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ASSESSMENT OF WATER QUALITY DUE TO MICROBIAL GROWTH IN DRINKING WATER DISTRIBUTION SYSTEMS IN BASRAH CITY

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ABSTRACT

Fifty-three drinking water samples were collected from 13 districts and 3 water treatment plants around Basrah City, during March-September 1999. The samples were distributed in accordance with the water treatment plant supplying the consequent districts. The results obtained from the water treatment plants samples were acceptable from the micro-biological view point whereas 12 genera and 19 species of bacteria and two genera and three species of fungi, most of them are opportunistic pathogens for human were identified from the water distribution systems, rendering the water unfit for human consumption.

INTRODUCTION

Microbiological growth in drinking water can degrade water quality and become a major area of concern for affected utilities. The term biologically stable water has been proposed to describe potable water that does not promote the growth of micro- organisms during distribution (Rice, *et al.*, 1991). The purpose of a water distribution system is to supply the consumer with potable water and to ensure good hygienic sanitary condition (Augoustinos *et al.*, 1992).

Drinking water contamination, resulting from a range of sources, such as, infiltration, break through at the treatment works or from the biofilm established within the pipe-work. Recent changes in the safe drinking water acts a total coliform rule, in the U.S.A., require that all potable water samples positive for total coliforms be analysed for fecal coliforms or *Escherichia coli* (Rice *et al.*, 1993). The occurrence of *E. coli* is considered an acute risk of public health, because its presence is directly associated with fecal contamination.

The aims of the present study are as follows:

(i) To evalute different selective media for isolation of *E. coli* from drinking water.

(ii) To insure the quality of the final drinking water and the ability of it's use for human purposes.

(iii) To conduct an in-depth study on the variation of bacterial population in drinking water system in the city of Basrah.

MATERIALS AND METHODS

Fifty- three drinking water samples were collected from thirteen districts in Basrah city during the period March-Septemper, 1999. These districts were supplied mainly from Bradhaaya (site I). Maakl (site II) and Hartha (site III) stations.

The samples were collected in 250ml Nalgene polycarbonate conical flasks to which a solution of 2.5 % (w/v) sodium thiosulphate was added as a reducing agent for any residual chlorine. The flasks were autoclaved at 1.5 bar for 15 min at 121 C°. Samples were collected in accordance with Standard Methods for the Examination of water and Wastewater (APHA, 1989) and analyzed in the laboratory within 1-2 h. following collection.

Microbiological analysis: water samples were analyzed for total plate counts (TPC), total coliform (TC), fecal coliform (FC), *Pseudomonas aeruginosa* and some fungi. The membrane filtration technique (APHA, 1989) was used by filtering two replicates of each sample through Milipore WCN type filters (Whattman Corp., Japan).

Culture media used include plate count agar (PCA), eosin methylene blue agar (EMB), m-FC$_5$broth (Al-Sulami *et al.*, 1995), MacConkey agar (MA), MacConkey broth (MB). (McFeters *et al.*, 1982), voilet red bile agar (VRBA). S.S. agar and Pseudomonas agar F (PAF) for isolation of *Pseudomonas aeruginosa.*

A total of 185 colonies of bacteria and 6 colonies of fungi (isolated on S.S. agar) were picked randomly from the initial growth and examined by a morphological and biochemical as followes: Gram stains, oxidase reaction, IMViC tests, hydrogen sulphide production, phenylalanine deaminase, Urea hydroloysis, arginine hydrolysis, lysine and ornithine decarboxylasc, malonate utilization and fermentation of lactose, sucrose, D-mannital , salicin, D-sorbitol, L-arabinose, raffinose, maltose, D. xylose and D(+)-mannose. The fungi were identified in the mycology laboratory , college of Education, University of Basrah.

Their biochemical profiles were compared with tabulated data (Farmer and kelly, 1991 and Holt *et al.*, 1994).

Chemical / Physical analysis : Samples were collected in clean 250 ml Nalgene polycarbonate flasks. Parameters tested included: PH, salinity (PH meter, WTW, Germany), turbidity (turbidity meter, Hana, singapore) and hardness (Gesamtharte test, Art. Nr.10032), which determined in the laboratory at room temperature.

RESULTS

During the first month of the study the samples collected from the three stations had showed no bacterial growth. Accordingly water samples were then collected from the districts supplied by these stations separately, and within a distance range of 500-3500m from each station.

Data obtained from each district were handeled in reference to the relevant station.

The isolation of total bacteria in all districts which study (Table 1) were uncountable (UC) per 100 ml.

Total coliforms varied from 0-UC colony forming units (CFU) per 100 ml and were isolated from all districts using different media. Fecal coliforms were also 0-UC CFU per 100 ml from all districts.

