Microbial Contamination of Eye Drops in out Patient in Iraq[#]

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Abstract

Acontaminated ophthalmic solutions represent a potential cause of avoidable ocular infection. This study aimed to determine the magnitude and pattern of microbial contamination of eye drops in out patient at the department of ophthalmology, at Baghdad national hospital, Iraq. Fifty four vials from the out patient clinic were obtained for microbial examination after an average use of 2 weeks. The dropper tip and the residual eye drop were examined for contamination. The specimens were cultured, the number of colonies counted, the organisms identified. Eight (15%) out of 54 analyzed vials were contaminated , most bacteria identified belonged to the normal commensal flora of the eye. Isolated contaminants were *Staphylococcus auereus, Micrococcus , Neisseria catarrhalis, Gram negative Rods, Candida albicans, and Staph epidermidus.*The dropper tip was more often contaminated (n=5) than the residual solution (n=2) and only one vial showed acontamination of both the drop and the tip (n=1) . Our data show acontamination rate of 15%, which is in the medium range of data puplished on the contamination of eye drops elsewhere (0.07% to 35.8%).

Key words : Microbial Contamination, Eye Drops

الخلاصة

يمثل تلوث قطرات العيون خطراً كبيراً لانه السبب الرئيسي في التهاب العين. ومخاطر ذلك قد يؤدي الى العمى احياناً لذلك تهدف هذه الدراسة الى تحديد مدى ومستوى التلوث المايكروبي لقطرات العيون للمرضى في عيادة الردهة الخارجية لقسم العيون في مستشفى بغداد العام في العراق. اربعة وخمسون قطرة جمعت وتم قياس التلوث المايكروبي لها بعد معدل استعمال اسبوعين. راس القطرة وبقية المحلول المتبقي خضع للفحص المايكروني وبعد زرع وعد المستعمرات تم تحديد انواع البكتريا الملوثة. ثمانية قطرات اي بمعدل (١٥٪) من مجموع (٥٤) قطرة تحوي بكتريا ملوثة معظمها تعود الى البكتريا الطبيعية الموجودة في العين او الجلد ومنها *Staphylococcus aureus, Micrococcus, Neisseria catarrhalis, Gram negative rods and Candida albicansand Staph epidermidus* رأس القطرة وجد ايضاً اكثر تلوثاً من بقية السائل المتبقي في القطرة وواحد فقط اظهر تلوث السائل و رأس القطرة.مستوى التلوث (١٥٪) يعتبر حد وسط للتلوث المايكروبي لقطرات العين من المعلومات المنشورة في انحاء العالم وهو ما حدد بـ(٣٥,٨-٠,٠٧)% .

Introduction

Contaminated eye drops and ophthalmic solutions are a potential cause of ocular infection. They can be associated with keratitis.[1] and corneal ulcers[2] and carry the risk of transmitting opportunistic micro-organisms, [3,4]as well as pathogenic organisms, such as *pseudomonas aeruginosa* and *Serratia marcescens*. [1] The published contamination rate of in-use ophthalmic solutions varies widely in the literature from 0.07% [5] to 35.8%.[3] A part from the risk of infection, bacterial contamination of eye drops may alter the pH of the solution and therefore reduce the efficacy of the drug [6].In order to prevent contamination, most preparations contain antimicrobial substances, unless the solution it self has an antimicrobial effect. These substances aim to preventing or inhibiting the growth of microorganisms which increase the risk of infection or degradation of the drug. The self sterilizing effect of eye drops caused by the presence of preservatives has been discussed controversially[7]. preservatives must meet several requirements (1) to be compatible with other ingredient ,(2) to be efficient during the entire duration of use of eye drops, and (3) to be non toxic. Commonly used preservatives of ophthalmic solutions are benzalkonium chloride, which also works as a detergent and therefore increases the penetration of the active ingrediente of the drug, thiomers; chlorhexdine; parahydroxy benzoate; phenylmercuric nitrate, EDTA, chlorobutanol; benzylalcohol; phenylethyl alcohol; and parabens[8,9]**.** As preservatives interfere with the metabolism and inhibit the growth of micro-organisms, they may have similar effects on human cells, explaining potential cytotoxic effects and inflammatory cell responses.[6] The antimicrobial activity is important for the rate of infection resulting from contamination during the process of instillation. Contact with fingers or lids, ciliaries conjunctiva and cornea are possible causes of contamination even if instilled by healthcare professionals.

