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Prevalence, Resistance to Quinolones/Fluoroquinolones of *Staphylococcus aureus* Strains Isolated in the Hospital and University Center of Brazzaville, Republic of Congo

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Abstract

Staphylococcus aureus is a gram-positive pathogenic bacterium responsible for various infections including skin suppurations. Quinolones are sometimes used last in infections. The *Staphylococcus aureus* objective of this study was to determine the prevalence and assess the level of resistance to quinolones of *Staphylococcus aureus* trains isolated in the Hospital and University Center of Brazzaville (CHU-B). 40 strains of *Staphylococcus aureus* from various biological products were isolated and identified by bacteriological methods. The study of the resistance of these different strains to antibiotics (Quinolones/Fluoroquinolones) was carried out by the phenotypic method, by antibiogram. A total of 40 *Staphylococcus aureus* strains were isolated including 13 (32.50%) community strains, 27 (67.5%) clinical strains distributed as follows: neonatal and pediatric service 29.62% and 25.92%, surgery 18.51%, contagious 14.81%, emergencies 7.4% and delivery block 3.7%. 60% of strains were isolated in women and 40% in men. The average age of those infected was 16 years and older. 90% strains were resistant to Nalidixic acid (AN), 72.5% to Ofloxacin (OFL), 67.5% to Ciprofloxacin (CIP), 70% to Norfloxacin (NOR), 62.5% to Levofloxacin (LEV), 47.5% to Moxifloxacin (MXF), and 40% strains were resistant to all antibiotics tested.

Keywords

Prevalence, *Staphylococcus aureus*, Antibiotics, Resistance

1. Introduction

Bacteria are microorganisms or single-celled living things without a nucleus. The human body is home to several microorganisms, which can be commensal or pathogenic. Once they become pathogenic, they can be responsible for many infectious diseases [1]. According to the World Health Organization (WHO), bacterial infectious diseases are the 3rd leading cause of death in developing countries after malaria and malnutrition [2]. The *Staphylococcus aureus* species is one of the most common pathogens involved worldwide, as well as in the Republic of Congo.

In recent decades, there has been a leading cause of hospital, clinical and community infections [3]. The strain was first isolated by Rosenbach. *S aureus* is a Gram-positive b, pathogenic and opportunistic bacterium of humans and various mammals [4] causing a wide variety of infections ranging from common infections to fatal infections, such as endocarditis, osteomyelitis and associated syndromes to the action of toxins [5] [6].

The treatment for bacterial infections is taking antibiotics. These bacterial infections are not at all easy to treat. Treatment failures occur despite an extensive arsenal of antibiotics and strong antiseptics. Bacterial infections remain a public health problem that the Congo's health system is hardly immune to. This is the case with the bacteria *Staphylococcus aureus* which is one of the most commonly isolated species in hospitals. This terrible pathogen has developed resistance to each new antibiotic placed on the market, in particular that of the quinolones/fluoroquinolones (FQ) family, from which infections linked to hospital require *Staphylococcus aureus* a heavy burden on healthcare resources. On the other hand, the choice of antibiotics to be administered during treatment against bacterial infections is an important factor in the management of infected hospitalized and non-hospitalized patients and also a means of compensating for treatment failures. This work is going to determine the prevalence and assess the resistance of isolated *Staphylococcus aureus* in hospital and community settings to quinolones/fluoroquinolones in the Republic of Congo.

2. Material and Methods

2.1. Site and Collection of Strains

Staphylococcus aureus strains collected at the biomedical analysis laboratory in the Hospital and University Center of Brazzaville (CHU-B) using various biological fluids (urine, vaginal swabs, blood, pus, and Pleural Fluid) of hospitalized patients (hospital strains) and non-hospitalized (community strains) were used.

Staphylococcus aureus strains were isolated during the period of April 2019 to July 2020.

