

The research projects listed below are funded in whole or part by the Beef Checkoff and are intended to help ensure a safe and nutritious beef supply.



Funded by the Beef Checkoff

Current Research Projects

July 2025

From Trim to Table: Tracking *Salmonella* Dynamics, Including Levels and Survival, from Beef Trim to Ground Product, Kansas State University and Cargill

This research will conduct an inoculation study to evaluate the influence of *Salmonella* levels in beef trim (including lymph nodes) on *Salmonella* levels in ground beef, providing valuable insights for managing *Salmonella* in final product. *Salmonella* survival will be assessed during standard industry storage periods to further understand its persistence and potential risks in products intended for commerce.

Historical analysis of *Salmonella* using pre-existing data: trends in outbreaks, genomics, and geographic factors, University of Georgia

Emerging pathogen monitoring programs, such as wastewater monitoring for COVID-19, have gained critical focus as a possible way to predict the next outbreak and assist in development of management tools before the first wave of illnesses hits. These approaches can be extended to beef-related *Salmonella* outbreaks by leveraging the wealth of surveillance data that is collected into the NCBI Pathogen Detection Isolates Browser. This study will develop a framework for understanding where, why, and how certain *Salmonella* strains emerge to cause outbreaks. A short-list of warning signs and monitoring recommendations will be developed to predict the next outbreak before it happens.

Reliability and repeatability of digital PCR and VAE assays for the enumeration of *Salmonella* in beef samples, Florida State University and U.S. Meat Animal Research Center USDA-ARS

This study intends to optimize digital PCR (dPCR) and varying amplification efficiency (VAE) assay workflows for the detection and quantification of *Salmonella* load in beef samples. The reliability and repeatability of dPCR and VAE assays will be validated and their applicability will be compared with an AOAC performance-tested method. The application of a partition-based digital PCR approach will enable highly reliable and repeatable detection and quantification of *Salmonella* load in contaminated beef samples. The VAE assay will enable simple estimation of *Salmonella* levels in beef samples.

Improving and validating the THERM model for predicting growth of *Staphylococcus aureus* in raw meat products during temperature abuse and come-up-time (CUT) deviations, University of Wisconsin – Madison

The current Temperature History Evaluation for Raw Meats (THERM) model, originally intended for use by small and very small processors who lacked refrigeration in their raw processing areas and now widely used at all levels of the meat processing industry to estimate pathogen growth in raw materials, may overestimate pathogen growth when following USDA temperature recommendations. This study will refine product categories and determine the growth of *Staphylococcus aureus* in uncured model meat products with varying pH levels and temperatures which represent come-up-time ranges where *S. aureus* may grow. A validated predictive model will be developed using a variety of different meat products and formulations.

Current Research Projects *continued*

Analysis of beef *Salmonella* outbreaks using the USMARC SNP analysis pipeline, U.S. Meat Animal Research Center USDA-ARS

Using the pipeline developed in a previously funded project (see Enhanced Characterization of Sequence Differences Among *Salmonella* isolates within SNP Clusters Identified by the NCBI Pathogen Detection System), genomes from previous beef *Salmonella* outbreaks will be evaluated to characterize the genomic variation between sequenced isolates related to the outbreak. The findings will detail the reliability of Beef *Salmonella* outbreak traceback.

Creating Alternative Support for Lethality and Stabilization for Heat Treated and Fully Cooked Meat and Poultry Products, University of Wisconsin, HansonTech

Nearly all meat processors in the United States utilize USDA, FSIS Appendix A and B to ensure adequate thermal lethality and stabilization is achieved for partially and fully cooked products. Through the development and release of updated versions in 2017 and 2021, and the realization that a host of potential food safety vulnerabilities exist, the widespread usefulness and in-plant practical application of these guidance documents has become a significant concern and practical challenge to implement. The primary objective of this study is to develop a scientific-based, regulatory-supported, and industry-useful thermal processing and cooling resource (e.g. cooking and cooling food safety handbook) for validating pathogen destruction and control, and regulatory compliance for partially and fully cooked meat products that can be used in conjunction with or in lieu of USDA, FSIS Appendix A & B.

