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**Email:** Umnashwa@yahoo.com

(أستلم 2018/ 12 /23 ؛ فُيل 2019/ 2 / 25)

(2017-2014)

40

:

<i>Escherichia coli</i>	21	56	
7 <i>Pseudomonas aeruginosa</i>		8 <i>Aeromonas hydrophila</i>	10
<i>Klebsiella pneumoniae</i>	<i>Klebsiella</i>	<i>Pseudomonas fluorescens</i>	
		4 6 <i>Klebsiella oxytoca</i>	
			100%
		<i>Aeromonas Pseudomonas</i>	

### **Contamination of Domestic Well Water in Nineveh Governorate with some Pathogenic Bacteria and Detection on its Ability to Biofilm Formation**

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#### **ABSTRACT**

Due to the unavailability of the drinking water in Mosul city in the period between (2014-2017), people started digging wells in their houses to use it in daily life for consumption and irrigation. Forty well water samples were chosen for this study in living quarters (AL-Dubat, Adan, AL-Mouthana, AL-Noor, AL-Zhoor, AL-Jamiea, AL-Falah, AL-Baker). Some pathogenic bacteria were isolated and identified from well water digged in different areas in the left side of Mosul city

and to study reconciliation to human consumption. Results showed that fifty-six bacterial isolates obtained included 21 of *Escherichia coli*, 10 of *Aeromonas hydrophila*, 8 of *Pseudomonas aeruginosa*, 7 of *Pseudomonas fluorescens*, and show two species of Klebsiella bacteria, *Klebsiella pneumonia* and *Klebsiella oxytoca* 6 and 4 respectively.

Then the ability of these bacterial isolates to form biofilm was studied by using two methods; Tube method and Congo red method, isolates gave 100% positive results in its ability to biofilm formation by tube method. While by congo red *Aeromonas* and *Pseudomons* were unable to form biofilm.

Also the study included determination of antibiotics sensitivity and resistance of bacterial isolates to different antibiotics as it is considered one of the important virulence factors , Results showed that there is variation in its sensitivity and resistance to these antibiotics.

**Keywords:** well water, bacterial contamination, biofilm.

.(OHA, 2015)

Dug Wells

(30-10)

Driven Wells

(400-100)

.(Waller, 2016)

(50-30)

%59

.(Maupin and Barber, 2005)

2

.(WHO, 2017)

%76

.(Guzman- Herradar *et al.*, 2015)

*Pseudomonas Escherichia coli Aeromonas hydrophila*

*Klebsiella oxytoca Klebsiella pneumoniae Pseudomonas fluorescens aeruginosa*

.(Forbes *et al.*, 2007)

2001

(1 )  
.2017

:1

WHO			
zero	1	cfu	( 35 24 100 )
zero	1	cfu	( 44 24 100) E.coli
zero	1	cfu	( 35 24 250) E.coli
zero	zero	cfu	( 35 24 1)

(2017-2014)

**Samples**

5 40 /  
: (2018/11/5) (2018/10/22)

**Culture media**

LAB, HIMEDIA, OXOID, :

(BACTO)

- . 15 121
- Nutrient agar
- MacConkey agar
- Nutrient broth
- Salmonella Shigella agar -
- (EMB) Eosine Methylene Blue agar
- Triple Sugar Iron agar
- Simmon Citrate agar
- Pepton Water
- (TSB)Tryptic Soy broth
- Muller Hinton agar
- (TCBS)Thiosulfate citrate bile salts sucrose agar

- Cetrimide agar
- Glucose Phosphate Pepton Water
- Semi Solid Nutrient agar
- Nutrient Gelatin
- Phenol Red agar
- Congo Red agar
- Blood agar
- Urea agar base

(Forbes *et al.*, 2002; Atlas, 2010; Tille, 2014)

(2 )

Bioanalyse

:2

/	/
30Mcg	(VA) Vancomycin
(20\10)Mcg	(AMC) Amoxicilin/Clavulanic acid
10Mcg	(TMP) Trimethoprim
10Mcg	(CN) Gentamicin
10Mcg	(AK) Amikacin
10U	(B) Bactiriacin

40

<sup>3</sup> 250

(5)

<sup>3</sup> 0.1

**Diagnostic Tests**

: :

