SUPPORTING MATERIALS (IN ORDER BY REFERENCE)

Purpose

The Research Triangle Institute (RTI) is conducting a multi-year evaluation of the *Pathogen Reduction; Hazard Analysis and Critical Control Point (HACCP) Systems; Final Rule* for the U.S. Department of Agriculture, Food Safety and Inspection Service (USDA, FSIS). As part of this evaluation, RTI is conducting a series of studies to evaluate consumer knowledge, behavior, and confidence, and consumer education programs. The first study was to conduct focus groups with adult consumers to (1) test FSIS food safety messages and (2) identify effective delivery mechanisms for these messages. In addition, we obtained information on participants' confidence in the safety of meat and poultry, their general food safety knowledge and use of safe handling practices, and their awareness of FSIS and government food safety initiatives.

This summary report is part of the Final Report, which provides detailed summaries of each focus group and an analysis of the Pre-Discussion Survey data. Participants completed the Pre-Discussion Survey—a short survey that included questions from the ongoing FDA/FSIS Food Safety Survey—prior to the focus group discussion.[1]

Key Findings

Our key findings from the Pre-Discussion Survey and the focus group discussions are summarized below.

[1]The FDA/FSIS Food Safety Survey is conducted every 5 years and collects information on consumer food safety perceptions, knowledge, and behavior. As part of our evaluation of the impact of the PR/HACCP Final Rule on consumer knowledge, behavior, and confidence we will compare the results from the Pre-Discussion Survey to the FDA/FSIS Food Safety Survey.

Consumer Confidence, Food Safety Knowledge, and FSIS Awareness

Participants discussed their confidence in the safety of meat and poultry, their knowledge and use of safe handling practices, and their awareness of FSIS. Participants

- Say their confidence in the safety of meat and poultry is increasing.
- Rely on a variety of sources for food safety information.
- Perceive that their knowledge and use of safe handling practices are increasing.
- Correctly identified meat and poultry as high-risk foods for foodborne illness.
- Are unfamiliar with the bacteria *Listeria monocytogenes* and *Campylobacter.*
- Are unaware that pregnant women are a high-risk group for foodborne illness.
- Have not heard of the Food Safety and Inspection Service (FSIS).

Food Safety Messages

Participants are familiar with and understand most of the food safety messages that we asked about, including expiration dates. However, there are some gaps in consumer awareness and understanding:

- Importance of using a food thermometer to check for doneness.
- Two-hour rule: refrigerate or freeze foods within 2 hours or less.
- Danger Zone graphic and concept: unsafe temperatures between 40°F and 140°F.
- Phrase "refrigerate leftovers immediately."

- Fight BAC!™[2]—unfamiliar with graphic; partially understand the four messages.
- Term "pathogens"—unfamiliar with term; prefer the phrase "harmful bacteria."

Delivery Mechanisms

Participants recommended the use of multiple delivery mechanisms to increase their food safety knowledge. They also suggested the following:

- Use the safe handling label as a mechanism for educating consumers.
- Target school-age children as a way to get food safety messages to parents.
- Make it convenient to use a food thermometer (e.g., a refrigerator magnet that has a holder for storing a thermometer).

Main Recommendations

Our main recommendations are listed below and discussed later in the recommendations section of this report.

- Target the following areas for food safety education:
  - ✓ Prompt refrigeration of leftovers.
  - ✓ Two-hour rule: refrigerate or freeze foods within 2 hours or less.
  - ✓ Danger Zone concept: unsafe temperatures between 40°F and 140°F.
  - ✓ Pathogens and the dangers posed.
- Promote food thermometer usage:
  - ✓ Continue Thermy™[3] campaign.
  - ✓ Make thermometer usage convenient.
- Reach consumers with a variety of approaches.
- Reevaluate the safe handling label as a mechanism for educating consumers.
- Target pregnant women with information on listeriosis.
- Target children as a way to get food safety messages to parents.
- Redesign the Danger Zone graphic.

This summary report briefly describes the study design, discusses the results of the Pre-Discussion Survey and the focus groups, and presents recommendations for increasing consumer knowledge and use of safe handling practices.

Study Design

We conducted a total of eight focus groups—two groups in each of four different locations with four different populations. For each population, we conducted one focus group with individuals who have a high school education or less and one focus group with individuals who have some college education (most participants have at least a 4-year degree).

We targeted the following populations:

- Young parents—individuals between the ages of 20 and 35 with at least one child who is 6 years old or younger
- Young adults—individuals between the ages of 20 and 30 who have no children

[2]The Fight BAC!™ campaign was launched in October 1997 by the Partnership for Food Safety Education. The focal point of the campaign is BAC and the four food safety messages of clean, separate, cook, and chill (see Figure 1).

[3]The campaign launched by FSIS in spring 2000, with Thermy™ as its food safety messenger, was designed to encourage consumers to use a food thermometer to ensure that food is cooked to a safe internal temperature (see Figure 3).

- Seniors—individuals 60 years old or older
- General—individuals between the ages of 30 and 59

We conducted the focus groups in Raleigh, North Carolina; San Antonio, Texas; San Diego, California; and Annapolis, Maryland. We selected the sites to provide geographic diversity and semi-rural/urban representation. In Raleigh and San Antonio we recruited individuals who live outside the city limits to provide a semi-rural population. Also, San Diego and Annapolis were selected because these cities were sites of a Fight BAC!™ saturation campaign.

Each focus group included a mix of males and females. We recruited participants to reflect the racial diversity of the area in which the group was conducted. All participants have primary responsibility or share responsibility for cooking in their household, prepare meals at least three times a week, save or eat leftovers, and are not vegetarians. Each group included 7 to 9 participants, for a total of 67 participants.

Results

Findings from the Pre-Discussion Survey are highlighted below, along with the qualitative information gathered in the focus group discussions.

Consumer Confidence

Participants' level of concern about foodborne illness varied. Young adult participants are not concerned about foodborne illness because they have never experienced it. Participants in the other groups are somewhat to very concerned about foodborne illness. Media reports on foodborne illness heighten concern. Regardless of the level of concern, participants say they take measures at home to keep food safe.

Participants offered mixed opinions as to whether the rate of foodborne illness is increasing or decreasing.[4] Some participants think it is increasing only because awareness of foodborne illness has increased and people are more likely to report it, while others attribute the increase to new bacteria. Some participants think the rate of foodborne illness is decreasing because consumers are more knowledgeable about safe handling practices and have made changes in how they handle food.

Most participants reported that their confidence in the safety of meat and poultry is increasing or about the same compared to 5 years ago.[5] They attribute their increased confidence to increased awareness of safe handling practices, improved labeling, and prepackaging of meat and poultry. Two senior participants said that their confidence has decreased over the past 5 years citing concerns about the educational level of food handlers and the lack of proper sanitation procedures. One of the 67 participants expressed negative feelings about the government's ability to keep food safe.

About 86 percent of all participants (includes participants from all four locations) are mostly or completely confident that the meat and poultry they prepare at home is safe to eat. Participants are confident because they take precautions to handle and prepare meat and poultry safely and have confidence in the grocery stores where they shop. Several participants said they

[4]This topic was only discussed in the San Antonio and San Diego groups. We added questions to collect information on consumer confidence to use in our evaluation of the impact of the PR/HACCP Final Rule on consumer knowledge, behavior, and confidence.

[5]Ibid.

know the meat they eat is safe because they overcook it.

Participants are more concerned about getting foodborne illness from eating out, especially at fast food restaurants, than from eating food prepared at home. They feel they have control over the safety of the food that they prepare.

Overall, consumer confidence in the safety of meat and poultry appears to be increasing. Most participants are confident that the meat and poultry they prepare at home is safe to eat. Participants are more concerned about getting foodborne illness from eating out because they have more control at home. However, discussions about their actual practices revealed that some participants unknowingly follow some improper handling practices when cooking at home.

Food Safety Knowledge

Most participants described themselves as being fairly knowledgeable about food safety and safe handling practices. They discussed the public's increased awareness about food safety. Some participants discussed how they have made changes in how they handle food after hearing about foodborne illness outbreaks.

Participants identified meat and poultry (83 percent of participants) and fish/shellfish (79 percent) as foods more likely to cause foodborne illness than other foods. About half of participants identified dairy products, eggs, and leftovers as high-risk foods.

Most participants understood that bacteria and improper handling of food are the causes of foodborne illness. Participants identified *E. coli* and *Salmonella* as bacteria that cause foodborne illness but had not heard of *Listeria monocytogenes* or *Campylobacter*.

Participants identified children, seniors, the immuno-compromised, and those with chronic illnesses (e.g., diabetes) as being at high risk for foodborne illness. Less than 25 percent of participants identified pregnant women as a high-risk population.

Participants rely on a variety of sources for information on food safety. The most common sources cited were

- family/friends,
- food labels/packaging,
- newspapers,
- magazines,
- television (news and news programs), and
- cookbooks and cooking shows.

Participants do not generally use the Internet or government sources (e.g., hotlines) for food safety information, although young adult participants suggested using the Internet to disseminate food safety information. Only one female participant recalled her doctor speaking with her about food safety while pregnant.

Safe Handling Practices

Participants discussed measures they take at home to protect their family from foodborne illness. The Pre-Discussion Survey also collected information on the prevalence of certain safe handling practices. The Pre-Discussion Survey findings on participants' use of safe handling practices are summarized below.

- 83 percent of participants wash cutting boards used for cutting meat or poultry with soap and/or bleach water before using the cutting board again; 5 percent use a different cutting board.

- 76 percent of participants wash their hands with soap after handling raw meat or poultry.
- 43 percent of participants own a food thermometer; most own a dial thermometer.
- 22 percent of participants refrigerate leftovers like soup or stew containing meat or poultry immediately.
- 17 percent of participants always or often use a food thermometer when cooking large cuts of meat, like roasts or turkeys.
- One participant uses a food thermometer when cooking hamburgers.

These findings are consistent with the results of the 1998 FDA/FSIS Food Safety Survey. While the majority of participants follow proper cleaning practices most do not refrigerate leftovers immediately or use a food thermometer to check for doneness.

Awareness of FSIS and Government Food Safety Initiatives

Participants identified the FDA (49 percent) and the USDA (22 percent) as the government agency responsible for the safety of meat and poultry.[6] No participants specifically mentioned the Food Safety and Inspection Service (FSIS). When asked, most participants considered this an appropriate name for the agency. Several participants suggested a shorter, simpler name or including "meat and poultry" in the name.

Most participants had not heard of any recent government initiatives to make meat and poultry safer. One participant had heard of HACCP, and one participant knew the meaning of the phrase "farm-to-table." Several participants guessed that "farm-to-table" meant eggs coming straight from the hen to the table (bypassing the grocery store) or produce fresh from the farm.

Understanding of Food Safety Messages

We asked participants about their awareness and understanding of the Fight BAC!™ concept (Figure 1). We also asked participants about key food safety messages. Some of these messages are part of the Fight BAC!™ campaign, while others are used on the safe handling label or FSIS educational materials.

Figure 1. Fight BAC!™

Fight BAC!™ One of the 67 participants had seen Fight BAC!™ prior to the discussion. Although participants were not familiar with Fight BAC!™, many liked the BAC character and the four-message graphic. Participants described it as colorful and eye-catching. They said that it covers the basic food safety messages and is easy to understand. Several participants offered

[6]This topic was not discussed in the Raleigh groups. We added questions to explore the findings from the American Customer Satisfaction Index (ACSI) survey that consumers are not aware of FSIS.

suggestions for improving the "separate" quadrant of the graphic. Some participants said the Fight BAC!™ concept would be effective with the general population, while others suggested targeting it to a younger, more inexperienced population such as those less than 16 years of age, young mothers, and new food preparers.

Wash hands and surfaces often. Participants were familiar with the "clean" message and understood the importance of washing hands and surfaces to prevent the spread of bacteria.

Separate: Don't cross-contaminate. Some participants were not familiar with the "separate" message or said that they had only recently learned about the importance of keeping raw and cooked foods separate. Those familiar with this message correctly discussed keeping raw meat and poultry products separate from ready-to-eat foods when cooking, when shopping, and in the refrigerator.

Cook to proper temperatures/Cook thoroughly. Some participants correctly defined "cook to proper temperatures" as cooking foods to a certain internal temperature to kill bacteria. Others thought "cook to proper temperatures" meant to cook to the temperature specified in the cooking directions or recipe, which is usually correct. A few participants confused internal temperature with oven temperature, which is incorrect.

Participants defined "cook thoroughly" as cooking meat until it is done or cooking all the way through. Participants discussed that consumers may have different preferences about doneness of meat; for example, to some a steak cooked rare is done.

Participants offered mixed opinions as to whether one phrase is more effective at conveying the message to cook meat and poultry to a temperature high enough to kill bacteria. Some participants preferred "cook to proper temperatures" because this indicates that a food thermometer is required to check for doneness, while others, particularly young adults, said they would never use a thermometer to check for doneness.

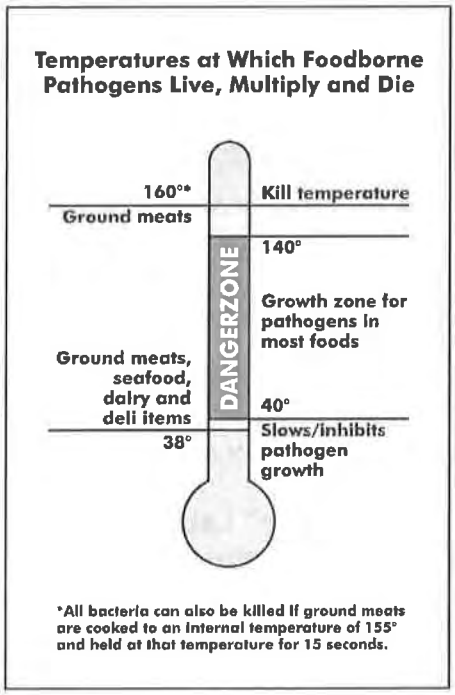
Refrigerate promptly/Refrigerate leftovers immediately or discard. Participants defined "refrigerate promptly" as refrigerating or freezing food after returning from the grocery store and refrigerating leftovers after eating. Some participants thought that "refrigerate leftovers immediately or discard" meant to refrigerate leftovers *after* cooling to room temperature. Many participants incorrectly let leftovers cool before putting them in the refrigerator. No participants divide leftovers into several shallow containers before refrigerating which is the recommended practice. Some participants were surprised to learn that cooling leftovers to room temperature is an unsafe practice and that putting warm foods in the refrigerator will not damage the refrigerator.

Danger Zone: unsafe temperatures between 40°F and 140°F. We showed participants two versions of the Danger Zone graphic currently used in FSIS educational materials and asked them to evaluate the graphics (Figure 2). Only one young adult participant who had recently completed a cooking class was familiar with the Danger Zone graphic and concept.

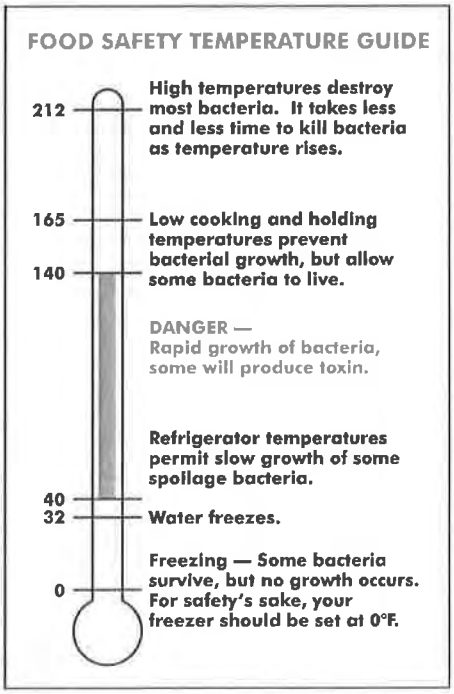
Participants found both versions of the graphic confusing and thought some combination of the two would be better. Participants did not mind that the thermometers used in the graphics do not resemble a food thermometer, saying the graphic used was the universal symbol for a thermometer. Some participants suggested integrating BAC or Thermy™ with the Danger Zone graphic.

Figure 2. Danger Zone

Version 1



Version 2



Keep hot foods hot and cold foods cold. Although not all participants had heard this phrase, participants were generally familiar with this concept.

Two-hour rule. Most participants were not familiar with the two-hour rule—that is, to refrigerate or freeze perishables, prepared foods, and leftovers within 2 hours or less. Participants wanted to know whether the rule applied to raw or cooked meat.

Thaw in refrigerator or microwave. Most participants knew to thaw foods in the refrigerator or microwave and recall seeing this information on poultry packaging. Some participants said they sometimes thaw turkeys in water in the sink. Most participants do not thaw foods at room temperature; several participants admitted that until recently they used to do this.

When in doubt, throw it out! Most participants were familiar with this message and practiced it at home. Participants discussed that they label leftovers with dates so they know when to dispose of them. Some participants said they use disposable storage containers so it is easy to dispose of old leftovers.

We also asked participants about expiration dates and other words and phrases used in food safety educational materials.

Expiration dates. Most participants knew the meanings and understood the differences between the sell-by, best-if-used-by/before, and use-by dates. Many participants refer to this information when shopping.

Poultry. Participants defined "poultry" to include chicken, turkey, duck, game hen, and any animal with feathers.

Ground beef. When asked what comes to mind when they hear "ground beef,"

participants said hamburgers, patties, and spaghetti.

Perishable. Participants correctly defined "perishable" as foods that spoil and foods that require refrigeration.

Pathogens. Some participants were not familiar with the term "pathogens." Those who were knowledgeable defined pathogens as germs, bacteria, or organisms that cause illness. Most participants preferred using the phrase "harmful bacteria" to "pathogens."

Participants had mixed opinions on whether it was effective to include the name of specific bacteria to get consumers' attention. Some participants said this would be appropriate on educational materials, but not on product labels. Others did not think it was necessary.

Irradiation. With the exception of one focus group, one or more participants in each group had heard of irradiation, although some did not know what it meant. Nine of the 67 participants expressed concern about irradiation and wanted to know if irradiated products are labeled.

Elderly. Participants in the seniors groups said that "seniors," "senior citizens," and "60 plus" are the best terms to use when referring to this population. Seniors do not like to be referred to as the "elderly."

Safe Handling Label

About 64 percent of participants recall seeing the safe handling label on raw meat and poultry products. Some participants compared it to the warning label on cigarettes—people see the label but they do not read it.

Suggestions for increasing the awareness and usefulness of the safe handling label included the following:

- Use color (e.g., red).
- Increase the size of the label and the font size of the text.
- Make the label a peel-off sticker that consumers can keep.
- Identify it as a warning by adding "Warning" in bold.
- Add instructions on the proper cooking temperature.
- Display a poster-size version of the label in the meat department.
- Use shorter phrases, like those used in the Fight BAC!™ graphic.
- Include Thermy™ on the label.

Using "Negative" Labels for Consumer Education

We asked participants what they thought about the following label:

> "Cook to 160°F. Cooking ground beef to 160°F eliminates harmful bacteria which could cause serious or fatal illness."

Participants had mixed opinions about the label. Some said the label would be effective at encouraging consumers, especially those just learning to cook, to cook ground beef to a safe temperature. Some said the label might be effective at first but that it would become like the cigarette warning labels that people do not pay attention to anymore. A few participants said such a label might discourage consumers from purchasing ground beef. Several participants said the label should be on all meat and poultry products, rather than singling out ground beef.

Promoting Thermometer Usage

When discussing the proper cooking of meat and poultry, some participants reported that they generally overcook meats, particularly hamburgers, to ensure doneness. About 1 in 5 participants uses a food thermometer to check for doneness of large cuts of meat. Others cut the meat to check the color or judge doneness based on the cooking time.

We presented the results of the Agricultural Research Service (ARS)/FSIS research on color of ground beef as it relates to doneness. Many participants were surprised to learn that color is not always a good indicator of doneness. Several participants from the seniors groups said that they plan to purchase and start using a thermometer in light of this research. Some young adult participants were not swayed by the research and said that they would not start using a thermometer. They said using a thermometer is impractical and unnecessary. They believe they can trust their own experience to ensure that meat is cooked thoroughly.

We asked participants what they thought about the Thermy™ messenger and slogan, "It's safe to bite when the temperature is right!" (Figure 3). Many participants liked Thermy™ and described it as cute, catchy, and attention-getting and thought it would appeal to the general population. A few participants described Thermy™ as too childish for such a serious subject and suggested targeting it to children.

Participants said that for them to start using a thermometer for all cuts of meat it would have to be easy and convenient. Even those who own a thermometer do not always use it, since it is often hidden in the back of a drawer. Participants suggested the following approaches for promoting thermometer usage:

Figure 3. Thermy™

- consumer education on the ARS/FSIS research on color of ground beef as it relates to doneness
- grocery store promotions at the store entrance or meat department; for example, display thermometers in the meat department and at the check-out line, offer free thermometers or discount coupons, or distribute literature/ coupons/recipe cards in conjunction with food samples or cooking demonstrations
- promotional items; for example, magnets with proper cooking temperatures
- labels on raw meat and poultry packaging with proper cooking temperatures
- disposable thermometers attached to meat/poultry packaging
- refrigerator magnet that has a holder for storing a thermometer (for ease of use and accessibility)

Delivery Mechanisms for Food Safety Messages

We asked participants about the best way to get information on food safety to them and others in their target population. Participants said that in this "information age" consumers are often overwhelmed with information, so the information provided must get their attention, be concise, and provide steps for immediate action. Participants suggested the following approaches for delivering food safety messages:

- television and radio commercials/public service announcements
- television news and news shows
- newspapers (weekly food section)
- magazines (e.g., cooking, women's, and general interest)
- cooking shows
- grocery store promotions
- labels on meat and poultry packaging
- home mailings with food safety information
- Internet Web sites (to reach young adults, in particular)
- senior center programs (to reach seniors, in particular)

Participants, especially young parents, suggested targeting children in food safety educational efforts because children will bring the message home to their parents. Participants suggested that educational efforts should start in elementary school. Participants suggested the following approaches for educating children about food safety:

- workshops at school
- food safety day at school
- commercials/public service announcements during children's programming
- free comic books/coloring books on food safety distributed via grocery stores
- literature distributed via school
- public service announcements using the *School House Rocks* format

Participants had mixed opinions about using cartoon characters like Thermy™ and BAC to promote food safety. Many participants thought such characters would be effective, especially with children. Some participants in the young adults and the general population groups did not like the use of animated commercials, saying such commercials would not be taken seriously.

Recommendations

Recommendations based on the focus group findings are summarized below.

Target These Areas for Food Safety Education

The focus groups identified the following areas where additional educational efforts are needed:

- Prompt refrigeration of leftovers.
- Two-hour rule: refrigerate or freeze foods within 2 hours or less.
- Danger Zone concept: unsafe temperatures between 40°F and 140°F.
- Pathogens and the dangers posed.

Many participants cool leftovers to room temperature before refrigerating, instead of refrigerating them immediately. Most participants were not familiar with the two-hour rule or the Danger Zone graphic and concept. Future educational efforts should target these areas.

Some participants were not familiar with the term "pathogens." We suggest that FSIS use the phrase "harmful bacteria" instead of "pathogens" in educational materials. Participants had mixed opinions on whether it was effective to include the name of specific bacteria to get consumers' attention.

Most participants were not familiar with the bacteria *Listeria monocytogenes* and *Campylobacter*. We recommend that FSIS educate consumers about the dangers of specific bacteria when necessary; for example, *Listeria monocytogenes* and the danger posed to pregnant women.

Promote Food Thermometer Usage

FSIS should continue their educational efforts on thermometer usage using Thermy™ as its "spokesperson" so that Thermy™ becomes a symbol of cooking food properly, like Smokey Bear or McGruff the Crime Dog. The Thermy™ campaign uses many of the ideas suggested by participants, such as grocery store promotions, refrigerator magnet with temperature chart, and educational materials on the ARS/FSIS research.

Participants said that for them to start using a food thermometer it would have to be easy and convenient. Even those who own a thermometer do not always use it. Possible approaches for making thermometers more accessible include disseminating refrigerator magnets that have a holder for storing a thermometer or providing disposable thermometers with meat/poultry packaging.

Reach Consumers with a Variety of Approaches

Participants suggested a myriad of ideas for promoting food safety, some of which are currently being used by FSIS. We recommend that FSIS continue to use multiple sources to educate consumers. Possible approaches for delivering food safety information include the following:

- Broadcast public service announcements on radio and television.
- Disseminate information on safe handling practices through newspapers (food section) and magazines.
- Disseminate information on ARS/FSIS research on color of ground beef as it relates to doneness via television news or news shows.
- Incorporate safe handling practices on television cooking shows.
- Disseminate information through schools to reach parents of school-aged children.
- Disseminate brochures covering the basics of food safety to individual households.
- Use labels on meat and poultry packaging to educate consumers.
- Provide animated graphics and interactive features, such as a self-administered quiz on food safety, on health/nutrition and food safety Web sites (to reach young adults, in particular).

Further analysis is required to determine which approaches would be most cost-effective.

Reevaluate the Safe Handling Label

Although many participants are familiar with the safe handling label, some no longer pay attention to it. We recommend that FSIS reevaluate the safe handling label to determine if the label needs to be revised. To increase visibility and awareness of the label, add color and shorten the messages, making them consistent with the four Fight

BAC!™ messages. We suggest that FSIS consider incorporating Thermy™ (as part of the label or a separate sticker) to get consumers' attention and to remind them to use a food thermometer to check for doneness. Any proposed changes to the safe handling label should be thoroughly tested with consumers.

Target Pregnant Women

About 75 percent of participants did not identify pregnant women as being at increased risk for foodborne illness. Also, participants were not aware of *Listeria monocytogenes* and the danger posed to pregnant women. We recommend that FSIS develop educational materials targeted to pregnant women. Additional research is required to determine how to effectively reach pregnant women with food safety messages.

Target Children

Participants suggested targeting children in food safety educational efforts because children will bring the message home to their parents. Possible approaches for educating children were listed earlier in the report (see page 10). Further research is required to determine the effectiveness of educating children as a delivery mechanism for food safety messages.

Redesign the Danger Zone Graphic

Many participants found the Danger Zone graphics confusing. We recommend that FSIS redesign the Danger Zone graphic, incorporating some of the changes suggested by participants.

Safe Minimum Internal Temperature Chart

Safe steps in food handling, cooking, and storage are essential in preventing foodborne illness. You can't see, smell, or taste harmful bacteria that may cause illness. In every step of food preparation, follow the four guidelines to keep food safe:

- **Clean**—Wash hands and surfaces often.
- **Separate**—Separate raw meat from other foods.
- **Cook**—Cook to the right temperature.
- **Chill**—Refrigerate food promptly.

Cook all food to these minimum internal temperatures as measured with a food thermometer before removing food from the heat source. For reasons of personal preference, consumers may choose to cook food to higher temperatures.

Product	Minimum Internal Temperature & Rest Time
Beef, Pork, Veal & Lamb Steaks, chops, roasts	145 °F (62.8 °C) and allow to rest for at least 3 minutes
Ground meats	160 °F (71.1 °C)
Ham, fresh or smoked (uncooked)	145 °F (62.8 °C) and allow to rest for at least 3 minutes
Fully Cooked Ham (to reheat)	Reheat cooked hams packaged in USDA-inspected plants to 140 °F (60 °C) and all others to 165 °F (73.9 °C).

Product	Minimum Internal Temperature
All Poultry (breasts, whole bird, legs, thighs, and wings, ground poultry, and stuffing)	165 °F (73.9 °C)
Eggs	160 °F (71.1 °C)
Fish & Shellfish	145 °F (62.8 °C)
Leftovers	165 °F (73.9 °C)
Casseroles	165 °F (73.9 °C)

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Food Safety and Inspection Service

Food Safety Handling Labels

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January 7, 2014

Food Safety and Inspection Service

Safe Handling Instructions (SHI) Background

- In 1993, a small team was tasked to identify information that would be included in a safe handling instruction statement.
- August 1993 FSIS issued an interim final rule on SHI.
- September 1993 Several Associations filed a complaint against FSIS for not following APA rules of notice and comment.
- November 1993 the interim final rule was withdrawn.
- A focus group study was performed prior to publishing a rule.
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- A proposal was published November 1993
- March 1994 the final rule was published, effective May 1994.

Food Safety and Inspection Service

SHI – Background (continued)

- At that time there were many minimum internal temperature requirements in the regulations and in consumer publications. For example, in the regulations cured poultry had to be cooked to 155°F, uncured poultry to 160°F (9 CFR 381.150) and Beef to 145°F (9 CFR 318.17). FSIS regulations now provide lethality measures.

