

UNITED STATES DEPARTMENT OF AGRICULTURE
FOOD SAFETY AND INSPECTION SERVICE
WASHINGTON, DC

FSIS NOTICE

04-20

1/10/20

LYMPH NODE TESTING TO COMPLEMENT THE NATIONAL ANTIMICROBIAL RESISTANCE MONITORING SYSTEM PROGRAM

NOTE: DO NOT IMPLEMENT THIS NOTICE UNTIL FEBRUARY 1, 2020.

I. PURPOSE

This notice provides instructions to Public Health Veterinarians (PHVs) at beef slaughter establishments for collecting a paired sample set, which includes mesenteric lymph nodes and cecal samples from the same carcass for microbiological and antimicrobial resistance testing under the FSIS National Antimicrobial Resistance Monitoring System (NARMS) Program.

II. BACKGROUND

A. Lymph node testing to complement the NARMS program is an effort to link NARMS mesenteric lymph node sampling data with associated cecal information from the same animal (dairy or beef cattle). This program is being conducted in support of the [National Action Plan for Combating Antibiotic Resistant Bacteria \(CARB\)](#) and the [USDA Antimicrobial Resistance \(AMR\) Action Plan](#).

B. Under this program, PHVs in beef and dairy livestock slaughter establishments subject to NARMS sampling will collect mesenteric lymph node samples from cattle (Dairy Cow, Beef Cattle). PHVs will collect mesenteric lymph node samples for microbiological testing from the same carcass as the cecal samples. The sample(s) will be shipped to the assigned field service laboratory (FSL) for analysis.

III. ESTABLISHMENT ELIGIBILITY FOR THE FSIS NARMS SAMPLING PROGRAM

Establishments that slaughter beef and dairy cattle will be subject to sampling under the Lymph Node Testing to Complement the NARMS Program. Beef and dairy cattle livestock slaughter establishments are subject to sampling based on the NARMS sampling eligibility criteria described in [FSIS Directive 10,100.1, FSIS Cecal Sampling Under the National Antimicrobial Resistance Monitoring System \(NARMS\) Surveillance Program](#). Sampling eligibility and frequency will be based on data in the Public Health Information System (PHIS).

IV. REVIEW OF TRAINING MATERIALS

Upon issuance of this notice, PHVs assigned to establishments subject to Lymph Node Testing to Complement the NARMS Program are to be familiar with the information provided in this notice and the following FSIS issuances:

1. [FSIS Directive 13000.2, Performing Sampling Tasks in Official Establishments Using the Public Health Information System](#);

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OPI: OPPD

2. [FSIS Directive 7355.1](#), *Use of Sample Seals for Laboratory Samples and Other Applications*; and
3. [FSIS Directive 10,100.1](#).

V. AWARENESS MEETING WITH PLANT MANAGEMENT

A. Upon issuance of this notice, PHVs at beef slaughter establishments are to review the information in this notice with establishment management during the next weekly meeting. The PHVs are to advise establishment management that sample collection for Lymph Node Testing to Complement the NARMS Program will begin on or after February 1, 2020.

B. During the meeting, the PHV is to inform the establishment that livestock carcasses selected for directed lymph nodes sampling are NOT required to be held or controlled by the establishment pending test results.

C. During the meeting, PHVs will work with establishment management to identify the following:

1. The point in the slaughter process where the viscera/mesenteric lymph nodes will be retrieved for sampling;
2. The location in the establishment where the sample selection will be performed;
3. The establishment employee designated to retrieve the selected viscera and transport it to the pre-determined sample collection location in livestock slaughter establishments; and
4. A cleanable work surface (i.e., table) in the sample collection location for staging the sampling supplies.

VI. LYMPH NODE SAMPLING TASK ASSIGNMENT

A. **PHIS Paired Sampling Tasks.** Notification of sampling requests will appear as alerts through PHIS. Paired sampling requests (one for NARMS cecal sampling and one for mesenteric lymph node sampling) will appear separately as distinct directed tasks on the establishment task list in PHIS. Both the NARMS cecal task and the lymph node task will denote that they are part of a paired sampling task. The following project codes are to be included with a paired lymph node task:

Project Code	Paired Project Code	Slaughter Class to Sample
NARMS_DC	NARMS_DC_MLN	Dairy Cow (DC)
NARMS_BC	NARMS_BC_MLN	Beef Cow (BC)
NARMS_ST	NARMS_ST_MLN	Steer (ST)
NARMS_HF	NARMS_HF_MLN	Heifer (HF)

NOTE: Establishments that are subject to NARMS sampling in other slaughter classes will continue to receive unpaired sampling tasks.

