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RESEARCH ARTICLE

ASSESSMENT OF ANTIBIOTICS RESISTANCE AND BIOFILM PRODUCTION AMONG BACTERIAL SPECIES ISOLATED FROM CONTACT LENSES

Eidha Bin-Hameed[1,2,*], Abdul Aziz Bin Beishr[1], Abdul Raheem Shames[1], Fatima Al-Salimy[1], Husam Habtoor[1], Mohammed Habtoor[1], Noha Al-assar[1], and Omar Al-amodi[1]

[1] *Dept. of Health Sciences, Faculty of Medicine and Health Sciences, University of Science and Technology, Aden, Yemen*
[2] *Dept. of Biology, Faculty of Science, Hadhramout University, Mukalla, Hadhramout, Yemen*

***Corresponding author: Eidha Bin-Hameed; E-mail: eidha6@gmail.com**

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Abstract

Contact lenses (CLs) wearing has been increased globally during recent decades, which is one of the main risk factors for developing several ocular infections. Resistant CLs bacterial infections are mainly due to the CLs contamination by bacteria producing biofilm. This study was aimed to assessment of antibiotics resistance and biofilm production among bacterial species isolated from contact lenses in Mukalla city, Hadhramout, Yemen. This cross-sectional study was carried out on 298 participants women during a period from October 2022 to January 2023. The CLs swab samples were collected, then inoculated onto culture media and incubated aerobically at 37°C for 24 hrs. The bacterial isolates were identified by conventional bacteriological methods of cultural characteristics, Gram staining and biochemical test. Antibiotics susceptibility testing was performed by disc diffusion method. Bacterial biofilm production on CLs was detected by tube method (TM) and Congo red agar (CRA) method. The prevalence of CLs bacterial infection was 54.4%. *Enterobacter spp*. 37.1%, followed by *Escherichia coli* 28.4%, *Pseudomonas aeruginosa* 11.7%, *Klebsiella pneumoniae* 6.8% were the most common Gram-negative isolated from CLs. *Staphylococcus epidermides* 3.7% and other coagulase negative staphylococci (CoNS) 12.3% were the most common species of Gram-positive bacteria isolated from CLs. The CRA method was found to be effective phenotypic screening method for detection of biofilm production of bacterial isolates from CLs. Prevalence of antibiotics resistance and multi-drug resistance (MDR) biofilm producing strains was found. In conclusion, there is a high prevalence of CLs use by females in Mukalla city, Hadhramout especially for cosmetic purposes. CLs infection due to improper care practices leads to eye complications. Increasing awareness is crucial to avoid identified risk factors for ocular infection.

Keywords: Contact lenses, Biofilm production, Antibiotics resistance, Bacterial species.

Introduction

Biofilm is a structured bacterial community, enclosed in a self-produced of polymeric matrix and adhered to biotic or abiotic surfaces [1]. Biofilm associated bacteria compared to their planktonic counterparts exhibit greater resistance to antibiotics [2]. This increased antibiotic resistance is mainly due to the limited diffusion of drugs through the matrix of biofilm and to physiological changes in bacteria due to the environmental conditions characterizing the biofilm [3].

Biofilm are the source of persistent infections of many pathogenic microbes. The higher incidence of biofilm-associated infections is contributed the frequent use of artificial implants and medical devices nowadays [4]. Both Gram positive bacteria such as *Staphylococcus aureus, S. epidermidis, Streptococcus pyogene, S. viridans, Enterococcus faecalis,* and Gram-negative bacteria such as *Escherichia coli, Klebsiella pneumonia, Proteus mirabilis* and *Pseudomonas aeruginosa* possess the ability to form biofilm [5]. The increasing trend of antibiotics resistance, along with the capacity of biofilm production on medical devices and tissue may cause the additional antibiotics resistance and fails the treatment [6].

CLs wear is now the most prevalent risk factor for new cases of ocular infection and corneal ulcers. The pathogenic property of microbes of biofilm forming on CLs surfaces plays a crucial role in developing CLs related eye infections [7]. Many reports showed that *P. aeruginosa* and *S. aureus* have been the most common frequently isolated organisms from CLs [8,9].

In the study area, there is insufficient knowledge of factors associated with CLs infection transmission. Also, in our knowledge, this is the first study was carried out to assessment of antibiotics resistance and biofilm production among bacterial species isolated from contact lenses among women in Mukalla city, Hadhramout governorate, Yemen. This research highlights the need to better understand the CLs related bacteria in order to improve the management of CLs bacterial infections. Therefore, the present study aimed to detect the presence of biofilm forming bacterial isolates from CLs and to explore their antibiotics resistance patterns.

