



# Microbial Contamination of Soft Contact Lenses Among Medical School Students in Southern Iraq

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**Background:** Contamination of CLs and accessories risks potentially devastating microbial keratitis. We aimed to explore the rate of microbial contamination and predisposing factors among a group of asymptomatic young medical students.

**Methodology:** The study included 115 healthy female medical students with a mean age of  $21.64 \pm 2.02$  years between January and November 2021. Information about CL use, wear and care was gathered. Each participant's CL and case were swabbed for microbiological identification. Univariable and multivariable analyses were conducted to look for associations with a range of factors.

**Results:** Overall, 91 participants (79.13%) had at least one contaminated sample (lens and/or case). The rates of contamination of CL and their cases were 66% and 76.5%, respectively. Higher contamination rates were caused by gram-negative bacteria (60% of the contaminated samples) with *P. aeruginosa* being the most common contaminant both for CL and cases, whereas *S. epidermidis* and *S. aureus* were the most common contaminants for the CL and cases, respectively, regarding gram-positive contamination. Multivariable analysis showed younger age as a significant predictor of pseudomonas contamination of the lenses and cases (OR: 12.302, 6.555 for CL & cases, respectively;  $P = 0.001$  for both). Older age was a significant predictor of *K. pneumoniae* contamination (OR: 4.154,  $P = 0.007$ ). Pseudomonas contamination of both lenses and cases was predicted by the type of solution used (OR: 10.8 and 13.5,  $P = 0.001$  and 0.003 for bottled and distilled water vs commercially available solutions for CL; OR: 4.5 and 5.8,  $P = 0.045$  and 0.004, respectively, for cases). Pseudomonas case contamination was associated with low frequency of solution change.

**Conclusion:** Microbial contamination rate of soft CL and their cases is high among young medical students in comparison to previously reported rates and was predisposed by several poor hygienic practices and wearing regimens.

**Keywords:** soft contact lens, contact lens case, microbial contamination, lens hygiene, lens wearing schedule, medical students

## Introduction

The advances in contact lens and lens-care solutions, materials and design have contributed to an increase in contact lens wear, for refractive and aesthetic reasons, to about 140 million users worldwide.<sup>1</sup> Nevertheless, CL wear is associated with a variety of potential complications including bacterial and fungal keratitis, acanthamoeba keratitis, papillary conjunctivitis, superior limbic keratoconjunctivitis, neurotrophic corneal ulceration and others.<sup>2-4</sup>

Contact lens-related bacterial keratitis has become a major cause of all new cases of bacterial keratitis, particularly in high socioeconomic populations such as the United Kingdom (65% of all new cases) and Japan (54.5%).<sup>5-7</sup>

Bacteria responsible for bacterial keratitis have been isolated from the surface of the contact lens or the contact lens-care solution.<sup>8-10</sup>

The contact lens disrupts the protective mechanism of the mucin layer of the tear film, hindering its antimicrobial activity on the corneal surface.<sup>11</sup>

Past studies have highlighted the rate of CL contamination and the variation of corneal pathogens with ambient temperature and humidity.<sup>12-14</sup> Hot and humid weather such as in Southern Iraq, where the present study was conducted, as well as lens care regimens and practices imposed by our study population may contribute to variation in CL bacterial contamination.

The aim of the current study was to evaluate the rate of contamination and to determine the types of microorganism in soft contact lenses and their cases among medical school students, and to analyze associations between these and the wearer's age, the type of contact lens, wearing schedule, frequency of change of CL, duration of wear, handling and cleaning processes, the type of CL solution, and its changing interval.

## Methodology

### Study Population

The study included 115 healthy female soft contact lens wearers attending medical colleges at the University of Basra between January and November 2021. The study followed the principles of the Declaration of Helsinki and was approved by the institutional review board at the University of Basrah (Ethical Standards and Professional Conduct Authority). Written informed consent was obtained from each participant before commencement of the study. Each participant was interviewed to complete a questionnaire related to their contact lens usage. Information involved personal data such as age, the type of contact lens, wearing schedule, frequency of change of CL, duration of wear, handling and cleaning processes, and the type of CL solution. Participants were excluded from the study if there were unwilling to participate or wearing daily disposable lenses (which do not require lens case) other types of CL (other than soft) or swimming with contact lens in or showing symptoms of ocular surface infection such as discharge and redness. The subjects were all females since no male subject had been encountered wearing soft contact lens during the study period.

