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Research Article

# Bacteriological Contamination in Indoor Swimming Pools: Prevalence and Pathogen Identification in Wasit, Iraq

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# **About** Article

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# ABSTRACT

Since tainted water may spread a number of waterborne illnesses, water quality is an important consideration in preserving public health. The pollution of swimming pools, a common characteristic of recreational facilities, especially in the warmer months, is one of the main issues. The purpose of the research was to find out how often bacterial contamination was in indoor swimming pool settings and water. During June and July of 2024, a total of 100 swab samples were gathered evenly from the water, pool ladders, pool margins, and changing areas of several swimming pools in Wasit province, Iraq. Following their independent cultivation on three different medium (Blood Agar, Eosin Methylene Blue Agar, and MacConkey Agar), the swabs were purified and identified using biochemical testing and morphological traits. In all, 37% of swab samples included different types of bacteria, which were detected in water (12%), pool ladders (44%), changing rooms (36%), and pool margins (56%). Following that, 68 (37.99%), 39 (21.79%), 45 (25.14%), and 27 (15.08%) bacterial isolates were found in water, changing areas, pool borders, and pool ladders, respectively. Twelve bacterial species were found in changing rooms, however Escherichia coli and Klepsellia aerogenes were the most important. Eleven bacterial species were identified in pool borders; notable increases were seen in Vibrio cholera, Aeromonus hydrophila, Escherichia coli, and Klepsellia pneumonia. Ten bacterial species, including Vibrio cholera, Escherichia coli, and Klepsellia pneumonia, were identified in relation to the pool ladders. Along with Vibrio cholera, Aeromonus hydrophila, Pseodomonus argenosa, Proteus mirabrillis, and, less significantly, Klepsellia pneumonia, Acintopacter baumannii, Klepsellia aerogenes, Proteus vulgaris, Salmonella epidermidis, Streptococcus faecalis, Anteropacter cloacae, Citropecter frndii, Edwardsiella tarda, and Serratia liquefaciens, 15 species of bacteria were identified in relation to water. According to the current study's findings, swimming pools are a source of diverse bacteria that swimmers may get, leading to a variety of diseases, including skin infections. Therefore, in order to reduce the danger of bacterial contamination in swimming pools, we recommended the adoption of thorough water quality monitoring programs, the use of cutting-edge diagnostic tools to detect and track possible pathogens, and the use of efficient disinfection tactics.

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## **1. INTRODUCTION**

Water is an essential resource for sustaining the life, and its contamination poses a significant threat to human health and the environment (Mishra, 2023). The prevalence of water-borne pollutants (physical, chemical and biological) has become a global concern since over one billion people lack access to clean water sources (Manetu & Karanja, 2021). One of the most widespread water quality issues is pathogen loading through the deposition of sewage and untreated wastewater, which leading degradation of aquatic ecosystems, and spreading of infectious diseases, with accumulation of hazardous substances such as heavy metals that can result in chronic poisoning and a variety of health problems (Akpor & Muchie, 2011; Karri et al., 2021; Jan et al., 2022). To address the issue of water contamination, a multifactorial approach is required involving regular monitoring, effective treatment technologies, and the implementation of stricter regulations to control the discharge of pollutants into water bodies (Altenburger et al., 2015; Brack et al., 2017).

#### 2. LITERATURE REVIEW

Swimming pools are ubiquitous features of many recreational facilities, providing a refreshing and enjoyable environment for people of all ages to engage in physical activity, socialize and beat the heat during the warmer months (Anderson et al., 2014; Pangrazi & Beighle, 2019; Chaúque et al., 2022). However, the very nature of swimming pools with their high bather loads and intermittent water circulation can make them susceptible to bacterial contamination posing potential health risks to swimmers (Barna & Kádár, 2012; Bonadonna & La Rosa, 2019). One of the primary concerns regarding bacterial contamination is the presence of pathogenic microorganisms that can cause a wide-range of infections even death (Okafor, 2011; Stec et al., 2022). Furthermore, the formation of biofilms by bacterial communities within the pool and existence of wild animals such as birds, dogs and cats can act as a source of contamination and enhance their ability to resist disinfection (Guida et al., 2016; Khan et al., 2016; Nguyen et al., 2024). Additionally, investigations have been revealed the presence of multidrug resistant bacteria on inanimate surfaces within the pool environment highlighting the potential cross-contamination and needing to rigorous cleaning and disinfection protocols (Cantón et al., 2013; Kramer & Assadian, 2014; Koeck et al., 2018; Ekowati, 2019).