Subjects and Methods

Study Design, Area and Population

A cross-sectional study was carried out among women used CLs in Mukalla city, Hadhramout during a period from October 2022 to January 2023.

Samples Collection

A total of 298 CLs samples were collected with sterile cotton swab moistened with sterile normal saline solution [10], then delivered with proper transport media to the medical microbiology department at Faculty of Medicine and Health Science, University of Science and Technology and processed.

Bacterial Culture and Identification

The samples were inoculated onto blood agar, MacConkey agar and nutrient agar (Himedia, India), then incubated at 37°C for 24 hours under aerobic condition. After 24 hours of incubation, each plate was examined, and negative plates were incubated for an additional 24 hours. Identification of bacterial species was obtained via colony specifications, Gram stain reaction and divers biochemical tests following standard methods [11].

Antibiotics Susceptibility Test

Kirby-Bauer disk diffusion method was performed to test each isolate for in vitro antibiotics susceptibility in accordance with the standards of the clinical and laboratory standards institute (CLSI) guidelines [12]. Briefly, the standard bacterial inoculum adjusted to 0.5 McFarland standard turbidity was uniformly distributed over the surface of Mueller Hinton agar (Himedia, India). Antibiotic disks (Himedia, India) including co-trimoxazole (25mcg), amikacin (30µg), cefixime (5mcg), clindamycin (mcg), ceftriaxone (30mcg), levofloxacin (5mcg), cloxacillin (1mcg), netillicin (30mcg), clarithromycin (15mcg), and vancomycin (30mcg) were applied on Mueller Hinton agar plates. Following overnight incubation at 37°C, the zone of inhibition was measured and interpreted as sensitive, intermediate sensitive or resistant per the standard criteria.

Biofilm Detection Methods

Tube Method (TM)

As described by Osungunna and Onawunmi [13], this qualitative method for biofilm detection was carried out as follows: a loopful of tested bacteria was inoculated in 10 ml of tryptone soya broth with 1% glucose in test tubes. Tubes were incubated at 37°C for 24 hours. After incubation, the tubes were decanted and washed with phosphate buffer saline (pH 7.3) and dried. The tubes were stained with crystal violet (0.1%), and excess stain was washed with deionized water. The tubes were dried in an inverted position. Biofilm production was considered positive when a visible film lined the wall and the bottom of the tube. The amount of biofilm formed was scored as weak/none, moderate, and high/strong.

Congo Red Agar (CRA) Method

The simple qualitative CRA method was performed as described by Triveni *et al.* [14] as follows, the CRA medium plates were inoculated with tested bacteria and incubated at 37ºC for 24 hours aerobically. Black colonies on medium indicates positive test for strong biofilm production, grayish black to deep red indicates moderate biofilm producers and red colonies are considered as weak/non biofilm producers.

Data Analysis

Data were analyzed using IBM Statistical Package for Social Sciences (SPSS) software (version 24; IBM SPSS Inc., New York, USA). Descriptive statistics was used to measure the frequencies and percentages. The association between different categories was measured and compared using Pearson Chi-square (χ^2) test. The level of statistical significance was set at P-value < 0.05.

Ethical Consideration Statement

Research ethical approval of this study was obtained from Department of Health Sciences, University of Science and Technology. The samples were taken from the participants after they agreed to it verbally with confidentiality of each participant.

Results

Prevalence of Contact Lenses Infection and Frequencies of Bacterial Species Isolated

In this study, women participants with CLs used leading to a prevalence rate of CLs bacterial infection. The CLs swabbed cultures yielded bacterial growth of 162(54.4%)

and the others 136(45.6%) showed no growth. Among Gram negative bacteria, *Enterobacter spp.* 60(37.1%) were the most common isolated followed by *E. coli* 46(28.4%), *P. aeruginosa* 19(11.7%) and *K. pneumoniae* 11(6.8%). *S. epidermides* 6(3.7%) and other CoNS 20(12.3%) were the only Gram positive bacteria isolated from CLs as given in table (1).