**Gram's Stain**

(Finegold and Martine, 1982)

**Necrosin stain**

.(Finegold and Martine,1982)

**Biochemical Tests**

:

- Oxidase test •
- Catalase test •
- Indol test •
- Methyl Red test •
- Voges Pros Kauer test •
- Citrate Utilization test •
- Sugar fermentation test •
- Triple sugar iron agar test+ H<sub>2</sub>S •
- Gelatinase test •
- Urease enzyme test •
- Motility test •

(Forbes *et al.*, 2002; Koneman *et al.*, 2006; Hassan *et al.*, 2011)

**APIE20**

: APIE20

.Biomerieux

APIE20

:

**Tube method**

.1

Hassan *et al.*, (2011)

. 24 37

%1

TSB

5 %1

. 24 37

phosphate buffer saline

Deionized water

(Christensen *et al.*, 1982;

.Hassan *et al.*, 2011)

Congo red agar

.2

24 37  
24 37

(Freeman *et al.*, 1989; Oliveira and

Cunha, 2010; Hassan *et al.*, 2011).

Kirby-

(Bactiriacin (Vandepitte *et al.*, 2003) Baure(1966)  
(B), Vancomycin (VA), Amoxicilin/Clavulanic acid (AMC), Trimethoprim (TMP), Gentamicin  
: (CN), Amikacin (AK)

1.5×10<sup>8</sup> 24 37  
3 0.1  
L-Shape

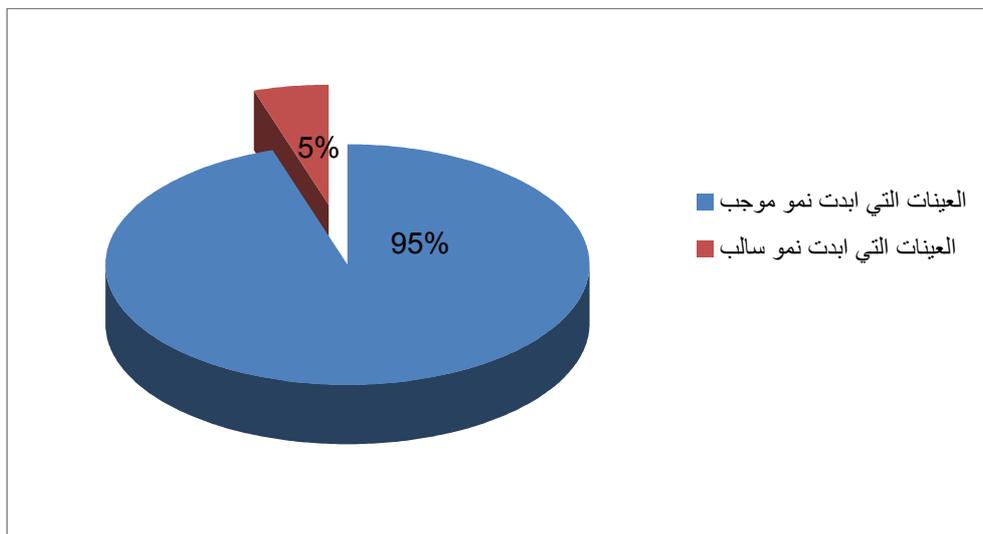
24 37

40

2 %95

38

%5



.....

(1)

5%

100%

(3 )

56

:3

<i>Klebsiella oxytoca , Aeromonas hydrophila , E. coli</i>	
<i>Klebsiella oxytoca, Klebsiella pneumoniae , Pseudomonas aeruginosa, E.coli</i>	
<i>Klebsiella pneumoniae , Pseudomonas fluorescens , Aeromonas hydrophila , E. coli</i>	
<i>Klebsiella oxytoca, Klebsiella pneumoniae , E. coli</i>	
<i>Klebsiella oxytoca, Klebsiella pneumoniae , Pseudomonas fluorescens , E. coli</i>	
<i>Klebsiella pneumoniae, Pseudomonas fluorescens</i>	
<i>Klebsiella pneumoniae, Pseudomonas aeruginosa, Aeromonas hydrophila, E. coli</i>	
<i>Pseudomonas aeruginosa, Aeromonas hydrophila , E. coli</i>	

