ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

Study on bacterial dissemination and pathological findings of Enteropathogenic Escherichia coli in white albino mice

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Manuscript Info

Manuscript History:

Received: 12 February 2014
Final Accepted: 25 March 2014
Published Online: April 2014

Key words:
: EPEC 0119 bacterial dissemination and pathology in mice .

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Abstract

In a study designed to identify the pathogenesis of Enteropathogenic *Escherichia coli* (EPEC) in white albino mice . EPEC strain was isolated from diarrhea of children in Al-KadhimiyaHospital , Baghdad , Iraq . The EPEC strain Isolated , biochemically and serologically typed as EPEC 0119 . Then two groups of mice (30 mice in each group) control group injected with phosphate buffer saline 0.1 ml S/C and infected group S/C injected with 0.1 ml of $10x9^{10}$ CFU/ml . Results revealed that heavy bacterial growth were disseminated in the internal organs ; intestine , liver , lungs , kidneys , Brain , heart & spleen during 48hrs post inoculation . Among pathological findings , thrombosis , edema and acute inflammatory cells response (neutrophils) infiltrate these internal organs of the mice

Conclusion

1- heavy bacterial growth disseminate in the internal organs of mice following S/C inoculation of EPEC in mice . 2- severepathogical lesions in the internal organs of mice following inoculation with these bacteria .

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Introduction

Escherichia coli is a common cause of gastroenteritis in children and infants and account of 30% of the total number of diarrhea pathogens in some regions (1) . There are more than 900 serotypes of EPEC and there are hundreds of strains causing diarrhea or hemorrhagic watery diarrhea in infants around the world (2) . EPEC are the cause of sever and persistent infants diarrhea both in developed and developing countries (3) . It is a major medical problem with serious consequences in children less than 3 months of age and its important cause of morbidity and mortality in weaned rabbits (4) . EPEC is also pathogenic in neonatal calves (5) . Also it is isolated from cases of post weaning diarrhea in swine (6) and there is increase the evidence of diarrheagenic role of EPEC in dogs (7) . The importance of the EPEC in these diseases processes, this study aimed at :

1- Study the dissemination of EPEC in internal organs of white mice .
2- Study the pathological findings associated with experimental infection of these laboratory animals with this microbial agent.

Materials and Methods

Forty stool samples were collected from diarrhea of infants and children under 2 years old in Al – KadhimiyaHospital , Baghdad , Iraq .*Escherichia coli* (EPEC) were isolated from these diarrhea samples on culture media , biochemically identified (8) and by using API-E , the isolates were EPEC (9) . Serotyping of EPEC were done using specific mononspecific antisera of *E. coli*(Bio Red mamas coquette trivalent 1) .The EPEC serotypes were 0119 and the LD50 of this serotype was $1x10^9$ CFU/ml (10). The two groups of white albino mice were taken, a control groups (30) were injected with 0.1 ml S/C of PBS and infected group were injected with 0.1 ml of 10 LD50 ($10x10^9$ CFU/ml) S/C .Then clinical signs , Bacterial dissemination in the internal organs of mice and pathological lesions were recorded and pieces of tissue lesions were taken for histopathology findings (11).

Results and Discussion

1- Clinical and Bacterial dissemination :

The infected group of mice showed different clinical signs and all mice in this group were died during the 48hrs post inoculation . Also heavy bacterial growth were isolated from these internal organs , intestine , lungs , liver , spleen , kidney , brain and heart (Table-1)

