

Prevalence, Antibiotic Resistance, and Biofilm Formation of Bacterial Species in Water Samples from Baghdad, Iraq

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ABSTRACT

Background: Our global health is seriously threatened by the emergence rapid spread of antibiotic resistance bacteria. The misuse of antibiotics has resulted in heightened selection pressure and the proliferation of multidrug-resistant pathogens, thereby diminishing the efficacy of treatments. Water sources serve as important reservoirs for the transmission of antibiotic-resistant bacteria. This study aimed to isolate and identify aquatic bacterial species from diverse sources in Baghdad, assess their antibiotic resistance profiles, and detect the biofilm formation of the most multidrug resistance bacterial species to find the relationship between slime layer production and resistance to antibiotics. **Methodology:** We conducted an analysis of bacterial species in water samples obtained from 80 varied locations between July 2023 and April 2024, employing chromogenic media, biochemical testing, and the API system. The antibiotic sensitivity test of the bacterial isolates was conducted using the Kirby-Bauer method for ten types of antibiotics from various groups, including (AMP, AMC, CTX, HLS, C, CIP, TR, K, AK and O. Biofilm production of the most resistance to the highest number of antibiotics bacteria was assessed using two methods: Congo red agar and adhesion to a smooth surface. **Results:** Findings indicate an isolation rate of 40 bacterial isolates, distributed among water samples as follows: 18 (45%) from rivers, 13 (32.5%) from stagnant water, 5 (12.5%) from turbid water, 4 (10%) from tank water, and none from tap water. The primary bacterial species include *Escherichia coli* isolates, *Pseudomonas aeruginosa*, *Enterobacter*, *Klebsiella oxytoca*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Acinetobacter*, and *Salmonella*. The most common bacterial species were *E. coli* and *P. aeruginosa*. The majority of isolates exhibited resistance to AMP, AMC, then HLS, K, AK, O, then CTX, C, TR, and CIP that has the lowest resistance percentage. The results indicate variability in slime layer production among the isolates, and *E. coli*, *P. aeruginosa* were high resistance to AMP and AMC. **Conclusions:** The findings reveal the existence of multiple bacterial species in Baghdad water, which show susceptibility to various antibiotics while exhibiting resistance to ampicillin, tetracycline, and kanamycin, and differ in their production of slime layers. The findings increase public health awareness of antibiotic resistance bacterial species and slime layer production load in aquatic environments.

Keywords: Aquatic Bacteria, MDR bacteria, Chromogenic agar, biofilm formation.

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INTRODUCTION

Water is known a conduit for the transmission of microorganisms affiliated with human health (1). Clean drinking water, an essential human right, can adversely affect health if contaminated with environmental opportunistic pathogenic microorganisms (2). Consequently, safeguarding human health necessitates maintaining drinking water

devoid of microbiological contamination (3). Surface water that had been not treated from rivers, streams, and reservoirs is directly utilized in rural areas for drinking and other household tasks (4). Runoff from rainfall and inputs from agricultural, when combined with animal waste and sewage effluents, can contaminate exposed water sources with microorganisms, rendering them unsuitable for human consumption (5).

All aquatic ecosystems rely on microorganisms, as they are essential to biogeochemical cycles and other critical environmental processes (6). Tigris and Euphrates Rivers with their tributaries are the main water resources in Iraq. Since the 1970s, the flow of these rivers has diminished because of hydrological projects implemented in riparian countries and climate change (7). Although Iraq possesses ample river resources and a substantial aquifer, it remains a desert nation where these resources have been excessively exploited for domestic, industrial, and agricultural uses. Bacterial, viral, and parasitic disease outbreaks have been associated with overloaded water distribution systems, antiquated wastewater treatment plants (WWTPs), and contaminated freshwater sources resulting from agricultural runoff, domestic and industrial waste, and the utilization of sewage water for vegetable irrigation (8).

