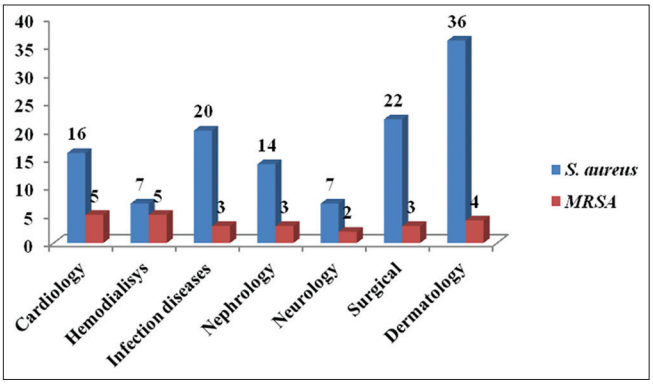

Figure 3: Distribution of positive cases with Staphylococcus aureus and methicillin-resistant S. aureus+

In Table 3, we have presented the antibiotics admitted to examination to see the sensitivity and resistant of MRSA cases.

Table 4 presents the prevalence of infection with *S. aureus* based to the source of clinical specimens.

Table 3: Antibiotics used for MRSA

Antibiotics	Type of *S. aureus* MRSA		
	Sensitive (%)	Resistant (%)	Intermediate (%)
Cefoxitin	10	90	0
Moxifloxacin	88	5	7
Cefixime	0	85	15
Norfloxacin	0	100	0
Imipenem	0	100	0
Doxycycline	50	29	21
Meropenem	0	100	0
Vancomycin	12	30	58
Rifampicin	77	13	10
Clindamycin	29	58	13
Amoxicillin	59	25	36
Levofloxacin	0	100	0
Ceftriaxone	0	90%	10
Gentamycin	0	75	25
Nitrofurantoin	78	0	12
Cefaclor	0	100	0
Ceftazidime	0	100	0
Cefotaxime	0	100	0
Cefuroxime	5	95	0
Amikacin	88	2	10

MRSA: Methicillin-resistant *S. aureus*, *S. aureus: Staphylococcus aureus*.

Table 4: Prevalence of infection with *S. aureus* based to source of clinical specimens

Cases collected from	Frequency (n)	Percentage (%)	Positive cases (n)
Urine infections	75	21.0	21
Skin and soft-tissue infections	96	27	32
Vaginal and urethral swab	47	13.2	15
From nasal, sputum and ear swab cases	58	16.3	18
Bloodstream, indwelling medical devices, and catheter-associated infections	80	22.5	36

Discussion

Nowadays, the prevalence of *S. aureus* and methicillin resistance of *S. aureus* (MRSA) is very common in most of the countries. The prevalence of MRSA varies from one geographic region to another and between different institutions in a given area [17] and the prevalence of infections and outbreaks due to of

S. aureus and MRSA has continued to increase in many of countries [17], [18], [19], [20]. Fact is that *S. aureus* and MRSA present different prevalence around the world and also around European countries [21]. The aim of this study was to evaluate the prevalence of *S. aureus* and MRSA in clinical specimens hospitalized to "Mother Theresa" University Hospital Center. This study is not the first prevalence survey on *S. aureus* infection and MRSA in patients admitted at "Mother Theresa" University Hospital Centre [22], [23], [24]. Overall, 356 samples tested for the presence of *S. aureus*, the prevalence of this infection among hospitalized patients resulted to be 34.2% (122/356). The prevalence of MRSA was 20.5% (25/122) of cases. In the previous study conducted in Albania, the MRSA in admitted patients was 14.2% MRSA nasal carriers in 2016, a study survey by Falzon *et al*. [22], 24% methicillin-resistant measured with MRSA-Screen in a study 2017 by Dako *et al*. [23]. Another study in 2003 by Faria *et al*. conducted in Mother Theresa Hospital Centre, the prevalence of *S. aureus* resulted 18.2% in admitted patients [24].

Our finding of MRSA prevalence (20.5%) is almost the same with another study conducted in Greece, which had a prevalence of 20% of MRSA in screened hospitalized patients [25]. However, we have a high prevalence if compare with the finding in Croatia that reports a MRSA 5.2% [26] and Serbia 11.8% [27].

Regarding the demographic characteristics data of patients admitted to the "Mother Theresa" Hospital Centre, presented in Table 1, the min age was 18 and the max 83 years old and the average was 54. 6 ± 17.3.

Male (59.8%) was the most predominant sex in this study compared to female (40.2%). The prevalence of S. aureus infection and MRSA divided by gender was 37.7% (S. aureus) with 36% (MRSA) for female and for male 62.3% (S. aureus) with 64% (MRSA); however, we did not find a significant association between gender and infection. If we compare the living areas of our patients, can be mention that more than half living in rural areas (64.9%) and the others in urban areas (35.1%). The prevalence of S. aureus in patients that lived in the rural area was distinctly the highest to those living in the urban area (66.4% and 33.6%, respectively). A strong significant association has found for residence area and presence of S. aureus for $p = 0.003$.

Faria *et al*., in their study, according to the means of logistic regression, proved that the age >40 years is an independent risk factor for *S. aureus* infection [24]. In our study, patients, more than 66 years old, presented the highest number of patients 32.9%, but we have not found any association between age groups and presence of *S. aureus* ($p > 0.05$). Married patients presented 75.3% of total cases, and in this demographic characteristic, we did not found any significant association between this infection and marital status of patients ($p > 0.05$).

However, on the other hand, in patients admitted to "Mother Theresa" Hospital Centre, we have found a strong association between the presence of *S. aureus* and school education ($p < 0.0001$) as well as the social level (for $p = 0.004$).

The prevalence of *S. aureus* on each wards resulted to be; 27.1% (16/59) for cardiology, 36.8% (7/19) dialysis, 37.3% (20/53) infection diseases, 34.14% (14/41) nephrology, 58.3% (7/12) neurology, 32.35% (22/68) surgical, and 34.6% (36/104) dermatology. The prevalence of MRSA was higher in cardiology wards (20%) than in dialysis (20%) and dermatology wards (16%). We did not found an association forward sample collection and presence of *S. aureus*, p value result > 0.05.

MRSA is an increasingly important clinical problem since MRSA is often multidrug-resistant and therapeutic options are limited. In this study, we have identified 25 MRSA isolates cases, in which 7 (28%) were susceptible to antibiotics, 6 (24%) demonstrated intermediate resistance and 12 (48%) were multi-drug resistant with resistance to nineteen antibiotics involved in the examination. In addition, seven of the 25 MRSA cases showed 100% resistance to norfloxacin, imipenem, meropenem, levofloxacin, cefaclor, ceftazidime, and cefotaxime.

Samples were taken out from the skin and soft-tissue infection, urine infection and from nasal, sputum, and ear swab and also from bloodstream, indwelling medical devices, and catheter-associated infections. Of all 122 cases isolated with *S. aureus*, 21 (17.2%) were from urine infections; 32 (26.2%) from skin and soft-tissue infections cases; 15 (12.3%) from vaginal and urethral swab; 18 (14.8%) from nasal and ear swab cases; and 36 (29.5%) from bloodstream and catheter-associated infections.

Conclusions

The rate of *S. aureus* in hospitalized patients on this study was 34% and the MRSA 20.5%. These results indicated that this type of infection is a significant concern for health services and patients included in the study. The highest percentage of *S. aureus* found in surgical and nonsurgical wound suggest that further investigation should be implemented. A screening of all hospitalized cases can lead to reduce the incidence of this infection in the hospital environment and also to control the risk factors.

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