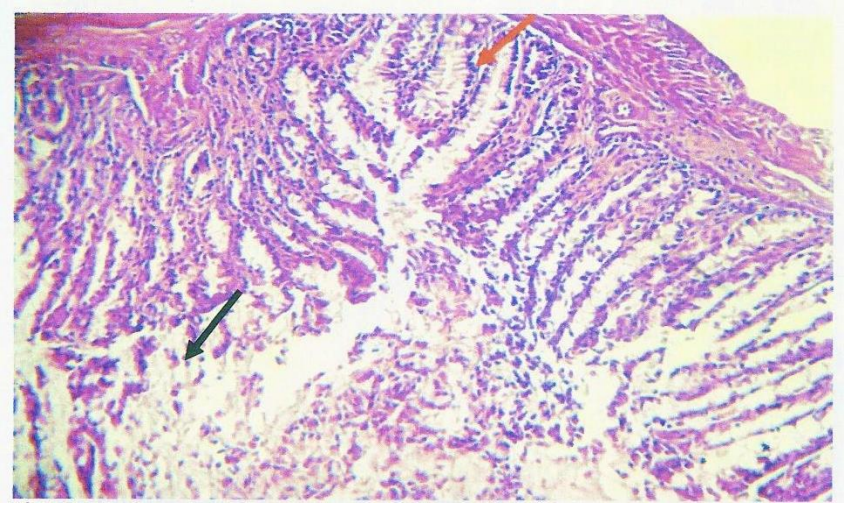
Group	Spleen	Liver	Kidneys	Lungs	Hear	Brain
Infected	++	+++	++	++	+	+
Control	-	-	-	-	-	-

Table -1: Bacterial dissemination in the internal organs of EPEC infected group died during 48hrs. post inoculation .

2- Pathological findings :

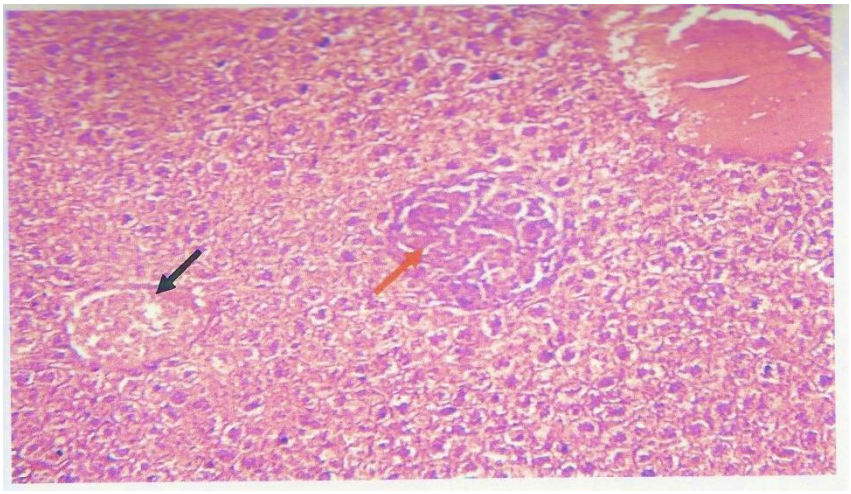
All the internal organs of the mice died during 48hrs post inoculation showed congestion , fluid oozing from cut section of the organsHistopathologically the organs showed :

Intestine : acute enteritis followed by extensive secretion of mucin , congestion of blood vessels and neutrophils infiltration in the villarepith and in the lumen of intestine (Fig – 1) .



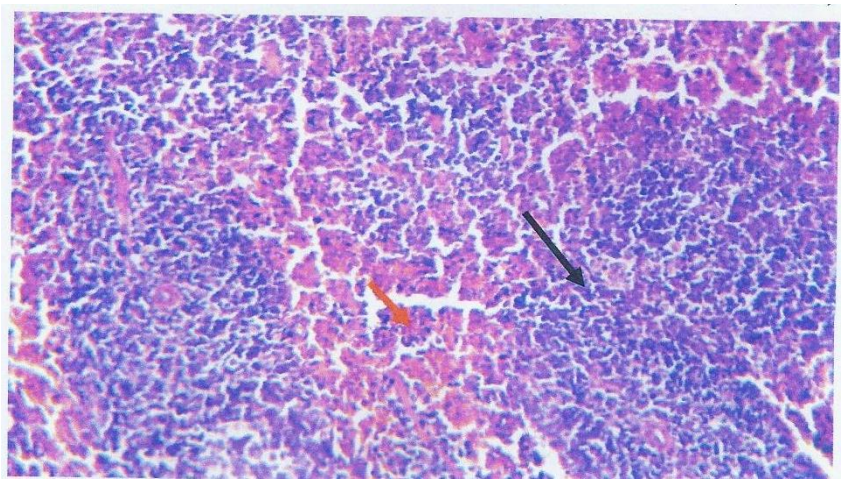
(Fig-1) : intestine showed , congestion , degeneration ; and neutrophils infiltration in villarepith.(H&E) x40

Liver : there is extensive congestion in the central vein , sinusoids and portal areas , neutrophils infiltration and prominence of kubffer cells (Fig – 2) .