In 2021, UNICEF reported that fewer than fifty percent of Iraqi schools had access to fundamental water resources, jeopardizing nutrition, health of children, future economic prospects, and cognitive development. Approximately 60% of children in the nation do not have access to safely managed water services (9). The efficiency of the distribution network is a dismal 32%, and it is deteriorating over time (10). According to the UN, 6 million individuals lacking access to clean water, and over 500,000 Iraqi children obtain the water that need from open wells, while more than 200,000 rely on rivers. In the first half of 2010, over 360,000 cases of diarrhea were documented, attributed to contaminated drinking water and a widespread lack of awareness regarding hygiene among the local populace, particularly among vulnerable demographics such as women and children. The report indicates that the Tigris River receives a minimum of 250,000 tons of raw sewage daily, jeopardizing sensitive water sources and the entire water distribution infrastructure (8). Contaminated drinking water supplies are a primary source of waterborne infections. *Acinetobacter*, *Chromobacterium*, *Salmonella*, *Alcaligenes*, *Staph. aureus*, *Flavobacterium*, *Clostridium botulinum*, *P. aeruginosa*, *Vibrio cholerae* and *E. coli* are the main human pathogen that contaminate water (11) .

The utilization of antibiotics in human and veterinary medicine has led to the emergence of resistant bacteria, adversely affecting disease treatment. Significant increases in antibiotic consumption via agricultural practices and human treatments have been recorded (12, 13). Consequently, antibiotic resistance has emerged as a significant public health issue, with substantial evidence of its presence in surface, wastewater, and potable water (14). The resistance of microorganisms to antibiotics heightens the risk linked to their pathogenicity. The emergence of multidrug-resistant organisms may escalate due to the processes of biological treatment in wastewater treatment facilities, potentially resulting in a particular proliferation of resistant bacteria (15). Chlorination diminishes the quantity of pathogens in potable water; however, some may still withstand the course of treatment and make their way into the distribution network (16).

Biofilms are communities of microorganisms that adhere to biotic and a biotic surface; secrete protective extracellular matrices that significantly impacting various fields including medicine, industry, and environmental science. Continuing challenges presented by biofilms in clinical medicine continue to render research in this area both dynamic and uncertain (17). The initiation occurs through a complex interplay of genetic factors and environmental, primarily associated with stress responses. External factors, including temperature, pH, environmental hazards, and nutrient availability, influence the transition of planktonic microorganisms to an adherent state (18). Individual bacteria within biofilms exposed to elevated antibiotic concentrations can endure and reform a further resistant biofilm, a phenomenon referred to as recalcitrance (19). Limited data are recorded on biofilm formation in Baghdad's water. The study aimed to isolate environmental bacteria from diverse sources in Baghdad, identify bacterial species through chromogenic media and biochemical tests, and assess their antibiotic resistance profiles, considering the potential impact of treated wastewater quality on public health and the importance of reducing pathogenic microorganisms in drinking water, and detect the biofilm formation of the most multidrug resistance bacterial species to combining antibiotic resistance and biofilm analysis in Baghdad's diverse water sources.

METHODOLOGY

1- Samples collecting: Between July 2023 and April 2024, eighty water samples were collected. Aseptically collected water samples were placed in sterile 150 ml glass bottles by submerging the bottles directly into the surface of multiple water sources (river, stagnant, tank, and turbid) from various sampling locations in early morning hours along the Tigris River in Baghdad. The samples should take one- third on the way below the surface if the depth is less than 0.5 Meter and 0.3 if the depth is greater than 0.5 Meter with avoiding debris and scums. Following a one-minute period of running the tap, the samples were collected and transferred into sterile bottles. All samples were labeled and transported to the Biology department laboratory on ice for analysis (20).

2- Identification of Isolates: The primary identification tests included culturing collected samples on Nutrient agar for bacterial species cultivation and colonial morphology, MacConkey agar to differentiate between gram-positive and gram-negative bacteria, Blood agar for the cultivation of both gram-positive and gram-negative bacteria and for haemolysis detection, Eosin Methylene Blue (EMB) agar to distinguish *E. coli* from other Enterobacteriaceae, and Triple Sugar Iron (TSI) agar to differentiate various Gram-negative bacteria. The Indole test, Citrate utilization test, Oxidase test, and culture on selective media, including Mannitol salt agar, Salmonella-Shigella agar, and Cetramide agar, were performed for further identification. MacConkey broth was employed for serial dilution, specifically from sewage, turbid and stagnant water samples (21).

