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Original Research Article

Causes of Bacterial and Fungal External Eye Infections and Their Antibiotic Susceptibility Patterns Among Children in Owerri, Imo State, Nigeria

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ABSTRACT

This study is a cross-sectional survey to isolate and identify the causes of bacterial and fungal external eye infections among children in Owerri, Imo State, Nigeria. It also considered the antibiotic susceptibility patterns of the organisms identified. Ocular samples of the discharge and corneal scrapings were collected from infected eyes of 460 children included in the study. Culture and microscopy with gram staining of samples, biochemical tests were used for the identification of bacterial and fungal colonies. Susceptibility tests were also carried out to ascertain the susceptibility pattern of the organisms. Statistical analysis was done with the statistical package for social sciences (SPSS), version 20.1, using Chi-square, analysis of variance (ANOVA), correlation test and simple percentage. P-value ≤0.05 was considered statistically significant. Out of the ocular samples collected, 132(14%) yielded positive culture growth. Of this number, 126 (95.5%) yielded bacterial growth, while 6(4.55%) yielded fungal growth. Among the bacterial isolates, gram positive bacteria were the most prevalent (88.1%). *Coagulase negative staphylococcus aureus* was the predominant isolate (34.8%) causing bacterial eye infections, followed by *staphylococcus aureus (*22.73%). *Aspergillus sp.* was the common cause of fungal infections on the cornea. Ceftriaxone had the highest susceptibility effect on all bacterial isolates (96%), followed by vancomycin (95.6%). Gram positive isolates were highly sensitive to ceftriaxone and ofloxacin (95.4% each), clindamycin (91.5%) and vancomycin (90.8%) while gram negative isolates were totally sensitive to ciprofloxacin (100%) and highly sensitive to clindamycin and ceftriaxone (97.4% each).

Keywords: Bacteria, Fungi, External eye, Isolation, Antibiotics, Susceptibility.

INTRODUCTION

Infection is an invasion of an organism's body tissues by causative agents. These infections can predispose the ocular structures to potential damage if left unattended, causing significant disabilities and possibly, blindness.[1] Indeed, earlier studies report external eye infections to be among the increasing causes of morbidity and vision loss around the globe.[2-4]

It is usually difficult to ascertain the agent of infection based on clinical presentation as they may or may not be distinguishing. Hence, microbiological procedures involving culture of clinical samples and identification of the causative agents remain the gold standard for diagnosis of causative agents of ocular infection.[3]

Also, deriving specific antimicrobial agents for effective treatment is becoming more tasking as resistance of infective ocular pathogens to commonly used antimicrobials is on the increase. Therefore, specificity of the antimicrobials is necessary to aid in effective management of these external eye infections. This study is aimed at determining the causative agents of bacterial and fungal external ocular infections among children in Owerri. It also ascertained the antibiotic susceptibility patterns of the isolated organisms.

MATERIALS & METHODS

This study was carried out in Federal University Teaching Hospital, Owerri, Imo State, Nigeria. The facility is a tertiary health care institution that serves patients from different parts of the South Eastern Nigeria.

The study was designed as a cross-sectional study that included children with suspected external eye infections. It was carried out in two phases: (i) on-site inspection, clinical evaluation and specimen collection and (ii) laboratory investigation.

Ocular samples were collected from children aged 1-17 years of age on presentation with external eye infections. Children who had no previous history of use of antibiotics were included while those who had antibiotic treatment less than 5 days or had undergone previous eye surgery within 3months before this study were excluded from this study. Ocular samples were collected from the 460 children included in the study.

Study variables

The isolates of bacterial and fungal external eye infections and their susceptibility patterns to antimicrobial agents were the dependent variables.

Research Instruments

The instruments for data collection included sterile gloves, sterile swab sticks, slit lamp biomicroscope, bard Parker blade #15, normal saline, topical anaesthetic, coolant ice packs and ice box.

