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Antibiotic Resistance Trends of Nasal Staphylococcal Isolates from Namibian School Children

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Abstract

Nasal colonization with *Staphylococcus* strains puts children at risk of developing difficult-to-treat staphylococcal infections. Antibiotic resistance data is limited in Namibia. Our study thus aimed to provide resistance trends for nasal staphylococci isolated from school children in the Mariental District. This is the first report on antibiotic resistance trends of staphylococci from Namibian school children. By Kirby-Bauer disk diffusion assay, 352 *Staphylococcus aureus* and 81 coagulase-negative staphylococci (CoNS) isolates from Namibian school children aged 6 - 14 years underwent susceptibility testing against seven antibiotics. Ninety-six percent *S. aureus* and 66.7% CoNS were resistant to ampicillin. Ampicillin resistance was significantly higher in *S. aureus* than in CoNS ($P < 0.0001$). Ciprofloxacin and gentamicin were the most effective against *S. aureus*. Ciprofloxacin was also the most effective drug against CoNS. Cefoxitin/methicillin resistance was seen in 14.5% *S. aureus* isolates and 8.6% of CoNS. Thirty-one antibiotic resistance patterns were observed, most frequently ampicillin (A), ampicillin-erythromycin (AP-E), and ampicillin-tetracycline (AP-T). Altogether 12.5% isolates (50 *S. aureus* and four CoNS) were multi-drug resistant. From the methicillin-resistant *S. aureus* (MRSA) isolates, 43.1% were multi-drug resistant. Methicillin-resistant CoNS were not multi-drug resistant, with the most common resistance pattern being ampicillin-rifampicin-cefoxitin (AP-RP-FOX). In conclusion, multi-drug resistance in our study was relatively low. However, some of the MRSA isolates were multi-drug resistant, which is of concern. Learners should be educated on the importance of handwashing and appropriate use of antibiotics to prevent spread of antibiotic-resistant bacteria within the community. Ciprofloxacin and gentamicin may effectively be used to treat staphylococcal infections in this study population.

Keywords

Staphylococcus, Resistance Trends, Children, Namibia

1. Introduction

Healthy school children under 16 years are potential carriers of *Staphylococcus aureus*, especially methicillin-resistant *S. aureus* (MRSA) and multi-drug resistant strains [1]. According to [2], children are asymptomatic reservoirs for community-associated MRSA (CA-MRSA) which enables these bacteria to rapidly spread within communities. Nasal colonization with *S. aureus* is a major risk factor for staphylococcal infection [3]. Most staphylococcal infections can easily be cleared with antibiotics, but bacteria that develop resistance towards certain antibiotics make treatment options limited, especially if antibiograms for reference purposes are unavailable. Methicillin-resistant *S. aureus* is resistant to beta-lactam antibiotics, while some strains are multi-drug resistant. Drug resistant strains are often responsible for chronic, persistent and recurrent infections, which is a challenge for healthcare practitioners.

Antibiotic resistance data is limited in Namibia and few reports on staphylococcal drug resistance exist. Our study aimed to make a contribution towards closing this gap in information by providing resistance data for 433 *S. aureus* and coagulase-negative staphylococci (CoNS) isolates from nasal swabs of children aged 6 - 14 years attending schools in the Mariental District. To our knowledge, this is the first report on resistance trends of nasal staphylococcal isolates from Namibian school children.

2. Materials and Methods

2.1. Study Area, Population and Sample Collection

This was a cross-sectional study in the town of Mariental, located southeast of Namibia's capital city of Windhoek on the B1 national highway. With written consent from their parents/guardians, the population that was screened for nasal staphylococci consisted of randomly chosen healthy learners attending five schools in the Mariental District. The children were divided into two age-groups: 6 - 10 years and 11 - 14 years, and consisted of 126 boys and 146 girls. Sample collection was done during March, September and October 2016. One nasal specimen was obtained from each child by gently rotating a sterile Amies transport medium swab (Labocare™, Johannesburg, South Africa) thoroughly around the perimeter of both nostrils. Specimens were kept frozen at −20˚C until transporting them to the University of Namibia's Biomedical Research Laboratory for processing.