Table (1): Frequencies and percentages of bacterial species isolate from CLs

Bacterial isolates	No.	%
Enterobacter spp.	60	37.1
E. coli	46	28.4
P. aeruginosa	19	11.7
K. pneumoniae	11	6.8
S. epidermides	6	3.7
Other CoNS	20	12.3
Total	**162**	**100**

Biofilm Production Detection Results

Tube Method (TM)

Positive results of biofilm produced by the TM was confirmed by visible thick film obtained inside the wall and the bottom of the tube indicating strong and moderate biofilm production, and the others indicate no biofilm formed with no color, figure (1).

Among Gram positive bacterial species, TM detected biofilm production in 1(16.7%) isolate of *S. epidermides* as strong biofilm producer, and 5(83.3%) identified as weak or non-biofilm producers isolates. Of other CoNS isolates, 2(10%) isolates were strong biofilm producers, 4(20%) of isolates were moderate biofilm producers, and 14(70%) showed weak or non-biofilm producers isolates.

For Gram negative bacteria, *E. coli* isolates showed 4(8.7%) isolates were strong biofilm producers, 17(37.0%) of isolates were moderate biofilm producers, and 25(54.3%) identified as weak or non-biofilm producers isolates. Two (18.2%) of *K. pneumoniae* isolates showed strong biofilm producers, 5(45.5%) of isolates were moderate biofilm producers, and 4(36.4%) were weak or non-biofilm producers. Of *Enterobacter spp.* isolates, 25(41.7%) isolates were strong biofilm producers, 14(23.3%) of isolates were moderate biofilm producers, and 21(35.0%) identified as weak or non-biofilm producers isolates. *P. aeruginosa* showed 3(15.8%) isolates were moderate biofilm producers, and 16(84.2%) identified as weak or non-biofilm producers. There was significant statistical analysis of TM method for screening biofilm production (*P-value* = 0.001) as shown in table (2).

Strong biofilm producer Moderate biofilm producer Weak/non biofilm producer

Fig. (1): Bacterial biofilm production by TM

Table (2): Biofilm production of bacterial isolated from CLs by TM

Bacterial species	No. (%)	Biofilm production by TM No. (%)			χ^2 value	P-value
		S	M	W/N		
S. epidermides	6 (3.7)	1 (16.7)	0 (0.0)	5 (83.3)	37.080	0.001*
Other CoNS	20 (12.3)	2 (10)	4 (20)	14 (70)		
Enterobacter spp.	60 (37.1)	25 (41.7)	14 (23.3)	21 (35.0)		
E. coli	46 (28.4)	4 (8.7)	17 (37.0)	25 (54.3)		
P. aeruginosa	19 (11.7)	0 (0.0)	3 (15.8)	16 (84.2)		
K. pneumonia	11 (6.8)	2 (18.2)	5 (45.5)	4 (36.4)		
Total	**162(100)**	**34(21.0)**	**43(26.5)**	**85(52.5)**		

**P-value* < 0.05 is considered statistically significant

S: Strong, M: moderate, W/N: Weak/None

Congo Red Agar (CRA) Method

Bacterial isolates gave black colonies on CRA indicates positive for strong biofilm production, grayish black to deep red indicates moderate biofilm producers and red colonies were considered as weak/non biofilm producers, figure (2).

CRA method detected biofilm production in 5(83.3%) and 14(70.0%) isolates of *S. epidermides* and other CoNS isolates as moderate biofilm producer respectively. Weak or non-biofilm producers isolates of *S. epidermides* and other CoNS identified as 1(16.7%) and 6(30.0%) isolates respectively of Gram-positive bacterial species.

For Gram negative bacteria, *E. coli* isolates showed 7(15.2%) isolates were strong biofilm producers, 28(60.9%) of isolates were moderate biofilm producers, and 11(23.9%) identified as weak or non-biofilm producers isolates. *P. aeruginosa* showed 1(5.3%) isolate was strong biofilm producers, 3(15.8%) were moderate biofilm producers, and 15(78.9%) identified as weak or non-biofilm producers. *K. pneumoniae* isolates showed 2(18.2%) of isolates were strong and moderate biofilm producers respectively, while 7(63.6%) isolates were weak or non-biofilm producers. Of *Enterobacter spp.* isolates, 12(20.0%) isolates were strong biofilm producers, 37(61.7%) of isolates were moderate biofilm producers, and 11(18.3%) identified as weak or non-biofilm producers isolates. There was significant statistical analysis of CRA method for screening biofilm production (*P-value* = 0.001) as given in table (3).