Laboratory procedure

Bacteriological Analysis

Each ocular specimen collected was subjected to bacteriological analysis where each sample was analysed for the presence of bacterial isolates. The swab sticks were subjected to 10 fold serial dilutions according to the method described by Fawole and Oso.[5] 0.1ml aliquots of the serially diluted samples respectively were inoculated into freshly prepared Nutrient agar, MacConkey agar and Mannitol salt agar for bacteria characterization and identification. The bacteriological assessments include; Total Heterotrophic count (THC), Total Coliform count (TCC) and Total Staphylococci count (TSC).

Identification of Isolates

Identification of Bacterial Isolates

Pure bacterial isolates were identified using methods of Cheesbrough.[6] Representative colonies of bacteria were picked from different plates after the incubation period. They were streaked on sterile nutrient agar plates for purification, followed by characterization using colony morphology, cellular morphology and biochemical tests.

Mycological Analysis

After the processing of the samples, spread plate technique was used in quantifying and detection of microfungi from the samples.[7]

Identification of fungal Isolates

To isolate molds associated with ocular infection examined for microorganisms, serial dilutions were carried out on the specimens.[8,9]

Susceptibility (Antibiogram) of bacterial isolates

The sensitivity of bacterial strains isolated was investigated using the Kirby-Bauer disk diffusion method. A suspension with turbidity equal to 0.5 McFarland standards was provided for each bacterial isolate. Then, the suspensions were placed in Nutrient agar medium and exposed to the different disks of antibiotics and incubated for 24 hours at 37^{o}C. Antibiotic disks included were Vancomycin (30µg), Tetracycline (20µg), Norfloxacin (10µg), Amoxicillin (20µg), Ofloxacin (5µg), Ampicillin (20µg), Clindamycin (30µg), Ceftriaxone (30µg), Ciprofloxacin (5µg) and Chloramphenicol (30µg). After this period of time, the diameter of growth inhibition zone of each sample was measured and categorized in two distinct groups of sensitive and resistant based on guidelines recommended by Clinical and Laboratory Standards Institute.[10]

STATISTICAL ANALYSIS

Statistical analysis was done with the SPSS version 20.1, using chi square, analysis of variance (ANOVA), correlation and simple percentage. P-value less than or equal to 0.05 was considered statistically significant.

Risk control

Adequate quality control measures were taken to ensure credible findings throughout the laboratory process. All materials, equipment and procedures were handled carefully, following stipulated guidelines for laboratory work. The questionnaire was written in English for consistency and required data on socio-demographic characteristics and ocular history were collected by the researcher. All specimens were collected by the researcher, while maintaining standard operating procedures (SOPs) for collection of external eye specimens. The sterility of the culture media was ensured by incubating un-inoculated media from each batch of specimens. The integrity of all prepared culture media was checked by inoculating standard strains.

RESULTS

Bacterial conjunctivitis was the major bacterial eye infection occurring among the participants (74.6%), followed by blepharitis (11.3%), bacterial keratitis (5%), hordeolum (3.9%), lid abscess (2.2%) while the least occurring bacterial infection was dacryocystitis (1.7%) (t=1.795, df=6, p=0.0156). Fungal keratitis (1.3%) was the only fungal infection seen among the participants.

Table 1: Distribution of bacterial and fungal isolates among children in Owerri.

Microorganism	Frequency (%)
CoNS	46(34.85)
S. aureus	30(22.73)
P. aeruginosa	24(18.18)
N. gonorrhoeae	10(7.58)
S. pneumonia	10(7.58)
K. pneumonia	5(3.79)
S. haemolytica	1(0.76)
Aspergillus sp.	6(4.55)
Total	132(100)

From the 132 isolates seen, 126 were bacterial isolates while the remaining 6 were fungal isolates The most frequent isolate identified was *Coagulase Negative Staphylococcus aureus* **(***CoNS)* 46(34.85%), and the least seen, *Staphylococcus haemolytica* 1(0.76%). Only one specie of fungal isolates was identified, *Aspergillus fumigatus* 6(4.55%) (table 1).

