CASE STUDY

GENE-UP® & Cattle Hide Swabs:

Real-World Comparative Performance for EHEC Detection

A study in conjunction with a leading US beef producer compared Shiga-toxin producing *Escherichia coli* (STEC) detection results from 62 cattle hide samples using both GENE-UP® and widely used current PCR methods. Current detection methods are not always accurate due to cross-reactions of naturally occurring pathogens on cattle hide. These tests have a high rate of unconfirmed screening results, which causes costly delays and recalls. USDA-FSIS considers the presence of *E. coli* O157:H7 in addition to the top six STEC serogroups: O26, O45, O103, O111, O121, and O145 in non-intact raw beef as adulterant. Each of these serogroups can cause severe illness and even death. Therefore, producers need the fastest, most sensitive, and most accurate STEC detection system. bioMérieux's GENE-UP® EHEC Series Solutions combines automated immunoselection with a highly specific and sensitive real-time PCR, using VIDAS® and Fluorescence Resonance Energy Transfer (FRET) technology.

BACKGROUND

- The presence of seven *E. coli* O-serogroups in beef trim is considered adulteration by the USDA-FSIS
- Current commercial PCR systems screen a high rate of unconfirmed positives
- The average direct cost of a recall due to adulteration is an estimated to be $10M (GMA 2012)

PROBLEM

- The economic impact of an inaccurate EHEC detection, even presumptive, is significant for beef producers
- Current PCR systems are less accurate than traditional culture testing due to cross-reaction issues

PURPOSE

- Comparatively evaluate different commercial PCR methods to screen for *E. coli* O157:H7 and the top six STEC organisms
- Identify PCR-based methods that more accurately screen results reducing false positives

PIONEERING DIAGNOSTICS

METHOD

All 62 cattle hide swabs under comparative testing were originally screened as positive for EHEC by current commercial PCR methodologies. For comparison, each enrichment was then screened for the presence of EHEC using the GENE-UP® EHEC Series Protocol. An upfront VIDAS®-based immunoselection and lysis (ESP1) was performed, followed by PCR analysis for *eae, stx,* EH1, and the top six serogroup PCR assays. All samples were culturally isolated, confirmed, and serotyped following bioMérieux protocols. Results were then compared against the original PCR results.

RESULTS

Using GENE-UP® EHEC series protocol, a wide variety of organisms were screened and isolated from each sample (Figure 1), However, only 9.7% of the cattle hide swab enrichments were found to be positive for organisms that are considered critical from a regulatory standpoint (Figure 2). GENE-UP® EHEC Series had 100% agreement, while the competitive PCR methods only had a 9.7% agreement to the culture confirmed results (Table 1). These findings are in accordance to what was observed in a previous study (Bosilevac et al., 2019). This study demonstrates the combination of VIDAS® and GENE-UP® EHEC offers a superior detection model.

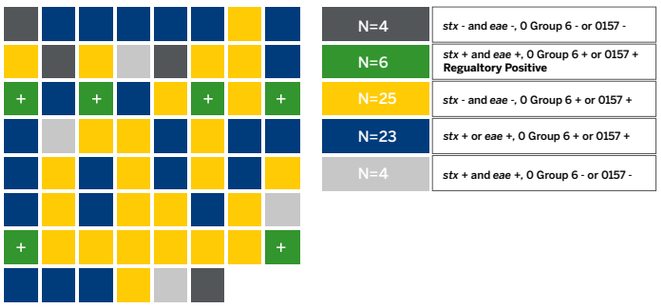

Figure 1: Summary of confirmed and serotyped results per sample

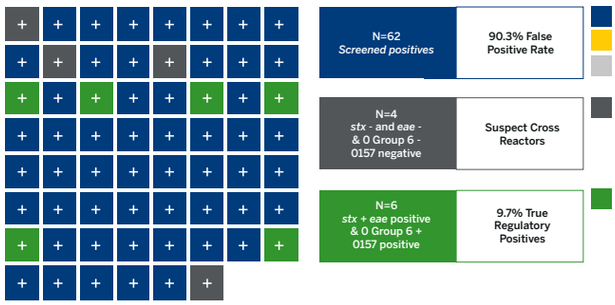

Figure 2: Visual summary of the competitive PCR system screening status for each hide swab sample

	BIOMERIEUX GENE-UP®	COMPETITIVE PCR METHODS
EHEC POSITIVE*	6	62
EHEC NEGATIVE*	56	0
AGREEMENT	100%	9.7%

Table 1: A comparison of screened PCR results.

* Culturally confirmed and serotyped via validated bioMérieux protocols.

Reduce false positive screens

Detects regulatory relevant genes

Specific and precise FRET-based PCR

Selects only the O groups of interest and targets *eae/stx*

Ensure confidence in screened result

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