

U.S. DEPARTMENT OF AGRICULTURE  
FOOD SAFETY AND INSPECTION SERVICE

**Pasteurized Egg Product Recognized Laboratory  
(PEPRLab) Program  
Salmonella Laboratory Quality Assurance Program  
Checklist**

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a valid OMB control number. The time required to complete this information collection is estimated to average 60 minutes per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection information.

LABORATORY NAME:

LABORATORY DIRECTOR:

LABORATORY REPRESENTATIVE(S) AT REVIEW:

REVIEWER:

DATE OF REVIEW:

**OPENING QUESTIONS:**

1. Who are your current clients?

Client: \_\_\_\_\_ Establishment No. \_\_\_\_\_

Client: \_\_\_\_\_ Establishment No. \_\_\_\_\_

2. Is this facility an in-plant laboratory?

\_\_\_\_\_

3. On average, how many *Salmonella* tests are conducted per week?

\_\_\_\_\_

4. How many of these tests are on USDA Official Surveillance Samples?

\_\_\_\_\_

5. When was the last time a pasteurized egg product sample was found to be positive for *Salmonella*?

\_\_\_\_\_

6. How many *Salmonella* positive pasteurized egg product samples have been found in the last 3 years?

\_\_\_\_\_

7. How soon and to whom were these reported?

\_\_\_\_\_  
\_\_\_\_\_

8. List all personnel involved in the *Salmonella* testing

\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_

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## Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program

### A. PERSONNEL REQUIREMENTS

1. Does the person in charge of microbiology have a baccalaureate degree in biology, chemistry, microbiology, food technology, medical technology, or other relevant science with at least 12 semester hours of course work in microbiology and/or at least 4 years of experience working in a public health, medical, food, or other related laboratory?  Yes  No  N/A
2. Are there training/education/experience records available for each analyst?  Yes  No  N/A
3. Is there a formal training program for employees working in microbiology that includes instruction in safety, technical procedures, and use of equipment?  Yes  No  N/A
4. Is there a record kept of this formal training?  Yes  No  N/A

### B. PHYSICAL FACILITIES

A laboratory should have sufficient work and storage space and the facilities to handle the overall workload in order to ensure the quality of work, and safety of the employees.

1. Are floors, benches, and storerooms clean, free of clutter, dust free, and well maintained?  Yes  No  N/A
2. Are the following facilities adequate:
  - a. Sinks?  Yes  No  N/A
  - b. Lighting?  Yes  No  N/A
  - c. Gas outlets/Bacti-cinerator?  Yes  No  N/A
  - d. Electrical outlets?  Yes  No  N/A
  - e. Incubator capacity?  Yes  No  N/A
  - f. Refrigerated storage space?  Yes  No  N/A
  - g. Ventilation?  Yes  No  N/A
3. Is there sufficient bench space for each analyst?  Yes  No  N/A
4. Are bench tops made of impervious materials?  Yes  No  N/A
5. Is the media preparation, glassware washing area separate from the analytical area?  Yes  No  N/A
6. Is unrelated traffic discouraged in the work area, and is the laboratory locked when analysts are not present?  Yes  No  N/A
7. Are samples that are stored at room temperature stored in sealed containers to prevent pests from infesting the laboratory?  Yes  No  N/A
8. Is there a pest control system in place for the laboratory?  Yes  No  N/A

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### C. SAMPLE RECEIPT AND HANDLING

Samples must be submitted to the laboratory in a condition that does not compromise the quality and validity of analytical results, and must be handled after receipt in the laboratory in a manner to maintain sample integrity.

1. Are samples inspected upon receipt in the laboratory for:
  - a. Leakage?  Yes  No  N/A
  - b. Thawed frozen samples?  Yes  No  N/A
  - c. Unsealed or ruptured containers?  Yes  No  N/A
  - d. Spoilage?  Yes  No  N/A
  - e. Evidence of tampering?  Yes  No  N/A
2. Are all samples (including rejected samples) recorded in a login system book, on worksheets, by computer, or in another permanent, accessible format?  Yes  No  N/A
3. Are unacceptable samples rejected for analysis and is the condition recorded in login system book, computer, etc?  Yes  No  N/A
4. If yes, are acceptable samples resubmitted?  Yes  No  N/A
5. Does sample information include at a minimum:
  - a. Lot number?  Yes  No  N/A
  - b. Date of collection?  Yes  No  N/A
  - c. Plant name and/or number?  Yes  No  N/A
  - d. Type of analysis requested?  Yes  No  N/A
  - e. Type of product/state of product?  Yes  No  N/A
  - f. Date of receipt?  Yes  No  N/A
  - g. Condition upon receipt?  Yes  No  N/A
6. Are liquid samples either analyzed on the same day received or refrigerated at 2.0 to 8.0°C until analyzed?  Yes  No  N/A
7. Is the maximum turnaround time for sample analyses:
  - a. 4 to 5 days for negatives (cultural isolation method)?  Yes  No  N/A
  - b. 5 to 7 days for positives (cultural isolation method)?  Yes  No  N/A
  - c. 2 to 3 days for negatives using a rapid screening procedure?  Yes  No  N/A
8. Are frozen samples either rapidly thawed in a water bath (preferably with agitation) at less than 45°C until the slush ice stage (for no longer than 30 minutes) or thawed at refrigerator temperatures (2.0 to 8.0°C) for no longer than 18 hours in the original container?  Yes  No  N/A
9. Are thawed frozen samples analyzed immediately?  Yes  No  N/A
10. Are dried egg samples analyzed upon receipt or stored at room temperature for no more than 24 hours?  Yes  No  N/A
11. If analysis of dried egg samples is delayed more than 24 hours, are they refrigerated at 2.0 to 8.0°C?  Yes  No  N/A
12. Are samples placed in appropriate storage after analysis and are negatives retained for at least one day after reporting and are positives retained for at least 30 days?  Yes  No  N/A

