Roka Bioscience 2014 IAFP Poster Abstract

Use of Non-Proprietary Harmonized Enrichment Media for the Detection of *Escherichia coli* O157:H7, non-O157 STEC, and *Salmonella* in 375-g Beef and Produce Samples with the Atlas® System

Anja Bubeck-Barrett[1], Bettina Groschel[1], Kristin Livezey[1], Hua Yang[1], William Kwong[1], Apolonia Huerta[1], Celina Puente[1], Steve Vaughn[1], Brett Maroni[1], Kevin Tsao[1], Julian Kougl[1], Michele Wisniewski[1], Joe Garcia[1], Bernadine Luong[1], Edgar Kamantique[1], Chad Fleischer[1], Brett Weaver[1], Jarrod Morgan[1], Michael Reshatoff[1], Michael Becker[1], Erin Crowley[2], Patrick Bird[2], Benjamin Bastin[2], Brian Kupski[3], and Wendy McMahon[3]

[1]Roka Bioscience, San Diego, CA [2]Q Laboratories, Inc., Cincinnati, OH [3]Silliker, Inc., Food Science Center, South Holland, IL

Introduction: Rapid methods are necessary for the detection of pathogenic *E. coli* O157:H7, non-O157 STEC, and *Salmonella* in the beef and fresh produce industry at appropriate sample sizes in accordance with industry needs. The use of a non-proprietary, harmonized enrichment media significantly streamlines the testing process for these pathogens as well as considerably reduces cost by eliminating the need to prepare multiple enrichments.

Purpose: To validate the use of a harmonized enrichment media to detect *E. coli* O157:H7, non-O157 STEC, and *Salmonella* in artificially inoculated ground beef and romaine lettuce samples with the Atlas E. coli O157:H7 EG2 and Salmonella G2 Detection Assays.

Methods: 375-g test portions were inoculated with low levels of *E. coli* O157:H7, non-O157 STEC, and *Salmonella* and diluted 1:5 in modified Tryptic Soy Broth + 10 g/L casamino acids (mTSB) for ground beef and Universal Pre-enrichment Broth (UPB) for romaine lettuce. All samples were enriched for 10 to 12 h at 42°C, transferred into a Roka G2 Sample Transfer Tube, and loaded onto the Atlas System. The Atlas System combines target capture, transcription-mediated amplification, and hybridization protection assay. The results of the candidate methods were compared to the FSIS and FDA reference methods.

Results: *E. coli* O157:H7, non-O157 STEC, and *Salmonella* were specifically detected after 10- to 12-h enrichment in 375-g ground beef and lettuce samples. All three Atlas Assays performed equally compared to the respective reference methods determined by POD analysis.

Significance: The Atlas E. coli O157:H7 EG2 Detection Assay, Atlas STEC EG2 Combo Detection Assay, and Atlas Salmonella G2 Detection Assay provide fast and highly accurate detection of *E. coli* O157:H7, non-O157 STEC, and *Salmonella* in 375-g ground beef and lettuce samples using a non-proprietary commercially available media. The harmonized media provides a simplified testing process with significant economic and efficiency advantages along with the highly accurate results required.

For more information or to download the full poster scan here.

20 Independence Boulevard | 4th Floor
Warren, New Jersey 07059

1.855.ROKABIO | 908.605.4700

www.rokabio.com

Roka Bioscience 2014 IAFP Poster Abstract

Method Verification for the Detection of *Salmonella enterica* by the Roka Atlas® Salmonella G2 Detection Assay in Produce Matrices

Tyler P. Stephens, W. Evan Chaney, Joseph Kabala, Tanushree Shah, and Erin Dreyling
Roka Bioscience, San Diego, CA

Introduction: *Salmonella enterica* has been implicated in over 1 million cases of salmonellosis annually in the United States according to the Centers for Disease Control and Prevention (CDC). Consequently, the FDA has increased its attention on produce safety, and FSMA regulations may increase testing volumes, thereby necessitating accurate and rapid methods to provide confident and timely results to the produce industry.

Purpose: The purpose of this study was to verify performance of the Atlas Salmonella G2 Detection Assay on additional produce matrices not previously submitted for AOAC-RI validation.

Methods: Produce matrices evaluated were spinach, iceberg lettuce, red leaf lettuce, fresh blueberries, and scallions. For each matrix, six 25-g samples and one 375-g sample were prepared and inoculated with ~8 CFU/sample of *Salmonella* Newport (Cornell S5-436), and six 25-g samples were prepared as uninoculated matrix controls. All samples were enriched with Universal Pre-enrichment Broth (UPB) at a 1:9 sample to media ratio and incubated at 42 ± 2°C for 10 and 24 h. Samples were collected at 10 and 24 h according to the Atlas Salmonella G2 Detection Assay Product Insert and loaded onto the Atlas System. All samples collected at 24 h underwent culture confirmation according to the FDA BAM Chapter 5 reference method.

Results: The Atlas System method specifically detected *Salmonella enterica* in all inoculated samples at 10 and 24 h, and all inoculated samples were culture confirmed. All uninoculated samples were negative according to the Atlas Salmonella G2 Detection Assay method at 10 and 24 h and culture at 24 h.

Significance: The Atlas Salmonella G2 Detection Assay method was verified for the propagation and detection of *Salmonella enterica* in five additional produce matrices using a 10-h enrichment and total time to result of 13.5 h with no false-positive, false-negative, or inhibited results. The results substantiate the efficiency and accuracy of the Atlas Salmonella G2 Detection Assay on foods outside the current AOAC-RI-approved matrices.