2.2. Bacterial Cultures Used

Two commercially obtained reference strains, *S. aureus* ATCC 25923 (an antibi-

otic susceptible strain) and *S. aureus* ATCC 33591 (a multi-drug resistant MRSA strain} (Microbiologics®, St. Cloud, US), as well as 433 staphylococcal nasal isolates from 272 Mariental school children, were used in antibiotic assays. Of these isolates, 352 were *S. aureus* and 81 were CoNS.

2.3. Antibiotic Susceptibility Testing

Antibiotic resistance testing on isolates obtained from the nasal specimens was carried out in 2017. The Kirby-Bauer disk diffusion assay was used to determine antibiotic susceptibility/resistance in isolates [4] [5] [6] [7]. **Table 1** shows the antibiotics that were used and interpretation of inhibition zones. *Staphylococcus aureus* ATCC 25923, *S. aureus* ATCC 33591 (MRSA), 352 nasal *S. aureus* isolates and 81 nasal CoNS were tested. Resistance towards cefoxitin (30 µg), in other words, an inhibition zone diameter < 22 mm, indicated MRSA or methicillin-resistant CoNS (MRCoNS), while resistance towards three or more different classes of antibiotics was an indication of multi-drug resistance in bacteria. By direct colony suspension, three to five well-isolated colonies from overnight tryptone soy agar (Scharlau Microbiology, Spain) plate cultures were inoculated into 10 ml sterile phosphate-buffered saline pH 6.8 - 7.4 (Skylabs, Johannesburg, SA) and adjusted to 0.5 McFarland standard (1.5×10^8 CFU/ml, absorbance reading 0.08 - 0.13 at 625 nm). Adjusted cultures were then swabbed onto Mueller-Hinton agar (Mast Diagnostics, Merseyside, UK) and left to dry for 5 - 10 minutes at room temperature before dispensing the antibiotic disks (Mast Diagnostics, Merseyside, UK) onto the plates. Plates were incubated at 35˚C for 18 - 20 hours and diameters were measured using a ruler. Isolates were classified as susceptible, resistant or intermediately resistant towards each antibiotic, according to the diameter (in millimetres) of their zones of inhibition. The reference strains *S. aureus* ATCC 25923 (susceptible) and *S. aureus* ATCC 33591

Table 1. Antibiotics used in this study and interpretation of inhibition zones of test cultures. Adapted from [6] and [7].

Chemical class	Antibiotic	Disk symbol	Disk content	Resistant	Intermediate	Susceptible
Aminoglycosides	Gentamicin	GM	10 µg	<18 mm *S. aureus* <22 mm CoNS	-	≥18 mm *S. aureus* ≥22 mm CoNS
β-lactams	Ampicillin	AP	25 µg	<18 mm	-	≥18 mm
Cephalosporins (also a *β*-lactam)	Cefoxitin	FOX	30 µg	<22 mm	-	≥22 mm
Fluoroquinolones	Ciprofloxacin	CIP	5 µg	<20 mm	-	≥20 mm
Macrolides	Erythromycin	E	15 µg	<18 mm	18 - 20 mm	≥21 mm
Tetracyclines	Tetracycline	T	30 µg	<19 mm	19 - 21 mm	≥22 mm
Other	Rifampicin	RP	5 µg	<23 mm	23 - 25 mm	≥26 mm

(multi-drug resistant MRSA) served as quality control strains. Clinical and Laboratory Standards Institute (CLSI) guidelines [6] and the European Committee on Antimicrobial Susceptibility Testing (EUCAST) breakpoint tables [7] were used to interpret results.

2.4. Statistical Analysis

A chi-square test for comparison of proportions with MedCalc statistical software (MedCalc statistical software version 16.4.3 {MedCalc software bvba, Ostend, Belgium; https://www.medcalc.org; 2016}) [8] was used to compare percentage differences in antibiotic resistance between *S. aureus* and CoNS. Statistical significant differences were indicated by a P-value of ≤ 0.05.