### Microbiological Technique

The contact lens was gently removed from one eye of each participant using sterile blunt tipped forceps by an experienced staff member at the same time the questionnaire was filled. In accordance with sterile aseptic technique, the concave side of the lens was swabbed using sterile cotton tipped swabs moistened with sterile 0.9% sodium chloride solution.

The lens case was also swabbed, and each was labelled, stored separately and transferred to the laboratory in less than 30 minutes to be incubated in a brain heart infusion broth at 37°C for 24–48 hours. Following this, 0.1 mL of the broth was inoculated into each of blood, MacConkey, chocolate and Sabouraud dextrose agar plates and incubated for 24–48 hours for bacterial isolation and for 2 weeks for fungi. A Vitek® system (bioMerieux Vitek, Hazelwood, Mo, USA) was used to isolate and identify bacterial colonies.

### Statistical Analysis

All data entry and subsequent statistical tests were performed using SPSS v26 (SPSS Inc., Chicago, Illinois, USA). Continuous variables were expressed as mean  $\pm$  standard deviation and categorical variables as frequencies and percentages. All percentages were calculated from the total number of participants. Crude association was tested using the chi-squared test or Fisher's exact test according to statistical assumptions. Predictors that were significant on univariable analysis were subsequently tested for association using binomial logistic regression analysis to calculate the adjusted odds ratio (OR). Significance was considered at P value less than 0.05.

## Results

The total sample size was 115 female subjects, with mean age of  $21.64 \pm 2.02$  years (range 18–26 years; median 22 years). [Table 1](#) summarizes the age and CL wear characteristics of the study population. Overall, 91 participants (79.13%) had at least one contaminated sample (lens and/or case). Of these, 76 (66.08%) had a contaminated CL and 88 (76.52%) had a contaminated lens case, with 73 participants (63.5%) demonstrating contamination of both CL and case ([Table 2](#)).

The bacteria *S. epidermidis* and *S. aureus* accounted for the highest rates of gram-positive contamination of CL (22 contaminated samples) and cases (15 contaminated samples), respectively ([Figure 1](#)). The highest rate of gram-negative infection was due to *P. aeruginosa*, for both lenses and cases where 27 and 39 samples were contaminated, respectively ([Figure 2](#)).

**Table 1** Demographic and Contact Lens (CL) Wear Characteristics of the Study Population

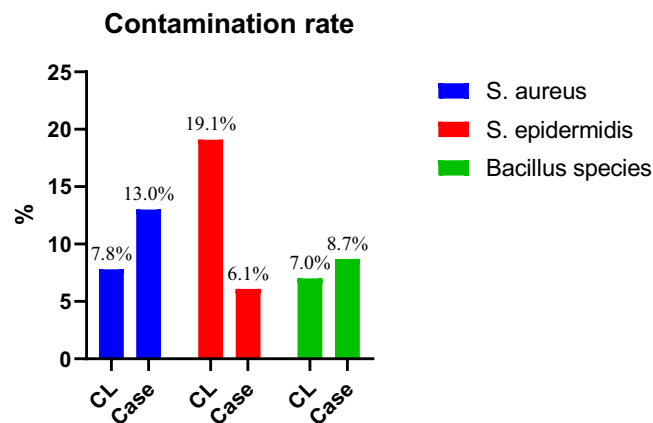
Variables		Number (%)
Age group (years)	18–22	69 (60)
	23–26	46 (40)
CL change interval	≤1 month	59 (51.3)
	>1 month	56 (48.7)
CL time (hours/day)	<6	54 (47)
	≥6	61 (53)
Type of solution	Commercially Available	64 (55.7)
	Bottled water	17 (14.8)
	Distilled water	34 (29.6)
Solution change frequency	Daily	19 (16.5)
	2–7 days	78 (67.8)
	>7 days	18 (15.7)

**Table 2** Contact Lens (CL) and Case Contamination Rates

		CL Case Contamination		Total
		Negative N (%)	Positive N (%)	
CL contamination	-ve N (%)	24 (20.0)	15 (13.0)	39
	+ve N (%)	3 (2.6)	73 (63.46)	76
Total		27	88	115

**Abbreviation:** N, number of contaminated samples.

More samples were contaminated by gram-negative (n = 69, 60%) than gram-positive bacteria (n = 45, 39%). The causative agent could not be identified in only seven (6.1%) samples, and 29 (25.2%) showed mixed contaminations, as shown by Figure 3.



