

A SURVEY STUDY OF SOME CHEMICAL CHARACTERISTICS AND PATHOGENIC BACTERIA OF THE EUPHRATES RIVER BASIN IN THE RAMADI CITY

Abdullah Abdul Al-Jaleel

Al-maarif University College,

Medical Laboratory Techniques, Al-anbar, Iraq

*abdullah.abduljaleel@uoa.edu.iq

Mohammed Shukry Nuaman

Al-maarif University College,

Medical Laboratory Techniques, Al-anbar, Iraq

Abstract

The present study was conducted along the bank of Euphrates river in Ramadi city in order to determine the concentration PH and electrical conductivity as well as diagnosing the type of pathogenic in the river water as well as circulatory system -five sites were identified before and after Ramadi hospital , as well asthree stations in the regulators , after which PH measurement andelectrical conductivity of the samples were taken, as well asbacteria were isolated and diagnosed.

The study showed the following results, the PH , rate was 7.7 , aswell as the electrical conductivity rate 1498, as well as two typesof pathogenic bacteria belonging to the Aeromonadaceae family which are the bacteria Aeromonas hydrophila and they Represented %99 percent and the bacteria Aeromonas caviae sufficient and represented the Proportion of %1 percent .

Introduction

Industrial and agricultural activities near water bodies for food Watch the pollutant rivers that turn into the most polluted It poses a great danger to the environment and human health. It is one of the most important chemicalpollutants Which The pollution of rivers is the acid and alkaline compounds, as the water is affected by what factories put out Chemicals ofa highly acidic or alkaline nature, such as fertilizer plants and oil refineriesPlants producing vegetable oils and many small factories overlooking the rivers. It leads to a change in the permissible pH and alkalinity ranges. and thus increase Organic matter in the water, which leads to an increase in the activity of microorganisms as pathogenic bacteria. As well as an increase in salinity in rivers, which affects the growth of Microorganisms, as well as human health by becoming water Not fit for drinking. As well as a change in the temperature of the water when the suspended matter increases Organic and inorganic, thus throwing waste water and factory cooling water an imbalance occurs in the ecological balance of river water and water bodies, as well as through Reducing the amounts of dissolved oxygen and the occurrence of metabolic processes and an increase in reactions Chemical, affecting various forms of life in the water

Objectives of the Study

- 1- Conducting chemical tests for the Euphrates River water in the city of Ramadi to determine the Acid properties (PH) and water salinity Ec
- 2- Conducting biological tests of the Euphrates River to identify some types of bacteria.

MATERIAL AND METHODS

Study Area

All the sites shown in figure 1 are in the Al-anbar government in the west of Iraq. Sites (1) before Ramadi General Hospital; (2) after Ramadi General Hospital; (3) Nazim Al-Warar/Wastewater flowing into the Nazim Al-Warr at the Ramadi Corniche; (4) Nazim Alwarar near Al Warar Bridge (Al Warar Dam); and (5) Ramadi Corniche near Al-Qasim Bridge.

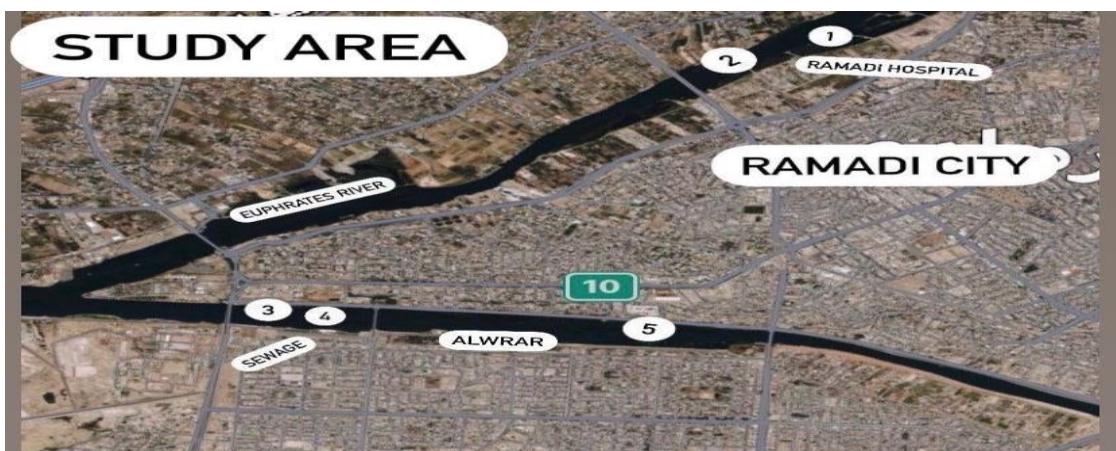


Figure 1. study sites at Euphrates River , Ramadi City.(Google Earth)

Samples Collection

We collected 5 Samples from the Euphrates River and Nazim Al-Warar . The samples were placed in plastic container. Samples were placed in the refrigerator for 24 hours. The research was conducted in the laboratories of the Department of medical laboratory techniques in Almaarif university college.



