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Bacteria associated with contact lenses among asymptomatic students wearers of university of Tripoli, Libya

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ABSTRACT

The use of contact lenses without special precautions and sterilization procedures results in keratitis and other eye infections. In Libya, contact lenses are common, and many people are unaware of their proper care and sterilization measures. The study aimed to evaluate the bacterial contamination in contact lenses among university students and determine the level of awareness regarding contact lens hygiene. This cross-sectional study was carried out from April to August 2022 on 50 medical students of the Faculty of Medical Technology at Tripoli University of Tripoli city Libya, who are asymptomatic and wear contact lenses. A questionnaire on personal information, contact lens wear, and lens handling procedures was distributed. The samples were taken from the lens with sterile swabs and quickly cultured, and the organism is identified using appropriate biochemical tests. and antimicrobial susceptibility tests by standard protocols. A total of 46 bacterial isolates were identified from 50 samples and the remaining 4 samples were found to have no growth. The common isolates were *Pseudomonas aeruginosa* 20 (83.3%), followed by *Staphylococcus aureus* 10 (45.5 %), *Staphylococcus epidermidis* 8 (36.4%), *Escherichia coli* 3 (12.5%), *Streptococcus viridans* 2 (9.1%), *Streptococcus agalactiae* 1 (4.5%), and *Streptococcus pneumoniae* 1 (4.5%), *Haemophilus influenza* 1 (4.2%). Our findings demonstrate that contact lenses are highly contaminated with bacterial strains and that could be due to improper care of contact lenses by wearers. Increasing awareness is crucial to avoid any identified risk factors to the eyes that can occur from organisms' contamination. Regular standard protocols lens care for the usage is, therefore, are essential.

Keywords: Contact lens infections; Bacterial strains; Contamination.

1. Introduction

Contact lenses (CL) are becoming more popular for cosmetic and therapeutic reasons. However, wearing a CL is linked to a number of potential side effects, such as neurotrophic corneal ulceration, bacterial and fungal keratitis, acanthamoeba keratitis, papillary conjunctivitis, superior limbic keratoconjunctivitis, and others [1]. Microbial keratitis is one of the serious complications of contact lens use and if not treated timely, may result in permanent visual damage to the cornea [2]. In developed countries, contact lenses are a frequent cause of microbial keratitis, especially in young individuals, with

about 12% of hospitalized cases necessitating corneal transplantation [2]. Microbial contamination of a contact lens container can affect contact lens usage and result in lens-related issues. Further, identical strains of bacteria have been isolated from both the corneal ulcer in microbial keratitis and the lens storage case, Observation of certain hygiene principles, such as washing the hands before using a lens, drying the lens case, using a lens case disinfectant, and more, may reduce lens and lens case contamination [3]. The major causative agents in contact lens-related microbial keratitis are gram-negative bacteria especially *Pseudomonas* species which is the most commonly isolated organism, then the following most frequently identified causative organisms are coagulase-negative *staphylococci* and *Staphylococcus aureus* [4]. Eye infections have been demonstrated to be caused by a lack of compliance with lens care instructions and inadequate cleanliness [1]. Contact lenses can cause complications that are unacceptable for the user, several types of research had reported the presence of microorganisms on contact lenses, especially bacteria. Furthermore, more problems may be linked to contact lens wear in developing countries than in industrialized countries [5]. When contact lenses become contaminated, the contact lens case becomes a potential source of pathogens, especially when it is not properly cleaned and disinfected. As a result, even cleansing and sterilizing contact lenses do not guarantee a safe environment because the source could be contaminated case. Many studies have shown that bacteria isolated from contact lenses associated with infectious eye disorders are identical to those isolated from the corresponding contact lens cases [6]. Cleaning the lens case with tap water will increase the contamination of the lenses with gram-negative bacteria. In addition, if the case is changed every two weeks, we will have the least amount of contamination [3]. Contamination of contact lens cases, solution, lens material, wearing schedule, and disinfection practices are major factors that influence infections associated with contact lens use. Contact lenses when inserted into the eye, rapidly accumulate proteins, glycoproteins, and lipids from tear film on its surface providing a conducive environment for bacterial adhesion. The ability of adhered bacteria to grow on the tear film components adsorbed on the lens surface is a pathogenic trait. Moreover, factors known to increase the risk of eye infections due to misusing contact lenses include; sleeping with contact lenses, swimming and showering with them, cleaning and keeping lenses in tap water, not using the lens case, late lens case replacement, use of contaminated solutions and cases, prolonged use, reuse, and poor hygiene [1,3,7]. Contact lens contamination and eye infections often occur as a result of not following the standard care procedures recommended by ophthalmologists, which include a series of steps for properly cleaning the lenses. This study aimed to detect the bacteria found in contact lens users and evaluate the isolated species' antibiotic susceptibility patterns. In addition, the present study was also planned to assess the knowledge of contact lens student users about the risk of microbial contamination associated with improper use and maintenance of contact lenses.

