# FSIS Ready-to-Eat Fermented, Salt-Cured, and Dried Products Guideline May 5, 2023

**Document ID:** FSIS-GD-2023-0002

This guideline provides information on the Agency regulatory requirements associated with safe production of ready-to-eat (RTE) shelf-stable, fermented, salt-cured, and dried products that rely on multi-hurdle approaches to achieve lethality and shelf-stability. It applies to small and very small meat and poultry official establishments although large establishments can also benefit from the information. It relates to 9 CFR part 417.

# Table of Contents

## Preface

This guideline represents FSIS' current thinking on this topic and should be considered usable as of this issuance. The information in this guideline is provided to assist meat and poultry establishments in meeting the regulatory requirements. The contents of this document do not have the force and effect of law and are not meant to bind the public in any way. This document is intended only to provide clarity to industry regarding existing requirements under the regulations. Under the regulations, meat and poultry establishments may choose to implement different procedures than those outlined in this guideline, but they would need to validate and support how those procedures are effective.

This guideline is focused on small and very small plants in support of the Small Business Administration's initiative to provide small businesses with compliance assistance under the Small Business Regulatory Enforcement Fairness Act (SBREFA). However, all meat and poultry establishments may apply the recommendations in this guideline. It is important that small and very small establishments have access to a full range of scientific and technical support, and the assistance needed to establish safe and effective Hazards Analysis and Critical Control Point (HACCP) systems. Although large plants can benefit from the information, focusing the guideline on the needs of small and very small establishments provides them with assistance that may be otherwise unavailable to them.

## Purpose

This guideline is designed to respond to commonly asked questions from small and very small establishments that manufacture RTE, shelf-stable, fermented, salt-cured, and dried meat and poultry products where cooking is not the primary lethality step about:

- Recommended targets for reduction or prevention of each hazard.
- The key steps in each process needed to ensure safety.
- The critical operational parameters associated with each step.
- The biological hazards associated with and scientific support available to help produce the following products:
    - Fermented dry and semi-dry fermented sausages including:
        - Lebanon bologna
        - Summer sausage
        - Pepperoni
        - Salami, including Genoa
        - Soudjouk
    - Salt-cured products including:
        - Basturma
        - Country cured ham
        - Bresaola
    - Dried products including:
        - Biltong
        - Droëwors.

Establishments can always seek guidance from State university extension service specialists and HACCP Coordinators on developing food safety plans not provided in this guideline to comply with HACCP regulatory requirements. Small and very small establishments can also seek guidance from the Niche Meat Processor Assistance Network, a university Extension-based community of practice.

## Reason for Issuing the Guideline

FSIS has several documents that address the safe production of ready-to-eat (RTE) meat and poultry products. However, based on questions received, FSIS determined that the current documents listed below do not adequately address the specific considerations related to supporting the lethality and shelf-stability of RTE shelf-stable fermented, salt-cured, and dried meat and poultry products. The available documents for RTE products include:

- The FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A) - provides guidance on using cooking as a lethality treatment.
- The FSIS Stabilization Compliance Guideline for Meat and Poultry Products (Revised Appendix B) - provides guidance on stabilization treatments (primarily cooling and hot-holding).
- The FSIS Compliance Guideline for Meat and Poultry Jerky Produced by Small and Very Small Establishments - provides guidance about how to safely produce jerky.
- The FSIS Guideline: Controlling *Listeria monocytogenes* in Post-Lethality Exposed RTE Meat and Poultry Products - provides guidance on complying with the requirements for 9 CFR 430 to address *Listeria monocytogenes* (*Lm*) contamination in the post-lethality exposed environment.
- FSIS Guideline for the Prevention and Control of *Trichinella* and Other Parasitic Hazards in Pork and Products Containing Pork - provides guidance on understanding the available options that are effective for the prevention and control of *Trichinella spiralis* and other parasitic hazards in pork.

FSIS had some information related to the safe production of fermented, salt-cured, and dried meat and poultry products that was previously available in the following documents and has been now incorporated into this guideline:

- "Food Safety Lessons Learned from the Lebanon Bologna Outbreak" – Although Lebanon bologna is a unique product which is fermented to a low pH, FSIS incorporated information from that guideline into this

### KEY DEFINITIONS

**Ready-to-eat** meat and poultry is defined by FSIS in 9 CFR 430.1 as a meat or poultry product that is in a form that is edible without additional preparation to achieve food safety and may receive additional preparation for palatability or aesthetic, epicurean, gastronomic, or culinary purposes.

**Shelf-stable** for the purposes of meat and poultry products is defined as the condition achieved when meat and poultry products can be stored under ambient temperature and humidity conditions; if the package integrity is maintained during storage, shipping, and display at retail and in the home; and the product will not spoil or become unsafe throughout the manufacturer's specified shelf-life.

larger document because a lot of the information can be applied to other semi-dry fermented products (Getty, 1999; Vignolo *et al.*, 2010).
- "Challenge Study – *Escherichia coli* O157:H7 in Fermented Sausage" – relevant information was incorporated into Appendix 15 of this document.

The guideline also includes lessons learned from two salmonella outbreaks in 2021 associated with ready-to-eat fermented, dried, and salt-cured meat Italian-style meat products. These lessons learned are also included in FSIS' Outbreak Investigation After Action Review.

## How to Effectively Use this Guideline

This guideline is organized to provide users with an overview of topics related to the safe production of RTE shelf-stable fermented, salt-cured, and dried meat and poultry products. Additional details about each topic are included in appendices. To use this guideline, FSIS recommends that readers review the overview of each of the topics and consult relevant appendices for more detail. Hyperlinks will quickly take you to the correct place in the document electronically and are also provided to other complementary guidelines. Users should note that words that are **bolded** throughout the document are defined in the Glossary in Appendix 16.

Appendices 12, 13, and 14, contain summaries of available scientific articles. These summaries are provided to help establishments identify relevant scientific support for supporting decisions in the hazard analysis. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

## How to Comment on the Guideline

FSIS is seeking public comment on this guideline 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 from publication of the *Federal Register* Notice and, as appropriate, the agency may update this guideline in response to comments. Although FSIS may make changes to the guideline in response to comments, this document reflects FSIS's current thinking, and FSIS encourages establishments producing products discussed in this document to review it.

Comments may be submitted by either of the following methods:

- Federal eRulemaking Portal Online submission at *regulations.gov*. This website provides a way to type short comments directly into the comment field on the webpage or attach a file to submit lengthier comments. Follow the online instructions at that site to submit comments.

- Mail and hand- or courier-delivered items: Send to Docket Clerk, U.S. Department of Agriculture (USDA), FSIS, 1400 Independence Avenue SW, Room 6065, Washington, D.C. 20250-3700.

All items submitted by mail or electronic mail must include the Agency name, FSIS, and document title: **FSIS Ready-to-Eat Fermented, Salt-Cured, and Dried Products Guideline.** Comments received will be made available for public inspection and posted without change, including any personal information, to http://www.regulations.gov.

## Questions Regarding Topics in this Guideline

If after reading this guideline you still have questions, FSIS recommends searching the publicly posted Knowledge Articles ("Public Q&As") in the askFSIS database. If after searching the database, you still have questions, refer them to the Office of Policy and Program Development through askFSIS and select **HACCP Deviation & HACCP Validation** as the Inquiry Type or by telephone at 1-800-233-3935.

Documenting these questions helps FSIS improve and refine present and future versions of the guideline and associated issuances.

# FSIS RTE Fermented, Salt-Cured, and Dried Products Guideline

## Background

Multi-hurdle products in the context of this guideline are those products that rely on a combination of hurdles or processing steps to eliminate pathogens of concern and result in a RTE shelf-stable product. These hurdles typically include processing steps such as fermentation, salt-curing, and drying that use a combination of factors such as reduced pH, reduced **water activity** (also referred to as $a_w$) over time, high brine, or salt concentration to kill bacteria and prevent their outgrowth during storage. Generally, one step is not sufficient on its own to eliminate pathogens of concern, instead a combination of processing steps or hurdles is needed.

## Products Covered by this Guideline

This guideline focuses on the safe production and supporting documentation for the following **RTE shelf-stable products** that are produced under the Not Heat-Treated Shelf-Stable HACCP category or Heat-Treated Shelf-Stable HACCP category. Examples of scientific support are included in appendices for products in **bold** because these are common products FSIS receives questions about and are products where scientific support is available**:**

| Product Group | Products in this Category Include… | For More Information (Go to page): |
|---|---|---|
| Fermented | **Genoa salami**, hard salami, **pepperoni**, turkey pepperoni, **summer sausage**, Abruzzese, **Lebanon bologna**, sopressata, thuringer, mettwurst, saucisson, chorizo, chourico, **soudjouk (sujuk or soujouk)**, pickled pigs' feet, bologna in vinegar, **landjager** | 15 |
| Salt-cured | Prosciutto ham, Parma ham, Westphalian ham, Bayonne ham, Serrano ham, Black Forest ham, **country ham**, pancetta, coppa, capocolla, **bresaola**, beef prosciutto, **basturma**, duck prosciutto, linguica, salchichon | 18 |
| Dried | dried beef, beef jerky[1], beef nuggets, steak tenders, kippered beef, meat sticks, turkey jerky, tasajo, pemmican, pipi kaula, **droëwors**, **biltong**, jamon (jambon), longanisa, (some) saucisson, (some) chorizo, dried soup mixes/soup bases, freeze-dried entrees, fried pork skins/rinds/cracklings/chicharrones, lard | 20 |

[1]Jerky is not covered in this guideline as explained on the next page.

## Products NOT covered by this Guideline

This guideline does not address the production of products classified as not ready-to-eat (NRTE) or not **shelf-stable;** however, labeling considerations are provided in Appendix 1 to address commonly asked questions.

This guideline does not cover jerky, which is considered a dried product, as most jerky processes rely on cooking alone (*e.g.*, by following the FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A)) to achieve lethality. Guidance regarding the production of jerky can be found in the FSIS Guideline for Meat and Poultry Jerky Produced by Small and Very Small Establishments.

## Biological Hazards of Concern in Multi-hurdle Lethality Products

The following section and the supplemental information in Appendix 2 are designed to complement FSIS' Meat and Poultry Hazards and Control Guide and to further assist establishments in conducting a hazard analysis for fermented, salt-cured, and dried products as required by 9 CFR 417.2(a)(1) and for supporting decisions in the hazard analysis as required by 9 CFR 417.5(a)(1).

### KEY DEFINITIONS

**Water activity**, abbreviated as $a_w$, is a measure of the concentration of moisture (*i.e.*, water) and its availability in a food. The amount of water available in a food depends on the total concentration of all dissolved substances in the product because they bind water. Thus, if ingredients such as salt or sugar are added to food, they compete with the bacteria for available water.

**Outgrowth of the following are hazards in raw products that fermentation, salt-curing, or drying steps should be designed to limit in the finished RTE product:**

- *Staphylococcus aureus* (*S. aureus*)
- *Clostridium perfringens* (*C. perfringens*)
- *Clostridium botulinum* (*C. botulinum*)

**The following are hazards present in raw products that the fermentation, salt curing, or drying steps should be designed to destroy in the finished RTE product:**

- *Salmonella*
- STEC
- *Lm*
- *Trichinella spiralis* (*T. spiralis*) and *Toxoplasma gondii* (*T. gondii*) (greater risk of infection for feral or non-confinement raised swine)

These hazards may be of concern at various points in the production process and multiple hurdles may be needed to address each hazard. Appendix 2 has some specific considerations that can help inform an establishment's hazard analysis decision-making. Information on mold as a potential biological hazard of concern can also be found in Appendix 2.

## Recommended Targets for Biological Hazards

For each biological hazard identified, establishments need to identify **targets** for reduction or prevention. Targets are quantifiable pathogen reduction levels or growth limits set by the establishment to produce safe products in the absence of regulatory performance standards. Targets are used by the establishment to demonstrate that the lethality and shelf-stability processes achieved by their food safety system prevents, eliminates, or reduces pathogens to acceptable levels. For example, FSIS recommends that the **lethality treatment** (*i.e.*, the combination of hurdles or steps) for RTE shelf-stable meat and poultry products achieve at least a 5.0-log reduction of *Salmonella*, STEC (in beef), and at least a 3.0-log reduction in *Lm*. For more information on lethality and shelf-stability targets see Appendix 3.

## Steps or Hurdles to Ensure Food Safety

For fermented, salt-cured, and dried products, lethality and shelf-stability are achieved using multiple process steps or hurdles. The following are the key process steps and hurdles used to achieve lethality and shelf-stability in fermented, salt-cured, and dried products. The key hurdles and critical operational parameters for each will be described in more detail for each process.

### KEY DEFINITIONS

**Lethality** is the process or combination of processes that ensures that no *Salmonella* organisms remain in the finished product, as well as reduces other pathogens and their toxins or toxin metabolites. Examples of lethality processes include cooking, fermentation, salt-curing, and drying.

**Stabilization** is the process of preventing or limiting the growth of spore-forming bacteria capable of producing toxins either in the product before consumption or in the human intestine after consumption. Establishments may use a variety different stabilization processes such as cooling, hot-holding, or meeting and maintaining certain pH or water activity levels.

**NOTE:** The processing steps used to achieve shelf-stability also often stabilize a product to prevent or limit the growth of spore-forming bacteria by reducing the pH or water activity. For more information on stabilization see FSIS' Stabilization Guideline for Meat and Poultry Products (Revised Appendix B).

**Table 1. Key Steps or Hurdles Used to Achieve Lethality and Shelf-Stability by Product Group**

| | **Seasoning/ Marination** | **Fermentation** | **Low-Temperature Heat Step** | **Salt-curing/ Equalization** | **Drying** |
|---|---|---|---|---|---|
| *Hurdle/ Product Group* | *Addition of salt and nitrite or nitrate/Lower pH* | *Lower pH/ Competitive Microflora/ Bacteriocin production* | *Heat* | *High brine/Reduced water activity* | *Reduced water activity over time* |
| *Fermented* | *X* | *X* | *Optional* | | *X* |
| *Salt-cured* | *X* | | *Optional* | *X* | *X* |
| *Dried* | *X* | | | | *X* |

In addition to applying multiple hurdles, it is also important that establishments understand and:

- Adhere to the basic principles of Sanitation Standard Operating Procedures (SSOPs) (9 CFR 416).
- Follow Good Manufacturing Practices (GMPs).
- Address product handling post-lethality.

## Validation – Element 1: Scientific Support

Once an establishment identifies the key steps in its process it should identify the scientific support available that closely matches the actual process and shows that the process achieves the desired lethality target as part of the validation process. There are two distinct elements to validation: 1) the scientific or technical support for the HACCP system design (design) and 2) the in-plant validation data (execution). This guideline focuses on the scientific support available to meet the first element of validation. As discussed in the FSIS HACCP Systems Validation Guideline, to meet the first element of validation (*i.e.*, the scientific or technical support) establishments may use:

- Published processing guidelines.
- Peer-reviewed scientific or technical data or information.
- Expert advice from processing authorities.
- Validated pathogen modeling programs.
- Challenge or inoculated pack studies.
- Data gathered by the establishment in-plant.
- Regulatory performance standards.
- Best practice guidelines.

Any of these types of support may be acceptable provided they are complete and the critical operational parameters match the establishment's process. Some specific considerations for each of the types of supporting documents when used to support lethality and shelf-stability of fermented, salt-cured, and dried products are given in Appendix 4.

Establishments should carefully identify the **critical operational parameters** used in the actual process and compare those to the ones used in the scientific support. Critical operational parameters are the specific conditions that the intervention must operate under for it to be effective. Examples of critical operational parameters used during fermentation, salt-curing, and drying include:

- Antimicrobial application.
- Fermentation temperature, target pH, time to reach target pH, and smoke (if used).
- Type and use of starter cultures.
- Curing time.
- Curing temperature.
- Salt coverage of exposed muscle tissue.
- Drying room temperature.
- Drying time (*i.e.*, days or weeks).
- Product characteristics including product formulation.

Establishments must implement critical operational parameters in the actual production process consistent with the parameters in the scientific or technical support or provide justification for why differences would not impact the efficacy of the intervention (9 CFR 417.5(a)(1)). When different levels of a critical operational parameter other than those in the support document are used, establishments should consider developing a decision-making document that explains the scientific rationale for why the different level would not affect the efficacy of the intervention or process or why the different studies support the effectiveness of the process when combined. This scientific rationale should include reference to data or scientific principles as support. Establishments may be able to use in-plant validation data when differences between the actual process and support are small. For more information see the Appendix 4 section about "Data gathered by the establishment in-plant". FSIS recommends establishments consider making a table to do a side-by-side comparison of the parameters in the actual process and those used in the actual study. An example is provided below in Table 2.

**Table 2. Example of Side-by-Side Comparison of Parameters Used in the Support vs. the Actual Process and Rationale for Differences**

| Process Step | Critical Operational Parameter | Level Used in the Scientific Support | Level Used in Establishment's Actual Process | Rationale for Why the Difference is Acceptable |
|---|---|---|---|---|
| *Post-fermentation low temperature heat step* | *Hold time and temperature* | *Internal temperature of **128**°F for 1h from Hinkens et al., 1996* | *Internal temperature of **134**°F for 1h* | *Actual process uses higher temperature and same dwell time which should result in same or higher reductions per FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A).* |

**CASE STUDY: The importance of using critical operational parameters consistent with the scientific support**

- *In March 2011, there was a recall of a Lebanon bologna product that was associated with a foodborne illness outbreak of E. coli O157:H7.*
- *FSIS' investigation revealed the establishment had not properly validated their process. The establishment's supporting documentation for the critical operational parameters did not match the actual commercial process used:*

| Critical operational parameter | Actual process | Supporting Documentation |
|---|---|---|
| *Diameter* | *52 to 119 mm* | *27 mm* |
| *Casing* | *Permeable casing* | *Impermeable sealed glass tube* |
| *Cooking equipment* | *Large smokehouse fitted with single heat source, humidity was not well-controlled* | *Well-controlled water bath* |

- *Differences likely led to a lower reduction in foodborne pathogens of concern in the actual process than what was demonstrated in the supporting documentation.*
- *This outbreak highlights the importance of identifying supporting documentation that is representative of the actual process so that the results can be repeatable.*

## Validation – Element 2: In-plant validation

As discussed in the FSIS HACCP Systems Validation Guideline, if establishments have adequate scientific support and implement the critical operational parameters in the actual process consistent with the scientific support then to meet the second element of validation (*i.e.*, the scientific or technical support) the establishment should collect in-plant data that demonstrates that the critical operational parameters are being met. Establishments should develop the appropriate in-plant data during the initial 90 days of implementing a new HACCP system, or whenever a new or modified food safety hazard control is introduced into an existing HACCP system (e.g., as implemented after a HACCP plan reassessment).

**NOTE:** For some salt-cured products, the initial validation period may extend beyond 90 calendar days due to the nature of the process and the length of time it takes to implement the critical operational parameters that impact lethality. To determine whether the system is properly designed and executed, even though the regulations provide 90 days for initial validation, an establishment needing more than 90 days can ask the district office, in writing, for additional time to collect at least 13 production days of records when it first starts operating, when it begins producing new product, or for a modified HACCP plan if the results of a reassessment indicate additional support is needed. In the request, an establishment should state why more than 90 days are needed to collect the in-plant validation data, and how it plans to gather at least 13 production days' worth of in-plant validation data. The request will then be evaluated on a case-by-case basis. The establishment should consider focusing validation activities

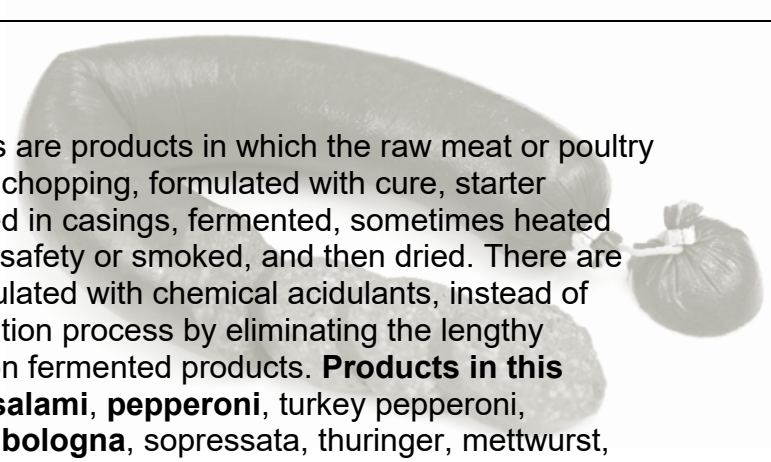
on the product produced most frequently within each HACCP category. In addition, the establishment may consider evaluating data collected for products across multiple HACCP categories to determine whether the data together can support its ability to meet critical operational parameters (80 FR 27557).

A discussion of how microbiological data gathered as part of initial validation can be used to support a process when differences between the scientific support and the actual process are small can be found in Appendix 4 under the section about “Data gathered by the establishment in-plant”.

The next part of the guideline includes an overview of each of the three following types of products: 1) fermented, 2) salt-cured, and 3) dried products:

- The biological hazards of concern for fermented, salt-cured, and dried products.
- The recommended targets to address those biological hazards.
- The steps or hurdles to ensure food safety.
- The types of scientific support that can support the recommended targets are met.
- The importance of understanding the critical operational parameters in your process and how they relate to those used in the scientific support.

## Overview of Fermented Products



RTE fermented meat and poultry products are products in which the raw meat or poultry are usually reduced in size by grinding or chopping, formulated with cure, starter culture, salt and seasoning mixture, stuffed in casings, fermented, sometimes heated with a low temperature heat step for food safety or smoked, and then dried. There are also RTE acidified products that are formulated with chemical acidulants, instead of starter cultures, to accelerate the acidification process by eliminating the lengthy fermentation step. This section focuses on fermented products. **Products in this category include: Genoa salami**, **hard salami**, **pepperoni**, turkey pepperoni, **summer sausage**, Abruzzese, **Lebanon bologna**, sopressata, thuringer, mettwurst, saucisson, chorizo, chourico, **soudjouk (sujuk or soujouk)**, pickled pigs' feet, bologna in vinegar, and **landjager**. See examples of scientific support for products in bold in Appendix 12.

**Table 3. Overview of hazards of concern establishments should consider in the hazard analysis during lethality and stabilization and typical controls for fermented products.**

| Hazard | Source | Adulterant (Yes/No) | Recommended Target | *Fermentation* | *Drying* |
|---|---|---|---|---|---|
| ***Salmonella* STEC (beef) *Lm*** | Raw meat and poultry, spices, herbs | Yes – zero tolerance | 5-log reduction for *Salmonella* and STEC<br>3-log reduction for *Lm*<br><br>For more information including on **alternative lethality targets** see Appendix 3 | Effectiveness for *Salmonella*, STEC, and *Lm* depends on:<br>Fermentation temperature, target pH, and time to reach target pH;<br>Starter culture;<br>Product characteristics;<br>Consider use of a low temperature heat step<br>See Appendix 6 | Effectiveness for *Salmonella*, STEC, and *Lm* depends on:<br>Drying room temperature<br>Drying time<br>Target water activity<br>Product characteristics<br>See Appendix 11 |
| ***S. aureus*** | Raw meat and poultry spices, herbs | Yes – depending on level | During production: no more than 2-log growth;<br>During storage: no outgrowth | For *S. aureus*, degree-hours concept during production<br>See page 37 and GMPs for Fermented Products | Drying is effective for *S. aureus* during storage after water activity is reduced See Appendix 5 |
| ***C. perfringens* *C. botulinum*** | Raw meat and poultry, spices, herbs | Yes – depending on level | No more than 1-log growth *C. perfringens*; no multiplication of *C. botulinum* | For *Clostridia*, starter culture, dextrose, nitrite, and salt<br>See page 38 | Drying is effective for *Clostridia* after water activity is reduced See FSIS' Stabilization Guideline |
| ***Trichinella spiralis* and *Toxoplasma gondii* (pork)** | Raw pork (greater infection risk: feral or non-confinement swine) | Yes – zero tolerance | Eliminate larvae | For *Trichinella,* fermentation can be effective in combination with salt and drying<br>See Porto-Fett (2010) and FSIS Trichinella Guideline | For *Trichinella*, drying can be effective in combination with salt, smoke, etc. See Sausage Treatment Methods 1-7 in FSIS Trichinella Guideline |
| **Molds** | Any food product | Maybe depending on type | No unintentional mold growth on finished product | May apply live mold culture to prevent growth of undesirable molds. Otherwise, rely on sanitation | Drying not effective for molds - rely on sanitation |

**Addressing *Salmonella*, STEC, and *Lm*:** Fermented products have been associated with *Salmonella and* STEC outbreaks (see Table 6), and FSIS has detected *Salmonella* and *Lm* in these products (see Appendix 2). The literature shows fermentation and drying alone do not generally achieve a 5-log reduction of STEC and *Salmonella* or a 3-log reduction of *Lm* (Faith *et al.*, 1997; Faith *et al.*, 1998a; Faith *et al.*, 1998b; Hussein *et al.,* 2022; Ihnot *et al.*, 1998). However, there are some conditions that have been found to result in a 5-log reduction as a result of additional interventions such as:

- A high fermentation temperature and a low final pH (Blue Ribbon Task Force, 1996).
- A low pH following fermentation that is maintained or decreases during drying along with a long drying time (Deibel Laboratories/Chr. Hansen, 2017; Gunvig *et al.*, 2017).
- After fermentation but before drying, apply a low-temperature heat step (Calicioglu *et al.*, 1997; Hinkens *et al.*, 1996) **or** FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A) time/temperature/humidity combination.
- Use of high pressure processing (HPP) prior to the fermented sausages reaching a water activity of less than 0.90 (Balamurugan, 2019).
- Addition of a finishing phase (Hussein *et al.,* 2022) or storage under vacuum at room-temperature for extended time following fermentation and drying (Calicioglu, 2002; Faith *et al.*, 1997; Faith *et al.*, 1998a; Faith *et al.*, 1998b; Ihnot *et al.*, 1998; Ingham *et al.*, 2004; Ingham *et al.*, 2005; Porto-Fett *et al.*, 2008).

**Critical Steps and Critical Operating Parameters:**

Fermentation (see Appendix 6 and Appendix 7 for more information):

- Fermentation temperature, target pH, time to reach target pH, and smoke (if used).
- Type and use of starter cultures.
- Product characteristics: Casing diameter and shape and product formulation including salt, sugar (type and level), and use of nitrite or nitrate.

Low-Temperature Heat Step (Optional) (see Appendix 8 for more information)

- Time and temperature: Heating come-up time (CUT), hold time, and temperature for low temperature heating step.
- Equipment used to generate heat.
- Product characteristics.