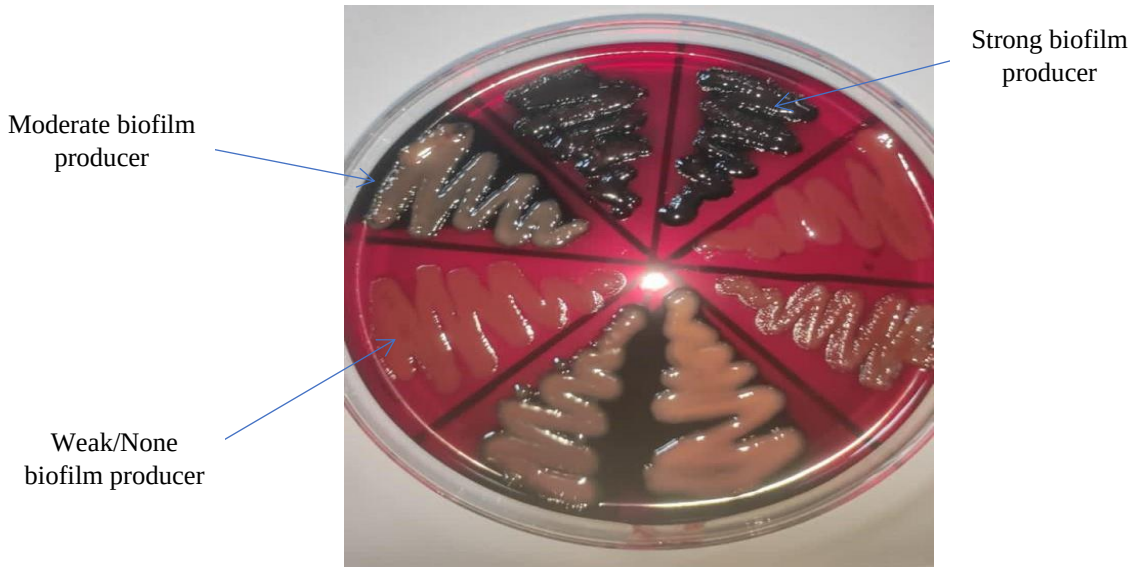

Fig. (2): Bacterial biofilm production by CRA method

Table (3): Biofilm production of bacterial isolated from CLs by CRA method

Bacterial species	No. (%)	Biofilm production by CRA No. (%)			χ^2 value	P-value
		S	M	W/N		
S. epidermides	6 (3.7)	0 (0.0)	5 (83.3)	1 (16.7)	38.857	0.001*
Other CoNS	20 (12.3)	0 (0.0)	14 (70.0)	6 (30.0)		
Enterobacter spp.	60 (37.1)	12 (20.0)	37 (61.7)	11 (18.3)		
E. coli	46 (28.4)	7 (15.2)	28 (60.9)	11 (23.9)		
P. aeruginosa	19 (11.7)	1 (5.3)	3 (15.8)	15 (78.9)		
K. pneumonia	11 (6.8)	2 (18.2)	2 (18.2)	7 (63.6)		
Total	**162(100)**	**22(13.6)**	**89(55.0)**	**51(31.4)**		

**P-value* < 0.05 is considered statistically significant
S: Strong, M: moderate, W/N: Weak/None

Antibiotics Susceptibility Testing Method

The results of the antibiotics susceptibility assay showed the presence of resistant patterns of the isolated bacteria to antibiotics. The highest antibiotics resistance was cloxacillin 149(92.0%), cefixime 148(91.4%), clindamycin 143(88.3%), vancomycin 134(82.7%) and ceftriaxone 108(66.7%). Antibiotics showed high sensitivity were levofloxacin 151(93.2%), neticillin 139(85.8%), amikacin 126(77.8%) and co-trimoxazole 119(73.5%), Table (4).

Table (4): The overall antibiotics susceptibility patterns of bacterial species isolated from CLs infections

Antibiotic	Susceptibility patterns No. (%)		
	Sensitive	Intermediate	Resistance
Vancomycin	23 (14.2)	5 (3.1)	134 (82.7)
Cefixime	12 (7.4)	2 (1.2)	148 (91.4)
Clindamycin	14 (8.6)	5 (3.1)	143 (88.3)
Neticillin	139 (85.8)	0 (0.0)	23 (14.2)
Co-trimoxazole	119 (73.5)	0 (0.0)	43 (26.5)
Cloxacillin	13 (8.0)	0 (0.0)	149 (92.0)
Clarithromycin	47 (29)	44 (27.2)	71 (43.8)
Ceftriaxone	31 (19.1)	23 (14.2)	108 (66.7)
Amikacin	126 (77.8)	17 (10.5)	19 (11.7)
Levofloxacin	151 (93.2)	3 (1.9)	8 (4.9)

