STEC Isolation Using Chromogenic Coliform Agar with the SureTect PCR Workflow

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INTRODUCTION

Isolation media such as CT-SMAC are effective for the detection of *Escherichia coli* O157:H7 but there are many challenges concerning the detection of non-O157:H7 Shiga toxin-producing *Escherichia coli* (STEC). There are few different biochemical traits between isolates of *E. coli* that can be utilized in plating methods to rapidly identify presence of non-O157:H7 STEC which makes STEC confirmation challenging.

The purpose of this study was to evaluate the performance of Thermo Scientific™ Oxoid™ Chromogenic Coliform Agar (CCA) for detection of non-O157:H7 STEC, including from meat, vegetables, fruit and dairy samples.

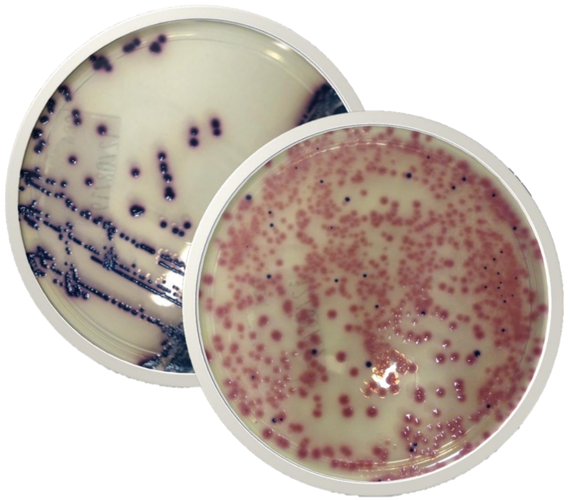


Figure 1: Typical *E. coli* growth on CCA (left) and in mixed culture with background flora (right)

Almost all *E. coli* possess the enzyme D-galactosidase and D-glucuronidase. These cleave the chromogenic compounds Salmon GAL and X-glucuronide to create dark blue/violet colonies. Coliforms do not possess D-glucuronidase and only cleave Salmon GAL, producing pink colonies.

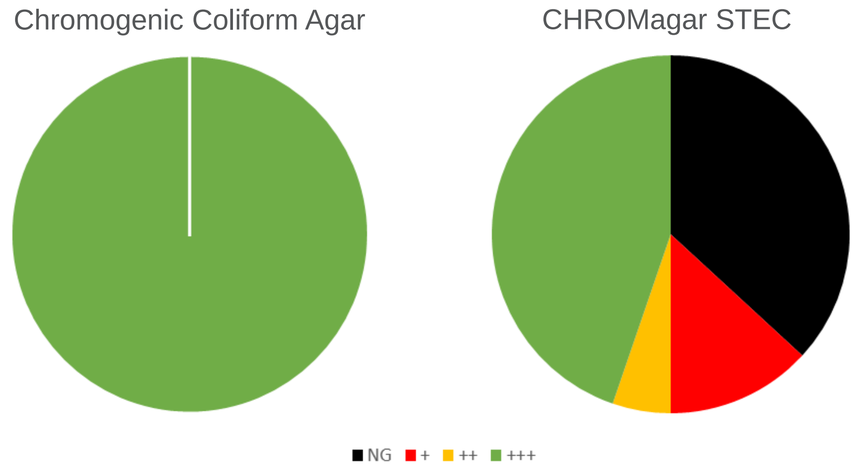
MATERIALS AND METHODS

Thirty-eight non-O157:H7 from the top six STEC serogroups were grown on CCA and CHROMagar STEC to test inclusivity of each medium. Food sample enrichments, including artificially contaminated with a low level (1-5 CFU/sample), were streaked directly onto the same agars for detection. Where direct streaking failed to isolate presumptive positive colonies, an IMS method was employed based upon results from a PCR screen for present serogroups.

RESULTS

Table 1: Serogroup breakdown of inclusivity data

Serogroup	Chromogenic Coliform Agar			CHROMagar STEC		
	+ to +++	No Growth	Correct Colour	+ to +++	No Growth	Correct Colour
O26 (n=10)	10	0	9	9	1	9
O45 (n=4)	4	0	4	1	3	1
O103 (n=7)	7	0	7	4	3	4
O111 (n=6)	6	0	5	4	2	6
O121 (n=6)	6	0	6	1	5	1
O145 (n=5)	5	0	5	5	0	5



Key: Growth in tertiary bed (+++), secondary bed (++), primary bed (+) and no growth (NG).

Figure 2: Proportion of test isolates that displayed various levels of growth on CCA and CHROMagar STEC

CCA displayed positive growth for 100% of non-O157:H7 isolates and displayed typical morphology of *E. coli* for 94.7% (36/38). CHROMagar failed to support growth for 12/38 isolates and displayed reduced growth for a further five. When isolating from food matrices, CCA displayed advantages over other media types with vegetables, fruit and unpasteurized dairy whereas CHROMagar showed advantages in selectivity when testing with meat.

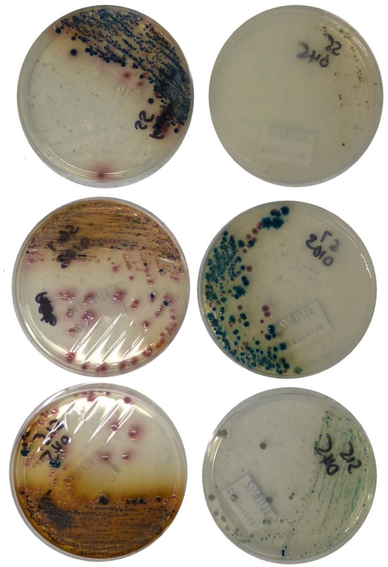


Figure 3: Difference in growth between positive samples of raw dairy for CCA (left) and CHROMagar (right)

Both CCA and CHROMagar were successful in correctly detecting STEC in this example from raw dairy (Figure 3). Differences in usability were noted including that CCA was less selective but allowed more growth of target colonies while CHROMagar removed more background flora and lowered the number of target colonies present.

CONCLUSIONS

The data indicates that CCA is a suitable alternative agar to previously identified options in confirmation workflows for the isolation of STEC from food samples. Using both CCA and CHROMagar STEC together provides both high selectivity and strong growth promotion for isolation of STEC from food samples.

TRADEMARKS/LICENSING

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