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### D. QUALITY ASSURANCE

A written quality assurance program for the laboratory should be available, and the quality control records should be reviewed at least weekly by the supervisor. Proper care of laboratory instruments and equipment is essential for satisfactory performance of laboratory tests. Maintenance must be performed on a regular basis by trained individuals. Monitoring must be performed at stated intervals by laboratory personnel to assure on-going reliability.

1. Is there a written Quality Assurance Program?  Yes  No  N/A
2. Is there documentation showing that records of procedure controls, instrument functions, scheduled maintenance, and equipment temperatures are reviewed at least weekly?  Yes  No  N/A
3. Is there a formal training program for employees working in microbiology that includes instruction in safety, technical procedures, and use of equipment?  Yes  No  N/A
4. Are quality control and maintenance records maintained for at least 3 years?  Yes  No  N/A
5. Is there a system for routinely reviewing the work to detect clerical or analytical errors, or unusual results?  Yes  No  N/A
6. Does the system provide for timely correction of errors?  Yes  No  N/A
7. Are laboratory results and analysts' worksheets retained for each sample including negative samples for a period of at least three years?  Yes  No  N/A
8. Are there records of internal reviews and, when indicated, corrective action(s) taken in response to unacceptable check-sample results?  Yes  No  N/A
9. Are thermometers checked for accuracy against a thermometric standard (National Institute of Standard and Technology/formerly National Bureau of Standards) before placing them in service?
  - a. thermometers calibrated annually?  Yes  No  N/A
  - b. Are correction factors listed on each thermometer?  Yes  No  N/A
  - c. Is the NIST traceable thermometer sent in for calibration at least every 5 years?  Yes  No  N/A
10. Are mechanical pipetting devices calibrated at least semi-annually to check accuracy of delivery?  Yes  No  N/A
11. Is there a scheduled, written preventative maintenance program for laboratory equipment and instruments?  Yes  No  N/A

12. Does the preventative maintenance program include the following:

#### I. AUTOCLAVES:

- a. Are acceptable temperature ranges defined for autoclaves?  Yes  No  N/A
- b. Are there recording thermometers, calibrated dials, or other recording devices present on autoclaves?  Yes  No  N/A
- c. Are temperatures checked and recorded at each use and is there documentation of corrective action for out-of-range results?  Yes  No  N/A
- d. Are autoclaves monitored with biological indicators at least monthly and are they monitored each time used with a physical indicator (indicator tape)?  Yes  No  N/A

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### II. WATERBATHS:

- a. Are thermometers suspended in distilled water?  Yes  No  N/A
- b. Are acceptable temperature ranges defined and available for waterbaths?  Yes  No  N/A
- c. Are temperatures checked and recorded at least daily, and is there documentation of corrective action for out-of-range results?  Yes  No  N/A
- d. Are water baths clean and free of debris, and is the water changed regularly?  Yes  No  N/A

### III. INCUBATORS:

- a. Are thermometers suspended in an approliquid such as sterile glycerin, distilled water or other acceptable medium?  Yes  No  N/A
- b. Are acceptable temperature ranges defined and available for each incubator?  Yes  No  N/A
- c. Are temperatures checked and recorded at least daily, and is there documentation of corrective action for out-of-rang temperatures?  Yes  No  N/A

### IV. REFRIGERATORS/FREEZERS:

- a. Are thermometers suspended in an appropriate?  Yes  No  N/A
- b. Are acceptable temperature ranges defined and available for refrigerators/freezers?  Yes  No  N/A
- c. Are temperatures checked and recorded at least daily and is there documentation of corrective action for out-of-range temperatures?  Yes  No  N/A