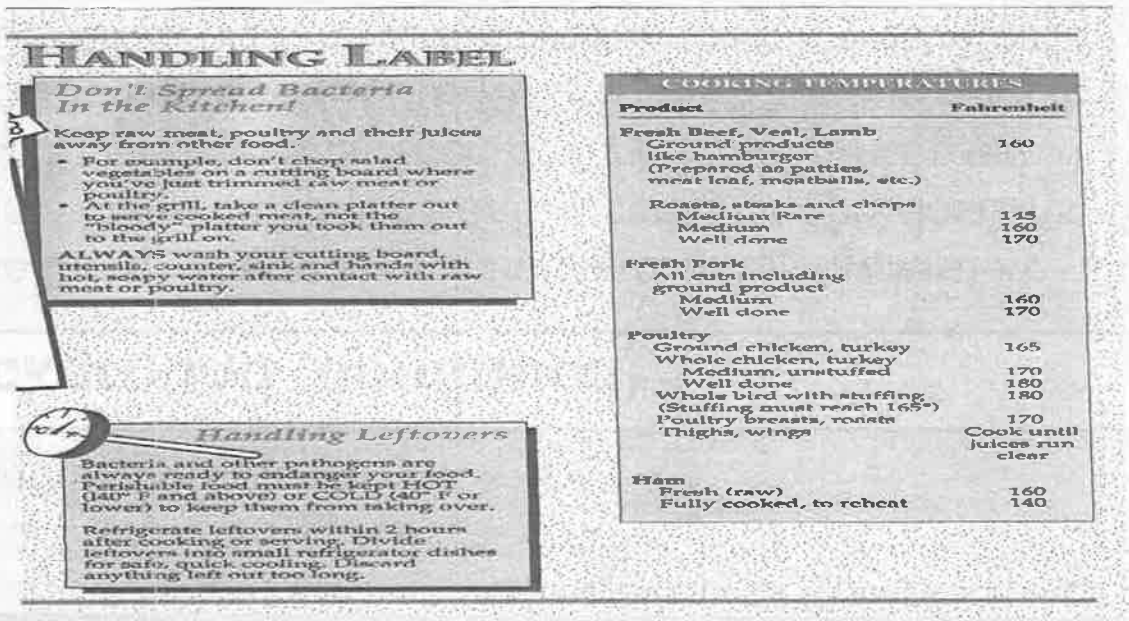
- In FSIS consumer publications and on labeling, consumers were given a variety of endpoint temperatures.

- The conclusion was that the only simple statement for SHI would be "cook thoroughly."

- Consumer endpoint cooking temperature recommendations at the time are shown in the next slide in a consumer brochure developed as outreach material upon implementation of SHI, March 1994.

Food Safety and Inspection Service

Brochure for Consumer Endpoint Temperatures in 1993



HANDLING LABEL

Don't Spread Bacteria In the Kitchen!

Keep raw meat, poultry and their juices away from other food.

- For example, don't chop salad vegetables on a cutting board where you've just trimmed raw meat or poultry.
- At the grill, take a clean platter out to serve cooked meat, not the "bloody" platter you took them out to the grill on.

ALWAYS wash your cutting board, utensils, counter, sink and hands with hot, soapy water after contact with raw meat or poultry.

Handling Leftovers

Bacteria and other pathogens are always ready to endanger your food. Perishable food must be kept HOT (140° F and above) or COLD (40° F or lower) to keep them from taking over.

Refrigerate leftovers within 2 hours after cooking or serving. Divide leftovers into small refrigerator dishes for safe, quick cooling. Discard anything left out too long.

COOKING TEMPERATURES

Product	Fahrenheit
Fresh Beef, Veal, Lamb	
Ground products like hamburger (Prepared as patties, meat loaf, meatballs, etc.)	160
Roasts, steaks and chops	
Medium Rare	145
Medium	160
Well done	170
Fresh Pork	
All cuts including ground product	
Medium	160
Well done	170
Poultry	
Ground chicken, turkey	165
Whole chicken, turkey	
Medium, unstuffed	170
Well done	180
Whole bird with stuffing (Stuffing must reach 165°)	180
Poultry breasts, roasts	170
Thighs, wings	Cook until juices run clear
Ham	
Fresh (raw)	160
Fully cooked, to reheat	140

Food Safety and Inspection Service

SHI – Final Rule Comments

- In comments to the proposal, six commenters stated that more explicit cooking instructions were needed.
- Some commenters recommended information on visual signs of doneness or final internal temperatures be required or at least permitted on SHI.
- One consumer group recommended visual keys over internal temperatures be included on SHI, as consumers may misunderstand internal temperatures to be cooking temperatures.

Food Safety and Inspection Service

SHI – Final Rule Comments

The FSIS response to these comments:

- FSIS does not believe it would be appropriate to add either an endpoint temperature or more comprehensive cooking directions because cooking temperatures and other visual indications of doneness vary by product; and

- SHI is not intended to replace comprehensive cooking statements that accompany many products.

Food Safety and Inspection Service

SHI – Final Rule

- FSIS concluded it was important to develop one SHI for all red meat and poultry products as opposed to different ones for different products.

- Given size limitations and the many varying endpoint temperatures depending on the product, "Cook Thoroughly" was the only simple, single statement that was appropriate.

Food Safety and Inspection Service

Current SHI

Label as described in Code of Federal Regulations, title 9, parts 317 and 381.

Safe Handling Instructions

This product was prepared from inspected and passed meat and/or poultry. Some food products may contain bacteria that could cause illness if the product is mishandled or cooked improperly. For your protection, follow these safe handling instructions.

Keep refrigerated or frozen. Thaw in refrigerator or microwave.

Keep raw meat and poultry separate from other foods. Wash working surfaces (including cutting boards), utensils, and hands after touching raw meat or poultry.

Cook thoroughly.

Keep hot foods hot. Refrigerate leftovers immediately or discard.

Actual size

Safe Handling Instructions

This product was prepared from inspected and passed meat and/or poultry. Some food products may contain bacteria that could cause illness if the product is mishandled or cooked improperly. For your protection, follow these safe handling instructions.

Keep refrigerated or frozen. Thaw in refrigerator or microwave.

Keep raw meat and poultry separate from other foods. Wash working surfaces (including cutting boards), utensils, and hands after touching raw meat or poultry.

Cook thoroughly.

Keep hot foods hot. Refrigerate leftovers immediately or discard.

Enlarged

Food Safety and Inspection Service

Current Recommended Cooking Temperatures for Consumers

- FSIS now recommends only 3 internal temperatures for consumers – 145°F and hold for 3 minutes for all whole muscle red meat, 160°F for ground red meat and 165°F for all poultry.
- With only 3 temperature recommendations, this information could be incorporated into the SHI requirements through rulemaking.

Food Safety and Inspection Service

SHI Changes Suggested

- Over the years FSIS has been approached by various organizations and consumer groups regarding the effectiveness of the current SHI.
- These organizations have recommended updates to the SHI to improve its effectiveness to consumers.

Food Safety and Inspection Service

Partnership for Food Safety Education

- In 2010 the Grocery Manufacturers Association, National Meat Association, and National Turkey Federation approached FSIS with a request for a waiver of the mandatory SHI label in order to conduct pilot testing that would utilize the Partnership for Food Safety Education's consumer tested Be Food Safe icon.

Food Safety and Inspection Service

Partnership for Food Safety Education

- The waiver was withdrawn with the intent to resubmit once new icons were developed and branded under USDA's new Food Safe Families campaign. The Food Safe Families campaign is a nationwide consumer food safety media campaign launched in 2011 as a collaborative effort the Ad Council, the Partnership for Food Safety Education, FDA, and CDC.

Food Safety and Inspection Service

Be Food Safe Icon for Use in Waiver

SAFE HANDLING INSTRUCTIONS

THIS PRODUCT WAS PREPARED FROM INSPECTED AND PASSED MEAT AND/OR POULTRY. SOME FOOD PRODUCTS MAY CONTAIN BACTERIA THAT COULD CAUSE ILLNESS IF THE PRODUCT IS MISHANDLED OR COOKED IMPROPERLY. FOR YOUR PROTECTION FOLLOW THESE SAFE HANDLING INSTRUCTIONS.

WASH WORKING SURFACES (INCLUDING CUTTING BOARDS, UTENSILS, AND HANDS AFTER TOUCHING RAW MEAT OR POULTRY.

KEEP RAW MEAT AND POULTRY SEPARATE FROM OTHER FOODS.

COOK THOROUGHLY. KEEP HOT FOODS HOT.

KEEP REFRIGERATED OR FROZEN. THAW IN REFRIGERATOR OR MICROWAVE. REFRIGERATE LEFTOVERS IMMEDIATELY OR DISCARD.

www.befoodsafe.org

Food Safety and Inspection Service

Proposal on Mechanically Tenderized Beef

On August 9, 2013 FSIS proposed to require validated cooking instructions on raw or partially cooked mechanically tenderized beef products.

- FSIS proposed to require that the validated cooking instructions include, at a minimum: (1) the method of cooking; (2) a minimum internal temperature validated to ensure that potential pathogens are destroyed throughout the product; (3) whether the product needs to be held for a specified time at that temperature or higher before consumption; and (4) instruction that the internal temperature should be measured by the use of a thermometer.

- FSIS is currently evaluating the comments on this proposal.

Food Safety and Inspection Service

Salmonella Action Plan

Focus the Agency's Education and Outreach Tools and Resources on Salmonella

- ❑ New approaches or routes for providing Salmonella-related food safety messages to the public could help decrease Salmonella illnesses. Therefore, FSIS will:
  - ✓ provide more detailed messages related to Salmonella, such as explaining why we recommend or require certain practices, and will clearly communicate why Salmonella is more challenging than some other pathogens (medium term);
  - ✓ provide easy to find information about Salmonella trends in easy to read formats (medium term); and
  - ✓ work with the Food and Nutrition Service (FNS) to provide audience-appropriate, multilingual, Salmonella-specific food safety information to consumers through FNS nutrition programs (long term).

Labeling is a means to reach consumers.

Food Safety and Inspection Service

Stakeholder Letter

- To gain feedback, on November 20, 2013, a letter was sent to consumer groups and other stakeholders posing questions about the current labels and how to revise.
- Nine comments were received, two from consumer groups, two from industry groups and five from universities.

Food Safety and Inspection Service

Comments Summary

- Comments stated that SHI needed to continue to be mandatory.
- Comments supported the need for consumer testing of any changes to SHI messages.
- Most suggested flexibility to add voluntary validated cooking instructions. But no consensus on what should be included in validated cooking instructions.
- Some wanted a stronger message about bacteria in food products.

Food Safety and Inspection Service

Additional Comments

- Some suggested more details while others wanted the SHI streamlined with less detail and larger fonts. (SHI was developed as a reminder to consumers of the importance of handling foods safely.)

- Some recommended web-links or QR codes be added to the SHI so consumers could easily obtain more information.

Food Safety and Inspection Service

Questions

- Should FSIS pursue changes to SHI? If so, what changes beyond those suggested by comments would you suggest?
- Should the SHI include validated cooking instructions?
- If FSIS required validated cooking instructions on specific products, is the general statement still necessary?
- Should SHI include instructions to use a thermometer to measure endpoint temperature?

Food Safety and Inspection Service

Questions

- How much flexibility should be built into the SHI if FSIS proposes changes?
- Should there be a variety of statements for different products?
- Should FSIS revisit the waiver submitted by industry to gather data on the proposed graphic included in the waiver?
- Should FSIS make changes to SHI to encourage use of food safety interventions?
- Should FSIS pursue measures beyond SHI for providing information to consumers on handling product safely? What off-label mechanisms would you suggest?

National Advisory Committee on Meat and Poultry Inspection
January 7 – 8, 2014
Washington, DC

Subcommittee on
Food Safety Handling Labels

Report and Recommendations based on FSIS proposed questions

1. Should FSIS pursue changes to Safe Handling Instructions (SHI)? If so, what changes beyond those suggested by comments would you suggest?

Yes - The subcommittee believes that FSIS should pursue changes to the existing SHI. The subcommittee believes there is value in continued requirement of SHI with modifications or refinements. Subcommittee recommendations include:

- Eliminating the first statement of the existing SHI. The second statement should be retained with the addition of wording directing the use of thermometers for checking temperatures.
- Highly prioritizing FSIS engagement with risk communication specialists from other agencies (local, state, federal), academic/extension specialists, and other food safety stakeholders to develop effective strategies.
- Federal food safety regulatory agencies seeking to increase consistency of SHI messaging for all foods
  An expert risk communications panel should determine the most effective mechanism(s) based on consumer/performance testing to include high risk groups.
- Federal food safety regulatory agencies seeking more simplification and standardization of SHI symbols with an overall goal of one universally recognized method for communicating safe handling of food.
- NACMPI members agree that communication of end point temperatures to consumers is vital. FSIS should strongly consider adding them to SHI labeling if supported by available consumer testing data.
- FSIS should place a very high priority on updating SHI content and format in a timely manner based on the best available information with feedback to the NACMPI committee.
- Ensuring risk based communication of content - emphasizing the consumer practices that most impact food safety.

2. Should the SHI include validated cooking instructions?

No.

A) If FSIS required validated cooking instructions on specific products, is the general statement still necessary?

Yes - The general SHI statement should still be included.

The subcommittee felt that, while validated cooking instructions should not be required in SHI, if there are not validated cooking instructions on the label, FSIS should consider including crucial end point temperatures (this would need to be consumer tested to determine effectiveness).

B) Should SHI include instructions to use a thermometer to measure endpoint temperature?

Subcommittee recommended:

- Conduct consumer testing to determine the most effective method (symbol and/or words) to communicate the importance of cooking foods to recognized end point cooking temperatures using a thermometer to monitor temperatures. Note: consumer representatives emphasized the importance of providing consumer end point temperatures. A temperature monitoring/thermometer symbol should be included to emphasize the importance of proper cooking.
- One concern noted: a universal temperature symbol might not effectively communicate differences between cooking, chilling and cold holding temperatures. This is another area that would need to be consumer tested to find the most recognizable and effective method of cold temperature handling.

3. How much flexibility should be built into the SHI if FSIS proposed changes?

Subcommittee recommended:

- **Against** providing flexibility for the revised SHI once it has been consumer tested and finalized. For consistency purposes, changes to the core content should not be made for various FSIS products.

- FSIS should identify core SHI symbols and/or text that should not be changed for consistency purposes but should allow industry flexibility to provide additional

label information using a well defined approval process that includes consumer testing / performance data. FSIS should consider how to develop efficient processes to review alternative labeling submissions. The subcommittee questioned if this additional label information should require formal FSIS review/approval or be handled under new generic labeling guidelines.

A) Should there be a variety of statements for different products?

No – SHI is awareness level information that should be consistently communicated across FSIS regulated products.

B) Should FSIS revisit the waiver submitted by industry to gather data on the proposed graphic included in the waiver?

Yes – This would be a good starting point for stimulating more rigorous evaluations of food safety risk communications and building partnerships. FSIS needs to develop an efficient process to evaluate possible alternative approaches that augment SHI that effectively communicates risk reduction behaviors to consumers. This should best be done in collaboration with risk communication experts from other agencies, academia and food safety stakeholders. Final models should be based on consumer testing / performance testing. They need to evaluate the data they have already gathered to determine if changes motivate desired behavioral changes of consumers. Consumer representatives stressed the importance of providing consumers with clear end point cooking temperatures. Their preference is to have this information on the label.

C) Should FSIS make changes to SHI to encourage use of food safety interventions?

No - The committee felt there were too many variables involved and would lead to even more consumer confusion.

4. **Should FSIS pursue measures beyond SHI for providing information to consumers on handling product safety? What off-label mechanisms would you suggest?**

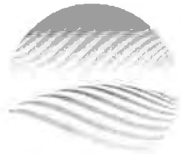
Yes – but since FSIS is a regulatory agency they should work in partnership with other appropriate groups to further educate consumers.

The committee agreed that SHI addresses only part of the food safety information needs of consumers. It was agreed that FSIS should work in partnership with risk communication experts from other agencies, universities, and food safety stakeholders to identify how SHI would fit within a comprehensive food safety risk communication strategy.

The key objectives should be to identify communication methods that effectively motivate consumers to use food handling/preparation behaviors that reduce food safety risks.

Possible mechanisms/elements of a comprehensive strategy:

- Partner with other food safety stakeholders to update food safety resources for use within schools
- Explore mechanisms to effectively leverage technologies to share food safety risk communication messages – examples: TV, radio, social media, extension outreach, food industry initiatives, websites
- Seek to establish/strengthen/better define a reasonably achievable consumer culture of food safety
- Establish/strengthen/create relationships with existing risk communication entities – increase consistency in messaging and sharing of expertise
- Consider more uniform use of an informational website on labels for consumers wanting more detailed information.
- Strongly urge the implementation/timing of any SHI changes be concurrent with new educational means to enhance consumer food safety practices



cfi THE CENTER FOR FOODBORNE ILLNESS RESEARCH & PREVENTION

Consumer Federation of America

U. S. Department of Agriculture
Food Safety and Inspection Service
1400 Independence Ave, SW
Washington DC 20250-3700

RE: FSIS-2015-0003
Notice of Request for New information: Gathering Sessions for Safe Food Handling Instructions

May 5, 20125

The Center for Foodborne Illness Research & Prevention[1] and Consumer Federation of America[2] appreciates the opportunity to comment on the Food Safety Inspection Service's (FSIS) proposal to collect data about consumer knowledge and perceptions related to the safe handling and preparation of raw or partially cooked meat and poultry products. FSIS will use this information to determine if a revision of the mandatory Safe Handling Instructions (SHI) label for meat and poultry products is warranted [Docket No. FSIS–2015–0003]. CFI and CFA agree that investigating the idea of revising the SHI label is warranted and encourages the approval of FSIS' plans to investigate this issue. These are initial comments and should not be considered complete.

Background:
In 1994, FSIS required all raw or partially cooked (*i.e.,* not considered ready-to-eat) meat[3] and poultry[4] products, destined for household consumers or institutional uses, to have a label with safe food-handling instructions affixed to the product. Since its adoption in 1994, this mandatory SHI label has not been revised.

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[1] The Center for Foodborne Illness Research & Prevention (CFI) is a national non-profit organization dedicated to advancing a stronger, more science-based food safety system that prevents foodborne illness and protects public health.
[2] Consumer Federation of America is a nonprofit association of 300 consumer groups, representing more than 50 million Americans, that was established in 1968 to advance the consumer interest through research, education and advocacy.
[3] Federal Register, Labels: Definition; Required Features. 35 FR 15580, Oct. 3, 1970.
[4] Federal Register, Special Handling Label Requirements, 37 FR 9706, May 16, 1972, as amended at 39 FR 4569, Feb. 5, 1974; 59 FR 14540, Mar. 28, 1994; 64 FR 746, Jan. 6, 1999.

Recently, FSIS has received inquiries from consumer groups and other stakeholders for more information about potential changes to the mandatory SHI label on meat and poultry products. In November 2013, FSIS sent a letter to consumer groups, industry groups and academia asking for feedback about the messages on the current SHI label and requested suggestions on potential revisions. Later, in January 2014, FSIS presented a summary of these stakeholders' responses to the National Advisory Committee on Meat and Poultry Inspection (NACMPI) and a subcommittee discussed the pros and cons of a potential revision.

The NACMPI Subcommittee on Food Safety Handling Labels issued a report indicating that FSIS should collect feedback from consumers about the types of revisions (to the SHI label) that would or would not be effective for improving consumer safe food handling and preparation of raw or partially cooked meat and poultry products.[5]

NACMPI members agreed on these points:

- The SHI label is important as a tool in FSIS' efforts to prevent foodborne. If a new SHI label is warranted, then the development and implementation of that new label should become a FSIS priority and be completed in a timely fashion.
- The objective of the label should be to effectively motivate consumers to use safe food handling/preparation behaviors to reduce food safety risks.
- The SHI label should contain well-researched food safety information that is consistently communicated throughout FSIS' food safety messages and programs.
- FSIS' proposed focus group strategy is fine, but it should include representatives from vulnerable populations or people who prepare food for vulnerable populations.
- The launch of the revised label (if consumer feedback indicates that a revision is warranted) should be timely and concurrent with a new educational campaign to enhance consumer food safety practices.

NACMPI discussed these topics at the January 2014 meeting:

End Point Temperatures

The current label says that meat and poultry products need to be "cooked thoroughly." While that statement is correct, FSIS may want to consider replacing vague instructions, such as "cook thoroughly," with specific end-point temperatures. Over the past twenty years, extensive research has documented that color is not a good indicator to determine if a meat or poultry product has reached a state of "thorough cooking," but many consumers still believe color is a good indicator of doneness.

On the other hand, multiple research studies have shown that the following end-point temperatures need to be achieved to kill internal foodborne pathogens:

- Poultry (whole birds, poultry parts and ground poultry) – 165 °F
- Ground meats – 160°F
- Whole-muscle meats – 145°F plus a stand time of 3 minutes

[5] NACMPI Subcommittee on Food Safety Handling Labels, January 7-8, 2014. Accessed on May 4, 2015 at http://www.fsis.usda.gov/wps/wcm/connect/70d59a26-32bb-4d25-9050-462b0b0cacb5/Food-Safe-Handling-Labels-NACMPI.pdf?MOD=AJPERES

To reduce the occurrence of foodborne diseases, consumers need to be aware of safe handling and preparation practices for meat and poultry products. Therefore, FSIS should consider listing the three end-point cooking temperatures on a revised meat and poultry SHI label. Consumers need to understand that temperature, not color, is the only way to determine if the product has been cooked thoroughly.

Thermometer Use

While the current label has a picture of a food thermometer on it, there is no text indicating that a thermometer must be used to determine if a meat or poultry product is cooked thoroughly. FSIS should consider adding information about thermometer use on a revised label.

Food Safety's Four Core Message Icons

Research conducted prior to the launch of USDA's 2012 *Food Safe Families* (FSF) campaign indicated that the new icons for the four core safe food practices (developed for the campaign) were preferred by consumers. Therefore, FSIS, in conjunction with other food safety agencies and stakeholders, should consider adopting FSF's icons or consider the benefits of conducting research on "icon effectiveness." The majority of NACMPI members, however, did not want "standardizing icons" to stand in the way of revising the SHI label. A revised SHI label will provide better meat and poultry safe handling information, which in turn will prevent the occurrence of foodborne illnesses.

Food Safety Contact Information

The current SHI label does not provide consumers with contact information for additional food safety information. FSIS should consider adding its website or a phone contact on the label, since some consumers may want more detailed information.

Comments

Each year in the United States, millions people are sickened, 128,000 are hospitalized and 3,000 die from foodborne illness. In addition, there are substantial economic costs incurred by the victims and retailers as they struggle to recover from food contamination events. Therefore, it is reasonable to plan and implement food safety education programs aimed at preventing foodborne disease.

Labels on food products have been effective in providing consumers with ingredient and nutritional information, so it is reasonable to also use labels on food to convey important food safety messages. Research conducted through the Centers for Disease and Prevention's Behavioral Risk Factor Surveillance System (BRFSS) showed that while less than half of consumers were aware of the safe food handling labels on meat and poultry products, 77% of the consumers who saw the label read it and 37% of those that read the label reported changing their handling and preparation practices based on the label instructions.[6]

[6] Centers for Disease Control and Prevention. Multistate Surveillance for Food-Handling, (1998) Preparation, and Consumption Behaviors Associated with Foodborne Diseases: 1995 and 1996 BRFSS Food-Safety Questions; *MMWR* 47(SS-4); 33-54. Accessed May 5, 2015 at http://www.cdc.gov/mmwr/preview/mmwrhtml/00054714.htm

In addition, the Partnership for Food Safety Education (PFSE) has conducted research on consumer perceptions and behaviors with regard to meat and poultry.[7] PFSE's research found consumers voicing these perceptions:

- Food safety, while important, is not as important as taste, i.e., cooks need to balance safety with the food's texture and moisture. Experience plays a big role in achieving this balance.
- Ground meat and poultry products are riskier foods than whole cuts.
- Food safety is more of a problem with pork and chicken because these products carry more "bacteria" or "pathogens" than beef does. Therefore, when determining if beef is completely cooked, there is more flexibility. Beef "doneness" is more of a preference decision, not a food safety one.
- Most consumers don't use a thermometer. While many consumers agree that thermometers are appropriate for large roasts or full birds, they also state that thermometers are not needed for other products. Experience and visual observations are other ways to determine doneness.
- USDA's recommended internal temperatures are higher than necessary for safety. Many consumers prefer the Food Network or other chefs' cooking recommendations.
- Allowing meat or poultry to "stand" after removing the food from its heat source is not related to food safety – instead, it is done to "seal the juices" and keep the product moist and tasty.
- "Stand time" does not complete the cooking process – once the product has been removed from the heat source, cooking stops. It is counterintuitive to think that cooking continues after removal from the heat source.

These perceptions reveal that consumers' knowledge about meat and poultry safety is limited and/or distorted. However, understanding the following key points about meat and poultry handling and preparation are critical in preventing foodborne illness:

- Cooking is the only "kill-step" that consumers have for destroying pathogens in raw and partially cooked meat and poultry products. Washing these products does not kill pathogens.
- Meat and poultry products need to reach a specific internal temperature for a specific amount of time in order to kill pathogens.
- Thermometers are the only method to determine if a meat or poultry product has reached a safe internal temperature. This is particularly important for foods containing ground meats (like hamburgers) or if the product has been tenderized (like is done with mechanically tenderized steaks and roasts).
- "Stand time" is required to ensure meat and poultry safety, since "stand time" completes the cooking process.

Given the major disconnects between consumer perceptions and verified safe handling information about meat and poultry products, it is likely that a revised SHI label would

[7] Partnership for Food Safety Education. PFSE New Cook Time Messaging Focus Groups Final Report. *Brand Amplitude®, LLC.* September 2, 2009

improve consumers' knowledge about important safe handling of meat and poultry products, thereby encouraging more consumers to adopt safer food handling and preparation practices.

Further, since cooking is the only "kill step" available to consumers, it is important that specific cooking instructions be provided to purchasers of raw and partially cooked meat and poultry products. By having the end-point temperatures listed on the SHI label, consumers should come to better appreciate the importance of using a thermometer to determine meat and poultry doneness. Further, given that consumers do not always have immediate access to USDA's recommended internal temperatures at the point of preparation, a label could become a primary source in verifying meat and poultry safety information.

Conclusion

According to the Federal Register notice, *Notice of Request for New information: Gathering Sessions for Safe Food Handling Instructions*, FSIS has contracted with RTI International to conduct six consumer focus groups to gather information on consumers' understanding and use of the current safe-handling instructions and responses to possible revisions to the instructions. To provide geographic diversity, FSIS will conduct two focus groups in three different geographic locations each with two focus groups (for a total of six). Locations will be representative of three of the four main geographical areas of the country (East Coast, South, Midwest, and West Coast). In each location, FSIS will conduct one focus group with English-speaking adults and one focus group with Spanish-speaking adults. The focus groups will include individuals at-risk for foodborne illness (*i.e.,* older adults, parents of young children, immune compromised individuals or their caregivers) as well as from the general population.

CFI and CFA agree that FSIS should be granted approval to collect data about consumer knowledge and perceptions related to meat and poultry safety in order to ascertain if a revision of the SHI label is appropriate. We also agree that the plans outlined in the above mentioned Federal Register notice are consistent with good research practices.

CFI and CFA appreciates the opportunity to submit these comments and looks forward to continuing our work with USDA/FSIS to develop appropriate and effective food safety education programs and messages for consumers.

Sincerely,

Patricia Buck
CFI Executive Director
Center for Foodborne Illness Research & Prevention

Chris Waldrop
Director, Food Policy Institute
Consumer Federation of America

Food Safety and Inspection Service:

Office of Public Affairs and Consumer Education

Requirements Gathering for Safe Handling Instructions

Chris Bernstein

Director, Food Safety Education

Office of Public Affairs and Consumer Education

Food Safety and Inspection Service

Food Safety and Inspection Service:

Timeline of Events

1993	• FSIS conducted focus groups to develop the SHI
1994	• FSIS published Final Rule requiring the SHI on raw and partially cooked meat and poultry products
2010	• GMA, NMA, NTF request waiver – Be Food Safe icon (now Food Safe Families)
2013	• FSIS sent stakeholder letter to obtain feedback on potential revisions to the SHI
2014	• FSIS solicited input at NACMPI meeting
FY2014	• FSIS initiated contract to gather information on consumer requirements for changes to the SHI
FY2016	• FSIS obtains results from requirements gathering contract.

Food Safety and Inspection Service:

Summary of Recommendations from 2014 NACMPI

- FSIS should pursue changes to the existing SHI.
  - Value in continued requirement of SHI with modifications or refinements.
- Validated cooking instructions should not be required in SHI.
- Include end point temperatures.
- No modification exemptions to the final revised SHI.

Food Safety and Inspection Service:

Requirements Gathering Contract Results

The Agency will consider moving forward with conducting a second round of consumer research to test alternative formats for the SHI if two or more of the following conditions are met:

	Conditions Meet?
Recommendation to Use a Food Thermometer	Yes
Recommendation to Add Endpoint Temperatures for Different Cuts of Meat/Poultry	Yes
Comprehension	No
Usefulness	Yes
Persuasiveness/Behavior Change	Yes
Recommendation to Add Source for More Information	Yes
Replace icons on current SHI with Food Safe Families (FSF) icons	No

Food Safety and Inspection Service:

Next Steps

- FSIS finished the Requirements Gatherings Contract this Spring.
- FSIS will work with a contractor to revise and/or redesign the SHI, including consumer testing.
- FSIS will work with the Social and Behavioral Science Team (SBST)
- FSIS will propose revisions for the new SHI and initiate rulemaking.

