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Care practices of contact lens solutions and microbial contamination among wearers in Ghana

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ARTICLE INFO	A B S T R A C T					
Keywords: Microbial contamination Contact lens care solutions Antimicrobial Susceptibility testing	<i>Purpose</i> : This study sought to assess contact lens solutions care practices, and their microbial contamination among contact lens wearers in Ghana and to profile their antibiotic susceptibility pattern. <i>Methods</i> : The study employed a biphasic approach which involved a cross-sectional design that investigated participants' habits related to care for the solutions with a two-part questionnaire and a microbiological analysis of samples of contact lens care solutions of the participants for microbial contamination. A snowball sampling method provided access to 32 different contact lens wearers in four care facilities in Ghana. In most cases, the participants had no pre-existing familial relationship with each other or with the care facilities. <i>Results</i> : Out of 32 samples of contact lens solution were found to be contaminated with <i>Enterobacter</i> sp. (34.80 %), <i>Pseudomonas</i> sp. (21.70 %), <i>Bacilli</i> sp. (21.70 %), <i>Klebsiella</i> sp. (17.20 %), and <i>Escherichia coli</i> (4.60 %). The duration of solution storage in the open bottle and nonadherence to manufacturer instructions for solution storage showed a statistically significant association with microbial contamination ($p \leq 0.05$). <i>Conclusion:</i> Contact lens care solutions have been found to harbour multiple antibiotic-resistant bacteria that are potentially pathogenic to the corneal surface. The contamination is associated with some unhealthy solution-care practices among wearers.					

1. Introduction

Contact lens (CL) wear is the leading risk factor for the development of microbial keratitis in otherwise healthy eyes [1]. The lens biomaterial serves as a substrate for the adherence of microorganisms increasing the risk for ocular surface infections [2]. More than half (approximately 56 %–65 %) of lenses are found to contain microorganisms, mostly bacterial when aseptically removed from the eye [3]. Some of the factors accountable for the development of microbial keratitis include the type of lens material [4], wearing schedule [5,6], adherence of microorganisms to the lens [7], and microbial contamination of lens care solutions [8,9].

Contamination of lens care solutions is of significant concern in contact lens wear [10], particularly in developing countries such as Ghana, where poor hygiene practices and limited access to clean water [11] can increase the risk of microbial contamination. In contrast to lens

contamination, which is almost exclusively bacterial, the microorganisms involved in solution contamination found in lens cases or on the internal wells are usually a mixture of bacteria, fungi, and protozoa [12,13]. Studies involving patients with contact lens-associated microbial keratitis have demonstrated contamination of the lens care solutions [14,15]. All types of solutions are at risk of microbial contamination, in experienced and compliant users [16] and even in unopened, factorysealed bottles (an incidence of 11.15 %) [17]. Contact lens cases or cleaning solutions have been found to be contaminated by Acanthamoeba sp. and other microbial organisms as a result of poor hand hygiene [18]. Microbial organisms that are responsible for the contamination of lens care systems can serve as a source of nutrition for other harmful organisms, including Acanthamoeba sp. [19]. Microbial contamination of the care system may result in the production of toxins that affect the eye [20]. In rabbits, intrastromal injection of endotoxins present in the cell walls of gram-negative bacteria has been linked to

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corneal ring infiltrates [21].

Several previous studies have established the risk of contamination of contact lens care solutions [22–24]. Taking into consideration hot and humid climates that promote the proliferation of bacterial populations, it is of utmost importance to frequently monitor the degree of microbial contamination in lens care solutions among contact lens wearers [10]. This study therefore aimed to assess contact lens care solution care practices and their microbial contamination among contact lens wearers in Ghana and to profile their antibiotic susceptibility pattern.

2. Materials and methods

2.1. Data collection procedure

This biphasic study employed a cross-sectional approach to investigate factors that influence contamination of contact lens solutions, and a laboratory analysis to assess the microbial contamination profile of contact lens care solutions. A two-part questionnaire was used to collect data on patient demographics, which included age, sex, and occupation, as well as information on habits related to caring for the solutions. The second phase involved the collection of in-use contact lens solutions from the identified wearers using a snowball approach. As an exclusion criterion, bottles with less than 1 ml of solution (almost finished) were not collected. No special instructions were given, and the participants were not informed beforehand that they would be recruited into the second phase of the study to avoid a possible modification in their solution care practices. A 2 ml sterile syringe was used to aspirate samples from the residual lens care solutions in the bottles into sterile vacuum tubes, which were then capped effectively and preserved under cold conditions (2-4 °C). Processing of the samples took place within a 12hour window. Control samples were obtained from unopened containers at the designated sites, for comparative purposes. Samples were collected between March and August 2023.