2. Method

This study cross-sectional was conducted at Tripoli University Faculty of Medical Technology from April to August 2022 and was approved by the ethical committee of the Faculty of Medical Technology, University of Tripoli, Libya. A total of 50 asymptomatic contact lens wearers among female medical students volunteered to be included in the study. Contact lens (CL) wearers with eye infections or under any therapeutic or diagnostic eye drops were excluded from the study. contact lens samples were collected from the concave surface of the contact lenses by using sterile cotton swabs moisturized with normal saline solution. The swab was immediately inoculated onto Nutrient agar, Blood agar, Mannitol salt agar, and MacConkey's agar plates.

The blood agar and MacConkey's agar were incubated at 37˚C for 24-48 h. Organisms were grown were identified using standard microbiological techniques using Gram's

staining, based on culture diagnosis by growing on selective media and performing biochemical tests including catalase, coagulase, and oxidase. and their antimicrobial sensitivities were done using Kirby-Bauer diffusion techniques.

A questionnaire was administered to participants to collect personal information and their contact lens Information including CL wear and their handling procedures.

2.1 Statistical Analysis

Data were presented as frequency and percentages. Statistical analysis was performed using Package of Social Sciences (SPSS) version 26. Student's Chi-Square Test was run to determine the correlation between two variables. The probability value (p) was considered significant if it was less than 0.05 and highly significant if was (<0.001).

3. Results

A total of 50 samples were collected from female students who are wearing contact lenses. The duration of the study was 4 months from April to August 2022, out of the 50 contact lenses collected from all subjects, 46 (92%) samples were contaminated and the remaining 4 (8%) samples showed no growth. The overall rate of microbial contamination among the total samples was Gram-negative 24 (52.2%). The rest of the isolates were from positive bacteria gram group 22 (47.8%), as shown in Table 1. Amongst the bacterial isolates, *Pseudomonas aeruginosa* had the highest occurrence with 20 (83.3%), followed by *Staphylococcus aureus* 10(45.5 %), *Staphylococcus epidermidis* 8 (36.4%), *Escherichia coli* 3 (12.5%), Whereas the last isolated species recorded with the lowest percentage were *Streptococcus viridans* 2 (9.1%), *Streptococcus agalactiae* 1 (4.5%), and *Streptococcus pneumoniae* 1 (4.5%), *Haemophilus influenza* 1 (4.2%).

Table 1. Bacterial strains isolated from contact lenses

Type of isolates	Gram-positive	Gram-negative
	22 (47.8%)	24 (52.2%)
Gram-positive isolate species		
Staphylococcus aureus	10(45.5 %)	
Staphylococcus epidermidis	8(36.4 %)	
Strepococcus viridans	2(9.1%)	
Streptococcus agalactiae	1(4.5%)	
Streptococcus pneumoniae	1(4.5%)	
Gram-negative isolate species		
Pseudomonas aeruginosa	20(83.3%)	
Escherichia coli	3(12.5%)	
Haemophilus influenza	1(4.2%)	