Food Safety and Inspection Service:

Questions

Food Safety and Inspection Service:

Office of Public Affairs and Consumer Education

Requirements Gathering for Safe Handling Instructions

Chris Bernstein

Director, Food Safety Education

Office of Public Affairs and Consumer Education

Food Safety and Inspection Service

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Food Safety and Inspection Service:

Questions

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doi:10.4315/0362-028X.JFP-11-058

Economic Burden from Health Losses Due to Foodborne Illness in the United States

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MS 11-058: Received 4 February 2011/Accepted 26 September 2011

ABSTRACT

The Centers for Disease Control and Prevention (CDC) recently revised their estimates for the annual number of foodborne illnesses; 48 million Americans suffer from domestically acquired foodborne illness associated with 31 identified pathogens and a broad category of unspecified agents. Consequently, economic studies based on the previous estimates are now obsolete. This study was conducted to provide improved and updated estimates of the cost of foodborne illness by adding a replication of the 2011 CDC model to existing cost-of-illness models. The basic cost-of-illness model includes economic estimates for medical costs, productivity losses, and illness-related mortality (based on hedonic value-of-statistical-life studies). The enhanced cost-of-illness model replaces the productivity loss estimates with a more inclusive pain, suffering, and functional disability measure based on monetized quality-adjusted life year estimates. Costs are estimated for each pathogen and a broader class of unknown pathogens. The addition of updated cost data and improvements to methodology enhanced the performance of each existing economic model. Uncertainty in these models was characterized using Monte Carlo simulations in @Risk version 5.5. With this model, the average cost per case of foodborne illness was $1,626 (90% credible interval [CI], $607 to $3,073) for the enhanced cost-of-illness model and $1,068 (90% CI, $683 to $1,646) for the basic model. The resulting aggregated annual cost of illness was $77.7 billion (90% CI, $28.6 to $144.6 billion) and $51.0 billion (90% CI, $31.2 to $76.1 billion) for the enhanced and basic models, respectively.

Accurate burden-of-illness estimates for foodborne diseases are useful for policy makers and others that seek to characterize and prioritize resources dedicated to addressing the problem of these diseases. Scallan et al. *(14, 15)*, in studies conducted for the Centers for Disease Control and Prevention, estimated that approximately 48 million new cases of food-related illness, resulting in 3,000 deaths and 128,000 hospitalizations, occur in the United States annually. These estimates, although confirming that foodborne illness continues to be a problem, are significantly lower than the previous estimates by Mead et al. *(12)* of 76 million cases, 5,000 deaths, and 325,000 hospitalizations. The burden associated with specific pathogens, relative to others, also has changed. For example, *Clostridium perfringens* is now believed to cause more than 26% of food-related bacterial illnesses, in contrast to the less than 6% estimated by Mead et al. *(12)*. Because of extensive methodological improvements employed by Scallan et al., it is not clear to what extent, if any, the differences in estimates are driven by true changes in the burden of illness in the population. Regardless of the reason for the differences, the results of the economic burden of foodborne illness studies conducted by Scharff et al. in 2009 *(18)* and Scharff in 2010 *(16)*, which were based on the estimates provided by Mead et al. in 1999, are now outdated.

A comparison of the 2011 and 1999 estimates. The Scallan et al. *(14, 15)* 2011 estimate for the burden of foodborne illness is not simply an update of the older 1999 Mead et al. *(12)* numbers. Major changes in both methodology and representation of risk are evident in the newer studies. For this reason, although the numbers are smaller, the authors caution readers not to see the difference as representing an overall downward trend in the burden of foodborne illness. Nevertheless, because updated data were included in the Scallan et al. analysis, any trends occurring during this period would have been subsumed in the final numbers presented. For example, most of the increase in the estimated number of cases of *Vibrio vulnificus* infection was due to a more than doubling of identified passive surveillance cases. Important methodological changes in the Scallan et al. study included the introduction of uncertainty, the disaggregation of underdiagnosis and underreporting factors, and the exclusion of travel-related illnesses. The inclusion of uncertainty in the Scallan et al. model is an important improvement that removes the false confidence implied by the Mead et al. point estimates. Scallan et al. estimated a 90% credible interval of 28.7 to 71.1 million illnesses, with a mean of 47.8 million cases. This characterization has the advantage of giving policy

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TABLE 1. *Burden of foodborne illness expressed as annual number of cases*[a]

Disease or agent	No. of illnesses	No. of hospitalizations	No. of deaths
Bacteria			
Bacillus cereus	63,400	20	0
Brucella spp.	839	55	1
Campylobacter spp.	845,024	8,463	76
Clostridium botulinum	55	42	9
C. perfringens	965,958	438	26
STEC O157:H7[b]	63,153	2,138	20
STEC non-O157	112,752	271	0
ETEC[c]	17,894	12	0
Other diarrheagenic *Escherichia coli*	11,982	8	0
Listeria monocytogenes	1,591	1,455	255
Mycobacterium bovis	60	31	3
Salmonella, nontyphoidal	1,027,561	19,336	378
S. enterica Typhi	1,821	197	0
Shigella	131,254	1,456	10
Staphylococcus aureus	241,148	1,064	6
Streptococcus group A	11,217	1	0
Vibrio cholerae, toxigenic	84	2	0
V. vulnificus	96	93	36
V. parahaemolyticus	34,664	100	4
Other *Vibrio*	17,564	83	8
Yersinia enterocolitica	97,656	533	29
Parasite			
Cryptosporidium spp.	57,616	210	4
Cyclospora cayetanensis	11,407	11	0
Giardia intestinalis	76,840	225	2
Toxoplasma gondii	86,686	4,428	327
Trichinella spp.	156	6	0
Virus			
Astrovirus	15,433	87	0
Hepatitis A	1,566	99	7
Norovirus	5,461,731	14,663	149
Rotavirus	15,433	348	0
Sapovirus	15,433	87	0
Total known	9,388,074	55,962	1,350
Total unknown	38,392,704	127,839	1,686
Grand total	47,780,778	183,801	3,036

[a] Data from Scallan et al. *(14, 15)*; see these references for estimates of uncertainty associated with these illnesses.
[b] STEC, Shiga toxin–producing *Escherichia coli*.
[c] ETEC, enterotoxigenic *E. coli*.

makers more information upon which to base potentially costly decisions. Scallan et al. also treated the effects of underreporting and underdiagnosis separately. The updated Scallan et al. estimates that were used in this study are presented in Table 1.

New economic burden of foodborne illness estimates. The federal agencies that employ economic cost data in regulatory analyses typically use either a basic cost-of-illness model that includes values for medical care, productivity losses, and mortality or a cost-of-illness model enhanced to include pain and suffering values. The former is the method used by the Economic Research Service of the U.S. Department of Agriculture (USDA), and the latter has historically been used by the Center for Food Safety and Applied Nutrition at the U.S. Food and Drug Administration *(6, 20, 22)*. Recently, the USDA Food Safety and Inspection Service has also used the enhanced method in at least one regulatory analysis *(21)*. There are advantages associated with each method. By including a value for pain and suffering, the enhanced model has the advantage of more fully accounting for economic costs associated with foodborne illness. This value is derived by monetizing quality-adjusted life years (QALYs) that have been designed to assess utility loss. The QALYs used in this study were based on individuals' trade-offs between the amount of time with good health and the amount of time with given symptoms and activity limitations (the time trade-off method). This approach yields a measure of the loss of well-being (typically between 0 and 1). Monetized QALY losses are the product of loss of well-being from a condition, the number of days with the condition, and the economic

value of 1 day (derived from the value of statistical life) *(23)*. Ideally, this measure would represent the ill consumer's willingness to pay to avoid these pain and suffering losses. However, this only occurs under restrictive conditions, leading to a split in opinion regarding whether this model should be used *(1, 8, 9, 13)*. In contrast, the basic model avoids the controversy over how QALYs should be used but does not provide a value for the legitimate economic costs associated with pain and suffering. In this article, estimates for both methods are provided.

Rationale and objectives. Previous estimates of the cost of foodborne illness based on the data of Mead et al. *(12)* are no longer valid following the release of the 2011 Scallan et al. articles *(14, 15)*. Thus, updated estimates are needed. The primary objective of this study was to provide updated estimates of the economic cost of foodborne illness based on an integration of the Scallan et al. and the Scharff et al. *(18)* models. Features of the integrated model include (i) a full replication of the Scallan et al. model; (ii) estimations of economic cost for both the basic and enhanced cost-of-illness model; (iii) use of updated cost data; (iv) employment of methodological improvements to the cost models; (v) characterization of uncertainty; and (vi) characterization of costs at the aggregate level and for specific pathogens.

MATERIALS AND METHODS

The methodological changes made by Scallan et al. *(14, 15)* clearly affected the predicted number of illnesses from foodborne sources. The Scallan et al. method also led to changes in the burdens associated with illness. These changes are likely to affect both the average cost per case and the aggregated annual social cost of foodborne illness. The following model derives updated estimates of the cost of foodborne illness by combining the model described previously by Scharff et al. *(16, 18)* (including limited methodological changes) with the Scallan et al. model.

Evaluation of uncertainty. Uncertainty associated with both the illness and economic components of the model is characterized using Monte Carlo simulation modeling in @Risk version 5.5 (Palisade Corp., Ithaca, NY). The characteristics of the respective models' measures of uncertainty must be accurately preserved because both the Scallan et al. and Scharff models incorporate uncertainty in a comprehensive manner. Given that the cost-of-illness model provides the base, the primary concern is how to accurately incorporate the uncertainty described in the Scallen et al. model. Two options for preserving uncertainty are available. First, one could simply construct Beta distributions that yield values that match the Scallan et al. means and credible intervals (CIs). However, this approach would ignore the shape of the distributions, an important omission given that the combinations of multiple distributions used by Scallan et al. yield, in some cases, multimodal distributions. To most closely preserve the characteristics of uncertainty in the Scallan et al. model, I instead replicated this model using the detailed technical appendices that were made available. My replication of the Scallan et al. model yields similar, though not identical, estimates. My estimate of known illnesses is within 0.1% of theirs, and hospitalizations and deaths are within 2.0 and 0.5%, respectively. These small disparities probably are due to differences in how the Monte Carlo analyses were performed. For the present study, the mean values were adjusted to correspond with the Scallan et al. estimates. This preserves both their values and the underlying distributions they used.

Two cost-of-illness models. Both the basic and enhanced cost-of-foodborne-illness models account for health-related economic costs associated with foodborne illness. In the basic model, economic costs from foodborne illness include both financial losses due to medical expenditures and lost productivity and lost utility (well-being) due to death. The losses associated with each given pathogen i are summarized in equation 1:

$$\text{Cost}_i = \text{Hospital}_i + \text{Physician}_i + \text{Pharma}_i + \text{Prod}_i + \text{CProd}_i + \text{VSL}_i + \text{Sequel}_i \quad (1)$$

Medical costs include costs of hospital services (Hospital_i) not including physician care, physician care (Physician_i) including the cost of lab work and both inpatient and outpatient care, and pharmaceutical costs (Pharma_i). Financial costs also are incurred when individuals are not able to work as a result of either their own illnesses or the illnesses of their children (Prod_i and CProd_i). The value of statistical life (VSL_i) figure used is based on a published meta-analysis of dozens of studies of individuals' trade-offs between fatality risk and money *(23)*. For example, if the most an individual is willing to pay for a reduction in fatality risk of 1/10,000 is \$500, the VSL would be \$5 million (\$500 = VSL/10,000). Because in some cases acute illnesses lead to other often chronic conditions, the costs associated with these conditions are included (Sequel_i). Specifically, costs associated with Guillain-Barre disease (*Campylobacter*), hemolytic uremic syndrome with or without end-stage renal disease (*Escherichia coli*), newborn complications (*Listeria*), and reactive arthritis (*Campylobacter, Salmonella, Shigella,* and *Yersinia*) are included. For each cost measure described above, uncertainty is fully incorporated into the model.

The enhanced cost-of-illness model also incorporates a value for pain and suffering (equation 2):

$$\text{Cost}_i = \text{Hospital}_i + \text{Physician}_i + \text{Pharma}_i + \text{CProd}_i + \text{QALY}_i + \text{VSL}_i + \text{Sequel}_i \quad (2)$$

The difference between the two models is that the enhanced model includes a measure for lost quality of life (QALY_i) but no measure for own-illness productivity loss (Prod_i). Prod_i is omitted because lost productivity from one's own illness is assumed to be accounted for in the more global QALY_i value (QALY losses are based in part on functional disability resulting from illness). The QALY variable, when it is used, includes losses from pain, suffering, and functional disability. It is estimated based on the value of a statistical life year (VSLY) adjusted for the number of days ill. VSLY is a measure of the implicit value that individuals place on the loss of a year of life and is estimated as the VSL divided by the discounted ($r = 3\%$) number of life years (L) remaining for the average person: $\text{VSLY} = (r \times \text{VSL})/[1 - (1 + r)^{-L}]$ *(24)*. For example, if you have an illness that results in a QALY loss of 0.1 for 3 days and the VSLY is \$357,000, the economic loss is calculated as $0.1 \times (3 \div 365) \times \$357{,}000 = \$293$. In the medical literature, QALYs are often implicitly valued at the lower rate of \$100,000 *(10)*. Estimates based on this valuation can be found in the sensitivity analysis.

Details regarding the sources and methods used to derive these costs were described more fully by Scharff et al. *(16, 18)* and in the appendix to this article (http://go.osu.edu/ehe-efa7d). Changes made to these base models are as follows. The most significant changes are based on data presented by Scallan et al. in 2011 *(14, 15)*. Specifically, the new Scallan et al. estimates for

TABLE 2. *Cost per case of foodborne illness by pathogen, 2010*

Disease or agent	Cost per case (U.S. dollars)						
		Productivity loss				Total cost per case	
	Medical care	Ill person	Caregiver	Quality of life	Death (VSL)	Prod	QALY
Bacteria							
Bacillus cereus	49	58	59	126	0	166	234
Brucella spp.	2,663	2,782	2,855	7,178	8,857	17,157	21,553
Campylobacter spp.	216	350	360	6,789	776	1,846	8,141
Clostridium botulinum	106,030	20,534	21,076	357,845	1,195,952	1,343,592	1,680,903
C. perfringens	50	73	75	160	197	395	482
STEC O157:H7[a]	693	398	409	849	8,097	9,606	10,048
STEC non-O157	90	398	408	868	0	896	1,366
ETEC[b]	58	398	408	868	0	863	1,334
Other diarrheagenic *Escherichia coli*	58	397	408	869	0	863	1,335
Listeria monocytogenes	59,377	1,819	1,867	46,197	1,174,628	1,272,279	1,282,069
Salmonella, nontyphoidal	401	647	568	7,421	2,697	4,312	11,086
S. enterica Typhi	2,499	933	861	8,128	0	4,293	11,488
Shigella	179	555	570	8,244	558	1,956	9,551
Staphylococcus aureus	87	133	136	289	183	539	695
Streptococcus group A	46	645	662	1,411	0	1,353	2,119
Vibrio cholerae, toxigenic	249	618	635	1,343	0	1,502	2,226
V. vulnificus	40,852	848	870	2,086	2,748,273	2,790,553	2,792,171
V. parahaemolyticus	90	500	514	1,095	853	1,957	2,551
Other *Vibrio*	91	500	514	1,094	3,322	4,426	5,020
Yersinia enterocolitica	126	883	906	8,125	2,176	4,186	11,334
Parasite							
Cryptosporidium spp.	82	722	741	1,581	511	2,056	2,916
Cyclospora cayetanensis	64	442	453	966	0	958	1,483
Giardia intestinalis	70	1,060	1,088	2,324	190	2,408	3,672
Toxoplasma gondii	2,735	2,650	2,719	6,760	27,655	35,759	39,869
Trichinella spp.	718	4,328	4,442	9,945	0	9,487	15,104
Virus							
Astrovirus	105	356	365	776	0	827	1,247
Hepatitis A	1,213	928	952	2,094	32,814	35,907	37,073
Norovirus	83	122	125	265	200	530	673
Rotavirus	188	303	311	654	0	802	1,154
Sapovirus	77	303	311	661	0	691	1,049
Total known	171	255	251	1,941	1,111	1,786	3,458
Total unknown	81	240	247	528	322	890	1,178
Grand total	99	243	248	805	477	1,068	1,626

[a] STEC, Shiga toxin–producing *Escherichia coli*.
[b] ETEC, enterotoxigenic *E. coli*.

hospitalization and death rates replace the older Mead et al. *(12)* estimates that had been used. For most pathogens, Scallan et al. defined the proportion of the population that "seeks care" as a variable for their underdiagnosis multiplier. The seeks care variable was used as a proxy for the probability of visiting a doctor, where it has been defined. This seeks care variable is an imperfect proxy because some patients may visit a doctor more than once for an illness, whereas others may visit the emergency room and be hospitalized without first visiting a doctor.

A second set of changes involves updating values to reflect the most recent data available. Estimates of hospital care costs and lengths of hospitalization from the Healthcare Cost and Utilization Project have been updated to include data through 2008 *(2)*. Productivity loss estimates have been updated to reflect the reported average hourly cost of compensation in September 2010 *(5)*. All estimates were updated to 2010 U.S. dollars using the consumer price index relevant to the economic sector at issue (e.g., the consumer price index for physician services was used to inflate 2009 physician visit costs to obtain 2010 values) *(4)*.

The final changes reflect independent improvements to the model. One change involves adjustment of the VSL (based on a 2003 estimate from a widely used meta-analysis) to account for income changes and inflation. In the 2009 Scharff et al. study *(18)*, this value was not adjusted. This simple assumption was replaced in the present study with the 2009 Bellavance et al. *(3)* estimate for the income elasticity of VSL (elasticity is assumed to be uniformly distributed between 0.84 and 1.08). Income elasticity of VSL is defined as the ratio of the percentage change in VSL to the

TABLE 3. *Economic cost of foodborne illness, basic cost of illness model, 2010*

Disease or agent	Cost per case (U.S. dollars)		Total cost (millions of U.S. dollars)	
	Mean	90% CI	Mean	90% CI
Bacteria				
Bacillus cereus	166	78–235	11	2–28
Brucella spp.	17,157	9,305–26,402	14	8–24
Campylobacter spp.	1,846	995–4,110	1,560	437–4,031
Clostridium botulinum	1,343,592	89,000–8,010,000	74	4–416
C. perfringens, foodborne	395	187–1,378	382	45–1,443
STEC O157:H7[a]	9,606	3,447–23,513	607	121–1,827
STEC non-O157	896	845–989	101	11–273
ETEC[b]	863	842–908	15	0–41
Other diarrheagenic *Escherichia coli*	863	842–908	10	0–28
Listeria monocytogenes	1,272,279	81,000–3,904	2,025	95–6,613
Salmonella, nontyphoidal	4,312	1,558–10,042	4,430	1,479–10,881
S. enterica Typhi	4,293	2,389–6,925	8	0–24
Shigella	1,956	1,285–5,439	257	38–768
Staphylococcus aureus, foodborne	539	318–1,992	130	29–434
Streptococcus group A, foodborne	1,353	1,342–1,365	15	0–112
Vibrio cholerae, toxigenic	1,502	1,311–1,762	0.1	0–0.3
V. vulnificus	2,790,843	585,000–5,297,000	268	54–538
V. parahaemolyticus	1,957	1,085–4,752	68	29–169
Other *Vibrio*	4,426	1,573–9,963	78	28–179
Yersinia enterocolitica	4,186	1,899–16,473	409	69–1,662
Parasite				
Cryptosporidium spp.	2,056	1,504–4,911	118	21–394
Cyclospora cayetanensis	958	933–1,059	11	0–39
Giardia intestinalis	2,408	2,249–2,617	185	128–267
Toxoplasma gondii	35,759	13,349–62,961	3,100	1,112–5,726
Trichinella spp.	9,487	8,818–10,748	1	0–4
Virus				
Astrovirus	827	794–864	13	5–22
Hepatitis A	35,907	9,509–68,261	56	13–125
Norovirus	530	369–709	2,896	1,545–4,728
Rotavirus	802	730–883	12	4–21
Sapovirus	691	672–713	11	4–18
Total known	1,976	894–3,200	16,865	8,436–29,230
Total unknown	890	619–1,363	34,182	21,047–51,404
Grand total	1,068	683–1,646	51,048	31,214–76,142

[a] STEC, Shiga toxin–producing *Escherichia coli*.
[b] ETEC, enterotoxigenic *E. coli*.

percentage change in income and is preferred to inflation as an adjustment measure because most VSL studies are based on trade-offs between risk and wages. VSL increased from $6.7 to $7.3 million between 2003 and 2010. Another change involves how QALY losses are estimated. The previous assumption was that an individual who becomes ill would have been in perfect health (QALY = 1) if not for the illness. However, Luo et al. *(11)* demonstrated that the average QALY in the United States is actually 0.87. As a result, a QALY of 0.689 (representing utility with a mild foodborne illness) now corresponds to a loss of 0.181 QALYs, as opposed to the previously estimated loss of 0.311 QALYs. Useful future research could further refine this measure to account for the preillness QALYs of persons who are afflicted with foodborne illnesses. Another change to the model was the addition of reactive arthritis as a sequela to *Campylobacter*, *Salmonella*, *Shigella*, and *Yersinia* infections. Economic costs were estimated based on the estimate by Townes et al. *(19)* for the risk of developing reactive arthritis following foodborne illness, medical cost data, productivity loss data, and QALY loss estimates adjusted to reflect age differences (as reported by FoodNet) using the Scharff and Jessup method *(7, 17)*. The inclusion of arthritis as a sequela increases the expected cost per case of foodborne illness for these pathogens by $99 in the basic model and by $5,979 (for *Campylobacter*) to $7,030 (for *Shigella*) in the enhanced model that includes a measure for pain and suffering.

RESULTS

Multiple estimates were produced using the specified models. Both the cost per case and the total cost of foodborne illness were determined for each pathogen studied and for foodborne illness as a whole. Although most of the Scallan et al. pathogens were included in the analysis, *Mycobacterium bovis* was not. However, this

TABLE 4. *Economic cost of foodborne illness, enhanced cost of illness model, 2010*

Disease or agent	Cost per case (U.S. dollars)		Total cost (millions of U.S. dollars)	
	Mean	90% CI	Mean	90% CI
Bacteria				
Bacillus cereus	234	37–517	15	1–46
Brucella spp.	21,553	8,097–38,337	18	7–35
Campylobacter spp.	8,141	1,793–19,764	6,879	1,134–20,129
Clostridium botulinum	1,680,903	200,000–8,403,000	93	10–435
C. perfringens	482	162–1,523	466	56–1,641
STEC O157:H7[a]	10,048	3,241–24,326	635	120–1,931
STEC non-O157	1,366	666–2,259	154	16–467
ETEC[b]	1,334	636–2,222	24	0–71
Other diarrheagenic *Escherichia coli*	1,335	636–2,226	16	0–48
Listeria monocytogenes	1,282,069	89,000–3,919,000	2,040	105–6,644
Salmonella, nontyphoidal	11,086	2,513–25,615	11,391	2,459–29,064
S. enterica Typhi	11,488	2,706–8,568	21	0–31
Shigella	9,551	1,860–23,609	1,254	105–4,526
Staphylococcus aureus	695	280–2,195	168	35–507
Streptococcus group A	2,119	986–3,565	24	0–170
Vibrio cholerae, toxigenic	2,226	1,126–3,623	0.2	0.1–0.5
V. vulnificus	2,792,171	585,000–5,300,000	268	54–538
V. parahaemolyticus	2,551	908–5,651	88	29–213
Other *Vibrio*	5,020	1,339–11,159	88	24–202
Yersinia enterocolitica	11,334	2,438–29,157	1,107	167–3,311
Parasite				
Cryptosporidium spp.	2,916	1,001–6,756	168	21–569
Cyclospora cayetanensis	1,483	702–2,475	17	0–63
Giardia intestinalis	3,672	1,516–7,107	282	108–597
Toxoplasma gondii	39,869	12,145–72,669	3,456	1,019–6,606
Trichinella spp.	15,104	7,024–25,507	2	1–6
Virus				
Astrovirus	1,247	558–2,396	19	5–44
Hepatitis A	37,073	9,033–71,112	58	12–130
Norovirus	673	301–1,106	3,677	1,424–6,912
Rotavirus	1,154	623–1,866	18	5–37
Sapovirus	1,049	515–1,766	16	5–34
Total known	3,458	1,012–7,201	32,462	9,542–66,780
Total unknown	1,178	499–2,168	45,208	18,128–84,939
Grand total	1,626	607–3,073	77,671	28,595–144,599

[a] STEC, Shiga toxin–producing *Escherichia coli*.
[b] ETEC, enterotoxigenic *E. coli*.

omission likely has a negligible effect on the overall cost of foodborne illness, given that only 60 domestically acquired cases of foodborne illnesses are annually attributable to this pathogen.

The cost per case for multiple cost categories is presented in Table 2, including the total cost per case for the basic (Prod) and enhanced (QALY) cost-of-illness models. The average cost associated with each case of foodborne illness is $1,068 (90% CI, $683 to $1,646) in the basic model and $1,626 (90% CI, $607 to $3,073) in the enhanced model. The total annual cost attributable to domestic infections from each pathogen (and for all foodborne illnesses) for the basic model is given in Table 3. The total health-related cost of foodborne illness in the United States is $51.0 billion (90% CI, $31.2 to $76.1 billion). The corresponding cost using the enhanced model (Table 4) is $77.7 billion (90% CI, $28.6 to $144.6 billion). The distribution of values produced by Monte Carlo simulation for each model is illustrated in Figure 1.

The uncertainty associated with the economic cost of each foodborne pathogen is provided in Tables 3 and 4. One improvement of the present model over previous models is that it includes uncertainty estimates for both economic factors and predicted illnesses. The result is a larger but more accurate CI for each estimate given. For example, in Scharff's 2010 study *(16)* the 90% CI for the total annual cost of *Yersinia* cases using the enhanced model was $150 to $1,369 million (mean, $674 million). The revised estimate is $1,107 million with a 90% CI of $167 to $3,311 million.

A sensitivity analysis was used to assess the relative effects of uncertain model parameters on the cost-of-illness outputs for the basic and enhanced models. In Figure 2, the

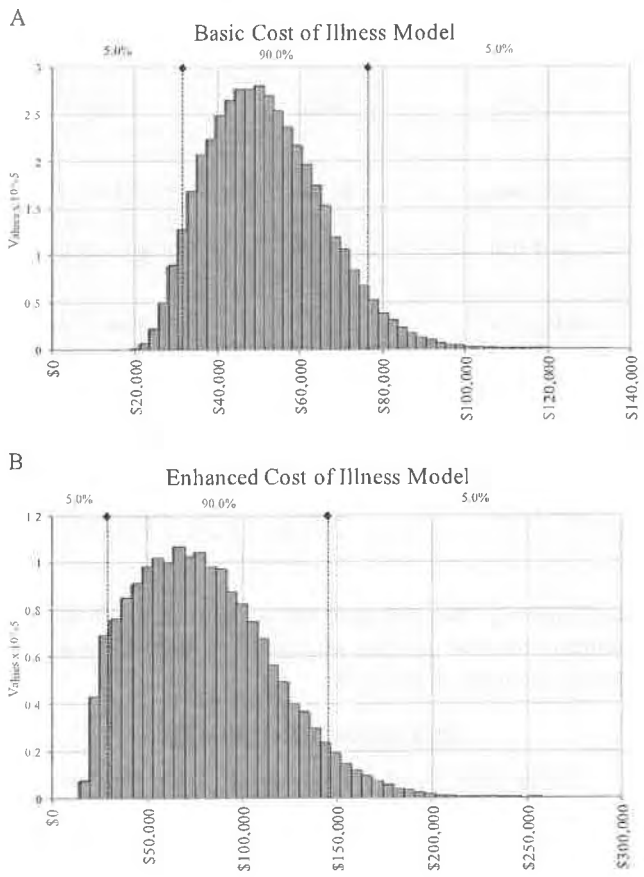

FIGURE 1. *Economic cost of foodborne illness in the United States (values given in millions of U.S. dollars).*

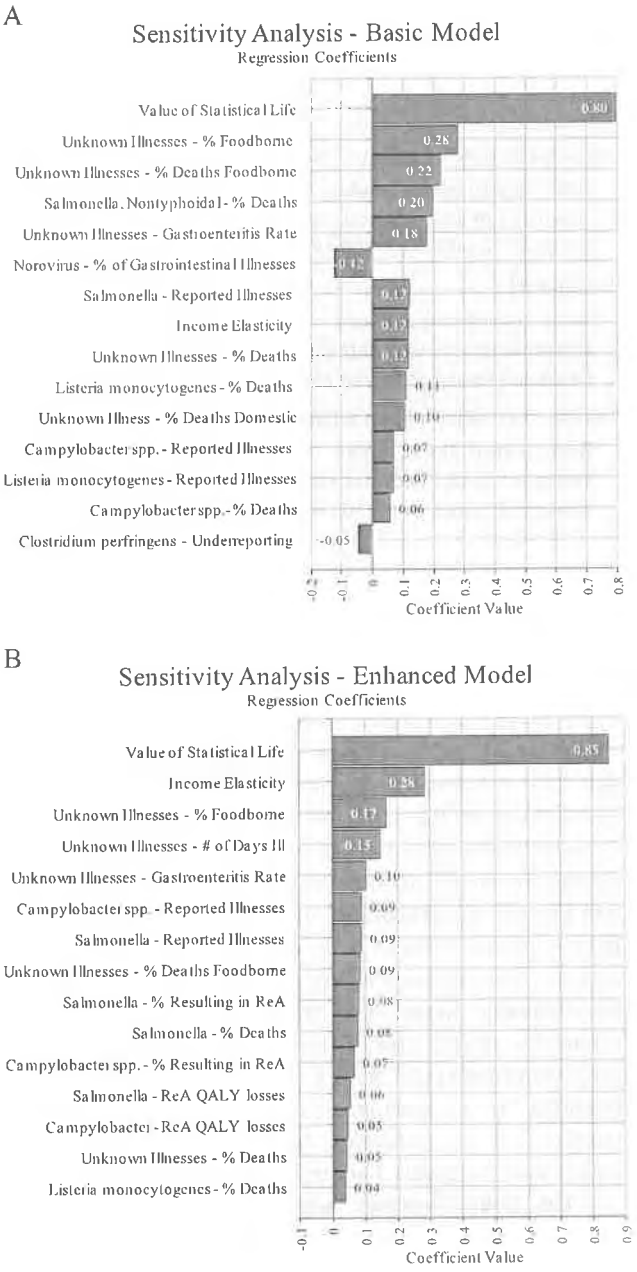

FIGURE 2. *Sensitivity of aggregate cost-of-illness estimates to model inputs.*

15 distributions (of a total of 800 used) that have the greatest effect on cost are illustrated for each model. For both models, the distribution used to estimate the VSL had the largest effect. This finding is not surprising given that death costs for all pathogens in both models (and QALY losses in the enhanced model) were estimated using this distribution. The only other economic measurement that made the list was the (related) income elasticity parameter. The bulk of the parameters that drove the results were related to illness disposition. Those pathogens associated with a high incidence of illness rate (unknown illness, campylobacteriosis, salmonellosis, and norovirus infection) and a high death rate (listeriosis) played a substantial role in each model, although a relatively larger role in the basic model. Conversely, those pathogens causing large quality of life losses, specifically those with large losses from reactive arthritis, played a larger role in the enhanced model (salmonellosis and campylobacteriosis). The importance of both economic and illness disposition distributions in the models illustrates the value of fully incorporating uncertainty for both burden of illness and economic components.