Table 2: Distribution of gram positive and gram negative bacterial isolates

Gram positive	Frequency (%)	Gram negative	Frequency (%)
N. gonorrhea	10(10.3)	*K. pneumonia*	5(17.24)
S. pneumonia	10(10.3)	*P. aeruginosa*	24(82.75)
S. aureus	30(30.93)		
CoNS	46(47.42)		
S. haemolytica	1(1.03)		
Total	97(76.98)		29(23.02)

Out of the 126 bacterial isolates, the gram positive isolates were predominant (76.98%) while there was a low prevalence of the gram negative bacterial isolates (23.02%) Among the gram positive bacterial isolates, *CoNS* (47.42%) was the predominant isolate while *Pseudomonas aeruginosa* 24(82.75%) was the predominant gram negative isolate identified (table 2).

Table 3: Distribution of bacterial and fungal isolates among clinical features

Microorganism	**Frequency (%)**	**Lid abscess (%)**	**Bacterial Conjunctivitis (%)**	**Blepharitis (%)**	**Bacterial Keratitis (%)**	**Dacryocystitis (%)**	**Hordeolum (%)**	**Fungal Keratitis (%)**
Neisseria gonorrhea	10(7.58)	0(0.0)	8(10.13)	0(0.0)	2(16.67)	0(0.0)	0(0.0)	0(0.0)
Klebsiella pneumonia	5(3.79)	0(0.0)	3(3.80)	2(11.11)	0(0.00)	0(0.0)	0(0.0)	0(0.0)
Streptococcus pneumonia	10(7.58)	0(0.0)	6(7.59)	2(11.11)	0(0.00)	2(40.00)	0(0.0)	0(0.0)
Staphylococcus aureus	30(22.73)	4(100)	12(17.72)	6(33.33)	2(16.67)	2(40.00)	2(25.00)	0(0.0)
CoNS	46(34.85)	0(0.0)	36(45.57)	4(22.22)	2(16.67)	0(0.00)	4(50.00)	0(0.0)
Pseudomonas aeruginosa	24(18.18)	0(0.0)	12(15.19)	4(22.22)	6(50.00)	0(0.00)	2(25.00)	0(0.0)
Staphylococcus haemolytica	1(0.76)	0(0.0)	0(0.0)	0(0.0)	0(0.00)	1(20.0)	0(0.0)	0(0.0)
Aspergillus fumigatus	6(4.55)	0(0.0)	0(0.0)	0(0.0)	0(0.00)	0(0.00)	0(0.0)	6(100)
Total	132(100)	4(3.03)	79(59.85)	18(13.64)	12(9.09)	5(3.79)	8(6.06)	6(4.55)

CoNS was the predominant isolate (45. 57%) causing bacterial conjunctivitis, blepharitis was mostly caused by *S. aureus* (33.33%), lid abscess was wholly caused by *S. aureus* (100%). For bacterial keratitis, it was mostly caused by *P.aeruginosa* (50%), while dacryocystitis was mostly caused by *S. pneumonia* and *S. aureus* (40% each) and hordeolum mostly caused by *CoNS* (50%). *Aspergillus sp.* was totally (100%) responsible for the few cases of fungal keratitis (table 3).

Table 4: Susceptibility of bacterial isolates of external eye infections to antibiotics among children in Owerri