The number of bacteria isolated on S.S. agar varied due to the site and incubation temperature, but in general, high numbers of bacteria were seen especially on site I and II. On PAF the number of bacteria varied from 560 CFU per 100 ml on site II to UC CFU per 100 ml on site I, while about 2950-UC CFU per 100 ml on site III.

Results of the chemical / physical parameters are summarized in Table (2). There were no differences in pH values between the three sites, but the salinity and turbidity were found to vary from less than 0.5 g/l to 1.6 g/l and from 2.16 FTU to 99.71 FTU respectively, while the hardness was more than 23 dH for all sites.

Identification of 160 isolates as presumptive entero- bacteriaceae (Table 3) on different types of media shown wide range of bacteria including *Escherichia coli* (6.25 %), *Enterobacter aggylomerans* (2.5 %), *E. taylorae* (2.5 %) , *E. gergoviae* (2.5 %), *E. asburiae* (1.87 %), *E. sakazaki* (1.25 %), *Klebseilla ornithinolytica* (3.75 %), *K. pneumoniae* (2.5 %) *Hafnia alvei* biogroup (3.12 %), *Proteus mirabilis* (1.25 %), *Rahnella aquatilis* (0.62 %), *Serratia tonticola* (1.87%); *S. iodonfera* biogroup 2 (2.5 %) *Citrobacter diversus* (1.25 %), *C. frundii* (1.25 %) , *Kluyvera ascorbata* (0.66 %),

Table 1: Comparison of counts of presumptive total and fecal coliform isolates from drinking water on different culture media.

Sample site	No.	PCA	m-FC5	MA		MB		EMB		VRBA		SS		PFA
			FC	TC	FC	TC	FC	TC	FC	TC	FC	35C	44.5C	
Site I	25	*UC	10-2770	10-UC	ND	16880	7440	UC	94	96-890	UC	0-UC	640-586	UC
Site II	13	UC	0-2380	290	160	ND	ND	ND	ND	0-10	ND	10	0-1390	560
Site III	15	UC	810-UC	+ND	300-UC	1360-UC	ND	UC	66	72-UC	1570-UC	UC	200-UC	2950-UC

* UC : Uncountable
+ ND : Not Done

Table 2: A summary of the chemical/ physical parameters of water samples collected from Basrah city.

Sample Site	No.	pH		Salinity ppt		Turbidity FTU		Hardness d H
		Mean	Range	Mean	Range	Mean	Range	
Site I	25	7.65	7.44-7.9	1.02	0.6-1.6	21.55	2.16-76	> 23
Site II	13	7.45	7.39-7.56	0.64	0.5-0.96	42.46	10.85-71	> 23
Site III	15	7.60	7.39-7.85	0.71	0.5-1.6	39.04	6.5-99.71	> 23

* d H : Germany degree 1 d H = 10 mg CaO/leter.

Table - Identification of bacteria isolates from drinking water using different types of media.

Species	No. of isolates						
	m-FC5	EMB	VRBA	MA	MB	SS	PAF
Escherichias coli	6	1	1	2			
Enterobacter agglomerans	3		1				
Ent. taylorae		2			2		
Ent. gergoviae				4			
Ent. asburiae						3	
Ent. sakazaki						2	
Klebseilla ornithinolytica	1	2	2		1		
K. pneumoniae				4			
Hafnia alvei biogroup 1		3				2	
Proteus mirabilis		2					
Rahnella aquatilis		1					
Serratia tonticola				3			
Serratia lodonfera biogroups 2						4	
Citrobacter divrsus						2	
C. frundii						2	
Kluyvera ascorbata						1	
Buttiauxella agrestis						2	
Budvicia aquatica						2	
Total *non-enteric bacteria*	6	10	15	13	12	8	
Pseudomonas euroginosa	4	5	12	8	6		15
Bacillus spp.							60
Aspergillus terrus						1	
A. flavus						3	
Penicillium sp.						2	

Buttiauxella agerstis (1.25 %), *Budvicia aquatica* (1.25 %) and non – enteric bacteria (40 %). While *Pseudomonas aeruginosa* was (21.86 %) as shown in (Fig.1).

The percentage of detection of *E. coli* on m-FC5 broth was 30 %, EMB 3.84 %, VRBA 3.22 % and MA 5.88 %. (Fig.2). While S.S. agar showed ability to enumerate fungi. Two genera were isolated *Aspergillus terrus, A. flavus* and *Penicillium* sp.