2.2. Isolation and Identification of Strains

The isolation of *Staphylococcus aureus* strains was made by culturing the biological products for 24 hours on a selective agar medium called Chapman * agar (hypersaline medium containing 7.5% Na Cl and 1% mannitol and containing red of phenol). A few drops of the biological liquid were spread on Chapman agar medium, making streaks. The identification of the strains was done by following the morphological characters by the gram staining technique, and by the biochemical characters obtained by the catalase and coagulase tests [7].

2.3. Study of Resistance to Quinolones

The resistance profile of *Staphylococcus aureus* was evaluated, by standard antibiogram (phenotypic method). The bacteria were cultured on the MUELLER HINTON (MH) medium in the presence of antibiotics (ATB) and incubate in a normal atmosphere, 35˚C ± 2˚C for 20 ± 4 h, and the consequences on their development and survival were observed [8] [9].

The reading and interpretation of the results was done by measuring with precision the different diameters of the zones of inhibitions and by comparing them with the critical values pre-established by the Antibiogram Committee of the French Society of Microbiology (CA-SFM 2019) [10].

The antibiotics tested were those of the quinolones/fluoroquinolones family of the four generations [11], for the first generation Nalidixic acid (AN) 30 µg, second generation Ciprofloxacin (CIP) 5 µg, Norfloxacin (NOR) 10 µg, and Ofloxacin (OFX) 5 µg, third generation Levofloxacin (LEV), 5 µg and fourth and latest generation Moxifloxacin (MXF) 5 µg.

2.4. Analysis of the Results

For statistical analyzes Microsoft Excel was used and this helped for statistical data graphics.

3. Results

3.1. Isolation and Identification of Strains

Figure 1 represents the percentage of community and clinical *Staphylococcus*

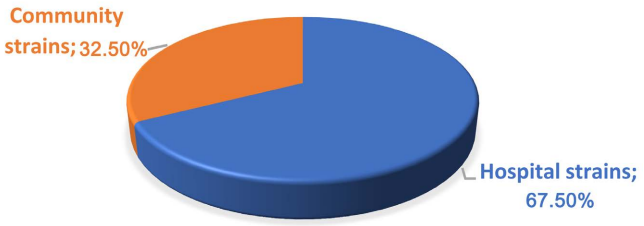

Figure 1. Distribution of isolated and identified strains of *Staphylococcus aureus*.

aureus strains isolated and identified. Forty (40) strains of *Staphylococcus aureus* were isolated and identified, including 67.50% clinical and 32.50% community samples from the services in the Hospital and University Center of Brazzaville (CHUB).

3.2. Statistical Analysis of Epidemiological Data

During the study period of April 2019 to July 2020, 40 strains of *Staphylococcus aureus* were isolated on Chapman Agar medium, 13 community-based and 27 clinics distributed as follows: neonatology and pediatrics department 29.62% and 25.92%, surgery 18.51%, contagious 14.81%, emergencies 7.4% and delivery block 3.7%. 60% of the strains were isolated from women (n = 24) and 40% from men (n = 16). The average age of those infected ranged from 16 years and older.

3.2.1. Distribution of *Staphylococcus aureus* Strains by Department

Figure 2 shows the distribution of ***Staphylococcus aureus*** by department. The services most colonized by *S. aureus* are the neonatology and pediatric services with percentages of 29.62% and 24.13%. The other services surgery, and contagious the frequency was respectively 18.51% and 14.81%. Emergency departments and the delivery block were the least colonized by ***Staphylococcus aureus*** at 7.4% and 3.7%.

3.2.2. Distribution of *Staphylococcus aureus* Strains by Sex

Figure 3 shows the distribution of clinical and community strains by sex. The strains were more isolated in women than in men in both cases with percentages of 59.25% and 40.75% respectively for clinical strains and 61.53% and 38.46% for community strains.

