

## THE DISTRIBUTION OF FECAL INDICATOR BACTERIA IN UMM QASR AND KHOR AL- ZUBAIR – BASRAH/ IRAQ

Asaad M. R. Al-Tae<sup>1</sup> Wesal F. Hassan<sup>2</sup>

<sup>1</sup>Department of Marine Environmental Chemistry, Marine Science Center, Basra, Iraq ([a.altaee@yahoo.com](mailto:a.altaee@yahoo.com)). <sup>2</sup>Department of Marine Environmental Chemistry, Marine Science Center, Basra, Iraq ([Wfhassan003@yahoo.com](mailto:Wfhassan003@yahoo.com))

### ABSTRACT

Maintenance of the microbiological quality of water has been used as an important means for preventing waterborne disease throughout the twentieth century. The commonest microbiological tests done on water are for coliforms and *Escherichia coli* (or faecal coliform). In this study 432 samples were taken for total and fecal coliforms and biological demand oxygen. The samples were collected at 12 stations from Khor Al-Zubair and Um Qasr port / Basrah- Iraq. From each site three samples were taken from different depths (surface, middle and bottom). The samples were collected in four periods at 6 hours during April- May 2009. Results indicated that the logarethmic no. of total coliforms ranged from (2.18- 2.84) CFU/100ml, while the logarethmic no. of fecal coliforms ranged from (0.86- 1.94) CFU/100ml. The biological oxygen demand (BOD<sub>5</sub>) values ranged from (16.47- 20.07) mg/L. These reults indicated that the recipient environment was polluted and poses a great concern.

### INTRODUCTION

Some bacteria, although naturally occurring, are known to cause diseases in humans, especially those with compromised immunity. For example, in the United States Gulf Coast areas, *Vibrio vulnificus* causes illness or even death in immunocompromised individuals who consume bacteria-contaminated shellfish [1,2, 3]. In coastal waters of New England in the United States, *V. parahaemolyticus*, another naturally occurring bacterium, has been implicated in leg gangrene and endotoxin shock in humans [4]. Several

types of disease causing viruses, protozoa, and bacteria are known to occur in sewage, human feces and fecally contaminated waters [5,6,7,8,9] Many of these pathogens (e.g., *V. cholerae*, *Salmonella spp.*, *Campylobacter jejuni*) originate directly from human and other warm-blooded animal sources, and are the causative agents of some of the most important waterborne diseases in the world, especially in developing countries where sanitation is generally poor and access to portable water is limited. *V. cholerae*, for example, occurs naturally in freshwater

and brackish waters [10,11,12] in associations with planktonic organisms, and is responsible for cholera epidemics and the associated deaths that have occurred in many countries [13,14,15].

The detection and enumeration of disease-causing organisms in surface waters is difficult, time consuming, and expensive; and for many of the pathogens, methods for their routine monitoring and isolation are nonexistent or the costs for their isolation and enumeration are very prohibitive. It is also impossible and impractical to identify all the enteric pathogenic organisms present in the water at any particular time. Moreover, because of their low densities in surface waters, the absence of pathogenic organisms in tested water samples does not guarantee that the organisms are not present in the water from which samples were collected. It is therefore important to identify harmless organisms that could be used as predictors of the presence of pathogenic organisms in groundwater, surface waters, or drinking water after treatment [16].

Coliform bacteria are a collection of related microorganisms that live and reproduce in large numbers in human and animal intestines. They can also be found in areas where human and animal fecal matter may be present such as in soil, surface water, and plants. Though coliform bacteria may be relatively harmless, the presence of a group of fecal coliform in water which is known as *Escherichia coli* may cause diseases such as cholera, typhoid fever, dysentery etc. [17].