**Figure 1** Gram-positive bacterial contamination rates.

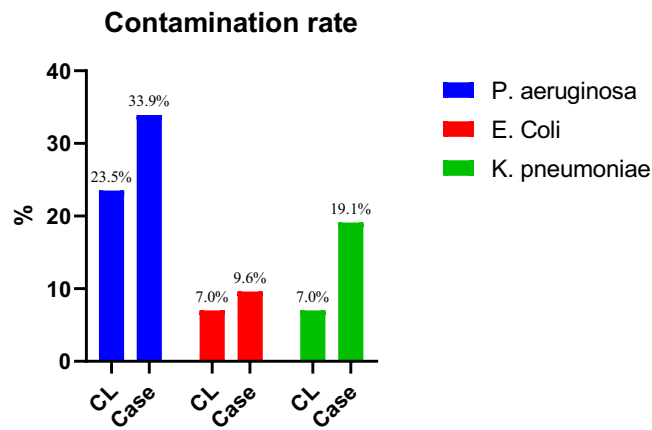


Figure 2 Gram-negative bacterial contamination rates.

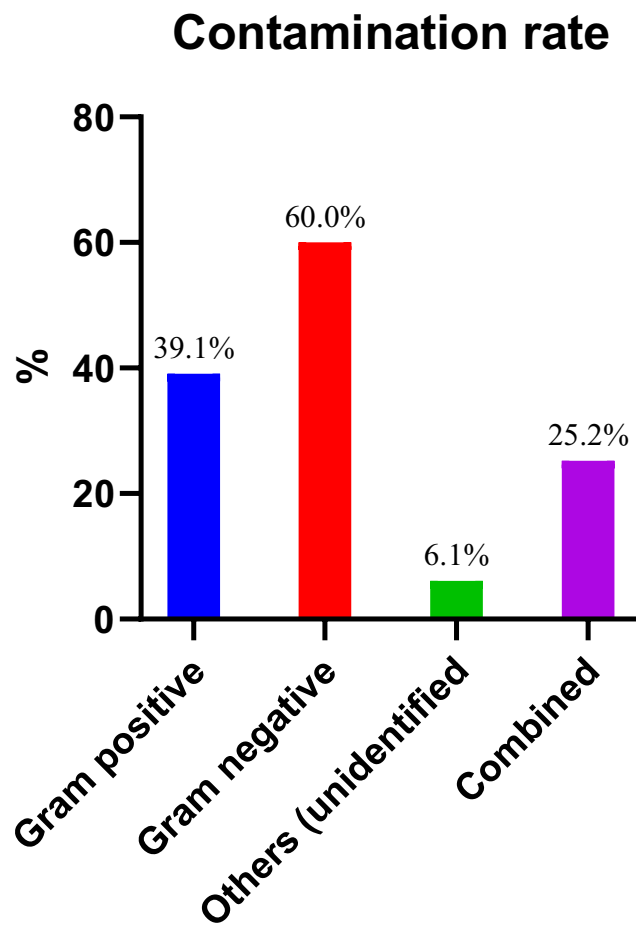


Figure 3 Distribution of contamination according to gram-stain.

Univariable analysis (Table 3) showed significant associations between *S. aureus* CL contamination and both CL change interval exceeding one month, and CL wear exceeding six hours per day ( $P = 0.015$  and  $0.035$  respectively); however, no such association was found for case contamination. *Staphylococcus epidermidis* CL contamination was significantly negatively associated with solution change frequency ( $P = 0.016$ ), but again no such association was found for case contamination.