In Iraq, the numbers of swimming pools have been increased significantly in last 20 years. Nonetheless, almost national studies have focused on water hygiene or water quality (Khalaf *et al.*, 1992; Aenab & Singh, 2012), and data described the microbiological characteristics of public swimming pools remain very rare of old (Ali *et al.*, 2009). Hence, the current study was conducted to investigate the almost prevalent bacterial contamination in indoor swimming pools in Baghdad and Wasit provinces (Iraq).

#### **3. MATERIALS AND METHODS**

### 3.1. Ethical approval

This study licensed by the Scientific Committee of the

Department of Biology, College of Education for Pure Sciences (University of Wasit).

## 3.2. Preparation of culture media and transport broth

In this study, three culture media (Blood Agar, Eosin Methylene Blue Agar and MacConkey Agar) and one transport broth, Brain-Heart Infusion (BHI), in addition to seven biochemical tests (catalase, coagulase, gas and indol production, sugar fermentation and urease activity), and Gram stain were prepared according to their manufacturers' instructions (HiMedia, India).

## 3.3. Samples

A total of 100 swab samples were collected from the changing rooms (total no=25), pool edges (total no=25), pool ladders (total no=25) and water (total no=25) of various indoor swimming pools found in Wasit province (Iraq) during June and July (2024). The swabs were kept in tubes containing the BHI broth and transported to the laboratory under cooled condition to be cultured as soon as possible.

## 3.4. Bacteriological examination

Under aseptic conditions, the swabs were streaked separately on the prepared three media, and incubated at 37°C for 24 hours, and the grown colonies were re-cultured additionally for purification (Ibraheim *et al.*, 2023).

#### 3.5. Morphological and biochemical characterization

Initially, all bacterial isolates were used to preparation the Gram stained slides that tested by light microscope (Zeiss, Germany) at a magnification of 1000X to identify their morphological characteristics. Finally, biochemical testing was done to the species of study isolates (Jasim *et al.*, 2024).

#### 3.6. Diagnosis of bacterial species via VITEK2 technology

The species of bacterial isolates were identified in current study using the VITEK2 Compact System (bioMérieux, France). Briefly, bacterial suspensions were employed in 45% sterile NaCl as similar to McFarland Standards (0.5x10<sup>8</sup> CFU/ml).

#### 3.7. Statistical analysis

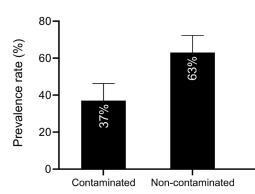
The t-test and One Way-Analysis of Variance (ANOVA) in the GraphPad Prism Software (version 8.0.2) was applied to detect significant differences between study values (mean  $\pm$  standard error) at p<0.05 (Gharban, 2024).

#### 4. RESULTS AND DISCUSSION

#### 4.1. Results

Our findings revealed that overall 37% (37/100) of swab samples were contaminated with various species of bacteria (Figure 1). Significantly (p<0.0283, CI = 7.430 to 66.57, r2 = 0.8409), pool edges were showed the higher rate of contamination [56% (14/25)] while water having the lower one [12% (3/25)] when compared to values of changing rooms [36% (9/25)] and pool ladders [44% (11/25)], (Figure 2).





**Figure 1.** Total results of testing the environments and water of swimming pools (total No=100)

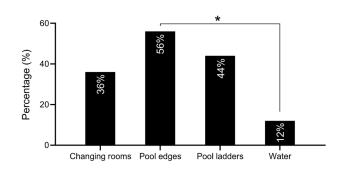
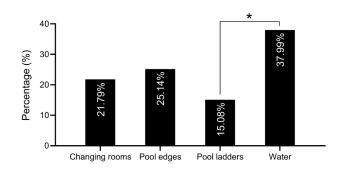


Figure 2. Distribution of contaminated samples according to type of swab sample

Subsequently, significant increase [p<0.0138, CI = 9.697 to 40.30, r2 = 0.9001] in number of bacterial isolates was identified in water [68 (37.99%)] whereas the lowest was observed in pool ladders [27 (15.08%)] in comparison to changing rooms [39 (21.79%)] and pool edges [45 (25.14%)], (Figure 3).



**Figure 3.** Number of bacterial isolates obtained from the environments and water of swimming pools

In changing rooms, a total of 12 species of bacteria was reported; in which, significant prevalence was seen in *Escherichia coli* (28.21%) as well as in *Klepsellia aerogenes* (12.82%) when compared to other bacterial species including

Pseodomonus argenosa (10.26%), Staphylococcus aureus (10.26%), Vibrio cholera (10.26%), Proteus mirabrillis (7.69%), Aeromonus hydrophila (5.13%), Klepsellia pneumonia (5.13%), Acintopacter baumannii (2.56%), Proteus vulgaris (2.56%), Salmonella epidermidis (2.56%), and Streptococcus faecalis (2.56%), (Table 1).