## MATERIALS AND METHODS

**Discription of Study Area:** Umm Qasr and Khor Al- Zubair are located between (40°51' – 47°53') East and (30°12' – 30°13') North as shown in Figure- 1. Umm Qasr is a port complex in southern Iraq. It stands on the canalized Khor AL-Zubair, part of the Khor Abd Allah which leads to the Arabian Gulf. It is a small fishing town located in the south of Iraq very close to the Kuwaiti border, used as a military port on a few occasions. The government of King Faisal II established a permanent port in Umm Qasr in 1950s (Iraq hopes for revival as an international highway, The Times, 15 May 1961). Umm Qasr was subsequently founded as a port in 1961 by the Iraqi ruler commander Abdul-Karim Qassem. It was intended to serve as Iraq's only "deep water" port, reducing the country's dependence on the disputed Shatt Al-Arab waterway that marks the boarder with Iran. The port facilities were built by a consortium of companies from West Germany, Sweden and Lebanon 7 with a railway line connecting it to Basra and Baghdad. The port opened for business in July 1967. Khor Al-Zubair is an extension of the Gulf waters in the lower reaches of Mesopotamia Figure-1. It has an approximate length of 42 km, a wide of 1 km at low tide, and an average depth of 10-20 m, During 1983 this water body was connected to an oligohaline marsh (Hor Al-Hammar) to the north by Shatt Al-Basra channel, changing the environment of the Khor from a hypersaline lagoon to an estuary one [18].

The lower (South) end is connected with Khor Abdullah. The topography of Khor Al-Zubair looks like a spindle with tapering ends, at the northern and southern ends. The northern end receives fresh water influx of average  $700\text{m}^3/\text{sec}$  throughout the tidal cycle. The current in the Khor is characterized by one directional through out the tidal cycle towards the southern end (Arabian Gulf), with a velocity exceeding  $2\text{m}/\text{sec}$  during ebb tide and  $0.66\text{ m}/\text{sec}$  in flood tide. At the Southern end, the water discharge reaches  $10000\text{ m}^3/\text{sec}$  with a velocity range of  $0.8\text{-}5.78\text{ m}/\text{sec}$  [19] with big tidal

range at Umm-Qasr reaching  $4.3\text{m}$ . Due to the low profile of the shore-line, the tidal flood penetrates the mudflats to a further distance, depending on the state of the tide, covering the halophytic vegetation.

**Sampling:** A total of 432 samples were collected from Khor Al-Zubair channel and Um Kaser in twelve sites, from each site three samples were taken from different depth (surface, middle and bottom). The samples were collected in four periods at 6 hours during April- May 2009(Fig. 1, Table 1).

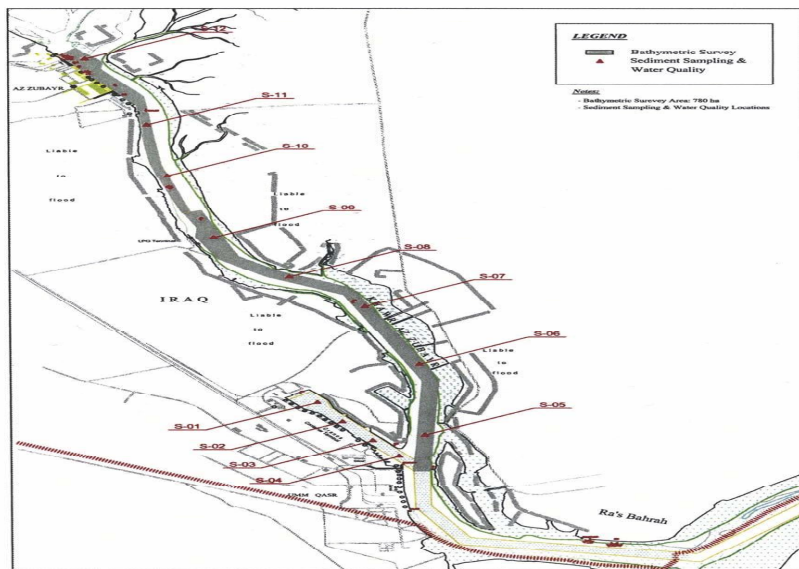


Figure- 1: Location of sampling sites for environmental survey.

Table - 1: Sampling sites, names and positions.

<b>Environmental Survey</b>			
<b>Sampling Point No.</b>	<b>Sampling Point Name</b>	<b>Latitude</b>	<b>Longitude</b>
S-01	Um Qasr	N 30 03 639	E 47 55 772
S-02		N 30 03 171	E 47 56 244
S-03		N 30 02 742	E 47 56 656
S-04		N 30 02 312	E 47 56 984
S-05	Khor AlZubair	N 30 03 275	E 47 57 410
S-06		N 30 04 686	E 47 57 210
S-07		N 30 05 650	E 47 56 735
S-08		N 30 06 805	E 47 55 502
S-09		N 30 07 579	E 47 54 588
S-10		N 30 09 056	E 47 54 046
S-11		N 30 10 600	E 47 53 871
S-12		N 30 12 258	E 4752812

**Bacteriological analysis:** The samples were collected in 250 ml sterile Nalgene polycarbonate conical flasks, and analyzed for total coliforms and fecal coliforms. The membrane filtration technique [20] was used by filtering two replicates of each sample through 0.45 $\mu$  pore size membrane filter (Sartorius, Germany).