: 4

37.5%	21	<i>E. coli</i>
17.9%	10	<i>Aeromonas hydrophila</i>
14.3%	8	<i>Pseudomonas aeruginosa</i>
12.5%	7	<i>Pseudomonas fluorescens</i>
10.7%	6	<i>Klebsiella pneumoniae</i>
7.1%	4	<i>Klebsiella oxytoca</i>

56

(4 )

10 *Aeromonas hydrophila* ( 37.5%) 21 *Escherichia Coli*  
 (14.3%) 8 *Pseudomonas aeruginosa* (17.9%)  
*Klebsiella* (12.5%) 7 *Pseudomonas fluorescens*  
 (7.1%) 4 *Klebsiella oxytoca* (10.7%) 6 *Klebsiella pneumoniae*  
*E.coli*

(Abdurrahman and Altahir, 2011)

(22.5%)

(U.S.EPA, 2015)

(UNESCO, 2012)

( Koneman *et al.*, 2006 )

*Escherichia coli*

EMB

(2)

(1)

APIE20

(3)



A

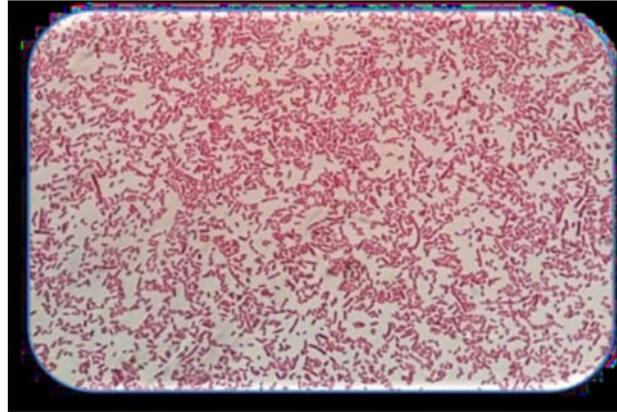
B

EMB :B

:A

*Escherichia coli*

:1



*Escherichia coli* :2



*Escherichia coli* API :3

*E.coli*

44

37

*Vebrio colerae*

2002

2002

) .

.(Atlas, 2010

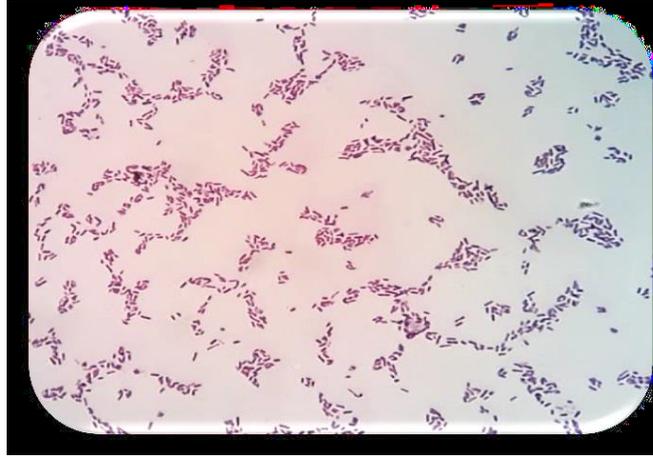
TCBS

*Aeromonas hydrophila*

.(4A)

(4B)





*Pseudomonas aeruginosa* :6

*Klebsiella oxytoca* *Klebsiella pneumoniae* *Klebsiella pneumoniae*  
 Endimiani *et al.*, (2009)

(5 )

:5

Glucose fermentation	Lactose fermentation	Motility	Urase	Gelatinase	Gas	H2S	Triple Sugar Iron Agar	Citrate Utilization	Voges Pros Kauer	Methyl Red	Indol	Catalase	Oxidase	العزلات
+	+	+	-	-	+	-	A/A	-	-	+	+	+	-	<i>Escherichia coli</i>
+	-	+	-	+	+	-	A/AK	-	-	+	+	+	+	<i>Aeromonas hydrophila</i>
-	-	+	V	+	-	-	AK/AK	+	-	-	-	+	+	<i>Pseudomonas aeruginosa</i>
-	-	+	V	+	-	-	AK/AK	+	-	-	-	+	+	<i>Pseudomonas fluorescens</i>
+	+	-	+	-	+	-	A/A	+	+	-	-	+	-	<i>Klebsiella pneumoniae</i>
+	+	-	+	-	+	-	A/A	+	+	-	+	+	-	<i>Klebsiella oxytoca</i>