Seventy-five isolates on PAF were identified as presumptive pseudomonad, most of these organisms (80%) were later identified as members of the genus *Bacillus* being aerobic, Gram-positive, spore-forming bacilli. The rest (20 %) were identified as *Pseudomonas aeruginosa.*

DISCUSSION

From data obtained in the present study, there were a variety in the numbers and types of bacteria isolated from drinking water. These variations came apparently from the presence of bacteria in the distribution systems. Systems differed much in their size, the area that they serve the size of population and the distance the water is carried from its source. Differences in the size of the initial water works, the source of water and in the type of water work, may affect the final water quality (Augoustinos *et al.* 1992). Also the drinking water distribution system is often not a totally closed or sealed entity. Due to this, a possibility of external contamination exists. So the water distribution system should be designed to prevent external contamination. Previous studies (Geledreich *et al.*, 1992, Sartory and Holmes, 1997) had reported that the distribution systems may become contaminated from storm water run-off or sewage infiltration. Attachment of bacterial cells to a solid surface (biofilm) may provide a more favourable environment for bacterial cell growth (Allen, 1980; Touvinen *et al.*, 1980).

A living organism, such as bacterial cell, can produce material to change its surface properties, and the properties of material surface nearby (Lion *et al.*, 1988). According to Tamper (1987) there are several mechanisms or factors, which enable bacteria to attach to surfaces, including Van der Waal's forces, ionic binding, covelant coupling, cross – linking, ingel, microcapsules and fibers.

Sartory and Holmes (1997) noticed that the bacteria, which associated with distribution pipe biofilms were more protected than those in the bulk water and more resistant to chlorination. Also the type of material used in the system had affected the growth of microorganisms.

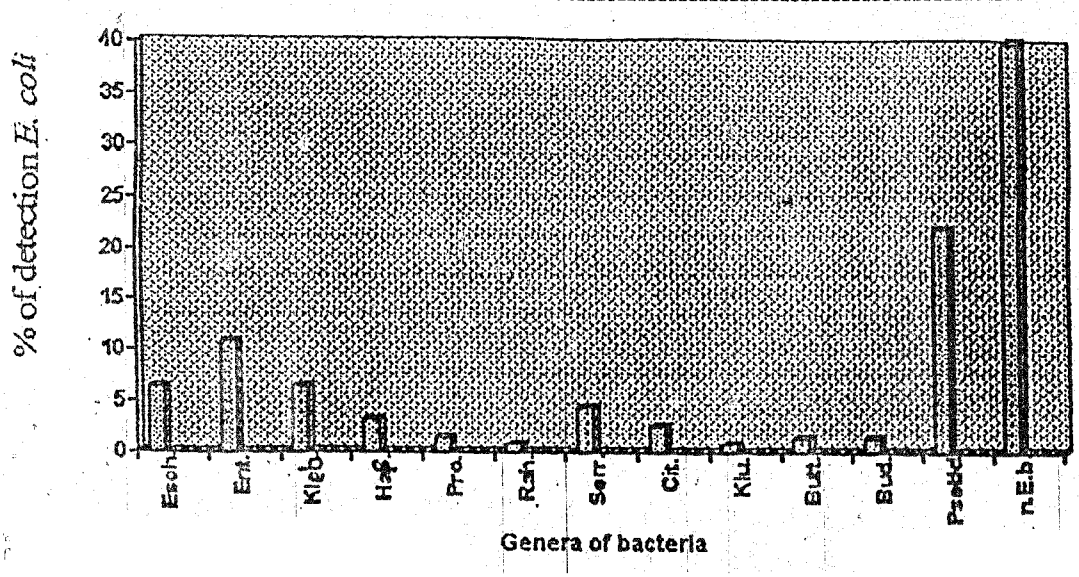

Fig. 1. Percentage of genera of bacteria isolated from drinking water.