Drying (see Appendix 11 for more information):

- Drying room temperature.
- Drying time.
- Target water activity.
- Product characteristics.

***Key Points – Fermentation and Drying***

Fermentation and drying alone are not particularly effective lethality treatments. When these steps are found to be effective, there are a lot of critical operational parameters that need to be implemented consistent with the scientific support. It is not enough to only meet degree-hours, follow a drying or salt-curing method for *Trichinella*, and achieve a final water activity for shelf-stability. Degree-hours are intended to control the outgrowth of *S. aureus*. To reduce levels of other pathogens such as *Salmonella*, products often need to be fermented to a lower pH than 5.3. Also, methods for controlling *Trichinella* have not been validated for controlling *Salmonella* and product may need to be dried longer to reduce levels of *Salmonella* in the product.

**Available Scientific Support:** Below is a list of available scientific support for the reduction of *Salmonella*, STEC, and *Lm* in fermented products**.** For detailed summaries of these common scientific support articles for fermented products see Appendix 12.

**Fermented Sausage**

- Nickelson, R., II, J. Luchansky, C. Kaspar, and E. Johnson. 1996. Update on dry fermented sausage *Escherichia coli* O157:H7 validation research. Blue Ribbon Task Force. Report No. 11-316.

**Summer Sausage**

- Calicioglu, M., Faith, N.G., Buege, D.R., and Luchansky, J.B. 1997. Viability of *Escherichia coli* O157:H7 in Fermented Semi-dry Low-Temperature-Cooked Beef Summer Sausage. J. Food Prot. 60(1): 1158-1162.

**Lebanon-Style Bologna**

- Getty, K.J.K, Phebus, R.K, Marsden, J.L., Schwenke, J.R., and Kastner, C.L. 1999. Control of *Escherichia coli* O157:H7 in Large (115 mm) and Intermediate (90 mm) Diameter Lebanon-style Bologna. J. of Food Sci. 64(6): 1100-1107.

**Salami**

- Deibel Laboratories/CHR. Hansen. 2017. Fate of *Salmonella Spp*., *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Staphylococcus aureus* Inoculated Into a Non-Heated and Dried Salami Product. Available from CHR. Hansen Inc. upon request. https://www.chr-hansen.com/en/contact-us
- Faith, N. G., Parniere, N., Larson, T., Lorang, T.D., Kaspar, C.W., Luchansky, J.B. 1998a. Viability of *Escherichia coli* O157:H7 in salami following conditioning of batter, fermentation and drying of sticks, and storage of slices. J. Food Prot. 61:377-382.
- Hussein, M.H., Burroughs, S., Emch, A.W., Waite-Cusic, J. 2022. Enhancing the Reduction of *Salmonella* and *Listeria monocytogenes* During Traditional Salami Processing by Adding a Finishing Phase. Food Control. 131: 108432. [2]
- Porto-Fett, A.C.S., Call, J.E., Shoyer, B.E., Hill, D.E., Pshebniski, C., Cocoma, G.J., and Luchansky, J.B. 2010. Evaluation of fermentation, drying, and/or high pressure processing on viability of *Listeria monocytogenes, Escherichia coli* O157:H7, *Salmonella* spp., and *Trichinella spiralis* in raw pork and Genoa salami. Int. Journal of Food Micro. 140: 61-75.

**Pepperoni**

- Hinkens, J.C., Faith, N.G., Lorang, T.D., Bailey, P., Buege, D., Kaspar, C.W., Luchansky, J.B. 1996. Validation of Pepperoni Processes for Control of *Escherichia coli* O157:H7. J. Food Prot. 59(12): 1260-1266.
- Ihnot, A.M., Roering, A.M., Wierzba, R.K., Faith, N.G., Luchansky, J.B. 1998. Behavior of *Salmonella typhimurium* DT104 during the manufacture and storage of pepperoni. International Journal of Food Microbiology. 40:117-121.
- Faith, N.G., Parniere, N., Larson, T., Lorang, T.D., Luchansky, J.B. 1997. Viability of *Escherichia coli* O157:H7 in pepperoni during the manufacture of sticks and subsequent storage of slices at 21, 4 and -20°C under air, vacuum and $CO_2$. Int. Journal of Food Micro. 37:47-54.

**Soudjouk**

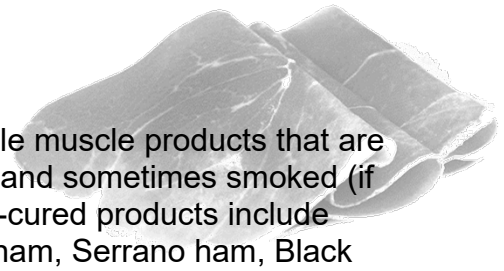
- Calicioglu, M., N. G. Faith, D. R. Buege, Luchansky, J.B. 2001. Validation of a Manufacturing Process for Fermented, Semidry Turkish Soudjouk to Control *Escherichia* coli O157:H7. J. Food Prot. 64(8):1156-1161.
- Calicioglu, M., N. G. Faith, D. R. Buege, Luchansky, J.B. 2002. Viability of Escherichia coli O157:H7 during manufacturing and storage of fermented, semidry soudjouk-style sausage. J. Food Prot. 65:1541-1544.
- Porto-Fett, A.C.S., Hwang, C.A., Call, J.E., Juneja, V.K., Ingham, S.C., Ingham, B.H., Luchansky, J.B. 2008. Viability of multi-strain mixtures of *Listeria monocytogenes*, *Salmonella typhimurium*, or *Escherichia coli* O157:H7 inoculated into the batter or onto the surface of a soudjouk-style semi-dry sausage. Food Microbiology. 25: 793-801.[2]

**Landjäger**

- Rivera-Reyes, M., Campbell, J.A., Cutter, C.N. 2017. Pathogen reductions associated with traditional processing of Landjäger. Food Control. 73: 768-774. [2]

[2] Studies did not achieve a 5.0-log reduction so either a finishing phase (Hussein *et al.,* 2022) or storage step under vacuum should be added (see Rivera-Reyes *et al.*, 2017 and Porto-Fett *et al.* 2008) or additional supporting documentation (*i.e.,* journal article or challenge study) should be provided.

## Overview of Salt-Cured Products



RTE salt-cured meat and poultry products are usually whole muscle products that are cured with salt and sodium nitrite or nitrate, then air dried, and sometimes smoked (if desired for certain flavor characteristics). Examples of salt-cured products include Prosciutto ham, Parma ham, Westphalian ham, Bayonne ham, Serrano ham, Black Forest ham, **country ham**, pancetta, coppa, capocolla, **bresaola**, beef prosciutto, **basturma**, duck prosciutto, linguica, and salchichon. Because many products rely on **dry-curing**, the guideline focuses on the critical operational parameters of dry-curing. See examples of scientific support for products in bold in Appendix 13.

**Table 4. Overview of hazards of concern establishments should consider in the hazard analysis during lethality and stabilization and typical controls for salt-cured products.**

| Hazard | Source | Adulterant (Yes/No) | Recommended Target | *Salt-curing/Equalization* | *Drying* |
|---|---|---|---|---|---|
| ***Salmonella* STEC (beef) *Lm*** | Raw meat and poultry, spices, herbs | Yes – zero tolerance | 5-log reduction for *Salmonella* and STEC<br>3-log reduction for *Lm*<br>For more information including on **alternative lethality targets** see Appendix 3 | Effectiveness for *Salmonella*, STEC, *Lm* depends on:<br>Curing temperature<br>Curing time<br>Salt coverage of exposed tissue<br>Product characteristics<br>See Appendix 9 | Effectiveness for *Salmonella*, STEC, *Lm* depends on:<br>Drying room temperature<br>Drying time<br>Target water activity<br>Product characteristics<br>See Appendix 11 |
| ***S. aureus*** | Raw meat and poultry spices, herbs | Yes – depending on level | No more than 2-log growth during production; no outgrowth during storage | For *S. aureus*, high enough brine concentration/low enough water activity prior to drying<br>See page 36 | Drying is effective for *S. aureus* during storage after water activity is reduced See Appendix 5 |
| ***C. perfringens* *C. botulinum*** | Raw meat and poultry, spices, herbs | Yes – depending on level | No more than 1-log growth *C. perfringens*; no multiplication of *C. botulinum* | For *Clostridia*, resting/equalization critical, high enough brine concentration/low enough water activity prior to drying, use of nitrite or nitrate<br>See page 38 | Drying is effective for *Clostridia* after the water activity is reduced See FSIS' Stabilization Guideline |
| ***Trichinella spiralis* and *Toxoplasma gondii* (pork)** | Raw pork (greater risk of infection for feral or non-confinement raised swine) | Yes – zero tolerance | Eliminate larvae | Effective for *Trichinella* with curing, equalization, and drying per methods for capocollo, hams and pork shoulder picnics, boneless pork loins in and country ham in FSIS Trichinella Guideline | Effective for *Trichinella* with curing, equalization, and drying per methods for capocollo, hams and pork shoulder picnics, boneless pork loins in and country ham in FSIS Trichinella Guideline |
| **Molds** | Any food product | Maybe depending on type | No unintentional mold growth on finished product | Not effective for molds – rely on sanitation | Not effective for molds – rely on sanitation |

**Addressing STEC, *Salmonella* and *Lm*:** Salt-cured products have been associated with *Salmonella* outbreaks (see Table 6) and FSIS has detected *Salmonella* and *Lm* in these products (see Appendix 2). There is limited literature available that supports salt-curing and drying alone achieve a 5-log reduction of STEC and *Salmonella* or a 3-log reduction of *Lm* (Ingham *et al.*, 2006; Genigeorgis and Lindroth, 1984; Reynolds *et al.*, 2001).

However, there are some examples that have been found to result in a 5-log reduction:

- Dry-curing beef strips, followed by a low-temperature heat step, followed by drying (Genigeorgis and Lindroth, 1984).
- Dry-curing hams with extended equalization and storage (Reynolds *et al.*, 2001).

**Critical Steps and Critical Operating Parameters:**

Dry-Curing and Salt-Equalization (see Appendix 9 for more information):

Dry-Curing

- Curing temperature.
- Curing time.
- Salt coverage of exposed muscle tissue.
- Product characteristics (*e.g.*, product size and formulation, including salt concentration).

Salt-Equalization

- Equalization temperature.
- Equalization time.
- Brine concentration and water activity after equalization.
- Product size (diameter or thickness).

Drying (see Appendix 11 for more information):

- Drying room temperature.
- Drying time.
- Target water activity.
- Product characteristics.

**NOTE:** High pressure processing (HPP) may also be used for salt-cured hams to add to the overall process lethality (Perez-Baltar, 2020).

**Available Scientific Support:** Below is a list of available scientific support for the reduction of *Salmonella*, STEC, and *Lm* in salt-cured products**.** For detailed summaries of these common scientific support used for salt-cured products see Appendix 13.

**Basturma**

- Ingham, S.C., Searls, G., Buege, D.R.. 2006. Inhibition of *Salmonella* serovars, *Escherichia coli* O157:H7, and *Listeria monocytogenes* during dry-curing and drying of meat: a case study with basturma. J. Food Safety 26: 160-172.[3]
- Genigeorgis, C., Lindroth, S. 1984. The Safety of Basturma, An Armenian-type Dried Beef Product with Regard to *Salmonella*. Proceedings of the 30th European Meeting of Meat Research Workers. Bristol, UK. 217-224.

**Country Cured Ham**

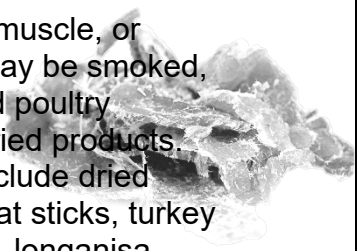
- Reynolds, A.E., Harrison, M.A., Rose-Morrow, R., Lyon, C.E. 2001. Validation of Dry Cured Ham Process for Control of Pathogens. J. Food Sci. 66:1373-1379.

**Bresaola**

- Watson, S.C., Gaydos, N.J., Egolf, S.R., Campbell, J.A. 2021. Fate of *Escherichia coli* O157:H7, *Salmonella spp.*, and *Listeria monocytogenes* During Curing and Drying of Beef Bresaola. Meat and Muscle Biology. 5(1): 14, 1-8.

[3] Study did not achieve a 5-log reduction so additional supporting documentation (i.e., additional journal article or challenge study) should be provided.

## Overview of Dried Products



RTE dried meat and poultry products can be comminuted, sliced whole muscle, or whole muscle products that may or may not be formulated with nitrite, may be smoked, are usually heated, and are air dried or oven dried. In addition, meat and poultry products that are freeze dried are also considered by FSIS to be RTE dried products. Examples of products that are dried as the primary lethality treatment include dried beef, (some) beef jerky, beef nuggets, steak tenders, kippered beef, meat sticks, turkey jerky, tasajo, pemmican, pipi kaula, **droëwors**, **biltong**, jamon (jambon), longanisa, (some) saucisson, (some) chorizo, dried soup mixes/soup bases, freeze-dried entrees, fried pork skins/rinds/cracklings/chicharrones, and lard. See examples of scientific support for products in bold in Appendix 14.

**Table 5. Overview of hazards of concern establishments should consider in the hazard analysis during lethality and stabilization and typical controls for dried products.**

| Hazard | Source | Adulterant (Yes/No) | Recommended Target | *Seasoning/Marination* | *Drying* |
|---|---|---|---|---|---|
| ***Salmonella* STEC (beef) *Lm*** | Raw meat and poultry, spices, herbs | Yes – zero tolerance | 5-log reduction for *Salmonella* and STEC<br>3-log reduction for *Lm.*<br>For more information including on **alternative lethality targets** see Appendix 3 | Effectiveness for *Salmonella*, STEC, *Lm* depends on:<br>Product formulation<br>Antimicrobial application<br>See Appendix 9 | Effectiveness for *Salmonella*, STEC, *Lm* depends on:<br>Drying room temperature<br>Drying time<br>Target water activity<br>Product characteristics<br>See Appendix 11 |
| ***S. aureus*** | Raw meat and poultry spices, herbs | Yes – depending on level | No more than 2-log growth during production; no outgrowth during storage | Effectiveness of seasoning/marination on *S. aureus* unclear | Drying is effective for *S. aureus* during storage after the water activity is reduced<br>See Appendix 5 |
| ***C. perfringens* *C. botulinum*** | Raw meat and poultry, spices, herbs | Yes – depending on level | No more than 1-log growth *C. perfringens*; no multiplication of *C. botulinum* | Effectiveness of seasoning/marination on *Clostridia* unclear | Drying is effective for *Clostridia* after the water activity is reduced<br>See FSIS' Stabilization Guideline |
| ***Trichinella spiralis* and *Toxoplasma gondii* (pork)** | Raw pork (greater risk of infection for feral or non-confinement raised swine) | Yes – zero tolerance | Eliminate larvae | Seasoning/marination not effective for *Trichinella* – see FSIS Trichinella Guideline for alternatives such as freezing | Drying not effective on its own for *Trichinella* – see FSIS Trichinella Guideline for alternatives such as curing or freezing |
| **Molds** | Any food product | Maybe depending on type | No unintentional mold growth on finished product | Seasoning/marination not effective for molds – rely on sanitation | Drying not effective for molds – rely on sanitation |

**Addressing STEC, *Salmonella,* and *Lm*:** Dried products have been associated with *Salmonella* outbreaks outside of the U.S. (see Appendix 14) and FSIS has detected *Salmonella* and *Lm* in these products (see Appendix 2). In general, literature does not support that drying alone achieves a 5-log reduction of *Salmonella* and STEC or a 3-log reduction in *Lm.* Therefore, additional interventions are often needed to produce a RTE product. Examples of interventions that may provide additional lethality include **antimicrobial interventions** such as lactic or acetic acid (*e.g.*, vinegar) or HPP. Establishments should be aware that HPP is less effective on intermediate moisture foods (*i.e.,* those foods that do not require refrigeration to control pathogens) (Balamurugan, 2019; Perez-Baltar, 2020).

**Critical Steps and Critical Operating Parameters:**

Marination/Seasoning (see Appendix 10 for more information):

- Product formulation.
- Antimicrobial application (*e.g.*, concentration, pH, coverage, contact time).

Drying (see Appendix 11 for more information):

- Drying room temperature.
- Drying time.
- Target water activity.
- Product characteristics.

***Key Point – Additional Interventions***

It is not appropriate to add up the results of two separate studies conducted for the same type of intervention (such as two acid dips) because the second time the intervention is used it will likely be less effective. This is because any bacteria that survive the first treatment are likely to be more tolerant to the second treatment.

**Available Scientific Support:** Below is a list of available scientific support for the reduction of *Salmonella*, STEC, and *Lm* in dried products. For detailed summaries of these common scientific support used for dried products see Appendix 14.

**Droëwors**

- Burnham, G.M., Hanson, D.J., Koshick, C.M., Ingham, S.C. 2008. Death of *Salmonella* Serovars, *Escherichia coli* O157:H7, *Staphylococcus aureus*, and *Listeria monocytogenes* During the Drying of Meat: a Case Study Using Biltong and Droewors. J. Food Safety. 28:198-209.[4]

**Biltong**

- Burnham, G.M., Hanson, D.J., Koshick, C.M., Ingham, S.C. 2008. Death of *Salmonella* Serovars, *Escherichia coli* O157:H7, *Staphylococcus aureus*, and *Listeria monocytogenes* During the Drying of Meat: a Case Study Using Biltong and Droewors. J. Food Safety. 28:198-209.[4]
- Karolenko, C.E., Bhusal, Ar., Nelson, J.L., Muriana, P.M. 2020. Processing of Biltong (Dried Beef) to Achieve USDA-FSIS 5-log Reduction of *Salmonella* Without a Heat Lethality Step. Microorganisms. 8(5): 791.
- Naidoo, K., Lindsay, D. 2010. Survival of *Listeria monocytogenes*, and Enterotoxin-Producing *Staphylococcus aureus* and *Staphylococcus pasteuri*, During Two Types of Biltong-manufacturing Practices. Food Control. 21:1042-1050.[4]

[4] Studies did not achieve a 5-log reduction or there were methodological issues so additional supporting documentation (*i.e.,* a journal article or challenge study) should be provided.

## Post-lethality Considerations

In addition to achieving lethality and shelf-stability using fermentation, salt-curing, and drying, it is important to ensure contamination and adulteration of products is prevented after the lethality treatment is complete. Even if the product is RTE shelf-stable, pathogens may still be able to survive on the product if it becomes contaminated during handling. RTE foods implicated in foodborne illness are commonly implicated due to post-processing contamination by bacteria such as *S. aureus* and *Lm* either by food handlers or from the environment.

To ensure that contamination and adulteration of products is prevented after the lethality treatment, establishments are required to:

- Develop and implement SSOPs (9 CFR 416.11-9 CFR 416.16).
- Maintain sanitation in the RTE area to ensure that food contact surfaces are free of contamination from *Lm* and other pathogens, such as *Salmonella*, in accordance with 9 CFR part 430.
- Support the safety of non-meat ingredients added part-way through the lethality treatment (*e.g.*, during lard application during drying of salt-cured hams) or after the final lethality step is complete (*e.g.*, coating dry or semi-dry sausages in pepper or rolling sausages in rice flour to give the appearance of an outer coating of mold). In accordance with 9 CFR 417.2(a)(1) and 417.5(a)(1):
    - Establishments must determine what potential hazards are associated with the non-meat ingredients at the step in the process when the ingredients are "received" into the food safety system.
    - Establishments must also document any controls needed to support decisions about those hazards such as **Letters of Guarantee (LOGs)**, **Certificates of Analysis (COAs)**, or other information (*e.g.*, sampling by the receiving establishment).

Because fermented, salt-cured, and dried products rely on multiple hurdles, establishments are required to identify the step in the process when lethality is complete according to their scientific support for purposes of complying with the *Listeria* Rule (9 CFR 430) which addresses control of *Lm* in the post-lethality environment. Identifying the step where lethality is achieved will ensure that:

- the establishment identifies all possible surfaces that contact the food after the post-lethality step for sampling (required under 9 CFR 430.4(b)(2)(iii)(A) and (3)(i)(A)); and
- any **post-lethality treatments** are designed and implemented appropriately.

Identifying when lethality ends and the post-lethality environment begins is particularly important since some treatments, such as drying, extended storage, or HPP, can be used as part of the multi-hurdle lethality or as a post-lethality treatment or both. For

example, if a study shows a 5-log reduction in STEC, *Salmonella*, and *Lm* occurs after 18 days of drying, then the establishment would identify the lethality treatment ends after 18 days of drying and the post-lethality environment begins when the product is in the drying room on day 19. The establishment would also identify any food contact surfaces that the product contacts after 18 days of drying (*e.g.*, racks, aprons, packaging machines, carts) in its *Lm* Control Program to address potential post-lethality *Lm* contamination. This would include any food contact surfaces the product contacts during drying that are also in contact after the 18 days of drying are complete, such as racks. The establishment may also choose to implement a validated post-lethality treatment after 18 days of drying, such as additional 60 days of storage under vacuum at 70°F to meet the requirements of Alternative 2, Choice 1 (Alt. 2a) [or Alternative 1 if the establishment can also support the product's water activity is below the growth limit of *Lm*]. In this example, drying would be part of the lethality treatment and extended storage would be a post-lethality treatment. Examples of post-lethality treatments that have been validated for fermented products include storage under vacuum at refrigerated temperatures (Faith *et al.*, 1997; Faith *et al.*, 1998a; Faith *et al.*, 1998b; Ihnot *et al.*, 1998; Ingham *et al.*, 2004) and pasteurization by submersion heating (Roering *et al.*, 1998). HPP has been validated as a post-lethality treatment for products such as salt-cured ham (Perez-Baltar *et al.*, 2020).

Further guidance on post-processing handling and sanitation for RTE products is available in the FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A) and the Guidelines to Control *Listeria monocytogenes* in Post-Lethality Exposed Ready-to-Eat Meat and Poultry Products. The *Lm* guideline also contains more information on post-lethality treatments and when fermentation and drying may be considered as antimicrobial processes.

## References


Acton, J.C., Keller, J.E. 1974. Effect of Fermented Meat pH on summer sausage properties. Journal of Milk and Food Technology. 11:570-576

Akköse A., Aktaş, N. 2014. Curing and diffusion coefficient study in pastırma, a Turkish traditional meat product. Meat Science. 96: 311-314.

Aksu, M.I., Kaya, M., Ockerman, H.W. 2005. Effect of modified atmosphere packaging, storage period, and storage temperature on the residual nitrate of sliced-pastırma, dry meat product, produced from fresh meat and frozen/thawed meat. Food Chemistry. 93: 237-242.

American Meat Institute Foundation. October 1997. Good Manufacturing Practices for Fermented Dry and Semi-Dry Sausage Products. Available at: https://meathaccp.wisc.edu/assets/Heat_Treated_Shelf_Stable/AMIF_degreehours.pdf.

Balamurugan, S. February/March 2019. High-pressure processing during drying of fermented sausages to enhance safe and stability. Food Safety Magazine. Available at: https://www.foodsafetymagazine.com/magazine-archive1/februarymarch-2019/high-pressure-processing-during-drying-of-fermented-sausages-to-enhance-safety-and-stability/.

Baccus-Taylor, G., Glass, K.A., Luchansky, J.B., Maurer, A.J.. 1993. Fate of *Listeria monocytogenes* and pediococcal starter cultures during the manufacture of chicken summer sausage. Poultry Science. 72:1772-1778.

Blankenship, L.C., 1978. Survival of a *Salmonella typhimurium* experimental contaminant during cooking of beef roasts. Applied Environ Microbiol. 35(6):1160-1165.; Borneman, D.L., Ingham, S.C., and Ane, C. 2009. Predicting growth/no-growth of *Staphylococcus aureus* on vacuum-packaged ready-to-eat meats. J. Food Prot. 72: 539-548.

Borowski, A.G., Ingham, S.C., Ingham, B.H. 2009. Validation of ground-and-formed beef jerky processes using commercial lactic acid bacteria starter cultures as pathogen surrogates. J. Food Prot. 72(6): 1234-1247.

Breidt, F, Andress, E.L., Ingham B.H. 2018. Recommendations for designing and conducting cold-fill hold challenge studies for acidified food products. Food Protect. Trends. 38(5): 322-328.

Buchanan, R.L. and Edelson, S.G. 1996. Culturing enterohemorrhagic *Escherichia coli* in the presence and absence of glucose as a simple means of evaluating the acid tolerance of stationary-phase cells. Appl. Environ Microbiol. 62: 4009-4013.

Buchanan, R.L, Stahl, H.G., Whiting, R.C. 1989. Effects and Interactions of Temperature, pH, Atmosphere, Sodium Chloride, and Sodium Nitrite on the Growth of *Listeria monocytogenes*. J. Food Prot. 52(12): 844-851.

Brul, S. and Coote, P. 1999. Preservative agents in foods: Mode of action and microbial resistance mechanisms. International Journal of Food Microbiology. 50: 1-17.

Burnham, G.M., Hanson, D.J., Koshick, C. M., Ingham, S.C. 2008. Death of *Salmonella* Serovars, *Escherichia coli* O157:H7, *Staphylococcus aureus*, and *Listeria monocytogenes* During the Drying of Meat: a Case Study Using Biltong and Droewors. Journal of Food Safety. 28:198-209. Available at: https://meathaccp.wisc.edu/validation/assets/Dry%20JFS%2028.pdf.

Cabrera-Diaz, E., Moseley, T.M., Lucia, L.M., Dickson, J.S., Castillo, A., Acuff, G.R. 2009. Fluorescent protein-marked *Escherichia coli* Biotype I strains as surrogates for enteric pathogens in validation of beef carcass interventions. J. Food Prot. 72: 295-303.

Calicioglu, M., Faith, N.G., Buege, D.R., Luchansky, J.B. 1997. Viability of *Escherichia coli* O157:H7 in fermented semi-dry low-temperature-cooked beef summer sausage. J. Food Prot. 60(1): 1158-1162. Available at: https://doi.org/10.4315/0362-028X-60.10.1158.

Calicioglu, M., Faith, N.G., Buege, D.R., Luchansky, J.B. 2001. Validation of a manufacturing process for fermented, semidry Turkish soudjouk to control *Escherichia coli* O157:H7. J. Food Prot. 64:1156-1161. Available at: https://doi.org/10.4315/0362-028X-64.8.1156.

Calicioglu, M., Faith, N.G., Buege, D.R., Luchansky, J.B. 2002. Viability of *Escherichia coli* O157:H7 during manufacturing and storage of fermented, semidry soudjouk-style sausage. J. Food Prot. 65:1541-1544. Available at: https://doi.org/10.4315/0362-028X-65.10.154.