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Plastic bottles have been reported to be more commonly contaminated near the bottle cap. this has been attributed to a lack of preservative at this area.[4] In a clinical study 220 in-use medications of 101 patients with non-microbial acular surface disorders were examined by cultivating the bottle caps. the authors concluded that acycle of contamination between in-use medications and conjunctiva may present an important risk factor for microbial keratitis in patients with ocular surface disease[10].The occurrence of bacterial ocular infection such as keratitis and endophthalmitis transmitted by contaminated eye droppers has been reported.[11] In a recent study the authors noted that some cases of bacterial keratitis in Iran are thought to be due to contaminated eye drops used on multiple patients [12]. Brudieu et al found abig difference in contamination rates between vials used by ophthalmological patients (17.7%) and vials used by medical and gerontological patients (35.8%). A positive correlation was also found for vial contamianation and the duration of use vials containing an antimicrobial agent were less likely to be contaminated than vials without antimicrobials. However no clinically relevant infection through such vial contamination was identified[3]. In a study comparing the contamination rate of drops used in an eye department and a nursing home no difference was found but the authors stated that the residual eye drop is more often contaminated than the tip of the bottle. They also noticed the presence of. Gram negative organisms in the nursing home [7]. Most studies, however, found the bottle tips to be more often contaminated than the solution.[2,4,13] Therefore, topical eye medications may present a potential risk of infection, aspecially if the ocular epithelial barrier is compromised. Minimising the contamination of eye drops and the transmission of infections is an important issue in clinical ophthalmology. Several regulations and suggestions have been made in this context. We conducted the following cross sectional study and analysed (54) ophthalmic solutions examining bacterial and fungal contamination. This study aimed to determine the magnitude and pattern of microbial contamination multi dose of eye drops in out patient.

Material and Methods

A total of (54) eye drop containers (in use) were obtained for microbial examination. The microbial analysis was performed on the dropper tip and the residual eye drop for each containers. The eye drops were obtained and cultured according to the following:

- A sterile cotton tip swab was moistened in sterile brain heart infusion (BHI) enriched medium before wiping the nozzle tip of the eye drop containers and then used to inoculate the culture plates[13].
- The vials were inverted and one drop was directly inoculated on each of the media and then spread across the plates.

All media except the sabouraud agar plates were incubated at 37 ^{0}C for 48hrs. and evaluated after 24 and 48hrs. the blood agar, chocolate blood agar plates were incubated in amicroaerophile environment.[15] The sabourand agar plates were incubated at 30 ^{0}C for up to 10 days and evaluated for growth on days 1, 5 and 10. The BHI broth was also incubated at 37 ^{0}C and subcultured on blood agar after 24 hours. All culture media except sabouraud dextrose agar (BioMerieux, F) were obtained from Biotec Laboratories Ltd, UK. A significant growth was considered a growth on the main inoculation site or on two or more streaks on the plate. The BHI was analysed for changes in colour and turbidity of the media. The colonies on solid media were counted and all organisms identified by microscopy after Gram staining and biochemical tests.

Results

A total of 54 medications were analysed; from the out patient clinic. as shown in table (1) the used of different preservatives found in the analysed specimens. They were grouped in four different categories *analgesics, antihistaminc, antibiotic and steroids.* . Over all, 8(15%) of the 54 analysed vials were contaminated table (2) at the bottle tip alone or with additional contamination of the solution or solution alone. Within the four categories the rate of contamination varied between 0% for analgesics and 17% ---> for both antibiotics and Steroids.The dropper tip was more often contaminated (n=5) than the residual solution (n=2). One bottle showed contamination of both the dropper tip and the medical solution. (n=1) Most of the identified organisms were part of the normal skin flora or conjunctival flora. Gram positive organisms were cultivated from five of the eight contaminated medications and two contaminated medications grew Gram negative organisms and one grew *Candida albicans.* One of the medications grew more than one bacterium as shown in table 3 . None of the medications were found to be past the expiry date.