Table 2 shows the present study's antibiotic resistance of various isolated strains. *P. aeruginosa* indicated high resistant to Erythromycin 20(100%), Fusidic Acid 12 (60%), Cephalexin 12 (60%), Cephradine 12 (60%), Bacitracin 11 (55%), Ampicillin 9 (45%). and least susceptible to Ciprofloxacin 2 (10%), and Amikacin (0%). *Staphylococcus aureus* was also most resistant to *Erythromycin* 8 (80%), Bacitracin 5 (50%), and Ampicillin 4 (40%). *Staph. epidermidis* exhibited utmost resistance 8 (100%) against Erythromycin and 4(50%) for all other antibiotics of Fusidic Acid, Cephalexin, and Cephradine. While *E. coli* showed a high resistance pattern with 3 (100%) for Fusidic Acid, Erythromycin, and Ampicillin. while 2(66.6%) for both Bacitracin and Cephradine.

S. viridnas strains displayed relatively high antimicrobial resistance to Fusidic Acid, Bacitracin, Cephalexin, and Cephradine with 2 (100%). *H. influenzae* was resistant to Fusidic Acid 1 (100%), Bacitracin 1 (100%), Ampicillin 1 (100%), and Cephradine 1 (100%). The least isolated strains *S. agalactiae* and *S. pneumoniae* were susceptible to all used antibiotics in this study.

Table 2. Antibiotic resistance pattern of microorganisms isolated from contact lens

ANTIBIOTIC	*p. aeruginosa (no=20)(%)*	*S. aureus (no=10)(%)*	*s. epidermidis (no=8)(%)*	*E, coli (no=3)(%)*	*s. viridans (no=2)(%)*	*H. influenzae (no=1)(%)*	*s. agalactiae (no=1)(%)*	*s. pneumoniae (no=1)(%)*
FA (10)	12 (60)	5 (50)	4 (50)	3 (100)	2 (100)	1 (100)	0	0
CIP (5)	2 (10)	0	0	0	0	0	0	0
E (15)	20 (100)	8 (80)	8 (100	3 (100)	0	0	0	0
BA (10)	11 (55)	5 (50)	2 (25)	2 (66.6)	2 (100)	1 (100)	0	0
AM (10)	9 (45)	4 (40)	3 (37.5)	3 (100)	0	1 (100)	0	0
CL (30)	12 (60)	2 (20)	4 (50)	0	2 (100)	0	0	0
CE (30)	12 (60)	3 (30)	4 (50)	2 (66.6)	2 (100)	1 (100)	0	0
AK (30)	0	0	0	0	0	0	0	0

(FA) Fusidic Acid, (CIP) Ciprofloxacin, (E) Erythromycin, (BA) Bacitracin, (AMP) Ampicillin, (CL) Cephalexin, (CE) Cephradine, (AK) Amikacin.

Table 3 illustrates the distribution of contaminated samples according to contact lenses practice and information among University Students. During the study, it was discovered that 31 (62%) of the respondents used them for cosmetic reasons, and 19 (38%) students used them to correct vision. The percentage of positive culture results, in cosmetic and therapeutic lenses, was 30(97%) and 16(48%), respectively. Moreover, 35(70%) students had worn CLs for ≥ 2 years and 2(4%) students had worn CLs for more than 3 years. Thus, the minority of participants in this study were experienced wearers. The high percentage of positive culture in CLs worn by students for ≥ 2 years was 32(91%), and the low percentage in CLs worn for more than 3 years was 1(50%).