 Table 1. Bacterial species identified changing rooms of swimming pools

Species	No.	%	_
Acintopacter baumannii	1	2.56	
Aeromonus hydrophila	2	5.13	
Escherichia coli	11	28.21 **	
Klepsellia aerogenes	5	12.82 *	
Klepsellia pneumonia	2	5.13	p<0.0038
Proteus mirabrillis	3	7.69	CI = 2.189 to 9.576
Proteus vulgaris	1	2.56	$r^2 = 0.4160$
Pseodomonus argenosa	4	10.26	
Salmonella epidermidis	1	2.56	
Staphylococcus aureus	4	10.26	
Streptococcus faecalis	1	2.56	
Vibrio cholera	4	10.26	

In pool edges, 11 bacterial species were recorded; in which, significant increases were seen in *Klepsellia pneumonia* (20%), *Escherichia coli* (17.78%), as well as in *Aeromonus hydrophila* (13.33%), *Vibrio cholera* (13.33%), *Proteus mirabrillis* (11.11%) in comparison to other bacterial species; *Acintopacter baumannii* (6.67%), *Klepsellia aerogenes* (4.44%), *Proteus vulgaris* (4.44%), *Salmonella epidermidis* (4.44%), *Serratia liquefaciens* (2.22%), and *Staphylococcus aureus* (2.22%), (Table 2).

 Table 2. Bacterial species identified pool edges of swimming pools

Species	No.	%	_
Acintopacter baumannii	3	6.67	
Aeromonus hydrophila	6	13.33 *	
Escherichia coli	8	17.78	
Klepsellia aerogenes	2	4.44	p<0.0023
Klepsellia pneumonia	9	20 **	p<0.0023 CI = 2.431
Proteus mirabrillis	5	11.11	to 9.331
Proteus vulgaris	2	4.44	$r^2 = 0.4494$
Salmonella epidermidis	2	4.44	
Serratia liquefaciens	1	2.22	
Staphylococcus aureus	1	2.22	
Vibrio cholera	6	13.33 *	



**Table 3.** Bacterial species identified pool ladders of swimming pools

Species	No.	%	
Aeromonus hydrophila	1	3.7	
Escherichia coli	5	18.52 *	
Klepsellia pneumonia	4	14.81	p<0.0099
Proteus mirabrillis	2	7.41	
Proteus vulgaris	1	3.7	CI = 1.620 to 10.14
Pseodomonus argenosa	1	3.7	$r^2 = 0.3485$
Roatinolia terrigena	1	3.7	
Salmonella epidermidis	3	11.11	
Staphylococcus aureus	1	3.7	
Vibrio cholera	8	29.63 **	

Regarding water, 15 species of bacteria were diagnosed; most significantly including *Escherichia coli* (19.12%) in addition to *Vibrio cholera* (13.24%), *Aeromonus hydrophila* (10.29%), *Pseodomonus argenosa* (10.29%), *Proteus mirabrillis* (8.82%), and less significantly in *Klepsellia pneumonia* (7.35%), *Acintopacter baumannii* (5.88%), *Klepsellia aerogenes* (5.88%), *Proteus vulgaris* (4.41%), *Salmonella epidermidis* (4.41%), *Streptococcus faecalis* (4.41%), *Anteropacter cloacae* (1.47%), *Citropecter frndii* (1.47%), *Edwardsiella tarda* (1.47%), and *Serratia liquefaciens* (1.47%), (Table 4).

Table 4. Bacterial species identified water of swimming pools

Species	No.	%	_
Acintopacter baumannii	4	5.88	0.29 .47
Aeromonus hydrophila	7	10.29	
Anteropacter cloacae	1	1.47	
Citropecter frndii	1	1.47	
Edwardsiella tarda	1	1.47	
Escherichia coli	13	19.12 ***	p < 0.0003 CI = 3.208 to 8.555 $r^2 = 0.5761$
Klepsellia aerogenes	4	5.88	
Klepsellia pneumonia	5	7.35 *	
Proteus mirabrillis	6	8.82	
Proteus vulgaris	3	4.41	
Pseodomonus argenosa	7	10.29	
Salmonella epidermidis	3	4.41	
Serratia liquefaciens	1	1.47	
Streptococcus faecalis	3	4.41	
Vibrio cholera	9	13.24 **	

