

Study of Microbial Contamination of Some Commercial Contact Lens Solutions

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Abstract

Microbial contamination is very common among contact lens users whether contamination involves the solutions or the lens. Many bacteria cause eye infections through this route and some may lead to serious outcomes to the eye. Most isolated bacteria from previous studies on the subject belong to Gram negative bacteria and few Gram positive bacteria. Our study was aimed at identifying microbial contamination of contact lens solutions from students at the College of Pharmacy/ University of Baghdad. The lenses used were varied from medical to cosmetic types. Both used and unused solutions were subjected for culturing to diagnose any microbial contamination. Thirty six (60%) of used samples showed bacterial growth. *Pseudomonas aeruginosa* accounts for the highest number of isolates (25%) followed by *E. coli* (21%), *Staphylococcus epidermidis* (6.6%), *Pseudomonas fluorescense* (5%) and *Proteus mirabilis* (1.6%) respectively. Only one unused (sealed) sample showed growth of *P. fluorescense*. Fungal growth was absent in both used and unused lenses. The bacterial contamination is likely to come from bad personal hygiene and improper or misuse of the solutions where these bacteria especially *P. aeruginosa* are frequently found in various environments from skin to solid materials and surfaces and are known to thrive in harsh environments. No relation was found between eye associated diseases and solution contamination among contact lens users. Special care should be paid in maintaining aseptic solutions and proper handling to avoid transmitting harmful bacteria to the eye where it may lead to serious eye infections.

Keywords: Contact lens solution, Microbial contamination, Eye infections, *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*

دراسة التلوث المايكروبي لبعض محاليل العدسات اللاصقة التجارية

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الخلاصة

يعتبر التلوث الميكروبي من الامور الشائعة عند مستخدمي العدسات اللاصقة بغض النظر اذا كان التلوث يحدث في المحاليل الخاصة ام العدسات نفسها. يوجد العديد من البكتيريا التي تسبب عدوى للعين عن طريق هذا المسلك من الاصابه و قسم منها قد تؤدي الى عواقب خطيرة للعين. اشارت الدراسات السابقة في هذا المجال الى ان اكثر البكتيريا المعزولة هي سالبة لصبغة كرام و القليل من المجاميع الموجبة لصبغة كرام. تهدف الدراسة الى تشخيص التلوث الميكروبي لمحاليل العدسات اللاصقة بين طلاب كلية الصيدلة في جامعة بغداد. تم جمع ستون عينة من محاليل العدسات اللاصقة المتوفرة تجارياً من طلاب كلية الصيدلة / جامعة بغداد. تتنوع أنواع العدسات المستخدمة من الطبية إلى التجميلية. تم وضعها في اوساط زرعية لتشخيص أي تلوث ميكروبي داخل المحاليل. كانت المحاليل المستعملة وغير المستعملة (الجديدة) خاضعة للزرع المايكروبي. أظهرت ست وثلاثون (60%) من العينات المستعملة نموًا بكتيريًا ، ولم يظهر أي نمو فطري. تمثل *Pseudomonas aeruginosa* أكبر عدد من العزلات (25%) تليها (*E. coli* (21%)، *Staphylococcus epidermidis* (6.6%)، *Pseudomonas fluorescense* (5%) و (*Proteus mirabilis* (1.6%) على التوالي. أظهرت عينة واحدة غير مستعملة (جديدة) نمو *Pseudomonas fluorescense*. من المحتمل أن يكون هذا التلوث البكتيري ناتجًا عن النظافة الشخصية السيئة وسوء استخدام المحاليل حيث توجد هذه البكتيريا وخاصة *P. aeruginosa* في بيئات مختلفة من الجلد إلى المواد الصلبة والأسطح ومن المعروف أنها تزدهر في البيئات القاسية. لم يتم العثور على علاقة بين الأمراض المرتبطة بالعين وتلوث المحلول ضمن مستخدمي العدسات اللاصقة. يجب إيلاء عناية خاصة في الحفاظ على المحاليل المعقمة والتعامل المناسب لتجنب انتقال البكتيريا الضارة إلى العين حيث قد تؤدي إلى التهابات خطيرة للعين.

الكلمات المفتاحية: محلول العدسات اللاصقة، التلوث المايكروبي، إصابات العين، *Pseudomonas aeruginosa*، *Staphylococcus epidermidis*

Introduction

Contact lenses are thin plastic lens worn between the eye and eyelid that may be used instead of eyeglasses. Actors, models, and others wear them for appearance, and athletes use them for safety and convenience. Contact lenses may also be used to correct certain abnormalities of the eye that cannot be corrected by regular glasses. It was invented in 1887 but was not used until 1938 when the first plastic contact lens was introduced. In 1950, the

corneal contact lens was introduced. It covered only the cornea of the eye, floated on the tears of the wearer, and could be worn all day without difficulty. Recent improvements include flexible lenses that shorten the initial period of adjustment for the wearer and porous lenses that do not have to be removed each day. Today, contact lenses that "breathe" have become popular. They allow oxygen to get to the cornea, preventing blurred vision due to the corneal exhaustion Syndrome^(1,2).

