DRAFT FSIS Compliance Guideline:
Modernization of Swine Inspection System

Developing Effective Microbiological Sampling Programs in Swine Slaughter Establishments to Assess Process Control and Sanitary Conditions

This guidance document is designed to help all swine slaughter establishments meet the sampling and analysis requirements under the proposed rule to modernize swine slaughter inspection.

This guidance is designed to assist establishments as they:

- Develop a microbiological sampling plan;
- Utilize microbial testing results to assess their ability to maintain process control; and
- Make decisions on process control throughout the swine slaughter process

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This Compliance Guideline follows the procedures for guidance documents in the Office of Management and Budget's (OMB) "Final Bulletin for Agency Good Guidance Practices" (GGP). More information can be found on the Food Safety and Inspection Service (FSIS) Web-page:

http://www.fsis.usda.gov/wps/portal/footer/policies-and-links/significant-guidance-documents

This is the **first** edition of the Compliance Guideline: Modernization of Swine Inspection Swine Inspection System - Microbiological Sampling In Swine Slaughter Establishments. Future editions will continue to reflect feedback received from all stakeholders.

This draft compliance guideline represents FSIS's current thinking on this topic and should be considered usable as of this issuance. Therefore, even though this is a draft document, FSIS encourages establishments slaughtering or producing raw pork products to incorporate information in this guideline in their decision making process. A final version of this guidance will be issued in response to public comments.

The information in this compliance guideline is provided as guidance to assist swine slaughter establishments, and is not legally binding from a regulatory perspective.

The effective date of these requirements will be determined once the final rule publishes for the Modernization of Swine Slaughter Inspection.

What is the purpose of this Compliance Guideline?

The purpose of this guidance document is to help swine slaughter establishments comply with the proposed regulatory requirements including the new microbiological sampling and analysis requirements that apply to all official swine slaughter establishments once the final rule publishes for the Modernization of Swine Slaughter Inspection.

Establishments may also find the information in this document helpful for developing programs prior to the implementation of the requirements related to the Modernization of Swine Slaughter Inspection. Establishments may also find the references listed at the end of this document useful for further resources as well as background on technical concepts.

Establishments can also seek guidance from University Extension Service specialists within the state that the establishment is located on how to assess process control, develop and maintain written sanitary dressing procedures, design sampling plans, how to collect samples, and how to test pork products and evaluate a process using statistical process control.

How can I comment on this Compliance Guideline?

FSIS is seeking comments on this guidance document as part of its efforts to continuously assess and improve the effectiveness of policy documents. All interested persons may submit comments regarding any aspect of this document, including but not limited to: content, readability, applicability, and accessibility. The comment period will be 60 days and the document will be updated in response to the comments received.

Comments may be submitted by either of the following methods:

Federal eRulemaking Portal Online submission at regulations.gov: This Website provides the ability to type short comments directly into the comment field on this Web-page or attach a file for lengthier comments. Go to http://www.regulations.gov and follow the online instructions at that site for submitting comments,

Mail, including CD-ROMs, and hand - or courier-delivered submittals: Send to Docket Clerk, U.S. Department of Agriculture (USDA), FSIS, OPPD, RIMD, Patriots Plaza 3, 1400 Independence Avenue SW, Mailstop 3782, Room 8-163A, Washington, DC 20250-3700.

All items submitted by mail or electronic mail must include the Agency name and document title: Compliance Guideline: Modernization of Swine Inspection Swine Inspection System - Microbiological Sampling In Swine Slaughter Establishments. Comments received will be made available for public inspection and posted without change, including any personal information to http://www.regulations.gov.

DRAFT FSIS Compliance Guideline:
Proposed Modernization of Swine Inspection
Microbiological Sampling In Swine Slaughter Establishments

Proposed Requirements for Written Procedures and Microbiological Sampling

Under the proposed rule to modernize swine slaughter inspection, The New Swine Inspection System NSIS, all swine slaughter establishments will be required to:

- Develop, implement, and maintain written procedures to prevent contamination of carcasses and parts by enteric pathogens and fecal material throughout the entire slaughter and dressing operation. To demonstrate effectiveness of such procedures using their Hazard Analysis and Critical Control Points (HACCP) system (i.e. HACCP, Sanitation Standard Operating Procedures (Sanitation SOP or SSOP),or pre-requisite programs), establishments are required to sample for microbial organisms and analyze results at prescribed locations and frequencies to assess the establishment's ability to maintain process control.

- Develop, implement, and maintain written procedures in their HACCP systems to prevent contamination of the pre-operational food contact surfaces by enteric pathogens

- Incorporate their written procedures, including their microbiological sampling plans, into their HACCP system.

Table 1. Current Requirements and Proposed Requirements for Microbial Sampling in Swine Slaughter Establishments

	Current Microbial Sampling Requirements for *E. coli* Biotype 1 (9 CFR 310.25)	**Proposed Requirements for Microbial Sampling (Indicator Organisms or *Salmonella*) under NSIS**	
Establishment size	**Minimum carcass sampling event frequency and location**	**Minimum carcass sampling event frequency and location**	**Minimum pre-operational sampling event frequency and location**
Very small (VS)	Starting June 1 of every year, establishments will take a minimum of one post-chill sample during each week of operation. The sampling plan may be modified after 13 consecutive weekly samples demonstrate effective process control.	Starting June 1 of every year, establishments will take a minimum of one post-chill sample per sampling event during each week of operation. The sampling plan may be modified after 13 consecutive weekly samples demonstrate effective process control.	1 times/month (monthly)
Very low volume (VLV)			
Small	Establishments will take one post-chill sample per 1,000 carcasses, with a minimum of one sample taken during each week of operation.	Establishments must analyze 1 sample at pre-evisceration[b] and 1 at post-chill per sampling event. Samples must be collected and analyzed at a frequency of once per 1,000 carcasses, with a minimum of one sampling event during each week of operation.	2 times/month (every 2 weeks)
Large			4 times/month (weekly)

[b] Establishments may choose to substitute alternative sampling locations if they are able to demonstrate that the alternative sampling locations are able to provide a definite improvement in assessing process control than at pre-evisceration and post-chill.

