Sampling Instructions for Enforcement, Investigations and Analysis Officers (EIAOs)
Food Safety Assessment RTE Product – Fully Cooked, Not Shelf Stable (FSA 03G)

I. Purpose

1. This document provides MPID Enforcement, Investigations and Analysis Officers (EIAOs) with instructions for scheduling and collecting samples under the Routine risk-based *Listeria monocytogenes* (RLm) sampling program. The RLm sampling program includes collection of product, food contact surface, and non-food contact surface samples. All samples are tested for the presence of *Listeria monocytogenes* (*Lm*) and non-*Listeria monocytogenes Listeria* species.

2. The following instructions are for use with:
   - MPID-Form 27.9 for Fully Cooked, Not Shelf Stable Product
   - MPID-Form 27.10, Product Contact Swabs
   - MPID-Form 27.11, Non-Contact Swabs

II. RLm Sample Scheduling

1. MPID EIAOs are to perform a Public Health Risk Evaluation (PHRE) as per FSIS Directive 5100.4, *Public Health Risk Evaluation Methodology*.

2. As part of the PHRE or Food Safety Assessment (FSA) for an establishment producing post-lethality exposed ready-to-eat (RTE) products the EIAO shall determine the product type and amount to be sampled as discussed in section **IV. Determination of Product to Sample.**

   NOTE: If the establishment's HACCP Establishment Size is listed as Small or Large in the Establishment Profile in PHIS, this will require correlation with the Area Supervisor to determine if the establishment produces post-lethality exposed RTE product on multiple production lines.

3. The EIAO is to correlate with the EIAO Supervisor if an establishment is listed as Small or Large in the PHIS Establishment Profile and if multiple production lines are used in the production of post-lethality exposed RTE product. The purpose of the correlation will be to decide if more than one sampling unit will need to be collected.

4. The EIAO is to correlate with the Area Supervisor to determine when the product type selected (i.e. 03G) for RLm sampling is produced.

5. The EIAO shall email MPIDSampling@ncagr.gov with the following information to schedule an RLm sample collection activity:

   i. The establishment name where sampling will occur.

   ii. The week that sample collection is desired.

   NOTE: It is best to provide a few options for collection dates as the laboratory may not be able to analyze samples during certain weeks.

iii. The product type that will be sampled:

a. 03G, Fully Cooked, Not Shelf Stable.

iv. The number of sample units to be collected.

a. One sample unit or 5 product samples, 10 product contact swab samples and 5 non-product contact swab samples in very small establishments or

b. A maximum of two sample units or 10 product samples, 20 product contact swab samples and 10 non-product contact swab samples in small establishments.

III. Prior to Sample Collection

1. Check contents of the shippers upon arrival. The shippers collectively should contain all items referenced below.

Per box received from the laboratory (the laboratory will send 3-5 boxes):

- □ 1 Foam Plug
- □ 1 FedEx US Airbill or FedEx Billable Stamp
- □ 2 ice packs
- □ 1 Cardboard mat
- □ 1 large white plastic bag (to place each sample box into while being stored in the establishment)
- □ 1 sample security seal (to seal each sample box after appropriately packaged for shipment or drop-off)

To facilitate product sample collection, for each sampling unit there will be:

- □ 1 Request for Sampling Form (1 form completed for all product samples)
- □ 5 clear plastic Whirl-Pak® bags (to place each individual product sample collected in)
- □ 5 sample security seals (to place over the flap of each Whirl-Pak® bag containing a product sample)

To facilitate product contact swab collection, for each sampling unit there will be:

- □ 1 Request for Sampling Form (1 form completed for all product contact swabs)
- □ 10 sample security seals (one seal for each sponge-stick)
- □ 10 sterile sponge-sticks
- □ 10 clear plastic Whirl-Pak® bags (to place each bag with sponge-stick into after sampling)
- □ 10 pairs of sterile gloves (one pair per sponge-stick)
- □ 10 tubes of Dey-Engley Broth (one tube per sponge-stick)

To facilitate non-contact swab collection, for each sampling unit there will be:

- □ 1 Request for Sampling Form (1 form completed for all non-contact swabs)
- □ 5 sample security seals (one seal for each sponge-stick)
- □ 5 sterile sponge-sticks
- □ 5 clear plastic Whirl-Pak® bags (to place each bag with sponge-stick into after sampling)
- □ 5 pairs of sterile gloves (one pair per sponge-stick)
- □ 5 tubes of Dey-Engley Broth (one tube per sponge-stick)

2. Contact the Raleigh Office at MPIDSampling@ncagr.gov immediately if the security seal is broken or missing on the outside of the box, supplies are missing, gloves are contaminated, or the boxes show evidence of being tampered with when you receive the supplies.