Figure 2. Site 1 Euphrates River before Ramadi General Hospital



Figure 3. Site 2 Euphrates River after Ramadi General Hospital



Figure 4. site 3 Nazim Al-Warar / Wastewater flowing into the Nazim Al-Warr at the Ramadi Corniche



Figure 5. site 4 Nazim Alwarar Near Al Warar Bridge (Al Warar Dam)



Figure 6. site 5 Ramadi Corniche Gray Cornis near Al-Qasim Bridge

Culture media

All media that used in the research shown in table 1.

Table 1. Culture media

culture medium	The amountg / L	Supplying company	Origin
Nutrient agar medium	23g / 100mlwater	Himedia	India
MaCconky agar medium	5.8 g /100mlwater	Biomark	India
Blood agar medium	4.5 g /100ml water	Himedia	India

Moist heat Sterilization

We used an autoclave. When sterilizing in this way, samples are placed into a steam chamber on a shelf or raised floor, and the chamber is closed and heated so that steam forces air out of the vents or exhausts. Pressure is then applied so that the interior temperature reaches 121 °C (250 °F), and this temperature is maintained for between 15 and 30 minutes. In this way, we sterilized the culture media that is not damaged by heat.

Cultivation of Samples

We make sure that the samples are not exposed to contamination after making sure we grow the samples on Petri dishes containing nutrient agar medium using a swap, and then we put the dishes in the incubator at a temperature of 37 °C for 24 hours.

Preparation the Nutrient agar medium. We choose the Nutrient agar medium Because it is a comprehensive and general medium for all bacteria.

Samples were taken and cultivated on Petri dishes containing MaCconky agar medium and Blood agar medium for the purpose of distinguishing between negative and positive bacteria for the purpose of identification them.

Identification of Samples

Cultural Characteristics

After growing the on the medium after incubating for 24 hours at a temperature of 37 ° C, the characteristics of the bacterial colonies were observed(color, shape, size).

Microscopic Characteristics

We used Gram stain to distinguish between positive and negative bacteria.

Gram stain

Gram stain has been used for staining samples and the Samples are placed on the slides With a drop of normal saline .

- 1- Crystal violet** Leave it on for (1) minute and then wash it off .
- 2- Iodine** Leave it on for (1) minute and then wash it off .
- 3- Alcohol** Leave it on for (10) Seconds and then wash it off .
- 4- Fuchsin solution** Leave it on for (1) minute and then wash it off .

Vitek Compact

The device was used to confirm the identification of bacterial isolates after conducting biochemical tests on them, the device was formed The device consists of a cassette holder and Reagent Cards containing 64 holes, each one representing the base material or medium for testing, and plastic tubes as well as a DensiChek device and an input and output unit.

Sensitivity Test

Preparation of the bacterial suspension. We take (2ml) from the normal saline and put it in a tube, and we take a bacterial smear by means of the sap, and then it is mixed, and then we take a smear from each tube and then put it in the dish (the culture medium) the antibiotics are Aztreonam, Ceftriaxone, Amoxicillin, Ampicillin and Chloramphenicol.

Chemical Examination

Chemical tests were carried out in Anbar Water Directorate (Quality Control Department). The necessary chemical tests were conducted on the five samples through the following devices and steps.

RESULTS AND DISCUSSION

Hydrogen Ion Concentration (PH)

PH is a significant environmental parameter which can be used in evaluating the acid_base balance of water (wlto, 2004) . it is important because many biological activities can occur only within a narrow range of PH . Therefore any variation beyond agreeable rang affected particular organism (Iqbal et al .2004) .

The values of PH in all sites are shown in figure (1) PH varied from (7.5 – 7.9) and the means value was (7.7) . The lowest value at site (1) while the highest value was recorded at site (5). The increase of PH in the river could be related to the groeth of aquatic plants and photosynthesis which exhausts CO₂ leads to arising in the values of PH (Yousry et al .2009) : This helps the growth of bacteria in the river water . This is in agreement with the fundings of the researchers (Al-Heety et al . 2011 · and Hussein Al-khafaji .2015) They found that the PH varied between (7.3-8) in the Euphrates River (figure 7).