[c] Establishments may choose to substitute alternative sampling frequencies if they are able to demonstrate that the alternative is an integral part of the establishments' verification procedures for their HACCP plans and are able to provide a definite improvement in assessing process control than at the prescribed frequency, i.e. a program in the HACCP plan.

Table 2. Establishment Size

Establishment size	Defined as
Very small (VS)	Fewer than 10 employees or annual sales of less than $2.5 million.
Very low volume (VLV)	Annually slaughter no more than 20,000 swine, or a combination of swine and other livestock not exceeding 6,000 cattle and 20,000 total of all livestock
Small	10 – 499 employees unless annual sales total less than $2.5 million
Large	500 or more employees

Microbial Sampling Plan for Carcass Sampling

Establishments that slaughter swine are responsible for determining which microbial organism will be most effective in assessing process control and developing their own sampling plans. FSIS recommends establishments choose an indicator organism that is able to provide meaningful data in assessing process control. Potential indicator organisms include Aerobic Plate Count (APC), generic *E. coli*, Enterobacteriaceae (EB) and total coliforms. FSIS recommends APC because APC is less specific than generic *E. coli* and therefore, sampling for it results in more positive results to assess variations in process control (Williams 2015). Having more positive numbers (enumeration) helps because establishments can plot this information and see trends over time. Generic *E. coli* on the other hand is a smaller group of bacteria and therefore testing for it tends to result in many negatives making it hard to identify trends from day to day.

Establishments that are participating in the *Salmonella* Initiative Program (SIP) may use the SIP microbial data as part of their sampling plan to assess their process control provided they meet minimum frequencies and location requirements.

Any establishment may choose to substitute alternative sampling locations if they are able to demonstrate that the alternative sampling locations are able to provide a definite improvement in assessing process control than at pre-evisceration and post-chill. FSIS recommends all establishments conduct carcass mapping (O'Connor 2012) to determine which sampling locations (i.e. bleed out, polishing, post-evisceration) and sampling sites (i.e. inside and/or outside ham, belly jowl, oral cavity, or rib cage) are most effective for each establishment to assess its process control and reduce the total number of pathogens that may be present. Establishments should provide support for the sampling locations and sampling sites (i.e. sampling data demonstrating the ability to assess process control) described in the sampling plan. FSIS also recommends establishments take into account contamination sources such as incised lymph nodes (Garrido 2014, Vieira-Pinto 2005), tonsils (Bonardi 2013), intestinal rupture and stick wounds when designing their sampling plan.

Definitions

Pre-evisceration refers to the location early in the slaughter process prior to evisceration of the hog.

Post-chill refers to a later point in the slaughter process after carcasses are chilled and all interventions have been applied prior to fabrication.

Very small and very low volume slaughter establishment operating under Traditional Inspection can choose to continue conducting generic *E. coli* testing at post-chill to meet these requirements. FSIS considers the requirements under the former regulations for generic *E. coli* testing of swine to be a scientifically valid "safe harbor" for assessing process control.

Former provisions that FSIS considers to be safe harbors:

A. Each very small or very low volume establishment that slaughters swine under Traditional Inspection may test for *Escherichia coli* Biotype I (also referred to as generic *E. coli*) at the post-chill point in the process.

B. To collect the sample, the establishment should collect an excision or swab sample of the ham, belly and jowl from the carcass at the end of the chilling process as described in the FSIS Compliance Guideline: Guidelines for *Escherichia coli* Testing for Process Control Verification in Cattle and Swine Slaughter Establishments

C. Laboratories analyzing the samples should use any quantitative method for analysis of generic *E. coli* that is approved as an Official Method of the AOAC International (AOAC) (formerly the Association of Official Analytical Chemists) or approved and published by a scientific body and based on the results of a

collaborative trial conducted in accordance with an internationally recognized protocol on collaborative trials and compared against the three tube Most Probable Number (MPN) method and agreeing with the 95 percent upper and lower confidence limit of the appropriate MPN index.

E. coli excised-sample results for swine tested in the FSIS baseline studies have been separated into three categories for the purpose of process control verification: acceptable, marginal, and unacceptable. In the Pathogen Reduction/HACCP Regulation, m and M, representing respectively the 80th and 98th percentile of sample results, leaving 18 percent of results in the marginal range denoted the upper limits for the acceptable and marginal ranges.

An establishment is operating within the criteria when the most recent *E. coli* test result does not exceed the upper limit (M), and the number of samples, if any, testing positive at levels above (m) is three or fewer out of the most recent 13 samples (n) taken, as follows:

Table 3. Performance criteria for generic *E. coli* from swine carcasses.

Lower limit of marginal range (m)	Upper limit of marginal range (M)	Number of Samples tested (n)	Maximum number permitted in the Marginal range
10 CFU/cm^2	10,000 CFU/cm^2	13	3

Note: The data in this table is only applicable for excise tissue samples from the ham, belly and jowl areas. This procedure is detailed in the FSIS Compliance Guideline: Guidelines for *Escherichia coli* Testing for Process Control Verification in Cattle and Swine Slaughter Establishments.

Appendix 1 on page 28 contains a self-assessment checklist that highlights the key elements that an establishment should address as part of their written microbiological sampling plan.

Random Selection and Sampling of Carcasses

Samples should be collected randomly at the frequency determined by the establishment as part of its sampling plan. At a minimum, under the proposed rule all swine slaughter establishments will be required to collect samples at the frequency specified in Table 1.If more than one shift is operating at the plant, the sample can be taken on any shift. Variations have been found from samples collected on different

shifts, therefore it is important to make sure all shifts have an equal opportunity for being selected.

Different methods of selecting the specific carcass for sampling could be used, but the method used should include the use of random numbers to ensure that testing data is not biased. Examples of methods include random number tables, calculator or computer-generated random numbers, or drawing cards.

The carcass that is sampled should be selected at random from all eligible carcasses. If there are multiple lines, randomly select the line for sample collection for that interval. Each line should have an equal chance of being selected at each sampling interval within the relevant time frame (based on the sampling frequency for the plant).

Carcasses should be selected at the identified points in the process. At the post-chill site, samples should be collected after the final wash and the application of any final antimicrobial interventions. A drip time of at least 60 seconds should be observed before sample collection to prevent excessive antimicrobial carryover in the collected sample.