Organism	**Frequency of Occurrence (%)**	**Sensitive/Resistant**	**Vancomycin (%)**	**Tetracycline (%)**	**Norfloxacin (%)**	**Amoxicillin (%)**	**Ofloxacin (%)**	**Ampicillin (%)**	**Clindamycin (%)**	**Ceftriaxone (%)**	**Ciprofloxacin (%)**	**Chloramphenicol (%)**
Neisseria gonorrhoeae	10(7.93)	S R	10(100) 0(0)	9(90) 1(10)	9(90) 1(10)	6(60.0) 4(40.0)	8(80.0) 2(20.0)	5(50.0) 5(50.0)	10(100.0) 0(0)	9(90.0) 1(10.0)	10(100.0) 0(0)	7(70.0) 3(30.0)
Klebsiella pneumoniae	5(3.97)	S R	4(80.0) 1(20.0)	4(80.0) 1(20.0)	4(80.0) 1(20.0)	3(60.0) 2(40.0)	4(80.0) 1(20.0)	2(40.0) 3(60.0)	5(100.0) 0(0)	5(100.0) 0(0)	5(100.0) 0(0)	4(80.0) 1(20)
Streptococcus pneumonia	10(7.94)	S R	9(90.0) 1(10.0)	4(40.0) 6(60.0)	8(80.0) 1(20.0)	6(60.0) 4(40.0)	10(100.0) 0(0)	6(60.0) 4(40.0)	10(100.0) 0(0)	9(90.0) 1(10.0)	10(100) 0(0)	6(60.0) 4(40.0)
Staphylococcus aureus	30(23.81)	S R	26(86.7) 4(13.3)	22(73.3) 8(26.67)	29(96.7) 1(3.3)	18(60.0) 12(40.0)	27(90.0) 3(10.0)	20(66.7) 10(33.3)	29(96.7) 13.3)	29(96.7) 1(3.3)	25(83.3) 5(16.7)	20(66.7) 10(33.3)
CoNS	46(36.51)	S R	43(93.5) 3(6.52)	41(89.1) 5(10.9)	40(87.0) 6(13.0)	36(78.3) 10(21.7)	45(97.8) 1(2.2)	34(73.9) 12(26.1)	40(87.0) 6(13.0)	44(95.7) 2(4.3)	43(93.5) 3(6.5)	41(89.1) 5(10.9)
Pseudomonas aeruginosa	24(19.05)	S R	20(83.3) 4(16.7)	19(79.2) 5(20.8)	21(87.5) 3(12.5)	21(87.5) 3(12.5)	24(100.0) 0(0)	21(87.5) 3(12.5)	23(95.8) 1(4.2)	24(100.0) 0(0)	24(100.0) 0(0)	21(87.5) 3(12.5)
Staphylococcus haemolytica	1(0.79)	S R	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	1(100.0) 0(0)	0(0) 1(10 0)	1(100) 0(0)
Total	126(100)	S R	113(89.7) 13(10.3)	100(79.4) 26(20.6)	112(88.9) 14(11.1)	91(72.2) 35(27.8)	119(94.4) 7(5.6)	89(70.6) 37(29.4)	118(93.7) 8(6.3)	121(96.0) 5(4.0)	117(92.9) 9(7.1)	99(78.6) 27(21.4)

The highest significant effect of vancomycin was on *N. gonorrhea* and *S. haemolytica* (p=0.0272). That of tetracycline, norfloxacin, amoxicillin and ampicillin was on *S. haemolytica* (p=0.0375, 0.0263, 0.0347, 0.0356 respectively), ofloxacin on *S. pneumonia*, *P. aeruginosa* and *S. haemolytica* (p=0.0138), clindamycin on *N. gonorrhea*, *klebsiella pneumonia*, *S. pneumonia* and *S. haemolytica* (p=0.0199), ceftriaxone on *klebsiella pneumonia*, *P. aeruginosa* and *S. haemolytica* (p=0.0259). Ciprofloxacin on *N. gonorrhoeae*, *klebsiella pneumonia*, *S. pneumonia* and *P.*

aeruginosa (p=0.0246). The highest significant effect of chloramphenicol was on *CoNS* (p=0.0395). Ceftriaxone recorded the highest sensitivity effect (96.0%) among all bacterial species isolated (r^2=0.3100, F=258.7, P =<0.0001) (table 4).

Table 5: Percentage Susceptibility of gram positive and gram negative bacterial isolates of external eye infections to antibiotics among children in Owerri

Organism	Frequency (%)	Sensitive/Resistant	Vancomycin (%)	Tetracycline (%)	Norfloxacin (%)	Amoxicillin (%)	Ofloxacin (%)	Ampicillin (%)	Clindamycin (%)	Ceftriaxone (%)	Ciprofloxacin (%)	Chloramphenicol (%)
Gram positive	97(76.98)	**S**	91.8	79.4	89.7	69.1	93.8	68.0	92.8	94.8	90.7	77.3
		R	8.2	20.6	10.3	30.9	6.2	32.0	7.2	5.2	9.3	22.7
Gram negative	29(23.02)	**S**	82.8	79.3	86.2	82.8	96.6	79.3	96.6	100	100	86.2
		R	17.2	20.7	13.8	17.2	3.4	20.7	3.4	0.0	0.0	13.8

The gram positive isolates were mostly sensitive to ceftriaxone (94.8%) and least sensitive to ampicillin (68.0% each) while the gram negative isolates were totally sensitive to ceftriaxone and ciprofloxacin (100% each) and least sensitive to tetracycline and ampicillin (79.3% each) (table 5).