The membrane filters were placed on the surface of M- endo agar contained in Petri dishes and incubated at  $37 \pm 1^\circ\text{C}$  for 24 hours. The coliform colonies will appear as pink to dark red spots with metallic (golden) sheen, which may vary in size from pinhead to complete colony coverage. While for fecal coliforms the membrane filters were placed on the surface of M-FC agar without rosolic acid

contained in Petri dishes and incubated at 44.5°C for 18 h in water bath. The colonies will appear as blue or light blue.

Water temperature and pH were measured at the sampling stations using a Multimeter (Multi 340i WTW Wissenschaftlich/Germny). The dissolved oxygen (DO) determined via oxygen sensitive membrane electrode [20]. The classic BOD assay is rather long and requires multiple steps. First, a given sample volume was introduced in a volumetric flask and made up to a fixed total volume with distilled water. The role of dilution is to ensure that there is a mass of oxygen sufficient to avoid any decline in bacterial activity. The flask can be shaken in order to ensure that the water is saturated with oxygen. To keep an appropriate (optimum) medium for development of the microorganisms, the pH of the sample should be between 6 and 8. A blank, i.e., dilution water was also prepared, and the same treatment as for the sample is applied. When necessary, both sample and blank must be seeded with a volume of water with microorganisms. A fraction of the total solution content was stored in a covered flask, avoiding the presence of air bubbles. The sample should be kept at 20° C and away from all light. Then DO was measured in both the diluted sample and blank, at the beginning and 5 days after the preparation.

## RESULTS

**Physicochemicals and Bacteriological Characteristics:** All physicochemicals and bacteriological data obtained during this study are summarized in Table 2. The

data recorded of water samples from stations S1, S2, S3, and S4 were located in Um Qasr, while the stations S5 to S12 were located in Khor Al-Zubair.

**Water Temperature:** Water temperatures at all stations ranged from 25.8°C at S1 to 27.56 at S12.

**pH:** The pH test measures the acidity of water body. The pH value of natural waters usually falls between 6.5 and 8.5 (if it is too low, too acidic, too high, too basic). In this study the pH value ranged from 7.93 at S1 to 8.12 at S5.

*Biological Parameters of Seawater*

**Biological Oxygen Demand:** A five-day biological oxygen demand (BOD5) concentration was measured regularly for the sea water samples from the study area. The highest concentration was 20.07 mg/L recorded at S5 and the lowest concentration was 16.47 mg/L at S11.

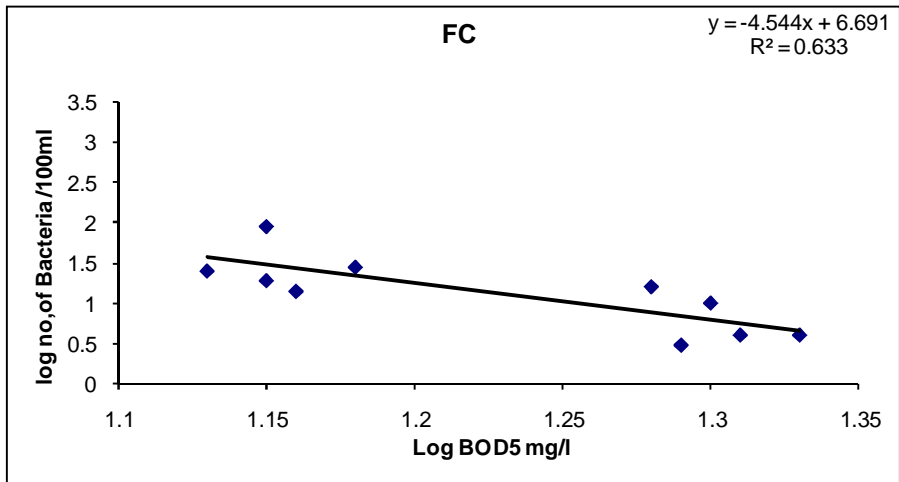
**Total and Fecal Coliform Bacteria:** The total and fecal coliform bacteria were measured using filtration technique. The highest logarithmic no. of TC and FC were 2.84/100ml and 1.94/ 100ml at S7 and S12, respectively. While the lowest logarithmic no. of TC and FC were 2.18 and 0.86 at S10 respectively.

