นิพนธ์ต้นฉบับ

Original Article

การปนเปื้อนของเชื้อแบคทีเรียและสิ่งแปลกปลอมของเลนส์สัมผัสสี ในอำเภอวารินชำราบ จังหวัดอุบลราชธานี ประเทศไทย

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บทคัดย่อ

วัตถุประสงค์ในการศึกษาครั้งนี้เพื่อศึกษาแบบภาคตัดขวางของการปนเปื้อนของเชื้อแบคทีเรียและสิ่งแปลกปลอมใน เลนส์สัมผัสสีเพื่อความสวยงามชนิดรายเดือนจากร้านจำหน่ายผลิตภัณฑ์เพื่อความงามจำนวน 4 ร้านค้าและ 1 ร้านค้าในตลาดนัด ในอำเภอวารินชำราบ จังหวัดอุบลราชธานี ทำการวิเคราะห์การปนเปื้อนของเชื้อแบคทีเรียด้วยวิธีการเพาะเชื้อบนอาหารเลี้ยงเชื้อ ตรวจสัณฐานวิทยาโดยการย้อมแกรมและทดสอบทางชีวเคมี และหาสิ่งแปลกปลอมและสิ่งปนเปื้อนอื่น ๆ ภายใต้กล้องจุลทรรศน์ ผลการศึกษาพบว่ามีการปนเปื้อนของเชื้อแบคทีเรียจำนวน 4 ตัวอย่างจากทั้งหมด 58 ตัวอย่าง ที่ระยะเวลาที่แตกต่างกันตั้งแต่ 24-72 ชั่วโมง โดยเชื้อที่พบปนเปื้อนในเลนส์สัมผัสสีเพื่อความสวยงามชนิดรายเดือน ได้แก่ *Bacillus sp., Staphylococcus aureus, Citrobacter sp.* และ *Staphylococcus epidermidis* นอกจากนี้ยังพบการปนเปื้อนของสิ่งแปลกปลอมอีกด้วย การศึกษาในครั้ง นี้แสดงให้เห็นว่าผู้บริโภคควรระมัดระวังในการซื้อและการดูแลเลนส์สัมผัสสีเพื่อความสวยงามชนิดรายเดือนมากขึ้นเพื่อลดความ เสี่ยงและเพิ่มความปลอดภัยที่เกิดจากการใช้เลนส์สัมผัสสี

คำสำคัญ : เลนส์สัมผัสสี สิ่งแปลกปลอม การปนเปื้อนเชื้อแบคทีเรีย ร้านจำหน่ายผลิตภัณฑ์เพื่อความงาม ตลาดนัด

Bacterial and Foreign Body Contamination of Colored Contact Lenses in Warin Chamrap, Ubon Ratchathani, Thailand

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Abstract

Objectives: This study aims to investigate bacterial and foreign body contamination in cosmetically monthly colored contact lenses from 4 beauty shops and a flea market in Warin Chamrap, Ubon Ratchathani, Thailand.

Methods: A cross-sectional study of colored contact lenses. The 58 samples, 11 brands and 2 vial sizes of monthly cosmetically colored contact lenses. The contact lenses and contact lenses solution was observed under light microscopic examination to find foreign bodies and other contamination. All of the colored contact lens samples underwent microbial culture using nutrient agar, a growth colony was made by biochemical tests and Gram's staining to confirm the isolated organism.

Results: From 58 samples, the results showed bacterial growth from 4 of 58 samples in both of contact

lenses and contact lenses solution in different duration from 24-72 hours. The results revealed that *Bacillus sp. Staphylococcus aureus, Citrobacter sp.*, and *Staphylococcus epidermidis* were contaminated in cosmetically monthly colored contact lenses in this study. The result the foreign body also found in another vial CL solution under light microscope. **Conclusion** : The study showed bacterial and foreign body contamination in cosmetically monthly colored contact lenses. This study shows that consumers should be more cautious when purchasing and caring for monthly colored CL to reduce the risks and increase the safety associated with the use of colored CL.

Keywords : colored contact lenses, foreign body, bacterial contamination, beauty shop, flea market