2.2. Participant selection

Eye care practitioners in Ghana have been reported to provide limited contact lens services [25], resulting in only a handful of eye care facilities and optometric practices offering contact lens services to the public.

The study therefore utilized snowball sampling to recruit 32 contact lens wearers. The snowball sampling method provided access to 32 contact lens wearers who were associated with four care facilities in Accra, Kumasi, and Cape Coast. A visit was made to each of the contact lens care facilities. The first contact lens patients identified in the facilities were first asked for their consent to participate in the study, and then, to refer the investigators to other contact lens wearers. In most cases, the participants had no pre-existing relationship with each other or with the care facilities. The contact lens wearers were recruited from four distinct eye care facilities in the country as a result of the dearth in the number of eye care facilities that offer contact lens services to the public. Inclusion criteria were applied stringently to individuals aged 18 and above, without gender bias, who had sought treatment at these facilities. Active use of contact lens solutions during the study period (had to wear CLs at least once a week either for vision correction or for cosmetic reasons) was also a crucial consideration. During the study, individuals experiencing symptoms such as redness and discharges indicative of bacterial corneal or conjunctival infections related to contact lens wear were also excluded to ensure that the investigation remained focused on the contamination of contact lens solutions.

2.3. Culturing and identification of microbes

A range of culture media, including nutrient agar and peptone water agar, was prepared meticulously and precisely following manufacturer instructions (Thermo Fisher Scientific, San Jose, CA, USA) to guarantee sterility. Additional media, including triple sugar iron agar, citrate agar, and motility agar, were employed to evaluate microbial biochemical activity. Mueller-Hinton agar (Thermo Fisher Scientific, San Jose, CA, USA) was used for culture and sensitivity tests and was prepared as contained in the manufacturer's instructions. To ensure accurate processing of the contact lens solution, 1 ml of each sample was pipetted and inoculated into a sterile Petri dish containing approximately 20 ml of culture media. The plates were gently swirled to ensure even distribution before solidifying. Subsequently, the plates underwent incubation: 24 h at 37 °C for bacteria and 120 h at 25 °C for fungi. Following this period, the visible colonies were enumerated, scrutinized for morphological traits, and categorized. Selected colonies were then segregated and re-cultured in nutrient agar for future identification. Microorganism isolates were identified through an array of measures encompassing biochemical, morpho-cultural characteristics, and Gram staining. The morpho-cultural properties were established by assessing the colony appearance on nutrient agar. Gram-negative bacteria were identified through biochemical tests, including catalase, indole, triple sugar iron reduction, citrate, and motility tests. Gram-positive bacteria, on the other hand, were identified using catalase, motility, and indole tests.

2.4. Antimicrobial susceptibility testing

The bacteria under observation underwent susceptibility testing to a range of antibacterial agents using the disc diffusion method. Antifungal susceptibility testing was not performed since no fungal contaminant was isolated. Mueller-Hinton agar served as the medium in combination with antibacterial discs. The antibacterial agents included Tetracycline (10 μ g), Gentamicin (10 μ g), Ciprofloxacin (5 μ g), Amoxiclav (30 μ g), Ampicillin (30 μ g), Amikacin (30 μ g), Vancomycin (30 μ g), Ceftriaxone (30 μ g), Chloramphenicol (30 μ g), and Cefuroxime (30 μ g) which were imported from Scientific Laboratory Supplies, United Kingdom.

2.5. Data analysis

The study data was transferred to the Statistical Package for Social Science (SPSS) software, version 26.0 for Windows (Chicago, USA) for statistical analysis. To ensure reliability, all reported values were thoroughly examined for missing variables, outliers, and normal distribution. Owing to the small sample size, the Monte Carlo exact test at a 99 % confidence interval for 10,000 samples was applied. The Monte Carlo exact test was used to determine factors associated with contamination of contact lens solutions, and provided an unbiased estimate of the exact p-value, without the requirements of the asymptotic method. Univariate and multivariate logistic regression were run to screen for predictors of the likelihood of contamination, and odds ratios (cOR: Crude Odds Ratio; aOR: Adjusted Odds Ratio) with 95 % confidence intervals (CI) were utilized. Following the purposeful selection of variables procedure [26], variables found to be associated with contamination of lens care solutions from the Monte Carlo exact test were included in the univariate logistic regression model, and afterwards, the multivariate logistic regression model. The model for the multivariate logistic regression was adjusted for "The duration solution has remained in the bottle" and "Storage of solution according to instructions", and interactions were not explored. A statistically significant result was obtained with $p \leq p$ 0.05.