3. Results and Discussion

3.1. Antibiotic Susceptibility/Resistance of Isolates

Altogether 433 staphylococcal isolates, as well as two reference strains, underwent antibiotic susceptibility testing against seven antibiotics (**Table 1**). Excluding the reference strains, 352 of these were *S. aureus* and 81 were CoNS. Only 4.8% of isolates were susceptible to all antibiotics tested. As expected, *S. aureus* ATCC 25923 was susceptible to all antibiotics, while *S. aureus* ATCC 33591 (MRSA) was resistant to cefoxitin and also multi-drug resistant.

Table 2 summarizes percentage susceptibility, intermediacy or resistance of the 352 *S. aureus* isolates to the antibiotics. Most isolates (96.0%) were resistant to ampicillin, rendering this antibiotic mostly ineffective against *S. aureus*. For CoNS, 66.7% of isolates were also resistant to ampicillin (**Table 3**). However, resistance towards ampicillin was significantly higher in *S. aureus* than in CoNS ($P < 0.0001$). Resistance towards ampicillin has become very common in staphylococci and can be ascribed to the action of the enzyme beta-lactamase which is under plasmid control. Plasmids containing resistance genes can be transferred from one bacterium to another [9]. Other studies also indicated high ampicillin resistance in *S. aureus*; in northeastern Brazil, for example, de Carvalho *et al.*, 2017 [10] observed 80.0% ampicillin resistance in *S. aureus* from nasal secretions of children attending public daycare. In our study, ampicillin was still effective against 33.3% of coagulase-negative isolates. Ampicillin should not be the drug of choice for treating infections caused by *S. aureus* and CoNS, unless used in combination with other drugs.

As indicated in **Table 2** and **Table 3**, respectively, a total of 83 (23.6%) *S. aureus* isolates and nine (11.1%) CoNS were resistant to erythromycin ($P = 0.0064$). These resistance rates are not that high and erythromycin is therefore expected to be effective against staphylococci in most instances. In comparison to our study, a study by Mengistu *et al.*, 2013 [11] using Namibia Institute of Pathology (NIP) data showed higher resistance (32.3%) to erythromycin in *Staphylococcus* isolated from cerebrospinal fluid (CSF) for the period 2009-2012. Another Namibian study undertaken by Iileka *et al.*, 2016 [12] reported a lower

Table 2. Percentage susceptibility, intermediacy or resistance for *S. aureus* isolates (n = 352).

Antibiotic	Susceptible (%)	Intermediate (%)	Resistant (%)
Ampicillin 10 µg	14 (4.0)	-	338 (96.0)
Cefoxitin 30 µg	301 (85.5)	-	51 (14.5)
Ciprofloxacin 5 µg	351 (99.7)	-	1 (0.3)
Erythromycin 15 µg	238 (67.6)	31 (8.8)	83 (23.6)
Gentamicin 10 µg	328 (93.2)	-	24 (6.8)
Rifampicin 5 µg	248 (70.4)	40 (11.4)	64 (18.2)
Tetracycline 30 µg	232 (66.0)	60 (17.0)	60 (17.0)

Table 3. Percentage susceptibility, intermediacy or resistance for CoNS isolates (n = 81).

Antibiotic	Susceptible (%)	Intermediate (%)	Resistant (%)
Ampicillin 10 µg	27 (33.3)	-	54 (66.7)
Cefoxitin 30 µg	74 (91.4)	-	7 (8.6)
Ciprofloxacin 5 µg	81 (100.0)	-	0 (0)
Erythromycin 15 µg	65 (80.2)	8 (9.9)	8 (9.9)
Gentamicin 10 µg	72 (88.9)	-	9 (11.1)
Rifampicin 5 µg	55 (67.9)	12 (14.8)	14 (17.3)
Tetracycline 30 µg	60 (74.1)	14 (17.3)	7 (8.6)

erythromycin percentage resistance (10.2%) than ours for *S. aureus* isolates obtained from various clinical samples over the time period 2012-2014. According to PathCare Namibia data [13] 11.0% *S. aureus* bacteria were resistant to erythromycin from 2014 to 2015, which is very close to the 10.2% indicated by Iileka *et al.*, 2016 [12]. In Brazil de Carvalho *et al.*, 2017 [10] observed a higher percentage (32.8%) erythromycin resistance for *S. aureus* compared to our results. Their percentage resistance is however almost the same as the 32.3% from Mengistu *et al.*, 2013 [11].