Canadian Food Inspection Agency (CFIA). 2020. Preventative control recommendations for manufacturing fermented and dried meat products. Available at: https://inspection.canada.ca/preventive-controls/meat/fermented-and-dried/eng/1522951036924/1522951037158#control.

Center for Disease Control (CDC). 1971a. Staphylococcal gastroenteritis associated with salami – United States. Morbidity and Mortality. 20(28): 253, 258.

Center for Disease Control (CDC). 1971b. Gastroenteritis associated with Genoa salami – United States. Morbidity and Mortality. 20(29): 261, 266.

Center for Disease Control (CDC). 1975. Staphylococcal food poisoning associated with Italian dry salami – California. 24(44): 374, 379.

Chikthimmah, N., Knabel, S.J. 2001. Survival of *Escherichia coli* O157:H7, *Salmonella*, and *Listeria monocytogenes* in and on vacuum packaged lebanon bologna stored at 3.6 and 13.0C. J. Food Prot. 64(7): 958-963.

Christiansen, L.N., Tompkin, R.B., Shaparis, A.B., Johnston, R.W., Kautter, D.A. 1975. Effect of sodium nitrite and nitrate on *Clostridium botulinum* growth and toxin production in a summer style sausage. Journal of Food Science. 488-490.

Deibel Laboratories/CHR. Hansen. 2017. Fate of *Salmonella Spp*., *Escherichia coli* O157:H7, *Listeria monocytogenes*, and *Staphylococcus aureus* Inoculated Into a Non-Heated and Dried Salami Product. Available from CHR. Hansen Inc. upon request. https://www.chr-hansen.com/en/contact-us

DeSouza, J.D., Ahmed, R., Strange, P., Barbut, S., Balamurugan, S. 2018. Effect of caliber size and fat level on the inactivation of *E. coli* O157:H7 in dry fermented sausages. Internal Journal of Food Microbiology. 266: 167-172.

Dierschke, S.E., Ingham, B.H., Ingham, S.C. 2010a. Use of Lactic Acid Bacteria as Pathogen Surrogates to validate commercial whole-muscle beef jerky process lethality against *Escherichia coli* O157:H7, *Salmonella* spp., *Listeria monocytogenes*, and *Staphylococcus aureus*. = Poster presented at the annual meeting of the Institute of Food Technologists. Chicago, IL. July 2010.

Dierschke, S.E., Ingham, S.C., Ingham, B.H. 2010b. Destruction of *E. coli* O157:H7, *Salmonella*, *Listeria monocytogenes*, and *Staphylococcus aureus* achieved during manufacture of whole-muscle beef jerky in home-style dehydrators. J. Food Prot. 73(11): 2034-2042.

Draper, A.D.K., Morton, C.N., Heath, J.N.I., Lim, J.A., Schiek, A.I., Davis, S., Krause, V.L., Markey, P.G. 2017. An outbreak of salmonellosis associated with duck prosciutto at a northern territory restaurant. Communicable Diseases Intelligence Quarterly Report. 41(1): E16-E20.

Eblen, D.R., Annous, B.A., Sapers, G.M. 2005. Studies to select appropriate nonpathogenic surrogate *Escherichia coli* strains for potential use in place of *Escherichia coli* O157:H7 and *Salmonella* in pilot plant studies. J. Food Prot. 68(2): 282-291.

Faith, N.G., Parniere, N., Larson, T., Lorang, T.D., Luchansky, J.B. 1997. Viability of *Escherichia coli* O157:H7 in pepperoni during the manufacture of sticks and subsequent storage of slices at 21, 4 and -20°C under air, vacuum and $CO_2$. International Journal of Food Microbiology. 37:47-54. Available at: https://www.fsis.usda.gov/sites/default/files/media_file/2021-09/Faith-1997.pdf.

Faith, N.G., Parniere, N., Larson, T., Lorang, T.D., Kaspar, C.W., Luchansky, J.B. 1998a. Viability of *Escherichia coli* O157:H7 in salami following conditioning of batter, fermentation and drying of sticks, and storage of slices. J. Food Prot. 61:377-382. Available at: https://doi.org/10.4315/0362-028X-61.4.377.

Faith, N.G., Wierzba, R., Ihnot, A.M., Roering, A.M., Lorang, T.D., Kaspar, C.W., Luchansky, J.B. 1998b. Survival of *Escherichia coli* O157:H7 in full- and reduced-fat pepperoni after manufacture of sticks, storage of slices at 4°C or 21°C under air and vacuum, and baking of slices on frozen pizza at 135, 191, and 246°C. J. Food Prot. 61:383-389. Available at: https://doi.org/10.4315/0362-028X-61.4.383.

Farber, J.M., Tittinger, F., Gour, L. 1988. Surveillance of raw-fermented (dry-cured) sausages for the presence of *Listeria spp*. Canadian Institute of Food Science and Technology. 21: 430-434.

Food and Agriculture Organization. 1990. Manual on Simple Methods of Meat Preservation. Available at: http://www.fao.org/docrep/003/x6932e/X6932E00.htm#TOC

Gamble, H.R., Hill, D. 2012. PORK Safety – Preharvest/Postharvest,Toxplasma Fact Sheet. National Pork Board.

Genigeorgis, C., Lindroth, S. 1984. The Safety of Basturma, An Armenian-type Dried Beef Product with Regard to *Salmonella*. Proceedings of the 30th European Meeting of Meat Research Workers. Bristol, UK. Pages 217-224.

Getty, K.J.K, Phebus, R.K, Marsden, J.L., Schwenke, J.R., Kastner, C.L. 1999. Control of Escherichia coli O157:H7 in Large (115 mm) and Intermediate (90 mm) Diameter Lebanon-style Bologna. Journal of Food Science. 64(6): 1100-1107. Available at: https://onlinelibrary.wiley.com/doi/epdf/10.1111/j.1365-2621.1999.tb12290.x.

Gök, V., Obuz, E., and Akkaya, L. 2008. Effects of packaging method and storage time on the chemical, microbiological, and sensory properties of Turkish pastirma – A dry cured beef product. Meat Science. 80: 335-344.

Goodfellow, S.J., Brown, W.L. 1978. Fate of *Salmonella* inoculated into beef for cooking. J. Food Prot. 41(8):598-605

Gunvig, A., Borggaard, C., Hansen, F., Hansen, T.B., Aabo, S. 2016. ConFerm - A tool to predict the reduction of pathogens during the production of fermented and matured sausages. Food Control. 67: 9-17.

Gunvig, A., Andresen, M.S., Borggaard, C. 2017. Staphtox predictor – a dynamic mathematical model for predicting the formation of staphylococcus enterotoxin during processing of meat. Poster presented at the International Committee on Predictive Modelling in Food (ICPMF) Annual Meeting. Cordoba, Spain. September, 2017.

Hew, C., Hajmeer, H.N., Farver, T.B., Riemann, H.P., Glover, J.M., Cliver, D.O. 2006. Pathogen survival in chorizos: ecological factors. J. Food Prot. 69(5): 1087-1095.

Hill, C., O'Driscoll, B., Booth, I. 1995. Acid adaptation and food poisoning microorganisms. Int. J. Food Microbiol. 28: 245-254.

Hill, D.E., Luchansky, J., Porto-Fett, A., Gamble, H.R., Fournet, V.M., Hawkins-Cooper, D.S., Gajadhar, A.A., Holley, R., Juneja, V.K., Dubey, J.P. Curing conditions to inactivate *Trichinella spiralis* muscle larvae in ready-to-eat pork sausage. Food and Waterborne Parasitology. 12: e00029.

Hinkens, J.C., Faith, N.G., Lorang, T.D., Bailey, P., Buege, D., Kaspar, C.W., Luchansky, J.B. 1996. Validation of pepperoni processes for control of *Escherichia coli* O157:H7. J. Food Prot. 59: 1260-1266. Available at: https://doi.org/10.4315/0362-028X-59.12.1260.

Honikel, K.O. 2010. Principles of Curing. In Toldra, F. Handbook of Meat Processing. Blackwell Publishing. 584 pages.

Hussein, M.H., Burroughs, S., Emch, A.W., Waite-Cusic, J. 2022. Enhancing the Reduction of *Salmonella* and *Listeria monocytogenes* During Traditional Salami Processing by Adding a Finishing Phase. Food Control. 131: 108432.

IEH. Evaluation of Process Parameters Used During the Fermentation and Drying of Italian-Style Salami. Unpublished report.

Ihnot, A. M., Roering, A.M., Wierzba, R.K., Faith, N.G., Luchansky, J.B. 1998. Behavior of *Salmonella typhimurium* DT104 during the manufacture and storage of pepperoni. International Journal of Food Microbiology. 40:117-121. Available at: https://www.fsis.usda.gov/sites/default/files/media_file/2021-09/Ihnot-1998.pdf.

Incze, K. 2010. Mold Ripened Sausages. In Toldra, F. Handbook of Meat Processing. Blackwell Publishing. 584 pages.

Ingham, S.C., Buege, D.R., Dropp, B.K., Losinski, J.A. 2004. Survival of *Listeria monocytogenes* during storage of ready-to-eat meat products processed by drying, fermentation, and/or smoking. J. Food Prot. (67)12: 2698-2702. Available at: https://meathaccp.wisc.edu/validation/assets/LM%20JFP%2067.pdf.

Ingham, S. C., Engel, R.A., Fanslau, M.A., Schoeller, E.L., Searls, G., Guege, D.R., Zhu, J. 2005. Fate of *Staphylococcus aureus* on vacuum-packaged ready-to-eat meat products stored at 21°C. J. Food Prot. 68(9):1911-1915.

IUFoST. 2012. A basic guide to drying fruits and vegetables. http://iufost.org/iufostftp/Guide%20to%20Drying-Full.pdf.

FSIS. 2022. *Salmonella* Outbreaks Linked to Italian-Style Meats: Outbreak Investigation After Action Review. Available at: https://www.fsis.usda.gov/sites/default/files/media_file/2022-04/FSIS-After-Action-Review-2021-09_2022-01.pdf.

Jay, J.M. 2000. Modern Food Microbiology. 6th ed. Gaithersburg, Md.: Aspen Publishing, Inc. P 445-446. 635 p.

Johnston, M.A., Pivinick, H., Samson, J.M. 1969. Inhibition of *Clostridium botulinum* by sodium nitrite in a bacteriological medium and in meat. Canadian Institute of Food Science and Technology Journal. 2(1): 52-55.

Juneja, V.K., Sofos, J.N. 2010. Pathogens and toxins in foods. ASM Press, Washington, D.C.

Karolenko, C.E., Bhusal, Ar., Nelson, J.L., Muriana, P.M. 2020. Processing of Biltong (Dried Beef) to Achieve USDA-FSIS 5-log Reduction of *Salmonella* Without a Heat Lethality Step. Microorganisms. 8(5): 791. Available at: https://www.mdpi.com/2076-2607/8/5/791/htm.

Karolenko, C.E., Wilkinson, J., Muriana, P.M. 2022. Validation of Biltong (Dried Beef) Process Lethality Using Non-Pathogenic Surrogate Organisms Associated with Beef. Poster presented at the annual meeting of the International Association for Food Protection. Chicago, IL. July 2010.

Kilic, B. 2009. Current trends in traditional Turkish meat products and cuisine. LWT Food Science and Technology. 42: 1581–1589.

Kneeling, C., Niebuhr, S.E., Acuff, G.R., Dickson, J.S. 2009. Evaluation of *Escherichia coli* Biotype I as a Surrogate for *Escherichia coli* O157:H7 for Cooking, Fermentation, Freezing, and Refrigerated Storage in Meat Processes. J. Food Prot. 72: 728-732.

Leistner, L. 2000. Basic aspects of food preservation by hurdle technology. International Journal of Food Microbiology, 55: 181–186.

Leyer, G.J., Wang, L.L., Johnson, E.A. 1995. Acid adaptation of *Escherichia coli* O157:H7 increases survival in acidic foods. Applied Environmental Microbiology. 61: 3752-3755.

Luchansky, J. B., Phebus, R.K., Thippareddi, H., Call, J.E. 2008. Translocation of surface-inoculated *Escherichia coli* O157:H7 into beef subprimals following blade tenderization. J. Food Prot. 71:2190-2197.

Luchansky, J. B., Glass, K.A., Harsono, K.D., Degnan, A.J., Faith, N.G., Cauvin, B., Baccus-Taylor, G., Arihara, K., Bater, B., Maurer, A.J., Cassens, R.J. 1992. Genomic analysis of *Pediococcus* starter cultures used to control *Listeria monocytogenes* in turkey summer sausage. Applied and Environmental Microbiology. 58:3053-3059.

Ma, L., Kornacki, J.L., Lin, C.M., Doyle, M.P. 2007. Development of thermal surrogate microorganisms in ground beef for in-plant critical control point validation studies. J. Food Prot. 70: 952-957.

Marshall, K. M., Niebuhr, S.E., Acuff, G.R., Lucia, L.M., Dickson, J.S. 2005. Identification of *Escherichia coli* O157:H7 meat processing indicators for fresh meat through comparison of the effects of selected antimicrobial interventions. J. Food Prot. 68: 2580-2586.

Mazuet, C., Sauteraue, J., Legeay, C., Bouchler, C., Bouvet, P., Popoff, M.R. 2015. An atypical outbreak of food-borne botulism due to *Clostridium botulinum* Types B and E from ham. Journal of Clinical Microbiology. 53(2): 722-726.

McKinney, S., Cutter, C., Campbell, J. 2019. Pathogen Reductions during Traditional Fermentation and Drying of Pork Salamis. Food Protection Trends. 39(1): 18-27.

Merialdi, G., Ramini, M., Parolari, G., Barbuti, S., Frustoli, M.A., Taddei, R., Pongolini, S., Ardigo, P., Cozzolino, P. 2016. Study on potential *Clostridium botulinum* growth and toxin production in parma ham. Italian Journal of Food Safety. 5: 5564.

Michet, C.J. 2015. Validation of a HACCP Program for the production of fermented dry cured pork products. Retrieved from the University of Minnesota Digital Conservancy, http://hdl.handle.net/11299/177030.

Mindlin, M. J., Lang, N., Maguire, H., Walsh, B., Verlander, N.Q., Lane, C., Taylor, C. Bishop, L.A., and Crook P.D. 2013. Outbreak investigation and case-control study: Penta-resistant *Salmonella* Typhimurium DT104 Associated with Biltong in London in 2008. Epidemiol. Infect. 141:1920-1927.

Mutz, Y.S., Rosario, D.K.A., Paschoalin, V.M.F., Conte-Junior, C.A. 2019. *Salmonella enterica*: A hidden risk for dry-cured meat consumption? Critical Reviews in Food Science and Nutrition, DOI: 10.1080/10408398.2018.1555132.

Naidoo, K, Lindsay, D. 2010. Survival of *Listeria monocytogenes*, and Enterotoxin-Producing *Staphylococcus aureus* and *Staphylococcus pasteuri*, during two types of Biltong-manufacturing practices. Food Control. 21:1042-1050.

Neser, A.T., Louw., A., Klein, S., Sacks, I. 1957. Fatal *Salmonella* food-poisoning from infected biltong. South African Medical Journal. 31(8): 172-174.

Nickelson, R., Luchansky, J.B., Kaspar, C.W., Johnson, E. 1996. Update on dry fermented sausage *Escherichia coli* O157:H7 validation research. An executive summary prepared by The Blue Ribbon Task Force of the National Cattlemen's Beef Association. Research Report No. 11-316. Available at: https://meatsci.osu.edu/sites/meatsci/files/imce/1996_dry_fermented_sausage.pdf.

Niebuhr, S.E., Laury, A., Acuff, G.R., Dickson, J.S. 2008. Evaluation of nonpathogenic surrogate bacteria as process validation indicators for *Salmonella enterica* for selected antimicrobial treatments, cold storage, and fermentation in meat. J. Food Prot. 71(4): 714-718.

Peck, M., Devlieghere, F., Membre, J. 2015. *Clostridium botulinum*: a recurrent emerging foodborne pathogen. Symposium conducted at the International Association of Food Protection: Portland, Oregon. July 26-29, 2015.

Perez-Baltar, A., Serrano, A., Montiel, R., Medina, M. 2020. *Listeria monocytogenes* inactivation in deboned dry-cured hams by high pressure processing. Meat Science. 160: 1-5.

Porto-Fett, A.C.S, Hwang, C.A., Call, J.E., Juneja, V.K., Ingham, S.C., Ingham, B.H., Luchansky, J.B. 2008. Viability of multi-strain mixtures of *Listeria monocytogenes*, *Salmonella typhimurium*, or *Escherichia coli* O157:H7 inoculated into the batter or onto the surface of a soudjouk-style semi-dry sausage. Food Microbiology. 25: 793-801. Available at: https://meathaccp.wisc.edu/validation/assets/Dry%20Food%20Micro%2025.pdf.

Porto-Fett, A.C.S., Call, J.E., Shoyer, B.E., Lücke, D.E., Pshebniski, C., Cocoma, G.J., and Luchansky, J.B. 2010. Evaluation of fermentation, drying, and/or high pressure processing on viability of *Listeria monocytogenes*, *Escherichia coli* O157:H7, *Salmonella spp.*, and *Trichinella spiralis* in raw pork and Genoa salami. International

Journal of Food Microbiology. 140: 61-75. Available at: https://www.fsis.usda.gov/sites/default/files/media_file/2021-09/Porto-Fett-2010-high-pressure.pdf.

Quintavalla, S. 2010. Plant Cleaning and Sanitation. In Toldra, F. Handbook of Meat Processing. Blackwell Publishing. 584 pages.

Reimann, H., Lee, W.H., Genigeorgis, C. 1972. Control of *Clostridium botulinum* and *Staphylococcus aureus* in semi-preserved meat products. Journal of Milk and Food Technology. 35(9): 514-523.

Reynolds, A.E., Harrison, M.A., Rose-Morrow, R., Lyon, C.E. 2001. Validation of dry cured ham process for control of pathogens. J. Food Sci. 66:1373-1379. Abstract available at: https://onlinelibrary.wiley.com/doi/abs/10.1111/j.1365-2621.2001.tb15217.x.

Rivera-Reyes, M., Campbell, J.A., Cutter, C.N. 2017. Pathogen reductions associated with traditional processing of Landjäger. Food Control. 73: 768-774.

Roering, A. M., Wierzba, R.K., Ihnot, A.M., Luchansky, J.B.. 1998. Pasteurization of vacuum-sealed packages of summer sausage inoculated with *Listeria monocytogenes*. J. Food Safety 18:49-56.

Ruhlman, M., Polcyn, B. 2012. Salumi: The craft of Italian dry curing. W.W. Norton Company, NY.

Saricoban, C., Karakaya, M., Caner, C. 2006. Properties of Turkish-style sucuk made with different combinations of beef and hen meat. Journal of Muscle Foods: 17(1): 1-8.

Sindelar, J.J. 2012. What's the deal with nitrates and nitrites used in meat products? University of Wisconsin Meat Science and Muscle Biology Lab.

Smith, J.L., Palumbo, S.A. 1978. Injury to *Staphylococcus aureus* during sausage fermentation. Applied and Environmental Microbiology. 36: 857-860.

Smith, J.L., Palumbo, S.A. 1983. Use of starter cultures in meats. J. Food Prot. 46: 997-1006.

Tatini, S.R., Lee, R.Y., McCall, W.A., Hill, W.M. 1976. Growth of *Staphylococcus aureus* and production of enterotoxins in pepperoni. J Food Sci. 41(2):223-225.

Tilkens, B.L., King, A.M., Glass, K.A., Sindelar, J.J. 2015. Validating the inhibition of *Staphylococcus aureus* in shelf-stable, ready-to-eat snack sausages with varying combinations of pH and water activity. J. Food Prot. 78(6): 1215-1220.

Theron, M.M. Lues, J.F.R. 2007. Organic acids and meat preservation: A review. Food Reviews International. 23(2): 141-158.

Toldra, F. 2002. Dry-cured meat products. Food & Nutrition Press. Trumbull, CT. 06611.

Tompkin, R.B. 1976. *C. botulinum*. In: Defigueredo MP and Splittstoesser DF. Food Microbiology: Public Health and Spoilage Aspects. West Point, Conn. AVI Publishing Co. 156-169.

Tornberg, E. 2005. Effects of heat on meat proteins – Implications on structure and quality of meat products. Meat Science. 70: 493-508.

Ulbrich, C.J., Lucia. L.M., Arnold, A.N., Taylor, T.M., Savell, J.W., and Gehring, K.B. 2015. Reduction of surrogates for *Escherichia coli* O157:H7 and *Salmonella* during the production of nonintact beef products by chemical antimicrobial interventions. J. Food Prot. 78(5): 881-887.

Vignolo, G., Fontana, C., and S. Fadda. 2010. Semidry and Dry Fermented Sausages. In Toldra, F. Handbook of Meat Processing. Blackwell Publishing. 584 pages.

Watson, S.C., Gaydos, N.J., Egolf, S.R., Campbell, J.A. 2021. Fate of *Escherichia coli* O157:H7, *Salmonella spp.*, and *Listeria monocytogenes* During Curing and Drying of Beef Bresaola. Meat and Muscle Biology. 5(1): 14, 1-8. Available at: https://www.iastatedigitalpress.com/mmb/article/id/11621/.

Whitworth, J. 2019, February 15. Denmark *E. coli* outbreak declared over; *Salmonella* investigation continues. Food Safety News. Retrieved from: https://www.foodsafetynews.com/2019/02/denmark-e-coli-outbreak-declared-over-salmonella-investigation-continues/.

Whitworth, J. 2020, September 4. *Salmonella* outbreak in France tied to sausage from Spain. Food Safety News. Retrieved from: https://www.foodsafetynews.com/2020/09/salmonella-outbreak-in-france-tied-to-sausage-from-spain/.

York, J. 2020, November 27. France recalls dried meats after *Salmonella* scare. The Connexion French News and Views. Retrieved from: https://www.connexionfrance.com/French-news/France-recalls-dried-meats-from-supermarkets-after-salmonella-scare.

## Appendix 1: Labeling Considerations for Not Ready-to-Eat (NRTE) fermented, salt-cured, and dried products.

The following are additional labeling considerations for NRTE fermented, salt-cured, and dried meat and poultry products.

Many of the products covered in this guideline can be classified by establishments as RTE or NRTE and as shelf-stable or not (i.e., it needs to be refrigerated or frozen by the consumer throughout storage or once the package is opened).
as described below:

- Fermented
    - Dry and semi-dry fermented sausages – intended use may be RTE or NRTE except for pepperoni and salami which typically have an intended use of RTE.

- Salt-cured
    - Basturma – intended use may be RTE or NRTE.
    - Country cured ham – intended use may be RTE or NRTE.
    - Bresaola – intended use is typically RTE.

- Dried
    - Biltong – intended use is typically RTE.
    - Droëwors – intended use is typically RTE.

For those products where the intended use may be NRTE, the product must bear safe handling instructions (SHI) per 9 CFR 317.2(k)(1) or 9 CFR 381.125. Additional labeling features are required for those products described in this guideline such as **dry and semi-dry fermented sausages, basturma, and country cured ham** that may have the appearance of an RTE product (*e.g.*, as a result of the fermentation, salt-curing, or drying steps) but that are classified as NRTE by the establishment. Because these products require cooking by the consumer for safety, FSIS requires a prominent statement on the principal display panel, which may include statements such as, "Uncooked, Ready to cook, Cook before eating, Cook and serve" or "Needs to be fully cooked."

To help ensure that NRTE products that appear RTE are cooked, FSIS also recommends that validated cooking instructions be provided on the label. The Guidelines to Control *Listeria monocytogenes* in Post-Lethality Exposed Ready-to-Eat Meat and Poultry Products contains further information on other labeling features that should be included on NRTE products that appear RTE. Establishments should be aware that many fermented, salt-cured, and dried products have product characteristics such as intermediate and low water activity that result in the typical consumer cooking instructions that are provided on the label being insufficient if the product is NRTE. For example, FSIS recommends that consumers cook raw beef sausages to 160°F. This recommendation is based on research demonstrating that achieving this temperature would result in at least a 6.5-log reduction in *Salmonella* as shown in FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A) (Blankenship, 1978; Goodfellow and Brown, 1978).

**NOTE:** It would not be appropriate to recommend a NRTE salt-cured and dried product such as basturma be cooked to 160°F using FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A) unless the intended use is to be cooked under moist conditions to rehydrate the surface.

Once a product is salt-cured or dried, any *Salmonella* remaining in the product would have increased thermal tolerance from surviving the drying process and additional heat would be needed to destroy remaining bacteria. Establishments that use consumer cooking in conjunction with its purchasing specifications and its own preventive measures employed during further processing to support decisions related to hazards in NRTE fermented, salt-cured, and dried products, need to have on-file documentation supporting their decisions (9 CFR 417.5(a)(1)). Such documentation may include documents describing the customary preparation practices for the safe consumption of the product and the basis for the establishment's determination that these practices constitute customary preparation. Such support could include a challenge study validating the cooking method recommended results in a safe product (*e.g.*, a 5-log reduction in *Salmonella*) or a science-based justification for why the cooking method recommended results in a safe product (*e.g.*, instructions that include a step where the product is immersed in water such as those typically used for country cured hams would rehydrate the product). For more information on conducting challenge studies see Appendix 15: Designing Challenge Studies for Fermented, Salt-Cured, and Dried Products.

If an establishment identifies the intended use as NRTE for products such as pepperoni, salami, bresaola, biltong, and droëwors where the intended use is typically RTE, the establishment must have on-file documentation supporting their decisions (9 CFR 417.5(a)(1)). This support must address how the establishment can ensure the consumer will properly cook the product (9 CFR 417.5(a)(1)), particularly if there is evidence such as marketing materials or recipes commonly indicating the product is RTE.

## Appendix 2: Biological Hazards of Public Health Concern for RTE Shelf-Stable Fermented, Salt-Cured, and Dried Products

The following are further considerations regarding hazards of public health concern for RTE shelf-stable fermented, salt-cured, and dried products that can help inform an establishment's hazard analysis decision-making.

### *Salmonella*, STEC, and *Lm*

Meat and poultry products may become contaminated with *Salmonella*, STEC (in beef), and *Lm*, from cross-contamination during the slaughter/dressing process or in the processing environment when insanitary conditions are present. In addition, spices and herbs can also be contaminated with *Salmonella*, STEC, and *Lm.* These pathogens can survive typical fermentation, salt-curing and drying treatments. **FSIS has detected *Salmonella* and *Lm* in fermented and dried products.**

- Between 2010-2018, the average percent positive for *Salmonella* in RTE fermented or dried products was 0.09% (12/12,684) compared to 0.05% for other RTE products (47/99,038).
- Between 2010-2018, the average percent positive for *Lm* in RTE fermented or dried products was 0.23% (31/13,609) compared to 0.32% for other RTE products (333/106,827).