Statistical Process Control and Indicator Organisms for Carcass Sampling

Statistical process control provides a powerful mechanism for establishments to assess and interpret the data collected for ongoing HACCP verification. Statistical process control can provide establishments with an early warning that their process may not be functioning as designed. This warning can allow establishments to take corrective actions or make other process modifications to bring their process back into control. Statistical process control can also provide establishments with reasonable assurance that their HACCP system is functioning as designed, and that they are likely to meet applicable performance standards.

A number of methods and approaches are available for establishments to follow. Establishments should consider available guidance and develop a statistically valid approach for interpreting sample results (Saini et al. 2011 and De Vries).

In cases where an establishment does not have the resources or capacity to develop and implement their own statistical control limits or procedures, establishments can utilize the results from FSIS nationwide swine surveys, provided in Table 4. These results come from a nationwide survey conducted in 2011[1]. During the survey, FSIS collected samples from two points during processing; swine carcasses at pre-evisceration and post chill. The tables show the geometric mean enumeration values for four common indicator bacteria: generic *E. coli*, APC, Enterobacteriaceae, and total

[1] FSIS Market Hog Survey

coliforms. The geometric mean is a type of average which indicates the central tendency of a set of numbers by using the product of their values. The geometric mean is preferred in this example as it decreases the influence of very large values when compared to the arithmetic mean (average).

Table 4 - Indicator Organism Geometric Mean Values for Market Swine

	Geometric Mean in CFU/cm^2 (log_{10})			
	Generic *E. coli*	**Aerobic Plate Count (APC)**	**Enterobacteriaceae (EB)**	**Total Coliform**
Carcass – *Pre-Evisceration*	603 (2.8)	645,654 (5.8)	1,023 (3.0)	832 (2.9)
Carcass – *Post Chill*	5 (0.7)	107 (2.0)	6 (0.8)	5 (0.7)

When establishments compare their sample results to the ones in the table, a sample value that is higher than the corresponding one listed in the table indicates the establishment may not be maintaining process control and may be less likely to meet applicable performance standards. Sample values lower than the one listed in the table indicate the establishment may be maintaining process control.

Charting and Interpreting Test Results for Carcass Sampling

Specific techniques of statistical process control include the use of a control chart, which plots data over time but also displays an upper control limit for specific measurements and a centerline, above and below which there is an equal number of sample results (the centerline is in effect an average). A sample result above the upper control limit would indicate the likely presence of a special cause of variation that should be addressed. Results within control limits indicate simply that the process is in control. Control charts are used to (1) analyze and understand variables that affect the process, (2) determine process capabilities, and (3) assess effects of the variables on the difference between target and actual performance. In most situations more than one type of control chart would be applicable. Detailed information on the use of control charts can be found in texts on statistical process control, under the topic “control charts”.

The following control charts are hypothetical examples of using microbiological test results, collected over time, to verify the effectiveness of a food safety system (Buchanan 2000).

Chart 1 - System under control

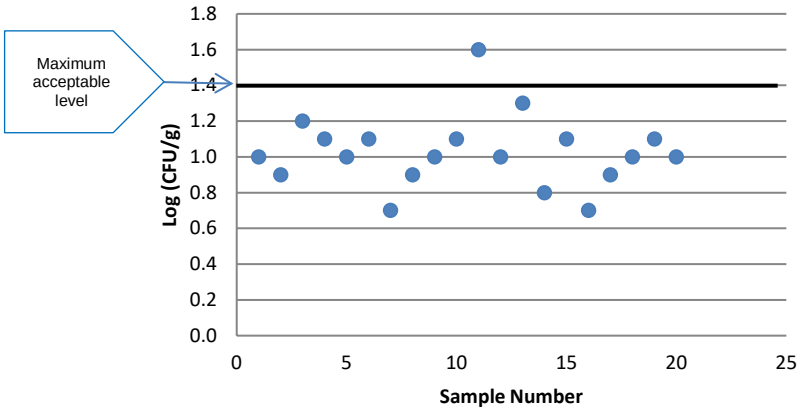

Chart 1 depicts a pattern of test results that would be seen in a well-controlled system.

In a well-controlled system, the majority of test results will be clustered around a central value.

It is important to note that even in a well-controlled system; there is some frequency of isolated results above the acceptable level.

Chart 2 - Lack of control due to excess variability

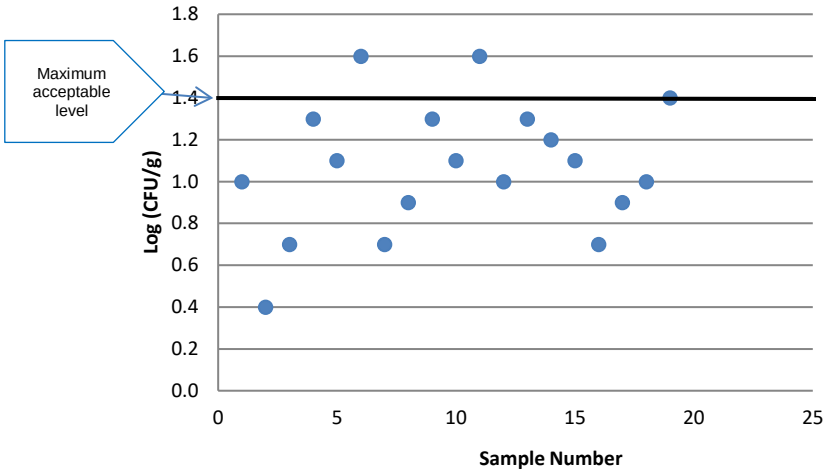

Chart 2 depicts a loss of process control due to excess variability. This is reflected in both an increased number of results above the maximum acceptable level and an increase in the scatter of points below the maximum acceptable level.

This chart suggests either a loss of control at a critical control point or the existence of another critical control point that had not been identified and controlled.