For the daily wearing time, 17 (34%) students wore CLs for five to twelve hours, and 6 (12%) of them wore CLs for more than 12 h daily; furthermore, 27 (54%) students reported that they wore CLs for less than five hours. Most of the contamination rates were in CLs that were worn by students for less than five hours with 26 (96%) and for five to twelve hours 15(88%). Regarding CL hygiene, the majority of wearers reported that they never washed their hands before handling CLs 28 (56%). Moreover, 35 (70%) students reported that they rinsed their hands, while 15 (30%) students reported negatively. The prevalence of the positive bacterial culture in hand washing, non-washing, hand drying, and non-drying every time dealing with lenses was 18(82%), 28(100%), 31(89%), and 15(100%), respectively. Sterilizing the contact lenses daily with the solution of CL occurred in 5 (30%) of wearers and the majority of wearers 31 (62%) sterilize CL just before using, and 4 (8%) of students sterilized their lenses twice a week. A total of 17 (43%) students had shared their CL with others. A high percentage of bacterial culture was also observed in CL that was sterilized just before using 29(93%) and not shared with others 29(88%). Among participants 24 (48%) students reported that they had eye redness which was at least one problem related to the use of CL, while 26 (52%) of them didn't have any redness symptoms. In 24(48%) students were suffering from eye redness the results of lens case culture were positive with 17(100%). According to the findings of Table 3, no significant statistical relationship was observed between the positive result rate of CL culture and all the questionnaire parameters of knowledge and practice regarding contact lens (CL) user's p-value > 0.05.

Table 3. The distribution of contaminated samples according to awareness and practices of CL usage among University Students

Variable	Parameter	N = 50 (%)	Bacterial n (%)	P value > 0.05
Reason for use	Cosmetic	31(62%)	30(97%)	0.74
	Therapeutic	19(38%)	16(48%)	
Duration of use	Less than 1 year.	13(26%)	13(100%)	
	Between 1 to 2 years.	35(70%)	32(91%)	0.86
	More than 2 years.	2(4%)	1(50%)	
Time length of use	Less than five hours.	27(54%)	26(96%)	
	Between five and twelve hours.	17(34%)	15(88%)	0.96
	More than twelve hours.	6(12%)	5(83%)	
Hands washing every time dealing with lens	Yes	22(44%)	18(82%)	0.63
	No	28(56%)	28(100%)	
Hands drying every time dealing with lens	Yes	35(70%)	31(89%)	0.75
	No	15(30%)	15(100%)	
Sterilize the CL with a solution	daily	15(30%)	13(87%)	
	Twice week	4(8%)	4(100%)	0.97
	Just Before using	31(62%)	29(93%)	
Share CL with other	Yes	17(34%)	17(100%)	0.76
	No	33(66%)	29(88%)	
Eye redness	Yes	24(48%)	24(100%)	0.68
	No	26(52%)	22(85%)	

4. Discussion

Across the world, CL is widely distributed among young adults, for reasons such as cosmetic or therapeutic, since its first use in 1887[8]. There is a continuous increase in the use of contact lenses in Libya because of the optical, occupational and cosmetic advantages to individuals. Several studies reported that the introduction of contact lenses was associated with an increase in ocular microbial complications [9,10]. The unique structure of the human eye, the use of contact lenses, and the constant exposure of the eye directly to the environment render it vulnerable to several uncommon infectious diseases before the invention of rare contact lenses. Thus, new opportunities were offered to these microorganisms when people started wearing contact lenses. Therefore, necessary precautions are required to protect the eye from these opportunistic organisms. Therefore, the identification of the microorganisms found in the contact lenses of wearers of paramount is importance.

In the current study, the frequency of positive microbial culture in cosmetic and therapeutic contact lenses was 30 (97%) and 16 (48%), respectively. Among the isolated bacteria observed in positive cultures, *Pseudomonas aeruginosa* 20 (83.3%) was the most common contaminant of contact lenses followed by *Staphylococcus aureus* 10 (45.5 %). This corroborates with other studies that also reported *P. aeruginosa and S. aureus* are the most frequently isolated organisms [11,12]. On the other hand, Hesam et al. [12] reported that *Pseudomonas aeruginosa* was the most prevalent causative agent of microbial keratitis accounting for (80%) of positive cultures and *Staphylococcus* was the second most common among laboratory-proven infectious keratitis with 12%. The most serious and sight-threatening complication associated with CL use is ulcerative keratitis.