Estimates based on three alternative economic and illness models are shown in Table 5. In the base Scallan et al. model, the values derived in the replication of the Scallan et al. model were adjusted to match the means presented by Scallan et al. The unadjusted model does not include this adjustment. In the population adjusted model, the number of illnesses from the base model (across all categories of pathogen and severity) was proportionally increased to reflect the population increase between 2006 (the year of the Scallan et al. estimates) and 2010. The differences in costs among these models are minimal. For each illness model, three economic models were used. The key difference between these models is whether utility losses from pain and suffering are included. The enhanced model with a full value QALY produced the highest estimated costs, whereas the basic cost-of-illness model and enhanced model with a $100,000 QALY model had the lowest values. Because QALYs are theoretically expected to include aspects of both productivity loss and pain and suffering, these low numbers provide evidence that the $100,000 QALY, which has been used by many researchers, is significantly undervalued.

TABLE 5. *Alternative estimates of the economic cost of foodborne illness, 2010*

Disease or agent	Cost per case (U.S. dollars)		Total cost (millions of U.S. dollars)	
	Mean	90% CI	Mean	90% CI
Adjusted to match Scallan et al.				
Basic cost-of-illness model	1,068	683–1,646	51,048	31,214–76,142
Enhanced model with full value QALY	1,626	607–3,073	77,671	28,595–144,599
Enhanced model with $100,000 QALY	1,029	638–1,620	49,158	29,695–73,899
Unadjusted				
Basic cost-of-illness model	1,094	684–1,710	51,300	30,844–77,461
Enhanced model with full value QALY	1,640	607–3,096	76,855	28,005–142,915
Enhanced model with $100,000 QALY	1,054	638–1,686	49,392	29,302–75,389
Scallan et al. adjusted and population adjusted				
Basic cost-of-illness model	1,068	683–1,646	52,712	32,232–78,623
Enhanced model with full value QALY	1,626	607–3,073	80,202	29,527–149,312
Enhanced model with $100,000 QALY	1,029	638–1,620	50,760	30,663–76,307

DISCUSSION

In this study, the estimated cost of foodborne illness was substantial: $51.0 billion in annual health-related costs in the basic model and $77.7 billion in the enhanced model. These values are lower than previous estimates of $102.7 and $151.6 billion, respectively, primarily because of the replacement of the estimates from Mead et al. *(12)* with those of Scallan et al. *(14, 15)*. Scallan et al. revised the total number of annual foodborne illnesses downward by 37% and lowered the probability of hospitalization and/or death from important sources (e.g., infections with *E. coli* O157, *Listeria*, and unknown agents). The improvements made to the economic model also reduced, although to a lesser extent, the overall cost-of-illness estimates. Not all pathogens were associated with a net decline in cost. Revised estimates for 12 pathogens (most notably *C. perfringens*, non-O157 *E. coli*, *Shigella*, *Yersinia*, and hepatitis A) had higher total economic burdens than in previous studies.

The costs presented here do not represent the full economic cost of foodborne illness. Although the largest categories of health-related costs have been included, the costs of some sequelae, such as congenital toxoplasmosis, thyroid disease, and postinfectious irritable bowel syndrome, were not examined in this study. Costs of foodborne illness to industry and public health agencies also were not addressed.

Although the estimates presented here are dramatic, there are limits to how these data should be used. The total cost figures are useful as measures of the scope of the problem, but the numbers do not by themselves provide economic justification for any particular program aimed at reducing foodborne illness. Whether a potential food safety program improves social welfare is dependent on three factors: the cost per case of foodborne illness, the number of cases expected to be averted by the program, and the cost of the program to government, consumers, and industry. When examining a particular program, social welfare will only be improved when the product of the cost per case and the number of cases averted exceeds the expected cost of implementing the program for society as a whole. The numbers for cost per case provided here are well suited for use in this type of analysis.

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Thermal inactivation of *Escherichia coli* O157:H7

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1. SUMMARY

Verotoxin-producing *Escherichia coli* O157:H7 is a cause of serious foodborne illness. It has a very small infectious dose and so it is vital to eliminate this pathogen from food. As heat treatment is the method of bacterial destruction most frequently used in food processing, accurate prediction of thermal death rates is necessary to achieve desired safety margins whilst minimizing processing. In most studies thermal inactivation has been described using first-order reaction kinetics and *D*-values. Whilst this approach does not seem justified on a theoretical basis, and may increase inaccuracy, there is no doubt that it is convenient and in many cases provides an adequate description of thermal death. A review of published data on the measured thermal inactivation of *E. coli* O157:H7 shows no strong evidence that a heat treatment of 70 °C for 2 min (or equivalent) fails to deliver a 6-decimal reduction in cell numbers.

2. INTRODUCTION

Although many strains of *Escherichia coli* are harmless inhabitants of the gastrointestinal tract, some can cause disease. The pathogenic strains at present of most interest are verotoxin-producing *E. coli* (VTEC). They produce two immunologically distinct types of verotoxin, VT1 and VT2, that are cytotoxic to vero cells (from the kidney of the African Green Monkey). Individual strains produce either one or both verotoxins (Chart 1998). VT1 is immunologically identical to and has the same biological activity as the shiga toxin of *Shigella dysenteriae* (O'Brien and Holmes 1987). VTEC have been associated with a range of human illnesses, varying from mild diarrhoea to serious illness, including severe bloody diarrhoea (haemorrhagic colitis), haemolytic uraemic syndrome (HUS) and thrombotic thrombocytopoenic purpura (TTP) (Coia 1998). Fatality rates can be high in the very young and elderly.

VTEC first came to prominence as a foodborne pathogen in 1982, when serotype O157:H7 was associated with two outbreaks of haemorrhagic colitis linked to the consumption of contaminated undercooked ground beef in the Western USA (Riley *et al.* 1982). Although serotype O157 strains are the most commonly reported cause of VTEC infection in the UK (Simmons 1997), more than 100 serotypes of *E. coli* can produce verotoxins (Coia 1998). Other serogroups associated with human infection include O26, O91, O103, O111, and non-H7 strains of O157 (Goldwater and Bettelheim 1998; Paton and Paton 1998). Food poisoning outbreaks have been associated with minced beef and other beef products, cured and fermented meat products, raw milk, milk products (e.g. cheese, yoghurt), apple juice, and raw vegetables and salads (Coia 1998). Person-to-person contact, environmental exposure (e.g. farm visits) and drinking or swimming in contaminated water are also thought to be important sources of infection. The largest outbreak of food poisoning associated with verotoxin-pro-

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ducing *E. coli* O157:H7 in the UK occurred in central Scotland in 1996. It involved separate but related incidents, including a church lunch, a birthday party and retail sales of contaminated meat. In total, 496 people are known to have been infected, of whom 27 developed HUS or TTP and 20 died (Pennington 1998).

Estimates of the infectious dose of VTEC are as low as 1–100 cells (Paton and Paton 1998). Consequently it is vital to eliminate this pathogen from food, rather than merely to prevent its growth. Heat treatment is the method of bacterial destruction most frequently used in food processing. Accurate information on thermal death rates is important to food processors in order to achieve the desired safety margins whilst avoiding over-processing.

If it is assumed that all cells in a population are identical and that heat acts by destroying a single target, perhaps a single molecule or single site per cell, the rate of death can then be described by first-order reaction kinetics. Here, plotting the logarithm of the number of survivors against heating-time results in a linear relationship, the so-called log-linear plot, in which the rate of destruction is independent of the number of cells or of time. From this line the *D*-value, the time taken to reduce the population by 90%, a one decimal reduction, can be calculated. This approach is straightforward and has been used extensively in calculations of the thermal inactivation of bacterial cells by wet heat. In an alternative approach, termed the 'end-point method', an assumption of first-order reaction kinetics is made, and the *D*-value is calculated from the heating-time required to bring about a defined number of decimal reductions. If the logarithm of the *D*-value against temperature is plotted, it is possible to calculate the *z*-value, the temperature increase required to reduce the *D*-value by a factor of 10. In this review published literature on the thermal inactivation of *E. coli* O157:H7 is summarized.

3. REVIEW OF PUBLISHED DATA

Many authors have studied the thermal destruction of *E. coli* O157:H7 in foods and in culture media. Data in the range of 50–70 °C are summarized in Fig. 1. The best straight line was fitted by linear regression. The equation of the fitted line, the correlation coefficient (R^2), and the *z*-values from the fitted lines are summarized in Table 1. A predictive model of the thermal inactivation of *E. coli* O157:H7 has also been developed as part of the UK Predictive Microbiology Programme and is available in Food MicroModel (Blackburn *et al.* 1997).

For all menstrua and for culture media and buffer, the correlation between heating temperature and measured *D*-value was weak (Fig. 1a,b, Table 1). Much of the observed variation is due to differences in test conditions and experimental procedures. The effect of factors such as adverse pH, antimicrobials and the use of different enumeration media have been widely tested. For example, the $D_{52\,°C}$-values for *E. coli* O157:H7 heated in Tryptone Soya Broth (TSB) at pH 6·0 and pH 4·8 were 75 and 46 min, respectively (Clavero and Beuchat 1996). Thermal death curve data from apple juice, all meat, poultry meat and red meat samples conform better to a linear relationship (Figs 1c–1f), with an R^2 of 0·75–0·86 (Table 1). This probably reflects a greater uniformity of experimental procedures. From the line of best fit for all meat the $D_{60\,°C}$ is 1·8 min and the *z*-value is 5·5 °C. The range of reported $D_{60\,°C}$ is 0·3–10·0 min (Fig. 1).

The *z*-values reported in the original literature or calculated from papers where thermal destruction was assessed at three or more temperatures in the same heating menstruum are summarized in Fig. 2 and Table 1. The highest *z*-value was 7·29 °C for cells heated in chicken slurry (Betts *et al.* 1993) and the lowest value for serotype O157:H7 was

Table 1 Summary of published data on the effect of different heating menstrua on the thermal destruction of *Escherichia coli* O157:H7

Menstruum	Parameters of lines of best fit*				Mean *z*-value (°C) from published data‡
	Equation for fitted line	R^2	*z*-value (°C)	$D_{60\,°C}$	
All menstrua	Log D = 8·04–0·13t	0·54†	7·6	1·6	4·9 (S.D. = 0·8, *n* = 72)
Broth and buffers	Log D = 5·96–0·09t	0·31†	10·5	1·7	5·5 (S.D. = 1·0, *n* = 11)
Apple juice	Log D = 8·02–0·14t	0·75	7·4	0·8	4·8 (*n* = 1)
All meat	Log D = 11·18–0·18t	0·85	5·5	1·8	4·8 (S.D. = 0·7, *n* = 60)
Poultry meat	Log D = 11·23–0·18t	0·81	5·5	1·7	5·1 (S.D. = 0·9, *n* = 22)
Red meat	Log D = 11·22–0·18t	0·86	5·5	1·9	4·6 (S.D. = 0·5, *n* = 38)

*From the lines of best fit in Fig. 1. †These correlations are weak. The equation for the fitted line and the *z*-value from the fitted line should not be used in thermal death calculations. ‡S.D. = standard deviation; *n* = number of *z*-values reported in the literature. Raw data for all menstrua are plotted in Fig. 2.

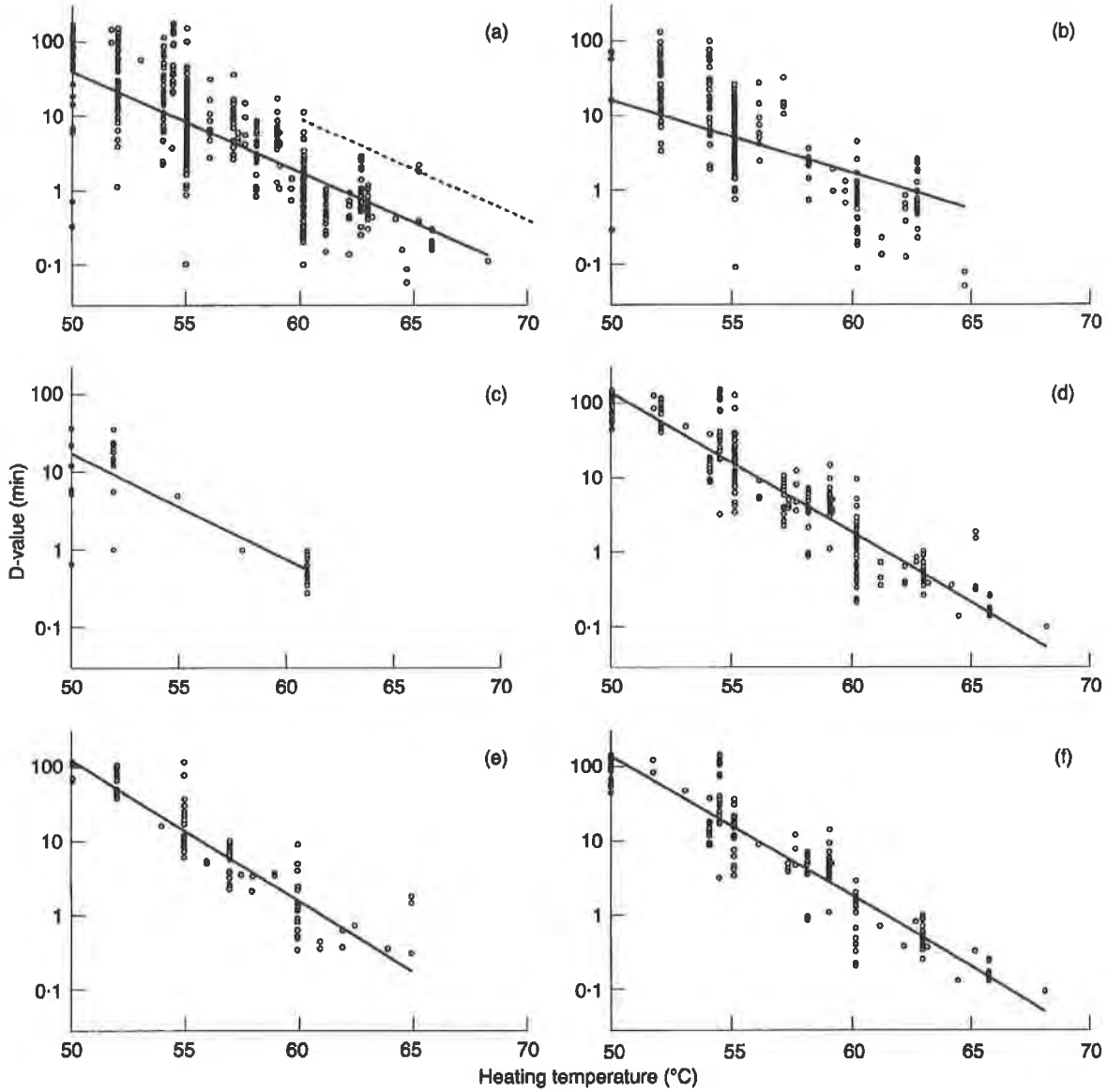

Fig. 1 Summary of published heat resistance data for *Escherichia coli* O157:H7 heated in all menstrua (a), broth and buffer (b), apple juice (c), all meat (d), poultry meat (e) and red meat (f). The solid lines indicate the line of best fit (see Table 1 for parameters of these lines), and the broken line on (a) indicates the 'line of safety' corresponding to a 1D reduction from the recommendations of the ACMSF (1995). Data are from: Doyle and Schoeni 1984; Line *et al.* 1991; Murano and Pierson 1992, 1993; Abdul-Raouf *et al.* 1993; Betts *et al.* 1993; Nishikawa *et al.* 1993; Todd *et al.* 1993; Ahmed and Conner 1995; Ahmed *et al.* 1995; Clavero and Beuchat 1995, 1996; Czechowicz *et al.* 1996; Jackson *et al.* 1996; Splittstoesser *et al.* 1996; Teo *et al.* 1996; Blackburn *et al.* 1997; Juneja *et al.* 1997a; Kotrola and Conner 1997; Kotrola *et al.* 1997; Orta-Ramirez *et al.* 1997; Williams and Ingham 1997; Clavero *et al.* 1998; Ingham and Uljas 1998; Juneja *et al.* 1998; Kaur *et al.* 1998; Quintavalla *et al.* 1998; Semanchek and Golden 1998; Williams and Ingham 1998; Veeramuthu *et al.* 1998; Duffy *et al.* 1999; George *et al.* 1999; Stringer *et al.* 2000

3·9 °C for cells heated in ground beef and enumerated on MEMB (Clavero *et al.* 1998). A *z*-value of 3·5 °C was reported for a VTEC strain of serotype O157:NM in beef (Todd *et al.* 1993). Of the 72 reported *z*-values, three were below 4·0 °C, 58 were 4·0–5·9 °C, nine were 6·0–6·9 °C, and two were 7·0 °C or above (Fig. 2). The mean *z*-value for all menstrua was 4·9 °C, with a standard deviation of 0·8 °C (Table 1). The mean *z*-value for each heating men-

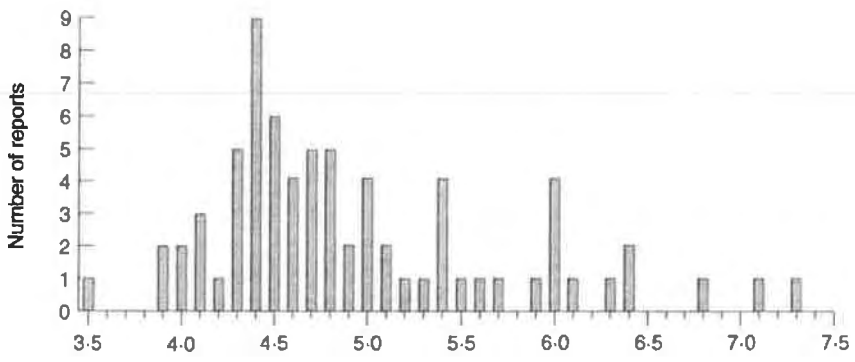

Fig. 2 Summary of published *z*-values for *Escherichia coli* O157:H7. Data are from references in Fig. 1

struum was in the range 4·6–5·5 °C (Table 1). It is noteworthy that whilst an average *z*-value seems to be in the region of 5 °C, the Advisory Committee on the Microbiological Safety of Food (ACMSF) used a *z*-value of approximately 7 °C (ACMSF 1995).

Although measured heat resistance can vary considerably with test conditions, none of the published data suggests that *E. coli* O157:H7 is unusually heat-resistant as compared with other nonspore-forming foodborne pathogens such as *Listeria monocytogenes* (Mackey and Bratchell 1989; George *et al.* 1998). In the UK, the ACMSF report on VTEC recommends that minced beef and minced beef products, including beefburgers, should be heated to an internal temperature of 70 °C for 2 min or its equivalent (Table 2). Similar recommendations have been made by the Food Safety Authority of Ireland (1999). These heat treatments were considered to be sufficient to give a 6D (six decimal) reduction in numbers of VTEC (ACMSF 1995). By dividing the times to a 6D reduction by six, the maximum safe *D*-value can be calculated for each temperature, and this is plotted as a "line of safety" in Fig. 1(a). From this figure, it is apparent that almost all of the reported *D*-values in the range 60–70 °C lie below the line of safety (Fig. 1a). The three points that lie above the line are from the study of Veeramuthu *et al.* (1998) who reported a $D_{60\,°C}$ of 10·0 min and $D_{65\,°C}$ of 1·7 min and 2·1 min in ground turkey. High heat resistance at temperatures above 70 °C has been reported. There was an estimated 5D reduction in *E. coli* O157:H7 in pepperoni on a frozen pizza baked in an oven at 191 °C (375 °F) for 20 min or at 246 °C (475 °F) for 15 min (Faith *et al.* 1998). It is difficult to compare these results with the data below 70 °C, because the temperature of the pepperoni was variable and did not reach that of the oven, and the bacterium may have been protected by a low water activity due to a high fat content and drying of the pepperoni.

If all the published data are taken into account, there is no strong evidence that a heat treatment of 70 °C for 2 min, or its equivalent, is insufficient to give a 6D reduction of *E. coli* O157:H7. From Fig. 1 it would, however, seem unsafe to extrapolate the ACMSF recommendations (ACMSF 1995) to temperatures below 60 °C.

The heat treatments recommended by the ACMSF for the UK differ from those specified in the USA Food Code published in 1997 (FDA 1997) and updated advice issued in 1998 (FSIS 1998). The advice was that, in restaurants, ground beef patties should be heated to an internal temperature of 68·3 °C (155 °F) for 15 s, and in the home burgers should be cooked to an internal temperature of 71·1 °C (160 °F), as measured using a food thermometer (FSIS 1998). In tests with a mixture of five strains, heating ground beef patties to an internal endpoint temperature of 68·3 °C (155 °F) was sufficient to give a 4D reduction of *E. coli* O157:H7 (Juneja *et al.* 1997b).

Table 2 Equivalent heat treatments recommended for minced beef products (ACMSF 1995)

Temperature (°C)	Heating time
60	45 min
65	10 min
70	2 min
75	30 s
80	6 s

4. FACTORS THAT AFFECT THE MEASURED THERMAL INACTIVATION OF *ESCHERICHIA COLI* O157:H7

The measured thermal resistance of a species can be influenced by many factors, including the growth conditions, such as the growth phase of the cells, composition, pH and water activity of the growth medium, growth temperature, holding period before heat treatment, heat shock, the heating method, for example use of open heating system and rate of heating, the heating menstruum, including its composition, pH, water activity and choice of humectant, and the recovery conditions (Olsen and Nottingham 1980). Heat resistance may also be affected by physical interactions such as attachment to solid surfaces. For example, the $D_{58\,°C}$ of *Salmonella typhimurium* attached to muscle was higher than that for free cells (Humphrey *et al.* 1997) and the presence of filterable pulp in apple juice enhanced the measured thermal tolerance of two strains of *E. coli* O157:H7 (Ingham and Uljas 1998). Several authors have quantified the effect of the experimental procedure on measured heat resistance and the conclusions from some of these studies are presented below.

4.1. Effect of growth conditions

The ability of *E. coli* O157:H7 to tolerate heat is strain-dependent (Betts *et al.* 1993; Ahmed and Conner 1995; Clavero and Beuchat 1995; Blackburn *et al.* 1997; Clavero *et al.* 1998; Quintavalla *et al.* 1998; Duffy *et al.* 1999; Fig. 3). A comparison of the heat resistance of seven strains of *E. coli* O157:H7 gave $D_{60\,°C}$ from 0·44 to 1·29 min, with strain 30-2C4 showing the highest heat resistance and strain W2-2 the lowest (Fig. 3). Clavero *et al.* (1998) compared the heat resistance of 10 strains at four temperatures. The ranking of the strains, in terms of heat resistance, varied with heating temperature. $D_{58·9\,°C}$ ranged from 4·9 to 15·0 min and $D_{62·8\,°C}$ ranged from 0·56 to 1·14 min.

Heat resistance was greater when cells were in the stationary rather than the logarithmic growth phase (Todd *et al.* 1993; Jackson *et al.* 1996; Kaur *et al.* 1998), and when cells were grown at 37 or 40 °C rather than 10, 23, 25 or 30 °C (Jackson *et al.* 1996; Kaur *et al.* 1998; Semanchek and Golden 1998; Fig. 3). For example, the heating time at 55 °C required for a 5D reduction was 7·3 min for exponential-phase cells grown at 23 °C, and 106·4 min for stationary-phase cells grown at 37 °C (Jackson *et al.* 1996). Storage conditions can also affect heat resistance. Whilst Jackson *et al.* (1996) reported that storage at 3 °C decreased heat resistance, and that the heat resistance of cells in meat that had been stored frozen was greater than for equivalent samples stored under refrigeration, Stringer *et al.* (2000) found that holding cells in TSB at 2 °C for 10 d brought about a small increase in heat resistance (Fig. 3). The thermal tolerance of two strains of *E. coli* O157:H7 decreased when they were held in apple juice (pH 3·4) at 4 °C for 24 h, while storage at 21 °C for 2 h decreased the thermal tolerance of only one of the strains (Ingham and Uljas 1998).

It has been reported that heat shock increases heat resistance, but its effectiveness depends on the environmental conditions. Heat shock increased the heat resistance of cells in TSB but not in beef slurry (Williams and Ingham 1997); heat shock at 42, 45, 48 or 50 °C had little effect on exponential-phase cells grown in nutrient broth (Kaur *et al.* 1998); heat shock at 42 °C for 5 min induced larger increases in heat resistance in cells grown aerobically than those grown anaerobically (Murano and Pierson 1992); heat shock at 45 °C for 5 min increased approximately 10-fold the number of cells recovered after heating at 59 °C for 5 min, while heat shock at 42 °C for 5 min had little effect (Bromberg *et al.* 1998). The increased thermotolerance gained by heat shock at 46 °C was maintained during a period of storage (Juneja *et al.* 1998).

4.2. Effect of heating method

To obtain good thermal inactivation data it is important to use a method of heat treatment that avoids local temperature variations, because a subpopulation of cells subjected to a lesser heat treatment will appear to be more heat resistant and tailing will occur. Teo *et al.* (1996) used an open system to examine the effects of high pH on the thermal death of *E. coli* O157:H7. Initially experiments were performed with heated flasks and the level of the water bath 2·0 cm above the surface of the heating menstruum and the cells mixed into the menstruum by hand. The resulting survivor curves had pronounced tails, and it is likely that the subpopulation in the tail did not receive the full heat treatment. Tailing was prevented by using a magnetic stirrer in the flask and increasing the level of the water bath to 5·0 cm above the heating menstruum. With these two methods, the heating times at 50 °C to give a 6D reduction were approximately 20 min and 5 min, respectively. Although some authors who used open heating systems have published log-linear survivor curves, it is best to avoid controversy and use closed systems. Even in closed systems, the heating method may affect measured heat resistance. A slow rate of heating may induce heat shock and so increase heat resistance. Careful measurement of the actual heat treatment is also required. It is not uncommon for reports to fail to take account of the come-up time.

