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Dramatic Change in the Apparent Epidemiology of Shiga-toxigenic *E. coli* Infection Associated with Introduction of CHROMagar™ STEC

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ABSTRACT

Objectives: Shiga-toxigenic *E. coli* (STEC) infection is common in Ireland. Comprehensive analysis of faecal samples for STEC is challenging. Molecular methods are the most comprehensive approach but there are challenges associated with routine application. We report evaluation of CHROMagarTM STEC for detection of STEC.
Methods. 1846 routinely submitted faecal samples were plated to CHROMagarTM STEC and CHROMagarTM O157 and incubated at 37°C. Suspect colonies (mauve) were confirmed as *E. coli* by indole production and morphology on Chromogenic UTI agar. *E. coli* were evaluated by agglutination with antisera (026, 0103, 0111, 0145 and 0157). Isolates agglutinating with specific antisera were referred to the national reference laboratory for molecular detection of *stx*1 and 2 and serotyping. Suspect colonies on CHROMagar O157 were confirmed as O157 by Oxoid Dryspot *E. coli* O157 and referred to the reference laboratory
Results: Mauve colonies were detected on 15% (277) of samples on CHROMagarTM STEC and 46 (2.5% of samples) isolates agglutinated with 1 of 5 specific antisera. STEC was confirmed in 23, with 18 O26, 3 O145, 1 O157 and 1 O111. 50% of isolates identified as suspect STEC from CHROMagarTM STEC in conjunction with *E.coli* serology confirmed. Specificity differed by serotype. Specificity for *E.coli* O26 was high at 81.8 and the single O157 isolate confirmed. However a high proportion of O145, O111 and O103 (respectively 67%, 90% and 100%) were not STEC. The single *E coli* 0157 was also isolated from ChromagarTM 0157. Six STEC O26 were *stx*1 and 2 positive, 11 were *stx*1 only positive and 1 *stx*2 only positive. STEC 0145 and 0157 isolates were *stx*2 positive and the STEC 011 was *stx*1 positive.
Conclusions: Use of CHROMagarTM STEC media was associated with a transformation in the apparent epidemiology of STEC infection in the region. It is now apparent that there is an predominance of non O157 STEC in particular O26. Prior notification data of laboratory confirmed STEC entirely underestimated the prevalence of infection. Under recognition remains a concern because of the limited range of antisera used in this protocol. In settings where routine application of molecular methods to all stool samples is not practical CHROMagarTM STEC can play valuable part in providing a more complete picture of the epidemiology of STEC infection. The change in practice has significant workload implications for the laboratory and for the Department of Public Health Medicine

INTRODUCTION

•Shiga-Toxigenic *E. coli (STEC)* is an important zoonotic pathogen associated with diarrhoea, haemorrrhagic colitis and haemolytic uraemica syndrome . *STEC is a notifiable infectious disease in Ireland*
•In 2011 270 cases of STEC were notified in Ireland, representing an incidence of 5.9/100,000 population. 69% of isolates were O157 and 31% were non 0157 - 17% 026 and 14% various other serotypes. Our laboratory reported 19 STEC - 79% O157 and 21% 026
•Detection of STEC from faecal samples is challenging because of the diversity of background of *E.coli* present in all stool samples and because of the diversity of STEC *E.coli* serotypes
•Increasingly non culture based methods based on toxin detection or detection of toxin genes are important in making the diagnosis and culture independent diagnosis is now accepted in ECDC case definitions for this pathogen
•Culture of the pathogen remains the gold standard however and at present remains the most widely used method in Europe.
•Our laboratory has used the selective O157 CHROMagar for a number of years but as noted we were concerned that we were not detecting non-O157 STEC
•We evaluated a novel chromogenic agar STEC CHROMagar that is reported to support detection of all of the more common serotypes of STEC including 0157,026,045,0103,0111 and 0145

MATERIALS AND METHOD

•Time period for the study was from 02/02/2012 to 10/06/2012
•Samples studied included 1846 routine clinical faecal samples
•Faecal samples were cultured on CHROMagar STEC in parallel with CHROMAgar 0157
•Suspect STEC colonies were those appearing mauve on CHROMagar STEC while suspect STEC 0157 colonies were those appearing pink on CHROMagar 0157
•Suspect STEC colonies were confirmed as *E.coli* by indole production and appearance on Chromogenic UTI agar
•*E.coli* were evaluated by agglutination with *E.coli* Pool 1 antiserum (containing 026,0103,0111,0145 and 0157 antiserum)
•Isolates which agglutinated with Pool 1 antiserum were then tested against each of the 5 individual component antisera to confirm agglutination with 1 of the 5 specific antisera
•*E.coli* isolates agglutinating with specific antisera were referred to the national public health reference laboratory for confirmation as STEC including molecular detection of stx1 and 2 and serotyping

RESULTS

•Suspect STEC colonies were detected in 277 (15%) of samples
•46 (2.5% of samples) of these confirmed as *E.coli,* agglutinated with specific antisera and were referred for confirmation
•23 of the presumptive STEC isolates referred were confirmed as STEC
•Of the 23 confirmed STEC isolates 18 (78%) were 026, 3 (13%) were 0145, 1(4.4%) was 0157 and 1(4.4%) was 0111
•Overall specificity of the CHROMagar in conjunction with *E.coli* serology was 50% but varied significantly by serotype – specificity for *E.coli* 026 was high at 81.8% and the single 0157 isolate confirmed but specificities for 0145, 0111 and 0103 were lower at 33, 10 and 0% respectively

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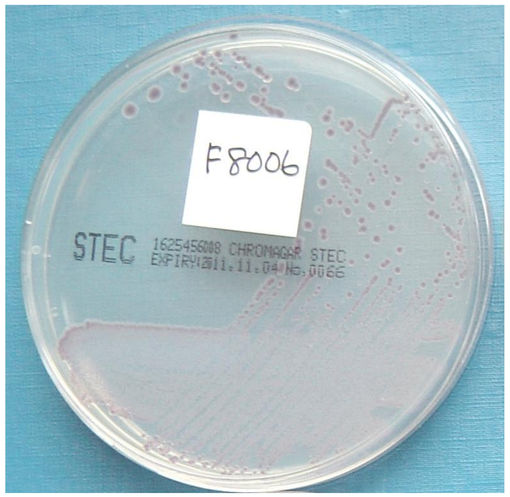
STEC mauve colonies on CHROMagar STEC™



CONCLUSION

•Adoption of STEC CHROMagar to replace O157 CHROMagar results in a transformation of the apparent epidemiology of STEC in our region
•O26 is the predominant STEC O group in our region similar to adjoining regions of Ireland
•The predominance of 026 in the region may reflect water borne transmission as many households in rural sectors of the region are served by untreated private water supplies (individual wells or small, unregulated group water supplies) which are liable to contamination with animal waste particularly following heavy rainfall
•CHROMagar supports detection of O157 and O26 STEC with relatively few false positive results
•Its performance for other STEC O groups is less satisfactory
•Under recognition of STEC remains a concern due to the limited range of antisera used in the protocol
•However, where routine application of molecular methods of detection to all clinical samples is not practical, CHROMagarTM STEC can play a significant role in enhanced detection of STEC infection and in providing a more complete picture of the epidemiology of STEC infection

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