For more information or to download the full poster scan here.

20 Independence Boulevard | 4th Floor
Warren, New Jersey 07059

1.855.ROKABIO | 908.605.4700

www.rokabio.com

Roka Bioscience 2014 IAFP Poster Abstract

Accuracy of Molecular Screening Methods for the Detection of *Salmonella enterica* in Production Ground Poultry Samples

W. Evan Chaney, Tyler Stephens, Joseph Kibala, Tanushree Shah, and Erin Dreyling
Roka Bioscience, San Diego, CA

Introduction: Accurate detection methods are essential in the poultry industry to detect *Salmonella enterica* in ground poultry products as a means to monitor baselines and verify process controls. Accuracy may be impacted by factors including sample preparation and detection method utilized.

Purpose: The purpose of these studies was to comparatively evaluate the accuracy of the Atlas® Salmonella SEN Detection Assay and BAX® *Salmonella* Assays (original and real-time) for the detection of *Salmonella enterica* in production ground poultry products.

Methods: Ground poultry products, consisting of ground turkey (n=39) and ground chicken (n=4), were collected by two poultry processors. Sample weights ranging from 25 to 325 g were enriched utilizing Buffered Peptone Water (BPW) in a 1:10 dilution at 35 ± 2°C. For the Atlas Salmonella SEN Detection Assay method, 400 µL was transferred into sterile Roka Sample Transfer Tubes at 12 h (Processor B) and at 18 to 24 h (Processor A). Both processors conducted BAX analysis as per routine procedure on the paired enrichments at 20 to 24 h. Duplicate 1.5- to 2.0-mL aliquots from the enrichments were sampled into sterile vials, maintained at 4°C, and shipped to Roka Bioscience for cultural analysis. Atlas Salmonella SEN Detection Assay and culture results were reported to collaborators at which time BAX results were disclosed.

Results: In total, *Salmonella enterica* was identified in 27.91% of samples by culture analysis, in 25.58% by the Atlas Salmonella SEN Detection Assay, and in 16.28% by the BAX Assay. The BAX Assay reported five false-negative results and the Atlas Salmonella SEN Detection Assay reported one false-negative result as compared to culture on ground poultry samples. Percent agreement of the screening tests to culture for the Atlas Salmonella SEN Detection Assay and BAX *Salmonella* Assays were 97.67% and 88.37%, respectively.

Significance: Screening method performance and accuracy can be adversely affected by matrix characteristics, variation in sample preparation procedures, and the rapid detection method utilized.

For more information or to download the full poster scan here.

20 Independence Boulevard | 4th Floor
Warren, New Jersey 07059

1.855.ROKABIO | 908.605.4700

www.rokabio.com

Roka Bioscience 2014 IAFP Poster Abstract

Accuracy of Molecular Screening Methods for the Detection of *Salmonella enterica* in Production Poultry Rinse Samples

W. Evan Chaney, Tyler Stephens, Joseph Kibala, Tanushree Shah, and Erin Dreyling
Roka Bioscience, San Diego, CA

Introduction: Accurate detection methods are essential in the poultry industry to detect *Salmonella enterica* in whole carcass rinse samples as a means to monitor prevalence and verify process controls. Variability in sample preparation and detection method may influence accuracy of results.

Purpose: The purpose of these studies was to comparatively evaluate the accuracy of the Atlas® Salmonella SEN Detection Assay and the BAX® *Salmonella* Assay for the detection of *Salmonella enterica* in carcass rinse samples from multiple processors.

Methods: Three poultry processors collected routine rinse samples according to USDA FSIS MLG 4.07 by rinsing each carcass with ~400 mL of Buffered Peptone Water (BPW). A 30 ± 0.6 mL post-rinse aliquot was combined with 30 ± 0.6 mL of sterile BPW and enriched for 20 to 24 h at 35 ± 2°C. Each sample was analyzed by Atlas Salmonella SEN Detection Assay, BAX Assay, and culture methods. For the Atlas Salmonella SEN Detection Assay method, 400 µL of enrichment was transferred into Roka Sample Transfer Tubes. For the BAX method, 1.5 to 2 mL of enrichment was transferred into sterile cryovials and held at 4°C. Samples were processed at Roka Bioscience on the Atlas System according to the approved Atlas System method and by culture according to MLG 4.07. Processors sampled paired enrichments for BAX analysis according to routine procedure. BAX results were disclosed after Atlas Salmonella SEN Detection Assay method and culture results were reported.

Results: The Atlas Salmonella SEN Detection Assay method reported one false-negative, whereas the BAX method reported nine false-negative and one false-positive results compared to culture. Percent agreement between culture and Atlas Salmonella SEN Detection Assay and BAX *Salmonella* Assays were 99.31% and 93.06%, respectively. Processors A, B, and C contributed 25.69%, 27.78%, and 46.53% of 144 samples. All discrepant results were attributed to the 25.69% of samples prepared by processor A.

Significance: Screening method performance and accuracy may be adversely impacted by the proficiency or complexity of operator handling requirements.

For more information or to download the full poster scan here.

20 Independence Boulevard | 4th Floor
Warren, New Jersey 07059

1.855.ROKABIO | 908.605.4700

www.rokabio.com