Chart 3 - Loss of control due to gradual process failure

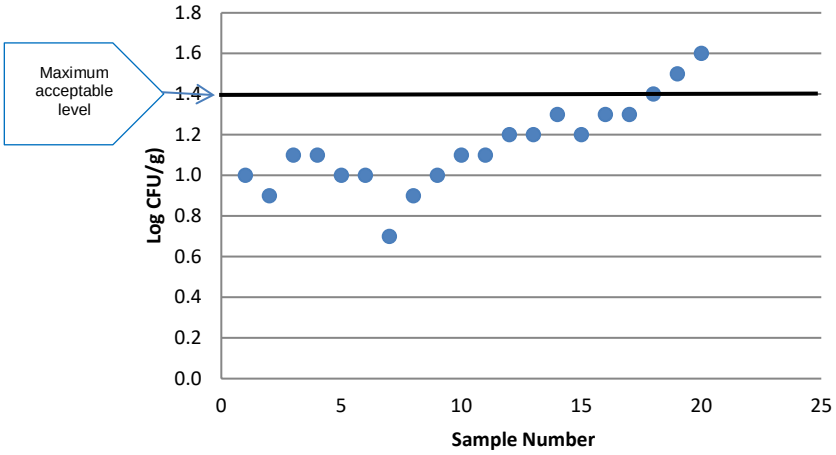

Chart 3 - depicts a situation where a component of the process is losing its effectiveness over time.

This loss of control is apparent by the upward trend in the data points toward the maximum acceptable level.

Chart 4 - Loss of control due to abrupt process failure

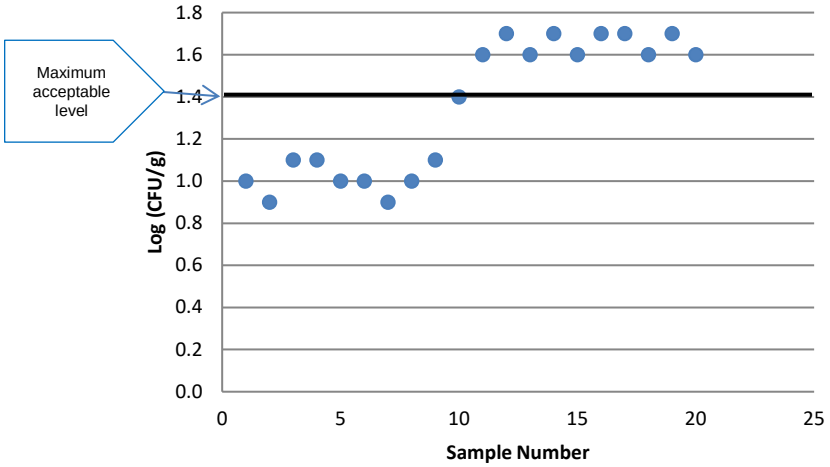

Chart 4 depicts a catastrophic loss of process control.

This pattern of test results would be encountered in a situation such as an abrupt failure of a key piece of equipment, such as an antimicrobial wash cabinet.

Chart 5 -Loss of control due to reoccurring transitory process failure

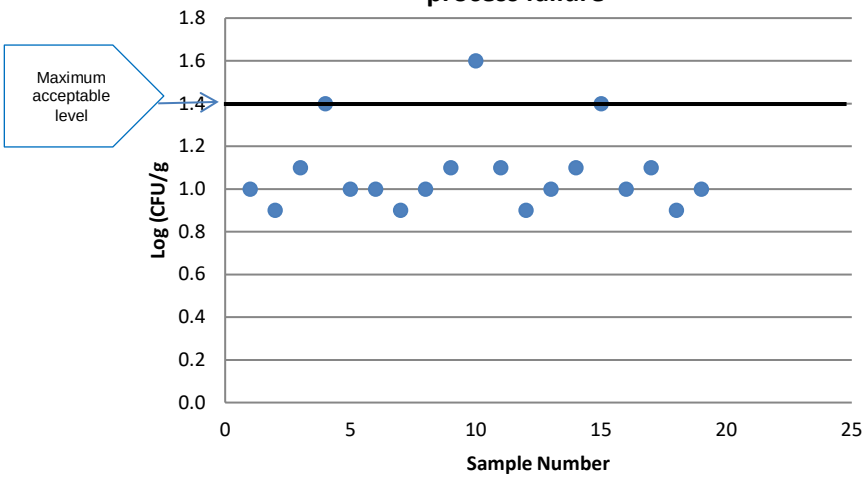

Chart 5 depicts conditions where there is the existence of an intermittent but reoccurring problem within the process. Note the distinct periodicity of the test results over time.

An example of a situation where this pattern may be observed is the dripping of condensation onto product as it travels down a conveyor belt

The test results should be plotted and evaluated in a series over time. The test result chart should be updated within the next business day following the reporting of test results by the testing laboratory. Every time a new test result is recorded, the oldest test in the series is dropped from the moving window. For example, an establishment may choose to evaluate their test results in a moving window of 13 tests. The establishment would use this series of 13 tests to evaluate their process control over the period of time represented by the series of 13 tests. The control chart would be updated with each new test result reported, adding the new test result and removing the oldest test result on the chart.

Microbiological testing provides a measure of the extent of control at the step being evaluated and all preceding steps. By performing microbiological analyses at several points within a process, it is relatively easy to identify the segment of the process where control has been lost. In addition, while it is not required, end-product testing and sanitary dressing verification can provide an integrated measure of the performance of

the entire process. Pre- evisceration and post chill results could be charted on the same graph with separate upper control limits to better correlate the samples and calculate the log reduction between the two samples.

Actions in Response to Process Control Results

As part of its process control procedures, an establishment should define the actions it will take if the test results obtained through its sampling are above the limits it has set. The establishment should delineate what its actions will be, who will take each action, how the outcome of these actions will be documented, and how it will be verified.

FSIS has made available the *FSIS Compliance Guideline for the Controlling Salmonella* in Market Hogs. The guideline summarizes known control points for *Salmonella* in the pre- and post-harvest production process. Establishments should use this compliance guide to improve management practices, to ensure effective sanitary dressing procedures and to assist in investigating when there is a loss of process control. When an establishment makes changes at the appropriate locations, process control should improve. As a result, establishments should produce raw pork products that have less contamination with pathogens, including *Salmonella*.