#### 4.2. Discussion

One of the most prevalent environmental issues worldwide is water pollution, which is a global cry to enhance people's

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lives and opportunities (Muhammed & Abubakar, 2022). Since many people use public indoor swimming pools on a daily basis, they may be contaminated with bacteria and other infectious agents, including a wide range of unicellular microorganisms (Papadopoulou et al., 2008; Gerba & Pepper, 2019). In addition to their potential to spread either directly or indirectly, bacteria are quite common in the environment and may quickly colonize, divide, and/or be found in nutrients, air, soil, and water (Dugan, 2022). 37% of the swab samples in this research included different types of bacteria, which were detected in the water (12%), pool ladders (44%), changing rooms (36%), and pool margins (56%). Following that, 68 (37.99%), 39 (21.79%), 45 (25.14%), and 27 (15.08%) bacterial isolates were found in water, changing areas, pool borders, and pool ladders, respectively. Twelve bacterial species were found in changing rooms, however Escherichia coli and Klepsellia aerogenes were the most important. Eleven bacterial species were identified in pool borders; notable increases were seen in Vibrio cholera, Aeromonus hydrophila, Escherichia coli, and Klepsellia pneumonia. Ten bacterial species, including Vibrio cholera, Escherichia coli, and Klepsellia pneumonia, were identified in relation to the pool ladders. Along with Vibrio cholera, Aeromonus hydrophila, Pseodomonus argenosa, Proteus mirabrillis, and, less significantly, Klepsellia pneumonia, Acintopacter baumannii, Klepsellia aerogenes, Proteus vulgaris, Salmonella epidermidis, Streptococcus faecalis, Anteropacter cloacae, Citropecter frndii, Edwardsiella tarda, and Serratia liquefaciens, 15 species of bacteria were identified in relation to water. According to our results, the top bacterial species found in swimming pool surroundings and water were Escherichia coli, Klepsellia spp. (K. aerogenes and K. pneumonia), and Vibrio cholera. According to Van Elsas et al. (2011) and Loayza et al. (2020), Escherichia coli is an intestinal pathogen that is spread from human or animal excrement to vulnerable hosts by contact with food and environmental compartments (water, surfaces, soil, hands, and flies). Since the presence of E. coli in water is often linked to an elevated risk of both enteric infections and diarrheal disorders, this bacterium is employed as an indicator to investigate the causes and outcomes of fecal pollution in the environment (Hunter, 2003; Gomes et al., 2016; Ercumen et al., 2017).

After a trailer park pool party, Friedman *et al.* (1999) found a cluster of gastrointestinal disorders, including one incidence of hemolytic-uremic syndrome and one culture-confirmed E. coli infection. Due to the fact that swimming pools are linked to a number of outbreaks because of the relative susceptibility of E. coli to sufficient levels of free chlorine, a report published in the United Kingdom details the investigation of an outbreak connected to a local leisure center pool and offers suggestions regarding the safe management of such facilities (Verma *et al.*, 2007).

Some bacteria are resistant to sodium hypochlorite, which is used to disinfect pools and is thought to be an opportunist microbe that causes various diseases, according to Rasti *et al.* (2012). These bacteria should not be disregarded when assessing the water quality of swimming pools, even if they are not thought to pose a serious risk and do not directly cause diseases in people (Purohit *et al.*, 2020; Nowicki *et al.*, 2021). Swimming pools may contain microorganisms linked to swimmers, such as fecal contamination of the water, unintentional fecal release or residual fecal material on bodies, and non-fecal shedding such as vomit, mucous, saliva, skin, mouth, and upper respiratory tract contamination, according to Ghasemi *et al.* (2019). According to earlier research, some bacteria may lead to a number of infections or illnesses affecting the respiratory, skin, or central nervous systems (Nichols, 2006; Papadopoulou *et al.*, 2008; WHO, 2009). The quality of pool waters during the working day has also been studied. According to some researchers, young children make up the majority of swimmers on weekends and holidays, so water sampling would be more effective during these periods (Fantuzzi *et al.*, 2010; Lévesque *et al.*, 2015; Carter *et al.*, 2019).

# **5. CONCLUSIONS**

According to the current study's findings, swimming pools are a source of diverse bacteria that swimmers may get, leading to a variety of diseases, including skin infections. Therefore, in order to reduce the danger of bacterial contamination in swimming pools, we recommended the adoption of thorough water quality monitoring programs, the use of cutting-edge diagnostic tools to detect and track possible pathogens, and the use of efficient disinfection tactics.

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