DISCUSSION

Several microbial pathogens can gain access to the eye from external sources, adjacent orbital tissues or through systemic circulation.[11] The eye is protected from infection by a combination of epithelial and mucus membranes which serve as anatomic, immunologic, microbiologic and mechanical barriers in warding off ocular infections and preventing the growth of microorganisms capable of causing diseases in the eye. At any rate, ocular infection, which occurs when these exogenous harmful organisms gain entrance to the eye, threatens the survival of the pathogenic species in the eye.[12,13] The results of this study report notable similarities in comparison to other studies conducted though certain dissimilarities abound in comparison to other likely studies.

Out of the 132 culture positive isolates, 126 (95.45%) bacterial isolates were identified and only 6 (4.55%) culture positive fungal isolates were seen. This is in line with a study conducted in India[13] where the level of bacterial isolates identified was higher (54.4%) than fungal isolates (33.3%). However, both studies do not tally in terms of magnitude.

The gram positive bacteria (76.98%) were the most prevalent isolates in this study as against the gram negative bacteria (23.02%). This result is in agreement with a study in Nigeria[1]. Also, in Quinghai province, China[14] gram positive bacteria made up a higher percentage (55.78%) of the bacteria isolated among children. It is also supported by several other studies in India,[15,16] Ethiopia[17-19] and Sudan,[20] where the most common bacterial isolates were gram positive. The high predominance of gram positive bacteria may be ascribed to infection of the eye from normal flora of the skin resulting from touching the eyes with hands.[18]

Among the gram positive bacteria, *CoNS* (34.85% equivalent to 47.42% of gram positive bacteria) was the most prevalent isolate, followed by *S. aureus* (22.73% equivalent to 30.93% of gram positive bacteria). This is consistent with findings by Suja et al.[13] Mazin et al.[20] Summaiya et al.[21] Muluye et al.[22] and Assefa et al.[23] In contrast, several other previous studies reported *S.aureus* as the prevalent isolated pathogen from ocular infections.[1,2,15,20,24] These variations may be caused by differences in climatic, geographic and environmental conditions and more opportunities for the conjunctival sac to get infected by the bacteria. Moreso, the high virulence characteristic of these pathogens such as a surface slime which may be involved in their pathogenesis. [2,13,14,24]

Gram negative bacteria were least isolated (23.02%) with *P. aeruginosa* being the most prevalent at 7.58%, equivalent to 82.75% of the gram negative bacterial isolates. This is in agreement with the study by Tesfaye et al.[17] but in contrast with the study by Mazin et al.[20] where *H. influenza* was the predominant gram negative bacteria with an overall prevalence of 17%, equivalent to 40% of the gram negative bacteria isolated. In some other studies, *E. coli*[22] and *Proteus sp.*[24] were the major isolates. The low prevalence of gram negative bacteria could be as a result of improved hygiene, as the major mode of transmission for enteric organisms is fecal-hand contamination of the eye.[19]

Generally, *S. pneumonia* had the least frequency of occurrence (0.76%) of all the isolates in this study. This finding does not support result of a study in Nigeria, where *N. meningitis* was the least occurring isolate.[1] It also disagrees with findings from other studies where *M. catarhalis*,[17,20] *H. influenza*[14] and *Enterobacter* [20] were the least occurring isolates. There was limited isolates of enteric bacteria, (*K. pneumonia*, 3.79%), which is in line with the studies by Getahun et al.[9] Ubani,[10] Tesfaye et al.[17] Suja et al.[13] and Mazin et al.[20] but in contrast to studies by Esenwah[25] and Anagaw et al.[18] This can be attributed to the factors relative to the surrounding conditions, period of study, environment and biological disparities of the isolates. It may also be ascribed to reduction in hand-fecal contamination of the eye resulting from accessibility of potable water supply in majority of households and neighbourhoods.[1]

Aspergillus fumigatus (4.55%) was the only fungal isolate seen in this study. Another study, however, reports *fusarium sp.* as the major fungal isolate identified.[13] This may be due to differences in environmental and ethnic conditions.