In the current study, only 17.0% *S. aureus* and 8.6% CoNS (P = 0.0595) were resistant to tetracycline, indicating its effectiveness against *Staphylococcus*. According to Mengistu *et al.*, 2013 [11] 29.6% of staphylococci from CSF displayed resistance towards tetracycline. In agreement with our results, Iileka *et al.*, 2016 [12] reported 17.4% tetracycline resistance in clinical *S. aureus* strains across the period 2012-2014. The resistance for *S. aureus* in our study towards this antibiotic is higher than the 4.3% found by de Carvalho *et al.*, 2017 [10] in Brazil.

Rifampicin resistance in our study was relatively low at 18.2% and 17.3% (P = 0.8495) for *S. aureus* and CoNS isolates, respectively. However, this is somewhat higher than the 7.0% indicated in other Namibian resistance data [13]. Rifampicin resistance in other countries may be higher. In India for instance, Bharathi *et al.*, 2014 [14] observed 64.7% resistance in nasal MRSA from school children.

Rifampicin is among the antibiotics commonly used to treat MRSA infections in India [14]. Staphylococci can quickly develop resistance to rifampicin [9].

According to our results, ciprofloxacin and gentamicin were most effective against *S. aureus*, with 99.7% and 93.2% of isolates that were susceptible to these drugs, respectively. Ciprofloxacin was also the most effective drug against CoNS, with 100.0% susceptibility. Resistance between *S. aureus* and CoNS for the two antibiotics was not statistically significant (P = 0.6220; P = 0.1886). In line with our results, de Carvalho *et al.*, 2017 [10] observed 92.9% *S. aureus* susceptibility towards ciprofloxacin in Brazil. In previous Namibian studies Mengistu *et al.*, 2013 [11] reported 19.0% ciprofloxacin resistance and 52.9% gentamicin resistance for *Staphylococcus* from CSF samples, whereas Iileka *et al.*, 2016 [12] reported a low 4.4% ciprofloxacin resistance for *S. aureus* clinical isolates. Namibian susceptibility data for 2011-2012 [13] showed *S. aureus* to be 96.0% susceptible to ciprofloxacin, while data for 2014-2015 indicated 95.0% susceptibility to gentamicin. Based on our findings, ciprofloxacin and gentamicin could be used to treat certain staphylococcal infections in our study population. However, taking into consideration the 52.9% resistance towards gentamicin in CSF *Staphylococcus* observed by Iileka *et al.*, 2016 [12], care should be taken by medical doctors when prescribing gentamicin for meningitis in Namibia.

In this study, cefoxitin was used to detect methicillin-resistant bacteria. These bacteria are also resistant to all beta-lactam antibiotics. A total of 51/352 (14.5%) *S. aureus* isolates were resistant to cefoxitin and therefore identified as MRSA, whereas 7/81 (8.6%) of CoNS isolates were MRCoNS. Cefoxitin resistance between *S. aureus* and CoNS were statistically insignificant (P = 0.1603). Our MRSA prevalence is close to the 13.8% cefoxitin-resistant *S. aureus* reported by Reta *et al.*, 2015 [15] in a community based cross-sectional study that involved children aged 6 - 12 years from nine primary schools in Bahir Dar Town, Ethiopia. It is also in line with other Namibian data that showed 13.6% MRSA (2010-2014) and 13.5% MRSA (2012-2014) obtained from various clinical specimens by Festus *et al.*, 2016 [16] and Iileka *et al.*, 2016 [12]. Considering these relatively low percentages, MRSA does not seem to be a major problem in Namibia yet. In general, there are not many studies available on methicillin resistance in CoNS. According to [17], MRSA and MRCoNS can be found together in the human nose and have similar antibiotics resistance genes that can be transferred between bacteria. We could not find studies involving nasal antibiotic resistant staphylococci from healthy school children in our neighboring countries (Angola, Zambia, Botswana and South Africa) to compare our results with.

