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Recovery and Detection of Shiga-Toxin Producing Escherichia coli and Salmonella spp. from Polyolefin Cloth (Microtally®) Using Non-Proprietary Media and a Loop-Mediated Isothermal DNA Amplification Method

Rocio Foncea¹, Gabriela Lopez Velasco¹, Christina Barnes¹, Eric Chlan¹

^{1.} Neogen Corporation, 620 Lesher Pl. Lansing, MI 48912, United States

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In January of 2023, the United States Department of Agriculture Food Safety and Inspection Service (USDA FSIS) issued a notice describing the use of a non-destructive surface sample collection method to collect raw beef manufacturing trimming and bench trim verification samples equivalent to the N60 excision sampling. Thus, it is important to assess the recovery and detection of foodborne pathogens from cloth material before a method is implemented for routine use in testing laboratories. The objective of this study was to assess the dual detection of low levels Shiga-Toxin Producing E. coli (STEC) and Salmonella from polyolefin cloth (Microtally) utilizing buffer peptone water ISO (BPW-ISO) and loop-mediated isothermal DNA amplification (LAMP). Beef trim was obtained from local supermarkets and placed into a tray (~10Kg) to mimic the sample collection process. The cloths (N=60) were used to massage the surface of the beef trim as described by FSIS (Notice 05-23). After massaging cloths were artificially spiked with a theoretical amount of 1 CFU of *E. coli* O157:H7 (*stx+/eae+*) S. enterica sv Typhimurium (ATCC14028) and refrigerated 48h for conditioning. Thirty swabs were enriched with 200 of BPW-ISO at 41.5°C and testing aliquots were analyzed after 8, 10, 12 and 24h. Additionally, thirty swabs were enriched with 200mL of modified tryptic soy broth at 41.5°C for 24 h. All samples were cultured confirmed. Based on our results, the relative limit of detection was calculated for the two enrichment schemes following AOAC guidelines, the p-value when comparing molecular screening and culture confirmation of *E. coli* O157, STEC and *Salmonella* was >0.05. The shortest enrichment (8h) evaluated, demonstrated to be sufficient to detect and recover both microorganisms from Microtally® cloths using BPW-ISO and LAMP. The significance of the study lies in the implementation of an enrichment scheme with non-proprietary media and LAMP detection, offering testing laboratories a fast method to analyze raw beef and bench trim verification samples.

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