If the establishment determines that the trends in its test results indicate a loss of process control, the establishment should take action to investigate the cause. As discussed in the previous section on process control, an establishment should consider how the different parts of its food safety system work together, and how they affect the entire food safety system. To do this, establishments should evaluate its process control procedures, sanitary dressing practices or sanitation procedures to determine whether a root cause can be identified and take steps to correct the problem. This determination should include a review of its process monitoring records as well as evaluation of the process during normal operations. The establishment should consider any implementation problems or changes in its practices, such as sanitary dressing procedures, including but not limited to:

- Procedures for routine cleaning and sanitizing of equipment, including hand tools that are used to remove contamination or to make cuts into the carcass;
- The design, configuration, and calibration of equipment to ensure proper function within operational parameters to prevent the contact between carcasses and parts and prevent contamination of carcasses during operation;
- Employee hygiene practices, ensuring that employees frequently wash hands and aprons that come in contact with carcasses and are properly trained when there are new or substitute employees on the line; and
- The implementation of antimicrobial or mechanical intervention treatments, such as carcass washes, sprays, or brushes, in accordance with the limits

selected by the establishment, including effective application to ensure coverage of the entire carcass.

Following its investigation, the establishment should respond appropriately to its findings through the use of decontamination procedures and antimicrobial intervention treatments as necessary to address any contamination that may have occur on the carcasses and parts. The establishment should also take steps to initiate any necessary equipment repair or recalibration and employee training when identified.

Written Microbial Sampling Plan for Pre-Operational Food Contact Surface Samples

FSIS is proposing this sampling requirement because in 2015, 152 people became ill after consumption of product produced at an establishment where FSIS found evidence during an investigation of insanitary conditions, including, but not limited to, pre-operational tables and knives that were contaminated with *Salmonella*. The proposed food contact surface sampling requirement would reduce the risk of cross-contamination from insanitary conditions and could identify harborage sites for *Salmonella* biofilms.

The presence and survival of *Salmonella* on surfaces within pork processing establishments has been shown to lead to cross contamination onto pork products (Botteldoorn *et al*., 2003; DeBusser *et al.,* 2013, EFSA, 2008; BIOHAZ, 2010; Van Hoek *et al*., 2012). *Salmonella* has also been found to survive longer in the presence of organic material (Allan *et al*., 2004; De Cesare *et al*., 2003; Møretrø *et al*., 2010) and desiccation could result in cross-protection of *Salmonella* to several disinfection treatments (Gruzdev *et al*., 2011; Kieboom *et al*., 2006). Therefore, it is important for establishments to develop a comprehensive sampling plan to verify cleaning and sanitizing efficacy and to identify potential biofilm harborage sites throughout the slaughter and processing environment. Harborage sites may be found throughout the slaughter and processing environment, including, but not limited to, food contact surfaces (FCS), reuse water, scalder, and de-hairer machines.

Under the proposed swine modernization, all official swine slaughter establishments will also be required to:

- develop, implement, and maintain in their HACCP systems written procedures to prevent contamination of the pre-operational food contact surfaces by enteric pathogens.

- incorporate their written procedures, including their microbiological sampling plans, into their Hazard Analysis and Critical Control Points (HACCP) plans, or Sanitation Standard Operating Procedures (Sanitation SOP), or other prerequisite program.

The pre-operational food contact surfaces (FCS) could include, hooks, conveyor belts, fabrication tables, injecting machines or equipment, including knives or saws, located in the edible food production area before slaughter and fabrication operations begin (to see a more detailed list of FCS go to Appendix 2). Under the proposed rule, the written procedures to prevent contamination will be required to include sampling and analysis of pre-operational food-contact surfaces for microbial organisms to ensure that the surfaces are sanitary and free of enteric pathogens. Establishments will not have to wait for sample results to begin operation. *Salmonella* positive pre-op food contact surfaces located towards the end of the slaughter process and the beginning of fabrication areas may indicate poor sanitation or process control at earlier locations of the slaughter process.

As part of their Pre-Operational Sampling Program, establishments should have both written routine and follow-up sampling plans. The routine sampling plan should include all of the sample collection procedures the establishment will follow when collecting routine samples. As part of the routine sampling plan, the establishment should identify the:

- sites they will sample,
- the frequency of sampling,
- the number of samples they will collect,
- the size of the sampling sites, and
- the sampling method.

Table 5. Minimum Routine Sampling Frequencies for Testing of Food Contact Surfaces

Establishment Size	**Defined As:**	**Minimum Frequency***
Very small	Fewer than 10 employees or annual sales of less than $2.5 million.	1 time/month/line (monthly)
Very Low Volume	Annually slaughter no more than 20,000 swine, or a combination of swine and other livestock not exceeding 6,000 cattle and 20,000 total of all livestock	
Small	10 – 499 employees unless annual sales total less than $2.5 million	2 times/month/line (every 2 weeks)
Large	500 or more employees	4 times/month/line (weekly)

*At least 3- 5 samples per production line should be sampled each time (monthly, biweekly or weekly).

Establishments can test for *Salmonella* or an indicator organism (e.g., APC, Generic E. coli and Enterobacteriaceae). FSIS recommends establishments choose *Salmonella*

since this is the most specific organism to test for and the purpose of the FCS sampling is to verify sanitary conditions. If an indicator organisms is chosen instead FSIS recommends selecting an organisms that targets a smaller group of bacteria (i.e. Enterobacteriaceae) but is still an indicator for *Salmonella*. The purpose of this sampling would be to determine the presence or absence of organisms on the pre-operational food contact surfaces, FSIS would not expect enumeration methods, however they would still be acceptable.

If an establishment chooses to sample for an indicator organism instead of *Salmonella* the establishment would be expected to take corrective actions and to follow up on the indicator organism positives, according to their sampling plan to verify the pre-operational food contact surfaces are sanitary and free of enteric pathogens. Establishments should identify the size and location of the sampling sites in order to follow-up on positive results and conduct effective correction actions. A finding of a *Salmonella* or indicator on a FCS indicated conditions where sanitation has been inadequate, but product produced that day would not be considered adulterated.

Sample Collection Considerations

Establishments should design their sampling plans so that they collect a combination of random and discretionary samples. Initially, samples should be collected at random, to ensure that all FCS have an equal chance of being sampled. The establishment should have plans in place so that representative samples of all FCS will be sampled over a specified period of time.