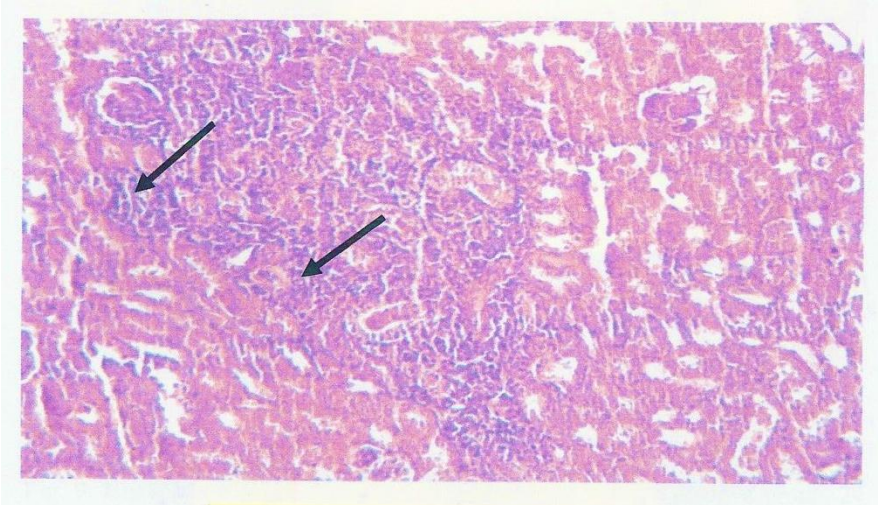
(Fig – 2): Liver showed congestion , neutrophils infiltration and prominence Kubffer cells .
(H& E) x40

Spleen : showed extensive congestion of the Red and white pulp , hemorrhage and oedema in the spleen trabeculae Also mild reactive hyperplasia of white pulp (Fig -3)



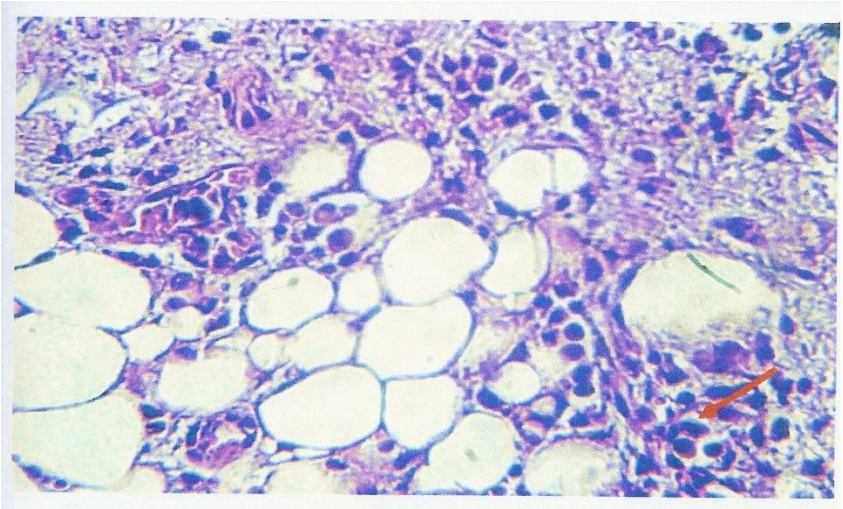
(Fig -3) : spleen : showed congestion , edema & hyperplasia of white pulp.
(H& E) x40

Kidneys : showed cloudy swelling of the renal tubules , dilation of Bauman space congestion and edema and neutrophils infiltration in the interstitial renal tissue (Fig -4)