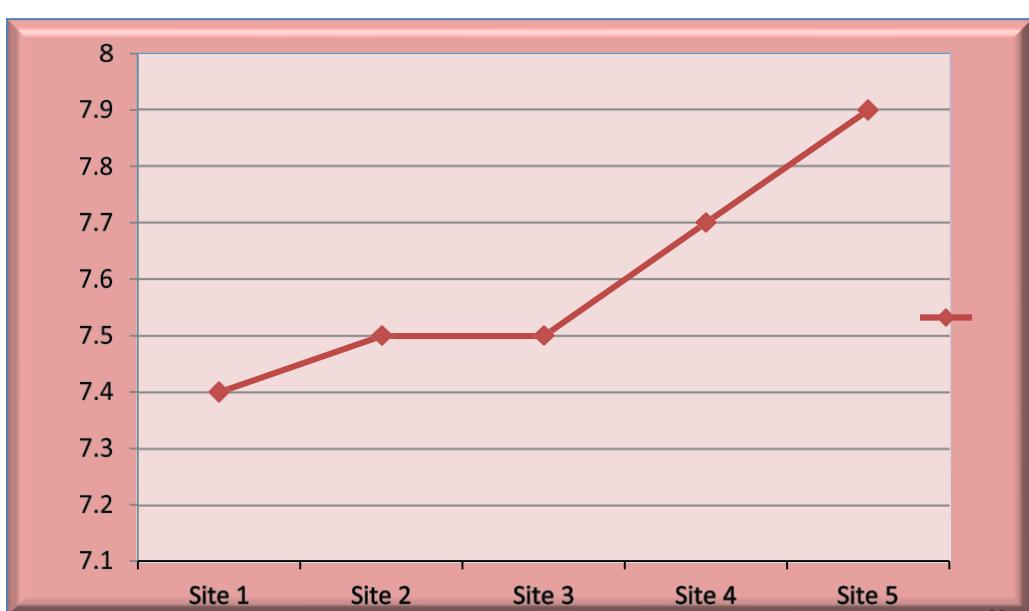


Figure 7 the values of pH during the study

Electrical Conductivity EC

Eelectrical Conductivity is related to the salt content dissolved in water it is a measure at the ability of water to carry electric current and it is sensitiveto variation in dissolved solids mostly mineral salts Usually the kinds of salt (Ion) which cause the conductivity are sodium magnesium calcium potassium chlorides carbonates and sulfate . (El – shakour and Mustafa 2012) the rang of electrical conductivity were (1169 – 2730) US \ CM the lowest conductivity values were recorded at site 2 and the highest values 2730 were recorded at site 3 natural water has low conductivity but will increase when the pollution exists the high values of electrical conductivity were recorded at site 3 due to the effect of sewage water andwaste water of household which

increase the EC and decrease in the rainfall this leads to an increase in the growth of plant and bacteria these results are in agreement with previous studies (AL – Mayah 2013 , Al – Heety et al . 2011) reported that increasing of the EC due to the strong effects of sewage water and decrease the rainfall on Euphrates Rivers (figure 8).