A few outbreaks associated with *Salmonella* and *E. coli* O157:H7 have occurred in the U.S. that were associated with fermented and salt-cured meats as shown in Table 6.

**Table 6. *Salmonella* and *E. coli* O157:H7 Foodborne Illness Outbreak History in Fermented and Salt-Cured, and Dried Ready-to-Eat Meat Products Produced in the U.S.**

| Products Affected | Year | Disease-Causing Organism | Case-Patients; States | Recall (Yes/No) | Process Type | Suspected Root Cause |
|---|---|---|---|---|---|---|
| **Salami sticks** | 2021 | *Salmonella* I 4,[5],12:i:- | 34; 10 states | Yes | Fermented and dried | Under-processing |
| **Italian-style Meats** | 2021 | *Salmonella* Infantis and Tyhphimurium | 40; 17 states | Yes | Fermented and dried | Under-processing |
| **Lebanon Bologna** | 2011 | *E. coli* O157:H7 | 14; 5 states | Yes | Fermented | Under-processing |
| **Prosciutto, Capocollo, calabrese, sopressata** | 2010 | *Salmonella* Montevideo | 272; 45 states | Yes | Salt-cured and Fermented and dried | Contaminated red and black pepper |
| **Lebanon Bologna** | 1995 | *Salmonella* Typhimurium | 26; 1 state | Yes | Fermented | Under-processing |
| **Salami** | 1994 | *E. coli* O157:H7 | 23; 2 states | Yes | Fermented and dried | Under-processing |
| **Basturma** | 1982 | *Salmonella* | Unknown; 1 state | Unknown | Salt-cured and dried | Unknown |

Several outbreaks have also been associated with fermented, salt-cured, and dried meat products outside of the U.S., with five outbreaks occurring in Europe between 2018 and 2020:

- In November 2020, a *Salmonella* outbreak in France was linked to a dry sausage produced in France (York, 2020).
- In September 2020, a *Salmonella* outbreak in France was linked to fuet sausage produced in Spain (Whitworth, 2020).
- In July 2019, a *Salmonella* outbreak in France was linked to coppa produced in Italy.
- In November 2018, a Shiga toxin-producing *Escherichia coli* (STEC) outbreak (O26:H11) in Denmark was attributed to a beef salami product (Whitworth, 2019).
- In October 2018, an outbreak of *Salmonella* Typhimurium in Denmark was potentially attributed to a spiced pork sausage.

Other outbreaks of note outside the U.S. include a 2015 *Salmonella* outbreak in Australia associated with duck prosciutto (Draper *et. al*, 2017) as well as a *Salmonella* outbreak associated with biltong consumption in London in 2008 (Mindlin, 2013).

**The presence of *Salmonella, Lm*, or *E. coli* O157:H7 in fermented and dried meat products may indicate under processing/insufficient lethality due to lack of a cooking step** (Farber *et al.*, 1988)**.** Although *Lm* contamination typically indicates post-lethality contamination, its presence in fermented, salt-cured, and dried meat and poultry products may also indicate under processing/insufficient lethality. This is because *Lm* can be present on raw meat and poultry products[5] and other ingredients and survive fermentation and drying because *Lm* is very tolerant to those types of lethality treatments. For more information on *Lm* prevalence in raw meat and poultry products, see FSIS' Baseline Data Reports.

**NOTE:** FSIS tested dry and semi-dry fermented sausages for *E. coli* O157:H7 prevalence between 1994 and 2011. During this time, FSIS did not have a positive *E. coli* O157:H7 test result from over 10,000 samples of these products. Therefore, FSIS discontinued testing. While FSIS is aware there can be problems associated with STEC in these products, such as the March 2011 Lebanon bologna *E. coli* O157:H7 outbreak, the Agency determined samples are not collected at a frequency high enough to detect such processing issues that tend to occur intermittently and at a low frequency. These problems are most often found during FSIS food safety assessments (FSAs) or during other in-depth establishment reviews.

### *Staphylococcus aureus*

*Staphylococcus aureus* (*S. aureus*) can contaminate raw meat and poultry from the animal hide, skin, or tissue during slaughter. After slaughter and after cooking, RTE meat or poultry products can be contaminated from handling by individuals carrying the

[5] FSIS baseline testing has identified *Lm* on all raw meat and poultry source materials at levels ranging from 4.1% on steer and heifer carcasses to 7.4% on market hogs and as high as 41.1% in raw ground chicken.

organism. *S. aureus* is of concern during storage of shelf-stable products because it is salt-tolerant and can grow at a lower water activity than other bacterial pathogens.

*S. aureus* is a concern during fermentation, salt-curing, and drying because the addition of the ingredients (salt, sodium nitrite, sodium nitrate) creates **microbial inversion** when the environment favors Gram-positive bacteria such as *S. aureus* as opposed to Gram-negative bacteria such as STEC and *Salmonella* (which grow better on raw product without ingredients added).

- For **fermented products**, the **degree-hours** concept is typically used to ensure that growth of *S. aureus* is limited during fermentation when the temperature is over 60 °F (15.6°C) (the critical temperature at which staphylococcal growth begins).
    - Degrees are measured as the excess over 60°F (15.6°C). Degree-hours are the product of time at a particular temperature and the "degrees." Degree-hours are calculated for each temperature used during fermentation. The limitation of the number of degree-hours depends upon the highest temperature in the fermentation process prior to the time that a pH of 5.3 or less is attained.
    - Good Manufacturing Practices for Fermented Dry and Semi-dry Sausage Products describes the degree-hours concept in detail.
    - The degree-hour guidance does not identify the maximum amount of growth of *S. aureus* expected when degree-hours are met; however, FSIS considers meeting degree-hours to limit growth to safe levels (*i.e.*, 2-logs or less) (Smith and Palumbo, 1978).
- For **salt-cured products**, the salt-cure is allowed to equilibrate so that it penetrates throughout the raw source materials so that *S. aureus* growth is prevented during drying when temperatures are elevated.
    - Generally, a 10% **brine concentration** following equalization will prohibit *S. aureus* enterotoxin production when the temperature is increased during drying (Reiman *et al.*, 1972).
    - *S. aureus* may also contaminate products post-lethality during handling and is also of concern during storage of shelf-stable products because it can grow at a lower water activity than other bacterial pathogens.

## KEY DEFINITIONS

**Degree-hours** is the amount of time in hours above 60°F (the critical temperature at which staphylococcal growth effectively begins) an establishment's fermentation process can take at a specific temperature to reduce the pH to 5.3 or below in order to control *S. aureus* growth.

Between 1994 and December 2002, FSIS tested 3,105 RTE products for the presence of staphylococcal enterotoxins (< 1 nanogram (ng) staphylococcal enterotoxins toxin per

gram or ml of sample). FSIS stopped testing RTE products for staphylococcal enterotoxins in January 2003 because FSIS did not find any positive samples. These negative findings are likely due in large part to the widespread use of commercial starter cultures and addition of fermentable sugars as well as education of producers by starter culture suppliers and trade associations in best practices for production of fermented meats (Smith and Palumbo, 1983), practices developed in response to outbreaks in the early 1970's (CDC, 1971a; CDC 1971b; CDC, 1975). For this reason, it is important that establishments continue to ensure processes are sufficient to limit *S. aureus* growth and enterotoxin production (*e.g.*, by following Good Manufacturing Practices for Fermented Dry and Semi-dry Sausage Products). For more information on FSIS' method for testing RTE products for the presence of staphylococcal enterotoxins see the Microbiology Laboratory Guidebook Method 39.03 available at: https://www.fsis.usda.gov/news-events/publications/microbiology-laboratory-guidebook.

### *Clostridium perfringens* and *Clostridium botulinum*

Meat and poultry products may become contaminated with *C. perfringens and C. botulinum* during the slaughter and dressing process as a result of cross-contamination in the processing environment when insanitary conditions are present. In addition, spices and herbs can contribute to the spore counts in raw formulated heat-treated meat and poultry products. For example, in one survey, *C. perfringens* spores were isolated from 43 of 54 (80%) different spices and herbs (Juneja and Sofos, 2010).

There have been several outbreaks in Europe associated with non-proteolytic *C. botulinum* and home-prepared (salted) ham (Mazuet *et al.*, 2015; Peck *et al.*, 2015). However, under commercial processing conditions, *s*pores of *Clostridia* will generally not germinate and grow out during production of dried meats because the microbiological environment of such products does not allow for the outgrowth of spores. Specifically, the acidity (reduced pH) and dryness (reduced water activity) of dried meats create conditions that prevent germination. FSIS' Stabilization Guideline for Meat and Poultry Products (Revised Appendix B) contains recommended pH and water activity values that will prevent the growth of *C. perfringens* and *C. botulinum*. Other controls include salt concentration, presence of lactic acid bacteria, and use of nitrate and nitrite. Use of sodium nitrite (50 parts per million (ppm)) or water activity of 0.92 or below are sufficient to ensure *C. botulinum* will not grow in these fermented, salt-cured, and dried products (Reynolds *et al.*, 2001; Johnston *et al.*, 1969; and Tompkin, 1976). The resting or equalization phase is a critical step for inhibiting *C. botulinum* growth in dry-cured hams (Merialdi *et al.*, 2016). In fermented products such as summer sausage, the use of a starter culture, dextrose, and nitrite were needed to inhibit *C. botulinum* growth (Christiansen *et al.*, 1975). The addition of at least 100 ppm nitrite/nitrate and at least 2.5% salt can also be used to control *C. botulinum* growth in fermented products during the fermentation and drying steps (CFIA, 2020).

### *Trichinella and Toxoplasma gondii*

*Trichinella spiralis* (*T. spiralis*) is a parasite that infects both humans and animals. Establishments producing RTE fermented, salt-cured, and dried pork products should be aware this is a hazard to consider as part of the hazard analysis, particularly if using pork from source materials derived from feral or non-confinement raised pigs. This guideline does not go into detail regarding how to address *T. spiralis* (or other potential

parasitic hazards such as *Toxoplasma gondii*) because control measures are addressed in the FSIS Guideline for the Prevention and Control of Trichinella and Other Parasitic Hazards in Pork and Products Containing Pork. FSIS' *Trichinella* Guideline contains one reference (Gamble and Hill, 2012) that addresses the effectiveness of curing for the destruction of *T. gondii* in pork products. *T. gondii* in dry-cured pork sausage may also be addressed with other scientific support (Hill *et al.*, 2018). Because there are no published studies comparing the lethality rate of *Salmonella* to the destruction of *Trichinella* in dried, salt-cured, or fermented products, it is not appropriate to apply scientific support conducted with *Salmonella* to support decisions related to *Trichinella* and it is also not appropriate to apply scientific support conducted with *Trichinella* to support decisions related to *Salmonella*. Relying on a minimum number of drying days for *Trichinella* elimination without validating the number of drying days needed for *Salmonella* reduction was a contributing factor in two *Salmonella* outbreaks associated with Italian-style fermented meats (FSIS, 2022).

### What About Mold?

Molds may grow on products like country cured ham (also known as country ham) during the curing process and drying process because the associated high salt, low temperatures, and environmental conditions do not inhibit these organisms. Most molds found on dry-cured products are harmless, although some molds are undesirable, and may cause allergic reactions and respiratory problems. A few types of molds, under the right conditions, produce mycotoxins, which are poisonous substances that can make consumers sick. Undesirable molds may also affect product quality as they can breakdown proteins and fats. Adherence to SSOPs is critical to prevent undesirable mold growth during processing and to reduce airborne mold spores in fermentation and aging rooms (Quintavalla, 2010). During storage, measures to prevent undesirable mold growth may include using short inventory pull dates, low pH, sufficiently low water activity, antimycotics, coatings, packaging, or any combination of these measures.

If mold is present during processing, depending on the type of mold, establishments may package the product with the mold on or scrub off the mold with a stiff vegetable brush to maintain wholesomeness. FSIS does not recommend washing mold off using hoses that can result in cross-contamination from the environment to the product. Dry-white mold, often seen on fermented and dried sausages, is generally considered to be a good mold because it can prevent "bad mold" from growing although color is not necessarily an indicator of "bad mold". Sometimes a commercial mold culture will be added prior to fermentation to actively apply a live mold culture to prevent the growth of undesirable molds and for flavor development. For more information on the impact of applying a commercial mold culture, see page 71. If a mold culture is not applied, and mold growth is desirable for quality reasons, establishments must determine the hazards reasonably likely to occur (9 CFR 417.2) and support the decision-made (9 CFR 417.5). As part of that decision-making the establishment must consider the effect mold plays in the process and needs to maintain documentation supporting that they are controlling or preventing any food safety hazards that might occur during this process.

# Appendix 3: Lethality and Shelf-Stability Targets

## Lethality Targets (*Salmonella*, STEC, and *Lm*)

The lethality treatment (*i.e.*, the combination of hurdles or steps) for RTE shelf-stable meat and poultry products should achieve at least a 5.0-log reduction of *Salmonella* and at least a 5.0-log reduction for STEC including *E. coli* O157:H7 for products containing beef as recommended in the FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A). In addition to *Salmonella*, the lethality treatment of RTE shelf-stable meat and poultry products should achieve at least a 3.0-log reduction in *Lm*, although a 5.0-log reduction or greater is desirable for providing an even greater safety margin for ensuring that *Lm* does not grow to detectable levels during storage. However, establishments are not required to validate their process achieves reductions in *Lm* (or STEC for products containing beef) if it achieves sufficient reductions in *Salmonella* as indicated in the FSIS HACCP Systems Validation Guideline.

Unlike cooking; however, research has shown that:

- STEC including *E. coli* O157:H7 and *Lm* are more tolerant than *Salmonella* during the fermentation and drying steps of dry/semi-dry fermented sausages (Hussein, *et al.,* 2022; Ihnot *et al.*, 1998; Porto-Fett *et al.*, 2010; McKinney, 2019).

- *Lm* is more tolerant than *Salmonella* during the drying step of dried and salt-cured meat and poultry products (Porto-Fett *et al.*, 2010; Reynolds *et al.*, 2001).

Therefore:

- If an establishment's scientific support is only based on reductions in *Salmonella* and the establishment has a STEC or *Lm* product positive either through its own testing or FSIS' testing or is associated with an outbreak of these pathogens, the Agency would require the establishment as part of its corrective actions to validate for the other pathogens unless it can support the cause of the positive was post-lethality contamination.

- FSIS also recommends establishments conducting new challenge studies (studies where the product is inoculated with bacteria to determine the processes effectiveness) determine the log reductions in *Salmonella*, STEC, (in products containing beef) and *Lm*. If an establishment is only able to include one pathogen, it may include either *Salmonella*, STEC, or *Lm* although FSIS recommends choosing *Lm* or STEC (for products containing beef) due to their increased survival in response to fermentation and drying.

- Because STEC have been shown to be more tolerant to fermentation and drying than *Salmonella* and have been shown to have similar tolerance to *Lm*, FSIS would not object to establishments using research that only demonstrates *E. coli* O157:H7 reductions in beef and does not include *Salmonella* or *Lm*.

- If an establishment's scientific support includes *Lm* and STEC, the support should demonstrate at least a 3.0-log reduction in *Lm* and at least a 5-log reduction in STEC to be considered adequate.

- If an establishment's scientific support only includes *Lm* (and not *Salmonella* or STEC), FSIS recommends the support should demonstrate at least a 5.0-log reduction in *Lm* in order to support at least a 5.0-log reduction in *Salmonella* and STEC (in beef) is achieved.

Establishments can demonstrate a 5.0-log reduction in *Salmonella* (and STEC in products containing beef) by using a combination of hurdles. However, if one of those hurdles that is used as part of the overall lethality is only applied to the meat or poultry raw materials (such as an antimicrobial intervention that is only applied to the raw meat or poultry component), establishments should provide support that the pathogen load on any non-meat ingredients has also been reduced or that those ingredients are free of pathogens (*e.g.*, by using COAs that report lot by lot test results).

## What Options do I have if my Steps/Hurdles don't Achieve a 5.0-log Reduction?

Establishments that are unable to demonstrate a 5-log reduction in *Salmonella* or STEC (in products containing beef) also have the option of applying **alternative lethalities.** One accepted alternative lethality is based on "Option #5" from The Blue Ribbon Task Force in which the raw batter of sausage is tested in conjunction with the application of a process that achieves at least a 2.0-log reduction in the hazard of concern. This option was developed for comminuted products where the entire raw batter with all the raw ingredients (*e.g.*, spices, flavor, salt, sugar, nitrite) are tested together. The raw batter testing option is discussed in detail in The Blue Ribbon Task Force document.

Importantly, when using this option, each and every lot of raw batter should be tested following the recommendations for sample size and number of samples collected per lot in The Blue Ribbon Task Force document. This option provides less assurance of product safety, so it is important that the raw material testing provides a high degree of confidence that there are no hazards of concern present. Although testing every lot of raw batter may not be practical for every small and very small establishment, it was recommended originally by the Blue Ribbon Task force as "the most practical solution to assuring the safety of certain dry fermented sausage products" and represents the best alternative when a 5.0-log reduction cannot be supported.

FSIS has received a number of askFSIS questions regarding "Option #5" from The Blue Ribbon Task Force and based on

### KEY DEFINITIONS

**Alternative lethality** is a lethality target or log reduction that is different from FSIS recommendations, but achieves an equivalent probability that no viable *Salmonella* organisms or other pathogens of concern remain in the finished product when properly implemented as described in the supporting scientific documentation.

these has the following guidance related to applying the option to dry/semi-dry fermented sausage products containing beef:

- Establishments can test the raw batter of beef products for STEC including *E. coli* O157:H7 only as described in the Blue Ribbon Task Force document.

- Because STEC are more tolerant than *Salmonella* during the fermentation and drying steps of dry/semi-dry fermented sausages as previously described, establishments producing dry/semi-dry fermented sausage products do not have to also test the raw batter for *Salmonella* or validate the process achieves a specific log reduction of *Salmonella*.

- FSIS also would not object to establishments testing the batter of beef containing products for *Salmonella* and support that the process achieves at least a 2.0-log reduction in *Salmonella* given *Salmonella* has been used as an indicator. If an establishment's scientific support is only based on reductions in *Salmonella* and the establishment has a STEC or *Lm* positive either through its own testing or FSIS' testing or is associated with an outbreak of these pathogens, the Agency would require the establishment as part of its corrective actions to validate for the other pathogens unless it can support that the cause of the positive was post-lethality contamination.

FSIS has the following guidance for products not containing beef (*e.g.*, those containing pork) that are unable to demonstrate a 5.0-log reduction in *Salmonella* or *Lm*:

- Establishments can test the raw batter for *Salmonella* and support the process achieves at least a 2.0-log reduction in *Salmonella* without demonstrating that specific reductions in *Lm* are achieved.
    - Because *Salmonella* is less tolerant than *Lm* during the fermentation and drying steps of dry/semi-dry fermented sausages, if an establishment's scientific support is only based on reductions in *Salmonella* and the establishment has a *Lm* product positive either through its own testing or FSIS' testing or is associated with an outbreak, the Agency would require the establishment as part of its corrective actions to validate for *Lm* unless it can support that the cause of the positive was post-lethality contamination.

- Establishments can test the raw batter for *Lm* and support the process achieves at least a 2.0-log reduction in *Lm* without demonstrating that specific reductions in *Salmonella* are achieved.
    - Because *Lm* are more tolerant than *Salmonella* during the fermentation and drying steps of dry/semi-dry fermented sausages as previously described, establishments producing dry/semi-dry fermented sausage products do not have to also test the raw batter for *Salmonella* or validate the process achieves a specific log reduction of *Salmonella*.

In terms of the specific testing, the option in The Blue Ribbon Task Force document recommends fifteen 25-gram (g) samples be taken from across the lot (regardless of lot size or weight) and indicates further research is needed to establish the limits of

compositing. For *Salmonella* and *E. coli* O157:H7, FSIS recommends compositing up to three – 25-g samples (total 75 grams) for a total of 5 analyses, although establishments may also be able to support compositing all fifteen – 25-g samples (total 375 grams). If establishments choose to test for *Lm,* FSIS recommends compositing up to 5 samples (total 125g) for a total of 3 analyses. When compositing, establishments should ensure the method has been validated for the larger test portion.

## How Can I Apply the Concept of Raw Batter Testing to Whole Muscle Products?

FSIS has identified that establishments producing products using whole muscle cuts (*e.g.*, biltong) are also unable to achieve at least a 5-log reduction of hazards of concern. FSIS recommends as an alternative, that every lot of the raw materials are tested in conjunction with a process validated to achieve at least a 2.0-log reduction. For whole muscle cuts, establishments should test both the meat or poultry and non-meat ingredients to support that the incoming pathogen levels are low and that a 2.0-log reduction would be sufficient to achieve product safety with confidence. For the non-meat ingredients, establishments may be able to support the incoming pathogen load of the raw materials is low through means other than their own testing. For example, when using spices, an establishment may receive a COA or LOG that indicates how each lot of ingredients is processed, tested, or otherwise treated to ensure its safety.

For the meat or poultry component, FSIS recommends establishments collect fifteen – 25-g samples of the meat or poultry whole muscle cut using **excision sampling**. This sample size is the same as that recommended for the raw batter testing option and the International Commission on Microbiological Specifications for Foods (ICMSF) case 13 sampling plan (for more information on the ICMSF sampling plans, see page 120 of the FSIS Guideline: Controlling *Listeria monocytogenes* in Post-Lethality Exposed RTE Meat and Poultry Products). FSIS recommends that excision samples are used for intact whole muscle cuts, but not for injected, vacuum-tumbled, or mechanically tenderized (by needle or blade) products. If the raw meat or poultry component is processed in such a way that it is made non-intact, the sampling should include excisions of both interior and exterior portions. The reason for this is because research has shown that such processing techniques push bacteria into meat and are less likely to be detected on the surface. One way to excise the surface and interior of meat is with a sanitized coring device (Ulbrich, 2015; Luchansky, 2008). For this sampling technique, establishments should use a sterile coring device with a 10 $cm^2$ diameter (surface area) to obtain core samples. Sanitized knife and forceps are recommended to cut segments of the exterior and interior portions of the core sample. For further guidance on this method see Luchansky *et al.*, 2008. Guidance regarding selection of organisms for testing and validation as well as for compositing of raw batter samples provided for raw batter testing provided earlier in this document on page 42 also applies to sampling and testing of whole muscle raw materials.

## Shelf-Stability Targets

Fermented, salt-cured, and dried products typically achieve **shelf-stability** through a combination of hurdles such as reduced pH, reduced water activity, or a combination of both, along with other extrinsic factors, such as reduced oxygen and packaging. *S. aureus* is the main pathogen of concern during storage of shelf-stable products because it can grow at a lower water activity than other pathogens. To achieve of shelf-stability, no outgrowth of *S. aureus* may occur in the product. Minimizing available water (*e.g.*, by achieving a sufficiently low water activity) is necessary to achieve shelf stability, provided measures are taken to address undesirable mold growth. Such measures to prevent undesirable mold growth may include using short inventory pull dates, low pH, **antimycotics** (such as potassium sorbate sprays and dips), coatings, packaging, or any combination of these measures.

For examples of scientific support that can be used to support shelf-stability see Appendix 5.

**NOTE:** FSIS has a regulatory definition of shelf-stability in (9 CFR 431.1) but this only applies to thermally processed, commercially sterile products and does not apply to the types of products described in this guideline.

## Can I Use Finished Product Testing Alone if I Don't have Scientific Support for my Process?

No. FSIS does not consider test and hold (also sometimes described as "Option #3" from The Blue Ribbon Task Force document) as acceptable support because it relies on finished product testing alone and does not support a specific log reduction in levels of target pathogens. So, relying on finished product testing alone is not consistent with the concepts of HACCP in which hazards are to be prevented or controlled by the HACCP system. In addition, water activity testing alone would also not be an appropriate way to support the safety of RTE meat and poultry products for the same reasons.

**NOTE:** An establishment may rely on finished product testing alone to support product safety for a limited time during the initial validation period (90 calendar days) if it is in the process of identifying alternative scientific support (*i.e.*, journal articles or challenge studies).

## KEY DEFINITIONS

**Moisture Protein Ratio (MPR)** expresses the percent moisture divided by the percent protein. MPR is commonly used in the U.S. to classify dried sausages and other meat products. Although MPR values indicate the degree of product drying, they are not necessarily indicative of microbial safety or product shelf-stability because they do not take into account availability of the water.

## Appendix 4: Considerations for Different Types of Scientific Support

The following addresses considerations for different types of scientific support that may be used to support the lethality or shelf-stability of fermented, salt-cured, or dried meat and poultry products.

### *Peer-Reviewed Scientific or Technical Data or Information*

Peer-reviewed journal articles, many of which are discussed throughout this document, are available to support the lethality and shelf-stability of fermented, salt-cured, and dried RTE meat and poultry products. When establishments are reviewing the literature, they should thoroughly evaluate whether the critical operational parameters used in the support match those used in the actual process. FSIS recommends that establishments use a single supporting document to support the effectiveness of its lethality steps. If multiple studies are used together to support the same process, the establishment will need to support that the new combination of parameters would be as effective as those studied in the individual articles or documents (80 FR 27557).

### *Pathogen Modeling Programs*

Establishments should not rely on the results of pathogen modeling programs alone unless those models have been validated. The following are validated models FSIS recommends for supporting decisions related to fermented, salt-cured, or dried meat and poultry products:

- **University of Wisconsin Shelf-Stability Predictor** available at: https://meathaccp.wisc.edu/ST_calc.html.
    - This model has been validated for estimating the likelihood of *S. aureus* and *Lm* growth.
    - Input parameters are pH and water activity.
    - For more information see Appendix 5.

- **Danish Technological Institute (DMRI) ConFerm model** available at http://dmripredict.dk (Gunvig *et al.*, 2016)
    - This model has been validated for estimating the reductions of *Salmonella*, STEC, and *Lm* in fermented products.
    - At most, the model can be used to support a 3.0-log reduction of *Salmonella*, STEC, and *Lm.*
    - Should only be used for products with matching critical operational parameters as described in Gunvig *et al.*, 2016.
    - Input parameters are: fermentation temperature, % salt in the formulation, ingoing nitrite ppm, pH at the start before fermentation, pH at 48 hours, final pH, % weight loss in product in the beginning and after drying, total process time (fermentation + drying), and water % in final product (as determined through laboratory analysis).