Among Gram-positive bacteria, *S. epidermides* and other CoNS were more resistance to cefixime and cloxacillin**,** whereas Gram-negative bacteria showed high resistance of *E. coli*, *P. aeruginosa*, *K. pnumoniae* and *Enterobacter spp.* to vancomycin, cefixime, clindamycin, cloxacillin and ceftriaxone. In this study, all bacterial species isolated from CLs infection showed multidrug resistance (MDR) to three or more types of antibiotics belonging to three or more different antibiotics classes such as clindamycin, cefixime, clarithromycin and cloxacillin.

Relationship of Antibiotics Susceptibility Patterns with Biofilm and Non-Biofilm Producing of Bacterial Species Isolated from Contact Lenses

Among 162 bacterial species isolated from CLs, biofilm producers isolates using TM showed high resistance rates to antibiotics used compared to non-biofilm producers isolates. Bacterial species biofilm producing isolates found highly resistant to clindamycin 73(45.1%), vancomycin 70(43.2%), ceftriaxone 45(27.8%), clarithromycin 27(16.7%), neticillin 18(11.1%), amikacin 11(6.8%) and levofloxacin 7(4.3%). There was significant statistical correlation of antibiotic resistance of vancomycin, clindamycin, neticillin, ceftriaxone and levofloxacin with bacterial biofilm production (*P-value* < 0.05). Also, biofilm producers of bacterial isolates using CRA method showed high resistance rates to antibiotics used compared to non-biofilm producers isolates. Bacterial species biofilm producing isolates found highly resistant to cloxacillin 99(61.1%), cefixime 97(59.9%), clindamycin 97(59.9%), vancomycin 90(55.6%), ceftriaxone 68(42.0%) clarithromycin 42(25.9%), co-trimoxazole 22(13.6%), neticillin 18(11.1%) and amikacin 13(8.0%). There was significant statistical correlation of antibiotic resistance of cefixime, co-trimoxazole, clarithromycin and ceftriaxone with bacterial biofilm production (*P-value* < 0.05), as shown in table (5).

Table (5): Antibiotics susceptibility test results of biofilm and non-biofilm producing bacterial isolates by TM and Congo red method

Antibiotic	Pattern	Biofilm tube method				Biofilm Congo red method			
		Producer	Non-producer	χ^2 value	P-value	Producer	Non-producer	χ^2 value	P-value
Vancomycin	S	3	20	14.274	0.001*	17	6	0.730	0.694
	I	4	1			4	1		
	R	70	64			90	44		
Cefixime	S	7	5	2.918	0.232	12	0	7.041	0.030*
	I	2	0			2	0		
	R	68	80			97	51		
Clindamycin	S	3	11	6.054	0.048*	12	2	3.836	0.147
	I	1	4			2	3		
	R	73	70			97	46		
Neticillin	S	59	80	10.150	0.001*	93	46	1.179	0.277
	I	0	0			-	-		
	R	18	5			18	5		
Co-trimoxazole	S	57	62	0.024	0.876	89	30	8.174	0.004*
	I	0	0			-	-		
	R	20	23			22	21		
Cloxacillin	S	6	7	0.011	0.917	12	1	3.708	0.054
	I	0	0			-	-		
	R	71	78			99	50		
Clarithromycin	S	19	28	4.754	0.093	33	14	6.556	0.038*

	I	27	17			36	8		
	R	31	40			42	29		
Ceftriaxone	S	21	10	6.568	0.037*	22	9	7.167	0.028*
	I	11	12			21	2		
	R	45	63			68	40		
Amikacin	S	61	65	3.096	0.213	86	40	0.038	0.981
	I	5	12			12	5		
	R	11	8			13	6		
Levofloxacin	S	70	81	7.926	0.019*	105	46	5.019	0.081
	I	0	3			3	0		
	R	7	1			3	5		

**P-value* < 0.05 is considered statistically significant
Key: (S) Sensitive, (I) Intermediate sensitive, (R) Resistant

Table (6): Relationship of biofilm production by TM and MDR

Bacterial biofilm	MDR bacteria									
	Tube method					Congo red method				
	Yes	No	Total	χ^2 value	*P-value*	Yes	No	Total	χ^2 value	*P-value*
Producer	68	9	77	1.137	0.286	90	21	111	4.706	0.030*
Non-producer	70	15	85			48	3	51		
Total	138	24	162			138	24	162		

