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Abstract

Background: Conjunctival bacterial infections are common among children living in internally displaced persons (IDP) camps due to poor hygiene and crowded living conditions. This study aimed to identify the bacterial isolates from conjunctival swabs of children residing in IDP camps in Makurdi, Benue State, Nigeria.

Materials and Methods: A cross-sectional study was conducted among children under 18 years old living in IDP camps in Makurdi. Conjunctival swabs were collected from 100 participants who met the inclusion criteria. The swabs were cultured on various media including blood agar, MacConkey agar, and nutrient agar. Bacterial identification was performed using Gram staining and biochemical tests. Data analysis was done using SPSS version 23.

Results: Out of 100 samples, bacterial growth was observed in 89 cases. The most frequently isolated bacteria were *Staphylococcus aureus* (39%), followed by *Bacillus spp.* (25%), and *Streptococcus spp.* (16%). Other isolates included *Escherichia coli* (6%), *Enterococcus spp.* (2%), and *Klebsiella pneumoniae* (1%). No bacterial growth was observed in 11% of the samples. Gender distribution showed a slightly higher prevalence of bacterial isolates among females (52%) compared to males (48%).

Conclusion: The high prevalence of *Staphylococcus aureus* indicates it as a primary bacterial pathogen associated with conjunctival infections in children at IDP camps. This suggests the need for improved hygiene and targeted interventions to reduce bacterial transmission.

Keywords: Conjunctival Swabs; IDP Camps; Bacterial Isolates; Staphylococcus aureus; Conjunctival Infections

Introduction

The conjunctiva, a thin, transparent mucous membrane covering the anterior surface of the sclera and lining the inside of the eyelids, serves as a primary barrier against microbial invasion. While generally equipped with effective defense mechanisms such as tears containing lysozyme, lactoferrin, and immunoglobulins, the conjunctiva can become vulnerable to bacterial infections under compromised hygiene, overcrowded conditions, and environmental stressors [1]. Conjunctivitis, commonly referred to as pink eye, is a leading manifestation of bacterial infections affecting the conjunctiva. The condition can cause discomfort, discharge, and vision impairment if left untreated, especially in children with limited access to healthcare [2].

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Children in Internally Displaced Persons (IDP) camps face increased risks of conjunctival infections due to crowded living conditions, poor sanitation, and inadequate medical services [3]. Such camps often serve as breeding grounds for the proliferation of pathogens due to close physical contact and environmental contamination. In this setting, bacterial conjunctivitis may go undiagnosed or untreated, leading to secondary complications such as corneal ulcers or systemic infections [4].

The epidemiology of bacterial conjunctivitis varies by region, influenced by climate, socioeconomic conditions, and healthcare access. Common bacterial pathogens implicated in conjunctival infections include *Staphylococcus aureus*, *Streptococcus pneumoniae* and *Haemophilus influenzae*. However, emerging resistance patterns among these bacteria present significant public health challenges. Surveillance and antibiotic susceptibility testing are critical in guiding appropriate therapy, particularly in resource-limited settings like IDP camps [5].

Despite the public health significance of bacterial conjunctival infections, there is limited research focusing on their prevalence and bacterial etiology in IDP camps, particularly in Nigeria. Makurdi, Benue State, hosts a considerable number of displaced persons due to communal conflicts and herdsmen crises, placing a strain on local resources and healthcare systems [6]. Understanding the bacterial profile and antibiotic resistance patterns in this population is essential for effective management and policy formulation.

Aim of the Study

The present study aims to isolate and identify bacterial pathogens from conjunctival swabs of children residing in IDP camps in Makurdi, Benue State, Nigeria. It seeks to fill the knowledge gap in the microbial landscape of conjunctival infections in displaced populations and provide evidence-based recommendations for improving child eye health in IDP settings.

Materials and Methods

Research design

The research is a cross-sectional study of which children living in IDP camps in Makurdi, Benue State who are below 18 years old. Demographic data and external examination were done. Conjunctival swabs were collected and taken to the laboratory for isolation, culturing, and identification.