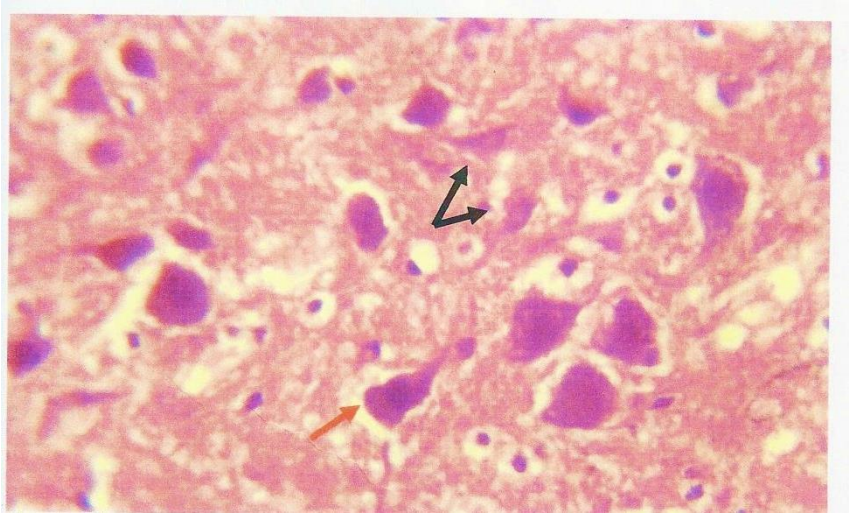
(Fig -4): Kidney showed congestion and neutrophils infiltration in the interstitial tissue
(H& E) x40

Perition : showed extensive peritonitis , characterized by infiltration of neutrophils and macrophages , congestion and edema in peritoneal tissue (Fig -5)



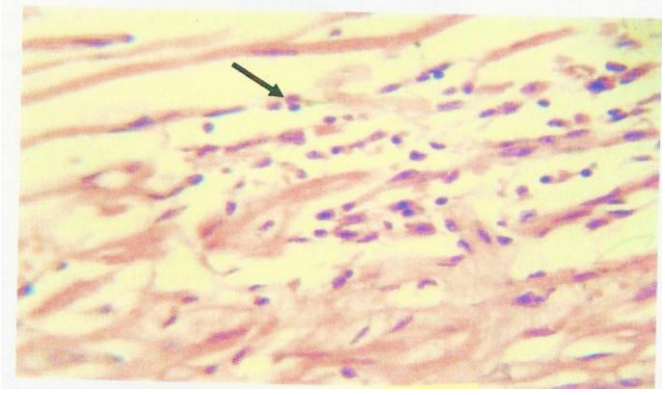
(Fig -5): periton : showed infiltration of neutrophils and few mononuclear cells in adipose tissue .(H& E) x40

Brain : showed shrinkage and degeneration of neurons and mild perivascular neutrophils infiltration and perineuronal edema (Fig-6)



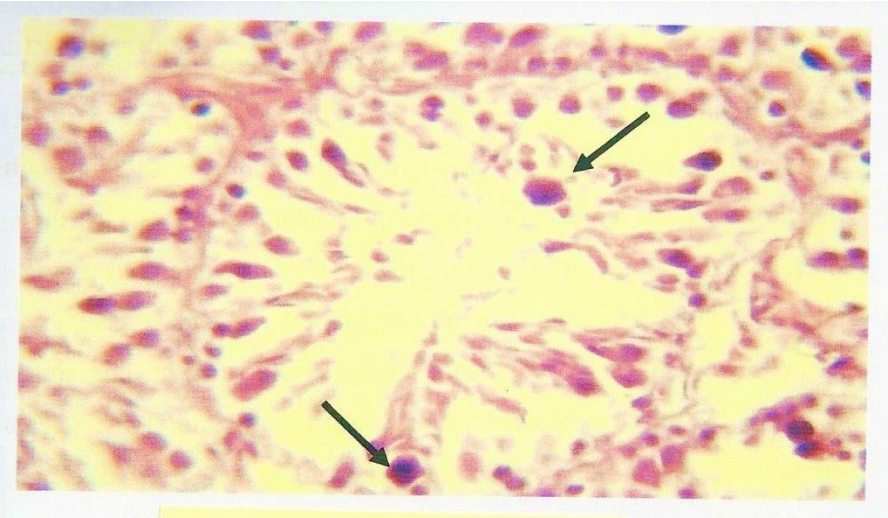
(Fig-6): Brain showed congestion ,perineuronal edema (H & E) x40