**P-value* < 0.05 is considered statistically significant

Relationship the Biofilm Production with Bacterial Multidrug Resistance

Among 77 biofilm producers of bacterial species isolated from CLs infection by TM, 68(88.3%) isolates were MDR, and 70(82.4%) out of 85 non-producers were also MDR. Statistically, there was no significant association between biofilm production by TM and MDR bacterial isolates (*P-value* = 0.006). By CRA method, 90(81.1%) isolates out of 111 bacterial species biofilm producers isolated from CLs infection were MDR. Among 51 non-biofilm producers, 48(94.1%) were MDR. Statistically, there was significant association between biofilm production and MDR isolates (*P-value* = 0.030), table (6).

Discussion

Despite the large number of young adults wearing CLs in Yemen, there is a lack of comprehensive data about the prevalence of CLs wear or about the knowledge of proper lens used and care. So, there is no previous studies has investigated the practice of dispensing CLs and study the risk factors associated with infections in Yemen.

CLs are widely distributed among young adults for reasons such as cosmetic or therapeutic. The current study revealed that high number of females in Mukalla city wore CLs for cosmetic. The prevalence of CLs use observed in some population-based studies can be even greater in number; for example, a study among medical students from Saudi Arabia indicated 40.5% of the students wore CLs [15]. Other study showed more than half of the students of Umm Al-Qura University students in Makkah, Saudi Arabia experienced eye complications such as ocular complaints, allergic reaction, dry eyes, corneal abrasions and corneal ulcer due to improper care of CLs [16]. There is a high prevalence 70.2% of CLs use by female university students in Saudi Arabia, especially for cosmetic purposes [17]. Infective keratitis secondary to soft lens wear was the most common complication, followed by epithelial keratitis and allergic conjunctivitis seen in public hospitals in Singapore [18].

In this study, the prevalence rate of CLs bacterial infection was 54.4%. *Enterobacter spp.* 37.1%, *E. coli* 28.4%, *P. aeruginosa* 11.7%, *K. pneumoniae* 6.8%, *S. epidermides* 3.7% and other CoNS 12.3% were the common isolates. Previous reports showed that *P. aeruginosa* and *S. aureus* were the most common frequently isolated organism [8,9,19]. In one study, *Pseudomonas* account for 24% of organisms related to CLs induced ulcer [9]. A study conducted in Iran revealed *P. aeruginosa*, *Staphylococcus spp.*, and *Serratia marcescens* were the three most common bacteria isolated from samples of patients with CLs-related bacterial keratitis. Overall, isolated bacteria were most sensitive to fluoroquinolones and aminoglycosides, especially ciprofloxacin and gentamicin respectively, and most resistant against penicillin and cephalosporins especially cefazolin and chloramphenicol [20].

A study carried out in Australia revealed the most common risk factor for keratitis was CLs wear, and the most commonly isolated organism was *P. aeruginosa*

[21]. Another study showed differences in the virulence factors of *P. aeruginosa* isolated from CLs and non-CLs-related keratitis and a strong biofilm production phenotype was found in some strains [22].

With the widespread use of CLs in in Saudi Arabia, corneal ulcer associated with CLs wear became more prevalent, and the study identified increasing awareness is crucial to avoid risk factors for corneal ulcer [16]. Other study revealed that CLs wearers are at high risk of getting ocular infections because of the contamination of CLs storage cases and CLs solution by microbes. The bacteria isolated from CLs includes *Corynebacterium spp.*, *S. aureus*, CoNS, *Bacillus spp.* and *Streptococcus spp*. Biofilm are the main cause of bacterial infection. The biofilm forming ability of these bacterial isolates is studied by CRA method and tube method. Best results were observed by TM [10].

The present study showed that 21.0% of bacterial isolated from CLs were strong biofilm producers, 26.5% were moderate and 52.5% were weak/none producers by TM. The biofilm producers showed in isolates by CRA method 13.6% as strong, 55.0% moderate and 31.4% weak/none producers. Other study observed 265 bacterial species isolated from CLs wearers included *S. aureus*, CoNS, *Pseudomonas*, non-fermenter Gram-negative bacilli, *Bacillus spp.*, Diphtheroids, Micrococci, *K. pneumonia*, *Klebsiella oxytoca*, *E. coli*, *Proteus mirabilis*, *Proteus vulgaris*, *Citrobacter koseri*, *Citrobacter freundii*, *Enterobacter cloacae*, *Moraxella* were moderately positive 53.5%, strongly positive 33.2% and negative 13.2% for biofilm production by TM, and 36.6% were moderately positive, 40% strongly positive and 23.3% negative for biofilm production by CRA method. TM and CRA exhibited significant statistical correlation and picked up a good number of biofilms-forming isolates, and hence may be used for detection of biofilm production [23].