4.3. Effect of heating menstruum

The medium in which the cells are heated can strongly influence the heat resistance of *E. coli* O157:H7. Heat resis-

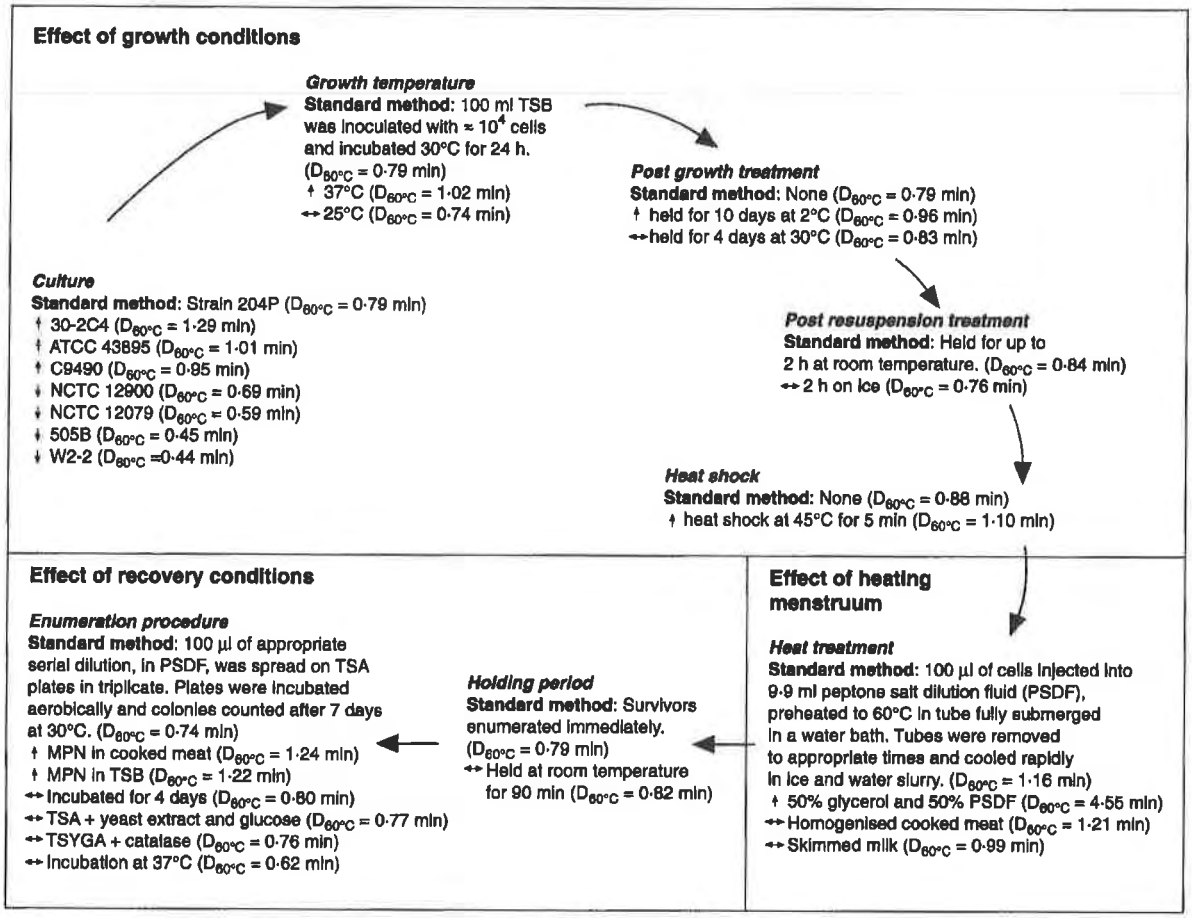

Fig. 3 Effect of procedural changes on the measured heat resistance of *Escherichia coli* O157:H7 at 60 °C. The arrows indicate whether changes significantly increased (↑), decreased (↓) or had no effect (←→) on measured heat resistance when compared to the standard method. Significant differences at the 95% level were determined using the *F*-test. From Stringer *et al.* (2000)

tance was greater for cells heated in lean ground beef than in lean ground chicken (Juneja *et al.* 1997a) and greater in beef and pork slurry than in TSB (Williams and Ingham 1997; Quintavalla *et al.* 1998; George *et al.* 1999). For three strains of *E. coli* O157:H7 heated in TSB and in pork slurry with curing agents, $D_{60\,°C}$ ranged from 0·28 to 0·33 min and from 0·94 to 1·05 min, respectively (Quintavalla *et al.* 1998). Cells can be more heat-resistant in fatty than in lean meat (Line *et al.* 1991), but this effect is not always observed. For example, cells were no more heat-resistant in turkey meat with 11% fat than in meat with 3% fat (Kotrola and Conner 1997). Ahmed *et al.* (1995) reported that heat resistance was greater in meat with a high fat content than in meat with a lower fat content, but they stated that as the moisture content decreased and the fat content increased, any affect could be related to water activity and not necessarily to the fat content. Sodium chloride concentrations up to 8·5% protected cells of *E. coli* O157:H7 from thermal inactivation in TSB (Blackburn *et al.* 1997) and *D*-values were increased when 8% NaCl was added to ground turkey breast (Kotrola and Conner 1997). However, Clavero and Beuchat (1996) found $D_{52\,°C}$ decreased when 8·6% or 16·8% NaCl was added to TSB. When 50% glycerol was added to the peptone salt dilution fluid used as the heating medium, $D_{60\,°C}$ increased from 1·2 min to 4·6 min (Fig. 3). Addition of a mixture of sucrose, glucose and fructose to apple juice to increase the Brix from 11·8° to 16·5° had no effect on heat resistance (Splittstoesser *et al.* 1996).

Heat resistance is also affected by the pH of the heating menstruum. Blackburn *et al.* (1997) found heat resistance was highest at pH 5·2–5·9 for cells heated in TSB. The pH

optimum depended on the heating temperature and NaCl concentration. Greater heat resistance was observed for cells in apple juice at pH 4·4 than at pH 3·6 or 4·0 (Splittstoesser *et al.* 1996), and in sodium carbonate buffer at pH 7 than in buffer at pH 10 or 11 (Teo *et al.* 1996). In these experiments adverse pH, either high or low, undoubtedly contributed to the kill. Cells were more sensitive to heat in the presence of sorbic, benzoic, acetic, citric or lactic acid (Abdul-Raouf *et al.* 1993; Splittstoesser *et al.* 1996; Blackburn *et al.* 1997) and a commercial mixture of polyphosphates (Kaur *et al.* 1998). The combined effect of water activity and pH were included in a predictive model of the thermal inactivation of *E. coli* O157:H7. The model provided fail-safe predictions of the effect of heating conditions, that is temperature (54·5–64·5 °C), NaCl (0–8·5%) and pH (4·2–9·8), on the thermal inactivation of *E. coli* O157:H7, and was suitable for use with many foods (Blackburn *et al.* 1997).

4.4. Effect of recovery conditions

The conditions under which cells are incubated after heat treatment and the incubation period can affect the measured heat resistance of cells. Measured heat-inactivation was increased when heat-treated cells were enumerated with pour plates rather than spread plates (Czechowicz *et al.* 1996), when cells were held on ice rather than at 20 °C before enumeration (Williams and Ingham 1997) and when incubation was at 20 or 25 °C rather than 30 °C (George *et al.* 1998). The measured heat resistance has been widely shown to depend on the media used to enumerate heat-treated cells (Abdul-Raouf *et al.* 1993; Murano and Pierson 1993; Ahmed and Conner 1995; Clavero and Beuchat 1995; McCleery and Rowe 1995; Clavero and Beuchat 1996; Clavero *et al.* 1998; Fisher and Golden 1998; Quintavalla *et al.* 1998; Duffy *et al.* 1999). It was usually lower if heat-treated cells were enumerated on media selective for *E. coli* O157:H7 rather than on non-selective media. For example, low counts were obtained with modified sorbitol MacConkey agar (Clavero and Beuchat 1995, 1996; Clavero *et al.* 1998). Heat-treated cells were also sensitive to 2% NaCl during enumeration (Semanchek and Golden 1998).

Measured heat resistance is higher if anaerobic (reduced) conditions, rather than aerobic (oxidized) conditions, are used for enumeration (Murano and Pierson 1992, 1993; Blackburn *et al.* 1997; Bromberg *et al.* 1998; George and Peck 1998; George *et al.* 1998, 1999). The measured heat resistance of *E. coli* O157:H7 was highest when anaerobic gas mixtures were used (time at 59 °C for a 6D reduction, 19–24 min), moderate when low oxygen concentrations (0·5–1%) were included (time for a 6D reduction, 13–17 min) and lowest when higher oxygen concentrations (2–40%) were included (time for a 6D reduction, 3–5 min) (George *et al.* 1998). In this study the oxidation–reduction (redox) potential of the media was altered by the use of different partial pressures of oxygen, but in a subsequent study in which the redox potential was manipulated with the oxidants potassium ferricyanide and 2,6-dichloroindophenol and the reductant dithiothreitol, the measured heat resistance was affected by the redox potential independently of the oxidant used to adjust it and therefore independently of the oxygen concentration (George and Peck 1998). When cells were heated so as to give a 6D reduction when enumerated in aerobic TSYGB, only a 3D reduction was due to thermal inactivation with the other 3D reduction related to sensitivity to oxidized conditions (Bromberg *et al.* 1998). Maintenance of these cells under anaerobic conditions at 30 °C for 4 h resulted in repair of sublethal damage and recovery of the ability of the bacteria to grow in oxidized media. Cells held at 5 °C did not recover in 816 h (Bromberg *et al.* 1998). Some authors have reported that catalase (McCleery and Rowe 1995) or pyruvate (Czechowicz *et al.* 1996) added to the enumeration medium increased recovery of heat-damaged cells, whilst others found little effect (Clavero and Beuchat 1995; Fig. 3).

To determine the true extent of thermal inactivation, it is important to use procedures that recover the greatest number of cells. For example, counting colonies after too short an incubation period, the use of certain selective media, or the use of aerobic counting techniques may overestimate the effectiveness of the heat treatment and hence the margin of safety that has been achieved. Failure to take full account of anaerobic recovery may be particularly important with regard to the margin of safety for foods that are packed under vacuum or a modified (low oxygen) atmosphere, have a low redox potential in spite of exposure to air, or support growth of other microorganisms that might lower the redox potential.

5. THERMAL INACTIVATION KINETICS OF *ESCHERICHIA COLI* O157:H7

5.1. Functions to describe thermal inactivation kinetics of bacterial cells

Although first-order reaction kinetics and *D*-values have been used extensively in calculations of thermal inactivation, the literature is full of examples where first-order reaction kinetics are not followed. Indeed Pflug and Holcomb (1991) estimated that only one-third of thermal destruction data follow such kinetics. Deviations take the form of shoulders or tails, and this subject has been reviewed (Moats *et al.* 1971; Cerf 1977). There are two basic explanations why first-order kinetics are not followed. The first is that shoulders or tails are artefacts that can be

Table 3 Possible explanations for shoulders or tails in thermal inactivation curves

	Explanation
Shoulders and/or tailing are artefacts and due to limitations in the experimental procedure	•Treatment heterogeneity (e.g. heating method, heating menstruum not uniform, sticking to the walls of vessels) •Use of mixed populations (mixtures of strains with an unequal resistance, cells at different stages of growth) •Clumping or clump separation during heat treatment or enumeration of survivors •Protective effect of dead cells •Method of enumeration (e.g. use of selective media, too short recovery period) •Poor statistical design (e.g. use of low counts)
Shoulders and/or tailing are a normal feature of the mechanism of inactivation	•Multiple hit mechanism (not single hit) •Natural distribution of heat sensitivity within the population (genetical or physiological heterogeneity) •Heat adaptation or heat shock/activation during heat treatment

attributed to limitations of the experimental procedure (Table 3). The second explanation is that shoulders or tails are a natural feature bound to the mechanism of heat inactivation or resistance of the population (Table 3).

For convenience, deviations from first-order kinetics are often ignored when calculating thermal resistance values, with straight lines fitted to all of, or sections of, curves. In these circumstances the use of log-linear plots is practical rather than theoretical and may be misleading. In an attempt to describe thermal death data more accurately, a number of alternative mathematical functions have been adopted. Some are based on understanding the physiological mechanism of inactivation. For example, Moats *et al.* (1971) assumed that targets are clumped at critical sites within the cell, Körmendy and Körmendy (1997) assumed that there is a distribution of resistance within the population, and Smerage and Teixeira (1993) assumed that heterogeneity occurs by adaptation to heat during heating. Other models involve empirical curve fitting (e.g. Pruitt and Kamau 1993; Stecchini *et al.* 1993; Whiting 1993). Although one function may appear to fit a thermal inactivation curve better than another, it is often difficult to prove that the model performance is better.

5.2. Discussion of the most appropriate function to describe the thermal inactivation kinetics of *Escherichia coli* O157:H7

When describing the thermal death of *E. coli* O157:H7, first-order reaction kinetics have generally been fitted to all or part of the curve. However, Blackburn *et al.* (1997) obtained curves that showed tailing and were poorly described by first-order reaction kinetics. These curves were better described by the log-logistic function (Cole *et al.* 1993). Predicted time to a 5D reduction was greater from the log-logistic model than the log-linear (*D*-value) model (Blackburn *et al.* 1997). An empirical approach developed by Whiting (1993) permits the application of up to three straight lines to thermal inactivation data giving different rate constants for the shoulder, exponential region and tail. Juneja *et al.* (1997a) used this model to determine *D*-values for subpopulations of *E. coli* O157:H7 heated in beef and chicken. Herremans *et al.* (1997) introduced a dynamic model to take account of the experimentally observed shoulder and tail in the thermal death curve of *E. coli* O157:H7 heated at 60 °C.

First-order reaction kinetics assume that the population is homogeneous and that heat acts by destroying a single target. From this, it may be expected that cells will exist in one of two states; either the target is undamaged and the cell is healthy or the target has been inactivated and the cell is dead. This theory does not allow the possibility of sublethally damaged cells. There are, however, many examples of heated *E. coli* O157:H7 cells showing sublethal damage. For example, delays have been reported in time to growth and of sensitivity to growth on media containing selective agents, sensitivity to 2% NaCl, and sensitivity to high redox potential and oxygen concentration during enumeration (as described earlier). Bromberg *et al.* (1998) identified two distinct periods of repair of sublethal injury during recovery and subsequent growth of heated cells of *E. coli* O157:H7. Cells were grown under strictly anaerobic conditions in TSYGB at 30 °C, heated in anaerobic TSYGB for 5 min at 59 °C, cooled in ice-water and held at 30 °C. At timed intervals samples were removed and counts performed in aerobic and anaerobic TSYGB by a MPN procedure (Fig. 4). Immediately after heating for 5 min and cooling, the aerobic count was 3×10^3 cells ml^{-1} and the anaerobic count was 3×10^6 cells ml^{-1}. After incubation for 4 h the aerobic and anaerobic counts were similar, indicating that all the cells had regained their ability to grow in the aerobic media. The anaerobic count did not change during this period (Fig. 4). There was no increase in cell numbers for approximately 13 h, when the aerobic and

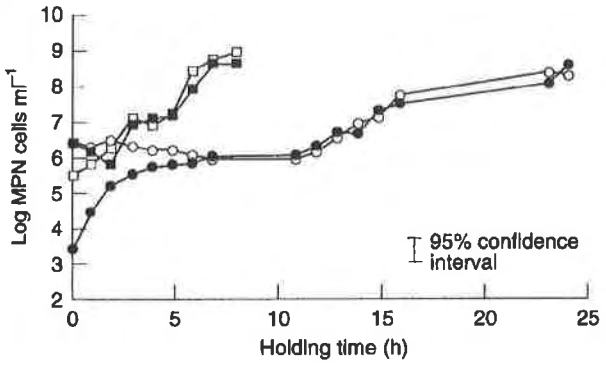

Fig. 4 Recovery and growth of sublethally heat-injured cells of *Escherichia coli* O157:H7 strain 204P. Cells were heated at 59 °C for 5 min, held at 30 °C in anaerobic TSYGB medium, and enumerated in aerobic TSYGB (●) and anaerobic TSYGB (○), or cells were unheated (but diluted), held at 30 °C in anaerobic TSYGB medium and enumerated in aerobic TSYGB (■) and anaerobic TSYGB (□). Results are the mean of four experiments (heated cells) and duplicate experiments (unheated cells). The 95% confidence interval was derived from the average of the individual confidence intervals. Data for heated cells are from Bromberg *et al.* (1998) and data for unheated cells are unpublished results of George and Peck

anaerobic counts increased at the same rate and reached 10^8–10^9 cells ml^{-1} within 24 h (Fig. 4). In a control test, cells that were diluted instead of heated, but were otherwise subjected to the same treatment, gave similar initial counts in aerobic and anaerobic TSYGB, and the cell numbers increased after 3 h, with both counts reaching 10^8–10^9 cells ml^{-1} after 7 h (Fig. 4). Heated cells of *E. coli* O157:H7 were therefore sublethally injured, and had two distinct periods of cell repair. The first period of repair took 4 h at 30 °C during which time sublethally injured cells regained their ability to grow in oxidized media. The second period of repair took a further 9 h at 30 °C, after which cell division commenced.

Sub-lethal injury has been studied in many bacteria, for example similar observations of the effect of heat treatment have been made with *L. monocytogenes* (e.g. Mackey and Bratchell 1989; George *et al.* 1998). These reports cast doubt on the validity of the mechanism that underlies the use of first-order reaction kinetics (and *D*-values) to describe thermal inactivation of *E. coli* O157:H7. Many of the thermal inactivation data for *E. coli* O157:H7 have, however, been adequately described by first-order reaction kinetics.

6. CONCLUSIONS

Accurate prediction of the thermal death rate of *E. coli* O157:H7 is important so food processors can ensure that appropriate safety margins are attained. Although there is some variation in the published heat inactivation data for *E. coli* O157:H7, there is no strong evidence that a heat treatment of 70 °C for 2 min or the equivalent, as recommended by the ACMSF, fails to deliver a 6D reduction in cells of *E. coli* O157:H7.

In most studies thermal inactivation has been described by first-order reaction kinetics and *D*-values. While this does not seem to be justified on a theoretical basis, there is no doubt that it is convenient, and in many cases provides an adequate description of thermal death. An important step forward would be to develop a mathematical function to describe thermal death that can be justified on a theoretical basis, that is one based on understanding the process of thermal death at the cellular level. A more accurate description of thermal death would benefit industry, regulators and consumers because it would allow a tighter control of safety margins.

7. ACKNOWLEDGEMENTS

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Thermal Inactivation of *Salmonella spp.* in Chicken Broth, Beef, Pork, Turkey, and Chicken: Determination of D- and Z-values

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ABSTRACT: The heat resistance of 35 *Salmonella* strains was determined at 55 to 65 °C. No correlation between the heat resistance and the origin of the *Salmonella* spp. could be established. D-values in chicken broth, using a linear regression, of an 8 *Salmonella* serotype cocktail were 4.87, 2.72, 1.30, and 0.41 min at 55, 58, 60 and 62 °C, respectively. Using a linear model, the D-values ranged from 4.86 min at 55 °C to 0.38 min at 62 °C. When the 8 *Salmonella* serotype cocktail was heated in meat, D-values at the common test temperatures of 58 and 60 °C calculated by both approaches were significantly higher ($p < 0.05$) than those observed in chicken broth.

Key Words: heat resistance, *Salmonella*, beef, pork, turkey, chicken

Introduction

SALMONELLA IS A LEADING CAUSE OF GASTROENTERITIS IN HUmans and continues to be of significant public health concern to the food industry. An important contributing factor which leads to outbreaks of foodborne illness, including salmonellosis, is inadequate time/temperature exposure during initial thermal processing (or cooking), and inadequate reheating to kill pathogens in retail food service establishments or homes (Roberts 1991; Bean and Griffin 1990). Inadequate cooking was cited as a contributing factor in 67% of the outbreaks in which *Salmonella* was an etiological agent (Bean and Griffin 1990). These outbreaks have implicated a variety of foods, including meat and poultry, milk, ice cream, cheese, eggs and egg products, chocolate, and spices, as vehicles of transmission (D'Aoust 1989). In an effort to eliminate this public health hazard, the U.S. Dept. of Agriculture has implemented a 6.5 - D reduction in population of *Salmonella* spp. for cooked beef, ready-to-eat roast beef, and cooked corned beef products, and a 7 - D *Salmonella* reduction in certain fully and partially cooked poultry products (USDA 1999). The thermal processing schedule to be used to meet this lethality performance standard is not specified.

Cooking remains a primary means of eliminating pathogens from ground muscle foods, and therefore serves to protect against foodborne disease. During cooking or thermal processing, the rate of destruction of a microbial population is generally considered to follow first order kinetics; that is, at a given temperature, the reduction in the log number of survivors occurs in a linear manner over time (Stumbo 1973; Tomlins and Ordal 1976). However, the traditional log-linear thermal-death-time model generally gives a good fit to the inactivation data only in situations when death is rapid. Significant deviations have been observed from the log-linear declines (logarithmic survivor curves) with characteristic lag periods or shoulders before any death occurs and a tailing or a subpopulation of more resistant bacteria that decline at a slower rate than the majority of the cells (Tomlins and Ordal 1976; Pflug and Holcomb 1983; Juneja and others 1997). Such nonlinear survivor curves generally cannot be accounted for by experimental artifacts, and there is presently no satisfactory, unifying explanation for the variability in inactivation kinetics of bacteria. In scenarios where the log-linear death model/linear regression approach gives a poor fit to the data, analysis of results using this approach could lead to underestimation of the time and temperature needed to achieve a desired reduction of bacterial numbers. Therefore, alternate approaches have been developed to account for the nonlinear decline in the log number of survivors with time. One such model for nonlinear survival curves is a linear model that incorporates a lag period (Buchanan and others 1993; 1994). This innovative approach is a valuable tool for the estimation of the heat resistance of bacteria, when deviations from the first order inactivation kinetics are observed.

Previous workers have conducted thermal inactivation studies of *Salmonella* spp. in aqueous media and foods (D'Aoust 1989). However, researchers have not addressed the question of identifying the most representative and appropriate strains of *Salmonella* to use for defining the heat resistance in specific meat products. Currently, there are more than 2,000 serotypes and considerably more strains of *Salmonella* known (Farber 1986). Accordingly, this study quantifies the heat resistance in chicken broth, beef, pork, turkey and chicken, as defined by D- and z-values, of different *Salmonella* serotypes isolated from beef, pork, chicken, and turkey, as well as human clinical isolates. D-values were determined by using (a) linear regression from the straight line portion of the survival curves; that is, first order kinetics, and (b) by a linear model that was fitted to the nonlinear survival curves to account for the lag period (Buchanan and others 1993; 1994). The heat treatment required for a specified lethality, that is, 6.5 or 7 $\log_{10}$ reduction of *Salmonella* in meat, detailed in this study, should assist food processors in designing thermal processes suitable for the inactivation of salmonellae in the different menstrua tested.

Materials and Methods

Bacterial strains

Salmonella spp. isolated from raw processed beef, pork,

chicken, and turkey, as well as human clinical isolates, were used in the study (Table 1). The strains were preserved at -70 °C in vials containing Tryptic soy broth (TSB; Difco, Detroit, Mich.) with 10% (v/v) glycerol (Sigma Chemical Co., St. Louis, Mo., U.S.A.) added.

Products

Commercially canned chicken broth (3% fat, Swanson brand), ground meat (beef, pork, chicken, and turkey), used as heating menstrua, were obtained from a retail supermarket. The proximate analysis of meats done by Lancaster Laboratories (Lancaster, Pa., U.S.A.) indicated that the beef contained 65.5% moisture, 12.45% fat, 0.82% ash, and 20.25% protein; pork contained 72.95% moisture, 6.95% fat, 0.90% ash, and 16.80% protein; turkey contained 70.20% moisture, 8.85% fat, 0.81% ash, and 19.30% protein; chicken contained 71.75% moisture, 8.45% fat, 1.14% ash, and 19.40% protein. The pH of the chicken broth and meats tested were determined, using a combination electrode (Sensorex semi-micro, A.H. Thomas, Philadelphia, Pa., U.S.A.) attached to an Orion model 601A pH meter. The meat was placed into appropriate barrier pouches (100g/bag) and vacuum sealed, frozen at −40 °C and irradiated (42 kGy) to eliminate indigenous microflora. Random samples were tested to verify elimination of microflora or to confirm sterility by diluting in 0.1% (wt/vol) peptone water (PW), spiral plating (Spiral Biotech, Bethesda, Md., U.S.A.; Model D) on Tryptic soy agar (TSA; Difco) and incubating at 30 °C for 48 h. The lower limit of detection using spiral plater is 21 cfu/ml.

Preparation of test cultures

To prepare the cultures, vials were partially thawed at room temperature and 1.0 mL of the culture was transferred to 50 mL of TSB in 250 mL flasks and incubated for 24 h at 37 °C. This culture was not used in heating studies, as it contained freeze-damaged cells. The inocula for use in heating studies were prepared by transferring 0.1 mL of each culture to 10 mL tubes of TSB and incubating aerobically for 24 h at 37 °C, to provide late stationary phase cells. These cultures were maintained by consecutive daily transfers in TSB for 1 wk. A new series of cultures was initiated from the frozen stock on a weekly basis.

On the day of the experiment, each culture was pelleted by centrifuging (5,000 × g, 10 min, 4 °C) and washed two times in PW. The cell pellets were then suspended in PW to a final concentration of 8 to 9 $\log_{10}$ cfu/mL. The population densities in each cell suspension were determined by spiral plating appropriate dilutions (in 0.1% PW), in duplicate, on TSA. Equal volumes of 8 different *Salmonella* serotypes isolated from different meat species were combined in a sterile test tube to obtain a *Salmonella* serotype cocktail (9 $\log_{10}$ cfu/mL) for the inoculation of chicken broth or the ground meats already described.

Sample preparation and inoculation

The individual *Salmonella* strains or *Salmonella* serotype cocktail inoculum was added (0.1 mL) to 10 mL chicken broth or (10 mL) to 100 g of thawed (over a period of 24 h at 4 °C) irradiated ground meat, to obtain a final concentration of approx. 8 $\log_{10}$ cfu/g. Each inoculated sample of chicken broth was vortexed and each bag of meat was blended (Seward laboratory stomacher 400) for 5 min, to ensure even distribution of the organisms in the respective menstruum. Duplicate 5 g ground meat samples were then weighed aseptically into 30 × 19-cm sterile filtered stomacher bags (Spiral Biotech, Bethesda, Md., U.S.A.). Bags containing meat samples inoculated only with 0.1 mL sterile PW were used as negative controls. Thereafter, the bags were compressed into a thin layer (approx. 1 to 2 mm thick) by pressing against a flat surface, excluding most of the air, and then heat sealed.

Thermal inactivation procedure

The broth suspensions were heated at 55, 58, 60, or 62 °C, using a submerged coil heating apparatus (Cole and Jones 1990). This apparatus is comprised of a stainless steel coil fully submerged in a thermostatically controlled water bath which allows microbial suspensions to be heated between 20 to 90 °C with a short time (< 1 s) to achieve temperature equilibrium. During the heating procedure, samples (0.2 mL) were removed at predetermined time intervals. Where low cell numbers were expected, 0.6-mL aliquots were removed. Samples were cooled rapidly in an ice slurry.

To simulate food industry storage conditions, the bags containing the meat samples were incubated for 90 min at 4 °C to achieve temperature equilibrium. Thereafter, the bags were placed in a basket and fully submerged in a temperature-controlled water bath (Techne, ESRB, Cambridge, U.K.) stabilized at 58, 60, 62.5, or 65 °C. The temperature was continuously monitored by two copper-constantan thermocouples inserted, prior to heat sealing, at the center of two uninoculated bags. The thermocouple readings were measured and recorded using a Keithyl-Metrabyte data logger Model DDL 4100 (Tauton, Mass., U.S.A.) connected to a microcomputer. The thermocouple signal was sampled every second, and the two readings averaged to determine the bag's internal temperature. Come-up times, which previous experiments have indicated are negligible (< 1 to 2 s), were included as part of the total heating time when used to calculate the D-values. Bags for each replicate were then removed at predetermined time intervals; sampling frequency was based on the heating temperature. After removal, bags were immediately placed into an ice-water bath, and analyzed for salmonellae within 30 min.

Enumeration of surviving bacteria

Heat-treated chicken broth or meat samples were combined with PW to obtain a 1:5 or 1:2 dilution, respectively, for the determination of surviving bacteria. Broth samples were vortexed and the meat slurry samples were pummeled for 2 min with a Seward laboratory stomacher 400. Decimal serial dilutions were prepared in PW and appropriate dilutions surface-plated in duplicate on TSA, supplemented with 0.6% yeast extract and 1% sodium pyruvate, using a spiral plater. Samples not inoculated with *Salmonella* spp. cocktail were plated as controls. Also, 0.1 and 1.0 mL of undiluted suspension were surface-plated where relevant. All plates were incubated at 28 °C for at least 48 h prior to counting colonies. Incubation temperature of 28 °C was used because researchers have reported that temperatures below the optimum for growth may enhance repair of heat damage (Katsui and others 1982). For each replicate experiment, an average cfu/g of two platings of each sampling point were used to determine the D-values.