Heart : showed congestion of blood vessels , edema and neutrophils infiltrates between muscle fibers (Fig -7)



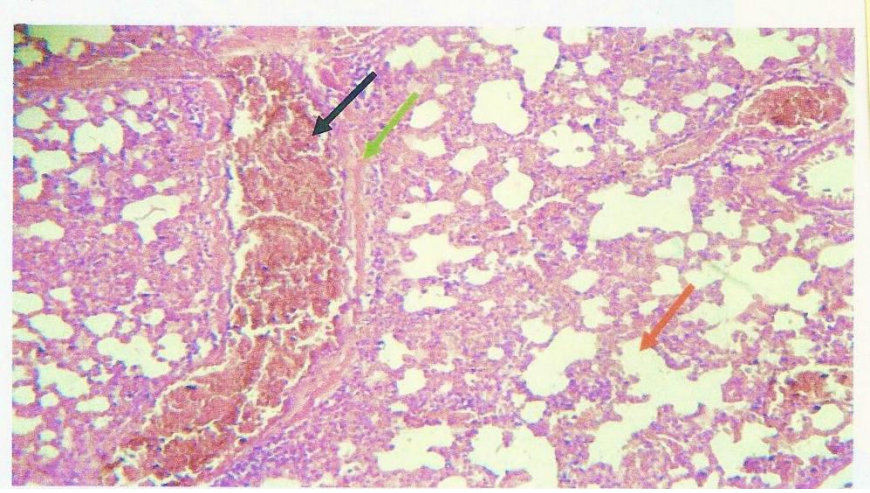
(Fig -7): Heart : showed edema and neutrophils infiltration between muscle fibers .
(H& E) x40

Testes : showed hydropic degeneration of seminiferous tubular epithelial lining , edema and congestion in the interstitial testicular tissue In addition spermatid giant cells formation .(Fig-8)



(Fig-8) : Testis showed degeneration of epithelial lining seminiferous tubules and spermatid giant cells (H & E) x40

Lungs : showed extensive congestion , edema and thrombus formation .
Also emphysema and extensive neutrophils infiltration in the interstitial pulmonary tissue (Fig-9) .



(Fig-9) : Lung showed congestion , thrombus formation, emphysema and neutrophils infiltration in the interstitial tissue . (H& E) x40

Control non infected group : showed normal clinically healthy animals , No bacterial isolation and No pathological lesion were seen in internal organs .
The death of infected group of mice during 48hrs post inoculation with EPEC , indicated that this group of animals were exposed to lethal dose of highly virulent strain of EPEC 0119 and this microbe rapidly disseminated following their S/C inoculation into the internal organs and cause septic shock , the similar finding were reported by (12) with the similar dose of EPEC 1/P in mice and the septic shock caused multifailureorgans manifested by sever congestion

, edema , hemorrhage , and neutrophils infiltrates in the internal organs of mice which is more evident in this study . Septic shock and septicemia occurred as a result of lipopolysaccharides a major constituent of gram negative bacteria (EPEC in this study) which resulted into diarrhea and enteritis in mice (12, 13) and in rabbits (14) and in children naturally (15) , all those authors found the enteritis accompanied by goblet cells proliferation and diarrhea and mucin secretion .

The congestion , hemorrhage , thrombus and edema were seen extensively in this study occurred as a result of septicemia and endotoxemia induced a septic shock in which inadequate blood perfusions in tissues resulted in cell dysfunction and death (16) .Also cell dysfunction and death occurred as a result of release of active oxygen species , peroxidase radicals by neutrophils that directly cause cell damage (12) which was evident in this study through neutrophils migration into the all internal organs of mice . The neutrophils migration occurred as a result of the complement components C3a and C5a which triggered under effect of endoxoxin of EPEC in complement pathway (17) .