B. **Scheduling Sample Collection.** The sample collection window for each NARMS sample task is 37 days. PHVs are to schedule sample collection as soon as possible to reserve laboratory capacity. Samples cannot be scheduled once the laboratory has met its daily testing capacity. PHVs are to schedule the paired sampling task of lymph node collection and cecal collection on the same day as they must be from the same animal (dairy cow, beef cow, steer, or heifer). The PHV is to collect the lymph nodes before collecting the paired cecal sample to prevent cross-contamination. The PHV is to ensure

completion of the scheduled PHIS task. For extenuating scheduling circumstances (e.g., intermittent producers in which there is short notice for a sampling opportunity), the PHV may contact the Eastern Laboratory to obtain a solution to be able to collect and submit a sample. PHVs are to use the laboratory inquiry mailing list to reach the NARMS point of contact: [FSIS – Laboratory Inquiry – Eastern Lab](#).

C. If a scheduled task will not be collected on the date reserved in the task calendar, the PHV is to remove or reschedule that task as soon as they become aware of the change so that laboratory capacity is as accurate as possible and space is free for additional PHV task scheduling.

D. For samples collected from Monday to Wednesday morning, the PHV is to schedule to ship the sample no later than Wednesday afternoon to ensure its arrival at the FSIS Midwestern Laboratory by Thursday. For samples collected between Wednesday afternoon and Friday afternoon, the PHV is to store and maintain the sample under refrigeration and under FSIS control and schedule to ship on Monday morning. Do not freeze the samples.

E. PHVs are to follow the instructions provided in [FSIS Directive 13,000.2](#), for accepting, scheduling, and completing a directed task using PHIS.

VII. LYMPH NODE SAMPLE SUPPLIES

A. **Shipping and Receiving Supplies.** The FSIS Eastern laboratory will continue to provide the sampling supplies for the NARMS cecal sample collection, and the FSIS Midwestern Laboratory will provide the sampling supplies for the NARMS lymph node sample collection. PHVs will receive supplies for the slaughter class to be sampled.

1. PHVs will receive sampling supplies one week prior to the start of the collection window after the directed NARMS sampling is added to the task list. PHVs are to only use the sampling supplies provided by the FSIS laboratories that are specific to this project and specific to the slaughter class scheduled for sample collection;
2. The PHV is to inspect the supplies upon receipt and verify all needed supplies have arrived and are intact; and
3. The PHV is to remove the gel coolant packs from the shipping container and place them in the freezer at least 24 hours before sample collection.

B. **Requesting Additional Supplies.** If supplies have not arrived 3 business days prior to the scheduled sampling day or the sample supplies received are not complete, PHVs are to send a request for the needed cecal supplies through the FSIS - Sampling Supplies – Eastern Lab mailbox in Outlook using the following address: SamplingSupplies-EasternLab@usda.gov, or the needed lymph node supplies through the FSIS – Sampling Supplies – Midwestern Lab mailbox in Outlook using the follow address: SamplingSupplies-MidwesternLab@usda.gov. PHVs are to use the subject heading “NARMS Lymph Node Supplies” in the e-mail and include the following in the message: the establishment number, name, and mailing address; the project code(s), the PHV’s contact name and telephone number, and a list of the supplies needed.

C. The PHV can also order sampling supplies through the PHIS Task Calendar. To order sample supplies, PHVs are to right-click on the scheduled NARMS sampling task and select **Order Supplies** from the drop-down menu. A pop-up window will appear that displays the project code and the name of the FSIS Laboratory that will fill the sampling supply request. When necessary, PHVs are to enter requests for specific supplies (e.g., extra gloves) in the **Comments** field and click **Submit Request**. A confirmation message will appear. The following sampling supplies (pictured below) for NARMS lymph node sample collection are provided by the FSIS Midwestern Laboratory:

1. Shipping container;
2. 7.5" X 15" Whirl Pak Sterile bag (1);
3. 2 Gallon-size (16" x 12") zipper lock bag (1);
4. Scissors (1 pair);

NOTE: All scissors must be returned to the FSIS Laboratory in the shipping container with the sample.