:+

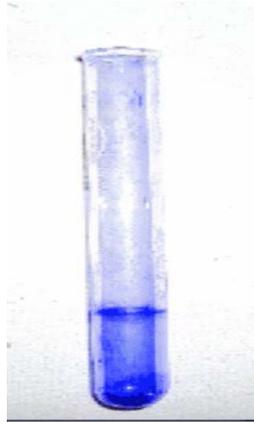
:-

:A

:AK

: V

(7)

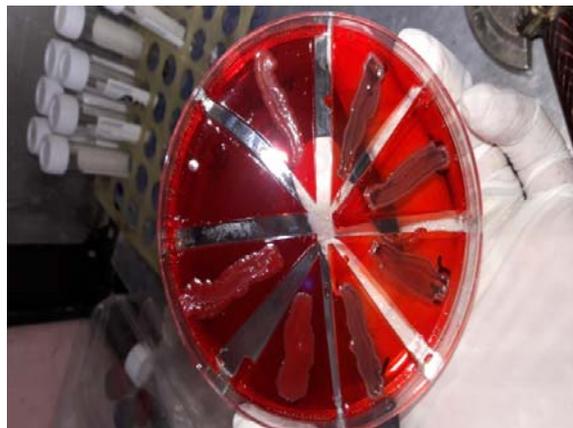


:7

(Christensen *et al.*, 1982 ; Hassan *et al.*, 2011)

(8)

(Oliveira and Cunha, 2010; Hassan *et al.*, 2011)



:8

(6 )

%100

Klebsiella

Aeromonas

.....

*Pseudomonas aeruginosa*,

*Aeromonas hydrophila* , *Pseudomonas fluorescens*

:6

++	+++	<i>Klebsiella pneumoniae</i>
++	+++	<i>Klebsiella oxytoca</i>
+	++	<i>Escherichia coli</i>
-	++	<i>Pseudomonas aeruginosa</i>
-	+	<i>Pseudomonas fluorescens</i>
-	+	<i>Aeromonas hydrophila</i>

(6)

8

.(Kirby Bauer method)

(7)

:7

VA	AMC	TMP	AK	CN	B	
R	R	R	16.28	R	R	<i>E.coli</i>
R	R	R	12	11	15	<i>E.coli</i>
R	R	9.79	16.98	16.98	R	<i>E.coli</i>
R	9.94	R	13.26	17.8	R	<i>Klebsiella pneumoniae</i>
R	15.31	18.40	17.28	18.96	R	<i>Klebsiella oxytoca</i>
R	R	R	16.89	17.17	R	<i>Pseudomonas aeruginosa</i>
12.50	R	9.79	17.37	16.55	R	<i>Pseudomonas fluorescens</i>
15.2	R	17	20.16	19.8	R	<i>Aeromonas hydrophila</i>
75%	75%	50%	0%	12.5%	87.5%	

*E.coli*

(Vancomycin, Amoxilin/Clavulanic acid) %100

*Klebsiella Pneumoniae*

.(Bactiriacin, Trimethoprim, Gentamicin )

(Vancomycin, Trimethoprim, Bactiriacin)

(Vancomycin, Bactiriacin)

*Klebsiella Oxytoca*

( Bactiriacin, Vancomycin, Trimethoprim,

*Pseudomonas aeruginosa*

<i>Aeromonas</i>	<i>Pseudomonas fluorescens</i>	(Amoxilin/ Clavulanic acid)	
		(Amoxilin/Clavulanic acid, Bactiriacin)	<i>hydrophila</i>
	87.5%		
	.75%	(Amoxilin/Clavulanic acid, Vancomycin)	(Bactiriacin)
		.50%	(Trimethoprim)
		(Amikacin)	(Gentamicin)
			12.5%
	(7 )	100%	
<i>Aeromonas</i>	<i>Klebsiella oxytoca</i>		(10 9)
			<i>hydrophila</i>



:10

*Aeromonas hydrophila*

:9

*Klebsiella oxytoca*

.(2002)

.126-118 (5) 15 .

.(2002)

.19-12 (1)13 .

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