2.6. Ethical consideration

The study protocol was approved by the institutional review board of the University of Cape Coast (UCCIRB/CHAS/2023/77). The samples were sent to the microbiology laboratory of the Department of Biomedical Sciences, University of Cape Coast, where they were assayed. Biosafety guidelines for the protection of personnel in the laboratory were observed.

3. Results

3.1. Characteristics of study participants

Thirty contact lens solutions were analysed, with an equal distribution of 15 samples each from male and female patients. The age range of the participants was 22–48 years, with a mean age of 30.93 \pm 7.37 years.

3.2. Microbial profile of contact lens solutions

In all, 32 samples of contact lens solutions were collected from the participants. Contact lens solutions from two participants were excluded because the volume of residual solutions was less than 1 ml. Out of the 30 samples of contact lens solution that were tested for microbial contamination, 23 (76.67 %) were found to be contaminated while the remaining 7 (23.33%) were not. The control samples, on the other hand, did not exhibit any contamination. During the study, participants only had access to multipurpose and saline contact lens solutions. The bottles of contact lens solutions had been open for varying durations, ranging from 3 to 212 weeks, with a mean duration of 46.47 \pm 45.03 weeks (95 % CI 29.65-63.28). No fungal growth was observed in any of the samples examined during the investigation. Further analysis of the contaminated samples identified the presence of five different genera of bacteria. These bacteria were identified as Enterobacter sp., Pseudomonas sp., Bacilli sp., Klebsiella sp., and Escherichia coli (Table 1). Most of the contact lens care solutions were found to be contaminated with at least one genus of bacteria and a maximum of three bacterial contaminants (Fig. 1).

3.3. Factors associated with contact lens solution contamination

The factors associated with bacterial contamination of the CL solution are presented in Table 2. The duration of solution storage in the open bottle showed a statistically significant association with microbial contamination, as did nonadherence to manufacturer instructions for solution storage. Storage of contact lens solutions always according to manufacturer instructions was not found to be a significant predictor of the likelihood of contamination of contact lens solutions, as determined by univariate (cOR: 0.00 [95 % CI: 0.00-0.00], p = 1.00) logistic regression. A duration of opening of less than 3 months (90 days) was found to be associated with a 91 % lower likelihood of contamination of contact lens solutions, as determined by univariate logistic regression (cOR: 0.09 [95 % CI: 0.02-0.56], p = 0.01), and an 88 % lower likelihood of contamination of contact lens solutions as determined by multivariate logistic regression (aOR: 0.12 [95 % CI: 0.02–0.95], p = 0.04). The other variable included in the multivariate logistic regression was "Storage of contact lens solutions always according to manufacturer instructions". The contamination rate was slightly skewed towards the male population, although a statistically significant relationship between sex and microbial contamination was not demonstrated.

3.4. Antimicrobial susceptibility testing

The isolates were subjected to a susceptibility test using a standard

Table 1

Incidence of microbial contaminants in lens solutions isolated from collected samples.

Organisms	N (%)
Bacilli sp.	5 (21.70)
Pseudomonas sp.	5 (21.70)
Enterobacter sp.	8 (34.80)
Klebsiella sp.	4 (17.20)
Escherichia coli	1 (4.60)

antibiotic disc. This classified each isolate as resistant, intermediate, or susceptible to the antibiotics present on the disc. Table 3 presents a summary of the antimicrobial susceptibility test results for the isolated bacteria. All isolates were susceptible to Gentamicin 10 μ g, Ciprofloxacin 5 μ g, Amoxiclav 30 μ g, Vancomycin 30 μ g and Amikacin 30 μ g. However, they were resistant to Ampicillin 10 μ g, Ceftriaxone 30 μ g and Cefuroxime 30 μ g. *Bacilli* sp. isolates were the most susceptible, with susceptibility to all antibiotics except Ampicillin 10 μ g, Ceftriaxone 30 μ g, and Cefuroxime 30 μ g.

4. Discussion

Despite their long history of use and proven disinfectant efficacy, lens care solutions themselves can be easily contaminated and become a reservoir of microbes that can contaminate lens storage cases, adhere to the lens, cause an inflammatory reaction, or infect the cornea [27]. This study hence sought to assess contact lens care solution care practices, microbial contamination among contact lens wearers in Ghana, and their antibiotic susceptibility profile pattern.