3.2.3. Distribution of *Staphylococcus aureus* Strains by Age

Figure 4 shows the distribution of clinical and community *S. aureus* strains according to age. Strains were more isolated in the 16-plus age group than 0 - 15

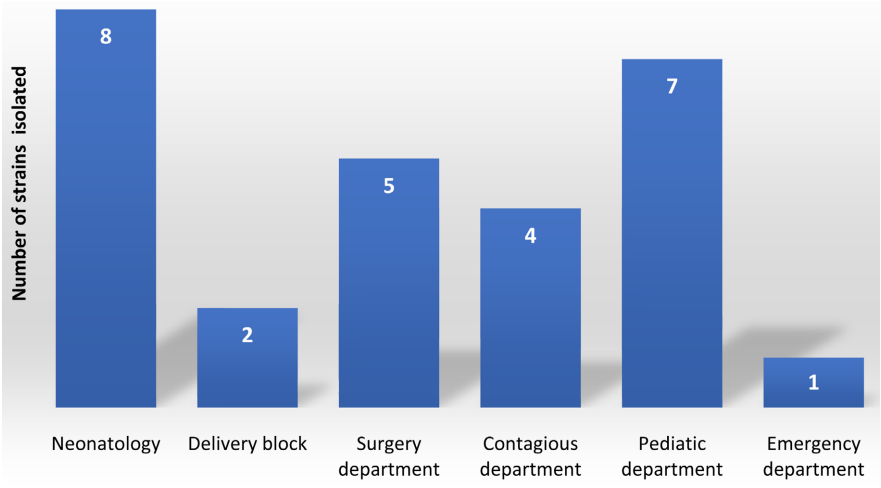

Figure 2. Distribution of isolated *Staphylococcus aureus* strains by department.

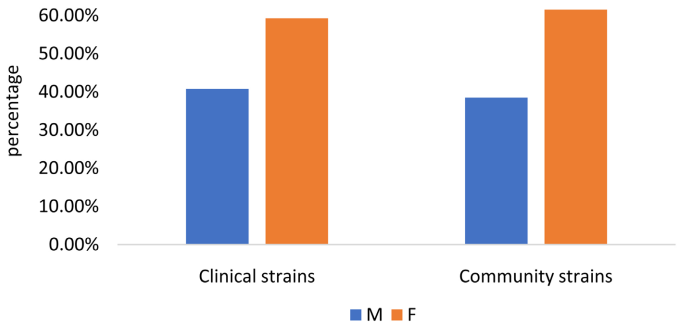

Figure 3. Distribution of clinical and community *Staphylococcus aureus* strains according to sex.

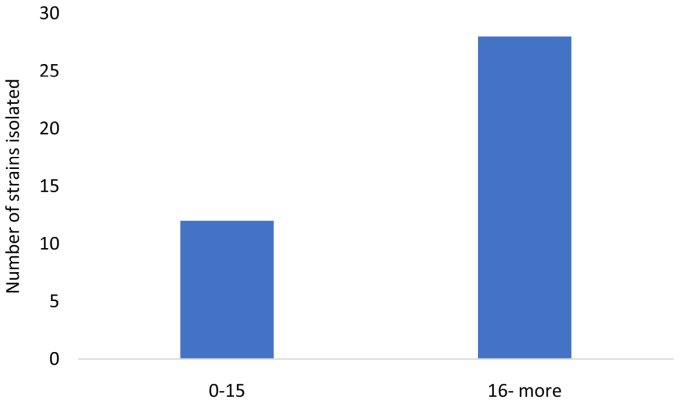

Figure 4. Distribution of community and clinical *Staphylococcus aureus* strains according to age.

with frequencies of 70% (28 strains isolated) and 30% (12 strains isolated), respectively.

3.2.4. Distribution of *Staphylococcus aureus* Strains According to the Nature of the Samples

Figure 5 represents the distribution of the strains isolated as a function of biological fluid.

Twelve (12) strains were isolated from urine and blood, nine (9) from vaginal swabs (PV), six (6) from pus and one (1) strain from pleural fluid (LP).

3.3. Phenotypes of Resistance

Figure 6 shows the different levels of sensitivity and resistance of the strains to the antibiotics tested, Ciprofloxacin (CIP), Norfloxacin (NOR), Levofloxacin (LEV), Ofloxacin (OFX), Moxifloxacin (MXF), Nalidixic acid (NA).

Figure 6(a) and **Figure 6(d)** show that the strains R68, H83 were sensitive to CIP, NOR, LEV, OFL, MOX and resistant to AN.

