VII-SUMMARY AND CONCLUSION

Enterohemorrhagic *Escherichia coli* (EHEC) are one of diarrheagenic *E.coli* that produce shiga toxins causing diarrhea, hemorrhagic colitis, and hemolytic uremic syndrome. *E. coli* O157:H7 is the most common and the one that can be identified in the clinical specimens of the EHEC species.

This study aimed to detect the role of *Shigella shiga* and *E. coli* O157 as a causative agent of hemorrhagic diarrhea, through isolation of both organisms and detecting cytopathic effect of verotoxin producing *E. coli* O 157 on tissue culture.

In the present study, 70 patients with hemorrhagic diarrhea were selected from the Outpatient Clinic of Benha University Hospital and Tanta Fever Hospital.

Patients were subjected to full history taking and clinical examination. Stool samples were collected from all patients in clean closed containers. Stool then cultured on MacConkey agar and sorbitol MacConkey agar media to screen for non-sorbitol fermenting *E.coli* that are presumptively *E.coli* O157. Isolated non-sorbitol fermenting *E.coli* were cultured on ChromIDTM 0157:H7 agar, to detect the specificity of *E. coli* O157 and to improve the detection rate of diagnosis of *E coli* O157, sorbitol-fermenting *E.coli* isolates were also tested for *E.coli* O111 and O26 antigen by *E.coli* O111 and O26 latex agglutination test

At the same time stool samples were cultured on selenite broth for 24 hours then subcultured from selenite on *Shigella Salmonella* agar, the suspected colonies of *Shigella* were subjected to conventional methods for isolation and identification.

This study group included 36 males (51.4%) and 34 females (48.6%), with age ranges from 2 - 60 years old.

As regard to the organisms isolated from the stool samples: *E. coli* were detected in 33 specimens representing 47.1% of cases, the frequencies of different serogroups of isolated *E. coli* cames as follows (7 specimens were detected *E. coli* O157 representing 10% of cases, 6 specimens were detected *E. coli* O111 representing 8.6% of cases , 4 specimens were detected *E. coli* O26 representing 5.6% of cases), and *Shigella* was detected in 28 specimens (40%) which existed as follows (7 specimens were detected *Shigella shiga* representing 10% of all cases and 21 specimens were detected other types of *Shigella* representing 30% of all cases). Other bacteria can be isolated from 9 specimens (12.9%).

Escherichia coli O157 were detected in 7 stool samples of patients with hemorrhagic diarrhea (10%) by ChromIDTM O157:H7 agar, all seven cases were non sorbitol fermenters on sorbitol MacConkey agar.

The diagnostic efficiency of CHROM was much better than that of sMac, with a 75% decrease in the number of colony picks (i.e., less false-positive growth on CHROM and better differentiation of O157 due to the green blue indicator), thus allowing presumptive identification from the primary isolation plate and differentiation from other organisms.

Evaluation of viability % of vero cells treated with *E. coli* O157 toxin using The tetrazolium-based colorimetric assay (MTT assay) revealed that 6 strains out of 7 strains (85.7 %) showed a reasonable toxicity to Vero cells post treatment with bacterial toxin while strain No 4 showed no cytotoxic effect on treated cells. And the sample toxicity to Vero cells were arranged as follow: Strain No (5) showed a higher toxicity followed by strains (2 and 6), (1 and 7) and 3. Strain No (4) showed no toxic effect.

In conclusion, *Escherichia coli* O157:H7 and *Shigella shiga* play role in the etiology of hemorrhagic diarrhea. In addition, stool culture on sorbitol MacConkey agar followed by ChromIDTM O157:H7 agar seems to be a simple and reliable method for *E. coli* O157:H7 detection.