Funded in part by the Meat Foundation

Rapid identification of ground beef and beef trim samples exceeding a *Salmonella* threshold level, Kansas State University and Cargill

The study will determine the impact of the *Salmonella* inoculation method on various *Salmonella* quantification and limit of detection (LOD) methods. Additionally, the study will determine the reliability and repeatability of various *Salmonella* quantification and LOD methods for the identification of non-intact beef samples harboring *Salmonella* equal to or above a threshold. Findings will address beef industry concerns regarding specific methods.

Effect of organic acid components on the inhibition of *Clostridium perfringens* during extended cooling of uncured meat products and the inhibition of *Listeria monocytogenes* during extended refrigerated storage, University of Wisconsin-Madison Food Research Institute

This research will compare the relative efficacy and synergistic effect of the active components (lactate, propionate, acetate/diacetate) found in clean label growth inhibitors (such as cultured sugar and vinegar-based products) on the inhibition of *Clostridium perfringens* in a model-uncured beef system, with primary focus on extending Phase 1 cooling (120 to 80°F). It will also validate inhibition of *Listeria monocytogenes* during extended refrigerated storage of a model deli meat system formulated with organic acid salts used to control *C. perfringens*.

Current Research Projects *continued*

Targeting Foodborne Pathogens in the Beef Industry through an Integrated Analysis of the Beef Processing System, Texas Tech University, University of Nebraska, U.S. Meat Animal Research Center USDA-ARS, and Auburn University

This research will take a comprehensive approach to targeting *Salmonella* and Shiga Toxin Producing *E. coli* (STEC) conducted through optimization of sampling techniques, microbial detection and microbial quantification methods combined with microbiome and metagenomic analysis to enable mitigation strategies and validation techniques to impact pathogen presence in beef products.

Assessing *Salmonella* status (prevalence, load, pathogenicity, and serotype complexity) through beef processing, University of Georgia and Texas Tech University

This study will integrate four advanced laboratory techniques, e.g. PiLOT, CRISPR-SeroSeq, to improve understanding of *Salmonella* dynamics in beef processing. Specifically, these data and techniques will enable (1) identifying steps in the processing chain which change the *Salmonella* communities either by load or loss or gain of subtypes (serotype, pathogenicity), or both and (2) describing which *Salmonella* subtypes (serotype, pathogenicity) are selected for, or which are lost, during processing steps. Findings will help build the research foundation needed for food safety protocols to produce integrated data sets. This data can then be applied to ensure that proposed regulatory frameworks meaningfully improve beef safety.

Recently Completed Research

Developing a Quantitative *Salmonella* Baseline from Ground Beef in the United States, Texas Tech University, Kansas State University, University of Georgia, U.S. Meat Animal Research Center USDA-ARS, Food Safety Net Services

The overall prevalence of *Salmonella enterica* from ground beef samples was determined over the course of 14 months. There was quarterly variation with the peak prevalence occurring in the Spring and Fall seasons. Demographic information indicated that *Salmonella* testing was not commonly done on beef trimmings prior to grinding or on final product.

Enhanced Characterization of Sequence Differences Among *Salmonella* isolates within SNP Clusters Identified by the NCBI Pathogen Detection System, U.S. Meat Animal Research Center USDA-ARS

This research intended to better understand the full picture of relatedness within critical *Salmonella* serovars of interest by performing a comparative genomic analyses on currently available data within the Pathogen Detection Isolates Browser (PDIB). An analysis pipeline was developed to catalogue *Salmonella* SNP cluster diversity in the NCBI PDIB. Relatedness analysis performed using the pipeline closely approximated the SNP differences reported by the PDIB and showed that the analysis can be done outside the database. The pipeline allows for a more complete understanding of the sequence similarity between isolates by identifying novel or missing elements in the genomes and cataloguing the sequence diversity.

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