- **DMRI Staphtox Predictor (Version 1.0)** available at: http://dmripredict.dk/
    - This model has been validated (Gunvig *et al.*, 2017) to predict the growth of *S. aureus* and potential toxin formation during mild heat treatment and during constant temperature fermentation of sausages ($a_w$>0.96).
    - Input parameters are NaCl in product, KCl in product, sodium nitrite in the recipe/ingoing, % water in product (as determined through laboratory analysis), time, pH, and temperature.
    - For more information on using this model to evaluate to evaluate product safety during fermentation deviations, see Appendix 7.

Other models are available for estimating reductions in bacterial pathogens of concern in fermented, salt-cured, or dried meat and poultry products, such as the *E. coli* O157:H7 survival models for fermented sausage that are part of the Agricultural Research Service (ARS) Pathogen Modeling Program (PMP) Online and the Meat and Livestock Model (MLA) *E. coli* inactivation model in Fermented Meat. However, these other programs are not considered validated at this time because the results of the modeling programs have not been compared to the results of actual studies to determine their accuracy. These models can be useful for providing an initial estimate of reductions in levels of pathogens prior to conducting a challenge study. Establishments should not rely on these models alone as scientific support unless they have additional support.

***Challenge or Inoculated Pack Studies***

When establishments want to use unique processes that are not supported by the literature, because there is no literature available or the process used is significantly different from that studied, a challenge study may be needed to demonstrate the safety of the process. General guidance on conducting challenge studies can be found on page 8 of the FSIS HACCP Systems Validation Guideline. Specific considerations for the design of challenge studies for fermented, salt-cured, and dried products are included in Appendix 15.

***Data Gathered by the Establishment In-Plant***

When an establishment has a scientific support document such as a peer-reviewed journal and it wants to use different critical operational parameters, it may consider collecting in-plant microbiological data to support the new combination of steps to achieve adequate reduction in bacterial pathogens of concern, particularly if data or scientific principles are not available to support the changes. For example, if an establishment producing a dried meat product identifies a journal article that matches its process, but it intends to use a slightly lower drying temperature (*e.g.*, 2 or 3°F lower than that used in the support), it may choose to collect in-plant microbiological data to provide additional support for its process. In this case, the establishment would take a statistically based number of samples **each day** it produces the product during a 90-calendar day period and analyze the finished product for *Salmonella,* STEC (including *E. coli* O157:H7 in products containing beef), and *Lm*. If the establishment produces the product less than 13 days within a 90-calendar day period, it should continue to test product until 13 different lots have been tested and found negative for *Salmonella,* STEC (including *E. coli* O157:H7 in products containing beef), and *Lm* using a method as sensitive and specific as FSIS'. Products could be shipped into commerce after test

results on each individual lot are received while the establishment is validating its process because the products would be considered to meet the definition of RTE in 9 CFR 430.1 (that it is in a form that is edible without additional preparation to achieve food safety). The establishment could also choose to produce and sample experimental products that are not offered for sale, are not introduced into commerce, and are not given away to potential customers. Because experimental products are not for introduction into commerce, they are not subject to inspection, are not eligible to bear the mark of inspection, and do not need to be produced in accordance with the meat and poultry products inspection regulations (9 CFR Part 300).

It would not be appropriate to use microbiological data gathered as part of in-plant validation data to support large differences between a journal article and an establishment's process, rather a new challenge study would likely be needed to support the unique combination of parameters. What is considered a large difference will depend on factors such as the parameter (*e.g.*, temperature, pH, water activity) and the sensitivity of the scale for measurement (*e.g.*, a 0.1 difference in temperature may not have as big of an impact as a 0.1 difference in water activity). For example, if an establishment producing a fermented meat product identifies a journal article that matches its process, but it intends to ferment to a higher pH (*e.g.*, 5.3 instead of 4.8 as shown in the support) it would not be appropriate to rely on in-plant microbiological data to support the differences. In this example, the establishment should conduct a challenge study to support its unique combination of parameters. While the challenge study is being conducted, products could be shipped into commerce during the 90 day initial validation period if each individual lot of product is tested as described in 2. above because the products would be considered to meet the definition of RTE in 9 CFR 430.1 (that it is in a form that is edible without additional preparation to achieve food safety).

## Appendix 5: Scientific Support Available for Shelf-Stability

Establishments typically need different scientific support to support using either water activity alone or a combination of pH and water activity to achieve shelf-stability than the support used to achieve lethality. This is because to support shelf-stability, establishments must have scientific support that these parameters allow no outgrowth of *S. aureus* in the product during storage (9 CFR 417.5(a)(1)):

- The **Food Standards and Labeling Policy Book** available at: https://www.fsis.usda.gov/guidelines/2005-0003 contains critical operational parameters such as certain levels of MPR, pH, water activity, and brine concentration for **shelf-stable sausages** or **Lebanon bologna.** Establishments can (but are not required to) use the criteria as support for shelf-stability because these criteria were based on ensuring no *S. aureus* growth would occur. Establishments that use other criteria may still need to monitor the MPR as part of in-plant validation and on-going verification activities for standard of identity purposes (e.g., salami must have an MPR of 1.9:1 or less) to not be considered false and misleading (9 CFR 317.8 or 9 CFR 381.129).

  **NOTE:** The regulatory standards of identity, 9 CFR 319.106, for country ham and dry cured ham do not ensure or support the product is shelf-stable. Rather, the criteria in the standard of identity were designed to ensure that the finished products have quality characteristics associated with hams and pork shoulders (42 FR 3299).

- The FSIS Guideline for Meat and Poultry Jerky Produced by Small and Very Small Establishments contains guidance on water activity parameters that support shelf-stability. Although that guideline is about jerky, the water activity parameters can be applied to other shelf-stable meat and poultry products.

- Research (Tilkens *et al.*, 2015**)** supports that a combination of ≤ pH 5.1 and water activity ≤ 0.96 in RTE snack sausages containing salt, dextrose, sodium nitrite, and sodium erythorbate does not support the growth of *S. aureus* and thus could be considered shelf-stable for this pathogen under anaerobic conditions (*e.g.*, packaged under vacuum). It also supports a combination of pH ≤ 5.1 and water activity ≤ 0.92 does not support the growth of *S. aureus* when products are stored aerobically although mold grew under these conditions in the study; therefore, establishments using these parameters should take additional measures to address mold growth.

- The **University of Wisconsin Shelf-Stability Predictor** available at: https://meathaccp.wisc.edu/ST_calc.html provides accurate predictions for *S. aureus* growth. It can be used alone as supporting documentation to support shelf-stability, provided that the establishment's product is like those the model was developed for (Borneman *et al.*, 2009; available on the shelf-stability predictor website).

## Appendix 6: Critical Operational Parameters for Fermentation

During fermentation, bacteria consume available carbohydrates (*e.g.*, dextrose or sucrose) and moisture in the meat or poultry to produce organic acids (*e.g.*, lactic acid and citric acid) among other compounds to lower the pH and reduce the amount of available water (reduce the water activity) in the meat. In meat or poultry, it is the lactic acid that is primarily responsible for the reduction in pH (Benito, 2007; Ortiz, 2014). **It is important to note that even with fermentation that results in lactic acid production and reduced pH, fermentation itself is not a particularly effective lethality treatment and pathogens such as *Salmonella*, STEC, and *Lm* can survive** (Faith *et al.*, 1997; Faith *et al.*, 1998a; Faith *et al.*, 1998b; Hussein *et al.,* 2022; Ihnot *et al.*, 1998). One reason is because bacteria become tolerant to the acid produced during the fermentation process particularly if it is a slow process (Leyer *et al.*, 1993; Chikthimmah and Knabel, 2001; Theron, 2007). The pathogens that do survive fermentation also become tolerant to other treatments like heat that would otherwise be lethal in a typical processed meat or poultry product (Brul, 1999).

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Examples of critical operational parameters for the reduction of *Salmonella*, STEC, and *Lm* during fermentation include:

- Fermentation temperature, target pH, time to reach target pH, and smoke (if used).
- Type and use of starter cultures.
- Product characteristics: Casing diameter and shape and product formulation including salt, sugar (type and level), and use of nitrite or nitrate.

In addition to these critical operational parameters during fermentation, how the raw batter is handled prior to fermentation can also have an impact on the effectiveness of the step. For example, research showed that raw batter conditioned by tempering at 55.4°F (13°C) for 2 hours, freezing, and thawing prior to stuffing and fermentation resulted in greater reductions in *E. coli* O157:H7 during fermentation than raw batter that was either refrigerated or frozen and thawed (but not tempered first) prior to stuffing and fermentation (Faith *et al.*, 1998).

Specific considerations for several critical operational parameters for the reduction of *Salmonella*, STEC, and *Lm* as related to fermented products are outlined below.

***Fermentation Temperature, Target pH, Time to Reach Target pH, and Smoke (if used)***

The establishment must be fermenting its product to the pH that is recommended in the supporting documentation or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Temperature impacts the final pH achieved during fermentation as well as the amount of time it takes to reach the target pH. In general, the higher the fermentation process temperature and humidity, the faster the fermentation (fermentation temperatures are typically no higher than 110°F).

However, the fermentation temperature should be at the optimum growth temperature of the added starter culture. The final pH will be affected by the added carbohydrate, the heating temperature after fermentation, and the drying conditions.

There are two key target pH values during fermentation:

- pH at which *S. aureus* growth is controlled (pH ≤ 5.3).
- Final product pH that contributes to lethality of *Salmonella*, STEC, and *Lm* (which will vary depending on the process and supporting documentation).

In addition to the target pH level itself, the time it takes the product to reach the desired pH is also important. For *S. aureus* control, it is important that the establishment monitor the time it takes the product to reach pH of 5.3. By monitoring the product pH and time it takes to reach the desired pH, the establishment can ensure its process is within an acceptable number of degree-hours and, if applicable, achieves a secondary pH in an amount of time consistent with the supporting documentation. If product takes longer to achieve the secondary pH than occurred in the supporting documentation, the process may not be as effective because it could allow time for the pathogens to adapt to the acidic conditions. In general, the lower the final pH and the faster it is achieved the greater reductions in pathogens such as *Salmonella* (Porto-Fett *et al.*, 2008) although as indicated on the previous page, fermentation itself is not a particularly effective lethality treatment.

Added smoke will sometimes inhibit fermentation at the product surface, but the extent to which this occurs will depend upon the product diameter.

**NOTE:** For those processes where fermentation is followed by cooking, FSIS does not object to an establishment using the worst-case chamber temperature (110 °F) to set the maximum allowable degree-hours and demonstrating 1) the total fermentation time plus cooking is 18 hours or less (maximum allowable degree-hours at 110 °F) and 2) the pH is 5.3 or less at the end of cooking.

***Type and Use of Starter Cultures*** - Today, most fermented meat processors either add lactic acid starter cultures and/or harmless staphylococci to the raw meat mix. The composition of the starter culture (*i.e.*, the strains) used in the product must be similar in composition to that used in the supporting documentation, to ensure that fermentation is achieved, and the rate of pH drop is as expected. If the starter culture is different the establishment must provide justification for why the different starter culture would be equally effective in order to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). The starter culture should be formulated to ensure microbial dominance of fermentation strains over any potential pathogens and to inhibit potential *S. aureus* growth during fermentation. In addition, the starter culture used for fermentation can affect whether **bacteriocins** are produced and the type of bacteriocins produced, which can affect the level of reduction for bacterial pathogens. For example, pediocin-producing *Pediococcus acidilactici* are more effective than non-pediocin-producing *Pediococcus acidilactici* at reducing *Lm* in chicken and turkey summer sausage (Baccus-Taylor *et al.*, 1993; Luchansky *et al.*, 1992). It is also important that the starter culture is well-dispersed throughout the raw batter.

**NOTE:** An alternative to commercial starter cultures for reducing the pH in sausage batters is **direct acidulation** by the addition of organic acids like lactic, citric, or glucono-delta-lactone (American Meat Institute, 1997). This guideline does not address the use of direct acidulation.

## Why is Using a Starter Culture Important?

If a starter culture and fermented sugars are not used, pathogenic bacteria will not be reduced in the product during fermentation (Calicioglu *et al.*, 2002; Smith and Palumbo, 1978; Smith and Palumbo, 1983). The use of a starter culture (*e.g.*, lactic acid bacteria) and fermentable sugars added to the initial fermented sausage mix assures microbial dominance over the potential pathogenic microorganisms. This means that in the absence of a starter culture *Salmonella,* STEC, and other undesirable microorganisms will instantly become the dominant microflora. To prevent pathogen dominance, establishments should use a starter culture to prevent or severely limit the growth of the bacterial pathogens of public health concern during the initial phase of the fermentation step of dry/semi-dry fermented sausages. The following factors are important to ensure food safety:

- Microbial competition.
- Production of lactic acid which lowers the product pH and inhibits the growth of pathogenic bacteria.
- Possible production of bacteriocins, which inhibit or reduce pathogenic microorganisms.

As described in Good Manufacturing Practices for Fermented Dry and Semi-Dry Sausage Products, there are two general ways in which lactic acid-forming bacteria for fermentation may be incorporated into the raw batter:

1. The preferred and most reliable method is to use a commercially prepared culture which is handled and used as prescribed by the manufacturer.
2. A less reliable procedure is the use of a portion of a previously fermented and controlled **mother batch**. Because this method is less precise than using a commercial culture, it is important that the inoculum derived from the mother batch be composed of a vigorous culture capable of producing a rapid pH decline.

> ***Key Point***
>
> A starter culture helps ensure that the fermentation is vigorous enough to lower the pH to 5.3 or less rapidly.

In addition to the concerns, identified in Good Manufacturing Practices for Fermented Dry and Semi-Dry Sausage Products, using a mother batch for fermentation is a less reliable procedure because it could lead to recycling of spoilage organisms and pathogens. Another concern with the use of a mother batch is the inconsistency in the starter culture. For these reasons, FSIS does not recommend this practice. If an establishment uses a mother batch, it must address the potential for recycling of spoilage organisms

and pathogens in its hazard analysis as well as the consistency of the starter culture (9 CFR 417.2(a)(1)). Establishments may consider characterizing the type and level of bacteria in the mother batch to support their decisions (9 CFR 417.5(a)(1). It is also particularly important the establishment address the potential for *S. aureus* growth during the fermentation and measure the degree-hours to ensure the pH is reduced to ≤ pH 5.3 within an acceptable number of degree-hours at a given fermentation temperature as described on page 37.

As indicated in the GMP document, a third method, used historically, relied on lactic acid bacteria which naturally occur in fresh meat to initiate the fermentation. While this practice had been used in the past and was the original art of making fermented sausage; the method is highly unreliable and **should not be used.**

***Product Characteristics: Casing diameter and shape and product formulation including salt, sugar (type and level), and use of nitrite or nitrate.***

The following product characteristics are important critical operational parameters to ensure the fermentation step is effective:

The **casing diameter** will impact the fermentation rate and final pH by affecting heat penetration and moisture migration in, and then out, of the sausage chub. Generally, large diameter products ferment slower due to slower heat transfer and they take longer to reach the desired final pH.

**Product formulation including salt, sugar (both the type of sugar and amount) and use of nitrite or nitrate** plays a role in the fermentation process and may also affect microbial tolerance to acid or heat. The establishment should understand the critical operational parameters associated with the product formulation (e.g., % salt, sugar type and level, moisture level, level of nitrite or nitrate or any other preservatives, and % fat) and should ensure that the materials used in the supporting documentation is like their product with respect to those critical operational parameters and ingredients. Reduced salt was a potential contributing factor in a 2021 salmonellosis outbreak associated with Italian-style meats (FSIS, 2022).

Although typically thought of for *Clostridial* control, nitrite is also a hurdle which inhibits *Salmonella* (Honikel, 2010). According to Honikel, 2010, “fermentation can be produced with only salt, but there is a greater microbial risk if no nitrite is used.” Nitrite alone may be used, or it may be used in combination with nitrate, which is often added as a reservoir for nitrite in long-term processing. Regardless, establishments must ensure the same amount of nitrite or nitrate, as well as any cure accelerators, such as ascorbate or erythorbate, is used as in their scientific support or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Establishments must also ensure the levels of sodium nitrite (whether from a natural or synthetic source) and other restricted ingredients are safe and suitable according to FSIS Directive 7120.1, *Safe and Suitable Ingredients Used in the Production of Meat and Poultry Products* and 9 CFR 424.21(c)).

If establishments are using **natural sources of sodium nitrite**, FSIS recommends that establishments use natural sources of sodium nitrite with known concentrations of nitrite. By knowing the concentration of nitrite, establishments can ensure they are

adding the same amount as used in the scientific support. Because natural sources of nitrite are not currently approved for use in 9 CFR 424.21(c) as curing agents, products that are required to contain curing agents and cure accelerators as part of a standard of identity in 9 CFR 319 or 9 CFR 317.17(b), but instead are formulated with natural sources of nitrite and ascorbate, must be labeled as “uncured” under 9 CFR 319.2. Also, the label must contain the statement “no nitrates or nitrites added” (9 CFR 317.17) that is qualified by the statement, “except for those naturally occurring in [name of natural source of nitrite such as celery powder]” as to not be considered misbranded due to false and misleading labeling under 9 CFR 317.8. Uncured products are also required to bear the handling statement, “Not Preserved - Keep Refrigerated Below 40 °F. At All Times” as per 9 CFR 317.17(c)(2). However, this handling statement is not required if an uncured product is processed to be shelf stable by fermenting or pickling to pH of 4.6 or less or drying to a water activity of 0.92 or less. For more information see FSIS’ Stabilization Guideline for Meat and Poultry Products (Revised Appendix B).

**Question:** ***Why are there so many critical operational parameters during fermentation? If I meet the degree-hours isn't that enough to show biological hazards are addressed?***

**Answer:** No, degree-hours are used to control the outgrowth of *S. aureus.* To reduce levels of other pathogens of concern in fermented products, such as *Salmonella*, STEC, and *Lm,* establishments often need to ferment these products to a lower pH. Other parameters, such as fermentation temperature, time to final pH, and formulation, also play a role in reducing other types of bacteria. All critical operational parameters within the scientific support must closely match the establishment's actual process or the establishment must provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)).

***Question: What if I meet the degree-hours and dry my product following one of the methods in FSIS's Trichinella Guideline and dry to a reduced water activity such as to a water activity below 0.85, isn't that enough to show biological hazards are addressed?***

**Answer:** No. Fermentation and drying alone are not particularly effective as lethality treatments. When these steps are found to achieve sufficient reductions in pathogens, there are a lot of critical operational parameters associated with both steps that need to be implemented in the actual process consistent with the scientific support. It is not enough to only meet degree-hours, follow the minimum number of drying days for eliminating *Trichinella*, and achieve a final water activity for shelf-stability. Degree-hours are intended to control the outgrowth of *S. aureus*. To reduce levels of other pathogens, such as *Salmonella*, STEC, and *Lm,* products often need to be fermented to a lower pH than 5.3. Also, the drying time and other factors are important to reducing bacteria during drying – not just the final water activity. The minimum number of drying days for eliminating *Trichinella* have not been validated to achieve any particular reductions in *Salmonella* and products often have to be dried longer to get significant reductions in *Salmonella*. In addition, smaller diameter products tend to dry faster than large diameter products, and the shorter drying time is often not enough time to achieve the same reductions in bacteria as larger diameter products even if it is enough time to achieve the targeted water activity (DeSouza, 2018).

Here are some citations that support the need for other critical operational parameters establishments should address during fermentation, in addition to degree-hours:

***Blue Ribbon Task Force of the National Cattlemen's Beef Association. May 1996. Dry Fermented Sausage and E. coli O157:H7 (Research Report No. 11-316).***

- Table 6 in the article shows that when large diameter (105 mm) salamis were fermented at 90°F to a pH ≤ 4.6 and held, the average *E. coli* O157:H7 log reduction was 4.72. In contrast, when the products were fermented at 90°F to a pH ≥ 5.0 and held in the chamber at that temperature for 7 days, a 2.87-log reduction was measured. The same findings, although less pronounced, were observed for large diameter salamis fermented at 110°F to a pH ≤ 4.6 and held.

The average *E. coli* O157:H7 log reduction was 6.42 as opposed to 6.03-log reduction for products fermented to ≥ 5.0.

***Porto-Fett, ACS, Hwang, C-A, Call, JE, Juneja, VK, Ingham, SC, Ingham, BH, Luchansky, JB. 2008. Viability of Multi-Strain Mixtures of Listeria monocytogenes, Salmonella Typhimurium, or Escherichia coli O157:H7 Inoculated into the Batter or Onto the Surface of a Soudjouk-Style Semi-Dry Sausage. Food Microbiology. 25: 793-801.***

- The authors fermented sausage with different levels of dextrose that resulted in a noticeable difference in pH values between sausages (pH 5.27 when formulated with 0.25% dextrose versus pH 4.81 when formulated with 0.60% dextrose).
- The authors reported the addition of a pedicococcal starter culture and added dextrose decreased the pH to approximately 5.3 or 4.8 following fermentation and drying, and the lower pH resulted in a quantifiable greater lethality.

These scientific support documents illustrate why meeting degree-hours is not enough to show biological hazards (*i.e., Salmonella*, STEC, and *Lm*) are addressed by the fermentation step.

For information related to fermentation deviations see Appendix 7.

***Other critical operational parameters during fermentation that do not need to be addressed***

A high relative humidity (85-90%) is preferred during fermentation to keep the product surface slightly moist (tacky) during fermentation and prior to subsequent drying. This avoids premature and uneven drying at the surface and the humidity enhances the fermentation by limiting undesirable evaporative cooling, increasing heat penetration, and speeding the reduction in pH. For these reasons, establishments should try to ensure that the relative humidity in the actual process is at least as high as the lower end of the relative humidity range used in the supporting documentation. It is recommended establishments determine the relative humidity levels in the actual process during the initial set-up of the system as part of in-plant validation as well as part of ongoing verification.

Establishments that are unable to determine the relative humidity level used in the scientific support or that are not able to meet the same or higher level of relative humidity from the support may also be able to use the implementation of other parameters such as final fermentation pH and time to reach pH to demonstrate that adequate relative humidity is present for the fermentation process. For example, The Blue Ribbon Task Force document, commonly used as scientific support for fermented sausage processes, does not report the level of relative humidity that was used. FSIS would not object to establishments using this document as support and concluding the relative humidity levels in the actual process are sufficient if the final fermentation pH and time to reach pH are consistent with the levels reported in The Blue Ribbon Task Force document. Indications of insufficient relative humidity include partial or inconsistent fermentation (for more information see Appendix 7: Fermentation Deviations) or indications of case hardening (*e.g.,* demonstrated by a lack of loss of weight during drying). If possible, FSIS recommends establishments still monitor the relative humidity levels as part of in-plant validation and ongoing verification so that

there is data available on typical levels.

The **casing type** influences moisture exchange. Products with impermeable, semi-permeable, or permeable casings exchange moisture with the environment differently and can influence the rate of product acidification, the penetration of heat into the interior of the product, and the maximum internal temperature reached by the product. However, several studies have found no differences in reductions for pork salami fermented and dried in natural, collagen, or fibrous casings (DeSouza, 2018; McKinney *et al.*, 2019). Therefore, establishments using scientific support conducted with a product fermented in one type of casing may be able to support applying that research to a product fermented in a different type of casing.

## Appendix 7: Fermentation Deviations

If the fermentation to a desired pH does not occur within the normal time, it can be due to a variety of reasons. If a fermentation problem does occur, it is often the result of:

- No fermentation (the pH does not change from its initial value of pH 5.6-6.0).
- Partial fermentation (the pH drops slightly to pH 5.4-5.6).
- Inconsistent fermentation (variation in fermentation activity from piece to piece or from location to location).

Below are some typical causes for inadequate or inconsistent meat and poultry product fermentation.

***No Fermentation***

- No starter culture added.
- No fermentable sugars added.
- Excessive salt added.
- Culture was thawed and refrozen.
- Culture premixed with cure, salt, chemicals.
- Antimicrobial agents added to formulation.
- Antibiotic residues in raw meat.

***Inconsistent or Partial Fermentation***

- Inadequate distribution of starter culture.
- Insufficient fermentable sugars added.
- Fermentation temperature above or below the optimal temperature for the starter culture.
- Inconsistent internal product temperature or processing temperature or humidity.
- Reduced fermentative activity of the starter culture.
- Out of code product or improper stock rotation.
- Culture was temperature abused.
- Antimicrobial agents added to formulation.
- Antibiotic residues in raw meat.

One or more of these causes may occur during the fermentation of a meat and poultry product, resulting in a fermentation deviation. Establishments should ensure that critical operational parameters within the scientific support that should closely match the establishment's actual process.

**Evaluation of Product Safety**

Establishments must be able to ensure that no product that is injurious to health or otherwise adulterated because of the deviation enters commerce, and to support its product disposition decisions (9 CFR 417.3(a) and (b)). In the event of a fermentation

deviation, establishments may use pathogen modeling (DMRI Staphtox Predictor) or testing to support product safety. Below are specific recommendations for either approach.

### *Pathogen Modeling Following a Fermentation Deviation*

Establishments may consider using the **DMRI Staphtox Predictor (Version 1.0)** available at: http://dmripredict.dk/ to assess the likelihood of *S. aureus* growth and toxin formation during a constant temperature fermentation deviation of sausages (water activity > 0.96). As explained on page 46, this model has been validated. The seven input variables and their ranges for entering information into the model are provided below:

Input variables and ranges:

1. NaCl in product: 1.8 to 4.2% in steps of 0.1%.
2. KCl in product: 0 to 4.2% in steps of 0.1%.
3. Sodium nitrite in the recipe/ingoing: 0-150 ppm in steps of 10 ppm.
4. % water in product as determined through laboratory analysis: 62-78% in steps of 1%.
5. Time: in hours.
6. pH: 4.6-6.0 in steps of 0.1 or pH change during fermentation of sausages.
7. Temperature: 15-40°C in steps of 1°C or time and temperature profile (maximum 500 data points or rows).

If modeling estimates **< 3.0-log growth of *S. aureus* and no toxin formation**, then modeling is adequate to show that the process prevented enterotoxin formation.

If modeling estimates **≥ 3.0-log growth of *S. aureus***, then product should be **tested** following the guidance below (even if the model predicts no toxin formation).
FSIS does not know of other validated pathogen modeling for estimating growth and toxin formation during a constant temperature fermentation deviation.

### *Testing Following a Fermentation Deviation*

FSIS recommends that establishments follow the recommendations in the Good Manufacturing Practices for Ferment Dry and Semi-Dry Sausage Products for evaluating product safety in the event of a fermentation deviation. Although the guidance document recommends at least three samples be collected per batch, FSIS recommends establishments use a statistically based sampling plan (*e.g.*, ICSMF sampling plans).