5. Plastic sterile tweezer (1);
6. Non-sterile examination gloves (8);
7. 2' x 3' plastic pad (1);
8. Sterile gauze pads (4);
9. 6" x 12" plastic sleeve for sampling form;
10. FSIS Form 7355-2A/2B (sample seals);
11. Absorbent pad for shipping container;
12. Cardboard separator(s);
13. Gel coolant pack (1);
14. Foam plug (1); and
15. Shipping airbill (pre-printed).



D. Isopropyl alcohol, isopropyl alcohol wipes or pads, or a caddy, are not included in the list of supplies; these should be procured locally. The PHV is to contact the Frontline Supervisor (FLS) for guidance on obtaining these supplies.

VIII. LYMPH NODE SAMPLE COLLECTION

A. **Collection Summary.** PHVs are to continue to collect NARMS cecal samples from cattle carcasses. The collection of cattle mesenteric lymph nodes is a new NARMS program. PHVs are to follow the instructions provided in this notice for mesenteric lymph node collection. PHVs are to follow [FSIS Directive 10,100.1](#) for cecal sample collection. Attachment I provides photograph-enhanced instructions for lymph node collection. Attachment II contains information on collecting cecal and lymph node samples as a paired sampling event. Sampling instructions are also available via [IPP Help](#).

B. General Instructions: PHVs are to:

1. Collect the lymph nodes from the same carcass that the corresponding NARMS cecal contents will be collected;
2. Collect lymph nodes before the paired cecal sample is collected to prevent cross-contamination;
3. Examine the cattle abdominal viscera that includes mesenteric lymph nodes (refer to Attachment I for pictures) to find and access the lymph nodes in the mesentery. Mesenteric lymph nodes are distributed from the duodenal or anterior part of small intestines to the ileocecal and colic

region; and

4. Collect six (6) mesenteric lymph nodes, preferably from the ileocecal/colic region from a single healthy animal.

C. Lymph Node Collection. PHVs are to:

1. Review the sampling steps applicable to lymph node sampling;
2. Wipe down the table that will be used as the work surface;
3. Lay out the plastic pad on the work surface to provide a clean surface and set out the sampling equipment;
4. Set the sterile bag on the plastic pad for ease of use while maintaining its sterility;
5. Place a sterile gauze pad on the plastic pad. Carefully clean the tips and blades of the scissors with alcohol wipes and position the blades of the scissors on the sterile gauze pad;
6. Wash hands with soap and water and aseptically don two pairs of gloves;
7. Rinse gloves and the mesenteric fat with 70% isopropyl alcohol;
8. With one hand, hold mesenteric lymph nodes one at a time in the ileocecal region of the intestine. This hand will be stationary and will hold the mesenteric lymph nodes throughout the procedure. With the other hand, carefully strip away the mesentery from mesenteric lymph nodes and carefully pluck/extract the lymph node one at a time while ensuring the integrity of the lymph node capsule;

NOTE: If the lymph node capsule is broken, or otherwise compromised, do not include it in the sample. Collect additional mesenteric lymph nodes to ensure six (6) intact lymph nodes are sampled.

9. When necessary, use the supplied scissors to separate and trim mesentery from the lymph nodes;
10. If one or both pairs of gloves become contaminated during initial collection of the viscera, remove the outer pair of gloves before rinsing the remaining gloves with isopropyl alcohol;
11. Gently wipe each separated lymph node using an alcohol wipe or pad;
12. Place each of the six lymph nodes in the sterile bag; and
13. Refrigerate (do NOT freeze) the entire sample immediately after collection and ship the same day using frozen gel packs.

IX. COMPLETING THE SAMPLING TASK AND SHIPPING THE SAMPLE

A. PHVs are to follow the instructions provided in [FSIS Directive 13,000.2](#), and in the [PHIS User Guide](#) for accepting, scheduling, and completing the paired sampling tasks using PHIS. PHVs may choose to print a draft copy of the NARMS sampling form from PHIS for use during sample collection and to document lot information to record in PHIS.

B. PHVs are to enter information for the Sample Collection data fields in PHIS for the paired samples collected. PHVs are to ensure that all requested information is submitted in PHIS. When sample

collection data entry is completed, the PHV are to click the **Submit to Lab** button, print the finalized forms, and sign and date the forms. PHIS will display a message stating that the sample collection information has been successfully submitted. PHVs are to place the signed sampling forms in the sample box with the corresponding sample.