A study showed of 32 MRSA isolates, 34.37%, 59.37%, and 81.25% showed positive results using CRA, TM or micro titer plate, respectively. Biofilm production was found to be reduced in the presence of ethanol or ethylenediaminetetraacetic acid (EDTA) and at extreme pH values [24]. Other study showed growth of biofilms on type of hard and soft lenses and lens cases of the organisms *S. aureus*, *P. aeruginosa* and *E. coli* [25].

Other study showed a lack of hygiene and improper care of CLs can predispose to the colonization the CLs surface with bacteria leading to biofilms production, especially with *P. aeruginosa* [19]. Culture-independent methods identified an association between disease severity and bacterial diversity in biofilms isolated from cases and lenses of patients with CLs-related corneal disease [26]. Other study showed *P. aeruginosa* adhered in higher numbers compared to *S. aureus* on CLs solutions [27].

Most of the ocular pathogens showed in vitro variation in adhesion between species and strains. *P. aeruginosa* can adhere to CLs the most of any bacteria test thus far, and this may be a reason it is the most predominant microorganism that causes CLs-associated MK [28]. Some soft CLs solutions may facilitate bacterial biofilm production and adhesion capability of *S. aureus* and *P. aeruginosa* [29]. A study revealed that *P. aeruginosa*, *S. marcescens*, and *S. aureus* form biofilms on CLs were resistant to the antimicrobial activity of five common multipurpose CLs care solutions and one hydrogen peroxide care solution [30].

The effective antibiotic therapy could be provided by antibiotic eye drops, the best choice is preventing the CLs related eye infections with efficient disinfection of CLs by multipurpose CLs solutions. The anti-biofilm activities of multipurpose were based on various factors, such as chemical ingredients and contact time of multipurpose, the type of infectious agent, and especially the CLs type and usage time [31]. So, the size of initial inoculum, nutritional content of media, and incubation period played significant roles in bacterial adhesion to CLs. Adhesion is more affected by the environment and numbers of bacteria initially applied to lenses [27].

Among 162 bacterial species isolated from CLs in the current study, biofilm producers isolates using TM showed high resistance rates to antibiotics clindamycin 45.1%, vancomycin 43.2%, ceftriaxone 27.8%, clarithromycin 16.7%, neticillin 11.1%, amikacin 6.8% and levofloxacin 4.3% compared to non-biofilm producers isolates. Also, biofilm producers of bacterial isolates using CRA method showed high resistance rates to antibiotics cloxacillin 61.1%, cefixime 59.9%, clindamycin 59.9%, vancomycin 55.6%, ceftriaxone 42.0% clarithromycin 25.9%, co-trimoxazole 13.6%, neticillin 11.1% and amikacin 8.0%. Another study showed antibiotic resistant strains of *P. aeruginosa* and *S. aureus* isolated from CLs [32].

One of the most important characteristics of biofilms is their increased tolerance to antimicrobial agents. It has been proved that biofilms can tolerate up to 100–1000 times higher concentrations of antibiotics and disinfectants than planktonic cells [6].

In the present study, among 77 biofilm producers of bacterial species isolated from CLs infection by TM, 88.3% isolates were MDR, and 82.4% out of 85 non-producers were also MDR. By CRA method, 81.1% isolates out of 111 bacterial species biofilm producers isolated from CLs infection were MDR. Among 51 non-biofilm producers, 94.1% were MDR. Other study showed 43.6% were non-biofilm producers while 56.4% produced biofilms. All biofilm producing isolates were sensitive to a limited spectrum of antibiotic classes. All moderate and strong biofilm producers and 81% of weak biofilm producers were MDR. In contrast, among non-

biofilm producers, only 11.8% were classified as MDR strains [33].

Conclusions

CLs wearers are at high risk of getting ocular infections because of the contamination of CLs, CLs storage cases and CLs solution contaminated by microbes. The worrying prevalence of antibiotics resistance and MDR biofilm producing strains represents a serious challenge to clinicians in the treatment and care of ocular infections. So, people should be educated for proper eye care to avoid chances of getting infection, and increasing awareness is crucial to avoid identified risk factors for ocular infections. New strategies for bacterial biofilm prevention and control will help in prevention of different diseases.