3- Culture on Chromogenic media: A colorless substrate found in chromogenic agar undergoes an enzymatic reaction to produce a recognizable color (22). HiCrome agar (Himedia/India) was used for detection and differentiated between bacterial species.

4- Analytical Profile Index (API) Test: The organisms were identified to the species level using the API guide, and the API test was carried out in compliance with the manufacturer's protocol (BioMérieux, France).

5- Measurement of (Most Probable Number): The standard medium that was preferred was MacConkey broths in tubes. Durham's tube made it easier to detect the production of gas. A measured volume of water samples was added to tubes of MacConkey purple broth. For 48 hours, the tubes were incubated at 37°C. When the medium's color changed from purple to yellow and gas accumulated in the Durham tube, the test was considered successful (23). Using McCrady's probability table, the coliform count per 100 milliliters of water was calculated based on the tubes displaying gas and acid production. The presumptive coliform count, which is the most likely number, was obtained from the analysis (24, 25). Since coliform bacteria can also come from environmental sources, their presence does not always indicate fecal contamination.

6- Antimicrobial Susceptibility Testing: By Kirby-Bauer disk diffusion method (26). Ten antibiotic discs (Himedia) at the final concentrations that are indicated were used include the following: ampicillin (AMP) 10 mcg, amoxycylav (AMC) 30(20/10) mcg, cefotaxime (CTX) 30 mcg, streptomycin (HLS) 300 mcg, chloramphenicol (C) 30 mcg, ciprofloxacin (CIP) 5 mcg, trimethoprim (TR) 5 mcg, kanamycin (K) 30 mcg, amikacin (AK) 30 mcg, and oxytetracycline (O) 30 mcg. These antibiotics were picked because prior research has documented microbial resistance to them or because they are utilized in both human and animal veterinary medicine (16).

Based on standard reference values set by the Clinical and Laboratory Standards Institute (CLSI), the results were used to classify isolates as susceptible, intermediate resistant, or resistant to a particular antibiotic (27).

7- Detection of Biofilm formation: The two methods that were used in this study were quantitative method for biofilm formation that includes:

1- Congo red agar: It was illustrated according to Freeman *et al.* (28). Bacterial isolates were cultured on medium plates and incubated for 24 to 48 hours at 37°C. Positive results were indicated by dry black colonies, moderate producers exhibited a gray coloration, while weak slime producers typically appeared pink (29).

2- Adherence on smooth surfaces: 10 ml of Tryptic soy broth (Himedia/ India) in glass tubes were inoculated with single colonies of the test bacterial strain using a sterile loop, the negative control was prepared by adding 10 ml of Tryptic soy broth to a culture tube.

Inoculated at 37°C for 24 to 48 hours. Subsequently, the contents of the tube were decanted, and 10 ml of a 0.1% safranin stain solution, previously prepared by dissolving 0.1 g of safranin in 100 ml of distilled water, was added to all tubes, including the negative control, the contents were carefully decanted. After that, the tubes were turned upside

down to make drainage easier (30).

8- Statistical Analysis

To find the distribution of difference factors in the parameters study; a statistical (2019) program was utilized (User's Guide to the Statistical Analysis System). In this study, the chi-square test was utilized to compare percentages (0.01 probabilities) in a significant way (31).

RESULTS

1-Water samples and identification of bacterial isolates: Eighty water samples were obtained from various water sources. Eighteen isolates were derived from river water, thirteen from stagnant water, five from turbid water, and four from tank water, representing eight bacterial species as illustrated in the Table 1 and 2. The bulk of these isolates originated from river, stagnant, turbid, and tank water, respectively; no bacterial isolates were acquired from tap water. All isolates were evaluated for growth on selective and differential media. Gram staining was used to identify the cocci, bacilli, and coccobacilli forms of the various colonies that were obtained. The majority of strains were identified by biochemical tests, cultivated on chromogenic agar Table 3. It was found that the water of these various samples contained distinct bacterial colonies. Colonies ranged from irregular, round borders, Figure 1 and 2.