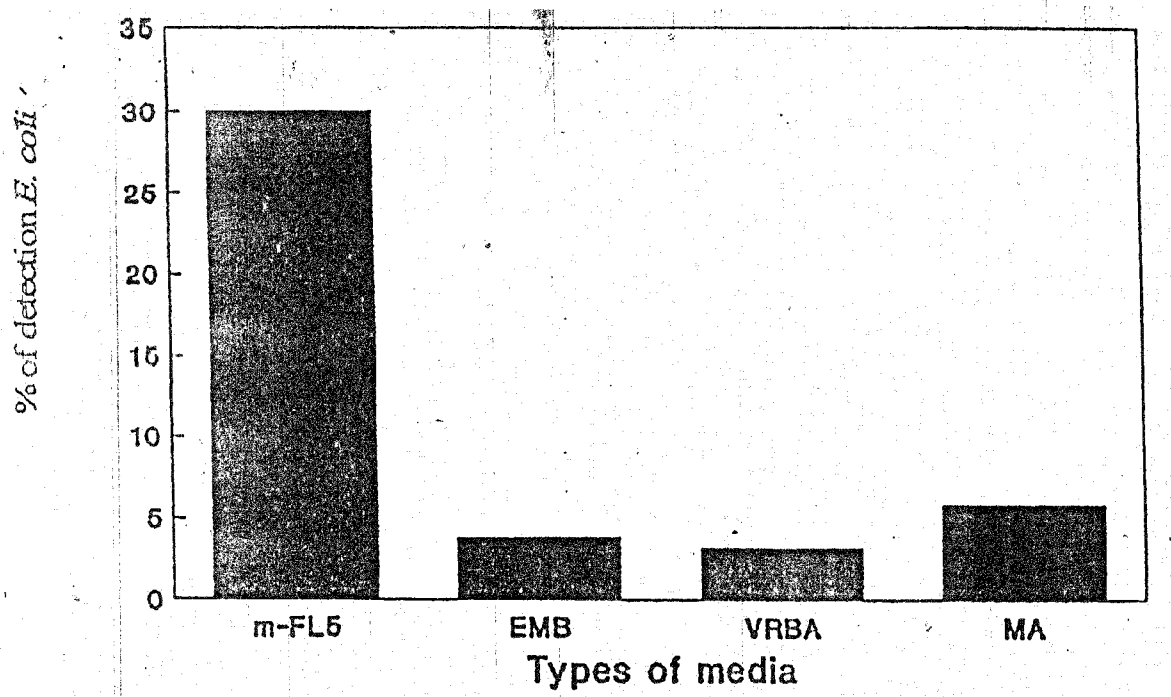

Fig. 2. Percentage of detection of E. coli using different type of media.

Augoustions *et al.* (1992) found that mild steel. PVC, and cement to be the best support for bacterial growth in water distribution system and copper the best inhibitor. Basrahs distribution system is a combing of the first three.

Physical and chemical parameters showed no differences in pH values between stations and the system, which indicate a delicate balance between bacterial growth in the distribution system. Martin *et al.* (1982) showed that, the increase in pH due to lime addition was highly effective in the elimination of coliform growth in the water distribution system.

There were variations in salinity values recorded at the three sites and this might be due to the difference, in water sources especially between site I and III, site I being the highest 1.02 g/l. Moreover, the turbidity level was generally high at all sites.

Turbidity has been used as a measure of water quality for many years. In the United State a limit of 1 NTU has been set for water leaving the water plant (EPA, 1976). In the present study all sites showed high values of turbidity and that usually is correlated with the presence of nutrients in the system which may result in microbial growth and deterioration of water quality (EPA, 1976).

Most workers (Herson *et al.*, 1984; LeChvalier *et al*, 1984. Ridgway and Olson, 1982), showed that, increased turbidity relates to high chlorine demand and decreased availability of residual disinfectant in the water distribution system. Moreover, they reported a close relationship between high levels of turbidity and increased bacterial growth. Increased turbidity means an availability of a matrix for the transport of microorganisms through the system or a way of introducing the microorganisms.

In the present study hardness was measured using a kit to determined the calcium oxide in water, all sites gave more than 23 dH (> 230 mg/l). The high percentage of calcium increases the hardness of water and release bad odor and its precipitation may cause blockages in the distribution net, encouraging bacterial growth on the inner surface of the pipes.

Large numbers of heterotrophic bacteria (table 3) were isolated from drinking water systems by using standard microbiological techniques. Many of these bacteria had been shown to be human secondary opportunistic pathogens (Augoustinos *et al.*, 1992). Moreover the growth of fungi and bacteria in water distribution systems had been shown to be of significance to human health. A part from direct health hazards, the growth of bacteria and fungi in the distribution system can introduce another problem, as their presence may mask other indicator organisms resulting from a real breakthrough of the treatment process.

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تقييم نوعية المياه اعتمادا" على النمو المايكروبي لمياه الشرب في شبكة التوزيع لمدينة البصرة

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الخلاصـــــة

جمعت 53 عينة من مياه الشرب من ثلاث عشر منطقة فـــي مدينـــة البصرة خلال الفترة من آذار–ايلول عام 1999. جدولت النتائج اعتماداً على ثلاث محطات رئيسية تغذي هذه المناطق. اظهرت نتائج الفحص الميكروبي وبعض الفحوصات الفيزياوية والكيمياوية ان المياه المجهزة الى المدينة ذات نوعية جيدة في وقت مغادرتـــها محطـــات التصفيـــة ولكـــن تتدنى نوعيتها خلال مرورها في شبكة التوزيع حيـــث وجـــد ارتفاع في معدلات البكتريا وقد عزل 12 جنســـاً و 19 نوعـــاً منـــها بالاضافة الى جنسين من الفطريات و ثلاث انواعاً منها . معظم هـــذه الاحياء المجهرية تعتبر مرضية انتهازية للانسان مما يجعل هذه المياه غير صالحة للاستخدامات البشرية في وقت وصولها الى المستهلك.