3. **At least 24 hours prior to sample collection**:

   i. Pre-chill all shipping containers (with lid open) in a cooler or refrigerator. Place the box in the enclosed large plastic bag before placing the shipping container in a cooler or refrigerator at the establishment. The shipping containers can get dirty during shipment to and from the laboratory. The bag will hopefully keep the shipping container from introducing any contaminants into the cooler. This should also assist in reducing moisture damage to the boxes.

   ii. Freeze the ice packs outside of the shipping container.

   NOTE: If the ice packs are left inside of the shipping container to freeze, the insulation insert, and foam plug will increase the amount of time it takes for the ice packs to completely freeze. This could result in the sample(s) arriving too hot for the laboratory to analyze, and therefore, rejected sample(s).

   iii. Refrigerate the Dey-Engley broth.

4. Place all other materials in a secure location until sample collection.

IV. Determination of Product to Sample

1. EIAOs are only to collect samples on days and shifts when the establishment is producing MPID regulated, post-lethality exposed (PLE), RTE meat or poultry products.

2. Collect five individual product samples per sampling unit from a particular line and processing lot.

   i. Using the maximums in section b. below, EIAOs are to collect each sampling unit from a single, PLE, RTE line.

   ii. When determining the number of sample units to collect, EIAOs are to:

      a. Sample a maximum of two lines on which PLE, RTE product is produced (two sample units) in small establishments; or

      b. Sample one line on which PLE, RTE product is produced (one sample unit) in very small establishments.

      NOTE: For small establishments, the sample units provided above is a maximum; it could be determined that collecting only one sample unit is necessary. Establishment HACCP size is defined as: small establishments – 10 or more employees but fewer than 500; and very small establishments – fewer than 10 employees or annual sales of less than $2.5 million. This information can be found in the Establishment Profile in the Public Health Information System (PHIS).

3. Collect products from the highest risk alternative and the highest risk PLE, RTE production line using the instructions in a and b below, whenever possible;

i. Select product from the highest-risk *Lm* control alternative (Risk: Alternative 3 > Alternative 2b> Alternative 2a > Alternative 1) as follows:

   a. For each sampling unit, select product from only one *Lm* control alternative, processing line, and production lot. For example, if one sampling unit is collected and the establishment produces products under all three alternatives, then select Alternative 3 product; and

   b. If more than one sampling unit is collected, then product from more than one alternative may be selected. However, all the individual samples in each sampling unit must be from the same *Lm* control alternative, processing line, and sampled lot.

4. When selecting production lines, consider other factors, including, but not limited to:

   i. Sampling history (results from both MPID sampling and the establishment's own sampling);

   ii. Noncompliance history or trends (e.g., noncompliance with 9 CFR 416, 417, and 430);

      a. Evidence of *Lm* harborage;

      b. Past outbreaks or links to illness; and

      c. Observations from IPP.

      NOTE: See FSIS Directive 10,240.5, Verification Procedures for Enforcement, Investigations and Analysis Officers for the *Listeria Monocytogenes* Regulation and Routine Risk-Based *Listeria monocytogenes* Sampling Program for more information regarding the selection of product for sampling.

V. Product Sampling Procedure (MPID-Form 27.9)

1. When possible, product eligible for this sampling procedure should be collected in its final intact packaged form.

   i. If the establishment only packages products in containers larger than one quarter pound (1/4 lb.), the establishment may short-weight or slack-fill a container. In such cases, the sample must be produced in the same way as the rest of the product it represents; the only difference would be the size of the package.

2. Collect five (5) individually packaged one quarter pound (¼ lb.) samples, for each sampling unit, of the chosen Fully Cooked, Not Shelf Stable RTE product. The total weight of the product submitted to the laboratory for analysis should be at least one and a quarter pounds (1 ¼ lbs.).

   i. The individual product samples should be collected at different times throughout the production of a specific sampled lot after all interventions listed in the establishment's HACCP plan and/or hazard analysis have been applied. However, if an establishment has any intervention based on microbiological test results, do not wait for the establishment to receive microbiological test results before sending the sample to the laboratory.