X. SAMPLE RESULTS REPORTING

Lymph node sample results from the NARMS Program will be posted in Laboratory Information Management System (LIMS)-Direct and PHIS.

XI. QUESTIONS

Refer questions regarding this notice to the Office of Policy and Program Development through [askFSIS](#), or by telephone at 1-800-233-3935. When submitting a question, use the Submit a Question tab, and enter the following information in the fields provided:

Subject Field: Enter **Notice 04-20**.

Question Field: Enter your question with as much detail as possible.

Product Field: Select **General Inspection Policy** from the drop-down menu.

Category Field: Select **Sampling-General** from the drop-down menu.

Policy Arena: Select **Domestic (U.S.) Only** from the drop-down menu.

When all fields are complete, press **Continue** and at the next screen press **Finish Submitting Question**.

NOTE: Refer to [FSIS Directive 5620.1](#), *Using askFSIS*, for additional information on submitting questions.



Assistant Administrator
Office of Policy and Program Development

FSIS Lymph Node Testing to Complement the NARMS Program
Attachment I: Photo-enhanced Instructions for Lymph Node Excision

1. Examine the entire intestinal tract from a ruminant on the bottom shelf of the cart to find and access the lymph nodes in the mesentery; the large organs will need to be moved. If one or both pairs of gloves become contaminated during initial collection of the viscera, remove the outer pair of gloves before rinsing the remaining gloves with isopropyl alcohol.



2. Evaluate the mesentery to locate, look, and feel the mesenteric lymph nodes. Rinse the mesenteric fat with 70% isopropyl alcohol.



3. When a mesenteric lymph node is located, as shown, identify a location to excise the lymph node without puncturing the outer capsule.



4. The tip of the knife points at the lymph node, prior to removal.



5. Another view of a mesenteric lymph node to show that lymph node appearance can change depending on the view.



6. Once the entire lymph node is visible and stabilized, start to excise or cutting out the stabilized lymph node. Gently wipe each separated lymph node using an alcohol wipe or pad before packing for shipment to the lab.

Note: If lymph node is punctured, please discard and excise another one.



FSIS Lymph Node Testing to Complement the NARMS Program

Attachment II: Paired Sampling of Livestock Summary Tables

Cecal Sampling: Beef and Dairy Cattle Instructions

Project	NARMS (cecal)
Program Dates	Ongoing
Analyzed for	Pathogens, indicator organisms, antimicrobial susceptibility.
Specimen to Sample	Cecal contents of the large intestine.
Collection Instructions	PHVs are to collect cecal specimens from ONE (1) randomly selected carcass. Aseptically milk approximately 30mL of the cecal contents into the sterile specimen cups and secure the lids. Place the specimen cups into the provided zipper lock bag and attach a barcoded seal.
Sample Request Form	Place the completed PHIS sample request form in a plastic bag and place it inside the shipping container underneath the foam plug.
Shipment	For samples collected from Monday to Wednesday morning, the PHV is to ship the sample no later than Wednesday afternoon to ensure its arrival at the FSIS Eastern Laboratory before Friday. For samples collected between Wednesday afternoon and Friday afternoon, IPP are to store and maintain the sample under refrigeration and under FSIS control and ship on Monday morning. Do not freeze the samples.
References	FSIS Directive 10,100.1

Paired Lymph Node Sampling: Beef and Dairy Cattle Instructions

Project	NARMS_MLN (lymph)
Program Dates	Ongoing; paired with NARMS cattle projects
Analyzed for	Pathogens, antimicrobial susceptibility.
Specimen to Sample	Mesenteric lymph nodes.
Collection Instructions	PHVs are to collect specimens from the ONE (1) carcass that was also selected for NARMS cecal sampling. Collect six (6) mesenteric lymph nodes. Place into sterile whirlpak and apply the barcoded seal label. Place bagged specimen into gallon zipper lock bag for secondary containment.
Sample Request Form	Fill out the form according to the instructions. Place the sample request form in a plastic bag and place it inside of the shipping container, underneath the foam plug.
Shipment	For samples collected from Monday to Wednesday morning, the PHV is to ship the sample no later than Wednesday afternoon to ensure its arrival at the FSIS Midwestern Laboratory before Friday. For samples collected between Wednesday afternoon and Friday afternoon, IPP are to store and maintain the sample under refrigeration and under FSIS control and ship on Monday morning. Do not freeze the samples.
References	IPP Help