Table 1 : Type of preservatives and number of medications

No.	Preservative	Number of medications
1	**Benzalkonium chloride0.05%**	**20**
2	**Benzalkonium chloride0.02%**	**14**
3	**Benzalkonium chloride0.01%**	**8**
4	**b.chloride 0.004%**	**4**
5	**Thiomersal 0.005%**	**4**
6	**Phenyl mereuric nitrate 0.001%**	**4**
	Total	**54**

Table 2 : Eye medication and contamination

Eye medication	Number tested	Contaminated %	Tip contamination	Tip/drop contamin active	(n=54)drop* contamination	Contaminant
Analgesic	**4**	**0(0%)**				
Antihistamine	**14**	**2(14%)**	**-**	**-**	**2(14%)**	**Two type colonies 1-white colonies coagulase(-). 2-Yellow colonies coagulase(+). *Staph. Aureus***
Antibiotics	**24**	**4(17%)**	**4(17%)**	**-**	**-**	**1-*Candida albicans* *2-Staph aureus* *3-Neisseria catarrhalis***
Steroid	**12**	**2(17%)**	**1(8%)**	**1(8%)**		***Micrococcus* Gram negative rods**
Total	**54**	**8(15%)**	**5(9%)**	**1(2%)**	**2(4%)**	

*(n= 54) total eye drops specimens

Table 3 : Contaminated medications and preservatives (n=54).

No.	Eye medication	Preservative	Contaminant
1	Antistin-privin	Benzalkonium chloride 0.02%	2types of colonies on blood white colonies and yellow (*Staph aureus*)
2	Antistin-privin	Benzalkonium chloride 0.02%	2types of colonies on blood white colonies and yellow *(staph aureus*)
3	Oflox	Benzalkonium chloride 0.05%	On MSA *Staph aureus*
4	Oflox	Benzalkonium chloride 0.05%	On blood agar :Candida albicans *Staph. aureus*
5	Oflox	Benzalkonium chloride 0.05%	*Neisseria catarrhalis*
6	Oflox	Benzalkonium chloride 0.05%	*Staph aureus*
7	Dexamethason 0.1%	Benzalkonium chloride 0.01%	*Micrococcus*
8	Dexamethasone phosphate. 0.1%	Benzalkonium chloride 0.01%	*G- Rods*

Discussion

We noticed amicrobial contamination of 8/54(15%) of out use application dispensers. The mean contamination rate of preserved eye drops described in the literature varies widely from 0.07%. [5] to 35.8%. Six different micro organismes were detected. As the containers were analysed on the day of collection, our results are likely to represent the specific clinical situation of that day. Five of eight of the identified organisms were Gram positive and 2/8 Gram negative and one *Candida* spp.Most of the organisms were part of normal commensal flora of the conjunctiva or the skin. The resident flora of the conjunctiva and eyelid mainly comprises of Gram positive bacteria, including coagulase negative *Staphylococci*, *Corynebacterium* spp. *Propionibacterium* spp. as well as *Staphylococcus aureus*, *Bacillus* spp. *Micrococcus* spp. and *Enterobacter* spp.[17,18] This is in accordance with several other published studies.[7,13,16] However, it differs from results puplished by Rahman etal,[19] who found only a small proportion of the microorganisms identified to be post of the normal commensals flora when studying the contamination of unpreserved eye drops.A cycle of contamination between the lids and dropper tips was suggested by Schein et al.[10] The contamination of eye drops and eye drop dispenser with the same microorganism, especially gram negative, has been described by the same group. This represents apotentially serious risk for ocular infection, especially in cases of compromised corneal epithelium as in extensive contact lens wear, ocular trauma or the use of topical steroids. In this study pathogenic organisms were rare and showed limited growth that probably did not represent a clinically relevant risk of infection. By using t.test we did not find any significant difference ($p<0.05$) in the contamination rate of eye drops/dropper tips and the residual volume which was left in the bottle. This supports the previously described self sterilising effect of many eye medications.The design of the containers might also influence contamination. Only bottles with a tip attached to the bottle itself were analysed in this study. The bottle tips were more often contaminated (n=5) than residual drops (n=2) with contamination of both the tip and the residual solution appearing in one specimen. These results are similar to the ones reported in earlier studies.[11,13,16] One reason for this pattern to be considered is the antimicrobial activity of preservatives of the solution itself. Such antimicrobial effects, however, may not act sufficiently on the tip itself as the contact time is limited. Further the tip provides a large surface for contamination from ocular structures or hands. Even dried crusts can sometimes be found on the bottle tips.The removal of such remnants with a sterile swab might further reduce the contaminator rate.[4] However the contamination of the solution itself has to be regarded as clinically more relevant since these get in direct contact with the patients eye.Non of the examined eye drops is expired and we recommend not storing any open bottles in the back of drawers or on top shelves but always keeping them handy and limited to those actually needed. Further more, we recommend noting the date of first opening on each container, as the duration of use might be another and possibly more relevant parameter rather than the expiry date[5].