**Table 3** Univariable Analysis of Gram-Positive Contamination

Type of Microorganism	Predictors Analyzed		Contact Lens Contamination N (%)		P value	Lens Case Contamination N (%)		P value
			Negative	Positive		Negative	Positive	
Staphylococcus aureus	Age group (years)	18–22	62 (58.5)	7 (77.8)	0.132	62 (62)	7 (46.7)	0.273
		23–26	44 (41.5)	2 (22.2)		38 (38)	8 (53.3)	
	CL change interval	≤1 month	<b>58 (54.7)</b>	<b>1 (11.1)</b>	<b>0.015</b>	55 (55)	4 (26.7)	0.053
		>1 month	<b>48 (45.3)</b>	<b>8 (88.9)</b>		45 (45)	11 (73.3)	
	CL wear duration (hours/day)	<6	<b>53 (50)</b>	<b>1 (11.1)</b>	<b>0.035</b>	49 (49)	5 (33.3)	0.257
		≥6	<b>53 (50)</b>	<b>8 (88.9)</b>		51 (51)	10 (66.7)	
	Type of solution	Commercially available	60 (56.6)	4 (44.4)	0.729	57 (57)	7 (46.7)	0.378
		Bottled water	15 (14.2)	2 (22.2)		13 (13)	4 (26.7)	
		Distilled water	31 (29.2)	3 (33.3)		30 (30)	4 (26.7)	
	Solution change frequency	Daily	19 (17.9)	0 (0)	0.363	19 (19)	0 (0)	0.149
		2–7 days	71 (67)	7 (77.8)		65 (65)	13 (86.7)	
		>7 days	16 (15.1)	2 (22.2)		16 (16)	2 (13.3)	
Staphylococcus epidermidis	Age group	18–22 years	58 (62.4)	11 (50)	0.278	64 (59.3)	5 (71.4)	0.70
		23–26 years	35 (37.6)	11 (50)		44 (40.7)	2 (28.6)	
	CL change interval	≤1 month	48 (51.6)	11 (50)	0.892	54 (50)	5 (71.4)	0.442
		>1 month	45 (48.4)	11 (50)		54 (50)	2 (28.6)	
	CL wear duration (hours/day)	<6	42 (45.2)	12 (54.5)	0.482	50 (46.3)	4 (57.1)	0.705
		≥6	51 (54.8)	10 (45.5)		58 (53.7)	3 (42.9)	
	Type of solution	Commercially Available	51 (54.8)	13 (59.1)	0.973	59 (54.6)	5 (71.4)	0.135
		Bottled water	14 (15.1)	3 (13.6)		15 (13.9)	2 (28.6)	
		Distilled water	28 (30.1)	6 (27.3)		34 (31.5)	0 (0)	
	Solution change frequency	Daily	<b>19 (20.4)</b>	<b>0 (0)</b>	<b>0.016</b>	18 (16.7)	1 (14.3)	1.0
		2–7 days	<b>62 (66.7)</b>	<b>16 (72.7)</b>		73 (67.6)	5 (71.4)	
		>7 days	<b>12 (12.9)</b>	<b>6 (27.3)</b>		17 (15.7)	1 (14.3)	
Bacillus spp.	Age group (years)	18–22	67 (62.6)	2 (25)	0.086	67 (62.6)	2 (25)	0.086
		23–26	40 (37.4)	6 (75)		40 (37.4)	6 (75)	
	CL change interval	≤1 month	54 (50.5)	5 (62.5)	0.717	53 (50.5)	6 (60)	0.743
		>1 month	53 (49.5)	3 (37.5)		52 (49.5)	4 (40)	
	CL duration (hours/day)	<6	51 (47.7)	3 (37.5)	0.721	49 (46.7)	5 (50)	1.0
		≥6	56 (52.3)	5 (62.5)		56 (53.3)	5 (50)	

(Continued)

**Table 3** (Continued).

Type of Microorganism	Predictors Analyzed		Contact Lens Contamination N (%)		P value	Lens Case Contamination N (%)		P value
			Negative	Positive		Negative	Positive	
	Type of solution	Commercially Available	58 (54.2)	6 (75)	0.363	56 (53.3)	8 (80)	0.240
		Bottled water	17 (15.9)	0 (0)		17 (16.2)	0 (0)	
		Distilled water	32 (29.9)	2 (25)		32 (30.5)	2 (20)	
	Solution change frequency	Daily	19 (17.8)	0 (0)	0.227	18 (17.1)	1 (10)	0.891
		2–7 days	70 (65.4)	8 (100)		70 (66.7)	8 (80)	
		>7 days	18 (16.8)	0 (0)		17 (16.2)	1 (10)	