Study area

The location of the study was an internally displaced persons (IDP) camp in Makurdi metropolitan city in Benue State, North Central Nigeria. The decimal latitude and longitude coordinates for Makurdi are 7.7322° and 8.5391°, and the elevation above sea level of 301 feet in Benue State. Makurdi has a population is 454,000 living and working there. Major ethnic groups include Tiv, Idoma, Igede, Jukun, Agatu, Etulo, Alago, and Igbo.

Population of study

For the context of this research, the population of study interest included children from internally displaced people camps in the Makurdi metropolis of Benue state, Nigeria. The age range is children under 18 years old and gender include both male and female.

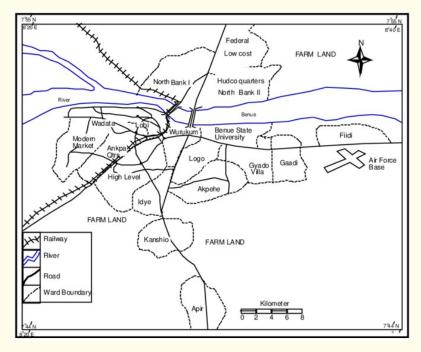


Figure 1: Map of Makurdi in Benue State [7].

Inclusion criteria

Those subjects that qualified to be part of the study include:

- 1. Children in official and unofficial internally displaced camps who are below 18 years old.
- 2. Children not clinically diagnosed with ocular infection.
- 3. Children that are willing to participate.

Exclusion criteria

- 1. Children who will not give consent.
- 2. Children who have lived less than 3 months in the camps.
- 3. Children who used antibiotics treatment past two weeks.

Sample size determination

The sample size was determined using the Cochran formula for estimating proportions in a population outlined by Airaodion., *et al.* [8]:

$$n = \frac{Z^2(Pq)}{e^2}$$

Where n = minimum sample size

Z = 1.96 at 95% confidence level,

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P = known number of children in IDP comps in Nigeria

e = error margin tolerated at 5% = 0.05

q = 1 - p.

According to Ukase and Terungwa [7], the total population of children in official and unofficial internally displaced people camps in Makurdi metropolis is 272,061. Considering this number, children make up 6.4% of the population people living in internally displaced people camps.

P = 6.4% = 0.064

q = 1 - p

= 1 - 0.064

= 0.936

 $n = \frac{(1.96)^2 (0.064 \ge 0.936)}{(0.05)^2}$

 $n = \frac{3.8416 \text{ x} (0.0.599)}{0.0025}$

 $n = \frac{0.2301}{0.0025} = 92.05$

The minimum sample size was 92 but was adjusted to 100 to account for a non-response rate of 10%.

Sample technique

For this research, a systematic sampling technique were used. The systematic sampling technique is where individuals from a larger population are to be selected according to a random starting point on a fixed periodic interval (the sampling interval).

Validation of instruments

The instruments used in this study were approved for laboratory use by the medical laboratory science council of Nigeria and the World Optometric Association; they are therefore valid for conducting clinical ocular examinations.

Procedure for data collection

Data collection is an important part of any type of research study. If collected inaccurately, data can impact the results of a study and ultimately lead to invalid findings. An informed consent were obtained from the subjects involved. The case history of the subject were taken to determine information about their socio-demographic status, ocular history and past medical history.

Penlight examination

External examination using penlight were conducted immediately after the case history has been taken on the subjects to observe the eyes.

Procedure for sample collection

Swabs were used to collect microorganism specimens using sterile cotton swabs from the conjunctiva of subjects.

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- 1. These swabs were held immediately in their containers to avoid contamination from air microbes for a controlled experiment during transportation to the laboratory.
- 2. In the laboratory, the agar plates would have been prepared for use and the samples and swabs were inoculated into the agar for culturing.