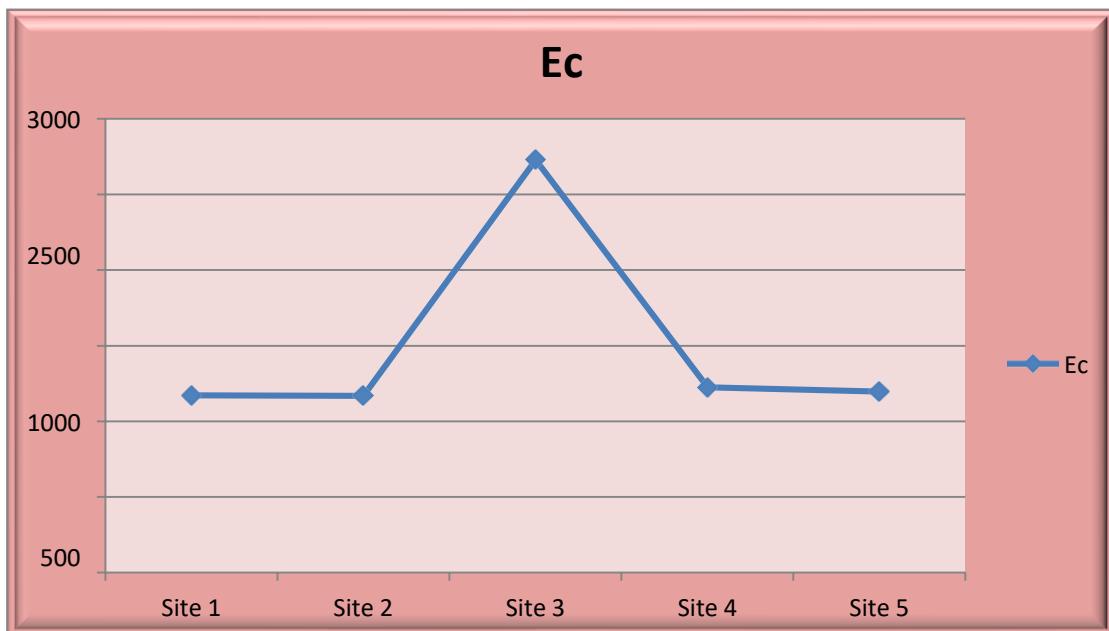


Figure 8 the values EC during the study .US

Bacteria

After performing phenotypic and biochemical tests of the isolates, Showed that there are types of isolated bacterial that were diagnosed in the waters of the Euphrates as shown in the table (2) bacteria belonging to the genus were isolated and identified Aeromonase spp are Aeromonas as hydrophilia bacteria and Aremononase cavine bacteria.

The reason for the wide spread of these types of genus Aeromononage Spp may be due to in the waters of rivers because it is endemic autochthonous in aquatic environments because they are moderately being facultative anaerobic, thermophilic as well as being which helps them tolerate anaerobic conditions in poorly ventilated locations (percival et al 2004) .

They are have the ability to move and may resist unfavorable conditions so it spreads quickly (Chenghesh et al 2007). If you conclude that these types of bacteria are prevalent in the Tigris River.

Aeromonas hydrophila are gram-negative, rod-shaped facultative anaerobes, & ranging in size from 0.3-1.0 μm wide by 1-0-3-5 μm -They are motile by a single polar flagellum. The bacteria can produce heat-labile enterotoxins, which can be associated with haemolysin and cytotoxin productions (Horneman and et al 2007, collier et al 1998, Burke et al).

Aeromonas ssp Free living, water is the main reservoirs It can also be found in mud, water, fresh produce, meat (beef, poultry, pork, Fish, shellfish, and shrimp) and dairy products (Hassan et al 2004). The infectious dose for humans and animals is greater than organisms (Rusin et al 1997 Kingombe et al 1999). Infection is spread via fecal-oral transmission during direct ingestion or drinking of contaminated water or foods (Udlch, 2004). Infection can also be transmitted eating con contaminated meat, dairy, shrimp, or fish (Horneman et al 2007) (figure 9).



Figure 9. Sample three after culture on Maconky agar medium that contains the Aeromonas

The tables 2,3, and 4 show the results of biochemical and the most bacteria that found in water and finally the type of bacteria.

Table 2 Biochemical diagnosis of bacterial isolates Biochemical Details

Hole number	Test name	result															
2	APPA	-	3	ADO	-	4	PyRL	-	5	IARL	-	7	dCEL	+	9	BGAI	+
10	H ₂ S	-	11	BNAG	-	12	AGLtp	+	13	dGLU	+	14	GGT	-	15	OFF	+
17	BGLU	+	18	dMAL	+	19	dMAN	-	20	dMNE	-	21	BXYL	-	22	BAlap	-
23	ProA	+	26	LIP	-	27	PLE	+	29	TyrA	+	31	URE	-	32	DsOR	-
33	SAC	+	34	dTAG	-	35	dTRE	-	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATK	+	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	pHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISe	-	56	CMT	+	57	BGur	-
58	O129R	+	59	GGAA	-	61	IMLTA	+	62	ELLM	+	64	ILATA	-			

Table 3 The percentage of bacteria isolated from watersamples

percentage	Number of samples	bacteria
99 %		Aeromonas hydrophila
1%	5	Aeromonas caviae

Table 4 Result of Gram stain on the five samples

sample	Results
Sample (1)	G+ Positive
Sample (2)	G+ Positive
Sample (3)	G-Negative
Sample (4)	G+ Positive
Sample (5)	G+ Positive

Conclusion

- 1 -The water of the Euphrates River tend to slightly alkaline
- 2 -The water of the Euphrates is low salinity.
- 3-The Euphrates river is pollinated with organic matter , plant and sewage.
- 4 -The water of the Euphrates river is contaminated with pathogenicbacteria that cause many human diseases .

