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

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Title: FSIS Laboratory Regulatory Sample Pathogen Methods Table and Definitions		
Revision: **1/31/2012**	Replaces: NA	Effective: 1/31/2012

Analyte	**Microbiology Laboratory Guidebook Chapter #**	**Screen Test**	**Confirmatory Tests** (following culturing on tube and plating media; and for *E. coli* O157:H7 IMS bead capture)	Days to Reporting**: Screen Negative**	Days to Reporting**: Potential + Result**	Days to Reporting **Presumptive + Result**	Days to Reporting **Final + Result**
E. coli O157:H7	MLG 5A MLG 5	BAX® PCR Assay (alternatives: lateral flow devices – RapidChek® or Transia™)	Serological confirmation: *E. coli* O157:H7 latex agglutination test kit (RIM® *E. coli* O157:H7 Latex Test Kit, REMEL) Biochemical confirmation: Vitek®GN/GNI/GNI Plus Cards (bioMerieux) Shiga toxin/toxin genes confirmation: Shiga Toxin test kit [Premier® EHEC, cat. # 608096 (Meridian Diagnostics, Inc)] or detection of toxin genes by PCR if needed	**Day 2**	**Day 2** (Limited distribution)	**Day 3**	**Day 4-6**
Non E. coli O157 STEC	MLG 5B	Multiplex RT PCRs: eae, stx then wzx genes	Latex agglutination Multiplex RT PCR typical colonies eae, stx then wzx genes Biochemical confirmation: Vitek® GN/GNI/GNI Plus Cards (bioMerieux)	**Day 2**	**Day 2** (Limited distribution)	**Day 3**	**Day 4-6**
Listeria monocytogenes	MLG 8 MLG 8A	BAX® PCR Assay	Tumbling Motility observation Biochemical confirmation: MICRO-ID® *Listeria*, *ListeriaAPI*®,Vitek 2 CAMP/CAMP Factor Test β-lysin CAMP factor discs (Remel) with MICRO-ID®; Genetic Identification Testing if needed for speciation – GenProbe Accuprobe® Ribosomal RNA-based *L. monocytogenes*-specific test system	**Day 3**	**NA**	**Day 4-5**	**Day 5-8**

Analyte	**Microbiology Laboratory Guidebook Chapter #**	**Screen Test**	**Confirmatory Tests** (following culturing on tube and plating media; and for *E. coli* O157:H7 IMS bead capture)	Days to Reporting**: Screen Negative**	Days to Reporting**: Potential + Result**	Days to Reporting **Presumptive + Result**	Days to Reporting **Final + Result**
Salmonella spp.	MLG 4 MLG 4C	BAX® PCR Assay	Serological confirmation: Somatic(O)Antigen Agglutination Tests (*Salmonella* polyvalent O antiserum); Flagellar (H) Antigen Agglutination Tests (Oxoid *Salmonella* Latex Test) Biochemical confirmation: Vitek® GNI/GNI Plus Cards (bioMerieux)	**Day 2**	**NA**	**Day 5** NA for HACCP	**Day 6-7** Depends on Vitek result available Day 6 PM; Day 7 AM
Campylobacter for Quantitative method	MLG 41	Direct Plating	Typical colonies subject to same day: Microscopic examination Latex agglutination	**Day 3**	**NA**	**NA**	**Day 3**

*Table doesn't include additional non regulatory testing (e.g.NVSL serotyping, PFGE subtyping, *Campylobacter* qualitative testing). On Day 1 sample arrives in the laboratory. Days listed do not include delays (e.g. restreak for purity, rare strains requiring additional testing in Outbreak Section Eastern Laboratory).

Definitions:

Potential positive *E. coli* O157:H7 – Enrichment medium from one or more subsamples yields a positive when screen tested.

Presumptive positive *E. coli* O157:H7 – One or more typical colonies on plating agar agglutinate when tested with O157 antiserum.

Confirmed positive *E. coli* O157:H7 – One or more isolates from the sample is a biochemically identified *Escherichia coli* that is serologically or genetically determined to be "O157" that meets at least one of the following criteria:

1) Positive for Shiga toxin (ST) production
2) Positive for Shiga toxin gene(s) (stx)
3) Genetically determined to be "H7"

Potential positive non *E. coli* O157 STEC – Enriched medium from a sample which yields a positive when screened on real-time PCR for each of the targeted genes (*eae*, *stx*1/2, and *wzx*) of one or more of six non-O157 serogroups (O26, O45, O103, O111, O121, O145).

Presumptive positive non *E. coli* O157 STEC – One or more typical colonies on modified Rainbow agar. Latex agglutination positive.

Confirmed positive non *E. coli* O157 STEC – Latex agglutination positive on Sheep Blood Agar, one or more isolates from the sample is confirmed positive on real-time PCR for the *eae*, *stx*, and *wzx* genes of one or more of six non-O157 serogroups, and biochemically identified as *Escherichia coli*.

Presumptive positive *L. monocytogenes* – A sample from which one or more typical colonies produces beta hemolysis on Horse Blood Agar.

Confirmed positive *L. monocytogenes* – A beta hemolytic isolate is Camp test positive, shows tumbling motility (optional) and is characterized biochemically as *L. monocytogenes*. Ribosomal RNA testing is occasionally required to resolve atypical strains.

Presumptive positive *Salmonella* spp. – A sample yields one or more isolates which show typical appearance on TSI and LIA slants and agglutinate salmonella somatic antisera.

Confirmed positive *Salmonella* spp. – *Salmonella* O group positive isolates are characterized biochemically as *Salmonella* spp.

Confirmed positive *Campylobacter* – Typical colony morphology, microscopic ID, latex agglutination positive for *C. jejuni, C. coli, and/or C. lari.*