### V. SPECTROPHOTOMETERS AND PHOTOMETRIC READERS:

- a. Are manufacturer's operation requirements followed for the spectrophotometer and photometric reader?  Yes  No  N/A
- b. Is the instrument calibrated according to manufacturer's requirement or kit manufacturer's requirements?  Yes  No  N/A

### VI. BALANCE:

- a. Is the balance checked with a certified weights at least weekly?  
**NOTE:** A 2000 gram balance must have a sensitivity of 0.1 grams with a 200 gram load?  Yes  No  N/A
- b. Is the balance checked annually by an authorized service representative using certified weights that are traceable to the National Institute of Standards and Technology?  Yes  No  N/A

### VII. pH METER:

- a. Are pH meters standardized with at least two appropriate standard buffer solutions covering the range of intended use prior to use and are the results recorded?  Yes  No  N/A
- b. If pH readings are going to be taken intermittently throughout the day, is the pH meter re-calibrated with fresh portion of buffers before each use?  Yes  No  N/A
- c. Are pH meter electrodes checked each time they are used to see if they are filled and not cracked?  Yes  No  N/A

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### E. MEDIA AND REAGENTS

All media, reagents, and chemicals must be prepared correctly, stored under appropriate conditions, and tested with reference organisms to assure satisfactory performance.

1. Are all purchased media, chemicals and solutions labeled with the date received and an expiration date?  Yes  No  N/A
2. Are all in-house prepared media, reagents, and solutions labeled with the name of product and expiration date?  Yes  No  N/A
3. Do media records contain complete QC information for each batch, including pH, sterility, and productivity? If pH paper strip is used for pH determination, the pH paper has to cover the pH range of use with pH gradation value  $\leq 0.2$  pH unit.  Yes  No  N/A

4. Are media, reagents, and/or solutions stored under appropriate conditions (i.e. refrigerated, away from daylight, in a cool or dry place and in appropriate laboratory containers)?  Yes  No  N/A

**Note:** The shelf life of prepared media will vary. In general, the maximum shelf life of prepared culture media in sealed tubes or bottles is 3 months in the refrigerator (2 - 8°C), or up to 1 month at room temperature (18 - 23°C). Media in vented tubes may be stored for up to 4 weeks if refrigerated or 2 weeks at room temperature. Plating media may be stored in the refrigerator for a maximum of 10 weeks in air-tight bags or for a maximum of 2 weeks if the bags are unsealed.

5. Are outdated materials discarded?  Yes  No  N/A
6. Is a sample of each batch of in-house prepared media checked for the ability to support growth (and for biochemical reactivity/selectivity, as appropriate to the media) by using reference organisms capable of evaluating pertinent characteristics of the media?  Yes  No  N/A

List the *Salmonella* media QA cultures used:

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7. Are reference organisms maintained under refrigeration on agar with at least monthly transfers, or by other appropriate methods?  Yes  No  N/A
8. Is an uninoculated control of each medium used and run concurrently with the sample?  Yes  No  N/A
9. Are all media in satisfactory condition upon visual examination (i.e. uncontaminated, hydrated, smooth, appropriate color and thickness) and results documented for each batch?  Yes  No  N/A
10. Are serological reagents tested with appropriate positive and negative controls? (Note: This may include culture controls, commercially produced antigen, or kit controls)  Yes  No  N/A
11. Are serological reagents refrigerated when not in use, inspected for clarity and color, and discarded when showing any turbidity, flocculation or color change?  Yes  No  N/A