In this study, *CoNS* was the major isolate causing conjunctivitis (45.57%) followed by *S. aureus* (17.72%), *P. aeruginosa* (15.19%), *N. gonorrhea* (10.13%), *S. pneumonia* (7.59%) and *klebsiella pneumonia* (3.80%). This is consistent with studies by Suja et al.[13] where *CoNS* was the major cause of conjunctivitis but contrasts with studies in Nigeria, by Esenwah[25] and Ubani.[1] as well as some other studies in Sudan and South India[16,20] where *S. aureus* was the predominant isolate causing conjunctivitis. However, the conjunctiva is prone to infection by several microorganisms and major routes of infection are airborne droplets, hand to eye contact and infection around the ocular adnexa including the lacrimal system, nose and paranasal sinuses.[16]

Blepharitis was mostly caused by *S. aureus* (33.33%), followed by *CoNS* and *P.aeruginosa* (22.22% each), *klebsiella pneumonia* and *S. pneumonia* (11.11% each). This result is in order with a atudy in Ethiopia,[25] where *S. aureus* was the major cause of blepharitis, followed by *CoNS.*

P. aeruginosa was the major isolate (50%) seen in bacterial keratitis in this study, followed by *N. gonorrhea*, *CoNS* and *S. aureus* (16.67% each). This is in support of the studies carried out in Nigeria and Jimma.[1,17] It also agrees with a study in China and Malaysia,[26,27] where *P. aeruginosa* was also found to be the predominant cause of microbial keratitis. This result is however, in contrast with studies in Ethiopia and India,[4,28] where *S.aureus* and *CoNS* were the major causes of keratitis. These variations may be attributed to variations in study population, environmental differences, period of study, corneal status/health and geographical location. Also, *P.aeruginosa* is a part of the normal flora of the cornea and can promote infection when there is mechanical trauma to the corneal epithelium, stroma or both. This causes the production of exotoxin A, which leads to necrosis of the tissue, resulting in corneal ulceration.[17,29,30]

The organisms implicated in dacryocystitis were *S. aureus* and *S. pneumonia,* which accounted for 40% each, followed by *S. haemolyticus*, accounting for 20% of the isolates identified. This supports previous studies where *S. aureus* and *S. pneumonia* were both implicated in equal proportion as the most predominant causes of dacryocystitis.[20,23] On the other hand, it contradicts studies in India and Ethiopia,[2,16,31] which reported *S. pneumonia* as the predominant isolate in dacryocystitis. Another study in contrast reported *S. aureus* and *CoNS* as accounting for 50% each of the predominant isolates in dacryocystitis.[4]

Among the isolates seen in hordeolum, *CoNS* was the predominant (50%) cause of the eyelid infection this was followed by *S. aureus* and *P. aeruginosa* (25% each). This result is in contrast with the study in Ethiopia,[2] where S. aureus was the predominant isolate causing hordeolum. All cases of lid abscess in this study yielded *S. aureus* (100%) as the only isolate. This finding is in line with previous studies conducted by Mazin et al.,[20] Blomquist[32] and Ackay et al.[33] and where *S. aureus* was the sole isolate in all case of orbital cellulitis with eyelid abscess. However, it contradicts the findings by Amsalu et al.[2] where *Serratia marcescens* was the major isolate in all cases of lid abscess, followed by *S. pneumonia.* Studies have shown that the eyelid margins create enabling environment for infection and generally involve the skin, eyelashes and associated glands anterior to the gray line or mucocutaneous junction.[16]

Aspergillus sp. was totally (100%) responsible for the few cases of fungal keratitis.