**Relationship between indicator bacteria and BOD5:** Results in Figure-2 showed the statistical relationship between the concentration of indicator bacteria and BOD5. All of the indicator bacteria were correlated with BOD5 with correlation coefficients  $r$ , ranging from 0.635 for FC, 0.808 for TC and 0.813 to FC/TC at  $p < 0.05$ .

Table-2: Physiochemical and bacteriological data (mean  $\pm$  standard deviation) for water from Um Qasr and Khor Al- Zubair ports.

Stations		Temp (°C)	pH	BOD5	Log no. of Bacteria/100 ml		
				mg /L	TC	FC	FC/TC
<b>1</b>	Mean	25.82	7.93	19.18	2.31	1.34	0.61
	Std. Deviation	1.82	0.04	2.34	0.76	0.72	0.38
	N	24	24	24	23	22	22
<b>2</b>	Mean	25.80	7.93	17.16	2.35	1.56	0.72
	Std. Deviation	1.75	0.02	3.71	0.69	0.60	0.35
	N	24	24	24	23	19	18
<b>3</b>	Mean	25.83	7.94	17.23	2.36	1.50	0.73
	Std. Deviation	1.80	0.02	4.46	0.81	0.61	0.52
	N	24	24	24	24	21	21
<b>4</b>	Mean	25.85	7.94	17.15	2.42	1.37	0.67
	Std. Deviation	1.84	0.03	3.28	0.72	0.65	0.45
	N	24	24	24	24	22	22
<b>5</b>	Mean	26.07	8.12	20.07	2.38	1.59	0.70
	Std. Deviation	1.84	0.08	8.95	0.64	0.67	0.32
	N	24	24	24	24	21	21
<b>6</b>	Mean	26.07	8.09	17.05	2.69	1.26	0.51
	Std. Deviation	2.09	0.09	4.43	0.59	0.74	0.36
	N	24	24	24	24	19	19
<b>7</b>	Mean	26.10	8.11	18.78	2.84	1.10	0.41
	Std. Deviation	2.15	0.07	4.50	0.37	0.78	0.36
	N	24	24	24	24	18	18
<b>8</b>	Mean	26.60	8.09	17.72	2.63	1.04	0.49
	Std. Deviation	2.47	0.05	4.11	0.66	0.59	0.56
	N	24	24	24	24	21	21

<b>9</b>	Mean	26.70	8.10	17.76	2.61	1.09	0.42
	Std. Deviation	2.48	0.04	3.46	0.69	0.69	0.30
	N	24	24	24	24	18	18
<b>10</b>	Mean	27.10	8.02	17.33	2.18	0.86	0.57
	Std. Deviation	2.34	0.10	4.82	0.88	0.58	0.57
	N	24	24	24	24	21	21
<b>11</b>	Mean	27.49	7.94	16.47	2.79	1.25	0.44
	Std. Deviation	2.79	0.08	4.06	0.63	0.76	0.28
	N	24	24	24	24	21	20
<b>12</b>	Mean	27.56	7.95	20.02	2.64	1.94	0.78
	Std. Deviation	2.45	0.08	6.12	0.63	0.61	0.33
	N	24	24	24	23	20	20



