

Evaluation of C-Reactive Protein in Iraqi Children Presented with Acute Enteropathogenic *Escherichia Coli* Associated Diarrhea with Special Emphasis to Age and Gender

Akut Enteropatojenik *Escherichia Coli* ile İlişkili Diyarenin Yaşa ve Cinsiyete Özel Olarak Iraklı Çocuklarda C-Reaktif Proteinin Değerlendirilmesi

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ABSTRACT

Background and Objectives: The aim is to determine the role of Enteropathogenic *Escherichia coli* (EPEC) serotypes (O111, O55, O26, O86, O119, O127, O114, and O142) in C-reactive protein (CRP) level in acute diarrhea among children; to determine the possible effect of age and gender in CRP level.

Methods: Culture of stool samples from 64 children in MacConkey agar for primary isolation of *E.coli* and confirmed via API20E. Sero-grouping of *E.coli* performed via specific antisera. CRP detected by latex agglutination.

Results: In this study, 64 children with diarrhea were involved. The mean age was 2.92 years. Males represent 34/64, 53.1%. The range of CRP level 12-96 mg/dl. EPEC1 and EPEC3 not detected. EPEC2 detected among 14/64, 21.9% and EPEC4 in 18.8%. Among the age group <1 year, EPEC2 detected in 9.38%, with mean CRP level 12mg/dl; 3.13%, 48 mg/dl 6.25%. EPEC4 detected in 2/64, 3.13%, with mean CRP level 12mg/dl. Age group 1-4 year, EPEC2 detected in 6/64, 9.38%, with mean CRP level 24mg/dl. EPEC4 detected in 6/64, 9.38%; with mean CRP level 12mg/dl, 24mg/dl, 48mg/dl 3.13% in each. Among the age group 4.1-8 year, EPEC4 detected in 4/64, 6.25%, with CRP mean level 48 mg/dl. EPEC not detected in 8> years age group. EPEC2 detected in male 28/64, 43.75% and the CRP level range from 12-96mg/dl. EPEC2 detected in 22/64 females, 34.38%, CRP level range from 12-96mg/dl. No significant difference or correlation between gender and CRP level detected. EPEC4 detected in males 26/64, 40.63%, CRP level range from 12-96mg/dl. EPEC4 detected in 26/64 females, 40.63%, CRP level range from 12-96mg/dl. No significant difference or correlation between gender and CRP level was detected. EPEC2 and CRP level significantly correlated with patients age. while infection with EPEC2 inversely correlated EPEC4. No significant correlation between gender and EPEC serotypes.

Conclusion: Although EPEC2 and EPEC4 frequently detected in diarrhea cases of 1-8 years old, only EPEC2 positively correlated with age. Even with a fluctuation in CRP level, it was positively correlated with age and not correlated with EPEC serotypes and gender.

Key Words: EPEC, age, gender, CRP

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ÖZET

Amaç: Amaç, çocuklarda akut diyarede C-reaktif protein (CRP) seviyesinde Enteropatojenik *Escherichia coli* (EPEC) serotiplerinin (O111, O55, O26, O86, O119, O127, O114 ve O142) rolünü; CRP düzeyine yaş ve cinsiyetin olası etkisini saptamaktır.

Yöntem: MacConkey agarda 64 çocuktan *E.coli*'nin primer izolasyonu için dışkı örneklerinin kültürü yapılmış ve API20E ile doğrulanmıştır. Belirli antiserumlarla *E.coli* sero grupları yapılmıştır. Latex aglütinasyonu ile CRP saptandı.