Based on the results and comparative analysis above, it is noteworthy that various organisms play a role in the pathogenesis of infections while targeting the susceptible organs in the population under study. Moreso, some of the organisms constitute the normal ocular flora, while some do not. Despite *CoNS, S. aureus, streptococcus* and other bacteria like some *Neisseria sp.* being part of the normal conjunctival flora, in certain conditions, they become implicated in ocular infections.[14,20]

The high rate of infection of infection of *CoNS* and *S. aureus* among the clinical features may be as a result of high virulence factor/pathogenicity such as exoenzyme and surface slime, which enable them to multiply and spread rapidly in tissues.[2,24] *S. aureus* generates numerous extracellular substances like coagulase, which deposits fibrin on the surface of the microorganism, decreasing the intake of the pathogen by phagocytic cells. These antiphagocytic capsules constitute the cell surfaces. The organism also secretes virulence factors like luekocidin and haemolysins (alpha toxin) which breakdown erythrocytes and destroy platelets.[29,34,35] *CoNS* can give rise to infections in patients with low immunity and exposure to use of medical devices.[36]

Pseudomonas is the most virulent corneal pathogen though seen as opportunistic. It possesses features like glycocalyx and pili for adherence and biofilms as coating which facilitate their adherence to targets.[37,38] They also give rise to melting of the corneal stroma due to effects of enzymes.[39] *N. gonorrhea's* virulence characteristics include porin, opa proteins and pili (van Vliet *et al.*, 2009). *Klebsiella* possesses conspicuous antiphagocytic capsule and MagA.[40]

Susceptibility testing in this study showed *N. gonorrhea* was completely sensitive to vancomycin, clindamycin and ciprofloxacin (100% respectively). It was also highly sensitive to tetracycline, norfloxacin and ceftriaxone (90% respectively), ofloxacin (80%) and chloramphenicol (70%). It was less sensitive to amoxicillin (60%) and ampicillin (50%). *Klebsiella pneumonia* was completely sensitive to clindamycin, ceftriaxone and ciprofloxacin (100% respectively). It was highly sensitive to vancomycin, tetracycline, norfloxacin, ofloxacin and chloramphenicol (80% respectively). It was sensitive to lesser extent to amoxicillin (60%) but was resistant to ampicillin to ampicillin (60%). *S. pneumonia* was completely sensitive to ofloxacin, clindamycin and ciprofloxacin (100% respectively). It was highly sensitive to vancomycin (90%), ceftriaxone (90%), norfloxacin (80%) and sensitive to lesser extent to amoxicillin, ampicillin and chloramphenicol (60% respectively) but was resistant to tetracycline (60%). *S. aureus* was highly sensitive to norfloxacin, clindamycin, ceftriaxone (96.7% respectively), ofloxacin (90%), vancomycin (86.7%) ciprofloxacin (83.3%) and tetracycline (73.3%). It was less sensitive to ampicillin and chloramphenicol (66.7% respectively). *CoNS* was highly sensitive to all antibiotics tested; ofloxacin (97.8%), ceftriaxone (95.7%), vancomycin and ciprofloxacin (93.5% respectively), tetracycline and chloramphenicol (89.9% respectively), norfloxacin and clindamycin (87% respectively), amoxicillin (78.3%) and ampicillin (73.9%). *P. aeruginosa* was completely sensitive to ofloxacin, ceftriaxone and ciprofloxacin (100% respectively). It was highly sensitive to clindamycin (95.8%), norfloxacin, amoxicillin, ampicillin and chloramphenicol (87.5% respectively), vancomycin (83.3%) and the least sensitivity being to tetracycline (79.2%). The single isolate of *S. haemolyticus* identified was completely (100%) sensitive to all the antibiotics used except ciprofloxacin to which it was completely (100%) resistant.