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The solutions that preserve contact lenses are subject to contamination by microorganisms such as bacteria and fungi and may lead to serious infections through the invasion of the cornea and conjunctiva⁽³⁾.

Staphylococcus aureus, *Pseudomonas*, *Bacillus*, *Proteus*, *Haemophilus*, *Enterobacter*, *Serratia*, and *Klebsiella* spp. are the most common isolated bacteria from eye infections related to contaminated medications⁽⁴⁾. These bacteria can damage important functional structures which might lead to vision loss and blindness⁽⁵⁾. Bacterial keratitis and endophthalmitis are some of these serious infections that were found to be associated with the use of contaminated topical medications⁽⁶⁻⁸⁾. The contamination rate of in-use ophthalmic solutions is variable according to published researches and ranges from 0.07% to 35.8%⁽⁹⁾. Most eye preparations may possess antimicrobial effect and if not they are provided with antimicrobial substances to prevent contamination. These substances prevent microbial growth to avoid infection of the eye and drug degradation⁽¹⁰⁾. In a study by Tsegawa *et.al* on 100 ophthalmic solutions used by patients and eye care workers cultured for microbial contamination, it was found that 11% were contaminated with various bacterial species and the significant findings of this study were the isolation of resistant *Staphylococcus aureus* and coagulase negative *Staphylococcus* both were methicillin resistant among other species⁽⁴⁾.

The current study focused on screening for microbial contamination in commercially eye lens solution used by students at College of Pharmacy/University of Baghdad.

Materials and Methods

Specimen collection

The samples were collected from October 2018 until May 2019. Sixty samples were collected from students at College of Pharmacy/University of Baghdad.

Sample characteristic

- 1-The samples were collected from 58 females and 2 males.
- 2-Fifty eight samples were extended wear, and 2 samples were daily wear.
- 3-Five samples have been used as medical lenses, while 55 samples have been used as cosmetic lenses.
- 4-Associated eye disease: one sample has a previous history of Toxoplasmosis retinitis infection in the right eye. While the other samples have astigmatism as diagnosed by ophthalmologists.
- 5-The manufacturing companies were the most common available in the Iraqi market and they were; BELLASOFT, NEOPLUS, FRESHEYE, SAFE & SOUND, HD, MY CHOICE, ANASTASIA, DESIO, CUTE, AQUASOFT, HEIDI, FRESHLOOK and they were manufactured mainly in Korea, Syria and France.

- 6-The collected solutions were divided into two groups: 20 new and 40 used solutions for comparison and culturing.

Culture media

The samples were cultivated on nutrient agar, MacConkey agar, Mannitol salt agar and blood agar to isolate the bacteria. Sabaraud Dextrose agar was used to isolate the fungi. All media were sterilized in autoclave at 121°C for 15 min. prior for sub culturing. Duplicate cultures were made for each sample.

Identification of microorganisms

Nutrient agar was used to identify gram positive bacteria, while MacConkey agar was used to identify gram negative bacteria. Hydrogen peroxide solution (3%) was used to distinguish between *Staphylococcus* and *Streptococcus*. Mannitol salt agar was used to differentiate *Staphylococcus aureus* growth from *Staphylococcus epidermidis*. Gram negative bacteria were isolated by MacConkey agar, Oxidase test and growth at 4°C/42°C(both were used to identify *Pseudomonas* species) and Triple Sugar Iron agar test also done for confirmation of the identification. The API 20E test system was used for the performance of 20 standard biochemical tests that differentiate various species of *Enterobacteriaceae* & other gram negative bacteria⁽¹¹⁾.

Results and Discussion

Results

Of the total 60 samples collected and cultured, all new 20 solutions (not used with contact lenses) showed negative growth except one sample showed growth of *Pseudomonas fluorescense* while the used solutions showed that 36 out of 40 samples were contaminated with bacteria. No fungal growth was observed in all samples. Table 1 shows the types of bacterial pathogens isolated from the samples with number and percentage of contamination analyzed through SPSS program.

Table 1. Microorganisms isolated from contaminated used lens solutions samples

Pathogen	No. of samples	%
<i>Pseudomonas aeruginosa</i>	15	41.6
<i>Escherichia coli</i>	13	36.1
<i>Staphylococcus epidermidis</i>	4	11.1
<i>Pseudomonas fluorescense</i>	3	8.3
<i>Proteus mirabilis</i>	1	2.7
Total	36 samples	100

Discussion

Pseudomonas aeruginosa was the most frequent isolated bacteria from samples that included in this study. Contamination with *Pseudomonas aeruginosa* may be due to many reasons, as bad storage of the solution, long time of the solution storage without replacing it frequently, bad hygiene means, when the lens removed from the eye and inserted directly into the solution bags without rinsing or washing the lenses with the solution.

Sometimes, the eye was infected before wearing the contact lenses. Contact lens with the persistence eye diseases may worsen the condition⁽¹²⁾.