Conclusion

Aclear instruction sheet about safe and effective used should accompany do when they are dispensed to patient. Patients who are unable to use eye drops in an aseptic way because of age or other physical (for example, poor vision) or mental limitations should be assisted by competent relatives or caretakers. The results of this study support the importance of a proper set of rules and the correct handling and application of eye medications.

References

1. Mayo MS, Schlitzer RL, Ward MA, et al. Association of Pseudomonas and Serratia corneal ulcers with use of contaminated solutions. J clin microbial 1987;25: 398-400.
2. Donzis PB. Corneal ulcer associated with contamination of aerosol saline spray tip. A.M J ophthalmol 1997;124:394-5.
3. Brudieu E, Duc DL, Masella JJ, et al. Bacteral contamination of multidose ocular solutions. Aprospective study at the Grenoble Teaching Hospital pathol Biol(paris) 1999;47:1065-70.
4. Clark PJ, ong B, stanely CB. Contamination of diagnostic ophthalmic solutions in primary eye care settings. Mil med 1997; 162. 501-6.
5. Wessels IF, Bekendamp, calvin WS, et al. open drops in ophthalmday Y offices: expiration and contamination. Ophthalmic Surg. lasers 1999;30:540-6.
6. Perry HD, Donnenfeld ED. Issues in the use of preservative free topical manag care 2003-; 12:39-41.
7. Raynaud C, Laveran H, Rigal D, et al. bacterial contamination of eye drops in

clinical use JFr ophthalmol 1997; 20:17-24.

8. Zanen A. prevention of infections in the ophthanology office. Bull socbelge ophthalmol 1996;260:9-16.

9. Soc. Belge Z. Antimicrobial agents in eye drops. Ceska slov farm 2004;53:107-16.

10. Schein OD, Hibberd PL, Starek T, et al. Microbial contamination of in use ocular medications. Arch ophthalmol 1992; 110; 82-5.

11. Coad CT, ostato MS, wihelmus KR. Bacterial contamination of eye drop dispensers. Am J ophthalmol 1984, 98: 548-51.

12. Jalali R, Zinolabedini F, Moradi M, et al. Bacterial contamination rate of eye drops. Comparison of hospital and a private out patient center in Kermanshah, Iran. In sight 2004, 29: 12-4.

13. Geyer O, Bottone EJ, podos SM, et al. Microbial contamination of medications used to treat glaucoma. Br. j opthalmol 1995; 79:376-9.

14. British National formulary. London; British Medical Assocation and Royal pharmaceutical society of Great Britain, 1997:441.

15. Schalla UC. Untersuchungen zur mikrobiologischen probengewinnuing und transport bei bakteriellen Entzundungen des äuBeren Auges. Munich: Ludwig-Maximilians-Universitat, 1998.

16. Hovding G, Sj ursen H. Bacteral contamination of drops and dropper tips of in use multidose eye drop botlles. Acta Ohthalnol (copenh). 1982; 60:213-22.

17. Ta CN, chang RT, Singh K, et al Antibiotic resistance patterns of ocular bacterial flora aprospective study of patients undergoing anterior segment surgery. Ophthalmology 2003; 110: 1946-51.

18. Leong JK, Shah R, McCluskey PJ, et al. bacterial contamination of the anterior chamber during phacoemulsification cataract surgery. J cataract Refract surg 2002; 28: 826-33.

19. Rahmam MQ, Tejwani D, Wilson, JA, et al. Microbial contamination of preservative free eye drops in multiple application containers. Br J ophthalmol 2006; 90. 139-41.