Bulgular: Bu çalışmaya 64 diyaresi olan çocuklar dahil edildi. Ortalama yaş 2.92 yıl idi. Erkekler 34/64,% 53.1'ini temsil ediyordu. CRP seviyesi 12-96mg / dl aralığında saptandı. EPEC1 ve EPEC3 saptanmadı. EPEC2 14/64,%21,9 oranında ve EPEC4 ise % 18,8 olarak tespit edildi. <1 yıl yaş grubunda EPEC2 % 9.38 oranda saptandı; % 3.13'ünde ortalama CRP seviyesi 12mg / dl; % 6.25'inde ise 48 mg / dl olarak saptandı. EPEC4 2/64, % 3.13 oranda, ortalama CRP seviyesi 12mg / dl tespit edildi. Yaş grubu 1-4 yıl arasında, EPEC2 6/64, % 9.38, ortalama CRP seviyesi 24mg/dl tespit edildi. EPEC4, 6/64, % 9.38; ortalama CRP seviyesi 12mg/dl, 24mg/dl, 48mg/dl olmak üzere herbirinde %3.13 olarak saptandı. 4.1-8 yaş grubunda, EPEC4 4/64, % 6.25, CRP ortalama seviyesi 48 mg/dl tespit edildi. 8 yaş üzeri grupta EPEC saptanmadı. 28/64 erkekte EPEC2 tespit edildi (% 43.75) ve CRP seviyesi 12-96mg/dl arasında tespit edildi. 22/64 kız çocuklarda EPEC2 tespit edildi (% 34.38), ortalama CRP seviyesi 12-96mg/dl arasındaydı. Cinsiyet ve CRP düzeyi arasında anlamlı bir fark veya korelasyon saptanmadı. EPEC4 erkeklerde 26/64, (% 40.63) tespit edildi, CRP seviyesi 12-96mg/dl arasındaydı. EPEC4, 26/64 kız çocukta (% 40.63), ortalama CRP seviyesi 12-96mg/dl arasında tespit edildi. Cinsiyet ve CRP düzeyi arasında anlamlı bir fark veya korelasyon saptanmadı. EPEC2 ve CRP düzeyi hastaların yaşı ile anlamlı derecede korele idi. EPEC2 ile enfeksiyon EPEC4 ile ters korelasyon gösterirken, cinsiyet ve EPEC serotipleri arasında anlamlı bir ilişki yoktur.

Sonuç: Her ne kadar 1-8 yaş arası diyare vakalarında EPEC2 ve EPEC4 sıklıkla tespit edilmiş olsa da, sadece EPEC2 yaşla pozitif olarak ilişkilidir.

Anahtar Sözcükler: EPEC, yaş, cinsiyet, CRP

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INTRODUCTION

Diarrhea is usually defined as the passage of loose or watery stools, usually at least three times in 24 hours and the importance is put onto the change in stool consistency rather than frequency, and the usefulness of parental insight in deciding whether children have diarrhea or not (1).

The diarrheagenic *E.coli* pathotypes that cause diarrhea include *EPEC*; enterotoxigenic *E.coli* ,ETEC ; enteroinvasive *E.coli* ,EIEC ; Shiga toxin-producing *E.coli* ,STEC ;diffusely adherent *E. coli* , DAEC and enteroaggregative *E.coli* , EAEC . These pathotypes are defined by the presence or absence of one or more definable *E. coli* virulence factors (2). *EPEC* strains are diarrheagenic *E. coli*, which usually are classified by a combination of oligosaccharides (O), flagella (H), and capsular (K) antigens. They are associated with outbreaks of infantile diarrhea among children in developing countries. In contrast to the limited importance of *EPEC* in industrialized countries, *EPEC* is a major cause of diarrhea in developing countries(3). *EPEC* strains continue to cause severe and sometimes fatal infantile diarrhea, particularly in Africa. Increased efforts at diagnosis, defining the clinical spectrum of disease, understanding pathogenic mechanisms, and delineating immune responses are desperately needed to develop new strategies to combat *EPEC*(4).

CRP is ring shaped, pentameric protein found in blood plasma, whose levels rise in response to inflammation. It is an acute phase protein of hepatic origin that increases following interleukin-6 secretion by macrophages and T cells. Its physiological role is to bind to lysophosphatidylcholine expressed on the surface of dead or dying cells and some types of bacteria in order to activate the complement system via the C1q complex(5). It enhances phagocytosis by macrophages via opsonin-mediated phagocytosis, which express a receptor for CRP. It plays a role in innate immunity as an early defense system against infections. CRP rises within 2-6 hours of the onset of inflammation, up to a 50,000-fold, and peaks at 48 hours(6). Its half-life of 18 -19 hours is constant, and therefore its level is determined by the rate of synthesis in the and production from liver as well as by other cells such as adipocytes and hence the severity of the precipitating cause(7) . Normal concentration of CRP in healthy human serum is between 5 and 10 mg/L, increasing with aging(8).