In the past, hard contact lenses were sterilized in boiling water. When they were replaced by soft lenses, boiling water sterilization was discarded due to the damage and deterioration that it causes to these soft ones. Furthermore, this type of sterilization causes allergic diseases⁽¹³⁾. Care system bottles can be easily contaminated and become a source of microbes that contaminate the lens storage cases^(14,15).

The results of the current study revealed that all new solutions showed no growth except one sample which was contaminated by *Pseudomonas fluorescens*. All types of care solutions, including hydrogen peroxide, have been reported to contain microorganisms even in experienced users or unopened factory sealed bottles^(16,17). It may be possible that contamination occurred during packaging in the factory. No gram positive bacteria or fungus were found in the cleaning solutions.

Gram negative bacteria have the best opportunity to contaminate lens care solutions due to their ability to adapt to various environments even with minimum nutritional requirements⁽¹⁸⁾.

The reason for such findings might be due to the fact that the moist tips of solutions might serve as a reservoir for microbial contamination, and this will put the risk of contaminating the eye drop with bacteria as a result of touching the dropper tips while opening it for usage, contact of dropper with the ocular tissues, and environmental factors⁽⁴⁾. Lens care solutions are provided with preservatives and some disinfectants such as Alexidine Dihydrochloride, Polyhexamethylene Biguanide, Oxylchlorite complex (sodium chlorite and hydrogen peroxide), benzalkonium chloride and other compounds. These are added to maintain low levels of microorganism during the product's life cycle whilst having minimal impact on the ocular surface⁽¹⁹⁾. Narayana *et.al* observed in their study that the antimicrobial activity of different solutions varies with respect to time of incubation, and also there was a marked difference in the activity of some solutions against some pathogens like *Pseudomonas aeruginosa* and *Staphylococcus aureus* and they

concluded that the users should store their lenses in solutions for longer periods of time⁽²⁰⁾. The stock

solutions are intended to prevent microbial growth due to their antimicrobial efficacy however, common pathogens of the eye such as *Staphylococcus aureus* and *Pseudomonas aeruginosa* are becoming increasingly resistant to some commercial solutions^(12,21).

Gram negative bacteria, especially *Pseudomonas aeruginosa* has the ability to form biofilms on various materials both living and non-living surfaces and the biofilm formation is an obstacle concerning the uses and design of ocular devices, such as contact lenses because it shelters the bacteria from harmful factors like antibiotics and the immune system thus leading to infection of the eye⁽²²⁾. Oliver *et.al* mentioned in his study that biofilms of *P. aeruginosa* resist multipurpose contact lens solutions and cause contamination but ozonized water and chlorohexidine reduced biofilm formation by reduction in bacteria numbers with ozonized water much more preferable as it doesn't deposit any toxic residues like chlorohexidine⁽²³⁾.

Escherichia coli are the 2nd most contaminant isolated in our study. It has been isolated from contact lens solutions from previous studies and cause a problem in daily wear lens users^(24,25). As mentioned by Demirbilek and Evrenin their study on multipurpose contact lens solutions, 5% Povidon iodine was more effective in elimination of this bacteria⁽²⁶⁾.

As for the single isolate of *Pseudomonas fluorescens*, this bacterium is common in soil and it is known to cause infections in immunocompromised persons such as cancer patients through blood transfusions and saline solutions contamination⁽²⁷⁾. It is likely that this bacterium contaminated the solution during the manufacturing and filling of the product within the factory limits as it was found in new contact lens solution sample (sealed).

While *Staphylococcus epidermidis* is a common commensal inhabitant of the skin at various sites, this bacteria is now widely studied and under monitoring due to increasing nosocomial infections caused by it. Some authors call it "the accidental pathogen" where it infects immune-compromised hosts and introduced to the system via instrument like urethral catheters. There is also an increasing resistance among nosocomial infections with these bacteria⁽²⁸⁾. The isolation of this bacterium among contact lens solutions most likely came from hands or contaminated bottle tips from the environment where these bacteria are commonly found. Correa *et.al* mentioned that *S. epidermidis* was one of the isolated bacteria from silicone hydrogel contact lens if they were used incorrectly or maladapted and lead to eye infection. Also, this may lead to

contamination of solutions if they were Bacteriocidally inefficient in eliminating them⁽²⁹⁾.

Proteus mirabilis is well known as one of the most frequent causative agent of urinary tract infections as well as causing wound infection, sepsis and pneumonia in hospitalized patients⁽³⁰⁾. It also can contaminate any material due to its actively swarming movement on solid surfaces. Lipener *et al* mentioned in his study that *P. mirabilis* was the second most frequently isolated bacteria along with *P. aeruginosa* from contact lens saline solutions with a percentage of 60%⁽³¹⁾. It is likely that these bacteria contaminated the solutions via hands with poor hygiene or improper handling of the solutions. There were no relation between eye associated diseases and solution contamination as both healthy and disease associated eyes showed contamination of contact lens solutions by their users.

Conclusion

Contact lens solutions users should pay more attention to restrictions and medical guidelines in maintaining aseptic solutions free from microbial contamination. Lens solution could be a source of eye infection if aseptic procedures were ignored. College students are likely to get contamination and infections due to the crowded environment where they study and that there are many sources and material that provide these microbes.

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