Table (1): The number and source of water samples with the no. of bacterial isolates, and the isolation percentage

Water sample source	No. of water samples (%)	No. of bacterial isolates (%)	P-value	Isolation percentage (%)
River	20 (25.00%)	18 (45.00%)	0.0335 *	90
Stagnant	20 (25.00%)	13 (32.50%)	0.294 NS	65
Turbid	20 (25.00%)	5 (12.50%)	0.0072 **	25
Tank	14 (17.50%)	4 (10.00%)	0.189 NS	28.57
Tap water	6 (7.50%)	0 (0.00%)	0.0264 *	0
Total	80	40	---	%
P-value	0.0001 **	0.0001 **	---	----
** (P≤0.01).				

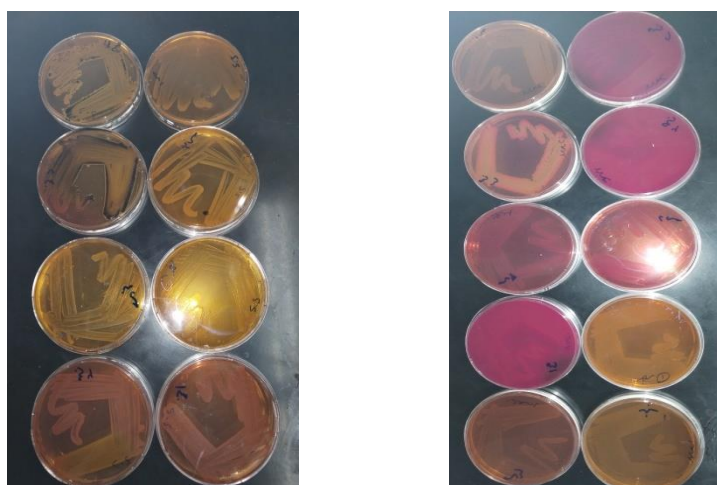
**** Highly Significant (P≤0.01). NS non- significant**

Table (2): Identification of the bacterial isolates from different water samples sources by using biochemical tests and chromogenic medium.

Water sample source	Bacterial isolates
River	<i>Escherichia coli</i> , <i>Pseudomonas aeruginosa</i> , <i>Staphylococcus aureus</i> , <i>Klebsiella pneumoniae</i> , <i>K. oxytoca</i> , <i>Enterobacter</i> , <i>Acinetobacter</i> , and <i>Salmonella</i>
Stagnant	<i>E. coli</i> , <i>P. aeruginosa</i> , <i>K. pneumoniae</i>
Turbid	<i>E. coli</i> , <i>K. pneumoniae</i>
Tank	<i>E. coli</i>
Tap water	non

Table (3): the morphological appearance of bacterial species on Chromogenic agar

Bacterial genus or species	Appearance of colonies
<i>Klebsiella</i>	metallic blue (+/- reddish halo)
<i>E. coli</i>	dark pink to reddish
<i>Enterobacter</i>	turquoise blue
<i>Pseudomonas</i>	translucent (+/- natural pigmentation cream to green)
<i>S. aureus</i>	golden, small opaque
<i>Acinetobacter</i>	Red
<i>Salmonella</i>	mauve (rose to purple)