Introduction

The colored contact lenses (CLs) allow its users to correct their eye anomalies and change the eye color to create a look that is subtle, bold or anywhere in between whether the users want to enhance their everyday look or decorate themselves for a special occasion¹. Cosmetically colored CLs are used purely for decorative purposes to change the eye color without having any vision re-correction properties². The use of cosmetically colored CLs is a familiar recent trend in the youngsters of middle to lower socioeconomic classes. Many of these young users are unaware and uninformed about the proper purchase and use of CLs³. Although the Thai FDA announcement regarding consumer safety in medical-purposed and cosmetic-purposes CLs sold in medical device stores or sold by contact lens importers/ sales representatives, incidences of eye infections from CLs use are rising worldwide, and also in Thailand⁴. Lack of compliance and poor hygiene towards lens care is strongly associated with foreign body and microbial contamination and has been proved to result in eye infections⁵. 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 cornea⁶. Bacterial colonization of CLs has also been implicated in CL-induced inflammation^{2,6} Specifically, CL acute red eye, CL peripheral ulcer, and infiltrative keratitis have all been associated with adherence of bacteria to hydrogel CLs⁷. Infiltrative keratitis and CL peripheral ulcer have been associated with Staphylococcus aureus, Streptococcus pneumoniae, Abiotrophia defectiva, Acinetobacter ssp, and others². Furthermore, microbial keratitis especially caused by Pseudomonas ssp., is the most serious complication associated with CL usage. It involves the entry and subsequent invasion of the corneal layers by the offending pathogens, which include structural as well as enzymatic components in which are responsible for the attachment of different bacteria to the cornea⁸. As a result of increased stress imposed by the contact lenses on the cornea, the latter is less able to defend itself against invading bacteria³. Thus, the bacterial effectively manages to overcome the eye's weakened defenses, precipitating a fulminant infection⁹.

Therefore, it is important to investigate the foreign body and bacterial contamination of the monthly cosmetically colored CL those sold in the beauty shops and a flea market around Warin Chamrap, Ubon Ratchathani, Thailand. As a result, the study was conducted to examine possible microbial contamination, the one of safety factors that affected the purchasing decision on cosmetically colored CLs, and to find out the guidelines for improving and developing the colored CL using in order to serve the needs of the consumers at present and in the future.

Materials and Methods

Cosmetically colored CL collection: This is a cross-sectional study that was conducted in Warin Chamrap of Ubon Ratchathani, Thailand. All untrained optical shops in this district that sell monthly colored cosmetic CL were included in this study. Every brand and vial size of monthly colored cosmetic CLs in each shop were sampled by an accidental sampling method. The 58 samples, 11 brands and 2 vial sizes of monthly cosmetically colored CLs were included in this study.

General appearance and physico-chemical properties of CLs: The CLs and CL solution was observed under light microscopic examination to find foreign bodies and other contamination. Specific gravity was measured by a refractometer. CL solution pH of each vial was evaluated by using pH meter and bacterial optical density (OD) were measured at 600 nm by spectrophotometer. Every sample was done in triplicate.

Bacterial culture: The examination of microbial contamination in this study was performed according to the Microbial limit test in the Thai Pharmacopoeia with some modifications. All of the colored CLs samples underwent microbial culture, we removed the lenses and CL solutions from their containers using sterile techniques and cultured them in nutrient agar at 37 degree Celsius for 24-72 hours. Every sample was done in duplicate. Afterwards, microbial growth was observed by clonal growth. After 72 hours of culture, the specimens that showed no growing colony were reported as no growth after 3 days.

Microscopic examination: The microscopic examination of a growth colony was made by Gram's staining to confirm the isolated organism.

Biochemical tests: The presence of specific pathogens was confirmed by biochemical testing. Biochemical tests were performed to identify the pathogens, including TSI agar, citrate agar, urea, motile, and indole according to standard methods.

Results

A total of 58 samples, cosmetically monthly colored CLs, were collected from 4 beauty shops and a flea market around Warin Chamrap of Ubon Ratchathani, Thailand. Each colored CL was contained in a vial sealed with an aluminum cap and no bottles were broken. In our previous study, external product defects, e.g. dirty products, unclear/unreadable labels, sticking on another label, and expired products were found. The inside products including CLs and CL solutions were investigated in this study.

General appearance and physicochemical properties of CLs

The result showed contamination in a CL (Figure 1A) and the foreign body also found in another vial CL solution (Figure 1B) under light microscope. We found

that all CL solutions from 11 brands were normal saline as label. Specific gravity of all 58 samples were not significantly different. The specific gravity of all solutions was between 1.004-1.010 (1.006 \pm 0.01).



Figure 1 Contamination in contact lenses and contact lens solution

(A) Contamination in contact lenses of sample No.16

(B) Contamination in contact lens solution of sample No.29

CL solution, normal saline solution, pH of each sample varied in range 6.65-8.40 (7.54 \pm 0.28) as shown in figure 2A. Interestingly, bacterial OD600nm of some samples were different as shown in Figure 2B.



Figure 2 pH and OD600nm of contact lens solution (A) pH of contact lens solution (B) OD600nm of contact lens solution

Bacterial culture

There was found bacterial growth from 4 of 58 samples (Table1) and also found in both of CL (1-2) and CL solution (3-4) in different duration from 24-72 hours (Table 2).