Microbial contamination of contact lens care solutions was found to be common in this study, as most of the contact lens care solutions were found to be contaminated with at least, one genus of bacteria (Fig. 1). The multipurpose solution was found to be the most commonly used among the participants, while the use of saline solution was from a participant who had possibly deviated from the instructions from the eye care professional pertaining to contact lens care. The bioburden of contamination in this study is significantly higher than the incidence rates of contamination of preserved lens care solutions reported in similar studies (contamination rates of up to 30 %) [28,29]. In contrast to this study where lens care solutions were stored for a minimum of approximately 29 weeks, the maximum duration of storage in the study by Sweeney et al. [27] was 28 days. The survival and replication of bacteria in contact lens solutions may lead to the formation of biofilm in contact lens cases [30].

The duration of solution storage in the open bottle showed a statistically significant association with microbial contamination, as did adherence to manufacturer instructions for solution storage. Also, a duration of opening of less than 3 months (90 days) was found to be associated with a lower likelihood of contamination of contact lens solutions. Lens care solutions were found to be stored for a minimum of approximately 29 weeks, suggesting that contact lens wearers change their solutions infrequently possibly due to infrequent lens wear. The length of time since a bottle was opened and used has been demonstrated in previous studies to influence the degree of contamination [10]. In a study by Donzis et al. [31], a used bottle were shown to harbour organisms five (5) days after opening. It has been recommended that solution manufacturers begin labelling solution bottles with a discard date (usually within 90 days of opening), as the activity of most lens care solutions against microbes falls with time [10]. The majority of patients are non-compliant in the use of contact lens care solutions and maintenance regimens [4,32]. In a study that evaluated microbial contamination of contact lens care accessories and compliance with care regimens in Nepal, subjects with medium or low compliance had highly significant rates of contamination both in CL cases and solutions than to subjects with high compliance [29]. Collins & Carney [33] correlated the presence of contact lens-related signs and symptoms with care and maintenance regimen compliance and found a positive relationship between poor compliance and the presence of lens-wearing complications such as corneal staining, lens deposition, and subjective symptoms. Improving compliance with lens care practices is crucial in reducing complications and enhancing the patients' success with contact lens wear [34]. Eye care practitioners should provide clear, concise, and detailed guidance on lens care and maintenance [35]. They should also follow up with patients to ensure compliance with the recommended regimen [35].

Contaminants identified in this study comprised pathogens that have

Pseudomonas sp. Enterobacter sp.

Escherichia coli

Klebsiella sp.

Bacilli sp.



Fig. 1. Population of bacteria isolated from the various contact lens solutions G-01 through G-30 represent the samples of contact lens care solutions obtained from the participants.

Table 2

Factors associated with the contamination of contact lens solutions.

Sex1.00
Male12(52.20)3(42.90)Female11(47.80)4(52.70)Type of solution100.00Multipurpose solution2(95.70)7(10.00)
Female11(47.80)4(57.10)
Type of solution1.00<
Multipurpose solution22(95,70)7(100.00)Saline(4.30)0(0.00) $ -$ Handwashing before handling the solution1.00 $ -$
Saline1(4.30)0(0.00) </td
Handwashing before handling the solution14(60.90)5(71.40)
Always14(60.90)5(71.40) $(-)$
Sometimes 9(39.10) 2(28.60) Never 0(0.00) 0(0.00) Frequency disinfection (69.60) 7(100.00) $ -$ Always 16(69.60) 7(100.00) $ -$
Never00.0000.00Frequency of disinfection 0.000 $ -$ Always1669.0000.00 $ -$ Sometimes4(17.40)00.00 $ -$ Never3(30.0000.00 $ -$ Sharing of solution $ -$ Always1(4.30)0(0.00) $ -$
Frequency of disinfection 0.46 - <td< td=""></td<>
Always 16(69.60) 7(100.00) Sometimes 4(17.40) 0(0.00) Never 3(130.00) 0(0.00) Sharing of solution 0.66 - - Always 1(4.30) 0(0.00) - - Sometimes 3(13.00) 0(0.00) - - - Never 19(82.60) 7(100.00) - - - - Reuse of previously used solution - 0.46 - - - - Always 1(4.30) 0(0.00) -
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Sometimes 2(6.70) 0(0.00) Never 0(0.00) 0(0.00) Topping up care solution 1.00 - - -
Topping up care solution 1.00
Always 2(8.70) 0(0.00)
Aiways 2(6.70) 0(0.00)
Sometimes 0(0.00) 0(0.00)
Never $21(91.50)$ $7(100.00)$
Checking of expiry date regularity 0.55 -
Aiways 14(60.90) 6(85.70)
Sometimes b(25.10) 1(14.30)
Never 3(13.00) 0(0.00)
Rinsing contact lenses with water 1.00 – – – –
Aiways 0(0.00) 0(0.00)
Sometimes 2(8.70) 0(0.00)
Never 21(91.30) 7(100.00)
The duration solution has remained in the bottle 0.01 0.09 0.02–0.95 0.04
< 3 months (within 90 days) 3(13.00) 5(71.40)
3 months to 1 year 11(47.80) 2(28.6)
> 1 year 9(39.1) 0(0.00)
Contact lens wear experience 0.13 – – –
< 2 years 16(69.60) 5(71.40)
> 2 years 7(30.40) 2(28.60)