Once the establishment has generated data demonstrating that their control system is effective, the establishment should adopt a more risk-based sampling plan. The risk - based sampling should include discretionary samples that are collected along with the random samples. These samples can be collected at the discretion of the sample collector based on positive results or other conditions as observed at the establishment. For example, if the establishment is collecting 3-5 samples per line as part of the routine sampling plan, 1-2 of the samples should be discretionary while the others should be collected randomly.

Establishments should also sample more frequently in areas where sanitation issues have been identified. Discretionary samples can also be collected to demonstrate the effectiveness of the establishment's corrective actions. The results from the discretionary samples can be linked to the sample collector's observations, providing more information about sources of harborage in the establishment.

If positive samples are found, the establishment should take corrective actions and collect follow-up samples as described in their sampling plan. In addition, the establishment should target the sites during future routine discretionary sampling, to ensure that the contamination has been addressed. For more information on follow-up sampling see Section Actions to Take in Response to a Pre-Operational FCS Positive.

Examples of FCSs may include:

- Knives,
- Hooks,
- Saw, and
- Evisceration Pans.

A table of other possible FCSs is provided in Appendix 2.

Frequency of Sampling and Explanation of this Frequency

The sampling frequency should be based on the following criteria:

a) Establishment size or volume (large, small, very small), and
b) Past history and observed patterns of contamination.

Frequency Determinations: How to use Table 5

When the establishment is using the sampling frequencies specified in the table, at least 3- 5 FCS samples per production line should be sampled each time (monthly, biweekly, or weekly). The samples should be taken at different days throughout the year, quarter, month, or week, to ensure that the samples are truly representative of processing conditions. The frequencies listed in the table are based on a typical slaughter schedule (5 days a week). Establishments that produce intermittently may be able to support sampling less frequently depending on the slaughter schedule.

Once an establishment has identified a sampling frequency, it should follow the frequency it has selected. If sampling is not performed at the stated frequency, the establishment would need to provide support that their surfaces are sanitary and free of enteric pathogens.

Appendix 1 on page 28 contains a self-assessment checklist that highlights the key elements that an establishment should address as part of their written microbiological sampling plan.

Considerations for Pre-Operational Sampling Methods

Using proper sampling collection technique is important to ensure that low levels of *Salmonella* or indicator organism are detected on pre-operational slaughter food contact surfaces. The establishment should provide written instructions for collecting food contact and the samples should be collected using aseptic techniques.

Establishments may use these methods, or adjust the methods based on the needs of the establishment. FSIS expectations for sampling and testing methods are provided below.

Sampling should be performed by a person trained in aseptic technique and samples should be collected using sterile sponges or other sampling devices.

Sample size
A 12”x12” area should be sampled, when possible, for FCS surfaces. If the surface area is smaller than 12”x12”, then the entire surface should be sampled.

Cotton-tip swabs and other smaller sampling devices are not recommended for sampling large areas (12”x12”) because they may become easily saturated with microorganisms. If these devices are used, FSIS recommends collecting a smaller sampling size per swab according to the manufacturer’s instructions to equal a 12”x12” area.

Sample collection
The sponge or sampling device should be hydrated with sterile neutralizing buffer, Dey Engley (DE) broth, or another sterile broth that contains components that can neutralize the effects of sanitizers that may be present in the sample.

Sample integrity
Samples should be stored under refrigeration (never frozen) before analysis. Samples should be properly labeled to avoid confusion regarding testing results.

Reuse water sampling
Some establishments reuse water in the slaughter environment. Depending on whether the reuse water is used on carcasses, the reuse water could be considered as a food contact sample.

***Question**: How soon after the samples are collected should they be analyzed to ensure the accuracy of the test results?*

***Answer**: To obtain the most accurate results, samples should be analyzed as soon after collection as possible. If samples must be transported to an off-site laboratory, they should be refrigerated and then shipped refrigerated, on the same day they were collected, via an overnight delivery or courier service to the laboratory. A sample should arrive at the laboratory and be analyzed no later than the day after it is collected.*

Sample compositing
FCS samples may be composited (combined) in order to conserve establishment’s resources. If compositing is performed, FSIS recommends that no more than 5 samples be composited, and separate sponges (or other sampling device) be used to collect each sample, to avoid possible cross contamination. One laboratory test can then be performed on the 5 separate samples, decreasing the cost to the establishment. In addition, individual locations for the composite sample should be noted to assist in determining the site of contamination to facilitate follow-up testing.

If a composited sample tests positive, the establishment should consider all the sites represented by the sample as positive and take corrective actions accordingly. During

follow-up sampling of FCSs, the sites should be re-sampled individually, along with additional swabs in the area.

The following are FSIS's expectations for pre-operational FCS testing methods:

1) An enrichment step is used to allow for recovery of injured organisms and growth of organisms to levels that can be detected by most testing methods. Many commonly used testing methods are unable to detect levels below 100 cells/sample. Therefore, it is important that the enrichment step be designed to allow low levels of cells that may be present in the sample to grow to detectable levels. It is also important to allow injured cells time to recover so that they can be detected by the testing method. In most cases, enrichment requires at least 8-hours to achieve adequate levels of microbial growth for detection. A one-hour resuscitation step is not an enrichment step, and would likely not be sufficient to detect low levels of organisms.

2) The entire sponge or sampling device is analyzed. Some methods involve testing just a small part of the broth or other diluent used to hydrate the sponge or sampling device. Studies have shown that bacteria are likely to be trapped on or in the interior of the sponge or other sampling device. Therefore, FSIS suggests that the whole sponge or sampling device be included in the enrichment step. Analyzing the entire sampling device will increase the likelihood of detecting cells.

3) The method has been validated. All screening methods should either be used by a regulatory body (e.g., FDA Bacterial Analytical Manual (BAM)), or validated by a recognized independent body (e.g., AOAC, AFNOR, ISO, NordVal, Microval). FSIS has provided a list of Foodborne Pathogen Test Kits Validated by Independent Organizations and has provided the compliance guideline, FSIS Guidance for Test Kit Manufactures, Laboratories: Evaluating the Performance of Pathogen Test Kit Methods.