References

1) Ali khani , M. ; Mirsalehian , A. ; Fatollahzadeh , B. ; pourshafie , M. and Asloni , M. (2007) . Prevalence of enteropathogenic and shiga toxin producing *E-coli* among children with and without diarrhea in Iran , J. Health Poplu. Nutr. 25(1) : 88-93
2) Keer , M. ; Fitzergerald , M. ; Sheridan , J. ; Medowell , , D. A. and Blair , I. S. (1999) survival of *Escherichia coli* 0157: H7 in bottled natural mineral water . J. App. Microbiol .87 : 833-841 .
3) Black , R. F. and Brown , K. H. (1982) longitudinal studies of infections diseases and physical growth of children in rural Bangladesh . Incidence of diarrhea and association with known pathogens . Am. J. Epidemiol. 115: 315-324 .
4) Blanco , J. E.; Blanco , M. ; Blanco , J. Mora , A. ; Balaguer , L. ; Mourino , M. ; Juarez, A. and Jansen , W. B. (1996) . oserogroups , biotypes and eae Agenes in *Escherichia coli* strain isolated from diarrhea and healthy rabbits . J. Clin .Microbiol. 34 : 3101 -3107 .
5) Fisher , J. ; Maddox, C. ; Moxley , R. ; Kinden , D. and Miller , M. (1994) Pathogenicity of a bovine attaching effacing *Escherichia coli* isolate lacking shiga – like toxins Am. J. Vet. Res. 55:991-999 .
6) Zhu , C. ; Harel , J. ; Jacques , M. ; Desautels , C. ; Donnenberg , M. S. ; Beaudry , M. and Fairbrother , J. M. (1994) . Virulence properties and attaching effacing activity of *Escherichia coli* 045 from swine postweaning diarrhea . Infect. Immun. 62 : 4153 – 4159 .
7) Turk , J. ; Maddox , C. ; Fales , W. ; Ostlund , E.; Miller . M. ; Johnson , G. ; pace , L. Turniquist , S. and Kreeger , J. (1998) . Examination for heat – labile , heat – stable , and shiga – like toxins and for the eae A gene in *Escherichia coli* Isolates obtained from dogs dying with diarrhea 122 cases (1992 – 1996) J. Am. Vet. Med. Assoc. 212 :1735 -1736 .
8) Quinn , P. J. ; Markey , M. E. and carter G. R. (2004) Clinical veterinary microbiology 6th ed. Mosby company year Book London UK. Wolf , London , PP: 261-267 .
9) Atlas , R. M. (1995) Principles of microbiology . Mosby year Book , St. Louis , Missor ; USA .
10) Dixon , W. J. (1980) Efficient analysis of experimental observations . Ann. Res. Pharmacol. Toxicol. 20: 441 – 462.
11) Luna , L. G. (1968) Manual of histological staining methods of the Armed Forces Institute of pathology , 3rd ed. McGraw – Hill Book company New York .
12) Joseph, E. P. (1993) Pathogenetic mechanism of septic shock . Engl. J. Med.328: 1471-1478 .
13) Christian , R. and chris , W. (2002) Lipopolysaccharide endotoxin , Annu. Rev. Biochem . 71: 635 – 700 .
14) Suzana , D. S.; Jennilee , V. ; Jerrold, R. T. ; Kristina , A. M. and Gail , H. (2005) mouse model of Enteropathogenic*Escherichia coli* infection . Infect. Immun. 73(2)1161 - 1170 .
15) Mundy , R. ; Pickard , D. ; Wilson , R. K. ; Simmons , C. P. ; Dougan , G. and Frankel , G. (2003) Identification of a novel type IV pilus gene cluster required for gastrointestinal colonization of *Citrobacterreodentium* . Mol. Microbiol , 48: 795 – 809 .
16) Nataro , J. P. and Kaper , J. B. (1998) Diarrheagenic*Escherichia coli* , Clin . Microbiol. Rev. , 11: 142 -201
17) Groisman , S. Y. (2001) Identification of novel cell envelop protein and assignment to a two – dimentional reference map. J. Bacteriol., 178 (16) : 5032 – 5038 .