**Note:** Bold text indicates significant results.

**Abbreviations:** N, number of contaminated samples; CL, contact lens.

Univariable analysis of gram-negative contamination (Tables 4 and 5) showed that *P. aeruginosa* contamination of both CL and case was significantly associated with younger age, longer interval (exceeding one month) between changes of CL, type of solution and its change frequency. *E. coli* contamination of CL was associated only with CL change intervals exceeding one month ( $P = 0.029$ ), while *K. pneumoniae* contamination of the case was associated only with older age ( $P = 0.003$ ).

**Table 4** Predictors of *Pseudomonas Aeruginosa* Contamination of Contact Lenses and Cases on Univariable Analysis

Predictors		Lens Contamination		P value	Case Contamination		P value
		Negative N (%)	Positive N (%)		Negative N (%)	Positive N (%)	
Age groups (years)	18–22	45 (51.1)	24 (88.9)	<b>&lt;0.001</b>	37 (48.7)	32 (82.1)	<b>&lt;0.001</b>
	23–26	43 (48.9)	3 (11.1)		39 (51.3)	7 (17.9)	
CL change interval	Less than 1 month	53 (60.2)	6 (22.2)	<b>&lt;0.001</b>	51 (67.1)	8 (20.5)	<b>&lt;0.001</b>
	More than 1 month	35 (39.8)	21 (77.8)		25 (32.9)	31 (79.5)	
CL duration (hours/day)	≤6	45 (51.1)	9 (33.3)	0.105	40 (52.6)	14 (35.9)	0.089
	>6	43 (48.9)	18 (66.7)		36 (47.4)	25 (64.1)	
Type of solution	Commercially Available	59 (67)	5 (18.5)	<b>&lt;0.001</b>	53 (69.7)	11 (28.2)	<b>&lt;0.001</b>
	Bottled water	10 (11.4)	7 (25.9)		9 (11.8)	8 (20.5)	
	Distilled water	19 (21.6)	15 (55.6)		14 (18.4)	20 (51.3)	
Solution change frequency	Daily	18 (20.5)	1 (3.7)	<b>0.005</b>	18 (23.7)	1 (2.6)	<b>&lt;0.001</b>
	2–7 days	61 (69.3)	17 (63)		53 (69.7)	25 (64.1)	
	>7 days	9 (10.2)	9 (33.3)		5 (6.6)	13 (33.3)	

**Note:** Bold text indicates significant results.

**Abbreviations:** N, number of contaminated samples; CL, contact lens.

**Table 5** Univariable Analysis of Other Gram-Negative Bacterial Contamination

Type of Microorganism	Predictors		Lens Contamination		P value	Case Contamination		P value
			Negative N (%)	Positive N (%)		Negative N (%)	Positive N (%)	
E. coli	Age group (years)	18–22	63 (58.9)	6 (75)	0.473	62 (59.6)	7 (63.6)	1.0
		23–26	44 (41.1)	2 (25)		42 (40.4)	4 (36.4)	
	CL change interval	Within a month	58 (54.2)	1 (12.5)	<b>0.029</b>	52 (50)	7 (63.6)	0.390
		More than a month	49 (45.8)	7 (87.5)		52 (50)	4 (36.4)	
	CL duration (hours/day)	≤6	52 (48.6)	2 (25)	0.279	50 (48.1)	4 (36.4)	0.459
		>6	55 (51.4)	6 (75)		54 (51.9)	7 (63.6)	
	Type of solution	Commercially Available	61 (57)	3 (37.5)	0.405	58 (55.8)	6 (54.5)	0.911
		Bottled water	15 (14)	2 (25)		15 (14.4)	2 (18.2)	
		Distilled water	31 (29)	3 (37.5)		31 (29.8)	3 (27.3)	
	Solution change frequency	Daily	19 (17.8)	0 (0)	0.115	19 (18.3)	0 (0)	0.243
		2–7 days	73 (68.2)	5 (62.5)		68 (65.4)	10 (90.9)	
		>7 days	15 (14)	3 (37.5)		17 (16.3)	1 (9.1)	
Klebsiella pneumoniae	Age group (years)	18–22	67 (62.6)	2 (25)	0.058	62 (66.7)	7 (31.8)	<b>0.003</b>
		23–26	40 (37.4)	6 (75)		31 (33.3)	15 (68.2)	
	CL change interval	Within a month	54 (50.5)	5 (62.5)	0.717	44 (47.3)	15 (68.2)	0.078
		More than a month	53 (49.5)	3 (37.5)		49 (52.7)	7 (31.8)	
	CL duration (hours/day)	≤6	48 (44.9)	6 (75)	0.145	41 (44.1)	13 (59.1)	0.205
		>6	59 (55.1)	2 (25)		52 (55.9)	9 (40.9)	
	Type of solution	Commercially Available	58 (54.2)	6 (75)	0.609	48 (51.6)	16 (72.7)	0.201
		Bottled water	16 (15)	1 (12.5)		15 (16.1)	2 (9.1)	
		Distilled water	33 (30.8)	1 (12.5)		30 (32.3)	4 (18.2)	
	Solution change frequency	Daily	18 (16.8)	1 (12.5)	0.638	15 (16.1)	4 (18.2)	0.280
		2–7 days	71 (66.4)	7 (87.5)		61 (65.6)	17 (77.3)	
		>7 days	18 (16.8)	0 (0)		17 (18.3)	1 (4.5)	