Calculation of D-values and z-values

D-values (time to inactivate 90% of the population) were calculated from the straight portion of the survival curves by plotting the log of survival counts compared with their corresponding heating times, using Lotus 1-2-3 Software (Lotus Development Corp., Cambridge, Mass., U.S.A.). Only survival

Table 1—Heat resistance (expressed as D-values in min)[a] for *Salmonella* spp. in chicken broth at 580 °C

Origin/Strain	Isolate	Method to Determine D-Value: Linear Regression D-Value (r^2)[b]	Linear Model D-Value	RMS error[c]
Beef				
Salmonella montevideo	FSIS 051	2.16 ± 0.20 (0.94)	1.75 ± 0.01	1.10
Salmonella typhimurium	FSIS 026	1.54 ± 0.08 (0.84)	1.41 ± 0.21	0.92
Salmonella kentucky	FSIS 074	1.82 ± 0.12 (0.90)	1.62 ± 0.10	1.11
Salmonella saint-paul	FSIS 039	1.94 ± 0.02 (0.92)	1.67 ± 0.00	1.01
Pork				
Salmonella copenhagen	8457	2.94 ± 0.22 (0.96)	2.50 ± 0.14	0.79
Salmonella derby	8453	1.29 ± 0.15 (0.82)	1.05 ± 0.09	1.42
Salmonella heidelberg	8456	2.06 ± 0.07 (0.92)	1.59 ± 0.07	0.89
Salmonella derby	F5109	1.68 ± 0.01 (0.90)	1.68 ± 0.01	0.62
Salmonella saint-paul	5130	2.23 ± 0.03 (0.85)	1.89 ± 0.03	0.85
Salmonella derby	5131	1.89 ± 0.03 (0.88)	2.24 ± 0.03	0.88
Salmonella heidelberg	F5038BG1	2.55 ± 0.05 (0.92)	2.54 ± 0.05	0.34
Chicken				
Salmonella kentucky	FSIS 062	2.29 ± 0.00 (0.88)	1.84 ± 0.01	1.40
Salmonella kentucky	FSIS 044	1.59 ± 0.02 (0.94)	1.59 ± 0.02	0.76
Salmonella heidelberg	FSIS 109	2.33 ± 0.03 (0.89)	1.85 ± 0.03	1.36
Salmonella heidelberg	FSIS 134	1.43 ± 0.02 (0.97)	1.37 ± 0.04	0.60
Salmonella hadar	FSIS 127	1.98 ± 0.13 (0.88)	1.59 ± 0.01	1.30
Salmonella hadar	FSIS 064	2.15 ± 0.07 (0.97)	2.01 ± 0.07	0.55
Salmonella thompson	FSIS 132	1.46 ± 0.13 (0.96)	1.27 ± 0.06	0.69
Salmonella thompson	FSIS 120	2.32 ± 0.30 (0.93)	2.15 ± 0.30	0.98
Turkey				
Salmonella muenster	MF 61976	2.03 ± 0.18 (0.95)	1.76 ± 0.12	0.89
Salmonella muenster	MF 59707	2.25 ± 0.02 (0.88)	1.89 ± 0.03	1.56
Salmonella reading	MF 58210	1.44 ± 0.04 (0.87)	1.39 ± 0.05	0.47
Salmonella reading	MF 63447	2.04 ± 0.18 (0.92)	1.77 ± 0.16	1.52
Salmonella hadar	MF 61777	1.61 ± 0.10 (0.90)	1.28 ± 0.06	1.40
Salmonella hadar	MF 60404	2.12 ± 0.10 (0.96)	1.95 ± 0.01	0.82
Clinical				
Salmonella newport	H 1073	1.48 ± 0.09 (0.97)	1.33 ± 0.04	0.65
Salmonella enteritidis phage type 13A	H 3527	2.98 ± 0.08 (0.90)	2.34 ± 0.00	1.22
Salmonella enteritidis phage type 4	H 3502	1.85 ± 0.10 (0.88)	1.56 ± 0.07	1.24
Salmonella enteritidis phage type 8	H 3526	2.02 ± 0.09 (0.84)	1.63 ± 0.11	1.55
Salmonella typhimurium	H 3379	2.30 ± 0.05 (0.98)	2.15 ± 0.02	0.48
Salmonella typhimurium DT104	H 3380	2.41 ± 0.02 (0.89)	2.29 ± 0.07	0.90
Salmonella thompson	H 2464	2.40 ± 0.01 (0.98)	2.39 ± 0.15	0.62
Salmonella hadar	110-96	2.09 ± 0.40 (0.89)	2.09 ± 0.41	0.91
Salmonella enteritidis	H 4386	2.10 ± 0.17 (0.95)	2.10 ± 0.18	0.98
Salmonella branderup	H0663	2.29 ± 0.13 (0.89)	2.28 ± 0.13	1.04

[a]D-values shown are the means of two replicate experiments and expressed as mean ± standard deviation.
[b]Correlation coefficients in parenthesis.
[c]Root mean squares error.

curves with more than five values in the straight portion, with a correlation coefficient (r) > 0.90, and descending more than 5 log cycles were used.

Also, regression lines were fitted to experimental data points that contributed to shouldering by a linear function (model) that allows for the presence of a lag period before initiation of an exponential decline in population density (Buchanan 1993; 1994).

$$Y = Y_0 \quad \text{For } T \leq T_L$$
$$Y = Y_0 + m\,(T - T_L) \quad \text{For } T \leq T_L$$

Where:

Y = Log_{10} count of bacteria at time T [log_{10} (cfu/mL)]

Y_0 = Log_{10} count of bacteria at time T = 0 [log_{10} (cfu/mL)]

m = Slope of the survivor curve. [log_{10} (cfu/mL))/min]

T = Time (min)

T_L = Duration of lag period to initiation of inactivation (min)

The survivor curves were fitted using ABACUS, a nonlinear curve fitting program that employs a Gauss-Newton iteration procedure (Damert 1994). D-values (time to inactivate 90% of the population) were calculated as the negative reciprocal of m.

The z-values were estimated by computing the linear regression of mean log_{10} D-values versus their corresponding heating temperatures using Lotus 1-2-3 software.

Statistical analysis

The heat resistance data were analyzed by analysis of variance (ANOVA) using SAS (SAS 1989) to determine if there were statistically significant differences among the treatment/systems. The Bonferroni mean separation test was used to determine significant differences ($p < 0.05$) among means (Miller 1981).

Results and Discussion

THE PH OF THE CHICKEN BROTH AND MEAT USED IN THE study was approximately 6.3 and 6.0, respectively. Surviving populations of *Salmonella* cells per mL of chicken broth or per gram of ground meat of different meat species were determined and D-values calculated. *Salmonella* cells heated at 58 °C in chicken broth exhibited log-linear decline in surviving cells with time. No obvious lag periods or shoulders and tailing were evident in any of the survivor curves of bacteria heated in this menstruum. Such linear survival curves would suggest that the pathogen population was homogenous in heat resistance. However, significant variation in the heat resistance among strains in chicken broth was observed (Table 1). The thermal resistance (D-values in min) of *Salmonella* heated in chicken broth at 58 °C ranged from 1.54 min (*S. typhimurium)* to 2.16 min (*S. montevideo)* for beef isolates, 1.29 (*S. derby)* to 2.94 min (*S. copenhagen)* for pork isolates, 1.43 (*S. heidelberg, FSIS #134)* to 2.33 min (*S. heidelberg,* FSIS # 109) for chicken isolates, 1.44 (*S. reading)* to 2.25 min (*S. muenster)* for turkey isolates, and 1.48 (*S. newport)* to 2.98 min (*S. enteritidis,* phage type 13A) for clinical isolates (Table 1). Regression curves calculated for 58 °C fit with an r^2 value of > 0.90. As shown in Table 1, D- values calculated by a linear model were very similar. Based on a minimal root mean square value, the thermal inactivation data could be fitted well to generate survivor curves. In the present study, no correlation between the heat resistance at 58 °C and the origin of the *Salmonella* serotype (food animal or human clinical) could be established due to significant variation in the heat resistance among strains. Understanding these variations in heat resistance is necessary in order to design adequate thermal inactivation regimes to eliminate *Salmonella* in thermally processed foods. In the next series of experiments, the D-values at 55, 60, and 62 °C of the *Salmonella* isolates from each species of meat and poultry, and clinical isolates, exhibiting highest heat resistance in chicken broth at 58 °C, were determined and are shown in Table 2. The D-values calculated by linear regression in chicken broth at 55 °C ranged from 5.86 min for *S. copenhagen* 8457 (pork isolate) to 3.77 min for *S. hadar* MF60404 (turkey isolate); the D-values at 62 °C were 0.40 and 0.32 min, respectively. The D-values calculated by a linear model were very similar to those obtained by linear regression. Again, no lag periods or shoulders and tailing were observed in any of the survivor curves observed at the four test temperatures. The heat resistance of the cocktail of the 8 *Salmonella* serotypes, representing isolates from each species of meat and poultry exhibiting highest heat resistance, was also assessed. Interestingly, the survivor curves exhibited no apparent shoulders or tailing. The D-values, using a linear regression, were 4.87, 2.72, 1.30, and 0.41 min at 55, 58, 60, and 62 °C, respectively (Table 2). When regression lines were fitted to the experimental data using the linear model, the D-values ranged from 4.86 min at 55 °C to 0.38 min at 62 °C.

When the 8 *Salmonella* serotype cocktail was heated in meat, D-values at the common test temperatures of 58 and 60 °C calculated by both approaches were significantly higher ($p < 0.05$) than those observed in chicken broth. Inactivation kinetics in meat, unlike in chicken broth, showed deviations from the first order kinetics; that is, survivor curves exhibited an initial lag period or shoulder before any death occurred in meat. The "shoulder effect" observed may be attributed to the poor heat transfer through the heating menstruum, or may be due to an initial requirement for the bacterial cells to sustain sufficient injury before the first order inactivation kinetics in the log number of survivors with time. Alternatively, the switch in the thermotolerance response from linear survival curves to nonlinear may be attributed to variability of heat resistance within a bacterial population (Hansen and Rieman 1963). Using the simple linear regression analysis, the D-values of the 8 *Salmonella* serotype cocktail in beef were 8.65, 5.48, 1.50, and 0.67 min at 58, 60, 62.5, and 65 °C, respectively. The D-values in pork, obtained by linear regression, ranged from 6.68 min at 58 °C to 0.87 min at 65 °C; the values ranged from 7.42 min to 0.80 min and 7.08 min to 0.59 min in turkey and chicken, respec-

Table 2—Heat resistance (expressed as D-values in min)[a] for *Salmonella* spp. in chicken broth at 55–62 °C

			Method to Determine D-Value		
Strain	Isolate	Temp °C	Linear Regression D-Value (r^2)[b]	Linear Model D-Value	RMS error[c]
Salmonella thompson	FSIS 120	55	4.05 ± 0.01 (0.92)	4.05 ± 0.02	0.81
		58	2.32 ± 0.30 (0.93)	2.17 ± 0.30	0.85
		60	0.83 ± 0.00 (0.91)	0.83 ± 0.00	0.85
		62	0.34 ± 0.02 (0.97)	0.34 ± 0.02	0.46
Salmonella enteritidis Phage Type 13A	H 3527	55	5.74 ± 0.09 (0.99)	5.34 ± 0.00	0.12
		58	2.98 ± 0.08 (0.90)	2.34 ± 0.00	1.12
		60	0.94 ± 0.01 (0.94)	0.83 ± 0.01	0.54
		62	0.39 ± 0.01 (0.97)	0.39 ± 0.01	0.90
Salmonella enteritidis Phage Type 13A	H 3527	55	3.81 ± 0.00 (0.49)	3.83 ± 0.02	0.39
		58	1.85 ± 0.10 (0.88)	1.56 ± 0.07	1.24
		60	0.89 ± 0.01 (0.01)	0.89 ± 0.00	0.59
		62	0.30 ± 0.00 (0.94)	0.30+0.00	0.59
Salmonella typhimurium DT104	H 3380	55	4.16 ± 0.03 (0.92)	4.17 ± 0.02	0.92
		58	2.41 ± 0.02 (0.89)	2.29 ± 0.07	0.90
		60	0.75 ± 0.09 (0.83)	0.44 ± 0.22	1.15
		62	0.27 ± 0.00 (0.90)	0.28 ± 0.00	1.01
Salmonella hadar	MF 60404	55	3.77 ± 0.11 (0.93)	3.28 ± 0.04	0.82
		58	2.12 ± 0.10 (0.96)	1.95 ± 0.01	0.82
		60	0.89 ± 0.01 (0.92)	0.89 ± 0.01	0.83
		62	0.32 ± 0.00 (0.96)	0.32 ± 0.00	0.42
Salmonella copenhagen	8457	55	5.86 ± 0.47 (0.99)	5.87 ± 0.45	0.17
		58	2.94 ± 0.22 (0.96)	2.50 ± 0.14	0.79
		60	0.99 ± 0.02 (0.94)	0.99 ± 0.02	0.80
		62	0.40 ± 0.01 (0.92)	0.40 ± 0.01	0.77
Salmonellla montevideo	FS18S 051	55	4.05 ± 0.03 (0.97)	4.05 ± 0.03	0.49
		58	2.16 ± 0.20 (0.94)	1.75 ± 0.01	1.10
		60	0.84 ± 0.00 (0.98)	0.84 ± 0.01	0.40
		62	0.24 ± 0.01 (0.93)	0.24 ± 0.01	0.88
Salmonella heidelberg	F5038BG 1	55	4.85 ± 0.01 (0.94)	4.84 ± 0.02	0.68
		58	2.55 ± 0.05 (0.92)	2.54 ± 0.05	0.34
		60	1.02 ± 0.02 (0.99)	1.03 ± 0.01	0.33
		62	0.34 ± 0.01 (0.92)	0.34 ± 0.01	0.88
Salmonella cocktail[d]		55	4.87 ± 0.10 (0.97)	4.86 ± 0.03	0.45
		58	2.72 ± 0.04 (0.88)	2.68 ± 0.02	0.74
		60	1.30 ± 0.02 (0.97)	1.31 ± 0.04	0.33
		62	0.41 ± 0.01 (0.97)	0.38 ± 0.01	0.50

[a]D-values shown are the means of two replicate experiments and expressed as mean ± standard deviation.
[b]Correlation coefficients in parenthesis.
[c]Root mean squares error.
[d]Mixture of 8 *Salmonella* serotypes.

Table 3—Heat resistance (expressed as D-values in min)[a] for an eight strain *Salmonella spp.* cocktail in beef (12.5% fat), chicken (7.0% fat), turkey (9.0% fat) and pork (8.5% fat) at 58 650 °C

		Method to Determine D-Value				
		Linear Regression		Linear Model		
Meat	Temp °C	D-Value	r^2[b]	D-Value	TL[c]	RMS error[d]
Beef						
	58	8.65 ± 0.03	.99	8.85 ± 0.01	0.2	0.21
	60	5.48 ± 0.04	.99	5.26 ± 0.06	2.32	0.08
	62.5	1.50 ± 0.01	.84	1.47 ± 0.01	1.82	0.15
	65	0.67 ± 0.04	.94	0.53 ± 0.06	1.15	0.17
Pork						
	58	6.68 ± 0.02	0.98	6.37 ± 0.01	2.65	0.24
	60	6.65 ± 0.05	0.99	6.60 ± 0.04	0.47	0.20
	62.5	1.62 ± 0.15	0.82	1.57 ± 0.05	0.60	0.15
	65	0.87 ± 0.22	0.96	0.73 ± 0.08	1.09	0.17
Turkey						
	58	7.42 ± 0.12	0.99	7.19 ± 0.00	2.49	0.24
	60	4.82 ± 0.04	0.99	4.82 ± 0.03	0.00	0.02
	62.5	1.51 ± 0.01	0.89	1.51 ± 0.00	0.08	0.39
	65	0.80 ± 0.01	0.97	0.73 ± 0.02	0.70	0.23
Chicken						
	58	7.08 0.2	0.99	7.07 ± 0.01	0.00	0.21
	60	5.20 0.15	0.99	5.19 ± 0.11	0.00	0.18
	62.5	1.36 0.01	0.87	1.35 ± 0.02	0.13	0.45
	65	0.59 0.07	0.92	0.45 ± 0.04	1.20	0.38

[a]D-values shown are the means of two replicate experiments and expressed as mean ± standard deviation.
[b]Correlation coefficients.
[c]Lag period.
[d]Root mean squares error.

Table 4—Heat resistance expressed as z-values[a] in °C for *Salmonella* serotypes in chicken broth at 58–62 °C

Method to Determine D-Value		Linear Regression	
		Linear Model	
Strain	Isolate	z-value (r^2)	z-Value (r^2)
Salmonella thompson	FSIS 120	6.41(0.95)	6.44(0.96)
Salmonella enteritidis phage type 13A	H 3527	5.86(0.96)	6.03(0.98)
Salmonella enteritidis phage type 4	H 3502	6.46(0.96)	6.43(0.97)
Salmonella typhimurium DT 104	H 3380	5.80(0.94)	5.53(0.93)
Salmonella hadar	MF 60404	6.56(0.95)	7.00(0.94)
Salmonella copenhagen	8457	5.91(0.97)	5.97(0.99)
Salmonella montevideo	FSIS 051	5.77(0.94)	5.85(0.96)
Salmonella heidelberg	F5038BGI	6.10(0.95)	6.11(0.99)

[a]Z-values were determined by the means of replicate D-values obtained in chicken broth and based on survivors on the recovery medium.

tively (Table 3). When survivor curves were generated by fitting the data to the linear function that allows for the presence of a lag period, the D-values of the 8 *Salmonella* serotype cocktails in beef were 8.85, 5.26, 1.47, and 0.53 min at 58, 60, 62.5, and 65 °C, respectively, and a maximum lag period of 2.32 min was observed at 60 °C. The D-values in pork, obtained by linear function, ranged from 6.37 min at 58 °C to 0.73 min at 65 °C; the values ranged from 7.19 min to 0.73 min and 7.07 min to 0.45 min in turkey and chicken, respectively. A maximum lag period of 2.65 min at 58 °C was observed in pork (Table 3). The increased thermal resistance of the *Salmonella* spp. in meat, compared to chicken broth, may be attributed to differences in composition (more solids in meat) between the substrates. A possible explanation for the slight differences in the D-values among the different meat species could be the effect of different meat species and the differences in fat content between the substrates. Jay (1986) indicated that food carbohydrates, fats, proteins, salt, and so on, confer protection to bacterial cells or spores against heat. In general, the heat resistance of any given microorganism is known to be affected not only by inherent genetic factors, but also by many environmental factors during heating such as the composition and pH of the heating menstruum (Tomlins and Ordal 1976; Hansen and Riemann 1963). Thus, it would be inappropriate to predict the thermal death time values or design thermal processes in one meat species from data obtained in other meat species or in broth.

Thermal death time curves for the 8 strains of *Salmonella* serotype cocktail were plotted from D-values obtained from heating bacteria in chicken broth and in different meat species to calculate z-values. For chicken broth, the z-values of all strains, including the cocktail, were very similar, ranging from 5.77 to 6.62 °C and 5.53 to 7.00 °C obtained using D-values calculated by linear regression and the linear model, respectively (Table 4; Figure 1). The z-values in meat ranged from 6.01 to 7. 10 °C (using D-values obtained by a linear regression) and 8.83 to 9.11 °C (using D-values calculated by a linear model; Figure 2). A possible explanation for the higher z-values in the latter case could be due to the heated cells exhibiting varying degrees of lag periods at different temperatures; such lag periods observed were added to the observed D-values. Our study indicates that larger changes in

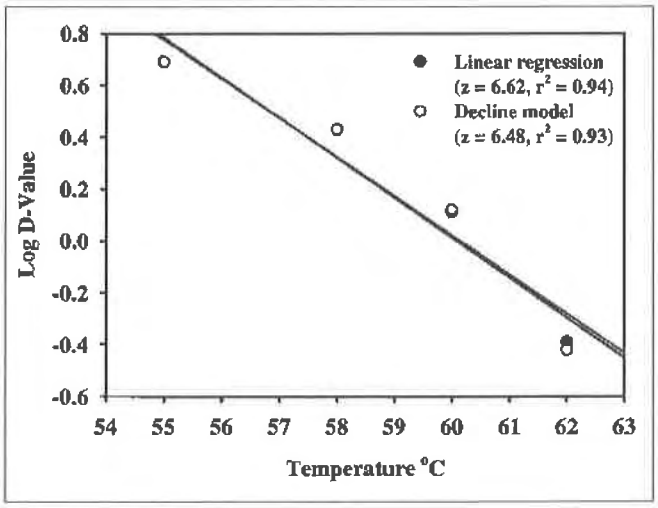

Figure 1—Thermal-death-time curves (z-values) for 8 *Salmonella* serotype cocktail over the temperature range 55 to 62 °C. The D-values in chicken broth, calculated by linear regression, and those calculated by curve fitting, used to determine the z-values were the means of two replicates and were obtained based on survivors on the recovery medium.

temperature are required to cause a 90% reduction in the D-value when a *Salmonella* spp. cocktail is evaluated in meat compared to chicken broth. Again, it would be inappropriate or invalid to determine z-values under one set of food formulation variables and applying to another set of parameters in foods.

It is feasible to compare the thermal inactivation data obtained in this study with those in the published literature on the heat resistance of *Salmonella* spp. The thermal inactivation data reported in this study were largely inconsistent with those reported elsewhere. Goodfellow and Brown (1978) reported D-values at 51.6, 57.2, and 62.7 °C of 61 to 62, 3.8 to 4.2, and 0.6 to 0.7 min, respectively, for a mixture of six *Salmonella* serotypes inoculated into ground beef. In a study by Orta-Ramirez and others (1997), when heat resistance of *S. senftenberg* in ground beef heated in thermal death time tubes was determined, the D-values ranged from 53.0 to 0.22 min at 53 to 68 °C, with a z value of 6.25 °C. In another study, Veeramuthu and others (1998) reported that the D-values for *S. senftenberg* in ground turkey (4.3% fat) heated in thermal death time tubes ranged from 211.35 min at 55 °C to 3.43 min at 65 °C and a z-value of 5.6 °C. While our D-values were lower than those reported earlier (mentioned above), the z-values were higher. It is worth emphasizing that, while we used a linear model for nonlinear survival curves to account for the lag periods and subsequently added the lag periods to the observed D-values, the previous studies (Goodfellow and Brown 1978; Orta-Ramirez and others 1997; Veeramuthu and others 1998) calculated D-values using only linear regression analysis for the best fit line of the survivor curve. When comparing the results obtained in the present study with those reported from other studies, it should always be kept in mind that meat species, muscle type, pH, fat content, and other environmental factors including the method of enumeration may influence the bacterial heat resistance results (Hansen and Riemann 1963; Stumbo 1973; Ahmed and Conner 1995). Also, certain strains of *Salmonella* are less resistant and are less tolerant to changes in temperature. These factors may have attributed to the inconsistency of results of *Salmonella*, spp. that is, heat resistance defined by D- and z-values, observed in the present study and those reported in the published literature.

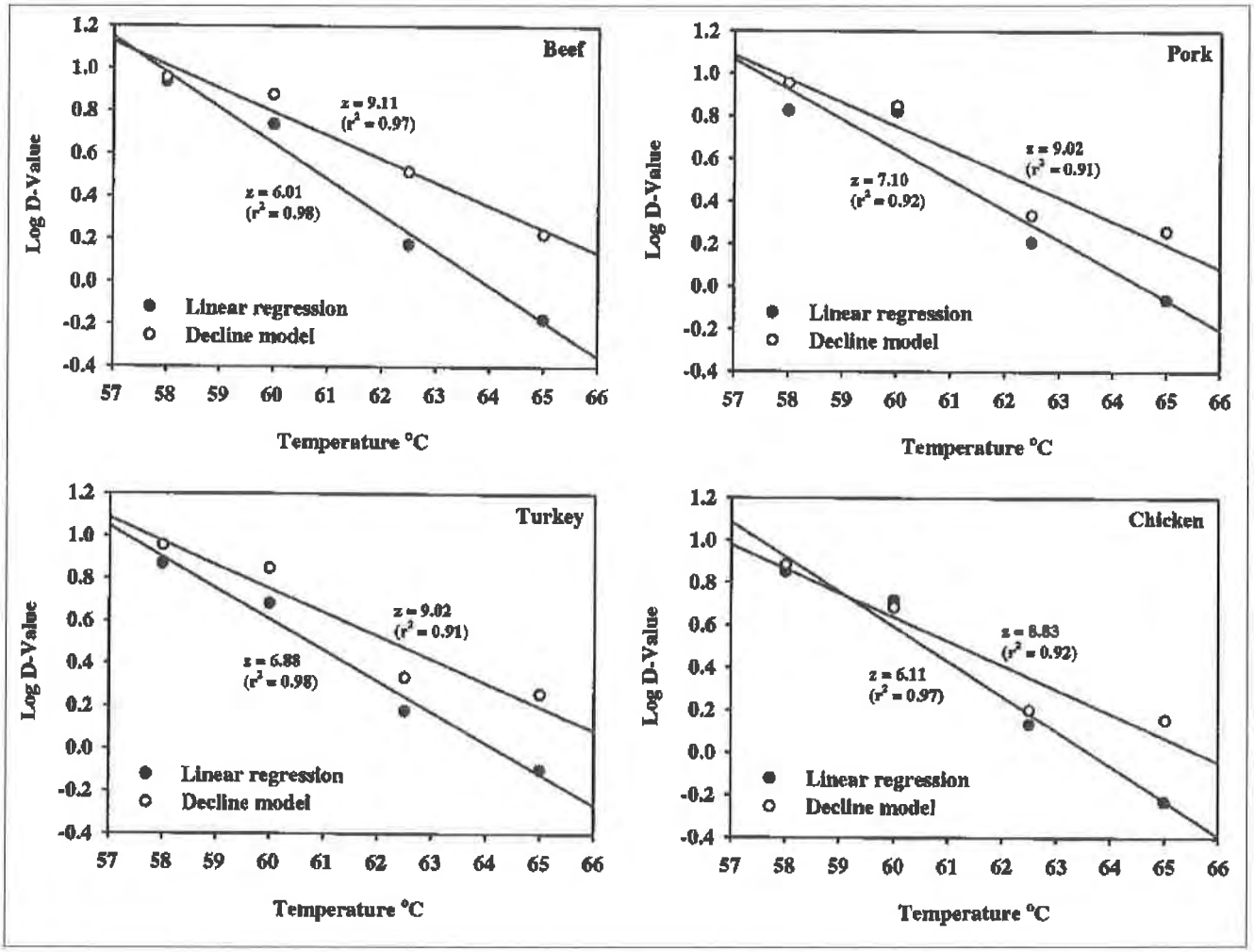

Figure 2—Thermal-death-time curves (z-values) for 8 *Salmonella* serotype cocktails over the temperature range 58 to 65 °C. The D-values in beef, pork, turkey, and chicken, calculated by linear regression, and those calculated by curve fitting, used to determine the z-values were the means of two replicates and were obtained based on survivors of the recovery medium.

Conclusions

THE RESULTS OF THE PRESENT STUDY CAN BE USED TO PREdict the time required at specified temperatures to achieve specific targeted $\log_{10}$ reductions such as 6.5 $\log_{10}$ or 7 $\log_{10}$ reductions of *Salmonella* spp. when heated in ground beef, pork, turkey, or chicken. Based on the thermal-death-time values determined in this study, contaminated ground beef should be heated to an internal temperature of 60.0 °C for at least 53.1 min, ground pork for 49.50 min, ground turkey for 33.74 min, and ground chicken for 36.33 min; this is based on the argument that thermal treatments must be designed to achieve a 7-D process for *Salmonella spp.* Thermal death time values from this study determined for the specific meat species will assist food processors in designing acceptance limits on critical control points that ensure safety against *Salmonella* spp. in cooked ground meat.

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Thermal Inactivation D- and Z-Values of *Salmonella* and *Listeria innocua* in Fully Cooked and Vacuum Packaged Chicken Breast Meat during Postcook Heat Treatment

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ABSTRACT Studies were conducted to determine thermal inactivation D- and z-values of *Salmonella* and *Listeria innocua* in fully cooked and vacuum-packaged chicken breast meat. Fully cooked chicken breast meat products that were obtained from three different sources with differing formulations were uniformly inoculated with a cocktail of *Salmonella* (including Senftenberg, Typhimurium, Heidelberg, Mission, Montevideo, and California) or *L. innocua* at approximately 10^7 cfu/g. The inoculated meat samples were vacuum-packaged and then heat-treated at a temperature of 55 to 70 C for 5 to 90 min. After heat treatment, the samples were immediately cooled in an ice-water bath. Survivors of *Salmonella* and *L. innocua* were enumerated for each sample. D- and z-values of *Salmonella* and *L. innocua* were determined for each product and compared among the products. Source and formulation did not cause significant differences in the D- and z-values of *Salmonella* or *L. innocua* among the three fully cooked and vacuum-packaged chicken breast meat products.

(*Key words*: *Salmonella*, *Listeria*, thermal inactivation, postcook, fully cooked chicken)

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INTRODUCTION

The market for fully cooked refrigerated meat and poultry products is rapidly increasing. Consequently, it becomes more and more important to ensure the food safety of these thermally processed retail products. Many of the fully cooked meat or poultry products were recalled due to concern of pathogens (USDA, 2001). Once a cooking process is validated, fully cooked meat or poultry products should be free of pathogens at the end of cooking. However, potential recontamination during postcook handling, prior to packaging, poses a concern. Such an event could introduce pathogens to packaged ready-to-eat meat or poultry products.

In-package pasteurization processes via steam or hot water have been used or considered for use on fully cooked meat and poultry products (Murphy, 2002, unpublished data). Murphy et al. (2001b) demonstrated that postcook pasteurization treatment via steam or hot water could effectively reduce the potential risk of *Salmonella* and *Listeria* in packaged ready-to-eat chicken breast meat products. Thermal pathogen destruction during pasteurization is time and temperature related and is usually described using the time required to cause a one $\log_{10}$ decrease in bacterial numbers at a given temperature (D) and the temperature difference required for the thermal inactivation curve to drop a logarithmic cycle (z). From a known z-value, process lethality (F) for a pathogen during pasteurization treatment can be calculated as

$$F = \int_0^t 10^{(T(t) - T(ref))/z} dt \qquad [1]$$

where T(t) is the product temperature at a time t and T(ref) is a reference temperature. To determine the thermal lethality for pathogens in a meat or poultry product that has gone through a pasteurization treatment, the z-values of bacteria in the product should be used. Thermal inactivation D- and z-values of pathogens in different raw meat or poultry products were reported in previous publications (Mazzotta, 2000; Juneja et al., 2001; Murphy et al., 2002). However, no data were generated regarding thermal inactivation kinetic values for pathogens in fully cooked and vacuum-packaged meat or poultry products, and these data are essential in validating postcook pasteurization process.