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12. Is Rappaport Vassiliadis broth (e.g. RV10, RVS, or RV Broth) prepared according to manufacturers instructions and autoclaved for 15 minutes at 115 or 116°C (12 lbs)?  Yes  No  N/A  
(NOTE: It is important not to overheat this medium.)
13. Is tetrathionate broth prepared according to manufacturers instructions and heated to a boil?  Yes  No  N/A  
(NOTE: It is important not to overheat this medium.)
14. Is only the basal medium of tetrathionate broth base stored?  Yes  No  N/A
15. Is the iodine - potassium iodide solution added to the tetrathionate broth base on the day of use?  Yes  No  N/A
16. Is selenite cystine broth prepared only by boiling, and is it used on the day of preparation?  Yes  No  N/A
17. Are Bismuth Sulfite Agar (BS) plates prepared, stored, and incubated only as follows:
- a. Are BS plates prepared (20 to 25 ml/plate) from dehydrated media that is smooth, free-flowing, and has been properly stored?  Yes  No  N/A
  - b. Are BS plates used on the day of preparation or no more than 1 day after preparation?  Yes  No  N/A
18. Are double modified lysine iron agar (DMLIA) plates used within three weeks of preparation (for FSIS method)?  Yes  No  N/A
19. In the preparation of XLT4 agar (FSIS METHOD), is one of the following used:
- a. XL Agar Base with Thiosulfate citrate and a 27 % solution (approximate) of the surfactant 7-ethyl-2-methyl-4-undecanol hydrogen sulfate, sodium salt, formerly produced by Union Carbide under the tradename of Tergitol 4?  Yes  No  N/A
  - b. XLT4 Agar Base with the XLT4 Agar Supplement (a 27 % solution (approximate) of the surfactant 7-ethyl-2-methyl-4-undecanol hydrogen sulfate, sodium salt, formerly produced by Union Carbide under the tradename of Tergitol 4).  Yes  No  N/A
20. Does the laboratory have a distillation, ion exchange, filtration, or other system available for producing or purchasing water, free from toxic or nutritive substances, to be used in media or reagent preparation?  Yes  No  N/A
21. Is the distilled water stored properly?  Yes  No  N/A
22. Is the water system monitored at least monthly and/or is there a certificate of analysis for purchased distilled water to ensure that each meet the following criteria:
- a. conductivity (< 1.0 Siemens) or resistivity (> 1 Megohm)?  Yes  No  N/A
  - b. bacteria (<1000 cfu /ml)?  Yes  No  N/A
23. Are other media used for *Salmonella* testing of pasteurized egg products?  
If so, list the media used:  
\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_
24. If so, are these media prepared and stored according to the manufacturer's instructions?  Yes  No  N/A

## Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program

### F. ANALYTICAL PROCEDURES MANUAL

To assure consistent laboratory results, a procedures manual should be available at each work station and should contain all procedures performed in the laboratory. Procedures should be written in sufficient detail to enable the analyst(s) to perform tests without referring to other publications.

(NOTE: Manufacturers' package inserts with specific product use instructions may be used in addition to the manual, but cannot replace the procedures manual.)

A recognized laboratory may use a rapid screening method in their testing program for FSIS official egg product surveillance samples only if that method is either an approved AOAC Official Method of Analysis of the AOAC INTERNATIONAL, validated for egg products, or the FSIS Rapid Screening Method as described in the MLG. All presumptive positives identified by rapid screening methods must be confirmed using one of the three accepted cultural methods listed below. Any recognized laboratory that does not use a rapid screening method in their testing program must use one of the following three cultural methods as their primary protocol for egg product analysis:

1. AMS Laboratory Methods for Egg Products - Section I ('93 rev.) and Section VII ('94 rev.) Reference AOAC 967.26, 967.27, 978.24, 989.12, 991.13.
2. FSIS MLG online, Chapter 4.
3. FDA BAM online, Chapter 5.

1. Is an Analytical Procedures Manual available in the laboratory?  Yes  No  N/A
  
2. For *Salmonella* testing, does the manual contain:
  - a. All of the procedures performed?  Yes  No  N/A
  - b. Only approved or accepted procedures?  Yes  No  N/A
  - c. Criteria for accepting or rejecting samples?  Yes  No  N/A
  - d. A section on media and reagent preparation?  Yes  No  N/A
  - e. Quality control procedures?  Yes  No  N/A
  
3. Does each procedure contain:
  - a. Step-by-step instructions?  Yes  No  N/A
  - b. Sample handling/preservation?  Yes  No  N/A
  - c. Expected reactions/results?  Yes  No  N/A
  - d. Corrective actions to be taken when expected reactions/results are not observed?  Yes  No  N/A
  - e. References?  Yes  No  N/A
  
4. Is the manual reviewed and updated annually?  Yes  No  N/A
  
5. Are changes in procedures approved and initialed by the supervisor?  Yes  No  N/A
  
6. Is there documentation to show that all analysts have read the procedures manual, including any revisions, and that only the most recent revision is being used?  Yes  No  N/A

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### G. PROCEDURES AND METHODS

Routine procedures for *Salmonella* detection permit recovery of small numbers of pathogens or debilitated organisms by pre-enrichment in lactose broth or buffered peptone water (BPW). Selective enrichment and plating procedures following that permit growth of *Salmonella* while limiting the growth of competing non-*Salmonella* organisms naturally present in food samples. Identification of an isolate as a member of the genus *Salmonella* depends on a combination of biochemical and serological parameters.