In the present study, the *Staphylococcus epidermidis* (36.4 %) was also isolated, coinciding with a study by Mohamed et al [13], which revealed that the most common isolated microorganisms were *Staphylococcus epidermidis* (36.3%) which was found to be more in normal conjunctival flora due to their virulence factors. Another similar study by Rahim et al in Pakistan confirmed the same finding, with slight variations in the percentages of occurrences [14].

In other studies, like Supriya S et al [15] the other organism isolated was *E. coli* (12.5%). These findings were consistent with the results of the present study that reported E. *coli* was isolated with a percentage of (12.5%). The

discovery of *E. coli* in the examined contact lenses may have resulted from the usage of contaminated water. Free-living amoebae had been isolated from dust, contact lenses, domestic water, and swimming pool [16]. The first case of Acanthamoeba keratitis in a female contact lens was described by Kamel and Norazah [17].

Due to the differences in the type of isolated microorganisms, and the type of antibiotics that are used, the pattern of antibiotic susceptibility and resistance varies in the different studies. However, the present study, indicates that all of the microorganisms were sensitive to amikacin. In comparison with the study done by Eslami F et al [3] ciprofloxacin was one of the most effective antibiotics against most strains isolated from CL.

Many authors reported that extended periods of asymptomatic lens wear did increase risk factors for ocular microbiota [18]. As a result, continuous use, particularly overnight, can cause hypoxia and hypercapnia of the corneal epithelium, resulting in ischemic necrosis and a corneal ulcer [19,20,21]. Similar to our finding, prolonged use of CL was associated with eye problems like redness. Failure to comply with instructions of use and poor hygiene is a risk factors for CL wearers in our study. Lack of hygiene and improper care of CL can predispose to the colonization of the CL surface with bacteria, leading to biofilm formation, especially with *Pseudomonas aeruginosa*. [22,23]. The potential CL user must be educated and given advice on how to properly care for their lenses, how long to wear them, and how to maintain good hygiene and prolonged wearing of CL, wearing it overnight, and swimming or taking a shower while wearing CL must be avoided. It has been shown that some microorganisms including potentially pathogenic species can survive for hours on contact lenses and have harmful effects on the ocular surface [8].

5. Conclusion

Finally, the individuals' lens care practices were poor, resulting in significant levels of contamination. In our findings, the most common bacteria that contaminate contact lenses was *Pseudomonas aeruginosa*. This is a result of bacterial adherence to contact lenses, which has been linked to the development of a number of unfavorable outcomes during wearing contact lenses. Hence it is proved in many studies that *P. aeruginosa* is the one that usually adheres to lenses in greater numbers than other strains. Therefore, the possibility of bacterial contaminants in contact lenses is common. therefore, creating awareness among the users about lens care practices and regular cleaning and replacements of lens cases are required. Prevention of bacterial contamination of contact lenses can reduce the risk of developing ocular infections.

6. References

1. Lim CHL, Stapleton F, Mehta JS. Review of contact lens-related complications. Eye Contact Lens. 2018;44(Suppl 2): S1–S10. DOI:10.1097/ICL.0000000000000481.
2. Hoddenbach JG, Boekhoorn SS, Wubbels R, Vreugdenhil W, Van Rooij J, Geerards AJ. Clinical presentation and morbidity of contact lens-associated microbial keratitis: a retrospective study. Graefes Arch Clin Exp Ophthalmol. 2014;252(2):299-306.
3. Eslami F, Ghasemi Basir HR, Moradi A, Bayat M. Microbiological analysis of contact lens cases and effective health behaviors. Immunopathol Persa. 2021;7(2): e17. DOI:10.34172/ ipp.2021.17.
4. Wang, J.-J.; Lai, C.-H.; Chen, C.-Y.; Liu, C.-Y.; Lin, M.-H.; Yang, Y.-H.; Wu, P.-L. Trends in Infectious Keratitis in Taiwan: An Update on Predisposing Factors, Microbiological and Antibiotic Susceptibility Patterns. Diagnostics. 2022, 12, 2095. DOI:10.3390/diagnostics12092095.
5. Abokyi, S., Manuh, G., Otchere, H., & Ilechie, A. (2017). Knowledge, usage and barriers associated with contact lens wear in Ghana. Contact Lens and Anterior Eye. 40(5), 329–334. DOI: 10.1016/j.clae.2017.05.006.
6. Wu YTY, Willcox M, Zhu H, Stapleton F. Contact lens hygiene compliance and