The aim of the present study is to determine the possible correlation between *EPEC* serotypes (O111, O55, O26, O86, O119, O127, O114, and O142) and CRP level in acute diarrhea among children . Also to determine the possible effect of age and gender in CRP level.

MATERIALS and METHODS

A total of (64) diarrheic child attended to outpatient's clinic of Baqubah teaching hospital during a period from January 2015 to December 2015 enrolled in this study. This study conducted according to the principles of Helsinki declaration. Dully filled consent obtained from all patient' s parents before participating in the study. Approval of ethical review Committee (Issue No. 1/232-15, January 2015), College of medicine –Diyala University-Iraq taken prior to initiation of the work.

Isolation and identification of E.coli

Gram staining technique applied for microscopic identification of gram-negative bacilli. Stool culture was done using MacConkey agar for determination of *E.coli* according to pinky colony morphology due to lactose fermentation (9)

E.coli typing

API 20E system from bio Merieux –France used for biochemical characterization of *E.coli* according to manufacturer instructions (10) . *E.coli* typed by direct slide agglutination technique in to four types using specific trivalent antisera from Bio-Rad-France. Antibodies specific for O111,O55 ,O26 antigens for *E.coli* type 1 ;antibodies specific for O86,O119,O127 antigens for *E.coli* type 2 ;antibodies specific for O125,O126,O128 antigens for *E.coli* type 3 ; antibodies specific for O114,O124,O142 antigens for *E.coli* type4(11) .

Principle

When a bacterial culture mixed with a specific antiserum directed against bacterial surface components, the cells are bound together through antigen-antibody bonds to form aggregates (agglutination).

This is usually visible to the naked eye as clumps in the suspension. By mixing specific antisera with an *E. coli* culture, the O- antigens are determined.

Procedure

Physiological saline used as a negative control. If the negative control is positive (agglutinates), the strain is auto agglutinating, i.e. O rough. Slide agglutination with O antisera according to statens serum institute procedure(12) .

The *E.coli* grown over night on a suitable agar medium not inhibiting motility. A small drop of antiserum 20 µl on a glass slide was applied. Culture from a single colony to each drop of antiserum was transferred via toothpick and mixed well and give milky turbidity . Titled the slide for 5 - 10 seconds. The reaction read with naked eye by holding the slide in front of a light source against a black background (indirect illumination).A positive reaction is seen as a visible agglutination. A negative reaction is persistence of the homogeneous milky turbidity. A late or weak agglutination considered negative.

CRP-latex agglutination

Reagents and samples allowed reaching room temperature. About 50 µL of the sample and one drop of each positive and negative controls were added into separate circles on the slide test. Then the CRP-latex reagent was mixed vigorously before using and 50 µL was added next to the samples to be tested. The drops mixed with a stirrer, spreading them over the entire surface of the circle. The slide was placed on a mechanical rotator at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test read later than two minutes. The approximate CRP concentration in the patient sample is calculated as follow(13) : 6 x CRP Titer = mg/L. The normal value of CRP in children was 0-10 mg/l (14).

Statistical analysis

Spearman's test (rho) for categorical and non categorical data used for correlation. The level of significance was 0.05 to 0.01 (two-tail) . The level of confidence limits was 0.095.Statistical analysis performed using SPSS for windows version 17 (15, 16)

RESULTS

In this study, (64) children with diarrhea were involved. As shown in table 1 the minimum age of infected children was 4 months while maximum age was 10 years and the mean age was 2.92 years. Males represent 34/64, 53.1% and the rest 30/64, 46.9% were females. The minimum CRP level 12mg/dl while maximum was 96mg/dl . As shown in table 2 , *EPEC*1 and *EPEC*3 not detected in all patients. *EPEC*2 detected among 14/64, 21.9% and *EPEC*4 in12 /64 , 18.8% . Table (3) shown that among the age group <1 year, *EPEC*2 detected in 6/64 , 9.38% , with CRP level 12mg/dl 3.13% ,48 mg/dl 6.25% . *EPEC*4 detected in 2/64 , 3.13% ,with CRP level 12mg/dl 3.13% .Age group 1-4 year, *EPEC*2 detected in 6/64 , 9.38% , with CRP level 24mg/dl , 9.38% . *EPEC*4 detected in 6/64 , 9.38% , with CRP level 12mg/dl, 24mg/dl, 48mg/dl 3.13% . Among the age group 4.1-8 year, *EPEC*4 detected in 4/64 , 6.25% , with CRP level 48 mg/dl. *EPEC* not detected in 8> years age group .