1. Is at least 100 g of sample tested for official surveillance samples?  Yes  No  N/A
2. Is a positive control culture run along with all *Salmonella* tests through any rapid screening test and confirmation tests?  Yes  No  N/A  
List the *Salmonella* control culture(s) used:  
\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_
3. Is every tube and plate throughout the test procedure appropriately labeled?  Yes  No  N/A
4. For each sample, are records maintained documenting each step of analysis for traceability? (i.e. analyst ID, media/kit/reagent lot number, incubation time and temperatures, equipment ID number, etc.)?  Yes  No  N/A
5. List the cultural method used for analysis and/or confirmation of official surveillance samples.  Yes  No  N/A
6. Is the laboratory using a rapid screening method that is either the FSIS MLG Method or an approved AOAC Official Method, validated for egg products?  Yes  No  N/A  
If yes, list the method below with its AOAC reference number:  
Rapid Screening Method: \_\_\_\_\_  
AOAC Official Method Reference Number: \_\_\_\_\_
7. Prior to implementing a new rapid method, were parallel tests conducted using both the rapid and conventional cultural methods and were the results documented?  Yes  No  N/A
8. In the parallel testing, did the methods show equivalency, agreeing at least 95 percent of the time?  Yes  No  N/A
9. Are all positive results that are obtained by rapid screening methods followed up by subculturing the sample and subsequently performing biochemical and serological identification of any *Salmonella* isolates?  Yes  No  N/A
10. Is the ratio of egg sample to pre-enrichment broth maintained at 1:10?  Yes  No  N/A

**Proceed to question #11 if your lab is using the AMS culture method. Go to question #12 if your lab is using the FSIS, MLG chapter 4 culture method. Go to question #13 if your lab is using the FDA, BAM chapter 5 culture method.**

#### 11. AMS Method - Laboratory Methods for Egg Products (Section I - 1993 rev.) and Section VII - 1994 rev.):

- a. Is the pH of the lactose broth/egg mixtures adjusted to 6.8 +/- 0.2 after being left for 1 hour at room temperature?  Yes  No  N/A

List the method of pH testing used: \_\_\_\_\_

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- b. After 24 +/- 2 hours incubation at 35°C is the lactose broth subcultured by transferring 1 ml into 10 ml of selenite cystine broth and an additional 1 ml into 10 ml of tetrathionate broth?  Yes  No  N/A
- c. After 24 +/- 2 hours incubation at 35°C are the selenite cystine and tetrathionate broths subcultured to selective differential agars, XLD, HE, and BS (or manufacturer' recommendation for rapid tests)?  Yes  No  N/A
- d. After 24 +/- 2 hours incubation at 35°C are up to three typical colonies (if available characteristic of *Salmonella* species, selected from each differential agar plate as follows:
- XLD** - pink/red colonies with/without black centers or all black colonies (atypical strains may appear yellow with or without black centers)?  Yes  No  N/A
- HE** - blue/blue-green colonies with or without black centers or all-black colonies?  Yes  No  N/A
- BS** - brown, black, or grey colonies, usually with a metallic sheen and darkening of the surrounding media or occasionally green colonies?  Yes  No  N/A
- e. Are all BS agar plates examined for typical or suspicious *Salmonella* colonies after 24 +/- 2 hours incubation at 35°C and, if negative, again at 48 +/- 2 hours incubation?  Yes  No  N/A
- f. Go to question #14. (page 19)

### 12. FSIS Method - Microbiology Laboratory Guidebook online (MLG), Chapter 4:

- a. After 20 - 24 hours of incubation at 35 +/- 2°C, is the buffered peptone water-sample mixture subcultured by transferring 0.1 ml. into 10 ml of Rappaport Vassiliadis (RV) Broth and by transferring 0.5 ml into 10 ml of tetrathionate (TT) broth, and are these broths then incubated at 42 +/- 0.5°C?  Yes  No  N/A
- b. After 22 - 24 hours incubation at 42 +/- 0.5°C are TT and RV broths subcultured to selective differential agars, BGS and either DMLIA or XLT4?  Yes  No  N/A
- c. After 18 - 24 hours incubation at 35 +/- 2°C are up to three typical colonies, (if available), characteristic of *Salmonella* species, selected from each differential agar plate as follows:
- BGS** - colonies that are pink and opaque with a smooth appearance and entire edge surrounded by a red color in the medium? (On very crowded plates, look for colonies that give a tan appearance against a green background.)  Yes  No  N/A
- DMLIA** - purple colonies with or without black centers? (Since salmonellae typically reverts to purple.)decarboxylate lysine and ferment neither lactose nor sucrose, the color of the medium  Yes  No  N/A
- XLT4** - black colonies or red colonies with black centers? (The rim of the colony may still be yellow in 24 h; later it should turn red.)  Yes  No  N/A
- d. Are all selective agar plates reincubated for an additional 24 +/- 2 hours and are all initially negative plates, as well as those yielding non-confirmed *Salmonella* colonies from the initial selection reexamined before discarding?  Yes  No  N/A
- e. Go to question #14. (page 17)