**Note:** Bold text indicates significant results.

**Abbreviations:** N, number of contaminated samples; CL, contact lens.

On multivariable analysis (Table 6), younger age was a significant predictor of *P. aeruginosa* contamination of both lenses and cases as well as *K. pneumoniae* contamination of the cases. Conversely, *Klebsiella* contamination was associated with older age. In addition, *Pseudomonas* contamination of the lens was predicted by the type of solution used but not solution change frequency. *Pseudomonas* contamination of the case could be predicted by using distilled water or bottled water compared to commercially available solution and by lens solution change intervals exceeding one

**Table 6** Multivariable Analysis of Bacterial Contamination of Contact Lenses (CL) and Their Cases

Microorganism	Sample	Predictors	Odds Ratio	95% Confidence Interval	P value
Staph. aureus	Contact lens	>1 month CL change interval	5.853	0.652–52.563	0.115
		>6 hours CL use per day	4.257	0.470–38.538	0.197
Staph. epidermidis	Contact lens	Solution change frequency (>7 days) vs daily	54.564	0.001–441.40	0.998
Pseudomonas aeruginosa	Contact lens	Age (18–22 years)	<b>12.302</b>	<b>2.708–55.876</b>	<b>0.001</b>
		CL change interval of >1 month	1.024	0.238–4.407	0.975
		Bottled water vs commercially available	<b>10.836</b>	<b>2.641–44.459</b>	<b>0.001</b>
		Distilled water vs commercially available	<b>13.487</b>	<b>2.451–74.233</b>	<b>0.003</b>
		Solution change frequency (>7 days) vs daily	17.469	0.999–305.628	0.050
	Contact lens case	Age (18–22 years)	<b>6.555</b>	<b>2.077–20.685</b>	<b>0.001</b>
		CL change interval of >1 month	2.004	0.589–6.818	0.266
		Bottled water vs commercially available	<b>4.451</b>	<b>1.032–19.188</b>	<b>0.045</b>
		Distilled water vs commercially available	<b>5.796</b>	<b>1.772–18.958</b>	<b>0.004</b>
		Solution change frequency (>7 days) vs daily	<b>26.616</b>	<b>1.833–386.565</b>	<b>0.016</b>
		1–7 days vs daily	5.323	0.562–50.421	0.145
E. coli	Contact lens	CL change interval of >1 month	13.647	0.608–306.331	0.100
K. pneumoniae	Contact lens case	Age group (23–26) years	<b>4.154</b>	<b>1.480–11.657</b>	<b>0.007</b>

**Note:** Bold text indicates significant results.

week compared to daily change. Other predictors that were significant on univariable analysis were not significant on multivariable analysis.