As shown in table 4 ,*EPEC*2 detected in 28/64 male, 43.75% and the CRP level range from 12-96mg/dl .*EPEC*2 detected in 22/64 females, 34.38% , CRP level range from 12-96mg/dl . No significant difference or correlation between gender and CRP level was detected. *EPEC*4 detected in 26/64 males, 40.63% , CRP level range from 12-96mg/dl . *EPEC*4 detected in 26/64 females 40.63% , CRP level range from 12-96mg/dl . No significant difference or correlation between gender and CRP level was detected. As shown in table 5 infection with *EPEC*2 and CRP level significantly correlated with patients age. Infection with *EPEC*2 inversely correlated *EPEC*4 . Infection with any *EPEC* serotypes was not correlated with patient gender.

Table 1: General Description of Patients' Age, Gender And CRP Level

Parameters		
Age	Minimum	4(months)
	Maximum	10 years
	Mean± SD	2.92 ± 2.16(years)
Gender	No.(%)	
	male	34(53.1%)
	female	30(46.9%)
	Total	64(100%)
CRP(mg/dl)	Minimum	12
	Maximum	96
	Mean± SD	46.8750 ± 33.71943

Table 2: Serotypes of EPEC In Diarrheic Children

Parameters		No. (%)
EPEC 1	Positive	0(0%)
	Negative	64(100%)
EPEC 2	Positive	14(21.9%)
	Negative	50(78.1%)
EPEC 3	Positive	0(0%)
	Negative	64(100%)
EPEC 4	Positive	12(18.8%)
	Negative	52(81.3%)

Table 3.EPEC Distribution According To Age And CRP Level In Diarrheic Children

Age group (year)	EPEC serotype	CRP level				Total	
		12mg/dl	24 mg/dl	48 mg/dl	96 mg/dl		
<1	EPEC1	Positive	0(0%)	0(0%)	0(0%)	0(0%)	
		Negative	12(18.75%)	0(0%)	4(6.25%)	0(0%)	16(25%)
	EPEC2	Positive	2(3.13%)	0(0%)	4(6.25%)	0(0%)	6(9.38%)
		Negative	10(15.62%)	0(0%)	0(0%)	0(0%)	10(15.62%)
	EPEC3	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	12(18.75%)	0(0%)	4(6.25%)	0(0%)	16(25%)
	EPEC4	Positive	2(3.13%)	0(0%)	0(0%)	0(0%)	2(3.13%)
		Negative	10(15.62%)	0(0%)	4(6.25%)	0(0%)	14(21.9%)
1-4	EPEC1	Positive	0(0%)	0(0%)	0(0%)	0(0%)	
		Negative	0(0%)	10(15.62%)	8(12.5%)	18(28.13%)	36(56.25%)
	EPEC2	Positive	0(0%)	6(9.38%)	0(0%)	0(0%)	6(9.38%)
		Negative	0(0%)	4(6.25%)	8(12.5%)	16(25%)	28(43.75%)
	EPEC3	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	0(0%)	10(15.62%)	8(12.5%)	18(28.13%)	36(56.25%)
	EPEC4	Positive	0(0%)	2(3.13%)	2(3.13%)	2(3.13%)	6(9.38%)
		Negative	0(0%)	8(12.5%)	6(9.38%)	16(25%)	30(46.88%)
4.1-8	EPEC1	Positive	0(0%)	0(0%)	0(0%)	0(0%)	
		Negative	4(6.25%)	2(3.13%)	4(6.25%)	0(0%)	10(15.62%)
	EPEC2	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	4(6.25%)	2(3.13%)	4(6.25%)	0(0%)	10(15.62%)
	EPEC3	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	4(6.25%)	2(3.13%)	4(6.25%)	0(0%)	10(15.62%)
	EPEC4	Positive	0(0%)	0(0%)	4(6.25%)	0(0%)	4(6.25%)
		Negative	4(6.25%)	2(3.13%)	0(0%)	0(0%)	6(9.38%)
>8	EPEC1	Positive	0(0%)	0(0%)	0(0%)	0(0%)	
		Negative	2(3.13%)	0(0%)	0(0%)	0(0%)	2(3.13%)
	EPEC2	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	2(3.13%)	0(0%)	0(0%)	0(0%)	2(3.13%)
	EPEC3	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	2(3.13%)	0(0%)	0(0%)	0(0%)	2(3.13%)
	EPEC4	Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)
		Negative	2(3.13%)	0(0%)	0(0%)	0(0%)	2(3.13%)