Figure 6(b) and **Figure 6(c)** show that strains H112 and S404 were sensitive to MOX and resistant to NOR, LEV, OFL, AN.

Table 1 represents the resistance rates of the community and clinical ***Staphylococcus aureus*** strains.

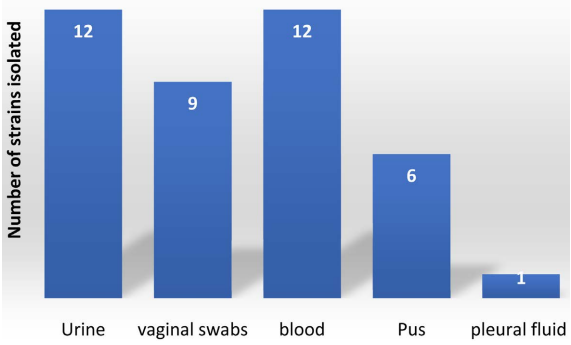

Figure 5. Distribution of *Staphylococcus aureus* strains according to the nature of the samples.

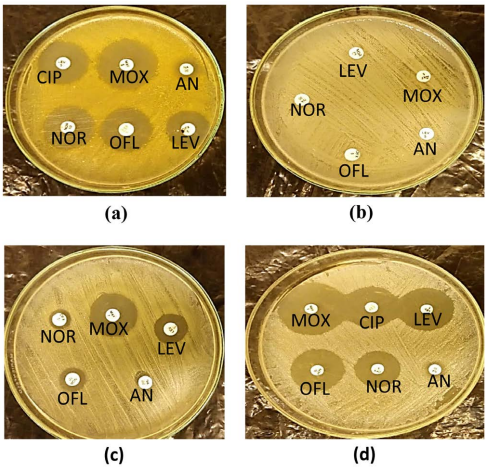

Figure 6. Resistance profile of *S. aureus* strains on MUELLER HINTON medium. (a) S R68; (b) S 404; S (c) H112; (d) S H83. S: Strain; NOR: Norfloxacin, CIP: Ciprofloxacin, MXF: Moxifloxacin, LEV: Levofloxacin; OFX: Ofloxacin, AN: Acid Nalidixic.

Table 1. Distribution of *S. aureus* strains according to origin and sensitivity to antibiotics.

	***S. aureus* clinical (n = 27)**			***S. aureus* community (n = 13)**		
ATBs tested	**R (%)**	**I (%)**	**S (%)**	**R (%)**	**I (%)**	**S (%)**
CIP	19 (70.37%)	0 (%)	8 (29.62%)	9 (69.23%)	0 (0%)	4 (30.76%)
NOR	19 (70.37%)	0 (%)	8 (29.62%)	8 (61.53%)	0 (0%)	5 (38.46%)
LEV	16 (59.25%	0 (%)	11 (40.74%)	8 (61.53%)	0 (0%)	5 (38.46%)
OFL	21 (77.77%)	0 (%)	6 (22.22%)	8 (61.53%)	0 (0%)	5 (38.46%)
MOX	12 (44.44%)	0 (%)	15 (55.55%)	6 (46.15%)	0 (0%)	7 (53.84%)
A N	26 (96.29%)	0 (%)	1 (3.70%)	13 (100%)	0 (0%)	0 (0%)

ATBs tested = Antibiotics tested; n = Number of strains. NOR: Norfloxacin, CIP: Ciprofloxacin, MXF: Moxifloxacin, LEV: Levofloxacin, OFX: Ofloxacin, AN: Acid Nalidixic.

All the strains were tested with different antibiotics from the same family broken down as follows: Ofloxacin, Ciprofloxacin, Levofloxacin, Nalidixic Acid, Moxifloxacin and Ofloxacin. It emerges from the analysis of the results of this table that 100% of the community strains were resistant to Nalidixic Acid,

69.23% to Ciprofloxacin 61.53% resistant to Levofloxacin, Norfloxacin, and Ofloxacin, on the other hand, Moxifloxacin was the most active molecule. Moxifloxacin was also the most active for clinical Staphylococcus ***aureus*** strains, moreover resistance rates of, 77.77% to Ofloxacin, 70.37% to Norfloxacin and Ciprofloxacin, 59.25% to Levofloxacin were observed and the highest rate 96.29% with Nalidixic Acid.