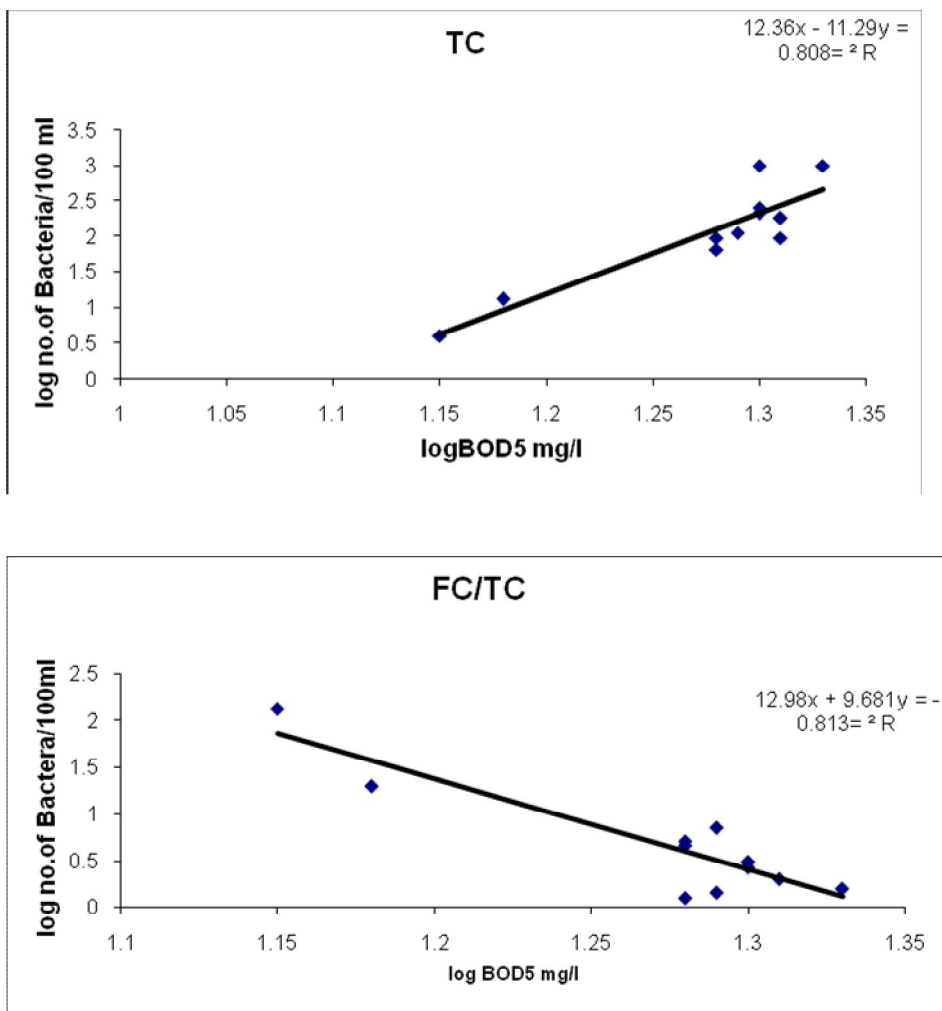


Figure – 2: Correlations between indicator bacterial concentrations and BOD5 in water sample



### **DISCUSSION**

The membrane filtration technique used is suitable for the determination of total and fecal coliforms in sea water. The advantage of this method is that to avoid the false-positive reading of anaerobic bacteria like *Clostridium perfringens*. Many of non-coliform bacteria will not grow on endo agar and m-FC agar and if grow can be excluded by special tests.

The result of this study demonstrated that localized pollution by pathogenic bacteria from waste and eutrophication effects can be severely along all sites and such conditions may prevail throughout the coastline, because the results of the total coliform and fecal coliform, recorded exceeds the permissible limit recommended by APHA, US EPA. The pollution resulted in damaging not only the landscape but also destroying the marine fauna and flora. The potential risk for these contaminants on public health and marine life are indeed of more attention at the national level.

Other factors playing a role in determining the number of bacteria, are the temperature especially high temperature in summer as it encourages the organisms to multiply and increase rapidly this is supported by experiment conducted by Carlucci and Pramer [21] who found that, the availability of nutrient, particularly phosphates, nitrates by marine bacteria which operates at high rate in summer and the ratio of die off rates of bacteria in the marine environment decreases. In addition there are factors of heavy tidal and flushing coursing waves

which accelerate the cleaning and rapid dispersion of wastes. Bacterial activity will exhaust the oxygen (decrease in dissolved in seawater) and increase the BOD.

The presence of total coliforms in seawater indicates only recent contamination by fecal materials and by soil in storm water runoff.

In conclusion, most of stations which studied were highly polluted with fecal materials and this phenomenon need for more intensive studies to determine the sources of pollution.

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