A Case-Control Study to Determine the Microbiological Spectrum and Antibiotic Sensitivity Patterns in Congenital Nasolacrimal Duct Obstruction

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Abstract

Purpose: To analyze the microbiological spectrum and antibiotic sensitivity patterns in children with congenital nasolacrimal duct obstruction (CNLDO).

Methods: One hundred thirty-four eyes of 123 children in the age group of 0–16 years with a diagnosis of CNLDO who underwent lacrimal surgical procedures were included in this prospective comparative study. Sixty-two children in the age-matched group planned for intraocular surgery with patent nasolacrimal duct were deemed controls. The conjunctival swab after performing Regurgitation on Pressure over the Lacrimal Sac in the CNLDO group and the conjunctival swab in controls were sent for microbiological analysis. Antibiotic susceptibility testing was done for commonly employed antibiotics by the Kirby Bauer disk diffusion method.

Results: Of 134 samples collected in the CNLDO group, 111 (82.8%) samples were culture positive. There were 165 bacteria isolated, among which 139 (84.24% of isolates) were Gram-positive bacteria, and 26 (15.75% of isolates) were Gram-negative. Fungal isolates were obtained in 2.23% of cases. The most common Gram-positive isolate was Staphylococcus epidermidis (S. epidermidis) (n = 51, 30.9% of total isolates), and the most common Gram-negative isolate was Haemophilus influenza species (n = 9, 5.5% of total isolates). Gram-positive isolates were sensitive mostly to gentamicin and vancomycin (95.5% each), and Gram-negative isolates to amikacin (92.3%). Both Gram-positive and Gram-negative isolates were susceptible to gatifloxacin (80% each). Probing outcomes were similar among Gram-positive (success, 84.6%) and Gram-negative (success, 84.0%) organisms.

Conclusions: There was a predominance of Gram-positive isolates in children with CNLDO with S. epidermidis being the most common. The microbiological profile did not have any effect on the outcomes of probing.

Keywords: Congenital nasolacrimal duct obstruction, Methicillin resistance, Staphylococcus epidermidis

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INTRODUCTION

Congenital nasolacrimal duct obstruction (CNLDO) is a common condition among infants with an estimated incidence varying from 1% to 20%.1 It is usually due to a membranous obstruction at the distal end of the nasolacrimal duct where it opens into the inferior meatus of the nose.2 This obstruction creates a mechanical blockage to the outflow of tears causing epiphora. Stagnation of tears in the lacrimal sac creates an ideal environment for microorganisms, predisposing to bacterial infection, and dacryocystitis.3 The clinical spectrum

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in CNLDO can vary from simple watering to acute or chronic dacryocystitis. Rarely, it may lead to preseptal or orbital cellulitis, lacrimal abscess, fistula, or even endophthalmitis. Hydrostatic lacrimal sac massage is the initial management of CNLDO up to 1 year of age as most cases resolve spontaneously by then. CNLDO usually presents with intermittent eyelid crusting and/or mucopurulent discharge. Short courses of topical antibiotics are commonly prescribed when the child presents with mucopurulent discharge during this period.

Prescribing the appropriate antibiotics based on culture sensitivities can prevent chronicity, antibiotic resistance, and NLD stenosis. Since the microbiological profile and especially their antibiotic sensitivity patterns keep changing with time, it is prudent to know the current sensitivity pattern of these organisms as it will guide the empirical choice of drugs in these patients, wherever needed. The present study aims to look for the prevailing organisms in the conjunctival cul-de-sac of patients of CNLDO and their antibiotic sensitivity pattern. We have also compared this microbiological pattern with the age-matched controls.

**Methods**

This was a prospective, nonrandomized, comparative study conducted after the approval of the institutional review board and adhered to the tenets of Declaration of Helsinki. The study was approved by the ethics committee of the institution. All patients underwent baseline ophthalmic evaluation. The diagnosis of CNLDO was based on positive Regurgitation on Pressure over the Lacrimal Sac test. Sixty-two eyes of children in the same age group planned for intraocular surgery with patent nasolacrimal duct were included as controls.