**Figure (1):** Primary isolation of bacterial isolates on selective and differential media

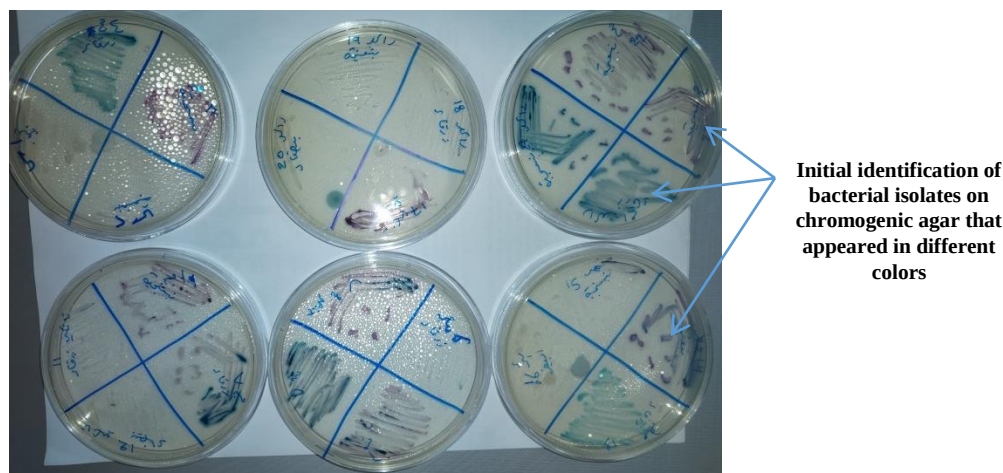


Figure (2): The appearance of aquatic bacterial isolates on chromogenic agar

It was found that the water of these various samples contained distinct bacterial colonies. The colonies, circular margins. Gram staining was used to identify the cocci, bacilli, and coccobacilli forms of the various colonies that were obtained. When examined under a microscope, the majority of strains showed that gram negative bacilli Predominated, followed by gram positive cocci.

2- Measurement of MPN: It was estimated that the MPN of all the water samples that were collected was extremely high (except tap water). The water's quality was subpar, with a significantly high coliform count. The amount of *E.*

coli in each 100 milliliters of water was also greater than 1, which further indicated the degree of contamination in these samples.

3- Antibiotics sensitivity: Ten antibiotics were used (ampicillin, cefotaxime, amoxyclav, streptomycin, chloramphenicol, ciprofloxacin, trimethoprim, kanamycin, amikacin, and oxytetracycline), the results show that all the isolates from different water samples were resist to ampicillin, while they sensitive to the other antibiotics in different degree as shown in Table (4, 5).

Table (4): Percentage of resistance of bacterial isolates to different antibiotics, the antibiotic sensitivity test by Kirby- Bauer disk diffusion, and the concentrations by (mcg)

Bacterial isolates	No. of isolates	% of resistance to antibiotics										P-value
		AMP (10)	AMC (30)	CTX (30)	HLS (300)	C (30)	CIP (5)	TR (5)	K (30)	AK (30)	O (30)	
<i>E. coli</i> (n= 13)	13	100	80	70	80	40	20	40	60	60	80	0.0001 **
<i>Pseudomonas aeruginosa</i>	8	100	100	50	80	50	20	60	80	80	80	0.0001 **
<i>Enterobacter</i>	4	100	80	50	70	20	0	50	40	60	80	0.0001 **
<i>Klebsiella oxytoca</i>	5	100	100	50	80	20	0	20	40	40	40	0.0001 **
<i>Klebsiella pneumoniae</i>	4	100	100	60	80	60	40	60	80	80	60	0.0001 **
<i>Staphylococcus aureus</i>	2	100	100	50	80	40	20	60	80	80	80	0.0001 **
<i>Salmonella</i>	2	100	100	50	70	40	20	40	80	60	40	0.0001 **
<i>Acinetobacter</i>	2	100	100	50	60	60	40	60	60	80	60	0.0001 **
P-value		1.00 NS	0.027 *	0.0085 **	0.0079 **	0.0001 **	0.0001 **	0.0001 **	0.0001 **	0.0001 **	0.0001 **	---
** (P≤0.01).												

****Highly Significant (P≤0.01).****Table (5):** Resistance of aquatic bacterial isolates to antibiotics, High resistance to AMP and AMC and low resistance to CIP observed across all isolates and

Antibiotic disc	Sensitive	Intermediate	Resistance	Chi-Square
AMP	4 (10%)	6 (15%)	30 (75%)	15.02 **
AMC	16 (40%)	2 (5%)	22 (55%)	9.75 **
CTX	16 (40%)	6 (15%)	18 (45%)	9.37 **
HLS	10 (15%)	12 (30%)	18 (45%)	8.45 **
C	16 (40%)	12 (30%)	12 (30%)	4.62 *
TR	19 (47.5%)	4 (10%)	17 (42.5%)	10.62 **
CIP	21 (52.5%)	14 (35%)	5 (12.5%)	10.89 **
K	9 (22.5%)	14 (35%)	17 (45%)	9.31 **
AK	10 (25%)	12 (30%)	14 (45%)	8.50 **
O	12 (30%)	14 (35%)	14 (45%)	4.19 *
Chi-Square	11.983 **	6.702 *	16.502 **	---