3.2. Resistance Patterns and Multi-Drug Resistance

Altogether 31 antibiotic resistance patterns were observed in this study (**Table 4**). Isolates with similar resistance patterns could be considered the same strain, unless they acquired these resistance genes from other strains, therefore sharing the same resistance pattern. For *S. aureus*, 27 different resistance patterns were obtained. For CoNS, there were 14 different patterns. Ten patterns (AP, RP,

Table 4. Resistance patterns for 433 *S. aureus* and CoNS isolates from school children aged 6 - 14 years, against seven antibiotics.

Number of isolates with this pattern	Number of *S. aureus* isolates with this pattern	Number of CoNS isolates with this pattern	Antibiotic resistance pattern
194	162	32	AP
2	0	2	E
2	0	2	GM
4	3	1	RP
2	0	2	T
52	50	2	AP-E
19	14	5	AP-GM
11	8	3	AP-RP
23	22	1	AP-T
8	8	0	AP-FOX
1	1	0	GM-E
2	2	0	T-RP
1	1	0	RP-E
1	1	0	AP-CIP-FOX
5	5	0	AP-E-FOX
3	3	0	AP-GM-E
5	4	1	AP-GM-RP
1	1	0	AP-GM-T
4	4	0	AP-RP-E
19	12	7	AP-RP-FOX
9	8	1	AP-T-E
7	7	0	AP-T-RP
3	3	0	AP-T-FOX
1	0	1	GM-T-E
1	1	0	AP-GM-RP-FOX
5	5	0	AP-RP-E-FOX
1	1	0	AP-T-E-FOX
2	1	1	AP-T-RP-E
11	11	0	AP-T-RP-FOX
3	3	0	AP-T-RP-E-FOX
1	1	0	AP-GM-T-RP-E-FOX

Key: Multi-drug resistant ; Multi-drug resistant MRSA ; AP—ampicillin (10 µg); CIP—ciprofloxacin (5 µg); (E—erythromycin (15 µg); FOX—cefoxitin (30 µg); GM—gentamicin (10 µg); RP—rifampicin (5 µg); T—tetracycline (30 µg).

AP-E, AP-GM, AP-RP, AP-T, AP-GM-RP, AP-RP-FOX, AP-T-E and AP-T-RP-E) were shared by *S. aureus* and CoNS. AP-RP-FOX was the only pattern shared

between MRSA and MRCoNS. The three most frequently encountered patterns were AP, AP-E and AP-T, with 194, 52 and 23 isolates having these, respectively.

Fifty-four out of 433 isolates (12.5%) were resistant against three or more classes of antibiotics and classified as multi-drug resistant. Of these, 50 isolates were *S. aureus* and four were CoNS. The most common multi-drug resistant pattern for methicillin-susceptible isolates was AP-T-E, displayed by eight *S. aureus* isolates, and one coagulase-negative isolate. Of the 51 MRSA isolates, 22 (43.1%) were multi-drug resistant, with AP-T-RP-FOX as the most encountered resistance pattern among them (See **Figure 1**). This is of concern, but is 14.5% less than the 57.6% multi-drug resistant MRSA isolated from Iranian children by Erami *et al.*, 2014 [18], and 20.5% less than the 63.6% from Brazilian children as reported by de Carvalho *et al.*, 2017 [10]. One of the multi-drug resistant MRSA isolates in our study showed resistance towards 6/7 antibiotics tested with only ciprofloxacin that was effective against it. Methicillin-resistant CoNS did not show multi-drug resistance, with the most common resistance pattern being AP-RP-FOX. Overall, our findings support the conclusion by Arali *et al.*, 2016 [1] that healthy school children under the age of 16 years are potential carriers of MRSA and multi-drug resistant strains.

4. Conclusion

Our study showed the presence of antibiotic-resistant strains among healthy school children. Overall, multi-drug resistance was relatively low. However, some of the MRSA isolates were multi-drug resistant, which is of concern. Learners should be encouraged to frequently wash their hands to prevent spread of antibiotic-resistant bacteria within the Mariental community and educated on