Microbial samples of children with CNLDO were collected by applying pressure over the sac area in the operating room and the refluxing discharge through the punctum was collected in a sterile cotton swab straightaway at the time of performing surgery. Care was taken that the lid margins were not touched during the sample collection to prevent contamination.

For controls, samples were collected by rubbing the swab over the inferior conjunctival fornix of age matched children planned for intraocular surgery and with a patent lacrimal passage. Sterile cotton tip applicators were used for both cases and controls, and they were sealed in long glass tubes and sent to the lab.

Patients complaining of watering due to other causes such as conjunctivitis, blepharitis, keratitis, and hordeolum were excluded from the study. Informed consent was obtained from the parents of the children enrolled in the study. Specimens collected were examined for bacteria and fungus using culture-based techniques. Direct Gram stain and potassium hydroxide mount were done on the collected material. The samples were then inoculated aerobically and anaerobically for 2 days onto different culture media-blood agar, chocolate agar, McConkey agar, brain heart infusion broth, brucella blood agar, and thioglycolate medium, the latter two being an anaerobic medium for bacterial growth. The samples were incubated at 37°C for 2 days. Sabaroud’s dextrose agar media was incubated at 25°C for 12 days to detect fungal growth. Standard phenotypic methods were used to identify bacteria and fungi. A culture was considered to be positive when the same organism was grown on two solid media, or confluent growth on one solid media correlated with the organism seen in the Gram stain.

Vitek-2 microbial identification system (Biomerieux, France) was used to identify rare bacterial isolates.

Kirby Bauer diffusion method was used for testing antibiotic susceptibility. Antibiotic sensitivity testing was done for each isolate, testing for commonly used topical antibiotics.

The antimicrobial spectrum included ciprofloxacin, norfloxacin, gatifloxacin, gentamicin chloramphenicol, cotrimoxazole, erythromycin, cefotaxime, penicillin, and amikacin. Amikacin sensitivity was done only for Gram-negative isolates, while vancomycin and penicillin sensitivity was done for Gram-positive isolates alone. The results of the antibiotic sensitivity test were recorded as sensitive or resistant.

Data were analyzed using the Statistical Package for Social Sciences version 14.0 (IBM, Chicago). One proportion Z test was used for statistical analysis of sensitivity patterns. Anticipating different microbial flora, analysis of the subjects was divided into four groups by age: younger than 2, 2–5, 5–8, and older than 8 years.

Fisher’s exact test was used to determine any statistical association between the age of the patient and the type of microorganism isolated. Univariate binary logistic regression analysis was used to assess the risk factors for treatment outcome failure. \( P < 0.05 \) was considered statistically significant.

**Results**

The study included 134 eyes of 123 children between 0 and 16 years of age who were referred to our Oculoplasty clinic with the diagnosis of CNLDO. The mean age at presentation was 3.41 ± 2.70 years. Among 123 patients, 85 (69.1%) were males, and 38 (30.9%) were females. Left eye (52.9%) involvement was more common. While 11 patients (8.9%) had bilateral involvement, previous probing history was noted in 21 eyes (15.7%).

Among the 134 collected samples in the CNLDO group, 111 (82.8%) had bacterial growth. There was no growth in the swab cultures from 23 eyes (17.1%). Among 111 samples with a positive culture, 45 (40.5%) were polymicrobial. There were 165 bacterial isolates in entirety. Table 1 summarizes the details of the bacterial isolates among cases in our study. One hundred thirty-nine isolates were Gram-positive bacteria accounting for 84.24% of isolates, and 26 (15.75%) were Gram-negative. Twenty different species of bacteria were isolated.
The most common Gram-positive cocci were *Staphylococcus epidermidis* (S. epidermidis) (30.9% of total isolates), followed by *Streptococcus pneumoniae* (S. pneumonia) (21.2%). The most common Gram-positive bacillus isolated was *Corynebacterium* species (13.3%). *Hemophilus influenzae* (H. influenza) (5.5%) was the commonest Gram-negative bacillus. No Gram-negative cocci were isolated. Three fungal isolates (2.23% of samples) were obtained. *Kocuria kristinae* (K. Kristina), *Stenotrophomonas maltophilia*, and *Achromobacter xylosoxidans* (one each) were some of the uncommon bacteria isolated.