3. Each of the individual samples, collected in their final intact packaging, should be placed in a separate Whirl-Pak® bag provided by the lab.

i. To open the Whirl-Pak® bag, remove the tear strip from the top, grasp the two small white tabs and pull apart.

ii. Once the sample is placed in the Whirl-Pak® bag, close it by folding the top of the bag down a few times, and folding the wire attached to the bag inward against the bag.

iii. Place a security seal over the top, so that the seal is sticking to the sides of the bag and over the top of the fold.

4. The EIAO should ensure that sample integrity and security is maintained at all times.

5. Refer to section **VIII. Completion of the Applicable MPID Forms** and section **IX. Final Packing and Shipment of the Samples** for additional instructions for the product collected.

NOTE: Product samples may be collected on a different day than the FCS and NFCS environmental samples, as long as the same production lot is represented by all three sample types within a sampling unit. For example:

An establishment produces tamales from scratch. All post-lethality exposure of the product to the environment occurs on a Monday. The establishment packages the product Monday afternoon; however, it is too late in the day to collect and ship the sample to the laboratory. The contact and non-contact swabs would be collected on Monday, but the product would be held on Monday, then collected and shipped on Tuesday.

VI. Collection Procedure for Product Contact Swabs (MPID Form 27.10) and Non-Contact Swabs (MPID Form 27.11)

1. For product contact swabs, the EIAO is to:

   i. Collect ten (10) swab samples per sampling unit.

      a. Collect most product swabs during establishment operations.

         (1) EIAOs may collect some product contact swabs at the end of pre-operational sanitation activities, before the start of production; and

         (2) EIAOs are to take post-operation samples as quickly after operations end as practical and before the implementation of establishment sanitation procedures.

2. For non-contact swabs, the EIAO is to:

   i. Collect five (5) swab samples per sampling unit.

      a. The samples should be collected in areas of the establishment where products are being processed, stored, or held, including smokehouses, coolers, and production rooms for the targeted production line(s).

   NOTE: Use a separate sponge-stick and separate pair of sterile gloves to swab each location. Do not swab more than one surface with the same sponge. Wash hands and change sterile gloves with each new swab following the instructions provided below.

Instructions for sample collection *without* a partner:

EIAOs are to:

1. Wash and sanitize hands and arms up to the mid-forearm. Dry with disposable paper towels.
2. Sanitize a work surface area using the establishment's cleaner / sanitizer. Let surface dry.
3. Using the ungloved hands:
    i. Open the bag containing the sponge-stick by pulling off the clear perforated strip at the top of the bag.
    ii. Grab and pull apart the white tabs to open the mouth of the bag.
    iii. Aseptically pour the Dey Engley broth into the bag to hydrate the sponge-stick, being careful not to contaminate the broth or sponge during this process.
    iv. By only touching the exterior of the bag, press the mouth of the bag back together.
    v. Evenly moisten the sponge-stick by massaging the outside of the bag in the area where the sponge is located.
    vi. Position the sponge-stick so that the handle sticks out of the bag. Press the top of the bag together around the handle.
4. A sterile glove shall be placed on the hand being used for swabbing:
    i. Position the glove package so that the L and R (L=Left and R=Right) are facing you. When the package is open, the gloves are folded, forming a cuff on the sleeve and are lying palm up. Leave them in the package until ready for use.
    ii. Hold the glove for the hand that will be used for swabbing by the inside cuff area, and insert the hand into the glove, palm side up, lifting the glove from the package.
    iii. Pull the glove completely on, touching only the fold cuff with your ungloved hand. Unroll the fold of the glove. Do not touch any non-sterile surface such as clothes, counter tops or the outside of the Whirl-Pak® bag with the sterile glove.
    iv. The other hand can be left ungloved to allow for the touching of non-sterile surfaces and materials.
5. Using the gloved hand:
    i. Carefully remove the sponge-stick from the bag by grasping the handle and swab the selected area being certain to utilize aseptic techniques. Do not allow your hand to go past the thumb stop on the handle of the sponge-stick.
    ii. Swab the chosen surface of the facilities and equipment in the post-lethality exposed RTE production area using firm and even pressure and by using the following technique:
        a. Swab vertically (approximately 10 times) on one side of the sponge,
        b. Swab horizontally (approximately 10 times) on the opposite side of the sponge and
        c. Swab diagonally, using the same surface side as was used for the horizontal swabbing (approximately 10 times).