Procedure for media preparation

Bacterial culture and identification methods

Upon the completion of the brain heart infusion broth preparation in the microbiology laboratory, the broth was subjected to an incubation process at a temperature of 37°C for 24 - 48 hours. Following the successful incubation of the BHI broth, the next step involved sub-culturing the broth on various cultural media, including blood agar plate (BAP), chocolate agar plate (CAP), mannitol salt agar (MSA), and MacConkey agar. The BAP, MSA, and MacConkey agar were inoculated and incubated at a temperature of 37°C for a duration of 24 to about 48 hours, while the CAP were incubated at a temperature of 37°C with 5% CO₂ for a duration of 24 - 48 hrs. After 48 hours, cultural media that exhibited no visible growth will eventually be discarded.

The preliminary identification of bacterial isolates were carried out through the investigation of their colony morphology, gram stain, and hemolytic reactions on blood agar plates. The discovery of bacteria will further be facilitated via the use of a series of routine biochemical tests. Based on the results of the gram reaction, catalase, and coagulase tests, gram-positive cocci were successfully identified. Furthermore, to distinguish gram-negative bacteria, a suspension of the test organism was prepared by inserting 3 - 4 colonies of the test species into 5 ml of nutrient broth. A loop full of the bacterial suspension will then be inoculated into Indole, citrate agar, triple sugar iron agar, lysine decarboxylase agar, urea agar, and motility medium, which were subsequently incubated at a temperature of 35 - 37°C for 24 hours.

The identification of gram-negative organisms was achieved through the observation of a shift in color for acid production, gas production, H₂S production, and medium motility turbidity.

Data analysis

The data were presented inform of tables and figures. It was uploaded into the statistical package for social sciences (SPSS) software version 23 for analysis. Analysis was done using the independent sample Test and paired sample Test.

Ethical consent

Ethical consent was obtained from the ethical committee of the School of Health Technology, Federal University of Technology, Owerri.

Results

A total of 100 subjects were included, with equal representation of males and females (50 males and 50 females). In the 4 - 8 years age group, 9 males (9%) and 4 females (4%) were recorded, accounting for 13% of the total sample. The 9 - 13 years age group had the largest representation, with 22 males (22%) and 21 females (21%), comprising 50% of the total. The 14 - 18 years age group included 19 males (19%) and 25 females (25%), accounting for 41% of the total subjects (Table 1).

Age group	Males		Females		Total		
	n	%	n	%	N	%	
4 - 8	9	9.00	4	4.00	13	13.00	
9 - 13	22	22.00	21	21.00	43	50.00	
14 - 18	19	19.00	25	25.00	44	41.00	
	50	50.00	50	50.00	100	100.00	

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The bacterial isolates were distributed among males and females, with some differences in frequency. The most common bacterial isolate overall was *Staphylococcus aureus* (39%), followed by *Bacillus spp.* (25%) and *Streptococcus spp.* (16%). Among males, *Staphylococcus aureus* was the most frequent (40%), followed by *Bacillus spp.* (24%). In females, *Bacillus spp.* was the most prevalent (26%), with *Staphylococcus aureus* (38%) slightly less common than in males. Other bacteria observed include *Escherichia coli* (6%), *Enterococcus spp.* (2%), and *Klebsiella pneumoniae* (1%), with a small number of subjects showing no bacterial growth (11%) (Table 2).

Bacteria		Male	Female		Total	
	n	%	n	%	n	%
Bacillus spp	12	24.00	13	26.00	25	25.00
Enterococcus spp	0		2	4.00	2	2.00
Escherichia coli	1	2.00	5	10.00	6	6.00
klebsiella pneumonia spp	1	2.00	0		1	1.00
Staphylococcus aureus	20	40.00	19	38.00	39	39.00
Streptococcus spp	10	20.00	6	12.00	16	16.00
No growth	6	12.00	5	10.00	11	11.00

Table 2: Frequency distribution of bacterial isolate among subjects.

Different bacteria showed variable growth on two media types. *Bacillus spp.* and *Staphylococcus aureus* showed growth only on Nutrient agar, while *Enterococcus spp., Escherichia coli*, and *Klebsiella pneumoniae* grew exclusively on MacConkey agar. *Streptococcus spp.* grew on Nutrient agar only. This indicates the differing media preferences for bacterial growth based on their characteristics (Table 3).