After colony isolation, Gram's stain (Figure 3),

Bacterial growth	Number of samples	Percentage
Growth	4	6.90
No growth	54	93.10
Total	58	

Table 1 Bacterial isolated from the contact lenses and contact lens solutions

Table 2 Duration of bacterial growth from the contact lenses and contact lens solutions

	Bacterial growth											
No. of plate	24 hours				48 hours				72 hours			
	1	2	3	4	1	2	3	4	1	2	3	4
25											\checkmark	
30							✓				\checkmark	
33			✓				✓				\checkmark	
34	\checkmark				\checkmark				\checkmark			

Note: \checkmark ; found colony growth

biochemical tests, and disc diffusion (Table 3) were done to identify the organisms. The results revealed that Bacillus sp. Staphylococcus aureus, Citrobacter sp., and Staphylococcus epidermidis were contaminated in monthly colored contact lenses in this study. **Discussions**



Figure 3 Gram's stain of bacterial colony isolation

										Standard		
No.	Gram's	Oxidase	Catalase	Coagulase		Bioc	hemical	Test		disc	Inhibition	Isolated
plate	stain	Test	Test	Test						diffusion	zone (cm.)	bacteria
					TSI	Indole	Motile	Citrate	Urea	Novobiocin		
25-3	Positive	Negative	Positive									Bacillus sp.
30-3	Positive	Negative	Positive	Positive	NN					sensitive	35	Staphylococcus aureus
	Negative	Negative	Positivo		KN	Negative	Motilo	Nenative	Negative			Citrobacter
33-3	Dopitivo	Nogativo	Positivo	Dopitivo	NN	i vogative	WOULD	i iogalive	INCYALINE	consitivo	27	sp. Staphylococcus
34-3	FUSILIVE	ivegalive	FUSILIVE	FUSILIVE	ININ					Sensitive	37	epidermidis

Abbreviation: NN; no change in slant/ no change in butt, KN; alkaline slant/ no change in butt

Nowadays, colored CLs wearers make up a significant and growing proportion of the contact lens wearing population in Asian countries, such as Taiwan, Korea, Singapore, Malaysia, Thailand, Hong Kong and China⁵. Cosmetically colored CLs, although originally developed for patients with disfiguring abnormalities of the iris and cornea⁹. Of these, contact lens-related microbial keratitis represents the most feared complication¹⁰. Apart from cosmetically colored CLs user behaviors, monthly colored CLs are unsafe because consumers might be uninformed about safety use CL⁵. In present study, the results from 4 beauty shops and a flea market around Warin Chamrap of Ubon Ratchathani, Thailand showed that ⁶.10 % of all cosmetically monthly colored CLs had Bacillus sp. S. aureus, Citrobacter sp., and S. epidermidis contamination. Our study was correlated with former study from Phitsanulok night bazaar and Naresuan University flea market which found Micrococcus spp. and *Bacillus spp*. Contamination⁴ while colored contact lenses sold on streets in Bangkok are uncontaminated¹¹. This study also correlated with previous results from female medical students in Shri Sathya Sai Medical College and Research Institute in India which reported S. aureus and Citrobacter koseri were contaminant of contact lenses¹². Last but not least, we were fortunate to find no Pseudomonas sp. contamination in our study. Interestingly, an unidentified foreign body had been observed in this study. The eye's foreign body is any abnormal object or substance that does not belong to the eye. The mechanical effects from foreign bodies may introduce the infection or cause specific reactions to the eye¹³. Ocular reactions from foreign bodies can be classified into 3 types depending on the substance which are the first, inorganic substances e.g. glass or plastic which cause no specific reaction except for mechanical irritation, exudative and fibroblastic isolation of the foreign body¹⁴. Secondly, a chemical reaction may cause a non-specific or occasional specific damage. The third, organic material e.g. vegetable material which causes the formation of granulation tissue with the giant cells¹⁵. Foreign bodies in the CL product may be the source of extraocular foreign bodies which affect the eyelid, sclera, conjunctiva or cornea when applied to this defected CL. CL wearers may feel eye irritation, sharp burning pain, tearing reflex with momentary blindness, blepharospasm which resulted in CL intolerance, CL dissatisfaction and discontinuation⁶. Moreover, the retention of foreign bodies on the ocular surface can introduce the infection, ocular surface damage, cause permanent vision damage and even occasional loss of the eye.

Conclusion

This is the first report that showed 4 isolations of bacterial contamination in cosmetically monthly colored CLs including *Bacillus sp. S. aureus, Citrobacter sp.,* and *S. epidermidis.* In our study we also found a foreign body in one of the vial CL solutions. From present study it can be concluded that the cosmetically monthly colored CLs sold in beauty shops and flea markets without proper care and storage by sellers should be aware. This study implied that reporting channels for complications associated with cosmetically colored CLs to regulatory authorities should also be established and promoted amongst CL sellers, CL wearers, eye care/ health professionals.

Conflict of interest

The authors declare no conflict of interest and declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Acknowledgements

This research is supported by a grant from the College of Medicine and Public Health (2020). In addition, Miss Phairao Sanwang is also gratefully acknowledged.

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