To increase profitability, most fully cooked and vacuum-packaged commercial meat and poultry products

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Abbreviation Key: ANCOVA = analysis of covariance; TSA = tryptic soy agar

are value-added or marinated prior to cooking processes. Generally, various food additives are used in marinade formulas. Differing accounts can be found in the literature regarding how product formulations affect the thermal lethality of pathogens (Kotrola and Conner, 1997; Doyle and Mazzotta, 2000). In a review paper, Doyle and Mazzotta (2000) indicated that some food additives, including polyphosphates, hydrogen peroxide, and the lactoperoxidase increased *Salmonella* sensitivity to heat. Also, these food additives could be more effective in culture media than in foods because they might interact with fat and protein, which reduced their availability to bacteria (Doyle and Mazzotta, 2000).

In a study using finely ground turkey breast meat (3% fat) as base heating medium, Kotrola and Conner (1997) found that some food additives, such as NaCl, sodium lactate, and polyphosphates increased heat resistance of *Escherichia coli* O157:H7. However, the heat resistance of *E. coli* O157:H7 in ground turkey breast meat was not affected by increasing fat content from 3 to 11% (Kotrola and Conner, 1997). In a previous study on different commercial meat products, Murphy et al. (2002) found that thermal inactivation D- and z-values for *Salmonella* and *Listeria innocua* were significant (at $\alpha = 0.05$) among different commercially formulated meat and poultry products, including chicken breast meat, chicken patties, chicken tenders, franks, beef patties, and blended beef and turkey patties. In the above studies, raw meat products were used.

No information could be retrieved from the literature on thermal inactivation D- and z-values of pathogens in fully cooked and vacuum-packaged meat or poultry products. It was also unknown how marinade formulation prior to cooking could affect pathogen thermal lethality in fully cooked and vacuum-packaged meat or poultry products during postcook heat treatment. The objective of this study was to determine thermal inactivation D- and z-values of *Salmonella* and *Listeria* in fully cooked and vacuum-packaged chicken breast meat products. Fully cooked chicken breast meat products from three different sources were evaluated, including (1) plain chicken breast meat that was cooked in a pilot-scale impingement oven, (2) product marinated with phosphate, salt, and water via vacuum tumbler and then cooked in a pilot-scale impingement oven, and (3) fully cooked chicken breast meat products that were commercially formulated, marinated, and cooked.

MATERIALS AND METHODS

Product

Three types of fully cooked chicken breast meat were used in this study, including (1) plain meat product without any additives, (2) product marinated with phosphate and salt prior to cooking, and (3) commercially formulated grilled chicken breast fillets. For plain meat product (1), fresh chicken breast fillets (4 C) were obtained from University of Arkansas Poultry Processing Pilot Plant and cooked in steam (99 C) via an impingement oven[2] to the minimum internal temperature of 71.1 C. For product that was marinated with phosphate and salt (2), fresh chicken breast meat fillets (4 C) were obtained from a commercial poultry processor. The meat was marinated for 60 min via a vacuum tumbler in a solution, containing 0.5% (wt/wt) sodium tripolyphosphate,[3] 2% (wt/wt) sodium salt[4], 70% (wt/wt) water, and 27.5% (wt/wt) ice. Marinated product was then cooked in steam at 99 C in an impingement oven[5] to the minimum internal temperature of 71.1 C. For commercially formulated product (3), chicken breast meat was marinated by injection, formed, and thermally processed by a commercial processor. Detailed product formulation and process information were proprietary to the processor. However, the marinade formulation contained NaCl (4%, wt/wt) and sodium tripolyphosphate (2.4%, wt/wt).

All three products contained about 20.11% protein, 73.64% moisture, and 1.27% fat. For the products (1) and (2), the endpoint internal temperatures of the products during cooking were correlated to cooking time. The correlation of product temperature and cooking time was predetermined via 20 cooking test trials. During the test trials, the internal temperatures of the products were monitored via thermocouple probes in a similar manner as described by Murphy et al. (2001a). The product internal temperatures were correlated to cooking time and verified using a computer simulation program (ThermoPro, Rong Murphy, Thermal Processing and Food Safety Program, University of Arkansas, Fayetteville, AR).

Bacteria

A cocktail of six *Salmonella* serotypes (Senftenberg, Typhimurium, Heidelberg, Mission, Montevideo, and California) were used in this study (Murphy et al., 1999). A nalidixic acid-resistant culture of each serotype was prepared as previously described by Murphy et al. (1999). Subcultures for use as inocula were prepared from the stock culture. A preliminary study was conducted to compare the thermal tolerance of nalidixic acid-resistant culture with the original nonresistance culture and found no significant difference (Murphy, 2001, unpublished data).

The same *L. innocua* M1 strain as that used by Murphy et al. (1999) was used in this study. *Listeria innocua* M1 has been used as a biological indicator for *L. monocytogenes* (Foegeding and Stanley, 1991; Fairchild and Foegeding, 1993). Lyophilized culture was revived in tryptic soy broth[6] plus 0.6% yeast extract[7] for 24 h at 35 C, plated on tryptic soy agar (TSA) plus 0.6% yeast extract, and incubated for 24 h at 35 C. The *Listeria* culture was resistant to 50 ppm rifampicin and 250 ppm streptomycin. A study by Walsh et al. (2001) indicated antibiotic-resistant

[2]Model 102, Stein-DSI, Sandusky, OH.
[3]George's Inc., Springdale, AR.
[4]George's Inc., Springdale, AR.
[5]Model 102, Stein-DSI, Sandusky, OH.
[6]Becton Dickinson and Company, Sparks, MD.
[7]Becton Dickinson and Company, Sparks, MD.

Listeria did not respond differently to thermal treatments in meat substrate from the original strain.

Heat Treatment

Fully cooked chicken breast meat was ground in a sterile Cuisinart™ food processor[8] and inoculated with *Salmonella* or *L. innocua*. An actively growing culture of *Salmonella* or *L. innocua* (10 mL) was used per 500 g of fully cooked chicken. The culture was individually maintained and grown overnight in tryptic soy broth plus 200 ppm nalidixic acid sodium salt[9] for *Salmonella* or tryptic soy broth plus 0.6% yeast extract, 50 ppm rifampicin[10], and 250 ppm streptomycin[11] for *L. innocua*. The six *Salmonella* cultures were mixed to form a cocktail just prior to the inoculation.

The inoculated samples were kept at 4 C for 30 min to allow attachment of the bacterial cells to the meat tissues. Samples of 25 g were then placed in a 152 mm wide × 254 mm long × 0.0508 mm thick gas/moisture barrier pouch[12] and sealed under 1 bar vacuum. The sealed samples were rolled flat into a thin layer with a rolling pin. The flatted samples filled all of the space in the pouch and had a thickness of approximately 0.5 mm.

The sample pouches were placed flat inside a stainless steel wire rack and immediately submerged in a circulated water bath that was maintained at 55, 57.5, 60, 62.5, 65, 67.5, or 70 C for 5 s to 90 min depending on the treatment temperature. In each trial, two inoculated and unheated controls were prepared to determine the initial inoculation values. After heat treatment, the samples were immediately placed in an ice-water bath. During heating and cooling, time temperature history was monitored via a thermocouple probe (40 gauge, type E) that was sealed in the pouch. Within 20 s of treatments, meat sample temperatures increased from 20 C to within 0.5 C of the heating-bath temperature. Eight seconds in the subsequent ice bath caused the meat sample temperature to fall below 20 C.

Enumeration

After cooling, pouches were blotted dry with paper towels, wiped with 75% ethanol, and a corner of the pouch was cut in 2.5 cm slot. Peptone[13] (0.1%; 60 mL) was slowly pipetted into the pouch. The sample solution mixture was manipulated by hand, transferred into a nylon mesh-lined bag, and blended in a Stomacher[14] for 2 min. Serial dilutions were plated in duplicate. *Salmonella* was plated on TSA overlaid with TSA containing 200 ppm of nalidixic acid sodium salt. *Listeria* was plated on TSA overlaid with TSA containing 0.6% yeast extract, 50 ppm of rifampicin, and 250 ppm of streptomycin. Inoculated unheated samples and uninoculated heated samples were included as controls.

All plates were incubated at 35 C and colonies were counted daily. Plates were returned to the incubator and recounted every day for 7 d until viable counts did not increase further.

Survival Models

For each product, the survivors, $\log_{10}(N)$, of *Salmonella* or *Listeria* were plotted against heating times at each temperature. The following linear primary model (Murphy et al., 2000) was used to model the thermal destruction of *Salmonella* and *Listeria* and to determine the decimal reduction time (D).

$$\text{Log}_{10}(N) = \text{Log}_{10}(N_0) \quad [t \le t_L] \qquad [2]$$

$$\text{Log}_{10}(N) = \text{Log}_{10}(N_0) + s\,(t - t_L) \quad [t > t_L] \qquad [3]$$

where N was colony-forming unit (cfu) per gram at time t, N_0 was cfu/g of inoculated unheated samples, s = slope of the survival curve, t = time (min), and t_L = duration of lag period (min).

D- and z-values

Triplicate thermal inactivation trials were performed at each temperature. The D-values for *Salmonella* and *Listeria* at each temperature were calculated by taking the negative inverse of the relevant s-value (Murphy et al., 2000). The z-values were determined as the negative inverse slope of the $\log_{10}$D vs. temperature plot.

Statistical Analyses

To test if D-values of *Salmonella* or *L. innocua* were equal at each temperature for the three products, assuming N_0 was the count of *Salmonella* or *L. innocua* at time = 0 and N was the count at time = t, for each temperature, an

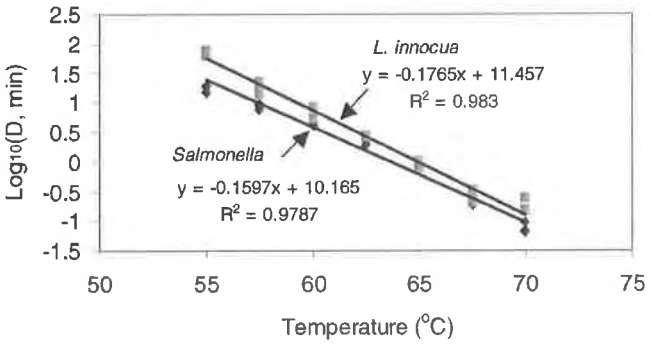

FIGURE 1. $\log_{10}$(D, min) of *Salmonella* and *Listeria innocua* vs. treatment temperature for fully cooked chicken breast products.

[8]Model CFP 5A, Robot-Coupe, Stamford, CT
[9]Sigma Chemical Co., St. Louis, MO.
[10]Sigma Chemical Co., St. Louis, MO.
[11]Sigma Chemical Co., St. Louis, MO.
[12]Tilia, Inc., San Francisco, CA.
[13]Becton Dickinson and Company, Sparks, MD.
[14]Lab Blender 400, Tekmar Co., Cincinnati, OH.

TABLE 1. The P values from the Analysis of Covariance for the effect of heating time and product on $\log_{10}(N/N_0)$ of *Salmonella* and *Listeria innocua* in fully cooked and vacuum-packaged chicken breast meat

Culture	Temperature (°C)	Source	*P*-value
Salmonella	55	Product	0.8449
		Time (min)	<0.0001
		Time × product	0.0881
	57.5	Product	0.4783
		Time (min)	<0.0001
		Time × product	0.0543
	60	Product	0.4030
		Time (min)	<0.0001
		Time × product	0.3756
	62.5	Proudct	0.1595
		Time (min)	<0.0001
		Time × product	0.4082
	65	Product	0.0972
		Time (min)	<0.0001
		Time × product	0.8848
	67.5	Product	0.5715
		Time (min)	<0.0001
		Time × product	0.7763
	70	Product	0.9575
		Time (min)	<0.0001
		Time × product	0.3185
L. innocua	55	Product	0.4468
		Time (min)	0.0002
		Time × product	0.9892
	57.5	Product	0.2450
		Time (min)	<0.0001
		Time × product	0.2950
	60	Product	0.5498
		Time (min)	<0.0001
		Time × product	0.2745
	62.5	Product	0.4662
		Time (min)	<0.001
		Time × product	0.9598
	65	Product	0.0852
		Time (min)	<0.0001
		Time × product	0.5308
	67.5	Product	0.9325
		Time (min)	<0.0001
		Time × product	0.3089
	70	Product	0.7718
		Time (min)	<0.0001
		Time × product	0.1539

Analysis of Covariance (ANCOVA) was performed where $\log_{10}(N/N_0)$ was the response, treatment was the effect of product formulations, and time was the covariate. The interaction between product formulation and time was also included in the test.

To test whether the z-value of *Salmonella* or *L. innocua* was equal, $\log_{10}(D)$ was the response, treatment was the effect of product formulations, and temperature was the covariate. The interaction between product formulation and temperature was also tested. The D-values of *Salmonella* and *L. innocua* were obtained at each treatment temperature for each product (plain, marinated with phosphate and salt, or commercially formulated). Letting D_S and D_L represent the decimal reduction time for *Salmonella* and *Listeria*, respectively, and the T represent the temperature, the $\log_{10}(D_S)$ or $\log_{10}(D_L)$ would be the response, product formulation would be the explanatory variable, and T would be the covariate. The data were fitted to the following linear regression model:

for *Salmonella*:

$$\log_{10}(D_S)_i = a + b\,\delta_{i2} + c\,\delta_{i3} + f\,(temp) + g\,(temp)\delta_{i2} + h\,(temp)\delta_{i3} \quad [4]$$

for *L. innocua*:

$$\log_{10}(D_L)_i = a + b\,\delta_{i2} + c\,\delta_{i3} + f\,(temp) + g\,(temp)\delta_{i2} + h\,(temp)\delta_{i3} \quad [5]$$

where i = 1 and 3 corresponds to fully cooked chicken breast meat that was commercially formulated, plain, and marinated with phosphate and salt, respectively, and $\delta_{ij} = \begin{cases} 1 \text{ if } i = j \\ 0 \text{ otherwise} \end{cases}$, where a, b, c, f, g, and h were the parameters to be determined.

Using SAS,[15] the Analysis of Covariance Type III SS (sum of square) tests were conducted to determine the effect of product formulation and heating temperatures

[15]Release 8.1, copyright 1999–2000, SAS Institute, Inc., Cary, NC.

TABLE 2. The *P* values from the Analysis of Covariance Type III SS test for the effect of temperature and product on the z-values of *Salmonella* and *Listeria innocua* in fully cooked and vacuum-packaged chicken breast meat[1]

Culture	Parameter	*P*
Salmonella	Temperature	<0 0001
	Product formulation	0 4364
	Temperature × product formulation	0 4360
L. innocua	Temperature	<0 0001
	Product formulation	0.6110
	Temperature × product formulation	0.5188

[1]R^2 was 0.9810 for *Salmonella* and 0 9875 for *L. innocua*.

on z-values. Paired comparisons were also conducted to determine the significant differences of the z-values between each pair of the products. The comparisons were also conducted using SAS.

RESULTS AND DISCUSSION

At each heating temperature, no difference in time and temperature history was observed among the three products. The analysis for the effect of heating time and product formulation on $\log_{10}(N/N_0)$ of *Salmonella* or *L. innocua* included a constant (intercept) term, a heating time (linear) term, and the effect of product on each term. The negative reverse of the linear term (slope) was the D-value for *Salmonella* or *L. innocua* at each corresponding temperature. Table 1 gives the result from ANCOVA analysis for $\log_{10}(N/N_0)$ of *Salmonella* and *L. innocua* at a heating temperature of 55 to 70 C. All of the *P* values for heating time were less than 0.0002, indicating that at temperatures 55 to 70 C, the heating time significantly (at $\alpha = 0.05$) affected the survivors, $\log_{10}(N/N_0)$, of *Salmonella* and *L. innocua*. The *P* values for all of the terms between time and product were greater than $\alpha = 0.05$, indicating that product formulation had no significant effect on bacterial numbers at $\alpha = 0.05$.

In Equations 4 and 5, $\log_{10}(D)$ of *Salmonella* or *L. innocua* was a linear function of treatment temperature. The negative inverse of the linear term (slope) for $\log_{10}(D)$ vs temperature was the z-value. Table 2 gives the results from ANCOVA Type III analysis that was conducted to determine the effect of the three products on the z-values of *Salmonella* or *L. innocua*. Treatment temperature affected the z-values of *Salmonella* and *L. innocua* ($P < 0.05$). However, there was no significant difference in the z-value of *Salmonella* and *L. innocua* among the three products. The product did not interact with treatment temperature ($P > 0.05$). Paired comparison tests were also conducted to determine whether the z-values of *Salmonella* or *L. innocua* between each two pairs of products were significantly different (Table 3). The z-values of *Salmonella* or *L. innocua* were not different between any two pairs of the products.

Since the D- and z-values of *Salmonella* or *L. innocua* were not significantly different among the three fully cooked chicken breast meat products, the data set for all three products was combined at each treatment temperature and time. At each heating temperature, linear regressions were performed for Equations 1 and 2 to obtain the plot of the survivors, $\log_{10}(N)$, of *Salmonella* or *L. innocua* vs. heating time. The D-values of *Salmonella* and *L. innocua* at each treatment temperature were obtained from these plots and are presented in Table 4. This resulted in 90 observations at each treatment temperature, and R^2 was greater than 0.86 for all of the regressions. The D-value of *Salmonella* ranged from 24.071 ± 1.852 min at 55 C to 0.097 ± 0.034 min at 70 C, and the D-value of *L. innocua* was from 56.169 ± 4.016 min at 55 C to 0.126 ± 0.038 min at 70 C. From the D-values at each temperature, linear regression was conducted for $\log_{10}(D)$ vs temperature and is shown in Figure 1.

The z-value was obtained from the slope of $\log_{10}(D)$ vs temperature plot and was 6.262 C for *Salmonella* and 5.666 C for *L. innocua*. The R^2 of the regression was greater than 0.98. Since no reports were found on D- and z-values of *Salmonella* or *L. innocua* in fully cooked chicken breast meat, it is difficult to compare this study with previous publications. In general, the D- and z-values of *Salmonella* and *L. innocua* from this study were in the same magnitude as those in cooking raw chicken meat products reported by Murphy et al. (2000, 2002).

TABLE 3. Paired comparison for the z-value of *Salmonella* and *Listeria* among different products

Culture	$Formulate_i$	$Formulate_j$	*P*-value for testing z of $Formulate_i$ = z of $Formulate_j$
Salmonella	Plain	PS[1]	0.9049
	Plain	Commercial	0 2475
	PS[1]	Commercial	0.2964
L. innocua	Plain	PS[1]	0.9699
	Plain	Commercial	0.3357
	PS[1]	Commercial	0.3180

[1]Fully cooked chicken breast meat marinated with phosphate and salt prior to cooking.

TABLE 4. D-values of *Salmonella* and *Listeria innocua* in fully cooked chicken breast meat products (N = 90 per temperature)

Temperature (C)	*Salmonella*			*Listeria innocua*		
	D (min)	SD	R^2	D (min)	SD	R^2
55	24.071	1 852	0 902	56.169	4 016	0 896
57 5	9.600	1 556	0 920	20 355	1 031	0 883
60	3.828	0 750	0 907	7.362	0 833	0 923
62.5	1.527	0 299	0 893	2.665	0 424	0.921
65	0.609	0.135	0.890	0.965	0.185	0.871
67.5	0.243	0.027	0.862	0.349	0.028	0 912
70	0.097	0.034	0.876	0 126	0.038	0.888

In the current study, the products tested were fully cooked, had similar compositions, and differed only on source, marinade formulations, and thermal processing methods. Therefore, the addition of food additives such as NaCl (<4%) and polyphosphate (<2.4%) may not affect the thermal inactivation of *Salmonella* or *L. innocua* in fully cooked commercial chicken products during postcook pasteurization.

The results from this study will be useful in validating postcook pasteurization processes for fully cooked chicken breast meat products. No significant difference was observed in thermal inactivation D- and z-values of *Salmonella* or *L. innocua* between the three chicken breast meat products with different sources and formulations. The D- and z-values obtained from this study should be useful in determining the process lethality of *Salmonella* or *L. innocua* during commercial postcook pasteurization of similar products.

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Growth and inactivation of *Salmonella enterica* and *Listeria monocytogenes* in broth and validation in ground pork meat during simulated home storage abusive temperature and home pan-frying

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Introduction

Salmonella enterica and *Listeria monocytogenes* are two of the most important foodborne pathogens; they are known to occur in raw meat, and are associated with foodborne outbreaks (Rose et al., 2002; EFSA and ECDC, 2015). Consuming contaminated raw or undercooked meat is believed to be one of the important vehicles of foodborne infection. The presence of these pathogens in meat can present a serious food safety threat. According to the strong-evidence foodborne outbreaks in Europe, up to 38.5% cases happened at households/domestic kitchens (EFSA and ECDC, 2015). Adequate refrigeration and thorough cooking are two points of attention to ensure microbiological safety of meat toward the end of the food chain.

Ground meat is a potentially hazardous type of fresh meat, it is particularly susceptible to bacterial contamination throughout its mass, and therefore, more likely to contain foodborne pathogens (Lianou and Koutsoumanis, 2009; Schlisselberg et al., 2013). Both retailers and consumers use low storage temperatures to minimize growth of spoilage and pathogenic microorganisms. However, *L. monocytogenes* can survive or even grow at low temperatures; *S. enterica* can grow when the storage temperatures are abused. Predictive models can be used to estimate the growth potential of microorganisms in the food chain. A number of models and software have been developed to predict the effects of temperature, pH or water activity on the growth of pathogens in ground meat (Mbandi and Shelef, 2001; Ingham et al., 2007; Pin et al., 2011;Velugoti et al., 2011). A limitation of these models is that they are based on the collection of data in sterile ground meat. Studies have demonstrated that the effects of competing microbiota on the growth of pathogens cannot be neglected (Zaher and Fujikawa, 2011; Møller et al., 2013). Turning our attention to the growth in ground pork meat, studies concerning the effect of natural microbiota on growth of pathogens have been performed by Ingham et al. (2007) and Møller et al. (2013) where ground pork was inoculated with relatively high levels of pathogens (3–5 log CFU/g). However, the actual initial contamination level of *S. enterica* and*L. monocytogenes* in ground pork is usually low (<10–100 CFU/g) (Ghafir et al., 2005; Thevenot et

al., 2006). Studies on chicken meat and fresh cut salads have indicated that the pathogens' initial densities had effects on their growth in the presence of natural microbiota (Oscar, 2007; Manios et al., 2013), and we expect a similar effect in ground pork meat. The growth of *S. enterica* and *L. monocytogenes* in ground pork meat with realistic levels of natural microbiota and low levels of inoculated pathogens is, as far as the authors are aware of, not available in literature or in the Combase Browser[1].

Home-cooking practice is an important and effective way to eliminate pathogens in meat. So far, thermal treatment remains the principal method of microbial inactivation for consumers at home (Álvarez-Ordóñez et al., 2008). It is recommended that ground pork or beef must be cooked to an internal temperature of 71 or 70°C for 2 min or its equivalent (Advisory Committee on the Microbiological Safety of Food [ACMSF], 1995; FDA, 2011b). However, most of the European consumers check the meat doneness visually, rather than using a thermometer (Bearth et al., 2014). Information used to establish cooking recommendations has largely been derived from *D* values in laboratory experiments (International Commission on Microbiological Specifications of Foods [ICMSF], 2005). Since the late 1990s, a number of studies have evaluated the heat resistance of *S. enterica* and *L. monocytogenes* in buffers or broth (Juneja et al., 2001; Sorqvist, 2003; Miller et al., 2009), and in meat and meat products (Juneja et al., 2001; Murphy et al., 2006; Halder et al., 2010;Vasan et al., 2014), but data collected using actual consumer-based handling and cooking processes are comparatively scarce. Thermal inactivation studies in the laboratory are usually performed at isothermal conditions, yet the cooking processes consumers use at home are generally non-isothermal: burgers are usually thermally treated for several minutes on each side in a frying pan in hot butter before being served for consumption. Furthermore, microorganisms in ground meat are immobilized and constrained to grow as colonies rather than planktonically, which may also have an effect on the observed thermal inactivation profiles. So far, no study has focused on the inactivation of foodborne pathogens, with the latter being previously allowed to grow in ground meat, providing, thus, the rationale for setting up and conducting the present study.

For assessing the food safety it is needed to estimate the growth and survival of pathogens in meat under reasonable foreseen conditions of pathogens' contamination level as well as storage conditions and subsequent thermal treatment prior to consumption. The average temperature of the fridge of Belgian households is 6.7°C and as much 10.8% (n = 3001) was even at temperatures larger than 10°C (De Vriese et al., 2005). Therefore, we conducted a systematic study to assess the behavior of *S. enterica* and *L. monocytogenes* in ground pork meat under 10°C refrigerator storage and subsequent consumer-based pan frying with, as usually practiced in Belgium, visual assessment of doneness. Ground pork with natural microbiota and inoculated with a low initial density (1–10 or 10–100 CFU/g) of *S. enterica* and *L. monocytogenes* was used to mimic naturally contaminated burgers. Meanwhile, for comparativeness, the growth and inactivation of these pathogens were also evaluated in brain heart infusion (BHI) broth. The

study will help to reduce the uncertainties in assessing the food safety threat of *S. enterica* and *L. monocytogenes* in ground pork meat. It will also permit to validate the applicability of the estimations derived from microbial growth and inactivation models often established in broth media and provide quantitative information on the behavior of *S. enterica* and *L. monocytogenes* in ground pork during reasonably foreseen home storage conditions and cooking practices.

Go to:

Material and Methods

Bacterial Strains and Culture Conditions

The following strains of *S. enterica* and *L. monocytogenes* were used for the growth and thermal inactivation test. Of *S. enterica*, three food-isolated strains selected were *Salmonella* Derby LFMFP 872 (pork isolate),*Salmonella* Enteritidis LFMFP 875 (poultry isolate) and *Salmonella* Typhimurium LFMFP 877 (poultry isolate). Three *L. monocytogenes* strains (LFMFP 392, serotype 4b, liver pate isolate; LFMFP 421, serotype 4b, clinical isolate, and LFMFP 491, serotype 1/2b, tuna isolate) were used. All stock cultures were kept at –75°C in Tryptone Soy Broth (TSB, Oxoid, Basingstoke, England), supplemented with 0.6% yeast extract (YE, Oxoid) and 15% glycerol (Prolabo, Heverlee, Belgium). Working stocks were stored refrigerated at 4°C on Tryptone Soy Agar (TSA, Oxoid) slants and were renewed monthly. Working cultures were activated by transferring a loopful from the slants into BHI (Oxoid) and incubated at 37°C for 18 to 24 h. The working cultures were prepared by transferring 0.1 ml of each culture into 10 ml of BHI and incubated at 37°C for 24 h. Immediately before inoculation, a cocktail containing three strains of *S. enterica* or *L. monocytogenes* was prepared individually by mixing approximately equal population of each strain and serially diluted in Peptone Physiological Salt Solution (PPS, containing 1 g/l neutralized bacteriological peptone and 8.5 g/l NaCl).

Growth Studies

Growth Studies in Broth

The growth curves of *S. enterica* and *L. monocytogenes* in broth at 10°C were determined in BHI. One milliliter of each pathogen cocktail dilution was inoculated into a 250-ml blue-cap bottle containing 99 ml of BHI to yield an initial dose of 1-10 (10^{-7} dilution) and 10–100 (10^{-6} dilution) CFU/ml. The broth was equilibrated overnight in the refrigerator to 10°C before inoculation. The incubation period was 24 days for *S. enterica* and 10 days *for L. monocytogenes*. At regular time intervals, aliquots (1 ml) of the culture were taken and serially diluted in PPS followed by plating on duplicated plates. The *S. enterica* and *L. monocytogenes* populations were determined by plating on Xylose Lysine Deoxycholate (XLD,

Oxoid) and Listeria Ottaviani and Agosti (ALOA, Biolife, Milano, Italy), respectively. Bacterial colonies were enumerated after incubation of the plates at 37°C for 24 and 48 h for *S. enterica* and *L. monocytogenes*, respectively.