Table 4. Distribution of *EPEC* According to Gender And CRP Level In Diarrheic Children

Sex		CRP(mg/dl)	CRP(mg/dl)				Total	χ^2	P value	R	P value
			12	24	48	96					
Male	<i>EPEC</i> 1	Negative	8(12.5%)	8(12.5%)	6(6.25%)	12(18.75%)	34(53.13%)	ND	ND	ND	ND
		Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)				
	Total	8(12.5%)	8(12.5%)	6(9.38%)	12(18.75%)	34(53.13%)					
Female	<i>EPEC</i> 1	Negative	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)	ND	ND	ND	ND
		Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)				
	Total	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)					
Male	<i>EPEC</i> 2	Negative	0(0%)	4(6.25%)	2(3.13%)	0(0%)	6(9.38%)	11.063	0.011	0.247	0.159
		Positive	8(12.5%)	4(6.25%)	4(6.25%)	12(18.75%)	28(43.75%)				
	Total	8(12.5%)	8(12.5%)	6(9.38%)	12(18.75%)	34(53.13%)					
Female	<i>EPEC</i> 2	Negative	2(3.13%)	2(3.13%)	2(3.13%)	2(3.13%)	8(12.5%)	1.705	0.636	-0.051	0.788
		Positive	8(12.5%)	2(3.13%)	8(12.5%)	4(6.25%)	22(34.38%)				
	Total	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)					
Male	<i>EPEC</i> 3	Negative	8(12.5%)	8(12.5%)	6(6.25%)	12(18.75%)	34(53.13%)	ND	ND	ND	ND
		Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)				
	Total	8(12.5%)	8(12.5%)	6(9.38%)	12(18.75%)	34(53.13%)					
Female	<i>EPEC</i> 3	Negative	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)	ND	ND	ND	ND
		Positive	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)				
	Total	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)					
Male	<i>EPEC</i> 4	Negative	2(3.13%)	2(3.13%)	2(3.13%)	2(3.13%)	8(12.5%)	0.654	0.884	0.092	0.607
		Positive	6(9.38%)	6(9.38%)	4(6.25%)	10(15.62%)	26(40.63%)				
	Total	8(12.5%)	8(12.5%)	6(9.38%)	12(18.75%)	34(53.13%)					
Female	<i>EPEC</i> 4	Negative	0(0%)	0(0%)	4(6.25%)	0(0%)	4(6.25%)	9.231	0.026	-0.072	0.706
		Positive	10(15.62%)	4(6.25%)	6(9.38%)	6(9.38%)	26(40.63%)				
	Total	10(15.62%)	4(6.25%)	10(15.62%)	6(9.38%)	30(46.87%)					

ND: Not

detected

Table 5. Correlation Between Age, Gender *EPEC2* And *EPEC4* In Diarrheic Children

Spearmen's rho		<i>EPEC 2</i>	<i>EPEC 4</i>	CRP
Age	rho	0.367	-0.157	0.325
	P value	0.003	0.214	0.009
Sex	rho	-0.109	0.130	-0.112
	P value	0.392	0.305	0.377
<i>EPEC 2</i>	rho		-0.254	0.051
	P value		0.043	0.690
<i>EPEC 4</i>	rho			-0.045
	P value			0.725

DISCUSSION

The rate of mortality from diarrheal diseases in the world has decreased, mainly because of better therapy and interventions that promote sanitary conditions and that educate inhabitants to encourage them to take part in primary health care activities (17). However, acute diarrheal diseases continue to be one of the major causes of morbidity and mortality in the developing world (18). In current study, the mean age was 2.92 ± 2.16 years which come in line with (19).