The proportion of Gram-positive and Gram-negative organisms among different age groups revealed no statistically significant difference \( P > 0.74 \) [Table 2].

Among 62 control eyes, 19 isolates were obtained, with *S. epidermidis* being the most common isolate (52.6%). Eighteen were Gram-positive (94.7%) and only one Gram-negative organism (5.26%) - *Neisseria* was isolated. No rare bacteria or fungus was isolated from control samples.

The sensitivity pattern of Gram-positive and negative organisms in the CNLDO group is as shown in Figure 1. Gram-positive organisms were most sensitive to gentamicin and vancomycin (95.5% each), followed by chloramphenicol (93.2%) and gatifloxacin (80%). Gram-negative organisms were most sensitive to amikacin (92.3%). Both Gram-positive and Gram-negative isolates were sensitive to gatifloxacin (78.1% and 80%, respectively). Both Gram-positive and Gram-negative organisms exhibited the highest degree of resistance to cotrimoxazole (54.5% and 57.9%, respectively), followed by erythromycin (42.3% and 35%, respectively) [Figure 2].

**Table 1: Details of species isolated in lacrimal sac samples in the congenital nasolacrimal duct obstruction group**

| Number of isolates, frequency, \( n (% \) |
|------------------------------------------|
| **Gram-positive cocci**                  |
| *S. pneumonia*                           | 35 (21.2) |
| Methicillin-sensitive *S. aureus*        | 13 (7.9)  |
| Methicillin-resistant *S. aureus*        | 2 (1.2)   |
| Methicillin-sensitive *S. epidermidis*   | 36 (21.8) |
| Methicillin-resistant *S. epidermidis*   | 15 (9.1)  |
| *S. viridans*                            | 8 (4.8)   |
| *E. faecalis*                            | 1 (0.6)   |
| *K. Kristina*                            | 1 (0.6)   |
| *S. pasteurii*                           | 1 (0.6)   |
| *S. saprophyticus*                       | 1 (0.6)   |
| *Beta hemolytic streptococcus*           | 2 (1.2)   |
| **Gram-positive bacillus**               |
| *Corynebacterium* species                | 22 (13.3) |
| *B. cereus*                              | 1 (0.6)   |
| *P. lactis*                              | 1 (0.6)   |
| **Gram-negative bacillus**               |
| *H. influenzae*                          | 9 (5.5)   |
| *H. parainfluenzae*                      | 5 (3)     |
| *K. pneumoniae*                          | 4 (2.4)   |
| *P. aeruginosa*                          | 3 (1.8)   |
| *P. stutzeri*                            | 2 (1.2)   |
| *E. coli*                                | 1 (0.6)   |
| *A. xylosoxidans*                        | 1 (0.6)   |
| *S. maltophilia*                         | 1 (0.6)   |

*S. pneumonia: Streptococcus pneumonia, S. aureus: Staphylococcus aureus, S. epidermidis: Staphylococcus epidermidis, S. viridans: Streptococcus viridans, E. faecalis: Enterococcus faecalis, K. Kristina: Kocuria Kristina, S. pasteurii: Staphylococcus pasteurii, S. saprophyticus: Staphylococcus saprophyticus, B. cereus: Bacillus cereus, P. lactis: Paenibacillus lactis, H. influenza: Haemophilus influenza, H. influenza: Haemophilus parainfluenza, K. pneumonia: Klebsiella pneumonia, P. aeruginosa: Pseudomonas aeruginosa, P. stutzeri: Pseudomonas stutzeri, E. coli: Escherichia coli, A. xylosoxidans: Achromobacter xylosoxidans, S. maltophilia: Stenotrophomonas maltophilia*