FSIS recommends establishments develop sampling plans that include:

- A case 11 or n=10 sampling plan be used in the event of a fermentation deviation, although establishments can provide support for other sample sizes (for more information on the ICMSF sampling plans, see page 120 of The FSIS Guideline: Controlling *Listeria monocytogenes* in Post-Lethality Exposed RTE Meat and Poultry Products).

- If the product is not cooked, the establishment should:
    - Test for the enterotoxin if any samples contain ≥ 1,000 CFU/g < 10,000 CFU/g of *S. aureus*.
    - Reject the lot and condemn the product if any samples contain ≥ 10,000 CFU/g.

- If product has undergone either a low- temperature or high-temperature heat step, establishments should test for the enterotoxin only as any vegetative cells of *S. aureus* would likely be destroyed by the heat treatment.

FSIS does **not** recommend:

- Compositing of samples because the purpose of testing following a fermentation deviation is to determine the level of *S. aureus* and compositing may dilute the sample.

## Appendix 8: Critical Operational Parameters for a Low-Temperature Heat Step

To achieve adequate reductions of pathogens in fermented products, while still maintaining the desired quality characteristics, a low-temperature heat step may be added after the fermentation is complete. For example, a fermentation and drying process for pepperoni achieved ≤ 2.0-log reduction of *E. coli* O157:H7; but, by adding a low-temperature heat step after fermentation (128°F for an hour) they were able to achieve a ≥ 5.0-log reduction of *E. coli* O157:H7 without changes to quality (Hinkens, *et al.*,1996). Dry-curing beef strips, followed by a low-temperature heat step, followed by drying has been found to result in a 5-log reduction of *Salmonella* in a salt-cured basturma product (Genigeorgis and Lindroth, 1984).

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Examples of critical operational parameters for the reduction of *Salmonella*, STEC, and *Lm* using a low-temperature heat step include:

- Time and temperature: Heating come-up time (CUT), hold time, and temperature for low temperature heating step.
- Equipment used to generate heat.
- Product characteristics.

### *Time and Temperature: Heating Come-up Time, Hold Time, and Temperature for Low-Temperature Heat Step*

The **temperature** that the product is heated to, and the amount of **time** the product is held at this temperature (**hold time and temperature**), are critical to ensuring that adequate lethality is achieved. In addition to the hold time and temperature, the **come-up time**, which is the time it takes the product to reach the target temperature for the post-fermentation **low-temperature heat step**, may be important to ensure product safety. Factors such as product diameter and relative humidity, affect heat transfer and the amount of time it takes the product to reach the target temperature. It is important for the establishment to understand how the actual temperature of the product, the CUT, and the amount of time the product is held at the target temperature compared to the supporting documentation. For example, if the CUT in the establishment's process is shorter than the time it takes in the study, the establishment's process may result in a lower level of pathogen reduction and not achieve product safety.

### *Equipment Used to Generate Heat*

Differences in equipment (*e.g.*, smokehouses and ovens) can influence the effectiveness of the process and the speed of fermentation or acidification and heating. For this reason, the establishment should gain an understanding of the pH and temperature profile of the product throughout the process. In addition, seasonality of atmospheric conditions, cold-spot determination, or heating consistency must be understood to support monitoring and verification procedures and the frequencies at which those procedures are monitored and verified (9 CFR 417.5(a)(2)).

***Product Characteristics: Casing Diameter and Product Formulation***

Product **casing diameter** is a critical operational parameter in fermented, dry and semi-dry processes because it affects heat transfer. It is important that the diameter of the product used in the establishment's process is the same or smaller than that of the product used in the supporting documentation. If the diameter of the establishment's product is larger than that of the product used in the supporting documentation, it is possible that the product core will take longer to reach the desired temperature and pH, and a lower level of pathogen reduction would be achieved.

See page 52 information on **product formulation**.

***Other critical operational parameters of interest during low temperature heating that do not need to be addressed***

***Relative Humidity During the Heating Step***

FSIS considers relative humidity to be inherently maintained and, therefore, does not need to be addressed, for products that are cooked in a casing (including a natural casing) even for products that are cooked and then dried. For more information on relative humidity during heating see page 25 of the FSIS Cooking Guideline for Meat and Poultry Products (Revised Appendix A).

See page 56 for information on **casing type**.

## Appendix 9: Critical Operational Parameters for Salt-Curing and Equalization

**Dry-curing** is the process of adding salt, sugar, nitrite, or nitrate by dredging the meat in the cure (known as the **salt box method**) or by applying a pre-weighed cure mixture to the surface of the product. FSIS recommends applying a pre-weighed cure mixture over the entire surface, instead of the salt box method, because this process ensures the same amount of ingredients are added in the actual process as was used in the supporting documentation. During this process, the salt, sugar, nitrite, and/or nitrate begin to migrate throughout the meat tissue. At the same time, moisture from the meat or poultry tissue is extracted by the salt surrounding the meat.

For dry-curing to be effective as a food safety measure, time needs to be given to allow **equalization**, which is the process where the cure mixture migrates throughout the meat tissue.

**NOTE:** High-salt tolerant starter cultures may be added directly to the brine or dry rub used for curing of the whole muscle to provide consistent flavor and improved color. Starter cultures may consist of non-pathogenic *Staphylococci* or *Lactobacilli* strains or a combination of both. The use of starter cultures on whole-muscle cuts is not addressed in more detail in this document since they are usually used for color and flavor development and do not generally impact product safety.

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Examples of critical operational parameters for the reduction of *Salmonella*, STEC, and *Lm* during the **curing** or **formulation step** include:

- Curing temperature.
- Curing time.
- Salt coverage of exposed muscle tissue.
- Product characteristics (*e.g.*, product size and formulation, including salt concentration).

### *Curing Temperature*

The curing temperature (typically ambient curing room temperature) is a critical operational parameter as it impacts the salt migration throughout the product. Specifically, a lower temperature during curing will slow salt migration throughout the product impacting the final brine concentration and water activity at the end of the curing step. During curing, establishments must ensure that growth of pathogens does not occur to significant levels (9 CFR 417.2(a)(1)). Storing at refrigeration temperatures (*e.g.,* by storing below 45°F or 41°F according to the Tompkin paper) is one way to support decisions at that step (9 CFR 417.5(a)(1)).

### *Curing Time*

Since cure migration occurs throughout the entire time the product is being cured, the length of curing time is critical to ensure that a sufficient cure penetration and adequate brine concentration are achieved throughout the product.

### *Salt Coverage*

Salt coverage is also critical as the amount of coverage, the amount of salt, and type of salt (discussed below) will ultimately impact the brine concentration achieved after the equilibration step. The coverage may be described in the supporting documentation either: (1) through complete coverage of the whole muscle cut or (2) the number of pounds per product or per surface area.

### *Product Characteristics: Product Size and Formulation*

**Product size** impacts the surface area, and therefore, the amount of cure needed. Product size also impacts curing and salt equalization times. A greater product surface area requires longer curing times to ensure sufficient cure penetration throughout the product.

**Product formulation** that impacts lethality includes the amount and use of:

#### *Salt*

Establishments must use the same or higher amount or concentration of salt as used in the supporting documentation to ensure the brine concentration and water activity throughout the product after equalization is like what was achieved in the support or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Establishments should use salt by weight rather than volume because the size of the salt grains make a big difference. Fine crystalline salt takes up less space (volume) and will weigh more than an equal volume of coarse granular salt. In addition to the concentration, **uniform salting over the entire surface** is critical to inhibit pathogens and spoilage microorganisms. Moreover, some products may be re-salted several times after the first salting step as the salt migrates into the tissue.

#### *Sodium Nitrite ($NaNO_2$)*

Nitrite is an important component during curing because it, in combination with salt, completely inhibits the growth of *C. botulinum*. Sodium nitrite, in combination with salt, also greatly inhibits the growth of *C. perfringens*, and slows the growth of many other pathogenic bacteria such as *Salmonella* and *Lm* (Buchanan *et al.*, 1989; Honikel, 2010; Sindelar, 2012). The amount of nitrite used in dry-cured products, such as coppa, country ham, prosciutto, and others, can be up to 625 ppm ingoing nitrite for dry-cured products and is based on the **green weight** of the meat in the product formulation. As discussed in Appendix 2, 50 ppm sodium nitrite is considered sufficient to ensure *C. botulinum* will not grow (Reynolds *et al.*, 2006; Johnston *et al.*, 1969; and Tompkin, 1976). The resting or equalization

phase itself is also a critical step for inhibiting *C. botulinum* growth in dry-cured hams (Merialdi *et al.*, 2016).

***Sodium Nitrate ($NaNO_3$)***

Sodium nitrate is used as a source of nitrite. If nitrate is used as the curing agent, the conversion (reduction) of nitrate to nitrite by bacteria in the meat or poultry or by a nitrate-reducing starter culture is a necessary step. The amount of nitrate that is reduced to nitrite is dependent upon the numbers of nitrate-reducing bacteria and several environmental conditions, such as temperature, moisture content, salt content, and pH. For this reason, the conversion rate and subsequent amount of nitrite that is formed is difficult to control. The poor control associated with the reduction of nitrate to nitrite, coupled with the fact that most processors today demand faster curing methods, has led to the diminished use of nitrate in meat and poultry products. FSIS recommends the use of nitrite instead of nitrate.

## ***Equalization***

After curing, products go through an equalization step (typically ≤ 45°F) to ensure salt migrates throughout the product and a sufficient brine concentration and water activity is achieved. Equalization is necessary to prevent the growth of *S. aureus* and other pathogens when temperature is increased during the drying step; this allows for sufficient salt penetration and equilibrium. Equalization often takes many weeks to achieve uniform salt distribution to a level greater than 10%.

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Examples of critical operational parameters for the reduction of *Salmonella*, STEC, and *Lm* during an **equalization step** include:

- Equalization temperature.
- Equalization time.
- Brine concentration and water activity after equalization.
- Product size (diameter or thickness).

### ***Equalization Temperature***

The equalization temperature is a critical operational parameter as it impacts the salt migration throughout the product. Specifically, a lower temperature during equalization will slow salt migration throughout the product impacting the final brine concentration and water activity. During equalization, establishments must ensure that growth of pathogens does not occur to significant levels (9 CFR 417.2(a)(1)). Storing at refrigeration temperatures (*e.g.,* by storing below 45°F or 41°F according to the Tompkin paper) is one way to support decisions at that step (9 CFR 417.5(a)(1)).

### *Equalization Time*

The equalization time is critical to ensuring sufficient brine concentration and water activity at the end of the process. If a process uses a shorter equalization time, then the final brine concentration and water activity may not be sufficient to prevent *S. aureus* growth when product begins drying at a higher temperature.

> ***Key Point***
>
> If a process uses a shorter equalization time than that in the supporting documentation, then the final brine concentration and water activity may not be sufficient to prevent *S. aureus* growth when product begins drying at a higher temperature.

### *Brine Concentration and Water Activity after Equalization*

Establishments must ensure that the product conditions during curing and equalization are such that the product has a high enough brine concentration and low enough water activity at the end of the equalization stage prior to drying to prevent *S. aureus* outgrowth during drying according to the scientific support. If the establishment is not using brine concentration and water activity as controls measured at the end of the equalization stage to prevent *S. aureus* outgrowth during drying then the establishment must provide other support for the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Staphylococcal enterotoxin production is inhibited at brine concentrations above 10% (Jay, 2000; Tatini *et al.*, 1976). Achieving a high enough brine concentration and low enough water activity will ensure that when the product moves into the drying step where temperatures are elevated that *S. aureus* growth is prevented or limited (≤ 2.0-log CFU/g). If the establishment is elevating temperatures during equalization then it must support the product has a high enough brine concentration and low enough water activity at the end of curing to prevent *S. aureus* outgrowth during equalization and drying according to the scientific support or must provide other support for the decisions in its hazard analysis (9 CFR 417.5(a)(1)).

### *Product Size*

Product size is also a critical factor. As product size increases, so does the equalization time since it will take longer for moisture to transfer from the center of the product to the surface and take longer to evaporate.

## ***Other critical operational parameters during salt-curing and equalization that do not need to be addressed***

### *Relative humidity*

Relative humidity in the curing environment impacts the amount of time it takes the cure to migrate throughout the product. If the relative humidity is too high, the curing process will be slowed, resulting in insufficient brine concentration/water activity throughout the product. Therefore, if the brine concentration/water activity at the end of curing/equalization meets the desired target, the establishment can support relative humidity does not need to be addressed.

### *Air Flow*

Along with relative humidity, air flow impacts the moisture loss from the surface of the product. If the air flow is too fast, the product may dry out faster. If air flow is too slow, the moisture loss may also be too slow when compared with the conditions that were studied. Therefore, if the product weight loss meets quality/yield targets, the establishment can support air flow does not need to be addressed.

### ***Drying***

After dry-curing and equalization, products are typically **dried** above refrigeration temperatures so that additional water (moisture) is removed from the product. Salt-cured products are dried to meet a water activity level sufficient to achieve shelf-stability by preventing the growth of microorganisms (*e.g.*, water activity ≤ 0.85), especially toxigenic microorganisms such as *S. aureus*. Drying to an intermediate or low water activity also may achieve additional reductions in *Salmonella*, STEC (in products containing beef), and *Lm*. **For an explanation of the critical operational parameters associated with drying, see** Appendix 11**.**

## Appendix 10: Critical Operational Parameters for Seasoning/Marination of Dried Products

During seasoning and marination salt is added to strips of meat as a dip or as a mixture with other ingredients (*e.g.*, spices, sugar, pepper, or sodium nitrite). The addition of ingredients during formulation and the method of application of antimicrobials (if used, for example, to add additional lethality) can impact their effectiveness at the time of application but also the effectiveness of the subsequent steps including drying.

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Therefore, the following critical operational parameters associated with seasoning and marination to reduce *Salmonella*, STEC, and *Lm* should be considered:

- Product formulation.
- Antimicrobial application (*e.g.*, concentration, pH, coverage, contact time).

### *Product Formulation*

Product formulation plays a role as the addition of ingredients (such as spices) may impart some antimicrobial activity. Product formulation also may affect microbial tolerance to acid or heat. The establishment should understand the critical operational parameters associated with the product formulation (*e.g.*, % salt, moisture level, level of nitrite or any other preservatives, and % fat) and must ensure that the material used in the supporting documentation is like their product with respect to those critical operational parameters or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)).

### *Antimicrobial Application*

Some dried products, such as biltong, contain a marination step with acids such as vinegar (acetic acid) that contribute to the overall reductions of pathogens. Critical operational parameters for antimicrobial application include the pH, temperature, pressure or flow rate (if applicable), coverage, and contact time. Establishments should be aware that it is not appropriate to add up the results of two separate studies conducted for the same type of intervention (such as two acid dips) because the second time the intervention is used it will likely be less effective. This is because any bacteria that survive the first treatment are likely to be more tolerant to the second treatment.

### ***Drying***

After seasoning and marination, products are typically dried at elevated room temperature so that additional water (moisture) is removed from the product. Products are dried to meet a water activity level sufficient to achieve shelf-stability by preventing the growth of microorganisms (*e.g.*, water activity ≤ 0.85), especially toxigenic microorganisms such as *S. aureus*. Drying to an intermediate or low water activity also may achieve additional reductions in *Salmonella*, STEC (in products containing beef), and *Lm*.

## Appendix 11: Critical Operational Parameters for Drying

Drying (sometimes called maturation for fermented products) is the process during which water (moisture) is removed from the product. Shelf-stable products are dried to meet a water activity level sufficient to prevent the growth of microorganisms, especially toxigenic microorganisms such as *S. aureus*. Drying is typically performed in a drying room to control the critical operational parameters of the process. Drying to an intermediate or low water activity also may achieve additional reductions in *Salmonella*, STEC (in products containing beef), and *Lm* (although *Lm* is more tolerant to drying than *Salmonella*). The lower pH achieved by fermentation aids in drying since the meat proteins are less able to bind water under acidic conditions.

Establishments must ensure that critical operational parameters within the scientific support closely match the establishment's actual process or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Examples of critical operational parameters for reduction of *Salmonella*, STEC, and *Lm* during a drying step include:

- Drying room temperature.
- Drying time.
- Target water activity.
- Product characteristics.

### *Drying Room Temperature*

It is critical that establishments use the same drying room temperature range that is used in the supporting documentation to achieve similar reductions in levels of pathogens of concern. As temperature increases the rate of drying will increase. Although this can result in similar reductions, care must be taken to ensure that meat surfaces do not become too dry while there is still a high moisture content inside the meat pieces (a phenomenon known as **case hardening**). Dry surfaces inhibit the further evaporation of moisture, which may result in products not uniformly dried and in microbiological spoilage starting from the areas where the moisture content remains too high (FAO, 1990). Research has also shown that storage under vacuum for extended time following fermentation and drying is more effective when done under room temperature than refrigeration (Ingham *et al.*, 2004).

### *Drying Time*

Establishments also need to ensure the drying time is consistent with the time used in the scientific supporting documents to reach the desired water activity. If product takes less time to dry, then the drying process will not be as effective as was shown in the support as the longer drying time can increase stress on the bacteria (Gunvig *et al.*, 2017; Mutz *et al.*, 2019). One main issue is that smaller diameter products tend to dry faster than large diameter products, and the shorter drying time is often not enough time to achieve the same reductions in bacteria as larger diameter products even if it is enough time to achieve the targeted water activity (DeSouza, 2018). The use of a shortened drying time for a smaller diameter product was a potential contributing factor in a 2021 salmonellosis outbreak associated with salami sticks (FSIS, 2022). If an

establishment is following a scientific support document performed with one size diameter product and they would like to produce a smaller diameter product, the establishment may consider holding the smaller diameter product for the total drying time needed for the larger diameter product. Ultimately, establishments need to determine the impact any differences between the support and the establishment's actual process would have on the expected reduction. If differences are large, a challenge study may be needed (for more information see Appendix 15).

### *Target Water Activity*

The target water activity must be the same at the end of the drying time as what was used in the supporting documentation or the establishment must provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)). Once the target water activity is reached within the desired time, establishments may continue drying product for a longer amount of time than in the scientific support to achieve a lower water activity (*e.g.,* for shelf-stability). Establishments should not assume because a product is dried to a lower water activity within the same amount of time as the scientific support that similar or higher reductions in pathogen levels will be achieved. For example, when chorizo sausages were dried to target water activity levels and held, the optimal death rate of *Salmonella* and *E. coli* O157:H7 occurred at a level above that of the lowest water activity tested and that "sweet spot" varied by bacteria (Hew *et al.*, 2006).

**NOTE:** Establishments must follow the procedure used to measure water activity in the scientific support or, if using a different procedure, must provide support for the procedure as required by 9 CFR 417.5(a)(2). For larger products, samples should be cut into small pieces. For most products, collecting representative samples from external and internal portions of the product is recommended. However, for thicker non-intact products (e.g., thicker biltong strips made from vacuum-tumbled meat), it may be more appropriate to collect internal samples that would represent a worst-case scenario (Karolenko, 2020).

### *Product Characteristics*: *Product Diameter and Product Composition*

**Product diameter** (thickness) impacts drying time with thicker or larger diameter products taking longer to dry. See drying time discussion above for further considerations.

### *Product Composition*

pH, amount of fat, and particle size also impact the drying rate (Toldra, 2002). Lowered pH increases the drying rate in fermented meats due to the decrease in water-holding capacity when the pH is around the **isoelectric point** of the proteins (Acton and Keller, 1974). Higher fat may result in reduced effectiveness of fermentation and drying (Faith *et al.*, 1998b). Establishments must ensure its product composition is like that of the product studied in the supporting documentation or provide justification for any differences to support the decisions in its hazard analysis (9 CFR 417.5(a)(1)).

For fermented products, the pH at the end of drying may also be a critical operational parameter. The pH most often stays the same or goes up slightly. pH increases occur

likely as a result of **proteolysis** and the production of non-protein nitrogen-containing compounds. In a few cases, pH decreases occur likely as a result of some fermentable sugar remaining and the specific starter culture still being active after fermentation (for example, if there is no low-temperature heat step) (Gunvig, 2016). A reduction in pH after drying has been associated with greater reductions in pathogens (Deibel Laboratories/Chr. Hansen, 2017).

**Question:** ***Why are there so many critical operational parameters during drying? If at the end of my study a certain water activity is reached, why isn't it enough just to know my product gets the same water activity?***

**Answer:** Because smaller diameter products tend to dry faster than large diameter products, and the shorter drying time is often not enough time to achieve the same reductions in bacteria in these products as in larger diameter products, even if it is enough time to achieve the targeted water activity (DeSouza, 2018). In addition to that drying time, drying temperature also plays a role in reducing bacteria.

Here are some citations that support there are other critical operational parameters in addition to water activity at the end of drying:

***DeSouza, J.D., Ahmed, R., Strange, P., Barbut, S., and Balamurugan, S. 2018. Effect of caliber size and fat level on the inactivation of E. coli O157:H7 in dry fermented sausages. Internal Journal of Food Microbiology. 266: 167-172.***

- The authors found that when large diameter sausages reached a water activity of 0.85 after approximately 55 days a 5.0-log reduction in *E. coli* O157:H7 had been achieved. However, when small diameter sausages with the same formulation and fermentation and drying schedule reached 0.85 after approximately 17 days, only a 2.0-3.0-log reduction in *E. coli* O157:H7 was achieved. To get to a 5.0-log reduction, the smaller diameter products had to be dried longer and to a lower water activity. If water activity is considered the only critical operational parameter, then insufficient reductions may be achieved at smaller diameters.

***Porto-Fett, ACS, Hwang, C-A, Call, JE, Juneja, VK, Ingham, SC, Ingham, BH, Luchansky, JB. 2008. Viability of multi-strain mixtures of Listeria monocytogenes, Salmonella typhimurium, or Escherichia coli O157:H7 inoculated into the batter or onto the surface of a soudjouk-style semi-dry sausage. Food Microbiology. 25: 793-801.***

- The authors found that as soudjouk-style sausages (fermented and dried) were stored, *Lm* numbers decreased over time.

***Gunvig, A., Borggaard, C.., Hansen, F., Hansen, T.B., and S. Aabo. 2016. ConFerm – A tool to predict the reduction of pathogens during the production of fermented and matured sausages. Food Control: 67: 9-17.***

- The authors found processing time to be a significant variable for predicting *Lm* reductions in fermented sausages.

These scientific support documents illustrate why meeting the water activity at the end of drying is not enough to show biological hazards (*i.e., Salmonella*, STEC, and *Lm*) are addressed by the fermentation step.

***Other critical operational parameters during drying that do not need to be addressed***

***Casing Type (for fermented products)***

Pore diameter of casing, as well as **casing type,** may impact the drying rate because it influences moisture exchange (Toldra, 2002). However, as described on page 56, several studies have found no differences in reductions for pork salami fermented and dried in natural, collagen, or fibrous casings (DeSouza, 2018; McKinney *et al.*, 2019).

***Presence of Mold***

Molds may be added to the surface of dry and semi-fermented products prior to fermentation to prevent the growth of undesirable molds and for other quality reasons, such as flavor development. Molds are commonly added commercially by dipping or spraying the sausage casing with a mold culture to ensure optimal mold growth/cover. The presence of molds on the external surface may result in more uniform drying with less chance for case hardening. Molds also may impact the drying rate particularly at the surface, resulting in slower drying (Incze, 2010), and they may also impact the pH at the surface of the product. Unless the drying rate/time for a product with mold is different than the drying rate/time for a product without mold, then the presence of mold is not considered a critical operational parameter.

***Relative Humidity***

It is important to maintain the relative humidity within a specified range. If the relative humidity in the chamber is decreased too fast, rapid drying will occur, which will also result in case hardening. When case hardening occurs, the interior moisture is prevented from escaping, which can result in spoilage (Ruhlman and Polcyn, 2013). If the relative humidity is too high, it can lead to undesirable mold growth. Unless there are issues with undesirable mold growth due to high relative humidity and the product weight loss does not meet quality/yield targets, the relative humidity does not need to be addressed.

***Air Flow***

The **air flow** or **speed** (or **air velocity**) at which the air moves through the dryer can be controlled by adjusting the speed at which the fan is operating. The velocity of the air must be sufficiently high to evaporate the moisture from the surface of the food in the dryer and sweep this moisture-rich air out of the dryer. When the heated air passes over the moist surfaces, it picks up water through the process of evaporation. The air then carries the water away from the food and eventually out of the dryer. As the air picks up moisture, it cools and its moisture content increases. This reduces its ability to pick up additional moisture as it continues its path through the rest of the dryer.

Air flows that are too low will not have the desired effect, and air flows too high may increase case hardening. Unless there are issues with undesirable mold growth due to low air flow and the product weight loss does not meet quality/yield targets, the air flow does not need to be addressed.

## Appendix 12: Scientific Support Available for Lethality in Dry and Semi-Dry Fermented Sausages

This guideline focuses on the safe production of fermented sausages, a category which includes such products as Lebanon bologna, summer sausage, pepperoni, salami including Genoa, and soudjouk. Fermented sausages are generally classified as dry or semi-dry sausages. Table 7 includes summaries of processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

### Table 7. Summary of Scientific Support Available for Lethality in Dry and Semi-Dry Fermented Sausages

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Lebanon bologna (beef) | 115 | *Pediococcus*, *Lactobacillus*, and *Micrococcus* spp. | 3.3% salt, 2.9% sugar, 0.8% dextrose, 0.14% potassium nitrate, 0.01% potassium nitrite | Stage 1: 80°F internal temperature for 8 hrs (CUT 5 hrs)<br>Stage 2: 100°F hold for 24 hrs (CUT 4 hrs)<br>Stage 3: 110°F for 24 hrs (CUT 2 hrs) Smoke applied during last two hours of stage 3. | 4.39 | 88 ± 2<br>80 ± 2%<br>80 ± 2 | n/a | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Getty, *et al.*, 1999. J Food Sci. 64(6): 1100-1107. |

[6] Target pH at the end of fermentation.

[7] RH = Relative humidity (although it is a critical operational parameter it does not need to be addressed).