In other words, vancomycin recorded 100% significant effect on *N. gonorrhea* and *S. haemolytica.* Tetracycline, norfloxacin, amoxicillin and ampicillin recorded 100% significant effect on *S. haemolytica,* respectively. Ofloxacin recorded 100% significant effect on *S. pneumonia*, *P. aeruginosa* and *S. haemolytica.* Clindamycin recorded 100% significant effect on *N. gonorrhea, klebsiella pneumonia, S. pneumonia* and *S. haemolytica.* Ceftriaxone recorded 100% significant effect on *klebsiella pneumonia*, *P. aeruginosa* and *S. haemolytica.* Ciprofloxacin recorded 100% significant effect on *N. gonorrhoeae*, *klebsiella pneumonia, S. pneumonia* and *P. aeruginosa*. The highest significant effect of chloramphenicol was recorded in *CoNS* (89.1%).

Ceftriaxone had the highest susceptibility effect on all bacterial isolates (96%). This is in contrast with findings by Amsalu et al.[2] where most of the bacterial isolates showed the highest susceptibility to vancomycin (95.6%).

The gram positive isolates were mostly sensitive to ceftriaxone (94.8%), followed by ofloxacin (93.8%), clindamycin (92.8%), vancomycin (91.8%), Ciprofloxacin (90.7%), norfloxacin (89.7%) and Tetracycline (79.4%), chloramphenicol (77.3%), amoxicillin (69.1%) and the least sensitivity to ampicillin (68.0%). This finding contrasts with that of Tesfaye et al.[17] and Bharati et al.[15] which reported ciprofloxacin to have the highest susceptibility on all gram positive isolates. It also contradicts the study by Mazin et al.[20] which reported highest susceptibility of gram positive isolates to vancomycin (95%), followed by chloramphenicol and ciprofloxacin (91% each) and then ceftriaxone (84%). However, this study and the study by Bharati et al.[15] Tesfaye et al.[17] and Amsalu et al.[2] agree in terms of these antibiotics having high susceptibility patterns on the gram positive isolates.

The gram negative isolates were mostly sensitive to ceftriaxone and ciprofloxacin (100% each), clindamycin and ofloxacin (96.6% each), norfloxacin and chloramphenicol (86.2% each), vancomycin and amoxicillin (82.8% each) and the least being tetracycline and ampicillin (79.3% each). This is consistent with the findings by Amsalu et al.[2] where ciprofloxacin showed higher susceptibility effect on the gram negative isolates. Nonetheless, it contradicts the study by Mazin et al.[20] which reported gram negative bacteria were highly susceptible to amikacin (92.7%) followed by ceftriaxone (87.3%) and then ciprofloxacin (78.2%).

The gram positive bacteria identified in this study were mostly resistant to tetracycline and ampicillin (20.7% each) and then to vancomycin and amoxicillin (17.2% each) whereas the gram negative isolates showed more resistance to ampicillin and tetracycline had equal and highest resistance effect (20.7% each). This result also contradicts a study in Ethiopia,[2] where amoxicillin and ampicillin had equal resistance effect (56.5%) on the gram negative isolates.

However, all strains of the isolated bacteria were mostly resistant to ampicillin (29.4%). This is in contrast with the study by Amsalu et al.[2] where major resistance of most of the bacterial isolates was seen with penicillin. The resistance to antibiotics may be due to initial exposure of the isolates to the drugs (termed first lined drugs) as there is common use of these antibiotics for ocular infections due to ease of purchase at low cost and without prescription in different localities which often result to increased drug resistance.[2,41]

CONCLUSION

Bacteria were the most frequent cause of external eye infections among the subjects, especially, gram positive isolates. *CoNS* were the major bacteria causing external eye infections among the participants and were highly susceptible to ceftriaxone while the gram negative isolates were mostly sensitive to both ceftriaxone and ciprofloxacin. In all, ceftriaxone had the highest susceptibility effect on all bacterial isolates identified. However, increased antibiotic resistance may occur due to indiscriminate use of antibiotics without prescription, inappropriate dosage regimen, abuse for viral and other non-bacterial infections, long term use and poor compliance.[4,15,42]

Declaration by Authors

Ethical Approval: Ethical clearance was gotten from the Federal University Teaching Hospital, Owerri, Imo State, Nigeria, for specimen collection from the eye and pediatric units. Consent was also gotten from subjects or their parents/guardians before inclusion in the study.

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