## Discussion

The results of the present study demonstrate a high rate of contact lens and case contamination among asymptomatic female medical students in Southern Iraq. Most of the participants (79.1%) had at least one contaminated sample. Previous related studies reported an overall rate of contamination ranging from 17% to 52%.<sup>15–19</sup> However, the present and previous studies were carried out on different populations and samples were collected using a range of methodologies. Poor hygienic practices were present among our study population such as wetting the CL with saliva before applying it to the eye in at least one participant. The risks associated with this practice are well documented. Contact lens cases had a higher contamination rate in our study than the lenses themselves (88 vs 76 samples). It is possible that micro-organisms flourish in the contact lens case first followed by secondary contamination of the lens.<sup>19</sup> Several other comparable studies from nearby regions have also reported a relatively high rate of contamination of CL cases (Table 7).<sup>16–19</sup> However, several factors other than lens care practice could potentially influence the rate of microbial contamination, including population age, gender, type of contact lens, environmental temperature and microbial isolation techniques.

It is known that bacterial adherence to contact lenses increases with decreased water content of the lens and that hydrophobic lenses are more susceptible to bacterial adherence than high water-content lenses.<sup>20</sup> High ambient



**Table 7** Rate of Microbial Contamination of Contact Lenses (CL) and Their Cases as Reported by Various studies<sup>16-19</sup>

Study Author (s)	Year Published	Location	Total Number of Participants	Rate of CL Contamination	Rate of CL Case Contamination
Yung et al	2007	China	101	9%	34%
Rahim et al	2008	Pakistan	100	65%	89%
Thakur et al	2014	India	50	56%	62%
Waleed et al	2021	Jordan	63	Not done	52%
Current study		Iraq	115	66%	76.5%

temperature in the range of 35 Celsius and above can contribute to dehydration of the lens and possibly increasing bacterial adherence and thereby account for the higher contamination rate observed in our study.<sup>21</sup>

Six different species of bacteria were identified in our study. Gram positive bacteria included *Staph. aureus*, *Staph epidermidis* and *Bacillus* species. Although these micro-organisms may provide a small contribution to the normal microbiome of the ocular surface, they can be involved in devastating keratitis especially under compromised ocular surface conditions which may be associated with chronic contact lens use.<sup>5,22,23</sup> In fact, there is some evidence that epithelial cells of the ocular surface respond weakly to colonization by bacteria that contribute to normal microbial flora, perhaps allowing more severe ocular infection when caused by these micro-organisms.<sup>24</sup> Multivariable analysis in the present study showed no significant association between the three identified species and any lens wear regimen or lens care practice.

Gram negative bacteria that were identified include *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *E. coli*. These micro-organisms are thought to rarely contribute to normal flora of the ocular surface; however, recent genomic evidence suggests that they may contribute significantly to the normal ocular microbiome and may therefore cause severe keratitis with devastating outcomes.<sup>24</sup> *Pseudomonas* in particular is known to be the most common cause of contact lens-related aggressive bacterial keratitis.<sup>5,17</sup> Consequently, identifying and addressing risk factors for *pseudomonas* contamination could potentially decrease the incidence of such aggressive forms of keratitis.

In our study, *Pseudomonas* contamination was significantly associated with younger participants who were six to 12 times more likely to have CL and case contamination, respectively. This can be attributed to the possible lack of sufficient knowledge about the risks associated with poor contact lens care in this age group and that older medical students may be more aware of anterior segment pathology and the need for careful CL care practice. The use of bottled or distilled water significantly increased the odds of *P. aeruginosa* contamination of both CL and case when compared to commercially available solution. This is expected since commercially available solutions are preserved; however, no currently available solution is immune to contamination.<sup>25</sup> Longer time intervals between solution changes were associated with *pseudomonas* contamination of cases only. The lack of significant association may be due to the limited number of contaminated CL samples.

Our study was limited by the number of samples contaminated by some species, and this may contribute to the lack of statistical significance with respect to those species.

## Conclusion

Microbial contamination rate of soft CL and their cases is high among young medical students in comparison to previously reported rates and was predisposed by several poor hygienic practices and wearing regimens. We recommend further larger population-based studies to identify and confirm the association between certain poor hygienic practices and CL wear and to educate the population in general and soft CL wearers in particular regarding the possibility of serious microbial infections that can result from high contamination rates due to poor hygienic practices.

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## Disclosure

The authors declare that they have no conflicts of interest regarding this work.

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