****Highly Significant (P≤0.01).**

4- Detection of biofilm formation

1-Congo red agar method: The result show that out of the ten most multidrug resistance bacterial isolates form different bacterial species (three isolates of *E. coli* , two *P. aeruginosa*, two *K. pneumoniae*, two *Staph. aureus*, and one *Salmonella*) three of them (30%) were weak, 5 of them (50%) were moderate and two (20%) were strong in adhesion test. These result according to the appearance of isolates on Congo red agar. The positive result was appeared as dark black colonies with a dry crystalline consistency as showed in Figure 5.

2-Adhesion to smooth surfaces: The result show that 2 (20%) of most resistance isolates to antibiotics were weak (+), 6 (60%) were moderate (++) and 2 (20%) *E. coli* isolates were strong (+++) in adhesion on smooth surfaces.

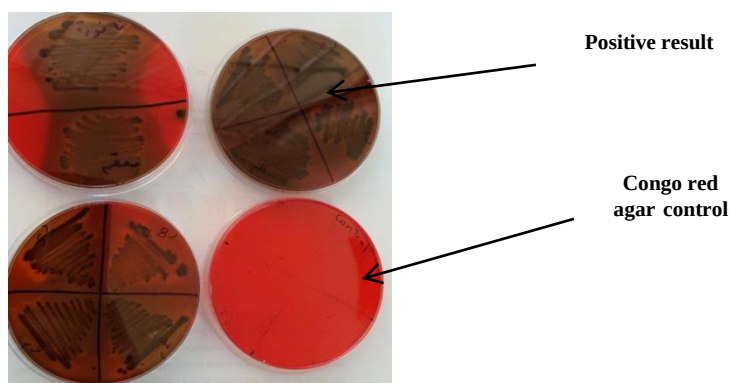


Figure (3): Congo red method show the slime layer production. Positive results were indicated by dry black colonies, moderate producers exhibited a gray coloration, while weak slime producers typically appeared pink

DISCUSSION

Isolating environmental bacteria from various locations and tap water in Baghdad, Iraq, was one of the study's goals. Several reports have already been made regarding the prevalence of pathogenic microorganisms, the diseases they cause, and their resistance to antibiotics that are of clinical interest. The highest isolation percentage was to *E. coli* and *P. aeruginosa* by using biochemical and chromogenic agar methods. Water is a vital nutrient that significantly contributes to digestion, nutrient absorption, and the excretion of waste through urine (32). Freshwater bodies are contaminated by various contaminants due to the rising in industrialization, human population, and urbanization. The application of biochemical tests in the identification of bacterial isolates is conventional. To identify pathogenic species or groups based on their enzymatic activity, chromogenic media use synthetic chromogenic enzyme substrates. This technology serves as an effective instrument for detecting specific microorganisms, even in mixed culture environments, enabling the user to achieve phenotypic detection based on the microorganism's expressed pattern, thereby facilitating final identification without sub- culturing and reducing the number of colonies to test, resulting in quicker and more efficient outcomes. Accessible readability and economies of scale: Economical and expedited approach (22). Numerous local investigations in Baghdad have recorded the isolation rates of bacterial species from various water samples. Hasan and Aburesha (2021) (33) reported that the isolation rate of *K. pneumoniae* was 18%, with 27 isolates obtained from 150 distinct water samples in Baghdad city. Mohammed and Flayyah (2018) (34) documented that out of 65 water samples collected from various surface locations in Baghdad city, 26 were identified, with 20 classified as *K. pneumoniae*, 4/26 (15.4%) identified as *Enterobacter cloacae*, and 1/26 (3.8%) identified as *K. oxytoca*.