Bacteria	MacConkey	Nutrient agar
Bacillus spp	-	+
Enterococcus spp	+	-
Escherichia coli	+	-
klebsiella pneumonia spp	-	+
Staphylococcus aureus	_	+
Streptococcus spp	_	+

Table 3: Frequency distribution of bacterial isolate growth on media.

The morphological characteristics of the bacterial isolates showed notable patterns. For example, *Bacillus spp.* was whitish, rodshaped, flat with undulated margins, and grew on Nutrient agar. *Staphylococcus aureus* appeared yellow, spherical, raised with entire margins, also growing on Nutrient agar. Similarly, *Streptococcus spp.* showed a creamy color, spherical shape, raised elevation, and entire margins, growing exclusively on nutrient agar. Other bacteria, such as *Escherichia coli* and *Klebsiella pneumoniae*, showed pink and grey colors, respectively, with similar rod shapes but varying elevations and margins (Table 4).

Microorganism	n	Color	Shape	Elevation	Margin	Media
Bacillus	25	Whitish	Rod	Flat	Undulated	Nutrient agar
Enterococcus	2	pink	Rod	Flat	Entire	MacConkey
Escherichia coli	6	Pink	Rod	Flat	Undulated	MacConkey
Klebsiella pneumonia	1	Grey	Rod	Flat	Undulated	Nutrient agar
Staphylococcus aureus	39	Yellow	Spherical	Raised	Entire	Nutrient agar
Streptococcus spp	16	Creamy	Spherical	Raised	Entire	Nutrient agar

Table 4: Frequency distribution of bacterial growth morphology characteristics for all subjects.

The bacterial morphology characteristics in females were consistent with the overall results, although some differences in frequency were noted. For example, *Bacillus spp.* in females was also whitish, rod-shaped, flat with undulated margins, and grew on Nutrient agar. The distribution of other bacteria such as *Staphylococcus aureus* (19 females), *Streptococcus spp.* (6 females), and *Escherichia coli* (5 females) followed similar trends as the general sample, with *Klebsiella pneumoniae* absent in females. This highlights the similarity in bacterial characteristics between genders, with slight variations in the frequency of specific bacterial types (Table 5).

Microorganism	n	Color	Shape	Elevation	Margin	Media
Bacillus	13	Whitish	Rod	Flat	Undulated	Nutrient agar
Enterococcus	2	pink	Rod	Flat	Entire	MacConkey
Escherichia coli	5	Pink	Rod	Flat	Undulated	MacConkey
Klebsiella pneumonia	0	Grey	Rod	Flat	Undulated	Nutrient agar
Staphylococcus aureus	19	Yellow	Spherical	Raised	Entire	Nutrient agar
Streptococcus spp	6	Creamy	Spherical	Raised	Entire	Nutrient agar

Table 5: Frequency distribution of bacterial growth morphology characteristics in female.

Discussion

This present study investigated the bacterial isolates from conjunctival swabs of children residing in Internally Displaced Persons (IDP) camps in Makurdi, Benue State, Nigeria. The demographic data showed that the majority of participants were between 9 to 18 years, accounting for 91% of the study population. Specifically, the age group 9 - 13 years had the highest representation at 50%, followed closely by 14 - 18 years at 41%. The younger age group of 4 - 8 years constituted only 13% of the total sample. This age distribution highlights that older children (9 - 18 years) are more susceptible to bacterial colonization of the conjunctiva or may reflect greater exposure to environmental factors that facilitate bacterial transmission. Gender distribution was equal, allowing for a balanced analysis of bacterial isolates between males and females.

The frequency analysis of bacterial isolates revealed a diverse spectrum of bacterial species, with *Staphylococcus aureus* being the most prevalent, identified in 39% of the total samples. This finding aligns with previous studies indicating *Staphylococcus aureus* as a common colonizer of the conjunctiva in children, particularly in environments with limited access to healthcare and poor hygiene practices [9,10]. The high prevalence of *Staphylococcus aureus* may be attributed to its ability to thrive on skin and mucous membranes, making it a frequent contaminant, especially in crowded living conditions such as IDP camps.