P*: p-values obtained from Monte Carlo exact test at a 99 % confidence interval for 10,000 samples.

P: p-values obtained from univariate logistic regression.

the potential to induce microbial keratitis associated with contact lenses, normal flora from the ocular surface, the gastrointestinal tract, the skin, as well as the surrounding environment. The strains of bacteria obtained from the contaminated samples included *Enterobacter* sp., *Pseudomonas* sp., *Bacilli* sp., *Klebsiella* sp., and *Escherichia coli. Bacilli* sp. Isolates were resistant to Ampicillin, Ceftriaxone, and Cefuroxime but most

Table 3

Antibiotic susceptibility pattern of the isolated bacteria.

Organism	GEN 10 µg	TET 10 μg	CIP 5 μg	CHL 30 µg	АМС 30 µg	AMP 10 μg	VA 30 μg	ΑΜΚ 30 μg	СТХ 30 µg	CRX 30 µg
Bacilli sp.	S	S	S	S	S	R	S	S	R	R
Pseudomonas sp.	S	I	S	R	S	R	S	S	R	R
Enterobacter sp.	S	S	S	R	S	R	S	S	R	R
Klebsiella sp.	S	R	S	Ι	S	R	S	S	R	R
Escherichia coli	S	Ι	S	R	S	R	S	S	R	R

AMP: ampicillin, CRX: cefuroxime, GEN: gentamicin, CIP: ciprofloxacin, CHL: chloramphenicol, AMC: amoxiclav, VA: vancomycin, AMK: amikacin, TET: tetracycline, CTX: ceftriaxone, R: resistant, S: susceptible, I: intermediate.

susceptible to Gentamicin, Ciprofloxacin, Amoxiclav, Vancomycin, and Amikacin. In a study by Sweeney et al. [36] on the bioburden of bacteria in preserved lens care solutions, coagulase-negative staphylococcus, *Bacilli* sp, *Corynebacteria* sp. and *Pseudomonas* sp. were found to be prevalent. However, whereas samples were aspirated from the residue of the lens care solution bottles in this study, the samples were collected from the nozzles and contents of the bottles in that study [36].

The ability of the isolated bacteria to thrive is based on the fact that the activity against them may fall during the storage of multipurpose solutions (MPS) [10]. In addition, some organisms may be able to utilize lens solution ingredients as nutrients for growth [10]. The presence of Gram-negative bacteria, even in small numbers, is considered an important bioburden, since these species are reported to be linked with corneal infection, swelling, and perforation, and are hardly ever isolated in asymptomatic subjects [37].

The limitation of this study was a lack of information on the type of multipurpose solution used, particularly, the types of active microbial components used, and therefore, it is recommended that future studies take into account the types of active microbial components used.

In light of the findings of this study, it is recommended that practitioners counsel their patients to follow the manufacturer's instructions for storage and disposal (after 90 days of use) of contact lens solutions and to practice proper hand hygiene before handling contact lens solutions and systems. In Ghana, many contact lens wearers only attend yearly follow-up visits [25], which undermines the importance of regular follow-up, especially considering the high contamination rate of contact lens solutions. Therefore, it is recommended that eye care practitioners counsel their patients to attend follow-up visits frequently. This will allow the practitioners to regularly reinforce the need for solution compliance and potentially reduce contamination.

5. Conclusion

Contact lens care solutions have been found to harbour multiple antibiotic-resistant bacteria that are potentially pathogenic to the corneal surface. The contamination is associated with some unhealthy solution-care practices among wearers. It is recommended that patients should be instructed to establish and maintain a lens-care regimen that adheres to the guidelines provided by the manufacturer of the products they utilize.

Conflict of Interest Statement

The authors declare no conflict of interest

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K.G. Owusu et al.

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