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Lebanon bologna (beef) | 90 | *Pediococcus*, *Lactobacillus*, and *Micrococcus* spp. | 3.3% salt, 2.9% sugar, 0.8% dextrose, 0.14% potassium nitrate, 0.01% potassium nitrite | Stage 1: 80°F internal temperature for 8 hrs (CUT 5 hrs)<br>Stage 2: 100°F hold for 24 hrs (CUT 4 hrs)<br>Stage 3: 110°F for 24 hrs (CUT 2 hrs) | 4.44 | 88 ± 2%<br>80 ± 2<br>80 ± 2 | n/a | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Getty, *et al.*, 1999. J Food Sci. 64(6): 1100-1107. |
| Beef sausage | 105 | *Lactobacillus plantarum* | | 70°F/4 days | ≥5.0 | | 1 hr at 100°F followed by 6 hrs at 125°F | n/a | n/a | n/a | n/a | >2.0-log <5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≤4.6 | | Hold at 90°F for 7 days | n/a | n/a | n/a | n/a | >2.0-log <5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≥5.0 | | Hold at 90°F for 7 days | n/a | n/a | n/a | n/a | >2.0-log <5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 55 | *Pediococcus acidilactici* | | 110°F/0.7 days | ≥5.0 | | Hold at 110°F for 7 days | n/a | n/a | n/a | n/a | >2.0-log <5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 110°F/0.7 days | ≥5.0 | | 1 hour at 100°F, 1 hr at 110°F, 1 hr at 120°F, and ending with 7 hrs at 125°F | n/a | n/a | n/a | n/a | >2.0-log <5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 55 | *Lactobacillus plantarum* | | 70°F/4 days | ≥5.0 | | 1 hr at 100°F followed by 6 hrs at 125°F | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Beef sausage | 55 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≤4.6 | | Hold at 90°F for 7 days | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 55 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≤4.6 | | 1 hr at 100°F followed by 6 hrs at 125°F | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≤4.6 | | 1 hour at 100°F, 1 hr at 110°F, 1 hr at 120°F, and ending with 7 hrs at 125°F | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 90°F/0.5 days | ≥5.0 | | 1 hour at 100°F, 1 hr at 110°F, 1 hr at 120°F, and ending with 7 hrs at 125°F | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 55 | *Pediococcus acidilactici* | | 110°F/0.7 days | ≤4.6 | | Hold at 110°F for 7 days | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 55 | *Pediococcus acidilactici* | | 110°F/0.7 days | ≤4.6 | | Hold at 110°F for 7 days | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef sausage | 105 | *Pediococcus acidilactici* | | 110°F/0.7 days | ≥5.0 | | Hold at 110°F for 7 days | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Blue Ribbon 1996 |
| Beef summer sausage | 66 | *Pediococcus acidilactici* | 2.5% salt, 0.3% dextrose, 0.26% curing salt (6.25% sodium nitrite), 0.054% sodium erythorbate | 1 hr at 86°F<br>1 hr at 90°F<br>1 hr at 95°F<br>1 hr at 100°F<br>1 hr at 105°F | 5.0 | 80 | 130°F | n/a | n/a | n/a | n/a | >5.0-log in *E. coli* O157:H7 | Calicioglu, *et al.*, 1997. J. Food Prot. 60(1): 1158-1162. |

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Pepperoni (beef and pork) | 55 | *Pediococcus acidilactici* | 3.33% salt, 0.63% dextrose, 2.0% cure mixture (156ppm $NaNO_2$), spice mixture (red pepper, clove, anise, garlic, oleoresin of paprika, BHA, BHT, citric acid). | 96°F/14 to 18 hr | ≤5.0 | 85 | 145°F internal or 128°F for 1 hr | 55°F /18 days | 65 | 0.9 | n/a | >5.0-log in *E. coli* O157:H7 | Hinkens, *et al.*, 1996. J. Food Prot. 59: 1260-1266. |
| Pepperoni (beef and pork) | 55 | *Pediococcus acidilactici* | 0.63% dextrose, 2.0% cure mixture (156ppm NaNO2), and 3% spice mixture | 96.8°F/16 to 20 hours | ≤4.8 | 92 | n/a | 55.4/18 days | 65 | 0.88 | Storage at 69.8 for 58 to 60 days | >5.0-log in *E. coli* O157:H7 | Ihnot *et al.*, 1998. J Food Micro. 40: 117-121. and Faith *et al.*, 1997. J. of Food Microbiology. 37: 47-54. |
| Genoa salami (pork) | 65 | *Pediococcus acidilactici, Pediococcus pentosaceous*, and *Kocuria varians* | 2.9% salt, 1% dextrose, 0.08% white pepper, 0.05% sodium ascorbate, 0.02% garlic powder, 0.015% sodium nitrate, and 0.005% sodium nitrite | 68°F/6 hrs 80.6°F/26 hrs | 4.58 | 90-95% | n/a | 68/40 hrs 62.6 until 0.92 (time not reported) | 65-75 | 0.92 | n/a | >2.0-log <5.0-log in *Salmonella* and >1.0-log <2.0-log in *E. coli* O157:H7 and >.01-log <2.0-log in *Lm* | Porto-Fett, *et al*., 2010. Int'l J. of Food Micro. 140: 61-75.[8] |

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Genoa salami (pork) | 105 | *Pediococcus acidilactici*, *Pediococcus pentosaceous*, and *Kocuria varians* | 2.9% salt, 1% dextrose, 0.08% white pepper, 0.05% sodium ascorbate, 0.02% garlic powder, 0.015% sodium nitrate, and 0.005% sodium nitrite | 68°F/6 hrs 80.6°F/26 hrs | 4.58 | 90-95 | n/a | 68/40 hrs 62.6 for 35 days | 65-75 | 0.92 | n/a | >2.0-log <5.0-log in *Salmonella* and >1.0-log <2.0-log in *E. coli* O157:H7 and >1.0-log <2.0-log in *Lm* | Porto-Fett, *et al.*, 2010. Int'l J. of Food Micro. 140: 61-75.[8] |
| Genoa salami (pork) | 65 | *Pediococcus acidilactici, Pediococcus pentosaceous*, and *Kocuria varians* | 2.9% salt, 1% dextrose, 0.08% white pepper, 0.05% sodium ascorbate, 0.02% garlic powder, 0.015% sodium nitrate, and 0.005% sodium nitrite | 68°F/6 hrs 80.6°F/26 hrs | 4.58 | 90-95% | n/a | 68°F /40 hrs 62.6 for 25 days | 65-75 | 0.88 | n/a | >2.0-log <5.0-log in *Salmonella* and >1.0-log <2.0-log in *E. coli* O157:H7 and >1.0-log <2.0-log in *Lm* | Porto-Fett, *et al.*, 2010. Int'l J. of Food Micro. 140: 61-75.[8] |
| Genoa salami (pork) | 105 | *Pediococcus acidilactici*, *Pediococcus pentosaceous*, and *Kocuria varians* | 2.9% salt, 1% dextrose, 0.08% white pepper, 0.05% sodium ascorbate, 0.02% garlic powder, 0.015% sodium nitrate, and 0.005% sodium nitrite | 68°F/6 hrs 80.6°F/26 hrs | 4.58 | 90-95 | n/a | 68/40 hrs 62.6 for 22 days | 65-75 | 0.94 | n/a | >2.0-log <5.0-log in *Salmonella* and >1.0-log <2.0-log in *E. coli* O157:H7 and >1.0-log <2.0-log in *Lm* | Porto-Fett, *et al.*, 2010. Int'l J. of Food Micro. 140: 61-75.[8] |

[8] Additional log reductions were achieved by use of high pressure processing (HPP) as described in the article.

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Soudjouk-style semi-dry sausage | 25mm, flattened to 11.4 cm w X 22.9 L (thickness not reported) | Bacteriocin-producing pediococcal starter culture | dextrose (0.60%), salt (1.9%), sodium nitrite (0.25% or 156 ppm), chopped fresh garlic (0.95%), and various spices (cumin (0.95%), paprika (0.42%), black pepper (0.42%), and all-spice (0.42%). | 75.2°F/72 hours | 4.8 | 90-95 | n/a | 71.6/72 hours | 80-85 | 0.915 | Stored at 69.8°F under vacuum for 30 days | >5.0-log in *Salmonella* and >2.0-log <5.0-log in *E. coli* O157:H7 and *Lm* | Porto-Fett, *et al.*, 2008. Food Micro. 25: 793-801. |
| Soudjouk-style semi-dry sausage | 3.65cm natural pork casings, after drying flattened to 1.25cm | *Pediococcus acidilactici* | dextrose (1%), sodium chloride (2%) and 4.7% spice mix (house blend, Kayseri Basterma, Inc., Poughkeepsie, N.Y.). Neither nitrate nor nitrite were used. | 71.6°F/72 hours | 4.9 | 50 | n/a (cooking caused unacceptable quality) | 48.2°F/18 hours | 40 | not reported | Conditioned at 71.6°F/50 % RH for 72 hours then stored under vacuum at 77°F for 21 days. | > 5.0-log in *E. coli* O157:H7 | Calicioglu *et al.*, 2002. J. Food Prot. 65: 1541-1544.[9] |

[9] Additional conditions including cooking were evaluated.

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Soudjouk-style semi-dry sausage | not reported | *Pediococcus acidilactici* and *Lactobacillus curvatus* | dextrose (1.5%), sodium chloride (1.9%), sodium nitrite (0.25% or 156 ppm), chopped fresh garlic (0.95%), and various spices (cumin (0.95%), paprika (0.42%), black pepper (0.42%), and all-spice (0.42%). | 75.2°F/72 hours | 4.55 | 90-95 | n/a | 71.6°F/72 hours | 80-85 | not reported | storage at 77°F for 14 days | >5.0-log in *E. coli* O157:H7 | Calicioglu *et al.*, 2001. J. Food Prot. 64: 1156-1161.[9] |
| Italian-style salami (pork) | 100 | *Pediococcus acidilactici* and *Staphylococcus carnosus* | Nitrite, nitrate, spices, and salt dextrose.<br><br>NOTE: Concentrations were not reported. | 104°F/24 hours | ≤4.8 | >95 | n/a | 55.4°F/30 days | not reported | ≥0.92 | n/a | >5.0-log reduction in *Salmonella*[10] | IEH. Evaluation of Process Parameters Used During the Fermentation and Drying of Italian-Style Salami. |
| Italian-style salami (pork) | 100 | *Pediococcus acidilactici* and *Staphylococcus carnosus* | Nitrite, nitrate, spices, and salt dextrose.<br><br>NOTE: Concentrations were not reported. | 104°F/24 hours | ≤4.8 | >95 | n/a | 59°F/30 days | not reported | ≥0.92 | n/a | >5.0-log reduction in *Salmonella*[10] | IEH. Evaluation of Process Parameters Used During the Fermentation and Drying of Italian-Style Salami. |

[10] The amount of non-meat ingredients used was not included and none of the trials within this study were repeated. Therefore, this study should not be used alone.

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Salami | 78 | BLC-007 | .57% dextrose, 0.13% white pepper, 0.19% black pepper, 0.014% sodium nitrite, and 0.005% sodium nitrate | 88-92°F (31-33°C) for 28 hours | < 4.9 | 90 | n/a | 67-70°F (19-21°C) for 24 hours and then at 56-59°F (13-15°C) for 29 days | 84-89<br><br>75-90 | < 0.92 | n/a | ≥ 5.0-$\log_{10}$ in *Salmonella*, *E. coli* O157:H7, and *Lm* | Deibel Laboratories/ CHR. Hansen. 2017. |
| Salami | 50 | BLC-007 | Salt, nitrite, spice mix<br><br>NOTE: Links were dipped in hydrated *Penicillium nalgiovense* spore solution | 90°F (32°C) for 24 hours | 4.5 | 95 | n/a | 59°F (15°C) for 31 days | 72 | 0.74 (after drying and finishing stage/60 days) | Finishing phase of 30 day storage at 70°F (21°C) with 50% RH | ≥ 5.0-$\log_{10}$ in *Salmonella and* 4.9-$\log_{10}$ in *Lm*[11] | Hussein *et al.,* 2022. Food Control. 131: 108432. |
| Salami | 80 | BLC-007 | Salt, nitrite, spice mix<br><br>NOTE: Links were dipped in hydrated *Penicillium nalgiovense* spore solution | 90°F (32°C) for 24 hours | 4.5 | 95 | n/a | 59°F (15°C) for 60 days | 72 | 0.83 (after drying and finishing stage/80 days) | Finishing phase of 20 day storage at 70°F (21°C) with 50% RH | ≥ 5.0-$\log_{10}$ in *Salmonella and* 4.9-$\log_{10}$ in *Lm*[11] | Hussein *et al.,* 2022. Food Control. 131: 108432 |

[11] Although the article does not demonstrate a 5.0-log reduction for *Lm*, FSIS would not object to its use as supporting documentation because the article demonstrated a 4.9-log reduction in *Lm* after the finishing phase.

| Product | Dia-meter (mm) | Starter Culture | Formulation | Fermentation Temperature (°F) + Time | pH[6] | RH[7] (%) | Low-Temperature Heat Step | Drying Room Temperature (°F) + Time | RH (%) | Final Water Activity | Additional Interven-tions | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Landjäger | Not reported, product was pressed | BLC-007 | 0.6% dextrose, 1.63% red wine, 2.93% salt, 0.32% black pepper, 0.16 granulated garlic, 156ppm nitrite | 74°F for 72 hours | < 4.8 | 61 | After fermentation sausages were smoked with hickory sawdust for 2 hours at 87°F (30°C). | 71°F (21.6°C) 3 days | 60 | < 0.88 | Raw trim was sprayed with 4.5% lactic acid (77°F/25°C ) for 45s and held for 30 min.<br><br>Stored under vacuum for 20 days at 73.4F (23°C) | ≥ 5.0-$\log_{10}$ in *Salmonella*, *E. coli* O157:H7, and *Lm* | Rivera-Reyes *et al.*, 2017. Food Control. 73: 767-774. |

## Appendix 13. Scientific Support Available for Lethality in Salt-Cured Products

### Basturma

Basturma is a traditional Turkish dry-cured beef product made from whole muscle. Basturma is known by different names depending on geographic location: pastirma, bastirma, pasterma, basterma. To make basturma, meat is dry-cured, dried, pressed and coated with çemen. Çemen (cement) is a paste made of spices and flavorings such as crushed cumin, fenugreek, garlic, and paprika (paste seasoning). Table 8 includes summaries of processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

**Table 8. Summary of Scientific Support Available for Lethality in Basturma.**

| Product | Source materials | Formulation | Dry-curing | Drying Room Temperature (°F) + Time + Relative Humidity (RH) | Second drying Step Room Temperature (°F) + Time + RH | Spice Paste and Third Drying Room Temperature (°F) + Time + RH | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|
| Basturma | Beef rounds | 6.0 kg of curing mixture per approximately 45.4 kg of meat. Curing mixture contained proprietary amounts of salt, sucrose, glucose, and sodium nitrite.[12] | Rounds were placed in plastic lugs in an alternating manner. Lugs were stored at 44°F (6.7°C), 50% RH, for 7 days, then, fluid was drained and rounds were manually dry-rubbed again with a second batch of curing mixture and allowed to cure for 14 more days. | Rounds were rinsed for 1 hour with tap water.<br><br>Dried at 70°F for 2 days<br><br>12 hours at 65% RH and 12 hours at 80% RH | Rounds were pressed for 12 hours under refrigeration then dried at 70°F for 4 days.<br><br>RH cycled 12 hours at 65% and 12 hours at 80% | Coated with a spice paste, and dried 70°F for 4 days<br><br>12 hours at 65% RH and 12 hours at 80% RH | Water activity: 0.87<br><br>MPR: 1.92: 1.00<br><br>pH: 5.6<br><br>% water-phase salt: 13.1 | >5.0-log in *E. coli* O157:H7 and *Salmonella* and ≥2.0-log <5.0-log in *Lm*[13] | Ingham, *et al.*, 2006. J Food Safety. 26: 160-172.<br><br>University of Wisconsin's Status Summary |

[12] Establishments wanting to use this reference should contact the authors for information on the formulation.

[13] None of the trials within this study were repeated. So, establishments should provide additional supporting documentation for the processes effectiveness.

| Product | Source mater-ials | Formulation | Dry-curing | Drying Room Temperature (°F) + Time + Relative Humidity (RH) | Second drying Step Room Temperature (°F) + Time + RH | Spice Paste and Third Drying Room Temperature (°F) + Time + RH | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|
| Basturma | Beef rounds | 6.0 kg of curing mixture per approximately 45.4 kg of meat. Curing mixture contained proprietary amounts of salt, sucrose, glucose, and sodium nitrite.[14] | Rounds were placed in plastic lugs in an alternating manner. Lugs were stored at 44°F (6.7°C), 50% RH, for 7 days, then, fluid was drained and rounds were manually dry-rubbed again with a second batch of curing mixture and allowed to cure for 14 more days. | Rounds were rinsed for 1 hour with tap water.<br><br>Dried at 75°F for 2 days<br><br>12 hours at 65% RH and 12 hours at 80% RH | Rounds were pressed for 12 hours under refrigeration then dried at 75°F for 4 days<br><br>Cycled 12 hours at 65% RH and 12 hours at 80% RH | Coated with a spice paste, and dried 75°F for 5 days<br><br>12 hours at 65% RH and 12 hours at 80% RH | Water activity: 0.95<br><br>MPR: 2.00: 1.00<br><br>pH: 6.0<br><br>% water-phase salt: 8.3 | >5.0-log in *E. coli* O157:H7 and *Salmonella* and ≥2.0-log <5.0-log in *Lm*[1] | Ingham, *et al.*, 2006. J Food Safety. 26: 160-172.<br><br>University of Wisconsin's Status Summary |
| Basturma | Beef rounds | 6.0 kg of curing mixture per approximately 45.4 kg of meat. Curing mixture contained proprietary amounts of salt, sucrose, glucose, and sodium nitrite.[15] | Rounds were placed in plastic lugs in an alternating manner. Lugs were stored at 44°F (6.7°C) , 50% RH, for 7 days, then, fluid was drained and rounds were manually dry-rubbed again with a second batch of curing mixture and allowed to cure for 14 more days. | Rounds were rinsed for 1 hour with tap water.<br><br>Dried at 81°F for 2 days<br><br>Constant 70% | Rounds were pressed for 12 hours under refrigeration then dried at 81°F for 4 days<br><br>Constant 70% | Coated with a spice paste, and dried 81°F for 6 days<br>Constant 70% | Water activity: 0.84<br><br>MPR: 1.52: 1.00<br><br>pH: 5.6<br><br>% water-phase salt: 18 | >5.0-log in *E. coli* O157:H7 and *Salmonella* and ≥2.0-log <5.0-log in *Lm*[1] | Ingham, *et al.*, 2006. J Food Safety. 26: 160-172.<br><br>University of Wisconsin's Status Summary |

[14] Establishments wanting to use this reference should contact the authors for information on the formulation.

[15] Establishments wanting to use this reference should contact the authors for information on the formulation.

| Product | Source mater-ials | Formulation | Dry-curing | Drying Room Temperature (°F) + Time + Relative Humidity (RH) | Second drying Step Room Temperature (°F) + Time + RH | Spice Paste and Third Drying Room Temperature (°F) + Time + RH | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|---|---|
| Basturma | Beef eye round whole or cut longitu dinally in half | Injected with 10% (weight/weight) brine made of 25% salt and 0.2% sodium nitrite. After injection dry salt added at levels of 4.7-10% (w/w) | 40°F for up to 6 days with frequent rotation. | Muscles hung individually in smokehouse and heated at an oven temperature of ≥ 149°F for at least 6 hours during which internal meat temperature reached ≥ 127 °F. | 68-77°F for 4 more days | Mixture of spices (flour, fenugreek, pepper, cumin, garlic, paprika, and food color) in water.<br>Dried at 68-77°F for 4 more days | | ≥5.0-log in *Salmonella* | Genigeorgis and Lindroth. 1984. European Meeting of Meat Research Workers. 217-224. |

## Country Cured Ham

Country cured ham is the dry cured hind leg of a pig. FSIS has a standard of identity for country ham in 9 CFR 319.106. The production process for country cured hams is similar to prosciutto and involves the three steps of curing, equalization, and drying. Table 9 contains a summary of processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

**Table 9. Summary of Scientific Support Available for Lethality in Country Cured Ham.**

| Product | Formulation | Cure application | Dry-curing | Equalization | Drying | Log Reduction | Finished Product Characteristics | Reference |
|---|---|---|---|---|---|---|---|---|
| Country cured ham | 1) 3.63 kg salt, 454 g sugar, 14.2 g sodium nitrite, 56.7 g sodium nitrate<br><br>OR<br><br>2) 3.63 kg salt and 454 g sugar | Cures were applied at a ratio of 42.53 g of cure mixture per 0.45 kg of ham, with a half portion each of this cure mixture applied in 2 separate hand rubs, on day 0 and day 10. | Hams were cured at 40°F (4.4°C) during dry salt curing (35 d), after which the excess salt was brushed off with no water added. | Hams were placed in stockettes and held at 40°F (4.4°C) for 14 d for salt equalization. | Hams were dry aged for 20 d at 85°F (29.4°C) (65% relative humidity) to meet *Trichinae* requirements in 9 CFR 318.10.[16] | ≥5.0-log in *E. coli* O157:H7 and *Salmonella* and ≥4-log in *Lm* when cured for 69 days.<br><br>Research also validated process resulted in sufficient control of *S. aureus* growth. | Finished product day = 120<br>Salt = 8.0%<br>water activity = 0.91<br>pH = 5.5<br>Brine concentration = 13.81% and 12.33% for cure mix 1 and cure mix 2, respectively. | Reynolds, *et al.*, 2001. Journal of Food Science. 66: 1373-1379. |

[16] Dry curing process was considered complete at this point (d 69). At d 69, hams were placed in ambient 68 to 75.2°F (20 to 24 °C) storage through d 120. Sufficient reduction of pathogens was achieved after d 69.

## Bresaola

Bresaola is traditionally produced with whole beef muscles, such as eye of round and inside round. These cuts are typically dry-cured with salt and spices, allowed to equalize, and then stuffed into casings and hung to dry (Watson *et al.,* 2021). Table 10 contains a summary of processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

**Table 10. Summary of Scientific Support Available for Lethality in Bresaola.**

| Product | Formulation | Cure application | Dry-curing | Equalization | Drying | Log Reduction | Finished Product Characteristics | Reference |
|---|---|---|---|---|---|---|---|---|
| Bresaola | Whole muscle intact beef rounds were sprayed with Beefxide (2.5% vol/vol) using a handheld sprayer for 15-20 sec on each side to coat all surfaces. A proprietary blend of salt (3.5% of meat weight) and cure ingredients (150ppm sodium nitrite and 100 ppm sodium nitrate). | The eye of round subprimals were cured by applying half of the cure mixture before dry-curing, after 7 days the subprimals were coated with the remaining half of cure mixture. | After being coated with dry ingredients, beef was placed in a meat lug at 39°F (4°C) for 7 days before being coated with the remaining half of the cure mixture. | The eye of round remained at 39°F (4°C) for 4 weeks. | Bresaola was stuffed into beef bung casings (115-130mm) and were dry aged at 54 to 57°F (12-14°C) (65-75% relative humidity) for 65 days. | ≥5.0-log in *E. coli* O157:H7, *Salmonella* and *Lm* when cured for 65 days.<br><br>Results were calculated on the surface and therefore, should only be applied when using whole muscle intact beef source materials. | Water activity = 0.88 | Watson, *et al.*, 2021. Meat and Muscle Biology. 5(1): 14, 1-8. |

## Appendix 14: Scientific Support Available for Lethality in Dried Products

### Droëwors

Droëwors is a dried beef sausage product developed in South Africa that can be made from various meat species (typically beef or game). Droëwors is made from ground meat stuffed into casings after being mixed with vinegar and seasoning blends. Table 11 includes a summary of the processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

**Table 11. Summary of Scientific Support Available for Lethality in Droëwers.**

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Droëwers | Grind size: Trimmings were ground twice, first through a plate with 10 mm size holes and then through a plate with 4mm size holes. | Vinegar/spices: Ground meat was stuffed into vacuum packed bags and proprietary amounts of vinegar and a spice blend were added. Then the mix was stuffed into lamb casings.[17] | Droewors sausage was hung on racks in an environmental chamber set to 72°F (actual range 68-71.6°F) with a target constant of 50% relative humidity (actual range 38-64%). Sausages were dried for 12-21 days. | After reaching a water activity of 0.60, sausages were vacuum packaged and stored at 68-71.6°F for 7 days. | Trial 1<br>pH: 5.5<br>water activity: 0.62<br>% water-phase salt: 19.1%<br><br>Trial 2<br>pH: 5.4<br>water activity: 0.60<br>% water-phase salt: 19.6%<br><br>Trial 3<br>pH: 5.4<br>water activity: 0.60<br>% water-phase salt: 22.2% | ≥2.0-log and < 5.0-log reduction in *E. coli* O157:H7 and *Salmonella* and a 1.5 to 2.7-log reduction in *Lm*.<br><br>Research also validated process resulted in sufficient control of *S. aureus* growth. | Burnham, *et al.*, 2008. Journal of Food Safety. 28: 198-209. |

[17] Establishments wanting to use this reference should contact the authors for information on their vinegar and spice formulation or should ensure that the pH and % water phase-salt reported match those listed in the "finished product characteristics" column.

## Biltong

Biltong is a dried beef product developed in South Africa. Traditionally, biltong is dried under ambient conditions; however, to achieve adequate lethality commercially produced biltong should be produced under controlled conditions. Biltong may be produced from various species (e.g., beef, poultry, venison, or game). In the U.S. biltong is most often made from strips of beef that are trimmed, salted, and dried. Consumption of biltong has been linked to at least one outbreak from *Salmonella* outside of the U.S. and in a 2008 outbreak, one case patient was an infant who was given the product for teething (Mindlin *et. al*, 2013). The intended use of biltong is typically considered to be RTE because consumers commonly consume biltong without further preparation for safety. Table 12 includes summaries of processes and the critical operational parameters of some of the processes that have been found to achieve lethality of pathogens. Establishments are not limited to using these articles as support, and the summaries are not adequate support on their own because they do not contain the details of each study and the establishment needs to determine if it is representative of the actual process. For this reason, establishments will need to have the full copy of the article on-file. Links to full copies of articles have been provided in the References section when available.