References

1. Abed ,H.H.kadhim , Z.Y. Abed.S.M.Mahdi .Z.S. (2016) . Levels of bacterial and chemical pollutant in Euphrates river in samawah . Iraq . World EXP Biosei 4.66-69 .
2. Al-Heety , E.A. , Turky , A.M., and Al-Othman , E.N.(2011) . Physico- chemical assessment of Euphrates river Nile at Rosetta branch : impact ofdrain discharge . Middle-East Journal of scientific Research , 12(4) 413- 423 .
3. Al-khafaji B.Y. and Hussein H.M.(2015) . concentration of some trace element in water, sediment and phragmites australis (Cav) ,in Al-Diwanya river Journal of Thi-Qar science , 5(2) , 42-53. In Arabic
4. Al-Mayah , W.T.J.(2013). Effect of domestic sweage of water quality onAl-Garraf River of Al-Haay city – MSC . thesis . college of science . university of Baghdad .
5. Burke , V., Robinson ,J., Atkinson , H.M ., Dibley , M., Berry , R.J., and Gracey , M . (1981) . Exotoxins of Aeromonas hydrophila . The Australian Journal of Expermintal Biology and Medical science 59 (pt 6) , 753-761
6. Chen, K., Yu, X. Ma , K. you and Q.Jiang (2005) . The association between drinking water source and colorectal cancer incidence in Jiashan county of chiria : a prospectivecohort study . Eur.J.public Health .15 :652-656

7. Collier, L., Balows , A., and Sussman , M. (Ed) . (1998) . Microbiology and Microbial infection : systematic Bacteriology (9th ed) . USA
8. El-shakour , E-H-A. and Mostafa , A. (2012) water quality assessment of river Nile at Rosetta branch : impact of drain discharge . Middle-East Journal of scientific Research , 12(4) 413-423 .
9. Gardner ,L.R. and C. Gorman . 1984 . The summertime net transport of dissolved oxygen , salt and heat in salt marsh basin , North Inlet .S.C. Estuarine , coas , shelf sci 19:331-339 .
10. Hassan , V., and et al .(2004) . outbreak of Aeromonas hydrophila wound Infection Associated with Mud Football . Clinical Infection Diseases ., 38 , 1084-1089 .
11. Horneman , A.J., Ali , and Abbott ,S.L. (2007) . Aeromonas . in P.R. Murray , E.J. Baron M.L. Landry , J.H. Jorgensen and M.A pfaller (Eds) Manual of Clinical Microbiology (gth ed., pp . 715-722) Washington , D.C: ASM press.
12. Iqbal , F., Ali, M., Salam , A., khan , B.A, Ahmed, s., Qamar , M., and umer , K. (2004) . seasonal variation of physico-chemical characteristics of River soan water at Dhoak pathan Bridge (Chakwal) Pakistan .international Journal of Agriculture and Biology 6(1) , 89-92
13. Islam ,M.S, A. siddika , M.N.H.Khan.M.M Golder , M.A. Sadique , A.N.M.H Kabir , A. Huq and R.R Colwell- 2001 . Microbiological analysis of tube -well water in rural area of Bangladesh-*Appl .Environ . Microbial.*
14. Kingombe , c., and et al (1999) . PCR Detection , characterization , and Distribution of Virulence Genes in Aeromonas spp . *Applied and Environmental Microbiology* 65(12) , 5293-5302
15. Langston (2000) . Toxic Effects of metals and the incidence of metal pollution in marine Ecosystems in R.W Furness 8-P.C Rainbow.
16. Rusin .., P.A; and et al . (1997) . Risk assessment of opportunistic bacterial pathogens in drinking water . *Rev Environ contam Toxical ., 152*, 57-83 .
17. Udeh , P.J . (2004) . A Guide to Healthy Orinking water . USA : I universe Inc .
18. World Health organization – (2004) . Guidelines for drinking-water quality : recommendations (vol.I) world Health organization .
19. Yousry , M., El-sherbini ,A.,heikal , M., and salem , T . (2009) . suitability of water quality status of Rosetta branch for west Delta water conservation and irrigation rehabilitation project . *water science ,46*, 47-60.