lens case contamination: a review. Contact Lens Anterior Eye. 2015;38(5):307-16.
7. Cope JR, Collier SA, Nethercut H, Jones JM, Yates K, Yoder JS. Risk Behaviors for Contact Lens-Related Eye Infections Among Adults and Adolescents - United States, 2016. Morb Mortal Wkly Rep. 2017;66:841-5. DOI: 10.15585/mmwr.mm6632a2.
8. Loh K, Agarwal P. Contact lens related corneal ulcer. Malays Fam Physician .2010;5:6-8.
9. Wu YT, Willcox MD, Stapleton F. The effect of contact lens hygiene behavior on lens case contamination. Optom Vis Sci. 2015; 92:167-74. DOI:10.1097/OPX.0000000000000634.
10. Ragupathy V, Zhao J, Wang X, Wood O, Lee S, Burda S, et al. Comparative analysis of cell culture and prediction algorithms for phenotyping of genetically diverse HIV-1 strains from Cameroon. AIDS Res and Therapy. 2009; 6:6405-6427
11. Anitha M, Mathivathani P, Ramya K, Vijay M. Assessment of Bacterial Contamination on Contact Lenses among Medicos. Int J Pharma. 2016; 4:1160-5. DOI: 10.21276/ijprhs.2016.03.03.
12. Hesam H, Ghaderpanah M, Rasoulinejad SA et al. (2015): Clinical Presentation and Antibiotic Susceptibility of Contact Lens Associated Microbial Keratitis. J Pathog. 5: 3-4.
13. Mohamed JA, Abdallah SM, Alatrouny AM, Newishy HM. Bacterial interaction among soft contact lens users and lens care solutions with antibiotic suseptibility pattern. Egypt J Hosp Med. 2017;68: 982-990. DOI: 10.12816/0038200.
14. Rahim N, Bano H, Naqvi S. Bacterial Contamination Among Soft Contact Lens Wearer. Pak J Ophthalmol. 2008; 24:93-6.
15. Supriya S. Tankhiwale, Sonali Dwidhmuthe and N.S. Tankhiwale. Risk factors and microbial colonization of soft contact lens storage cases and conjunctiva of asymptomatic lens users IJAMBR.2015; 3: 31-35.
16. Anisah N, Yusof S, Wan Norliana A, Noraina AR, Norhayati M. Acanthamoeba spp. isolated from salt water in the west coast of peninsular Malaysia. Trop Biomed. 2004;21:109-111.
17. Kamel AGM, Norazah A. First case of Acanthamoeba keratitis in Malaysia. Trans Royal Soc Trop Med and Hyg. 1995;89:652.
18. Dart JK, Radford CF, Minassian D, Verma S, Stapleton F. Risk factors for microbial keratitis with contemporary contact lenses: A case-control study. Ophthalmology. 2008;115:1647-54, 1654.e1-3.
19. Efron N, Morgan PB. Rethinking contact lens associated Rethinking contact lens associated keratitis. Clin Exp Optom. 2006;89:280-98.
20. Stapleton F, Keay LJ, Sanfilippo PG, Katiyar S, Edwards KP, Naduvilath T. Relationship between climate, disease severity, and causative organism for contact lens-associated microbial keratitis in Australia. Am J Ophthalmol .2007;144:690-8.
21. Behlau I, Gilmore MS. Microbial biofilms in ophthalmology and infectious disease. Arch Ophthalmol .2008;126:1572-81.
22. Toutain-Kidd CM, Kadivar SC, Bramante CT, Bobin SA, Zegans ME.Polysorbate 80 inhibition of Pseudomonas aeruginosa biofilm formation and its cleavage by the secreted lipase LipA. Antimicrob Agents Chemother .2009;53:136-45.