Bacillus spp. was the second most common isolate, found in 25% of samples. The presence of *Bacillus spp.* is often linked to environmental exposure since it is ubiquitous in soil and dust [11]. The relatively high percentage of *Bacillus spp.* indicates potential environmental

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contamination, underscoring the need for improved sanitation and hygiene in IDP camps. The detection of *Streptococcus spp.* in 16% of samples further supports this observation, as these bacteria are commonly associated with respiratory and skin infections that can spread through close contact in overcrowded settings [12].

Notably, *Enterococcus spp.* and *Escherichia coli* were found in 2% and 6% of the samples, respectively. The low prevalence of these isolates may reflect their less common association with conjunctival infections in this demographic. However, their presence indicates possible fecal contamination, highlighting inadequate hand hygiene practices, which is a known risk factor in IDP settings [13]. The detection of *Klebsiella pneumoniae* in only one sample suggests that while this bacterium is not a common conjunctival colonizer, it can still be present in cases of systemic or respiratory infections, which are prevalent in such environments.

The assessment of bacterial growth on MacConkey and Nutrient agar revealed significant patterns. *Bacillus spp., Staphylococcus aureus,* and *Streptococcus spp.* demonstrated growth on Nutrient agar but not on MacConkey agar, indicating their gram-positive nature. Conversely, *Escherichia coli* and *Enterococcus spp.* showed growth on MacConkey agar, confirming their gram-negative classification [14]. This growth pattern is consistent with the selective nature of MacConkey agar, which favors gram-negative organisms, and Nutrient agar's ability to support a broad range of bacterial species.

The growth of *Klebsiella pneumoniae* exclusively on Nutrient agar underscores its adaptation to nutrient-rich environments, typical of mucosal surfaces, and highlights its potential as an opportunistic pathogen in compromised hosts [15]. The absence of growth in 11% of the samples suggests that some conjunctival swabs were either devoid of culturable bacteria or contained fastidious organisms that require specialized media, such as those tailored for anaerobic or specific fastidious species like *Haemophilus influenzae* [16].

The most frequently isolated bacterium across all samples was *Staphylococcus aureus*, accounting for 39 out of 89 (43.8%) isolates. This high prevalence aligns with previous studies that identify *S. aureus* as a dominant pathogen in conjunctival infections, particularly in environments with poor hygiene and limited access to healthcare [17,18]. The spherical shape, yellow pigmentation, raised elevation, and entire margin morphology observed on nutrient agar are typical characteristics of *S. aureus*, consistent with the morphological descriptions in similar studies [19].

The dominance of *S. aureus* is concerning due to its known role in causing bacterial conjunctivitis and other ocular infections. Children in IDP camps are particularly susceptible due to overcrowded living conditions, limited water supply, and poor sanitary practices, which increase the risk of transmission [20]. Additionally, *S. aureus* can develop resistance to commonly used antibiotics, raising concerns about the potential for treatment challenges [21].

Bacillus species were the second most common isolates, with 25 occurrences. The whitish, rod-shaped colonies with flat and undulated margins on nutrient agar are indicative of *Bacillus* spp., which are commonly found in soil and dust. Their presence may be linked to environmental exposure, especially in camps with substandard hygiene and frequent dust exposure. While *Bacillus* is typically non-pathogenic, certain species like *Bacillus cereus* can cause ocular infections, particularly in immunocompromised individuals [22].

Analysis of bacterial isolates by gender revealed distinct variations in prevalence. Among female subjects, *Staphylococcus aureus* was the most prevalent (n = 19), followed by Bacillus (n = 13). Interestingly, no *Klebsiella pneumoniae* was isolated from female subjects, suggesting potential gender-related differences in conjunctival flora or exposure risk factors. The presence of *Escherichia coli* (n = 5) and *Enterococcus* (n = 2) among females is also noteworthy, given their typical association with fecal contamination [23]. The prevalence of these organisms could be indicative of poor hygiene practices and inadequate handwashing, which are prevalent in crowded and resource-limited IDP camps.