NOTE: If possible, swab at least a 1'x1' square of the area chosen. If the available surface area is less, swab the entire surface.

6. Open the bag that the sponge-stick originally came out of and insert the sponge back into the bag.

7. Grip the sponge-stick through the bag and bend the handle of it back and forth with slight force, while gripping the sponge through the bag. The stick should break easily within the sponge. Discard the broken handle.

8. Squeeze as much air out of the bag as possible and fold the top of the bag down at least 3 times. Fold in the tabs to lock the fold in place.

9. Refer to section **VII. Labeling and Packaging Procedure for Product Contact Swabs (MPID Form 27.10) and Non-Contact Swabs (MPID Form 27.11)** and **VIII. Completion of the Applicable MPID Forms** to ensure adequate labeling of the bag directly containing the swab and the associated form to be submitted to the laboratory.

Instructions for sample collection *with* a partner:

Person A represents the sample collector.
Person B represents the assisting person.

1. *Persons A* and *B* shall wash and sanitize hands and arms up to the mid-forearm. Dry with disposable paper towels.

2. *Person A* or *B* shall sanitize a work surface area using the establishment's cleaner / sanitizer. Let surface dry.

3. *Person A* opens sterile gloves onto this surface and applies them using aseptic technique.

    i. Position the glove package so that the L and R (L=Left and R=Right) are facing you. When the package is open, the gloves are folded, forming a cuff on the sleeve and are lying palm up.

    ii. Hold the glove for the hand that will be used for swabbing by the inside cuff area, and insert the hand into the glove, palm side up, lifting the glove from the package.

    iii. Pull the glove completely on, touching only the fold cuff with your ungloved hand. Unroll the fold of the glove. Do not touch any non-sterile surface such as clothes, counter tops or the outside of the Whirl-Pak® bag with the sterile glove.

    iv. The other hand can be left ungloved to allow for the touching of non-sterile surfaces and materials.

4. *Person B*:

    i. Opens the sterile sponge-stick bag and Dey-Engley broth.

    ii. Aseptically pours the Dey-Engley broth into the bag, being careful not to contaminate the broth or sponge during this process.

    iii. Should hydrate the sponge-stick by massaging the outside of the bag in the area where the sponge is located.

    iv. Opens the bag to expose the handle of the sponge-stick without touching it.

5. *Person A*:

    i. Grasps the handle of the sponge-stick to take it out of the bag.

    ii. Swabs the chosen non-contact surface of the facilities and equipment in the post-lethality exposed RTE production area using firm and even pressure and by using the following technique:

        a. Swab vertically (approximately 10 times) on one side of the sponge,

        b. Swab horizontally (approximately 10 times) on the opposite side of the sponge and

        c. Swab diagonally, using the same surface side as was used for the horizontal swabbing (approximately 10 times).

6. *Persons A and B* collectively:

    i. *Person B* holds the sterile sponge-stick bag and *Person A* inserts the sponge.

    ii. *Person B* grabs the sponge from the outside of the bag while person A bends the handle of it back and forth with slight force. The stick should break easily within the sponge after a few bends. Discard the broken handle.

    iii. *Person B* should squeeze as much air out of the bag as possible, close it by folding the top of the bag down a few times, and folding the wire attached to the bag inward against the bag.

7. Refer to section **VII. Labeling and Packaging Procedure for Product Contact Swabs (MPID Form 27.10) and Non-Contact Swabs (MPID Form 27.11)** and **VIII. Completion of the Applicable MPID Forms** to ensure adequate labeling of the bag directly containing the swab and the associated form to be submitted to the laboratory.

    NOTE: For more information on utilizing aseptic techniques see the Aseptic Sampling document included in the Sampling section of IPP – Help (must be connected to the VPN to access). For visual instructions on donning sterile gloves, see the How to Put on Sterile Gloves: Safe & Easy Steps article on WikiHow.

VII. Labeling and Packaging Procedure for Product Contact Swabs (MPID Form 27.10) and Non-Contact Swabs (MPID Form 27.11)

1. Each bag containing the sponge-stick shall contain a label on the outside of it for recording the following information:

    i. Date: record the date of collection

    ii. Environmental Testing (circle): If not already done by the laboratory, circle the “Contact” option for product contact swabs or the “Non-Contact” option for non-product contact swabs

    iii. Establishment #: record the establishment’s assigned inspection number

    iv. Location code: Write the sample/swab number (swab _____ of _____) that corresponds to the number on the sample request form. A very brief description of the actual sample site can be included as well.

    v. Insp. Initials: record the collector’s initials

NOTE: If the label is not included on one or more of the sponge-stick bags, utilize a permanent marker to record the information requested in 1. (a-f) above.