### 13. FDA Method - Bacteriological Analytical Manual online (BAM), Chapter 5:

- a. Is lactose broth used for pre-enrichment of dry egg products?  Yes  No  N/A

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- b. Is TSB with ferrous sulfate (35 mg ferrous sulfate per 1000 ml TSB) used for pre-enrichment of liquid egg products?  Yes  No  N/A
- c. Is the pH of the pre-enrichment broth/egg mixtures adjusted to 6.8 +/- 0.2 after being left for hour at room temperature?  Yes  No  N/A
- d. After 24 +/- 2 hours incubation at 35°C is the lactose broth subcultured by transferring 0.1 ml into 10 ml of RV broth and an additional 1 ml into 10 ml of tetrathionate (TT) broth?  Yes  No  N/A
- e. Is the RV broth incubated 24 h +/- 2 h at 42 +/- 0.2°C?  Yes  No  N/A
- f. Is the TT broth incubated 24 h +/- 2 h at 35 +/- 2.0°C?  Yes  No  N/A
- g. After 24 +/- 2 hours incubation are the RV and TT broths subcultured to selective differential agars, XLD, HE, and BS by streaking 10 ul from each broth onto each of the three selective differential agars?  Yes  No  N/A
- h. After 24 +/- 2 hours incubation at 35°C are at least 2 typical colonies (if available), characteristic of *Salmonella* species, picked to TSI and LIA slants from each differential agar plate as follows:
- XLD** - pink/red colonies with/without black centers or all black colonies (atypical strains may appear yellow with or without black centers)?  Yes  No  N/A
  - HE** - blue/blue-green colonies with or without black centers or all-black colonies?  Yes  No  N/A
  - BS** - brown, black, or grey colonies, usually with a metallic sheen and darkening of the surrounding media or occasionally green colonies?  Yes  No  N/A
- i. the BS plates give atypical reactions on TSI and LIA, are at least 2 Are BS plates re-incubated an additional 24 +/- 2 h and, if the original colonies from additional typical colonies picked, if available?  Yes  No  N/A
- j. Are selective agar plates stored at 2 - 8°C until completion of confirmation steps?  Yes  No  N/A
14. If suspicious colonies are not well isolated, are they re-streaked for purification directly onto selective agar plates before inoculating Triple Sugar Iron (TSI) and Lysine Iron Agar (LIA) slants?  Yes  No  N/A
15. Are characteristic colonies inoculated to TSI slants and LIA slants by inoculating the slants in tandem with a single pick from a colony, and by stabbing the butts and streaking the slants in one operation?  Yes  No  N/A
16. After incubation at 35 +/- 2°C for 24 +/- 2 hours with caps loosened, are TSI and LIA slants with the following characteristics of *Salmonella* selected for further analysis:
- a. **LIA** - Alkaline slant and butt (purple throughout) with or without hydrogen sulfide (H<sub>2</sub>S) production?  Yes  No  N/A  
(Note: Some strains will produce an acid butt, along with a typical TSI slant.)
  - b. **TSI** - Alkaline (red) slant and acid (yellow) butt with or without hydrogen sulfide (H<sub>2</sub>S) production?  Yes  No  N/A
17. Are TSI/LIA cultures, which appear to be mixed, streaked for isolation before additional biochemical or serological tests are performed?  Yes  No  N/A
18. Before reporting a presumptive positive sample as negative, at least six TSI/LIA cultures (if available) picked as below are subjected to further biochemical and serological testing :
- a. For FSIS MLG 4 method: are at least three well isolated colonies from each of two plating media picked to TSI/LIA pairs and subject to confirmation testing before a sample is reported as negative?  Yes  No  N/A

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- b. For AMS or FDA method, are at least two well isolated colonies from each of three plating media picked to TSI/LIA pairs and subject to confirmation testing before a sample is reported as negative?  Yes  No  N/A
19. Is a rapid/miniatuized biochemical test system used for identifying *Salmonella*?  
If yes, list the test system below with its AOAC reference number:  
Biochemical Test System: \_\_\_\_\_  Yes  No  N/A  
AOAC Reference Number: \_\_\_\_\_  Yes  No  N/A
20. Are the manufacturers' guidelines for miniaturized biochemical systems followed for inoculum preparation incubation, and interpretation of results?  Yes  No  N/A
21. Are sufficient biochemical tests performed to presumptively identify atypical isolates as *Salmonella*? (i.e. urease, dulcitol, lactose, and sucrose fermentation, and malonate utilization)  Yes  No  N/A
22. When performing slide agglutination tests, are all materials and equipment brought to room temperature before testing?  Yes  No  N/A
23. Is a saline control included to detect autoagglutination when performing the polyvalent or group somatic (O) antigen slide agglutination test?  Yes  No  N/A
24. Are polyvalent flagellar (H) antigen screening tests performed by a tube method using formalinized cultures prepared from:  
a. Brain-heart infusion broth incubated at 35°C for 4 to 6 hours for same-day testing?  Yes  No  N/A  
b. Trypticase soy broth incubated 24 hours at 35°C for next-day testing?  Yes  No  N/A
25. For H antigen testing is a negative control of formalinized saline with the formalinized culture included in the testing?  Yes  No  N/A
26. Are H antigen tests incubated at 48 - 50°C for 1 hour?  Yes  No  N/A
27. Are diluted *Salmonella* H antisera prepared in quantities sufficient only for daily use, and any remaining diluted antisera discarded at the end of the day?  Yes  No  N/A
28. If the Oxoid kit or SSI H antiserum is used, are manufacturer's instructions followed?  Yes  No  N/A

