INTRODUCTION

- Shiga toxin-producing *Escherichia coli* (STEC) cause a range of illnesses from gastroenteritis to life threatening haemolytic uraemic syndrome.
- CHROMagar™ STEC (CHROMagar, Paris, France) is a selective and differentia media for the detection of STEC in faecal specimens. Enterobacteriaceae appear colourless, blue, or are inhibited, while STEC colonies appear mauve.
- The SHIGA TOXIN QUIK CHEK™ (TECHLAB, Blacksburg, Virginia, United States) assay is a rapid membrane immunosay that can detect and differentiate the presence of Shiga toxins 1 and 2 (Stx1 and Stx2) in stool specimens directly, or from broth and plate cultures of stool samples.
- Here we compare the performance of CHROMagar™ STEC and the SHIGA TOXIN QUIK CHEK™ assay to a Vero cell cytotoxicity neutralization assay using a panel of STEC isolates.

MATERIALS & METHODS

- **Isolates:** An in-house library of 142 diarrrheagenic *E. coli* isolates from various host organisms (human, cow, pig, rabbit, and buffalo) was used for this study. Of these, 100 isolates belong to the Big 6 serotypes (O26, O45, O103, O111, O121, and O145) and O157 which are classified as adulterants by the United States Department of Agriculture (USDA). Isolates were obtained from Thomas S. Whittam STEC Center at Michigan State University, Statens Serum Institut, USDA, and the Uniformed Services University of the Health Sciences.
- **SHIGA TOXIN QUIK CHEK™:** “QUIK CHEK” assays were performed according to package insert instructions for broth culture testing.
- **CHROMagar™ STEC:** Medium was prepared per CHROMagar leaflet instructions.
- **Vero Cell Assay:** Toxin production was confirmed by Vero cell cytotoxicity assay and positive cultures neutralized with specific antisera against Stx1 and Stx2 to confirm Shiga toxin cytotoxicity. This is the recognized gold standard for Shiga toxin detection.

EXPERIMENTAL APPROACH

- Perform Vero Cell Assay
- Screen on SHIGA TOXIN QUIK CHEK
- Visually inspect CHROMagar™ STEC plates
- Streak CHROMagar™ STEC plate
- Incubate overnight (225RPM @ 37°C)
- Incubate one colony into GN broth
- Incubate 4hr (225RPM @ 37°C)
- Streak Blood agar plate
- Incubate overnight @ 37°C
- Inoculate 0.4mL of starter culture into new GN broth
- Chop Meat Stock (n = 142)
- Inoculate overnight @ 37°C
- Inoculate one colony into GN broth
- Perform Vero Cell Assay

RESULTS

- **QUIK CHEK vs. Vero**
  - n=142  
  - Stx + 83 0  83% 0  0
  - Stx - 0 59
  - Sensitivity: 100%
  - Specificity: 100%
  - PPV: 100%
  - NPV: 100%
  - Correlation: 100%

- **CHROMagar™ STEC vs. Vero**
  - n=142  
  - Stx + 67 11  81% 48
  - Stx - 16
  - Sensitivity: 81%
  - Specificity: 81%
  - PPV: 86%
  - NPV: 75%
  - Correlation: 81%

- **Serotype Isolates**
  - SHIGA TOXIN QUIK CHEK™
  - Sensitivity
    - O4 1
    - O15 1
    - O22 1
    - O26 14
    - O36 1
    - O45 7
    - O55 15
    - O91 1
    - O103 10
    - O104 2
    - O111 28
    - O113 2
    - O121 5
    - O128 14
    - O145 6
    - O149 1
    - O154 1
    - O157 30
    - O174 1
    - Unknown 1
  - Total (n)= 142

COMMENTS

- Overall, the SHIGA TOXIN QUIK CHEK™ assay out performed the CHROMagar™ STEC when compared to a Vero cell cytotoxicity neutralization assay (gold standard).
- Recent work with whole genome sequencing of STEC outbreak isolates has shown that aside from the production of Shiga toxin, no single virulence factor is common to all pathogenic STEC isolates, therefore, detecting the toxins themselves is of the utmost importance.
- Current guidelines for STEC testing suggest broth enrichment of samples which leads to multi-day procedures. The SHIGA TOXIN QUIK CHEK™ assay can be performed following broth enrichment or using direct faecal samples. Screening faecal samples directly may be beneficial to look for the presence of toxin quickly before attempting multi-day culture testing. This would allow for prompt diagnosis and eliminate unnecessary culturing attempts.
- CHROMagar™ STEC may prove useful for obtaining isolates for downstream testing, but the better sensitivity and specificity of the SHIGA TOXIN QUIK CHEK™ assay allows for detection of a wider range of virulent STEC.

REFERENCES


DISCLOSURE

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