A total of ten antibiotics from various classes were utilized, the majority of examined organisms were responsive to eight antibiotics but resistant to ampicillin and amoxyclave. Ciprofloxacin showed the lowest resistance, suggesting it may still be effective, and the differences were highly significant ($P \leq 0.01$). This high resistance to AMP and AMC may be due to the overuse of these antibiotics in Iraq. The resistance percentage was 100 % for amoxicalv and zero for the meropenem, imipenem, ceftazidime, cefotaxime and ceftriaxone for *K. pneumoniae* isolated from different water samples (33). In addition to being susceptible to streptomycin and ciprofloxacin, a significant percentage of the environmental isolates exhibited resistance to erythromycin, trimethoprim, and amoxicillin. Additionally, resistance to tetracycline, trimethoprim, and cephalothin was noted, and isolates from municipally treated tap and raw water samples also showed resistance to *Aeromonas* and *Pseudomonas* (16). Additionally, freshwater *Aeromonas* isolates exhibited ampicillin resistance (35).

Certain bacteria have the ability to adhere to surfaces by forming a slime layer (extracellular polymeric substances, or EPS), which is the fundamental building block of biofilms. Our result show that the production of these slime layers was varies among antibiotics resistance bacterial isolates from water samples. The variation in biofilm formation may be due to the small sample size ($n=40$ isolates) and potential biases (e.g., seasonal variation in sampling). Also, low flow rate or water stagnation and availability of nutrients that are favor in the formation of biofilm. This, in turn, makes it easier for harmful bacteria to spread. People can come into contact with these bacteria via different ways, direct contact, swimming and this water borne bacteria may also be conduct to the medical devises and hospitals when non-sufficient cleaning and filtration method used. Therefore, hospitals must resort to manual water filtration using membrane filters, especially for immunocompromised patients that may be stop some waterborne bacteria like *P. aeruginosa*.

The organism's attachment to the host surface is a crucial initial stage in the colonization process. Biofilms are linked to decreased susceptibility to antimicrobial treatment, and they may serve as a natural barrier shielding bacteria from such treatments (36). The finding are agree with other studies done by Koskeroglu *et al.* in 2023 (37), according to their study the biofilm formation by Congo red agar method, Eight isolates (53.3%) had moderate biofilm ability, while seven (46.6%) had negative biofilm ability. However, 53.3% of the isolates also had weak biofilm found by the microplate method, which supports the presence of multidrug-resistant bacteria with ESBL genes in water sources and suggests that these bacteria can spread to different environments and become a growing public health concern. Also a local study done by Al-Maeni (38) showed that there is a powerful correlation between antibiotic resistance and construction of biofilm.

CONCLUSION

This study has revealed that the analyzed water sources are polluted with various bacterial species. Bacteria are predominantly found in all environments affected by humans, with contamination sources arising from both human and animal pathogens. *E. coli* and *P. aeruginosa* were the most prevalent bacterial species found in diverse water samples, succeeded by *S. aureus*, *K. pneumoniae*, *K. oxytoca*, *Enterobacter*, *Acinetobacter*, and *Salmonella*, respectively. They were highly resist to AMP and AMC, while Ciprofloxacin showed the lowest resistance, suggesting it may still be effective. Their antibiotic resistance is concerning due to the potential contamination of the community with resistant bacteria, which raises alarms, particularly regarding the proliferation of MDR bacteria. Determining the significance of antibiotic resistance in waters for public health is essential given the rise in antimicrobial resistance. The result of biofilm formation indicate that the most multidrug resistance bacteria isolated from different water samples vary in slime layer production that may effected by the location the nutrients and the physical and chemical factors of water samples. This study sought to enhance awareness of bacterial contamination in local aquatic ecosystems and to investigate alternative water sources in Iraq, categorizing them as acceptable or unacceptable for human activities including drinking, fishing, swimming, and bathing. To better understand the risk, describe the issue, and create solutions to fight waterborne bacteria that are resistant to antibiotics, urgent need for improved water treatment to reduce MDR bacteria, and further research were needed (e.g., longitudinal studies to track resistance trends).