Direct plating methods (e.g., media that is added directly to an agar plate or dehydrated media) that do not include an 8-hour enrichment step would be unlikely to detect low levels of organisms.

Actions to Take in Response to a Pre-Operational FCS Positive

When a FCS is positive for *Salmonella* or an indicator organism it indicates that sanitation was inadequate. The establishment should conduct corrections (i.e. reevaluating the sanitation standard operating procedures (SSOP), switching sanitizers, re-training employees, or replacing hard to clean equipment) and perform follow-up samples to verify the effectiveness of the corrective actions. The establishment should specify the number of samples it will collect during follow-up sampling. FSIS recommends that 3-5 samples are collected from the site of the original FCS positive and the surrounding area. These may include other FCSs that are upstream from the original positive. It would be useful for the establishment to record the rationale for selecting follow-up sampling sites. For example, if a hand tool tests positive, the establishment may choose to sample other hand tools to verify sanitization of similar

items. Follow-up sampling could also include other FCSs on the same piece of equipment that were not previously tested. The establishment should also include a brief description of corrective and preventive actions that will be taken in response to positive results (details can be included in the Sanitation SOP) and response to positive results (next steps).

Pre-sampling preparation and aseptic technique

Extraneous organisms from hands, clothing, sampling equipment, or the processing environment may contaminate samples and lead to erroneous analytical results. Aseptic sampling techniques should be followed to ensure accurate results that are representative of the product and process.

Before beginning sample collection, it is important to assemble sampling supplies, such as sterile gloves, sterile sampling solutions, and sanitizing solution. Sterile sampling solutions, such as Dey Engley (DE) broth, should be stored according to the manufacturer's instruction at room temperature; however, at least the day before sample collection, check such solutions for cloudiness and do not use solutions that are cloudy or turbid or that contain particulate matter.

An area should be designated as a staging site for preparing the sampling supplies. A sanitizable surface, such as a stainless steel table or wheeled cart, can be used. A small plastic tote may also be useful for transporting sampling supplies to sample collection sites.

Sterile gloves should be used when handling carcasses or sterile sampling equipment (e.g., sampling sponge) during the sample collection process. Care should be taken to prevent contamination of the external surface of the gloves prior to or during the sample collection process.

Sample analysis

The establishment should ensure that microbiological testing meets its food safety needs. An establishment needs to determine whether sample analysis will be performed by an outside laboratory or in its own microbiological testing laboratory onsite (if available).

Because of the costs and the logistics involved with maintaining an onsite microbiological testing laboratory, establishments may choose to have samples analyzed by an outside laboratory. FSIS has made available the compliance guideline, *Establishment Guidance for the Selection of a Commercial or Private Microbiological Testing Laboratory*. This guidance document should be particularly useful to very small establishments when they are selecting a commercial or private laboratory to analyze establishment microbiological samples. Establishments should clearly communicate their needs to the testing laboratory and direct them to any necessary testing protocols

or other guidance, including this document, on the FSIS Web site. Establishments that select a laboratory that does not apply appropriate testing methods or effective Quality Control/Quality Assurance (QC/QA) practices may not receive reliable or useful testing results or be able to support decisions made in their hazard analysis. Establishments are responsible for ensuring the appropriate methods are selected and to convey this information to the laboratory. FSIS has also made available a list of *Foodborne Pathogen Test Kits Validated by Independent Organizations* for the detection of relevant foodborne pathogens (i.e., *Salmonella*, *Campylobacter*, *E. coli* O157:H7, and *Listeria spp*. including *L. monocytogenes*). These lists are intended to be informational and are not an endorsement or approval of any particular method, regardless of its inclusion in the list.

To prevent cross contamination, FSIS recommends that a microbiological testing laboratory be segregated from manufacturing areas, and that access to the laboratory space be limited. If the establishment tests for pathogens onsite, then they should have the following additional safeguards in place to ensure food safety and security:

- Follow requirements for Biosafety Level II laboratory operation as outlined in *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) available at: http://www.cdc.gov/biosafety/publications/bmbl5/BMBL.pdf;
- Restrict access to the laboratory to trained staff; and
- Ensure the laboratory is operating under the supervision of a qualified microbiologist or equivalent.

Establishments can (and often do) analyze samples for non-pathogenic organisms such as generic *E. coli* and aerobic plate counts (APC) on-site. The test method used should be validated for the target organisms and for the sample matrix being analyzed to ensure accuracy of the results. It should also be a method validated by a recognized independent body, such as the AOAC.

To obtain the most accurate results, samples should be analyzed as soon after collection as possible. If samples must be transported to an off-site laboratory, they should be refrigerated and then shipped refrigerated, on the same day they were collected, via an overnight delivery or courier service to the laboratory. Multiple samples collected on the same day can be shipped together to the laboratory in the sample shipping container. Multiple samples collected on the same day should be analyzed individually and not composited into one sample. A sample should arrive at the laboratory and be analyzed no later than the day after it is collected.

If sample collection, pick-up or shipment, and laboratory analysis cannot be carried out within this timeframe, the carcass or product selected for sampling should be held under refrigeration until the process can be accomplished in the appropriate span of time. The same principle applies for samples that are analyzed in-plant: If a carcass cannot be sampled, and the sample analyzed, by the day after it is taken, the carcass should be held under refrigeration until this is possible. Samples should not be held for an extended period of time. They should be either analyzed in-plant the same day as it is

collected or by the following day or immediately shipped for overnight delivery to the laboratory that will conduct the analysis. Sponge or tissue samples should be held at refrigerated temperature, not frozen, and shipped cold to the laboratory in an insulated shipping container with frozen gel packs. Lastly, the identity of the sample should be maintained during testing to ensure that sites are correctly identified.

Recordkeeping

Upon implementation of the sampling plan the establishment should maintain records sufficient to document the implementation and monitoring of sample collection; the testing procedures, including support for the adequacy of the testing frequency, and the test results. Records should include information such as the:

- Time, date, and location of the sample collection.
- Sample collector's name.
- Name or description of the product or sample source.
- Lot information and producer.

All entries should be dated and initialed by the sample collector immediately upon completion of the entry. If an outside laboratory is used for testing, then these records should also include information such as date the sample was shipped to the laboratory for analysis. The outside laboratory should document the:

- Date received;
- Condition of the sample upon receipt, including sample temperature, if applicable;
- Date the analysis was started and completed; and the
- Analytical result.