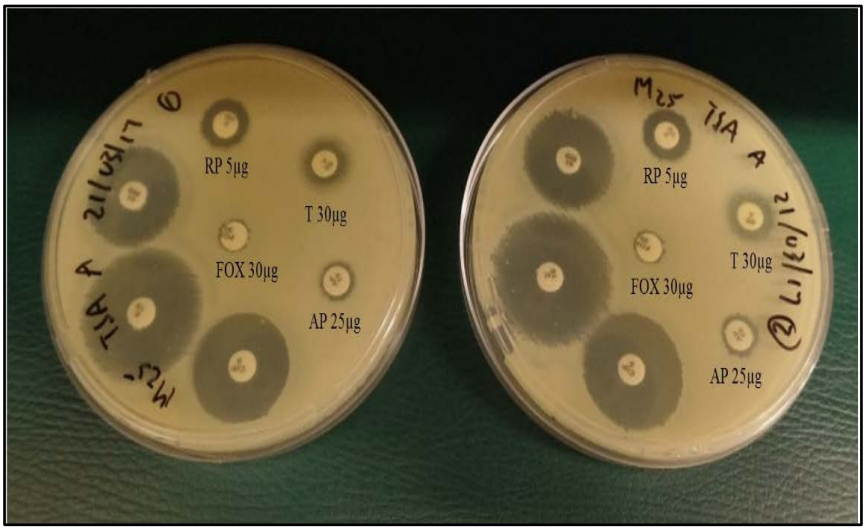

Figure 1. Duplicate Kirby-Bauer disk diffusion assay on Mueller-Hinton agar showing reduced inhibition zones of multi-drug resistant MRSA isolated from a 9-year-old girl (resistance pattern AP-T-RP-FOX). The left three germicidal circles indicate susceptibility of the bacteria against 10 µg gentamicin (top), 15 µg erythromycin (middle) and 5 µg ciprofloxacin (bottom).

the appropriate use of antibiotics. Ciprofloxacin and gentamicin may effectively be used to treat staphylococcal infections in this study population.

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Conflicts of Interest

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

References

[1] Arali, S.M., Kulkarni, V., Manjula, N.G., Gaddad, S.M., Jayaraj, Y.M. and Shivannavar, C.T. (2016) Prevalence and Antibiotic Susceptibility Profile of MRSA Isolated from the Anterior Nares of School Going Children in Gulbarga. *International Journal of Medical Science and Public Health*, **5**, 620-626. https://doi.org/10.5455/ijmsph.2016.0805201592

[2] Alzoubi, H.M., Agel, A.A., Al-Sarayreh, S.A. and Al-Zayadneh, E. (2014) Methicillin-Resistant *Staphylococcus aureus* Nasal Carriage among Primary School-Aged Children from Jordan: Prevalence, Antibiotic Resistance and Molecular Characteristics. *Journal of Egyptian Public Health Association*, **9**, 114-118. https://doi.org/10.1097/01.EPX.0000454671.83406.e8

[3] Reid, M.J.A., Fischer, R.S.B., Mannathoko, N., Muthoga, C., McHugh, E., Essigmann, H., Brown, E.L. and Steenhoff, A.P. (2017) Prevalence of *Staphylococcus aureus* Nasal Carriage in Human Immunodeficiency Virus-Infected and Uninfected Children in Botswana: Prevalence and Risk Factors. *The American Journal of Tropical Medicine and Hygiene*, **96**, 795-801. https://doi.org/10.4269/ajtmh.16-0650

[4] Harley, J.P. and Prescott, L.M. (2002) Laboratory Exercises in Microbiology. 5th Edition, McGraw-Hill, New York.

[5] British Society for Antimicrobial Chemotherapy (BSAC) Methods for Antimicrobial Susceptibility Testing Version 6.0.

[6] Clinical and Laboratory Standards Institute (CLSI) (2013) Performance Standards for Antimicrobial Susceptibility Testing: Twenty-Third Informational Supplement. CLSI Document M100-S23. CLSI, Wayne.

[7] European Committee on Antimicrobial Susceptibility Testing (EUCAST) (2017) Breakpoint Tables for Interpretation of MICs and Zone Diameters Version 7.1. http://eucast.org

[8] MedCalc (2016) MedCalc Statistical Software Version 16.4.3. https://www.medcalc.org

[9] Brooks, G.F., Carroll, K.C., Butel, J.S., Morse, S.A. and Mietzner, T.A. (2013) Jawetz, Melnick & Adelberg's Medical Microbiology. 26th Edition, McGraw-Hill, New York.

