

PREVALENCE OF NON-O157 ENTEROHAEMORRHAGIC ESCHERICHIA COLI IN RETAIL GROUND BEEF IN THE UNITED STATES

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Purpose Enterohaemorrhagic *Escherichia coli* (EHEC) are associated with human diseases ranging from diarrhea to hemolytic-uremic syndrome (HUS). *E. coli* O157:H7 is the EHEC most often isolated in patients in the US causing an estimated 73,000 illnesses annually. Non-O157 EHEC also causes illness and outbreaks. More than 37,000 illnesses are attributed to non-O157 EHEC serotypes. In the US the most common non-O157 EHEC serogroups in humans are O26, O45, O10, O111, O121, and O145. The incidence of non-O157 EHEC is increasing despite limited laboratory testing. The purpose of this study was to determine the prevalence of non-O157 EHEC in retail ground beef in the US.

Materials and Methods Meat samples were selectively enriched for *E. coli* O157, EHEC, and *Salmonella* spp. then tested by IEH Polymerase Chain Reaction (PCR) test kits. Sample enrichments screened positive by PCR were culture confirmed by isolating the target organisms. The organisms were confirmed and samples were scored as positive.

Each sample (200g) was weighed into a sterile Whirlpak, 69oz. filter bag followed by the addition of 200-250ml IEH Enrichment Broth. The sample was macerated by hand homogenizing. An additional portion of IEH Enrichment Broth was added to bring broth volume to 500ml. The bag was mixed until uniformly homogenized. Enrichment bags were incubated at 42°C for 24 hours. To minimize contamination, after enrichment incubation time was completed, the broth was well mixed and a 10ml aliquot was removed to sterile test tube as working stock.

Aliquots of enrichment were tested for typical gene signals using IEH-*E. coli* EHEC Test System. Presumptive positive cultures were subjected to cultural confirmation. Presumptive colonies (lactose positive, *E. coli* type colonies) were picked to a gridded Washed Sheep Blood Vancomycin-Cefsulodin-Cefixime (BVCC) plate to test for hemolysin activity. All presumptive target positive colonies were tested by PCR test kit for gene markers indicative of EHEC. Colonies testing positive for EHEC by PCR were checked for purity on MacConkey agar, confirmed as latex agglutination negative for *E. coli* O157, and confirmed by PCR for *E. coli* serotype specific markers. Colonies confirmed as EHEC by both PCR and negative latex agglutination were scored as positive.

Results and Discussions A total of 1216 retail ground beef samples were tested for the presence of EHEC. Twenty-three samples (1.9%) were positive for non-O157 EHEC strains. Serotypes included O26 (n=6), O103 (n=7), O113 (n=1), O121 (n=6) and O145 (n=3). All but the EHEC isolate serotype O113 were *Stx* and *eae* positive. The O113 strain was *Stx*2d_{act} and Subtilase positive.

Prevalence of 1.9% non-O157 EHEC in the retail ground beef supply shows the need for public health agencies in the US to increase awareness regarding these pathogens. The data clearly show that clinical and public health laboratories should routinely screen human and environmental specimens for the presence non-O157 EHEC.