Test results should also be recorded and linked to the sample collection records by a sample number, form number, or some other unique identifier. These records should be maintained in a way that ensures the integrity of the data. These records can be maintained in an electronic format, provided there are measures in place to ensure the security of the information. These records should be readily accessible for review by plant and FSIS inspection program personnel upon request.

Finished Product Standards (FPS) Waivers

On January 28, 2008 FSIS announced the *Salmonella* Initiative Program (SIP) as a voluntary program to provide incentives to establishments to maintain consistent process control to minimize *Salmonella* levels and to conduct microbial testing to demonstrate that they are maintaining process control (73 FR 4767, Jan. 28, 2008). In return, establishments received one or more waivers of certain provisions of the regulations, such as those on use of alternative Finished Product Standards (FPS) procedures.

These waivers were authorized under 9 CFR 303.1(h), which provides that the FSIS Administrator may, in specific classes of cases, waive any provisions of the swine inspection regulations for limited periods in order to permit experimentation so that new procedures, equipment, and processing techniques may be tested to facilitate definite improvements, provided that such waivers are not in conflict with the purposes or provisions of the Federal Meat Inspection Act (FMIA).

FSIS has granted waivers to establishments with respect to testing and other provisions in the FPS regulations, so that establishments could collect data and assess whether this other data would facilitate definite improvements.

The proposed rule to modernize swine slaughter inspection plans to amend the swine regulations to establish an additional inspection system, called the New Swine Inspection System (NSIS), for market swine establishments.

For establishments that choose to operate under NSIS, the proposed rule replaces FPS with a requirement that establishments maintain records to document that swine products resulting from its slaughter operation meet the definition of ready-to-cook (RTC) swine. Thus, all waivers to the following regulations 9 CFR 310.11 and 310.14 will be terminated when this proposed rule is finalized. The purpose of the waivers was to gather information on how non-food safety defects should be handled. The Agency's decision on this matter, to go the ready-to-cook standard in NSIS, was based on the information obtained under these waivers. Therefore, the reason for granting the waiver has been fulfilled. Establishments that are operating under FPS waivers and that would like to continue to use their alterative FPS procedures will need to convert to the NSIS.

Establishments that notify FSIS of their intent to operate under NSIS may continue to operate under the waiver from FPS requirements until they start operating under NSIS. If establishments choose to operate under traditional inspection their FPS waiver will end on a yet to be determined date. FSIS will give 30 days written notice of the termination of that waiver. Otherwise, establishments will need to submit a request for a new waiver from FPS requirements under traditional inspection with information on how the waiver would provide new information that would facilitate definite improvements (9 CFR 303.1(h)).FSIS expects that it will be difficult for establishments to meet requirements necessary to obtain a waiver once NSIS is available.

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Appendix 1- Microbiological Sampling Plan Self-Assessment Checklist

1.	**Written microbiological sampling plan**
	a. Sample Collection ❑ Procedure for random selection of carcasses or FCS for sampling ❑ Location within process where samples are collected. ❑ Pre-evisceration ❑ Bleed Out ❑ Other ❑ Post-chill ❑ Pre-op FCS ❑ Employee knife ❑ Frequency of sample collection ❑ Aseptic technique for gloving and sample collection ❑ Description of sample collection procedure Sponge sampling ❑ Designated employee to collect the sample ❑ Date and time collected
	b. Sample Handling and Shipping ❑ Proper sample handling and packaging to ensure sample integrity ❑ Sample identification ❑ Held under refrigeration/not frozen ❑ Packed in an insulated shipping container with cold packs ❑ Shipped to the testing laboratory on same day as collected ❑ Name of person or service (e.g., FedEx or courier service) transporting the sample ❑ Chain-of-custody documentation when samples transported from the establishment to an off-site laboratory (e.g., by a delivery service such as FedEx or courier) ❑ Holding time met (time from collection to analysis)
	c. Testing method and Test Results Reporting ❑ Description of the testing method used by laboratory ❑ Microbiological test results report received from testing laboratory ❑ Results reported in appropriate units of measure ❑ Test results recorded on a control chart (moving window format) ❑ Interpretation of results based on defined process control criteria ❑ Acceptable

	❑ Unacceptable ❑ Actions taken in response to test results and trends in results over time
2.	**Testing Laboratory**
	a. Establishments should refer to the FSIS *Establishment Guidance for the Selection of a Commercial or Private Microbiological Testing Laboratory* for guidance on selecting a microbiological testing laboratory. The checklist provided in the guidance is intended to assist establishments to determine whether a microbiological laboratory is capable of producing accurate and reliable results. Some of the general criteria to consider in selecting a testing laboratory include: ❑ Personnel ❑ Facilities ❑ Equipment ❑ Operations ❑ Analytical methods
	b. Laboratory testing method FSIS has made available a list of *Foodborne Pathogen Test Kits Validated by Independent Organizations* for the detection of relevant foodborne pathogens (i.e., *Salmonella*, *Campylobacter*, *E. coli* O157:H7, and *Listeria spp*. including *L. monocytogenes*). This list is intended to be informational and is not an endorsement or approval of any particular method, regardless of its inclusion in the list. Some of the general criteria to consider when selecting a method include: ❑ Sample size analyzed ❑ Microorganism tested for (e.g., *Salmonella*, APC, generic *E. coli*) ❑ Analytical method used (e.g., AOAC, NordVal) ❑ Date test was received at the laboratory ❑ Date analysis was started ❑ Date analysis was completed ❑ Analytical results recorded and reported to establishment ❑ Corrective actions related to test results, such as laboratory error, unacceptable sample temperature upon arrival

Appendix 2 – Possible Food Contact Surface Sites (This is not an exhaustive list)

Aprons
Belts
Blades
Carts
Coats
Conveyors
Cutting boards
Equipment surfaces
Equipment shields
Fabrication tables
Fabrication saws
Gambrel table
Gloves
Guiding bars
Injecting machines
Knives
Packaging materials
Polisher
Racks
Saw table
Saws
Scales
Scoops
Steam Vac
Thermometers
Utensils