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The isolation of *Escherichia coli* and *Klebsiella pneumoniae* in this study is of particular concern due to their association with severe ocular infections such as keratitis and endophthalmitis. *E. coli* (6 isolates overall; 5 among females) exhibited pink, rod-shaped colonies on MacConkey agar, consistent with its lactose-fermenting capabilities. The presence of *E. coli* in conjunctival swabs may indicate direct contamination from the environment or poor hygiene, such as touching the eyes with unwashed hands. Previous studies have documented similar findings, linking *E. coli* with conjunctival infections in underprivileged populations [24].

Klebsiella pneumoniae was isolated only once in this study. Although infrequent, its detection is significant due to its potential to cause severe ocular and systemic infections. The pathogen's role in hospital-acquired infections is well documented, and its occurrence in the community setting raises concerns about transmission dynamics and infection control, especially in the confined environment of an IDP camp [25].

The identification of *Enterococcus* (n = 2), with its characteristic pink colonies and flat elevation on MacConkey agar, suggests potential fecal contamination. *Enterococcus* spp. are indicators of poor sanitation and are often associated with fecal-oral transmission. The presence of this organism underscores the need for improved hygiene practices and access to clean water in IDP camps to mitigate the risk of conjunctival and other infections [26].

Furthermore, the detection of *Streptococcus spp.* (n = 16 overall; n = 6 in females) suggests potential for both commensal and pathogenic roles. *Streptococcus* species are normal inhabitants of the human upper respiratory tract but can cause conjunctivitis when there is a disruption in the ocular surface defense mechanisms, which may occur due to environmental irritants or underlying health conditions [27].

The bacterial profile identified in this study shares similarities with findings from other regions with similar socio-economic conditions. For instance, a study by Yusuf, *et al.* [28] in IDP camps in Northern Nigeria reported a high prevalence of *S. aureus* and *Bacillus spp.* in conjunctival swabs, correlating well with our findings. However, the lower incidence of *Klebsiella pneumoniae* and *Enterococcus* in our study could be due to differences in environmental exposure or demographic factors specific to the Makurdi IDP camps.

Limitations of the Study

This study highlights the significant presence of pathogenic bacteria in the conjunctiva of children in IDP camps, indicating a need for targeted interventions. Limitations include the inability to perform antibiotic susceptibility testing, which could provide insights into the potential resistance patterns of these isolates.

Conclusion

The findings indicate a higher prevalence of Gram-positive bacteria, suggesting that the children in these IDP camps may be at increased risk of bacterial conjunctivitis due to poor hygiene, overcrowding, and limited access to healthcare services. The high rate of *Staphylococcus aureus* indicates its role as a major pathogen in ocular infections, which could be due to its ability to colonize the skin and mucosal surfaces. The presence of *Bacillus spp.* and *Escherichia coli*, typically environmental and enteric bacteria, respectively, suggests potential contamination from external sources, likely due to unsanitary living conditions in the camps. These findings underscore the need for urgent public health interventions to prevent and manage bacterial ocular infections in this vulnerable population.

Recommendations

1. Improvement of sanitation and hygiene: Implementing programs to improve personal hygiene, especially handwashing practices among children, is essential. Educational campaigns can raise awareness about the importance of maintaining eye hygiene to reduce the risk of bacterial infections.

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- 2. Routine ocular screening and health checks: Regular ocular health screening for children in IDP camps should be established to detect and treat infections early. This can help in reducing the burden of preventable eye diseases.
- **3. Provision of medical supplies and antibiotics:** Supply of basic antibiotics and eye care medications to the healthcare centers in the IDP camps should be prioritized. This can help in the timely treatment of bacterial conjunctivitis and prevent complications.
- 4. **Training of healthcare workers:** Healthcare workers in IDP camps should receive training on identifying and managing ocular infections. This includes proper collection and handling of conjunctival swabs for accurate diagnosis.
- 5. Enhanced access to clean water and sanitation facilities: Efforts should be made to provide access to clean water and improved sanitation facilities within the IDP camps to reduce environmental contamination that could contribute to the spread of bacteria.
- **6. Strengthening public health policies:** Policymakers should focus on improving the living conditions in IDP camps by addressing the underlying factors that contribute to poor hygiene and overcrowding, thereby reducing the risk of bacterial infections.

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