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انتشار المقاومة للمضادات وتكوين الغشاء الحيوي لأنواع البكتيريا المعزولة من عينات مياه من بغداد ، العراق

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الخلاصة

خلفية البحث: صحتنا العالمية مهددة بشكل خطير بسبب سرعه ظهور وانتشار البكتيريا المقاومة للمضادات الحيوية الناتج من سوء استخدام المضادات الحيوية مما ادى انتشار مسببات الأمراض المقاومة للأدوية المتعددة والذي بدوره يؤدي الى تقليل من فعالية العلاجات. وتعمل مصادر المياه كمستودعات مهمة لانتقال البكتيريا المقاومة للمضادات الحيوية. **الهدف من البحث:** تهدف هذه الدراسة الى عزل وتحديد الأنواع البكتيرية من مصادر مائية متنوعة في بغداد، وتقييم خصائص مقاومتها للمضادات الحيوية، والكشف عن تكوين الأغشية الحيوية لأكثر الأنواع البكتيرية المقاومة للأدوية المتعددة لإيجاد العلاقة بين إنتاج الطبقة للزجة ومقاومة المضادات الحيوية. **المواد و طرق العمل:** أجرينا تحليلاً للأنواع البكتيرية في عينات المياه التي تم الحصول عليها من 80 موقعاً متنوعاً في الفترة ما بين يوليو 2023 وأبريل 2024، باستخدام اكار مولد اللون، والاختبارات البيوكيميائية، ونظام API. تم إجراء اختبار الحساسية للمضادات الحيوية لل عزلات البكتيرية باستخدام طريقة كيربي باور لعشرة أنواع من المضادات الحيوية من مجموعات مختلفة، بما في ذلك الأمبيسلين، الأموكسيسلاف، السيفوتاكسيم، الأميكاسين، الأوكسيتتراسيكلين، الستربتوميسين، التريميثوبريم، الكاناميسين، السيروفلوكساسين، والكلورامفينيكول. وتم تقييم إنتاج الأغشية الحيوية في البكتيريا الأكثر مقاومة للأدوية المتعددة باستخدام طريقتين: أكار الكونغو الأحمر والالتصاق بالاسطح الملساء. **النتائج:** تشير النتائج إلى نسبة عزل 40 عذلة بكتيرية، موزعة على عينات المياه على النحو التالي: 18 (45%) من الأنهار، 13 (32.5%) من المياه الراكدة، 5 (12.5%) من المياه العكرة، 4 (10%). من ماء الخزان، وصفر من ماء الصنبور. تشمل الأنواع البكتيرية الأولية عزلات الإشريكية القولونية، والزائفة الزنجارية، والأمعائية، والكلبسيلا الأوكسيتوكا، والكلبسيلا الرئوية، والمكورات العنقودية الذهبية، والمكورات العنقودية الجلدية، والراكدة، والسالمونيلا. الاجناس الاكثر شوعا هي الزوائف الزنجارية والاشريكية القولونية. أبدت غالبية العزلات مقاومة للأمبيسلين والاموكسيسلايف ثم الستربتوميسين، والكاناميسين و الاميكاسين والاكساسيلين ثم السيفوتاكسيم و الكلورامفينيكول والترابيمومثريم ثم السبرودار الذي اظهر اقل درجة مقاومه وعزلات الزوائف الزنجارية والاشريكية القولونية كانت اكثر مقاومه للامبيسلين والاموكسيسلايف. تشير النتائج إلى التباين في إنتاج الطبقة الغروية بين العزلات الأكثر مقاومة للمضادات. **الاستنتاجات:** كشفت النتائج عن وجود أنواع بكتيرية متعددة في مياه بغداد، والتي تظهر قابليتها على المقاومة للمضادات الحيوية المختلفة بينما تظهر مقاومة للأمبيسلين، التتراسيكلين، والكاناميسين، وتختلف في إنتاج الطبقات الغروية. تزيد النتائج في زيادة الوعي الصحي العام بالمحتوى البكتيري والأنواع المقاومة للمضادات الحيوية والمكونه للطبقات للزجة في البيئات المائية .

الكلمات المفتاحية: بكتيريا معزولة من المياه ، بكتيريا مقاومة للمضادات ، اكار مولد اللون ، تكوين الغشاء الحيوي.