Safety issues are not within the scope of the PEPRLab Program audit. Therefore, the laboratory is not required to report a corrective action for any observations and/or recommendations resulting from this part of the review. This segment is conducted out of concern for the health and safety of laboratory personnel.

U.S. DEPARTMENT OF AGRICULTURE  
FOOD SAFETY AND INSPECTION SERVICE

## Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program

### H. SAFETY:

Facilities need to be designed and equipped to meet established OSHA safety standards. Protective equipment should be available to personnel and a comprehensive safety program should be included in laboratory procedures.

1. Is there an ongoing, documented safety education program that includes, but is not limited to, instruction on:
  - a. Location and use of fire extinguishers, blankets, and other safety equipment?  Yes  No  N/A
  - b. Fire drills and evacuation routes?  Yes  No  N/A
  - c. Handling emergency situations?  Yes  No  N/A
  - d. Basic first aid procedures?  Yes  No  N/A
  - e. CPR training?  Yes  No  N/A
  - f. The labeling of all cancer suspect agents?  Yes  No  N/A
  - g. Lifting heavy items?  Yes  No  N/A
  - h. "Right to Know" laws?  Yes  No  N/A
2. Is there a safety manual available in the laboratory?  Yes  No  N/A
3. Does it include procedures for:
  - a. Handling spills of contaminated materials?  Yes  No  N/A
  - b. Disposal of biological waste?  Yes  No  N/A
  - c. Disposal of chemical waste?  Yes  No  N/A
  - d. Handling toxic materials?  Yes  No  N/A
4. Are Materials Safety Data Sheets (MSDS) available in the laboratory for all chemicals used in the laboratory?  Yes  No  N/A
5. Is there a designated safety officer in the laboratory?  Yes  No  N/A
6. Does the safety officer conduct periodic safety inspections using a checklist?  Yes  No  N/A
7. Are safety deficiencies and corrective actions documented?  Yes  No  N/A
8. Are accidents documented and reported to the safety officer?  Yes  No  N/A
9. Is emergency medical help readily available if needed by laboratory personnel?  Yes  No  N/A
10. Are emergency phone numbers (i.e. fire, ambulance, police) posted in a conspicuous place on or near the phone?  Yes  No  N/A
11. Are personnel ever alone in the laboratory?  Yes  No  N/A
12. Does the laboratory have at least two exits and are all exits and hallways free of obstructions?  Yes  No  N/A
13. Can the doors be locked from both sides?  Yes  No  N/A

## Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program

14. Are the following in the laboratory:
- a. Fire extinguishers (CO<sub>2</sub>, dry chemical)?  Yes  No  N/A
  - b. Fire blanket?  Yes  No  N/A
  - c. Eyewash station?  Yes  No  N/A
  - d. Overhead shower?  Yes  No  N/A
  - e. Fire alarm system?  Yes  No  N/A
  - f. Sprinkler system?  Yes  No  N/A
  - g. First aid kit?  Yes  No  N/A
15. Are fire extinguishers and other safety equipment regularly inspected, certified to be in working order, and their condition documented?  Yes  No  N/A
16. Is an EPA-approved disinfectant available to clean biohazardous spills and disinfect bench tops daily?  Yes  No  N/A
17. Is the disinfectant prepared and used according to the manufacturers instructions?  Yes  No  N/A
18. Are bio-hazardous materials discarded in leak-proof, tear-resistant plastic bags marked with a biohazard symbol?  Yes  No  N/A
19. Are biohazardous waste materials steam sterilized at 121°C for at least 45 minutes, with biohazard bags vented to effect complete sterilization as required by the manufacturer, or else incinerated prior to disposal in landfills?  Yes  No  N/A
20. Are biohazardous materials removed from the laboratory daily and contained in a manner to minimize accidental spills during storage and transport and to exclude rodents and vermin?  
(i.e. Bags are tied and placed in covered, rigid containers such as buckets, cans, or cardboard boxes, and liquids are placed in capped or tightly stoppered bottles or tubes.)  Yes  No  N/A
21. Are janitors and other maintenance personnel instructed in proper methods of disposal, and are disposal areas located well away from the building and protected from trespassers?  Yes  No  N/A
22. Are personnel instructed not to taste chemicals at all, and not to directly smell chemicals?  Yes  No  N/A
23. Is mouth pipetting strictly prohibited (with no exceptions for sterile solutions)?  Yes  No  N/A
24. Are eating, drinking, and smoking prohibited in the laboratory, and is labware prohibited from use for any of these purposes?  Yes  No  N/A
25. Is food prohibited from refrigerators that are used for reagents, samples, etc.?  Yes  No  N/A
26. Are personnel instructed to wash hands after handling samples, working with cultures, handling chemicals, and/or before leaving the laboratory?  Yes  No  N/A
27. Are laboratory personnel required to confine long hair?  Yes  No  N/A
28. Are aprons, gloves, and goggles available for handling hazardous materials?  Yes  No  N/A
29. Are heat-resistant gloves available near the autoclave and in the media preparation area?  Yes  No  N/A
30. Are laboratory coats or other protective clothing worn only in the laboratory?  Yes  No  N/A
31. Are bunsen burners turned off when not in use?  Yes  No  N/A

## Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program

32. Are chipped, broken, or etched glassware discarded in a specially marked, puncture proof, sealed container?  Yes  No  N/A
33. Is broken glassware always cleaned up with a dust pan/brush and never picked up with the hands?  Yes  No  N/A
34. Are heavy plastic carriers available for transporting acids or other corrosive chemicals?  Yes  No  N/A
35. Are bottles of acid (HC1) always tightly capped and rinsed on the outside after being used and/or before being opened?  Yes  No  N/A
36. Have laboratory personnel been taught to always pour acid into water, never water into acid?  Yes  No  N/A
37. Is a safety cabinet or room available for storing large containers of hazardous chemicals?  Yes  No  N/A
38. Are electrical connections covered with a heavy rubber coating?  Yes  No  N/A
39. Are extension cords grounded and, if running across the floor, are they taped down?  Yes  No  N/A
40. Are all electrical cords, receptacles, and switches in good condition and located away from water sources?  Yes  No  N/A
41. Does the laboratory use mercury thermometer(s)? If so, is a mercury spill kit available?  Yes  No  N/A
42. If located near water sources, are electrical outlets protected with ground-fault circuit interrupters?  Yes  No  N/A

**Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program**

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**I. SUMMATION AND COMMENTS**

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**Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program**

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**REFERENCES**

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1. EPA Guide for Infectious Waste Management, May 1987. United States Environmental Protection Agency, National Technical Information Service, Springfield, VA.
2. Handbook of Microbiological Media, 3<sup>rd</sup> Edition, 2004, CRC Press, Boca Raton, FL.
3. Biosafety in Microbiological and Biomedical Laboratories, 4<sup>th</sup> ed. May 1999. Centers for Disease Control, U.S. Department of Health and Human Services, Atlanta, GA.
4. Compendium of Methods for the Microbiological Examination of Foods, 4<sup>th</sup> ed. 2001. APHA, Technical Committee on Microbiological Methods for Foods, Washington, DC.
5. Good Laboratory Practice Regulations, Code of Federal Regulations (CFR), 21 CFR Part 58, U.S. Food and Drug Administration, 5600 Fishers Lane, Rockville, MD.
6. Difco & BBL Manual, 1st ed., 2003, Becton, Dickson and Company, Sparks, Maryland.
7. Official Methods of Analysis of AOAC INTERNATIONAL, Current AOAC Internet Version
8. Laboratory Methods for Egg Products - Section I (1993 revision) and Section VII (1994 revision), U. S. Department of Agriculture, Agriculture Marketing Service, Washington, D. C.
9. Microbiology Laboratory Guidebook online (MLG), Chapter 4, U. S. Department. of Agriculture, Food Safety and Inspection Service, Washington, D.C.
10. Bacteriological Analytical Manual online (BAM), Chapter 5, U.S. Food and Drug Administration, Washington, D.C.

**Pasteurized Egg Products Recognized Laboratory (PEPRLab) Program**

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**Instructions for completing the form**

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1. Answer all questions on the checklist by checking the appropriate response or by filling in the blank. Responses are based on observations or information supplied by laboratory personnel. Questions pertaining to services, equipment, instruments, methods or procedures not used routinely by the laboratory should be marked as not applicable (N/A).

2. Scan and submit the completed form to: [PEPRLab@fsis.usda.gov](mailto:PEPRLab@fsis.usda.gov)

3. Alternatively, mail the completed form to:

Program Manager, Pasteurized Egg Products Recognized Laboratory Program  
USDA, FSIS, OPHS, LQAD  
950 College Station Road  
Athens, Georgia 30605  
Phone: (706) 546-3559  
Fax: (706) 546-3453