United States Department of Agriculture Food Safety and Inspection Service, Office of Public Health Science

SOP No: MLG 41 Appendix 2.04 Page 1 of 2		
Title: FSIS Laboratory Specific Flow Chart for Campylobacter jejuni/coli/lari Enrichment Analysis		
Revision: .04	Replaces: MLG 41 Appendix 2.03	Effective: 03/07/22

Laboratory Guidebook Notice of Change

Chapter new, **revised**, or archived: MLG 41 Appendix 2.04

Title: FSIS Laboratory Specific Flow Chart for Campylobacter jejuni/coli/lari Enrichment Analysis

Effective Date: 03/07/22

Description and purpose of change(s):

This flow chart was revised in association with the revisions made in MLG 41 to reduce sample turn-around time.

- The double strength blood-free Bolton enrichment Broth plus supplements was replaced with Hunt Broth plus supplements, antibiotics and blood components.
- The 48 \pm 2h enrichment media incubation time was reduced to 24 \pm 2 h.
- Screened negative results can now be reported in 2 days and confirmed positive results can now be reported in 4 days.

FSIS Laboratory Specific Flow Chart for Campylobacter jejuni/coli/lari Enrichment Analysis

PRIMARY Poultry Rinse or Sponge or Raw Poultry Product Samples + Enrichment Broth (Sample receipt temperature of $0 - 15^{\circ}C$ is required) **ENRICHMENT** Transfer 30 mL of poultry rinse + 30 mL of Hunt Broth 1 to a vented culture flask or Whirl-Pak® bag and mix well. Day 1 Transfer 30 mL of raw poultry product diluted 1:6 in BPW² + 30 mL of Hunt This chart represents the best-case Broth to a vented culture flask or Whirl-Pak® bag and mix well. scenario. Analysis may take longer due Add 25 mL of Hunt Broth to carcass sponge sample containing 25 mL of to normal analytical circumstance such as re-streaking isolates for purity. transport media and mix well. Incubate for 24 ± 2 hrs @ 42 ± 1 °C in a sealed container applying appropriate microaerobic conditions. Perform Campylobacter screen using 3MTM Molecular Detection System **SELECTIVE ISOLATION MEDIA** Report as Negative Day 2 Streak sample to sufficiently dried Campy-Cefex plate for isolation. Place plate into a sealed container applying the appropriate microaerobic conditions. Incubate for 48 ± 2 hrs @ 42 ± 1 °C. When typical Campylobacter colonies are found, pick at **READING** least one typical colony from the plate in preparation for RESULTS isolate confirmation. Perform Bruker® MALDI Biotyper analysis. **CONFIRMATORY ANALYSIS** Day 4 **CONFIRMED INCONCLUSIVE CONFIRMED POSITIVE** NEGATIVE Whole Genome Sequencing (WGS) is **CONFIRMED** performed on confirmed **POSITIVE** positive and inconclusive **ISOLATES** isolates at each FSIS FSL³ and results are reported. ¹ Hunt Broth = Hunt Broth plus supplements, antibiotics, and blood ² BPW = Buffered peptone water

³ FSIS FSL = Food Safety and Inspection Service Field Service Laboratories