**Table 2: Distribution of Gram-positive and Gram-negative organisms in different age groups in the congenital nasolacrimal duct obstruction group**

|                  | <2 years, \( n (%) \) | 2-5 years, \( n (%) \) | 5-8 years, \( n (%) \) | 8-13 years, \( n (%) \) | Total   |
|------------------|------------------------|------------------------|------------------------|------------------------|---------|
| Gram-positive organisms (\( n \)) | 42 (30.2)              | 69 (49.6)              | 20 (14.4)              | 8 (5.8)                | 139     |
| Gram-negative organisms (\( n \)) | 5 (19.2)               | 15 (57.7)              | 4 (15.4)               | 2 (7.7)                | 26      |
| Total            | 47                     | 84                     | 24                     | 10                     | 165     |

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| Total            | 47                     | 84                     | 24                     | 10                     | 165     |
sensitivity to vancomycin (n = 13, 86.7%), followed by chloramphenicol (n = 12, 80%), tetracycline, gentamicin, and gatifloxacin (n = 11 each, 73.3%).

In the control group, all 18 Gram-positive isolates were sensitive to gentamicin, tobramycin, gatifloxacin, and vancomycin. Five of eighteen (27.8%) were resistant to ciprofloxacin. One MRSE that was sensitive to vancomycin, tobramycin, and gatifloxacin was isolated. Neisseria was sensitive to all the antibiotics tested [Table 3].

Of 134 eyes that underwent a surgical procedure, two had dacryocystorhinostomy, one had balloon dacryoplasty, and the remaining children underwent probing. One hundred thirteen (84.4%) eyes had a successful outcome after a single surgery, while 21 (15.5%) failed. Treatment outcomes were similar among Gram-positive (success rate, 84.6%) and Gram-negative (success rate, 84.0%) organisms (P = 0.82). No statistically significant association was seen between treatment outcome and the presence of polymicrobial growth on culture (P = 0.723).

**Discussion**

The nasolacrimal ducts are not patent in about 20% of full-term neonates. Blocked lacrimal ducts allow bacteria, tears, and other debris to pool on the conjunctiva acting as a medium for bacterial growth. Among infants with CNLDO, Pollard reported 2.9%, and Ffooks reported 1.6% of dacryocystitis. Frequent lacrimal sac massage along with topical antibiotics is considered a conservative method of treatment during infancy. Empirical and irrational broad-spectrum antibiotic therapy is a common practice without culture results that risks the development of resistant microbial flora in the conjunctiva cul-de-sac. Hence, it becomes pertinent to identify the microorganism and their sensitivity patterns. This study’s objective was to determine the current microbiological spectrum among children having CNLDO and antibiotic therapy based on the sensitivity results.

Male sex (n = 85, 69.10%) was predominantly affected similar to noted in earlier studies. The majority of patients had unilateral (n = 122, 91.05%) involvement, and 8.9% (n = 12) cases were bilateral, similar to the reports by Barejaand Soler Machin et al. We noted a slightly higher incidence of the disease on the left side (n = 71, 52.98%) as compared to the right side (n = 63, 47.01%). This too correlates well with an earlier study.

Different microbial spectrums among different age groups are usually expected due to environmental influences, antibiotic exposure in older children, microbiome maturity, and immunologic tutoring. However, Usha et al. did not notice any significant difference in the microbial distribution among different age groups.

Among our cases, 17.1% were culture negative, indicating that the presence of discharge may not always be a sign of infection. Kuchar et al. had negative cultures in 30% of samples among children with CNLDO (n = 50). The majority of earlier studies have shown Gram-positives as a predominant isolate with recent trends towards Gram-negative organisms. We found that Gram-positive organisms (84.24%) were more common, with Gram-negative bacteria comprising only 15.75% of the samples consistent with earlier studies. The results though differ from the study done in the South Indian population by Usha et al., where only 57% of the isolates were Gram-positive.