[10] de Carvalho, S.P., de Almeida, J.B., Andrade, Y.M.F.S., da Silva, L.S.C., de Oliveira,

M.V., Timenetsky, J. and Marques, L.M. (2017) Community-Acquired Methicillin-Resistant *Staphylococcus aureus* Carrying SCCmec Type IV and V Isolated from Healthy Children Attending Public Daycare in Northeastern Brazil. *The Brazilian Journal of Infectious Diseases*, **21**, 464-467. https://doi.org/10.1016/j.bjid.2017.04.001

[11] Mengistu, A., Gaeseb, J., Uaaka, G., Ndjavera, C., Kambyambya, K., Indongo, L., Kalemeera, F., Ntege, C., Mabirizi, D., Joshi, M.P. and Sagwa, E. (2013) Antimicrobial Sensitivity Patterns of Cerebrospinal Fluid (CSF) Isolates in Namibia: Implications for Empirical Antibiotic Treatment of Meningitis. *Journal of Pharmaceutical Policy and Practice*, **6**, Article No. 4. https://doi.org/10.1186/2052-3211-6-4

[12] Iileka, A.E.K., Mukesi, M., Engelbrecht, F. and Moyo, S.R. (2016) Antimicrobial Susceptibility Patterns of *Staphylococcus aureus* Strains Isolated at the Namibia Institute of Pathology 2012 to 2014. *Open Journal of Medical Microbiology*, **6**, 116-124. https://doi.org/10.4236/ojmm.2016.63016

[13] PathCare Namibia (2015) Antimicrobial Susceptibility Data. http://196.25.36.50/webfiles/files/ANTIMICROBIAL_SUSCEPTIBILITY_DATA_Staphylococcus_aureus.pdf

[14] Bharathi, M., Lakshmi, N., Kalyani, C.S. and Padmaja, I.J. (2014) Nasal Carriage of Multidrug-Resistant MSSA and MRSA in Children of Municipality Schools [Letter to the Editor]. *Indian Journal of Medical Microbiology*, **32**, 200. http://www.ijmm.org/text.asp?2014/32/2/200/129840 https://doi.org/10.4103/0255-0857.129840

[15] Reta, A., Gedefaw, L., Sewunet, T. and Beyene, G. (2015) Nasal Carriage, Risk Factors and Antimicrobial Susceptibility Pattern of Methicillin-Resistant *Staphylococcus aureus* among School Children in Ethiopia. *Journal of Medical Microbiology and Diagnosis*, **4**, 177. https://www.omicsonline.org/peer-reviewed/nasal-carriage-risk-factors-and-antimicrobial-susceptibility-pattern-of-methicillin-resistant-staphylococcus-aureus-among-school-c-42812.html

[16] Festus, T., Mukesil, M. and Moyo, S.R. (2016). The Distribution of Methicillin Resistant *Staphylococcus aureus* Isolated at the Namibia Institute of Pathology in Windhoek, Namibia. *Indian Journal of Medical Research and Pharmaceutical Sciences*, **3**, 1-8. https://pdfs.semanticscholar.org/7b04/5d14227962c4ae3671ca82eafe43ec9e062c.pdf

[17] Jayadev-Menon, P., Humphreys, H. and Fitzgerald-Hughes, D. (2015) Could Coagulase Negative Staphylococci Be an Evolutionary Source of Resistance Genes for *Staphylococcus aureus? BMC Proceedings*, **9**, A43. http://www.biomedcentral.com/1753-6561/9/S1/A43 https://doi.org/10.1186/1753-6561-9-S1-A43

[18] Erami, M., Soltani, B., Ardakani, A.T., Moravveji, A., Rezaei, M.H., Soltani, S. and Moniri, R. (2014) Nasal Carriage and Resistance Pattern of Multidrug Resistant *Staphylococcus aureus* among Healthy Children in Kashan, Iran. *Iranian Red Crescent Medical Journal*, **16**, e21346. https://www.researchgate.net/publication/280977442_Nasal_Carriage_Rates_of_Staphylococcus_aureus_and_CA-Methicillin_Resistant_Staphylococcus_aureus_among_University_Students https://doi.org/10.5812/ircmj.21346