Affected males represent 53.1% and 46.9% were females which come in line with (19). The mean CRP level 46.88 ± 33.72 mg/dl, which considered higher than other reports (20, 21). In current study, *EPEC1* and *EPEC3* not detected in all patients. *EPEC2* was isolated from (21.9%) of infected children while *EPEC4* was recovered from (18.8%) which come in agreement with previous study in Kut city, Iraq (22).

In current study, among age group <1 year, *EPEC2* detected in (9.38%), with CRP mean level fluctuated from 12mg/dl that was reported in 3.13% while 48 mg/dl was reported in 6.25% of *EPEC2* positive cases which come in agreement with that reported by (19). *EPEC4* detected in 3.13%, with CRP level 12mg/dl which reported in 3.13% of infected children. These results come in line with (19), and come closely to that reported by (23) who found that *EPEC* represent 5.2%. Current results were lower than that reported in Iran by (24), who revealed that *E. coli* associated diarrhea represent 40.4% in children under 1 year and collectively the CRP level among infected children was 14.3mg/dl (24). In another study from Nigeria, *EPEC* detected in 49.02% of children with diarrhea (25). This could be related to the onset of exposure to environment and increased introduction of solid foods to children whose immune system is still developing. All these factors represent a contributory beside the fact that older children are probably more mobile and playful than younger children (17). They have a higher chance than children below six months do to get diarrhea from hand-contamination, especially while playing in the ground; playing with their toys or other objects; and unknowingly putting their dirty fingers into their mouth. In addition, the risk of ingesting contaminated materials is high, especially in unhygienic environments (26).

Among age group 1-4 year, *EPEC2* was detected in 9.38%; the CRP level of infected children represent 9.38% at the level 24mg/dl. On the other hand, fluctuation in levels of CRP was reported among *EPEC4* infected children 12mg/dl, 24mg/dl, 48mg/dl at 3.13%. Among older age group (4.1-8) year, *EPEC4* detected in 6.25% of infected children, with CRP level 48 mg/dl. These results come in line with (22) and lower than that reported in Iran (24). *EPEC* not detected in 8> years age group and this may attributed to the development of immunity or the loss of receptors for some specific adhesion molecules (27). The *EPEC* serogroups come in line with that detected in other studies (26). *EPEC* commonly transmitted via the fecal-oral route in a poor hygienic environment (28). Results of studies carried out in areas with different health and socio-demographic characteristics showed that strains of *EPEC* were the major cause of diarrhea in developing countries (29). The current results come in line with a study conducted in Iran showed that most cases of acute diarrhea were due to sero groups of *EPEC* including O127, O86, O126, O142, O55, O119, O128. Also current result come in accordance with Nigerian study in which the frequent *EPEC* serotypes were O26, O111, O119, O127, O128, O44, O55, O125, O126, O114 and O142 serotypes (30).

In current study, *EPEC2* was detected in 43.75% of males in which, the CRP level range among them was 12-96mg/dl. *EPEC2* detected in 34.38% of females in which, CRP level range (12-96mg/dl). These result come in contrary with (22) although CRP level was not determine.

In current study, *EPEC4* detected among 40.63% of males and females. CRP level range 12-96mg/dl. Fluctuation of CRP levels reflects the low grade inflammatory response for noninvasive *EPEC*. No significant correlation between gender and CRP level was detected. This may attributed to the limited or even absent role of hormones at this stage of life. *EPEC2* and CRP level significantly correlated with patients age and this come in line with others (31). *EPEC2* inversely correlated with *EPEC4* and this may attributed to the competition on binding receptors that expressed on intestinal mucosa. No significant correlation between gender and *EPEC* serotypes reflects the importance of predisposing factors and availability of receptors for attachment and colonization regardless of patients' gender.

CONCLUSION

Although *EPEC2* and *EPEC 4* frequently detected among diarrhea cases of 1-8 years old, only *EPEC2* positively correlated with age. Even with a fluctuation in CRP level, it was positively correlated with age and not correlated with *EPEC* serotypes and gender.

Conflict of interest

No conflict of interest was declared by the authors.

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