The most common Gram-positive isolates among cases in our study were *S. epidermidis*, *S. pneumoniae* and *Corynebacterium* species while *H. influenzae* was the most common Gram-negative isolate. Huber-Spitzy et al. found *S. aureus* (45%) and *S. epidermidis* (20%), followed by the Gram-negative *Pseudomonas aeruginosa* and *Escherichia coli*. The data are given in other studies, not in our study. In our study, the most common Gram-negative isolate was *Staphylococcus epidermidis* (52.6%), followed by *S. maltophilia* (30.9%), *H. influenzae* (21.2%) and *S. pneumoniae* (12%).

**Table 3: Comparison of microbiological spectrum and antibiotic sensitivity between cases and controls**

| Cases, n (%) | Controls, n (%) |
|-------------|----------------|
| Gram-positive isolates | 139 (84.24) | 18 (94.7) |
| Gram-negative isolates | 26 (15.75) | 1 (5.26) |
| Most common Gram-positive isolate | *S. epidermidis* (30.9) | *S. epidermidis* (52.6) |
| Most common Gram-negative isolate | *H. influenzae* (5.3) | Neisseria spp. (5.26) |
| Antibiotic sensitivity | Gentamicin and vancomycin (95.5 each), chloramphenicol (93.2), tobramycin (100 each) |
| Antibiotic sensitivity | Gentamicin, vancomycin and tobramycin (100 each) |
| Antibiotic sensitivity | Amikacin (92.3) | Sensitive to all tested antibiotics |
| Rare isolates | *A. xylosidans*, *S. maltophilia*, *K. Kristina* | Nil |

*S. epidermidis*: Staphylococcus epidermidis, *S. pneumoniae*: Streptococcus pneumoniae, *H. influenzae*: Haemophilus influenza, *A. xylosidans*: Achromobacter xylosidans, *S. maltophilia*: Stenotrophomonas maltophilia, *K. Kristina*: Kocuria Kristina

Figure 2: Antibiotic resistance patterns among Gram-positive and Gram-negative isolates in the congenital nasolacrimal duct obstruction group

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coli as the predominant isolates in CNLDO. As per the study by Usha et al., the most frequent Gram-positive isolate was *S. pneumoniae*, and the most frequent Gram-negative isolate was *H. influenzae*. A study by Dave et al., demonstrated that the frequent use of azithromycin and fluoroquinolones alters conjunctival flora increasing *S. epidermidis* isolates. Though we have not taken into account the fact that many of these patients would have used topical antibiotics before presenting to us, it is reasonable to assume that the topical antibiotic used likely affects the spectrum of microbiology found in these patients.

In our study, 33.5% of the samples had polymicrobial growth. The presence of two or more isolates has been reported in as many as one-third of cases of dacryocystitis in some studies. Though few studies point towards polymicrobial growth in older individuals, we did not notice any such trend. No anaerobic bacteria were isolated in our study. Previous studies have reported anaerobic bacteria to be between 0 and 13 percent of isolates.

We had one isolate of *K. kristinae*. Very few cases of infections caused by this pathogen have been documented. Uncommon Gram-negative bacteria like *S. maltophilia* (one case) and *A. xylosidans* (one case) also were isolated. These uncommon species are thought to be emerging opportunistic ocular pathogens. Only 2% of our samples revealed fungal growth. The three fungal isolates in our study were *Aspergillus Niger*, *Candida stellatoidea*, and *Candida tropicalis*. Ghose and Mahajan studied the spectrum of fungal flora in congenital dacryocystitis, and fungi were isolated in 30.2% of the patients, differing from our study.

The conjunctival flora normally comprises noninfectious microorganisms in healthy individuals which play a role in ocular defense mechanism against infections. Bacterial isolation has been noted at a rate of 15%–100% from the conjunctiva of a healthy population. Bacterial growth was seen in about 30.64% (19 of 62) of our controls. Ocular surface flora consists predominantly of Gram-positive microorganisms, especially *S. epidermidis*, *S. aureus*, and diphtheroids. Analysis of the distribution of the microorganisms in both cases and controls, Gram-positive isolates were predominant with the most common isolate being *S. epidermidis* (30.9% in cases and 52.6% in controls). About 35 *S. pneumoniae* were isolated among cases whereas only one was isolated among controls. This difference could be attributed to the fact that the normal nasolacrimal system acts as a barrier against ascending pneumococcal colonisation.