4. Discussion

Statistical analysis of the epidemiological data from the study shows that the strains from hospital or community settings have the same characteristics and cause the same type of infection, and there is a predominance of isolation of hospital strains with 67.50%. This predominance can be explained by the fact that the study was conducted in a hospital setting. We were able to collect more clinical strains than community strains. These results are close to those of [12], different from those obtained by [13], which shows a predominance of community strains.

The proportions of strains isolated in different departments vary according to the type of activities of the hospital departments. The neonatology and pediatrics department, where newborns and children are respectively hospitalized, are the most colonized by ***Staphylococcus aureus***. As the samples taken in these services are blood, this dissemination is associated with health care and can be explained by the lack of hygiene and the lack of control of paramedical care by health personnel (the change of gloves, the control of asepsis on the treatment cart and correct and multiple hand washing). It can also be caused by the use of venous catheters during the administration of care; because these appear to be a risk factor for infection of ***Staphylococcus aureus*** in the blood when they are inserted into a vein and remain there for a long time [14]. Similar results have been reported by [15]. However, the data on this subject vary in the literature, including more men than women [16].

The most isolated ***Staphylococcus aureus*** are those from blood and urine with a percentage of 30%. Regarding community strains, the results obtained with the urine are consistent with those obtained by [17] with an isolation rate of ***Staphylococcus aureus*** of 34.3%. The reason for the presence of ***Staphylococcus aureus*** in pus is that this strain is more common in wounds and circulates easily in the blood.

Like most drugs, fluoroquinolones have been shown to have a favorable safety profile against, which has been verified with the different classes of Quinolones/Fluoroquinolones used. The resistance profile shows strong resistance to almost all tested antibiotics Nalidixic Acid (NA) Levofloxacin (LEV), Ciprofloxacin (CIP), Norfloxacin (NOR) and Ofloxacin (OFX). Resistance to these different antibiotics could be explained by the fact that it is acquired, progressive, and varies over time. The latter can also be explained by the fact that the wide use of Fluoroquinolones is a factor in the emergence and dissemination of strains resistant to

Fluoroquinolones [18]. Resistance rates were higher 96.29% and 100% with AN respectively for clinical and community strains, this is explained by the natural resistance of the genus staphylococcus to this antibiotic [19] [20]. On the other hand, the variability in the rate of resistance from one antibiotic to another within the family of Fluoroquinolones observed (77.77% at OFL, 70.37% at CIP, 70.37% at NOR, 59.25% at LEV, 44.44% at MOX for the clinical strains observed) could be explained by the impact of the activity of the antibiotic against the germ and function of the generation to which belongs the antibiotic. These results agree with those described in the literature (microbiology review) [19].

5. Conclusion

The study made it possible to determine the prevalence of strains of ***Staphylococcus aureus*** isolated in certain CHU-B services and in the community of the city of Brazzaville. In the sampling used, the strains were isolated more from hospitalized patients than from the community with rates of 67.50% and 32.50% respectively. A predominance of isolation was observed in women with 60%, the average age of those infected was the age range of 16 years and over.

From the point of view of resistance of the strains isolated to the antibiotics of the Quinolones/Fluoroquinolones family used, 40% of the isolated strains were resistant to all the antibiotics tested, and the clinical and community strains were resistant to Nalidixic acid (NA), Levofloxacin (LEV), Ciprofloxacin (CIP), Norfloxacin (NOR), Ofloxacin (OFX) and Moxifloxacin (MXF) sensitive. Ninety (90%) strains were resistant to AN, 72.5% to OFX, 67.5% to CIP, 70% to NOR, 62.5% to LEV, 47.5% to MXF. Resistance was more observed with NA with resistance rates of 96.29% and 100% respectively for clinical and community strains.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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