2. Each bag containing the sponge-stick shall be placed in a separate Whirl-Pak® bag.

3. Squeeze as much air out of the Whirl-Pak® bag as possible and fold the top of the bag down at least 3 times. Fold in the tabs to lock the fold in place.

4. Place a security seal over the top, so that the seal is sticking to the sides of the bag and over the top of the fold.

VIII. Completion of the Applicable MPID Forms

1. Complete the applicable sections of MPID-Form 27.9 for "Food Safety Assessment RTE Product Fully Cooked, Not Shelf Stable (FSA 03G)."

    i. Fill in the Collector Name, Collection Date, Shipping Date, Product Name, Production Date, Production Lot Code, Production Volume, along with the establishments' Contact Name and Phone Number or Email. Check yes or no to indicate whether product is being held.

2. Complete the applicable sections of MPID Form 27.10 for "Food Safety Assessment (FSA PC) – Product Contact Swabs"

    i. Fill in the Collector Name, Collection Date, Shipping Date, Production Date, Production Lot Code, along with the establishments' Contact Name and Phone Number or Email. Check yes or no to indicate whether product is being held.

3. Complete the applicable sections of MPID Form 27.11 for "Food Safety Assessment (FSA NC) – Non-Contact Swabs"

    i. Fill in the Collector Name, Collection Date, Shipping Date, Production Date, Production Lot Code, along with the establishments' Contact Name and Phone Number or Email. Check yes or no to indicate whether product is being held.

IX. Final Packing and Shipment of the Samples

1. Shippers shall be packed to return to the laboratory in the following manner:

    i. One (1) to three (3) shippers shall contain the Product Samples
    ii. One (1) shipper shall contain the Product Contact Swabs
    iii. One (1) shipper shall contain the Non-Contact Swabs

2. Place the samples and other contents in the shipper in the following order:

    i. Place the ice packs in the bottom of each shipper.
    ii. Place the cardboard mat on top of the ice packs in each shipper.
    iii. Add the samples.

iv. Place the styrofoam plug on top.

v. Place the completed Request for Sampling Forms on top of the styrofoam plug.

vi. Use the second security seal to seal the inside flap of the shipping container. Your sample will not be processed by the laboratory without the appropriate seals affixed. See the MPID Notice entitled Instructions for Use of Sample Seals.

vii. Close the shipping container using the velcro tabs.

3. If you are performing sampling procedures in a location convenient to the ASC Laboratory (4400 Reedy Creek Rd., Raleigh, NC 27607), the shipping containers (with sampling contents) can be dropped off in lieu of scheduling FedEx pickup by following the procedures below:

   i. At least one day prior to sample collection, email MPIDSampling@ncagr.gov requesting to drop the supplies off at the ASC Laboratory on a certain date. A member of the team will reach out to the laboratory to notify them of the request and will confirm with the requester whether the drop-off request has been approved.

   ii. The Shipping/Receiving loading dock is open Monday through Friday from 7:30 AM to 4:30 PM, excluding state holidays. Do not leave the shipping containers at the loading dock outside of these hours.

4. If you are performing sampling procedures in a location not convenient for sample drop-off at the laboratory, complete the pre-addressed FedEx US Airbills or FedEx Billable Stamps. Attach the completed Airbill or Billable Stamp to the exterior of each sample box.

   i. Samples must be picked up by FedEx on the same day that the sample is collected (exceptions below). However, if this is not possible, and a sample must be held overnight, it should be refrigerated**.**

      a. Exceptions:

         (1) If a product is too warm to be mailed the same day as it is produced, refrigerate the sample, then collect and mail the sample the next day.

         (2) Samples ready for collection after FedEx pickup Monday through Wednesday should be held refrigerated, collected and shipped the next day.

         (3) If a product is made only on Friday, refrigerate the sample, then collect and mail the sample via FedEx on Monday.

   ii. Ship the sample as soon as FedEx service is available by calling 1-800-463-3339 or drop off at FedEx drop off site.

   NOTE: Samples should **not** be collected and shipped the day before a State Holiday or on a Friday. They will be discarded. If product is made only on Friday, then the sample should be held, and then collected and mailed from the establishment via FedEx on Monday.