The antimicrobial agents selected for our study was based on the commonly used drugs in our population. Most of the available approved topical antibiotics are based on clinical studies performed in adults. There are no guidelines yet for topical antibiotics usage in children. In our study, Gram-positive organisms exhibited the highest rate of sensitivity to gentamicin (95.5%), followed by vancomycin (95.5%) and chloramphenicol (93.2%), while Gram-negative organisms did so for amikacin (92.3%). Unlike previous studies, sensitivity to fourth-generation fluoroquinolone-gatifloxacin was also studied. Both Gram-positive and Gram-negative isolates were sensitive to gatifloxacin (78.1% and 80%, respectively). Gatifloxacin is a fourth-generation fluoroquinolone that is effective against a broad spectrum of bacteria causing ocular infections. Although gatifloxacin has been withdrawn for systemic use due to its toxicity, no toxicity has been noted when given topically. The effectiveness of Chloramphenicol against Gram-positive bacteria and of fluoroquinolones (ciprofloxacin and ofloxacin) against Gram-negative bacteria in CNLDO has been reported in earlier studies. Resistance against gentamicin was noted in earlier mentioned studies in contrast to our study where isolates showed higher sensitivity to gentamicin. Gram-positive as well as Gram-negative organisms demonstrated the highest degree of resistance to cotrimoxazole and erythromycin, which is in accordance with most of the previous studies.

Amongst our cases 13.34% of *S. aureus* and 29.41% of *S. epidermidis* infections demonstrated resistance to methicillin. The common risk factors for MRSA infection include recent or prolonged hospitalization, recent or prolonged antibiotic use, any invasive procedures, and MRSA colonisation. Mills et al. noted 21.7% of dacryocystitis-related *S. aureus* infections to be MRSA. Kodi reported a case of MRSA in association with chronic dacryocystitis secondary to CNLDO in an 8-month-old infant. Frequent and prolonged antibiotic exposure in children could be a risk factor for developing MRSA infections and hence should be avoided. Both our MRSA cases were resistant to ciprofloxacin and erythromycin. Vancomycin is considered to be the drug of choice in MRSE cases, and our isolates, too, demonstrated the highest degree of sensitivity to vancomycin.

Treatment outcomes were similar among Gram-positive and Gram-negative organisms in the present study, and there was no statistically significant association between treatment outcome and the presence of polymicrobial growth on culture similar to the study by Al-Faky et al.

The microbial spectrum was found to be varying in different studies, and this very fact makes it essential to know the current microbial profile and their sensitivity patterns in a geographical area. In a country like India where most of the topical drugs are available as over the counter medications, there is a looming risk of development of antibiotic resistance. The present study guides us regarding the prevailing organism in the country and their sensitivity profile. The comparative and the prospective design of the study make the study stronger as compared to the previously available literature.

This study has several limitations. The sample size is relatively small and may not be an exact representation of the community. The control size is disproportionately small compared to the cases group. Although children of the same group were included as controls, they were not necessarily similar to the cases in every aspect, and hence, a true matching cannot be ascertained.
To conclude, Gram-positive organisms are predominant in CNLDO with *S. epidermidis* and *S. pneumoniae* being the major isolate in little over half of the cases (52.1%). The most effective antibiotics are gentamicin and amikacin for Gram-positive and Gram-negative organisms, respectively. Gatifloxacin has proven to be sensitive to both. The pattern of microbial growth does not affect the outcomes of probing. The results of our study will provide guidance in choosing the appropriate antibiotics having broad coverage against common pathogens.

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**Conflicts of interest**

There are no conflicts of interest.

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