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Conflict of Interest

No conflict of interest associated with this work.

Author's Contribution

The manuscript was prepared, written and approved in collaboration with all authors.

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مقالة بحثية

تقييم المقاومة للمضادات الحيوية وإنتاج الغشاء الحيوي للأنواع البكتيرية المعزولة من العدسات اللاصقة

عيظه بن حميد[1،2،*]، عبدالعزيز بن بشر[1]، عبدالرحيم شامس[1]، فاطمة السالمي[1]، حسام حبتور[1]، محمد حبتور[1]، نهى العصار[1]، و عمر العمودي[1]

[1] قسم العلوم الصحية، كلية الطب والعلوم الصحية، جامعة العلوم والتكنولوجيا، عدن، اليمن
[2] قسم علوم الحياة، كلية العلوم، جامعة حضرموت، المكلا، حضرموت، اليمن

*** الباحث الممثّل: عيظه بن حميد؛ البريد الالكتروني: eidha6@gmail.com**

استلم في: 19 يونيو 2023 / قبل في: 26 يونيو 2023 / نشر في 30 يونيو 2023

المُلخّص

ازداد ارتداء العدسات اللاصقة عالمياً خلال العقود الأخيرة، وهو أحد عوامل الخطورة الرئيسية لتطور الإصابة بالعديد من عدوى العين. وتُعزى مقاومة عدوى العدسات اللاصقة البكتيرية بشكل أساسي إلى تلوث العدسات بالبكتيريا المنتجة للغشاء الحيوي. هدفت هذه الدراسة إلى تقييم المقاومة للمضادات الحيوية وإنتاج الغشاء الحيوي للأنواع البكتيرية المعزولة من العدسات اللاصقة في مدينة المكلا، حضرموت، اليمن. أجريت هذه الدراسة المقطعية على 298 امرأة مشاركة خلال الفترة من أكتوبر 2022 إلى يناير 2023. جمعت عينات مسحات العدسات اللاصقة ثم حقنت إلى الأوساط الزراعية وحضنت هوائياً عند درجة حرارة 37 درجة مئوية لمدة 24 ساعة. تم التعرف على العزلات البكتيرية بالطرق البكتريولوجية التقليدية للخصائص المزرعية وصبغة جرام والاختبارات الكيموحيوية. تم إجراء اختبار الحساسية للمضادات الحيوية بطريقة الانتشار من الأقراص. كما تم الكشف عن انتاج البكتيريا للغشاء الحيوي على العدسات اللاصقة بطريقة الأنبوب وطريقة أجار صبغة الكونغو الحمراء. بلغ معدل انتشار العدوى البكتيرية للعدسات اللاصقة 54.4%. مثلت بكتيريا الامعائيات 37.1%، تليها الايشريشية القولونية 28.4%، والزائفة الزنجارية 11.7%، والكلبسيلا الرئوية 6.8% الأكثر شيوعاً للبكتيريا السالبة لصبغة جرام المعزولة من العدسات اللاصقة. وكانت بكتيريا المكورات العنقودية البشروية 3.7% والمكورات العنقودية السالبة لإنزيم التلزن الأخرى 12.3% أكثر الأنواع شيوعاً للبكتيريا موجبة الجرام. وُجِد أن طريقة أجار صبغة الكونغو الحمراء كانت فعالة للكشف عن التنميط الظاهري لتكوين الغشاء الحيوي للبكتيريا المعزولة من العدسات اللاصقة. كما وُجَد انتشار مقاومة ومقاومة متعددة للسلالات البكتيرية المنتجة للغشاء الحيوي للمضادات الحيوية. يستنتج من الدراسة أن هناك انتشار كبير لاستخدام العدسات اللاصقة من قبل النساء في مدينة المكلا، حضرموت خاصة للأغراض التجميلية. تؤدي عدوى العدسات اللاصقة بسبب ممارسات العناية غير المناسبة إلى مضاعفات بالعين. لذا فإن زيادة الوعي يُعد أمراً بالغ الأهمية لتجنب عوامل الخطورة بعدوى العين.

الكلمات المفتاحية: العدسات اللاصقة، انتاج الغشاء الحيوي، المقاومة للمضادات الحيوية، النوع البكتيري.

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