**Table 12. Summary of Scientific Support Available for Lethality in Biltong.**

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong | Thickness: Biltong strips were obtained from intact beef bottom round sub-primal cuts which were trimmed of external flat, squared off to produce a uniform shape, and sliced to 2.5 cm thickness. | Vinegar/spices: Proprietary amounts of vinegar and spice blend. Tumbled for 30 minutes.[18] | Biltong strips were hung on racks in an environmental chamber set to 72°F (actual range 68-71.6°F) with a target of 50% relative humidity (actual range 38-64%). Strips were dried for 17-26 days. | After reaching a water activity of 0.60, strips were vacuum packaged and stored at 68-71.6°F for 7 days. | Trial 1<br>pH: 5.6<br>water activity: 0.75<br>% water-phase salt: 15.4%<br><br>Trial 2<br>pH: 5.6<br>water activity: 0.75<br>% water-phase salt: 15.8%<br><br>Trial 3<br>pH: 5.5<br>water activity: 0.62<br>% water-phase salt: 21.5% | ≥ 2.0-log reduction and < 5.0-log reduction for *Salmonella and E. coli* O157:H7 and ≥ 1.0-log reduction and ≤ 4.0-log reduction for *Lm*. Also validated the process resulted in sufficient control of *S. aureus* growth. | Burnham, *et al.*, 2008. Journal of Food Safety. 28: 198-209. |

[18] Establishments wanting to use this reference should contact the authors for information on their vinegar and spice formulation or should ensure that the pH and % water phase-salt reported match the actual process.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong (tradit-ional method) | Beef steak portions (30cm length x 15cm width x 2.5cm thickness). | Marinated in undiluted apple cider vinegar (100 ml) for 30 seconds per side. Excessive vinegar was allowed to drip off and 16 grams biltong spice (predominantly consisting of black pepper, coriander, salt and brown sugar) was spread onto each side of the meat. Pieces were then marinated for 20 hours under refrigeration (40°F). | Meat pieces were dried in a Biltong buddy home-dryer fitted with a 40W light bulb that generated a constant temperature of 77°F. To achieve adequate reductions pieces were dried for 84 hours. Air flow and other critical operational parameters of the dryer were not reported.[19] | n/a | Not reported. | ≥ 5.0-log reduction for *Lm*.[20]<br><br>The research also validated the process resulted in sufficient control of *S. aureus* growth. | Naidoo, *et al.*, 2010. Food Control. 21: 1042-1050. |

[19] Critical operational parameters used in the study including the temperature were not reported. Therefore, establishments should provide additional supporting documentation for the effectiveness.

[20] Establishments should provide additional supporting documentation for the effectiveness because only one strain of *Lm* was used and no history or justification was given for why the particular *Lm* strain was chosen.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong (modern method) | Beef steak portions (30cm length x 15cm width x 2.5cm thickness) | 16 grams biltong spice (predominantly consisting of black pepper, coriander, salt and brown sugar) was combined with 16ml apple cider vinegar (1:1 g/ml ratio) and was spread onto each side of the meat and sealed and shaken for 1 minute to ensure full coverage. Pieces were then marinated for 20 hours under refrigeration (40°F). | Meat pieces were dried in a Biltong buddy home-dryer fitted with a 40W light bulb that generated a constant temperature of 77°F. To achieve adequate reductions pieces were dried for 96 hours. Air flow and other critical operational parameters of the dryer were not reported.[21] | n/a | Not reported. | ≥ 5.0-log reduction for *Lm*.[22]<br><br>The research also validated the process resulted in sufficient control of *S. aureus* growth. | Naidoo, *et al.*, 2010. Food Control. 21: 1042-1050. |

[21] Critical operational parameters used in the study including temperature were not reported. Therefore, establishments should provide additional supporting documentation for the effectiveness.

[22] Establishments should provide additional supporting documentation for the effectiveness because only one strain of *Lm* was used and no history or justification was given for why the particular *Lm* strain was chosen.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong | Boneless beef round was trimmed to remove excess fat and trimmed to 1.9 cm thick X 5.1 cm wide X 7.6cm long. | Raw beef strips were dipped in acidified calcium sulfate (Mionix RTE-01) adjusted to 10% lactic acid for 30 seconds.<br><br>Strips were then tumble-marinaded for 30 minutes under vacuum in a marinade containing salt (2% of formulation) and vinegar (2% formulation). | 77°F (25°C) and 55% RH for at least **4 days.** | n/a | 0.90 water activity[23] | ≥ 5.0-log reduction for *Salmonella* | Karolenko, *et al*., 2020. Microorganisms. 8(5): 791. |

[23] Results for each trial were not reported. Water activity estimated based on one trial reported in Figure 7. Product may need to be dried further for shelf-stability.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong | Boneless beef round was trimmed to remove excess fat and trimmed to 1.9 cm thick X 5.1 cm wide X 7.6cm long. | Raw beef strips were dipped in acidified calcium sulfate (Mionix RTE-17) adjusted to 5% lactic acid for 30 or 60 seconds or 5% lactic acid for 60 seconds.<br><br>Strips were then tumble-marinaded for 30 minutes under vacuum in a marinade containing salt (2% of formulation) and vinegar (2% formulation). | 77°F (25°C) and 55% RH for at least **6 days.** | | 0.85 water activity[24] | ≥ 5.0-log reduction for *Salmonella* | Karolenko, *et al*., 2020. Microorganisms. 8(5): 791. |

[24] Results for each trial were not reported. Water activity estimated based on one trial reported in Figure 7.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong | Boneless beef round was trimmed to remove excess fat and trimmed to 1.9 cm thick X 5.1 cm wide X 7.6cm long. | Raw beef strips were dipped in 5% lactic acid or 3% sodium acid sulfate for 30 seconds.<br><br>Strips were then tumble-marinaded for 40 minutes under vacuum in a marinade containing salt (2% of formulation) and vinegar (2% formulation).<br><br>Marinaded beef was held overnight (16-18hours) at 41°F (5°C). | 77°F (25°C) and 55% RH for at least **4 days** | | 0.90 water activity[25] | ≥ 5.0-log reduction for *Salmonella* | Karolenko, *et al*., 2020. Microorganisms. 8(5): 791. |

[25] Results for each trial were not reported. Water activity estimated based on one trial reported in Figure 7. Product may need to be dried further for shelf-stability.

| Product | Product Characteristics | Formulation | Drying | Post-drying Storage | Finished Product Characteristics | Log Reduction | Reference |
|---|---|---|---|---|---|---|---|
| Biltong | Boneless beef round was trimmed to remove excess fat and trimmed to 1.9 cm thick X 5.1 cm wide X 7.6cm long. | Raw beef strips were dipped in water for 30 seconds.<br><br>Beef strips were tumbled (no vacuum) for 5 min with spices (95-96% beef, 4-5% spice which included salt at 2.1% total formulation). Dry spiced-beef strips were placed in stainless steel pans and liquid marinade was slowly poured over (vinegar comprised 73% liquid marinade and 10% total formulated weight). Marinaded pieces were held at 41°F (5°C) and turned after 30 min and 8-12 hours (total time 16-20 hours). | 73°F (22.8°C) and 55% RH for at least **6 days** | | 0.85 water activity[26] | ≥ 5.0-log reduction for *Salmonella* | Karolenko, *et al*., 2020. Microorganisms. 8(5): 791. |

[26] Results for each trial were not reported. Water activity estimated based on one trial reported in Figure 7.

## Appendix 15: Designing Challenge Studies for Fermented, Salt-Cured, and Dried Products

If no literature is available or the process or method an establishment uses is significantly different than used in literature, establishments may decide to conduct a **challenge study**. When establishments want to use unique processes, a challenge study may be needed to demonstrate the safety of the process. Before conducting a challenge study, establishments may consider reaching out to the HACCP Coordinator in their state who can provide technical advice, assistance, and resources to support HACCP implementation in small and very small plants.

For general guidance on conducting challenge studies see the FSIS HACCP Systems Validation Guideline as well the article, Parameters for Determining Inoculated Pack/Challenge Study Protocols, published by the National Advisory Committee on Microbiological Criteria for Foods (NACMCF) in the Journal of Food Protection in 2010. For guidance on how to select a microbiological testing laboratory please review FSIS' Establishment Guidance for the Selection of a Commercial or Private Microbiological Testing Laboratory. Specific considerations related to fermented, salt-cured, and dried meat and poultry products that are not addressed in other guidance are included below.

A well-designed and documented challenge study should include details about:

- Product studied including formulation information.
- The types and number of strains of microorganisms.
- Methods of production, enumeration, and standardization of inoculum.
- Size of the inoculum to be used.
- Method of inoculation.
- Sample size, sampling time, and number of samples to test.
- Methods of microbial analysis.
- Number of replicates.
- Measurement of critical operational parameters and intrinsic factors at each key stage of production.

### *Product Studied Including Formulation Information*

The study should clearly identify the type of product studied as well as details about the raw meat or poultry components used (*e.g.*, biltong strips were obtained from intact beef bottom round sub-primal cuts which were trimmed of external flat, squared off to produce a uniform shape, and sliced to 2.5 cm thickness).

The challenge study should identify all ingredients used in the product formulation which may affect the inactivation or growth of bacterial pathogens in or on the product to include ingoing levels of sodium nitrite, phosphate, preservatives, and any antimicrobial agents (*e.g.*, organic acid salts).

### *The Types and Number of Strains of Microorganisms*

The types and number of strains of microorganisms used as an inoculum should be included in the protocol as well as an explanation of why the strains (including surrogates) were chosen.

If an establishment chooses to conduct a challenge study in a **laboratory**, the study should use at least five strains of the pathogenic microorganisms of concern (*e.g.*, five strains of *Salmonella*, five of STEC, and five of *Lm*) including strains associated with human foodborne illness and strains isolated from meat and poultry products.

If an establishment chooses to conduct a challenge study in a **plant environment** to best represent actual processing conditions, the establishment should choose non-pathogenic surrogate organisms that have been found to respond similarly to the pathogens of interest (*e.g.*, *Salmonella*, *S. aureus*, etc.) under the mode of lethality being evaluated (*e.g.*, fermentation, salt-curing, or drying). See Table 13 for examples.

FSIS recommends establishments target the same reductions in surrogates as they would for pathogens (*e.g.,* a 5-log reduction for surrogates found to respond similarly to *Salmonella* and STEC and a 3-log reduction for surrogates found to respond similarly to *Lm*). It may be possible for establishments to use adjustment factors to account for differences in tolerance to lethality treatments between a surrogate and pathogen. However, using adjustment factors is not recommended by FSIS unless the establishment also has performed a challenge study in the laboratory to support the effectiveness of the lethality treatment in reducing pathogens under laboratory conditions.

An establishment that chooses to do a challenge study may use a surrogate organism to measure change, but it should do so only after giving careful consideration to specific precautions. These precautions include ensuring that a microbiologist trained in food science and in the design of inoculated-pack studies introduces the non-pathogenic cultures within the establishment. In addition, the establishment should ensure that the introduction of the non-pathogenic cultures does not create an insanitary condition in the facility or cause the food to become adulterated. Whenever possible, the establishment should also avoid the use of bacterial surrogate strains that have been 'marked' using antibiotic resistant genes. Finally, establishments should ensure that the non-pathogenic cultures are necessary and proven to be effective for the intended purpose.

To better ensure that insanitary conditions are not created, establishments are encouraged to apply surrogate indicator organism cultures in a manner to ensure that the establishment can conduct a full cleaning and sanitizing of the facility and equipment after the stage in the food safety application being evaluated. Generally, product containing the surrogate indicator organism cultures would not automatically be considered adulterated.

Additional information on the strengths and limitations for using a particular surrogate indicator organism is provided in scientific research and should be considered.

**Table 13. Potential surrogates for lethality challenge studies conducted in-plant.**

| If you produce… | the following surrogate(s) have been validated… | to respond similarly to… | for the following processing step(s)… | as supported by…[27] |
|---|---|---|---|---|
| Summer sausage and biltong | Five American Type Culture Collection (ATCC) non-pathogenic *E. coli*:<br>• BAA-1427<br>• BAA1428<br>• BAA-1429<br>• BAA-1430<br>• BAA-1431 | *Salmonella, E. coli* O157:H7<br><br>For biltong, also *Lm* and *S.aureus* | Fermentation [28] | Kneeling *et al.*, 2009<br>Niebuhr *et al.*, 2008<br>Karolenko *et al.*, 2022<br>https://askusdacontactcenter.force.com/s/article/Use-of-Non-pathogenic-Escherichia-coli-E-coli-Cultures-as-Surrogate-Indicator-Organisms-in-Validation-Studies |
| Fermented and dried salami, Dry-cured whole muscle pork products | *Enterococcus faecium* | *Salmonella* | Fermentation, drying, and dry-curing[29]<br><br>See NOTE below | Ma *et al.*, 2007<br>Michet, 2015<br>Guidelines for Using *Enterococcus faecium* as a Surrogate Microorganism in Almond Process Validation |
| Whole-muscle and ground and formed jerky, Biltong, Droëwors | Saga 200 (Lactic acid bacteria – *Pediococcus acidilactici*) | *Salmonella, E. coli O157:H7, Lm,* and *S. aureus* | Antimicrobial treatment and drying<br>See NOTE below | Borowski *et al.*, 2009<br>Dierschke *et al.*, 2010a<br>Dierschke *et al.*, 2010b |
| Biltong | *Carnobacterium spp.* | *Salmonella, E. coli O157:H7, Lm,* and *S. aureus* | Antimicrobial treatment and Drying | Karolenko et al., 2022 |

**NOTE:** Research by Karolenko *et al.,* 2022 showed *Enterococcus faecium* and Saga 200 to be much more tolerant to drying during biltong processing compared to *Salmonella* and other pathogens. Other surrogates such as *Carnobacterium spp.* and the Five American Type Culture Collection (ATCC) non-pathogenic *E. coli* were found to behave more similarly to *Salmonella* and other pathogens. For this reason, FSIS recommends establishments consider other surrogates such as *Carnobacterium spp.* or the Five American Type Culture Collection (ATCC) non-pathogenic *E. coli* for in-plant validation studies used to validate drying.

[27] Additional research may be available to support the use of these (or other) surrogates for these (or other) processing steps.
[28] The surrogates have also been validated for freezing, refrigerated storage, and antimicrobial treatment of carcasses and ground beef.
[29] The surrogate has also been validated for cooking of ground beef.

### *Methods of Production, Enumeration, and Standardization of Inoculum*

FSIS recommends that strains used in challenge studies for processes that rely on low pH to achieve lethality (*e.g.,* fermentation) be exposed to acid during inoculum preparation (Breidt *et al.*, 2018; Buchanan and Edelson, 1996; Hill *et al.*, 1995). Adding glucose is one way to ensure that the inoculum is pre-adapted for acid tolerance, because the cells ferment the glucose, thereby lowering the pH of the media. Specifically, for studies conducted in a testing laboratory using STEC and *Salmonella*, individual cultures of each strain should be prepared by inoculating an appropriate growth media, such as tryptic soy or trypticase soy broth, supplemented with 1% glucose and incubating for 18 to 24 hours at 98.6 °F (37 °C) without agitation (shaking) to obtain stationary phase cells. Cultures should be grown the day prior to product inoculation with a minimum holding period prior to actual use. Each strain should be centrifuged, washed, and resuspended in 0.1% peptone broth. Dilutions of each strain should be made to yield approximately equal numbers of each of the five strains. The five strains should be thoroughly mixed prior to being used as an inoculum. After the mixed working inoculum is prepared, the viable count of the mixture should be determined by direct surface plating on sorbitol-MacConkey agar. Each of the individual strains in the inoculum should contribute about 20% of the total inoculum (Eblen *et al.*, 2005).

### *Size of the Inoculum to be Used*

The final concentration of *Salmonella*, STEC, or *Lm* in the product mixture should be no less than $2.0 \times 10^7$ CFU/g of meat mixture. The actual inoculum level in the product mixture should be confirmed by sampling the inoculated meat mixture immediately after inoculation. At this concentration, product can be serially diluted and direct plated without the need for enrichment to recover low levels of inoculum. The initial inoculum level was chosen to allow direct enumeration of at least a 5.0-log reduction in the level of the inoculum between the initial count in the product mixture and the finished product.

### *Method of Inoculation*

The inoculum should be added to the meat (or poultry) mixture prior to the addition of the other ingredients or a starter culture.

The following procedure is recommended for fermented sausages:

- Add inoculum to meat or poultry while grinding or chopping to desired consistency.
- Mix in cure (if used), salt, and spices.
- Blend in starter culture near end of mixing cycle.
- Stuff batter into casings.

**NOTE:** For practical reasons, establishments may formulate their products at the establishment and send the batter to their private lab where the inoculum is mixed in prior to stuffing. FSIS has no objection to this procedure provided all replicates are prepared in the same manner.

For fermented products, inoculated product should be stuffed into casings as usual to approximate normal production procedures. A shorter length in casings may be used as long as the length is approximately twice the diameter of the stuffed casing.

For fermented sausages, **beaker sausages** (where the sausage batter is fermented in a test tube) may be used to give a general estimate of the effectiveness of a process but are not recommended as the basis of a model system in a study used as the sole support because of differences in geometry and size of the test tube compared to the product produced and because of differences in the processing conditions, including the humidity. See pages 13 of this document for an explanation of how use of impermeable, sealed glass tubes as a model system may have contributed to an outbreak in a Lebanon bologna product.

***Sample Size, Sampling Time, and Number of Samples to Test***

A minimum of two samples for microbiological analysis should be collected at Time 0 and at the end of all multi-hurdle steps. Time 0 should be the point after the inoculum is added to the product but before the processing steps are applied. Time 0 should not be the inoculum level in the test tube before it is applied to the product.

For dry or semi-dry fermented sausage, establishments should select two sausage sticks at the end of drying (finished product). From each stick selected, cut multiple cross-sectional slices from multiple locations on each stick to a final analytical sample weight based on the method (*e.g.*, many methods are validated for 25 g).

***Methods of Microbial Analysis***

Methods should be specific or fit for the intended purpose to detect the target microorganism in the sample.

***Number of Replicates***

As recommended by NACMCF, at least two replicates should be conducted when three or more samples are tested at each time interval. According to NACMCF, replicates should be independent trials using different lots of product and inoculum to account for variations in product, inoculum, and other factors. When the number of samples analyzed at each time interval is only two, it is better for the study to be repeated (replicated) more than two times.

For guidance on evaluating the results of a challenge study or other scientific literature see FSIS' Cooking Guideline, page 56, "Acceptability of Challenge Study Results".

***Measurement of Critical Operational Parameters and Intrinsic Factors at Each Key Stage of Production***

Although samples for microbiological analysis are recommended to be collected at Time 0 and at the end of all of the multi-hurdle steps (*e.g.*, fermentation and drying or salt-curing and drying), FSIS recommends data be gathered on the critical operational parameters (*e.g.*, time, temperature, and relative humidity) during each processing step (*e.g.*, stuffing, fermentation, low-temperature heat step, if used, and drying). In addition,

data on the product's intrinsic factors (*e.g.*, salt and brine concentration, pH, and water activity) should be gathered at the end of each key processing step (*e.g.*, after stuffing, fermentation, low-temperature heat step, if used, and after drying). Parameters assayed for at the end of each step may also include moisture, fat, protein, and titratable acidity.

The challenge study should be designed to closely match the critical operational parameters (*e.g.*, time, temperature, and relative humidity) and product's intrinsic factors (*e.g.*, salt and brine concentration, pH, and water activity) in the establishment's actual process. As with journal articles and other types of support, it is very important that the critical operational parameters used during the actual experiment are documented, as these should then set the range for the establishment's actual process critical control point (CCP) critical limits and program limits. For example, if a challenge study is conducted to measure the effectiveness of a drying step and the product is dried in a drying room where the temperature ranged from 50-60 °F, the relative humidity ranged from 45-55%, and the air flow ranged from 0.5 to 1 m/sec, then the establishment should set its CCP limits to ensure the temperature does not go below 50 °F or above 60 °F, the relative humidity should not go below 45% or above 55%, and the air flow does not go below 0.5 m/sec or above 1 m/sec. If the establishment wants to use different limits, then it should provide a science-based justification for why these different limits would result in reductions consistent with those observed in the challenge study.

## Appendix 16: Glossary

**Air velocity:** rate of motion of air in a given direction.[30]

**Alternative lethality:** A lethality target or log reduction that is different from FSIS recommendations but achieves an equivalent probability that no viable *Salmonella* organisms or other pathogens of concern remain in the finished product when properly implemented as described in the supporting scientific documentation.

**Bacteriocins**: toxins produced by bacteria that inhibit the growth of other similar bacteria.

**Basturma:** a traditional Turkish product made from whole muscle that is dry-cured, dried, pressed and coated with çemen, a paste made of spices and flavorings, such as crushed cumin, fenugreek, garlic, and paprika. Other names for the product include pastirma, bastirma, pasterma, basterma.

**Biltong:** a dried beef product developed in South Africa that is often made from strips of beef that are trimmed, salted, and dried.

**Bresaola:** is a salt-cured and dried product traditionally produced with whole beef muscles, such as eye of round and inside round. These cuts are typically dry-cured with salt and spices, allowed to equalize, and then are stuffed into casings and hung to dry (Watson *et al.,* 2021).

**Brine Concentration:** is a measure of the amount of salt in the water phase of the product. Brine concentration can't be determined by the formulation; it is a value calculated from the total salt content and total water content values obtained by a lab analysis. Refer to FSIS Processing Inspectors' Calculations Handbook Chapter 14 for more information.

**Case hardening:** a shell of hardened protein at the surface of a meat or poultry product.

**Casing type:** the type of material that encloses the filling of a sausage.

**Certificates of Analysis (COAs)**: A document that includes test results or other analyses to show a product conforms to its specifications.

**Challenge study:** a type of study used to simulate what happens to a product during processing, distribution, and subsequent preparation and handling should it become contaminated[31].

**Country ham:** the dry-cured hind leg of a pig that is produced meeting the standard of identity in 9 CFR 319.106.

[30] https://www.mindat.org/glossary/air_velocity

[31] https://www.foodsafetymagazine.com/magazine-archive1/aprilmay-2001/do-you-need-microbial-challenge-testing/

**Critical operational parameters:** the specific conditions that the intervention must operate under for it to be effective.

**Degree-hours**: the amount of time in hours above 60 °F (the critical temperature at which staphylococcal growth effectively begins) an establishment's fermentation process can take at a specific temperature to reduce the pH to 5.3 or below to control *S. aureus* growth. Degree-hours were specifically designed to control *S. aureus* growth during fermentation.

**Direct acidulation:** the process of reducing the pH by the direct addition of organic acids.

**Dried products:** comminuted, sliced whole muscle, or whole muscle products that may be formulated with nitrite, may be smoked, are usually heated, and are dried.

**Droëwors:** a dried beef sausage product developed in South Africa that is made from ground product stuffed into casings after being mixed with vinegar and seasoning blends.

**Dry-curing:** process where the cure ingredients are rubbed onto the food surface or mixed into foods.

**Excision sampling:** sampling method that involves cutting samples of meat from the surface.

**Fermented products:** products in which the raw meat or poultry component is usually reduced in size, formulated with cure, starter culture, salt and seasoning mixture, stuffed in casings, fermented, sometimes heated with a low temperature heat step for food safety or smoked, and then dried.

**Genoa salami:** A type of dry sausage product prepared with all pork or with a mixture of pork and a small amount of beef. For more information see the entry in FSIS Food Standards and Labeling Policy Book available at: https://www.fsis.usda.gov/guidelines/2005-0003.

**Green weight:** the weight of the meat and/or poultry product or meat and/or poultry byproduct (meat block) before processing.

**Isoelectric point:** the pH where positive charges are equal to negative charges. For meat or poultry, once the pH has reached the isoelectric point of major proteins, the positive charges are equal to negative charges resulting in a net charge of the protein of zero. These positive and negative groups within the protein are attracted to each other and can result in a reduction in the amount of water than can be attracted and held by that protein.[32]

**Lebanon bologna:** A type of coarse ground, fermented, semi-dry sausage. The production of Lebanon bologna typically involves combining curing ingredients (such as

[32] https://swine.extension.org/water-holding-capacity-of-fresh-meat/

salt and sodium nitrite) and a "starter" culture of lactic acid bacteria with coarse ground beef. This mixture is placed in casings, fermented, and further dried.

**Lethality:** the process or combination of processes that ensures that no *Salmonella* organisms remain in the finished product, as well as reduces other pathogens and their toxins or toxin metabolites. Examples of lethality processes include cooking, fermentation, salt-curing, and drying.

**Letters of Guarantee (LOGs):** A LOG is a document that provides details for components that are used in the areas of food processing, handling, and storage.

**Moisture Protein Ratio (MPR)**: a measure that expresses the percent moisture divided by the percent protein. MPR is commonly used in the U.S. to classify dried sausages and other meat products. Although MPR values indicate the degree of product drying, they are not necessarily indicative of microbial safety or product shelf-stability because they do not take into account availability of the water.

**Mother batch:** previously fermented and controlled batch of product.

**Pepperoni:** A type of dry sausage prepared from pork or pork and beef. For more information see the entry in available at: https://www.fsis.usda.gov/guidelines/2005-0003.

**Proteolysis:** the breakdown of proteins or peptides into amino acids by the action of enzymes.

**Ready-to-eat** meat and poultry product is defined by FSIS in 9 CFR 430.1 as a meat or poultry product that is in a form that is edible without additional preparation to achieve food safety and may receive additional preparation for palatability or aesthetic, epicurean, gastronomic, or culinary purposes.

**Salt-cured products:** usually whole muscle products that are cured with salt and sodium nitrite and/or nitrate then dried and sometimes smoked (if desired for certain flavor characteristics).

**Shelf-stable** for the purposes of shelf-stable meat and poultry products is defined as the condition achieved when meat and poultry products can be stored under ambient temperature and humidity conditions; if the package integrity is maintained during storage, shipping, and display at retail and in the home; and the product will not spoil or become unsafe throughout the manufacturer's specified shelf-life.

**Soudjouk:** a highly spiced, Mediterranean-style, semi-dry sausage made by mixing ground meat, spices, and curing salts, and then stuffing the resulting batter into a natural or artificial casing that is flattened and subsequently fermented and dried at ambient temperature over several days (Saricoban *et al.*, 2006). Other spellings include sujuk, sucuk, soujouk, and sudjuk.

**Stabilization** is the process of preventing or limiting the growth of spore-forming bacteria capable of producing toxins either in the product before consumption or in the human intestine after consumption. Establishments may use a variety different

stabilization processes, such as cooling, hot-holding, or meeting and maintaining certain pH or water activity levels.

**Summer sausage:** refers to semi-dry sausages, especially Thuringer Cervelat. For more information see the entry in Food Standards and Labeling Policy Book available at: https://www.fsis.usda.gov/guidelines/2005-0003.

**Targets:** quantifiable pathogen reduction levels or growth limits set by the establishment to produce safe products in the absence of regulatory performance standards. As required by 9 CFR 417.2(c)(3), critical limits shall, at a minimum, be designed to ensure that applicable targets or performance standards established by FSIS, and any other requirement pertaining to the specific process or product be met.

**Water activity:** abbreviated as $a_w$, water activity is a measure of the concentration of moisture (i.e., water) and its availability in a food. The amount of water available in a food depends on the total concentration of all dissolved substances in the product because they bind water. Thus, if ingredients, such as salt or sugar, are added to food, they compete with the bacteria for available water.


https://www.fsis.usda.gov/contact-us/askfsis

**FSIS/USDA**
**www.fsis.usda.gov**
**2023**