Growth Studies in Ground Pork Meat

Ground pork meat was purchased at a local store and analyzed for the presence of *S. enterica* and *L. monocytogenes*, and was found to be absent in 25 g of meat samples (see below). The analysis of characteristics of the meat was performed as described by Lahou et al. (2015). It indicated that the ground pork contains about 8.1% fat and 1.5% sodium salt. The measured pH and water activity were 5.6 and 0.98, respectively. The meat was divided into portions (9.9 g) and aseptically transferred into a stomacher bag for growth studies. A diluted culture (0.1 ml) of the cocktail of *S. enterica* or*L. monocytogenes* was inoculated individually. The initial pathogen density aimed for was 1–10 or 10–100 CFU/g, which is similar to the level expected in naturally contaminated meat. The negative control samples were inoculated with 0.1 ml PPS. After the inoculum was added, the bags were hand mixed for 30 s, stomached for 2 min, compressed into a thin, uniform layer, loosely heat sealed, and then stored in a 10°C refrigerator. At selected times of incubation samples were added with 90 ml of PPS and were thoroughly homogenized in a stomacher (Lab Blender 400, Seward Laboratory, London, UK). Each sample was then serially 10-fold diluted with PPS for determination of bacterial density. The enumeration of the total plate count (TPC) in Plate Count Agar (PCA, Oxoid) was derived from ISO 6222 (5 days incubation at 22°C). Presumptive lactic acid bacteria (LAB) count was determined on Man Rogosa Sharp Agar (MRSA, Oxoid) with an overlay according to ISO 15214 (3 days incubation of MRS at 30°C) and the enumeration of coliforms was performed using Violet Red Bile Lactose (VRBL, Oxoid) Agar overlaid with the same medium according to ISO 4832 (24 h incubation of VRBL at 37°C). *S. enterica* and *L. monocytogenes* was, respectively, plated on XLD and ALOA plates. Suspected *S. enterica* colonies were further confirmed using Crystal E/NF ID (BD Benelux N. V, Erembodegem, Belgium).

Thermal Treatments

Thermal Treatment in Broth

Two methods were compared to evaluate whether different test methods used to measure thermal inactivation would influence the results. The schematic diagram of the two methods is shown in **Figure Figure11**. In Method I (**Figure Figure1A1A**), a 0.1 ml portion of the stationary phase culture was added directly into 9.9 ml BHI in test tubes (125 mm × 15 mm), resulting in an initial population of approximately 7.0 log CFU/ml. This method is termed the reference method. Method II is referred to as an alternative method. In method II 1-ml portions of culture were inoculated to 9 ml of BHI along the inner wall of the thin-walled test tube (160 mm × 15 mm) (**Figure Figure1B1B**). In both methods the test tubes were submerged in a water bath

(Memmert, WB 10, Germany) preheated to the target inactivation temperature of 60 ± 0.1°C. The temperature of the broth was monitored in a test tube throughout the duration of the thermal treatment with Testo 177-T4 temperature data logger (Testo AG, Lenzkirch, Germany). After the treatment, all the tubes were transferred to an ice water bath within 30 min before plating on XLD or ALOA plates for survivors.

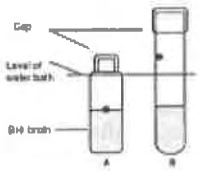

FIGURE 1

Schematic diagram of method I (A, reference method) and method II (B, alternative method) used to assess the heat resistance of pathogen strains in the water bath. The dots are the spots where cultures were injected.

Heat resistance of all the bacterial strains was compared in standard BHI broth (pH 7.3, 0.5% NaCl) and in BHI adjusted to pH 5.6 with lactic acid and NaCl 1.5% (w/w) as the intrinsic conditions in the ground pork meat. The added volume of lactic acid did not significantly affect the volume of the media. The stationary phase cultures of each tested strain were separately diluted with the challenge media (standard or adjusted BHI) to around 6 log CFU/ml. For the heat resistance test, 1-ml portions of the diluted culture were thermal treated as described in method II previously.

Thermal Treatment in Pork Meat Burgers upon Simulated Home Pan-frying

One milliliter strain mixture dilution of *S. enterica* or *L. monocytogenes* was individually inoculated into 99 g portions of ground pork in a stomacher bag for an initial dose of 10–100 CFU/g. The stomacher bags were massaged as described previously. Burgers (8.5 cm by 1.5 cm) were prepared in sterile Petri dish. Individual burgers were placed in stomacher bags, heat sealed, stored at 10°C for 5 days, and subjected to microbial analysis and simulated home pan-frying.

The inoculated pork burgers were baked in a frying pan of TEFAL S.A.S® with a diameter of 24 cm on an electrical heating plate (SCHOTT® instruments, model: SLK2, 1800 W, heated zone diameter of 20 cm). The standardized cooking procedure and time was established based on preliminary tests as to obtain a visual well-done cooked pork burger (Lahou et al., 2015). The pan was preheated at heating state 7 (the highest heating state of the heating element was 9). Then a total of 10 g of butter (Belolive®) was melted for another two minutes at state 7 until skim disappeared. One burger per experiment was put in the pan and fried at heating state 5 for 4.5 min for each side (total cooking time 9 min). The fried burger was lifted out of the pan and cooled down for 10 min on a plate followed by determination of the weight. During the process of pan-frying, geometric center and surface temperatures (both top and bottom surface) of three

additional burgers were monitored and recorded with a data logger (Testo 177-T4). The thermocouples were bent and inserted at ca. 3 mm depth in the burger so that they could measure temperature in a relatively small top/bottom surface layer of the burger. This temperature is henceforward called burger surface temperature. As a side-remark, it should be noted that the surface of a pork meat burger is not a flat and smooth surface and temperature of the (sub) surface of the burger may be very location specific. As soon as the burger was turned, the probes were immediately put back in. To measure the core temperature, a wireless temperature logger (DS1922T iButton, Maxim Integrated Products, Sunnyvale, CA, USA) was placed into the center of the burger. The burger core temperature profile was used to calculated the process lethality (*F*-value) using an Excel spreadsheet[2] based on the formula below

$$F=\int_{t0} 10^{(T-T_{ref})/z} dt$$

where T is the core temperature (°C) at a time t (min) and T_{ref} is a reference temperature (60°C was used in this study). According to a previous study (Murphy et al., 2006), in ground pork the z value is 5.89°C for *Salmonella* and 5.92°C for *L. monocytogenes*.

A representative 10 g sample, a strip of ca. 1 cm wide from the middle of the fried burger, was taken for microbial analysis. Enumeration of *S. enterica* or *L. monocytogenes*, TPC, total coliforms and LAB was performed as described above. For the samples where no surviving *S. enterica* or *L. monocytogenes* were found by enumeration, duplicate 25 g samples were used to test a complete inactivation of pathogens by the enrichment method. The enrichment of *S. enterica* and *L. monocytogenes* was carried out as previously described by Siro et al. (2006). For *S. enterica*, a 25 g sample was blended with 225 ml of Buffered Peptone Water (BPW, Oxoid) and incubated at 37°C for 24 h. From the primary enrichment, 0.1 ml of the aliquot was transferred into 10 ml of Rappaport-Vassiliadis broth (RVS, Oxoid) and incubated at 42°C for a further 24 h before plating out on XLD plates. For *L. monocytogenes*, the primary enrichment was done in Demi-fraser enrichment broth (Oxiod) at 30°C for 24 h. Then a 0.1-ml of the primary enrichment broth was subcultured into the secondary enrichment broth (10 ml of Fraser) and incubated at 37°C for 24 h. Afterward samples were streaked onto ALOA plates.

Data Analysis

Growth and inactivation studies for both pathogens were performed in triplicates. The mean of the duplicated plate counts per repetition was determined and converted to $\log_{10}$ values, and plotted versus time. Growth curves were fitted with "DMFit online[2]" using the Baranyi and Roberts (1994). Cell counts below the detection limit of 5 CFU/g were excluded in the calculation of curves, but indicated as separate data points on x-axis in the same figure. The growth parameters including lag time (λ), maximum growth rate (μ_{max}), and maximum population density (y_{max}) were determined. Inactivation data were analyzed by linear and non-linear models by the software GInaFiT (version 1.6) (Geeraerd et al., 2005). The goodness of fit of the models

was assessed using adjusted regression coefficient (R^2_{adj}). The kinetic parameters from the best fit model were reported. Statistical interpretation of differences among parameters was determined using ANOVA analysis (SPSS statistical software, Inc., Chicago), using 95% confidence limits.

Go to:

Results

Growth of *S. enterica* and *L. monocytogenes* in Broth

Growth curves of a cocktail of three strains of *S. enterica* or *L. monocytogenes* in broth exhibited a classical sigmoidal behavior (not shown). Variation among replications was found to be not significant ($P > 0.05$), and thus the growth data were averaged. At both initial densities, the maximum growth rate of *S. enterica* and *L. monocytogenes* was estimated to be about 0.021 and 0.066 $\log_{10}$ CFU/ml/h, respectively. Due to the longer lag time (ca. 60 vs. 17 h) and lower growth rate, the time needed to reach stationary phase for *S. enterica* was more than double that of *L. monocytogenes*.

Growth of *S. enterica* and *L. monocytogenes* in Ground Pork with a Natural Microbiota

The initial concentration of TPC, LAB, and coliforms in the ground pork were ca. 4.6, 4.4, and 1.5 log CFU/g, respectively, which indicated satisfactory initial microbial quality of the ground pork meat. Growth curves of TPC, coliforms, and LAB with different inoculum levels of *S. enterica* or *L. monocytogenes* at 10°C are presented in **Figure Figure22**. After ca. 4 to 5 days all the indigenous bacteria reached the stationary phase of growth. TPC reached its maximum value of ca. 8.9 log CFU/g, LAB at 8.3, whereas 5.9 log CFU/g for coliforms. The maximum growth rates of the indigenous bacteria were similar to each other ($P > 0.05$) regardless of their initial levels or inoculated pathogens (**Tables Tables11** and **22**).

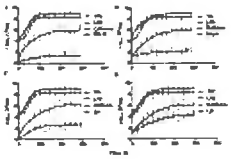

FIGURE 2

Growth of indigenous microbiota and *S. enterica* (SALM) (A,B) or*L. monocytogenes* (LM) (C,D) at low initial densities (A,C ~1 CFU/g; B,D ~10 CFU/g) at 10°C in ground pork meat. Solid lines are regression lines fitted with Baranyi ...

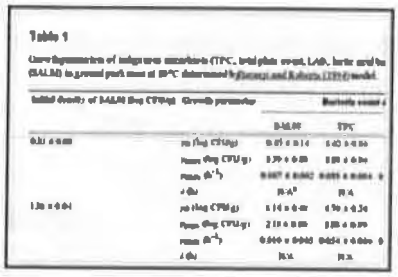

Table 1

Growth parameters of indigenous microbiota (TPC, total plate count; LAB, lactic acid bacteria) and *S. enterica* (SALM) in ground pork meat at 10°C determined by Baranyi and Roberts (1994) model.

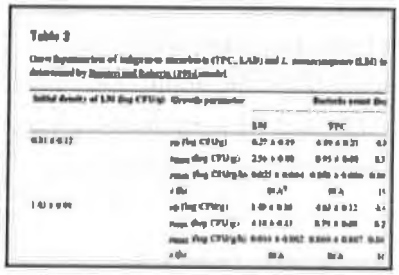

Table 2

Growth parameters of indigenous microbiota (TPC, LAB) and *L. monocytogenes* (LM) in ground pork meat at 10°C determined byBaranyi and Roberts (1994) model.

Salmonella enterica cells were able to multiply at both inoculum levels. However, the population increased by less than one log unit only, even after enforced long time (12 days) storage at this abusive temperature of 10°C. Increase of *S. enterica* starting from ca. 20 CFU/g occurred with limited variation (SD < 0.5 log CFU/g, **Figure Figure2B2B**) compared with the samples starting from a few (ca. 2) CFU/g which ranged from <0.7 (detection limit) to 2.1 log CFU/g (**Figure Figure2A2A**). Under the same enforced abusive storage conditions *L. monocytogenes* grew exponentially (**Figures 2C,D**) up to a maximum value of 2.6 and 4.2 log CFU/g, respectively (**Table Table22**) after 12 days at 10°C. The increase of *L. monocytogenes* starting from ca. 2 and 27 CFU/g was 2.3 and 2.8 log units, respectively. The variation of the observed values of *L. monocytogenes*among replicates was lower than for *S. enterica*.

Thermal Inactivation of *S. enterica* and *L. monocytogenes* in Broth

Survival curves of *S. enterica* and *L. monocytogenes* strains obtained by the reference method are shown in**Figures 3A,B**. The *S. enterica* curves were fitted by the log-linear model. For all regressions, the $R2_{adj}$values were larger than 0.95 (data not shown). Decimal reduction time or *D* values were determined from the maximum inactivation rate (k_{max}, *D* value = $\ln(10)/k_{max}$). *D* values of *S. enterica* strains ranged from 0.20 to 0.24 min (**Table Table33**).

Shoulders were observed on all inactivation curves of *L. monocytogenes* and were fitted to a log linear model with a shoulder (Geeraerd et al., 2000). The fittings yielded R^2_{adj} values from 0.97 to 0.99. The shoulder length (S_l) ranged from 0.52 to 1.13 min. *D* values of *L. monocytogenes* were more than twice higher than those of *S. enterica*. In general a minimum process of 6*D* reductions in the numbers of target microorganisms is recommended for pasteurized foods (International Commission on Microbiological Specifications of Foods [ICMSF], 2005; FDA, 2011a). The t_{6D} values, expressing the time needed to obtain six decimal reductions (Buchanan et al., 1993) of *S. enterica* and *L. monocytogenes* are given in **Table Table33**. Since shoulders were observed on *L. monocytogenes* inactivation curves, t_{6D} of *L. monocytogenes* strains are larger than six times the *D* values.

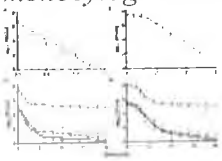

FIGURE 3

Inactivation curves of *S. enterica* LFMFP 872 (A,C) and *L. monocytogenes* LFMFP 392 (B,D) at 60°C with different challenge methods (reference method (∘), model (····); method II (Δ), model (···)) and broth ...

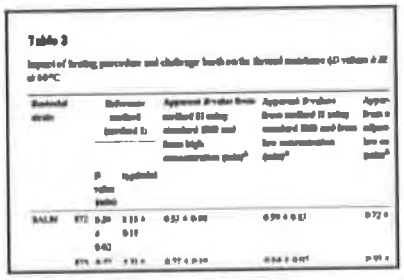

Table 3

Impact of heating procedure and challenge broth on the thermal resistance (*D* values ± *SD*) of SALM and LM heated at 60°C.

The inactivation curves obtained by the method II of thermal treatment (inoculated via the inner wall in the tube instead of immediately in the suspension) showed a biphasic shape. Typical curves are shown in**Figures 3C,D**. Survivor curves showed initially 2 to 3 log reductions, followed by prolonged tailing in which the numbers only slightly decreased further. A zero point was not achieved even after 20-min thermal challenge at 60°C. It deserves attention that the apparent *D* values, which were calculated from the initial log-linear part of the biphasic curves obtained by method II, were 1.5- to 2.9-fold larger than those obtained using the reference method I (**Table Table33**). This is important to be noticed as the exact laboratory procedure to determine *D* values is not always described in detail in scientific literature and this highlights the

fact that small deviations in elaborating the laboratory procedure for *D* values determination may impact the outcome.

When the pathogen cells were thermally treated at an initial concentration of ca. 10^5 CFU/ml, inactivation curves showed the same pattern as the high initial concentration (ca. 10^8 CFU/ml) (**Figures 3C,D**). The apparent *D* values were more or less invariable (**Table Table33**). Apparent *D* values of each strain thermally treated in standard and adjusted BHI are also listed in **Table Table33**. The strains treated in adjusted BHI (pH 5.6, 1.5% NaCl) showed higher apparent *D* values than those in standard BHI, especially for *S. enterica*.

Inactivation of *S. enterica* and *L. monocytogenes* in Pork Meat Burger by Simulated Home Pan-frying

The simulated home pan-frying procedure used in this study resulted in 30.4 ± 1.7% weight loss of the burgers. It was similar as a standard pan-frying procedure applied by Danowska-Oziewicz (2009) where the cooking loss was 28%. The temperature profiles of three burgers during pan-frying and cool-down at ambient temperature on the serving plate are presented in **Figure Figure44**. The temperatures of the burgers bottom rose sharply to the maximum (93.9–100.6°C) before flipping, while the increase on the top was very limited. The bottom temperatures were higher than the core temperatures, and this difference increased with time. After flipping, the (new) bottom temperature increased quickly while the (new) top temperature decreased gradually. During cooling down on the serving plate the bottom temperatures of the burgers immediately started to decrease exponentially, while the core temperature still slightly increased due to heat conduction. The peaks of the core temperatures, which ranged from 69.0 to 71.9°C, were reached at ca. 0.3 min after taking the pork meat burgers out from the pan.

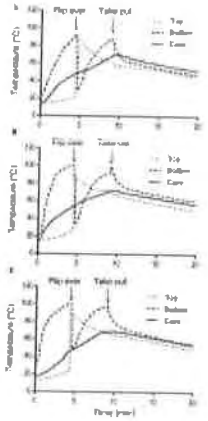

FIGURE 4

Temperature profiles of three replicate pork meat burgers (A,B,C) during simulated home pan-frying.

To evaluate the efficacy of thermal treatment during this simulated home pan-frying on the inactivation of pathogens in the meat, *F* values were calculated in pork burger as the equivalent time needed to reduce *S. enterica* or *L. monocytogenes* at 60°C. *F* values were obtained according to the core temperature profiles of the burgers (**Figure Figure44**). The calculated *F* values for *S. enterica* were 115, 282, and 123 min for three replicates, respectively; and for *L. monocytogenes* 113, 276, and 121 min. All the *F* values were obviously much higher than the expected time needed for 6 log reductions of both pathogens. After the pan-frying procedure pathogens are thus expected to reduce to undetectable levels as in the present study the initial contamination levels of *S. enterica* and *L. monocytogenes* in pork meat burgers (after prior storage for 5 days at 10°C) were ca. 1.95 log CFU/g and 3.10 log CFU/g, respectively (**Table Table44**). As expected no *S. enterica*were recovered from all the samples after enrichment in 25 g of pan-fried pork meat burger. Accordingly, at least a 3.3-log unit reduction of *S. enterica* was obtained. However, the presence of *L. monocytogenes* was detected in three out of six of the 25 g pan-fried pork burger samples, so 2.4- to 4.5-log units reduction was achieved in these three burgers, but no 6-log unit reduction was obtained. As for the indigenous microbiota the number of surviving bacteria was significantly reduced. The mean reductions of TPC and LAB were all over 6 log units. Regarding coliforms, this microbial group was, in all cases below the detection limit of 5 CFU/g.

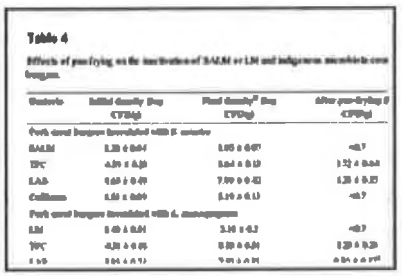

Table 4

Effects of pan frying on the inactivation of SALM or LM and indigenous microbiota count (TPC, LAB) in pork meat burgers.

Go to:

Discussion

In this work, we studied the growth of *S. enterica* and *L. monocytogenes* in artificially contaminated ground pork meat during storage under reasonably foreseen temperature abuse at 10°C. Subsequently, the inactivation of these pathogens – which were allowed to grow for 5 days at 10°C in the pork burger – was determined using a pan-frying procedure routinely practiced in Belgian domestic settings. The growth and inactivation results in the pork meat burgers were compared with those obtained in laboratory media such as BHI broth.

The survival and growth of *S. enterica* and *L. monocytogenes* in ground pork meat was monitored for up to 12 days of storage at 10°C. It is obvious that the meat was spoiled as of day 5: the TPC reached maximum levels. Monitoring of pathogens' behavior was continued to assess whether there was still outgrowth or rather survival or die-off of *S. enterica* and *L. monocytogenes* in presence of competition with these maximum levels of indigenous microbiota and their metabolites. Also this enabled maximum comparison between behavior in the meat versus BHI broth and predictions obtained by the mathematical models. The growth parameters of *S. enterica* and *L. monocytogenes* in BHI were generally in agreement with previous selected reports from Combase database and literature when selecting experimental conditions comparable to those in the present study (culture media of pH 7–7.5, aw 0.99–1.00, incubated at 10°C). The Combase reported growth rates of *S. enterica* in broth at 10°C varied from 0.020 to 0.030 log CFU/ml/h, with an average of 0.028 (4 reported values). As for *L. monocytogenes*, the growth rates ranged from 0.041 to 0.082 log CFU/ml/h with an average of 0.054 (21 reported values). In our study, at both initial densities, the maximum growth rate and y_{max} of *S. enterica* or *L. monocytogenes* was estimated to be similar. Thus results in the present study agreed with previous reports where the growth of pathogens in sterile broth was usually independent of initial density and y_{max} is usually not greatly affected by growth conditions (Buchanan and Klawitter, 1991).

As shown, both *S. enterica* and *L. monocytogenes* have the ability to multiply in ground pork at 10°C in the presence of a substantial numbers of indigenous microbiota. Still, it was observed that the growth of pathogens ceased when the indigenous microbiota reached its maximum population density. This is probably due to microbial competition between pathogens and the indigenous microbiota. This phenomenon has been referred to as the "Jameson effect" (Jameson, 1962). It is noted that for both *S. enterica* and *L. monocytogenes* in the pork meat, y_{max} was dependent on the initial dose; y_{max} was higher at higher initial pathogen contamination level, which is inconsistent with the results obtained in BHI broth. The difference iny_{max} could also be attributed to the Jameson effect by the indigenous microbiota in ground pork meat. A number of studies have been done on the growth of pathogens in sterilized ground meat where no competition occurred. Velugoti et al. (2011) studied the growth of *Salmonella* sp. in sterile ground pork meat. At 10°C, *S. enterica* reached a maximum population of 8.3 log CFU/g with a maximum rate of 0.018 log CFU/g/h, both of which were much higher than those values obtained in the present study. Mbandi and Shelef (2001) investigated the growth of *S. enterica* and *L. monocytogenes* in sterile ground beef at 10°C: numbers of both pathogens increased from 3.5 to approximately 8.0 log CFU/g after 20 days of storage. Indigenous microbiota in raw ground meat are thought to consist of a variety of microorganisms that can inhibit the growth of pathogens. Ingham et al. (2007) studied the growth of pathogens in meat with relatively low levels of indigenous biota (≤3.5 log CFU/g) and relatively high levels of inoculated pathogens (4.6 log CFU/g). An online software for evaluating the safety of meat was

developed based on their study[4]. This online tool predicted for *Salmonella* a growth of 6.6 log units in ground pork after 12-days storage at 10°C. However, we observed only less than one log unit increase of *S. enterica* and ca. 2.5 log units increase of *L. monocytogenes*. Similarly, Oscar (2007) reported that at 10°C, the growth of *S. enterica* from a low initial density in ground chicken with a natural microbiota was also very limited, from 1.1 to 1.8 log MPN or CFU/g.

Thermal inactivation of *Salmonella* and *L. monocytogenes* has been studied extensively resulting in a wide range of *D* values. It is well known that the inactivation dynamics may be influenced by various factors including the bacterial strain of the species, the physiological state of microbial cells, heating and recovery conditions (Smelt and Brul, 2014). Average *D* values of *Salmonella* and *L. monocytogenes* at 60°C as reported in broth or buffers (pH 7–7.5, a_w 0.99–1.00) were listed and compared to the ones estimated in the present study (**Table Table33**). The average published *D* values for *Salmonella* and *L. monocytogenes* were 0.75 and 1.32 min, respectively. Thus, the *D* values obtained in the BHI broth in the present study were within the same order of magnitude.

For almost one century, the food industry assumed that thermal inactivation followed first-order kinetics during the estimation of the outcome of a thermal treatment on the survival of microorganisms. However, there is growing evidence to support that the inactivation of microbial cells does not always follow the traditional first-order kinetics, especially during a mild thermal treatment (Augustin et al., 1998; Valdramidis et al., 2006). In the present study, shoulders were observed on *L. monocytogenes* survival curves. It has been a consensus that *D* values should be used with care when the isothermal survival curves are not really log-linear (Peleg, 2006). However, in many published articles, no inactivation curves are shown, but only *D*values. It is not clear if the original data were indeed log-linear so that the derived *D* values can have a clear meaning. Therefore, it is recommended that the '*D* values', including the ones reported in literature are critically assessed. The t_{xD}, an alternative concept for thermal microbial inactivation, was developed to describe microbial heat resistance (Buchanan et al., 1993). It describes the time *t* required for *x* log units reductions in the microbial population. In this concept, the deviations from the first-order kinetics were taken into account when estimating the effectiveness of a thermal treatment instead of excluding any shoulders and tails. Meanwhile, the use of t_{xD} rather than *D* values when communicating the performance of food inactivation processes has been accepted by many researchers (Heldman and Newsome, 2003; Valdramidis et al., 2005).

As established in the present study, the heat resistance may be affected by the heating method. Various methods of thermal treatment have been applied in evaluating heat resistance of bacteria in a laboratory media, e.g., heating in water baths using capillary tubes, test tubes, glass ampoules completely immersed in the water, and heating using pasteurization, submerged-coil heating apparatuses etc. (Sorqvist, 2003). The test tube method is one of the commonly used due

to the advantage of easy handling. The two thermal treatment methods applied in our study produced different patterns of inactivation curves and *D* values. Similar observations for bacterial cells or mold spores have been reported in previous studies when the test organism was heated in incompletely submerged capped tubes (Schuman et al., 1997; Zimmermann et al., 2013). The cells coating the walls above the level of the water bath were regarded to be responsible for this tailing phenomenon; these cells were not exposed to the intended temperature of inactivation. The pathogens' strains also showed higher heat resistance in broth with pH adjusted to 5.6 and an increased (1.5%) NaCl concentration. The effect of the pH on the heat resistance was similar to that observed previously (Blackburn et al., 1997; Mañas et al., 2003; Arroyo et al., 2009). There was an optimum pH for survival of cells, increasing acidity or alkalinity increased the rate of inactivation. It has been reported that maximum heat resistance of several pathogens is obtained at slightly acidified media (Blackburn et al., 1997). Furthermore, 1.5% NaCl in adjusted BHI had a heat protective effect. Based on the above, it needs to be recognized that the thermal inactivation kinetics of bacterial pathogens can be affected by the test procedures and types of challenge media. It is important to use suitable methodology in assessing the thermal resistance and clearly state the test conditions.

Based on the *6D* values of each three strains obtained in BHI broth in this study, 1.5 and 5.5 min thermal treatment at 60°C are deemed to be sufficient to achieve a 6-log unit reduction for *S. enterica* and *L. monocytogenes*, respectively. However, considering the increased heat resistance of pathogens in a food matrix versus laboratory media (Kenney and Beuchat, 2004), longer time may be needed in meat burgers to get 6 log units reductions for pathogens. O'Bryan et al. (2006) summarized the thermal resistance of *S. enterica* and *L. monocytogenes* in meat and poultry and great variation was shown. At 60°C, *D* values of *S. enterica* varied from 3.83 to 8.5 min and *L. monocytogenes* varied from 0.31 to 16.7 min. Even when the highest *D* values were used for the worst-case scenario considered, the pan-frying process should be sufficient to result in a 6 log reduction of both pathogens based on the calculated *F* values. However, the presence of *L. monocytogenes* in 25 g was detected in three out six of the pan-fried pork meat burgers samples. This result may be explained by several facts. Firstly, the pathogens in the pork burgers in this study were inoculated at a low level (ca. 10^2 CFU/g) and grew at 10°C for 5 days on the meat particles. It has already been reported that food type and composition (e.g., percentage fat) may have a protective effect on thermal inactivation. For example, Murphy et al. (2000) observed increased *D* values for a mixture of six*Salmonella* serotypes and *Listeria innocua* M1 when comparing the inactivation in chicken breast meat patties and a peptone agar aqueous solution. Secondly, the bacteria were constrained to grow as colonies. InVelliou et al. (2013) it was shown how *E. coli* K12 and *Salmonella* Typhimurium, grown as colonies of various sizes in a matrix gelled with xanthan gum, display a higher thermal resistance when compared with planktonic cells. The surviving *L. monocytogenes* after pan-frying may be a potential risk for food safety. Nevertheless, it is supposed that a concentration of *L.*

monocytogenes not exceeding 100 CFU/g of food at the time of consumption poses limited risk to the consumers (Nørrung, 2000).

Based on the growth and inactivation results in ground pork meat as obtained in the present study, it was established that *L. monocytogenes* grow faster and reaches a higher population density, and there were survivors after a simulated home-frying procedure. As such, it can be inferred that a thermal process that ensures destruction of *L. monocytogenes* in ground pork will also provide an adequate reduction of natural microbiota and other less heat resistant pathogens such as *Salmonella* possibly present in the pork meat burger. This coincides with previous recommendations that *L. monocytogenes* can be considered as the target organism for thermal inactivation (Rocourt et al., 2000; International Life Sciences Institute [ILSI], 2012).

Go to:

Conclusion

Results of this study in particular demonstrated that growth and thermal inactivation data based on laboratory experiments executed in broths show a clear difference with that of what can be expected in actual food. In the present study, both an overestimation of the extent of growth and an overestimation of the extent of inactivation was noticed. The former overestimation leads to a fail-safe situation, however, the latter overestimation is a fail-dangerous outcome. When applying outcomes from models based on laboratory media and condition to foods it is thus important to validate these models carefully and take into account differences that might occur due to other composition, texture and physico-chemical characteristics of the food matrix and indigenous competing microbiota, described as different types of *errors* in Pin et al. (1999)and Miconnet et al. (2005). In the present study the *intermediate error* includes the competition with the natural microbiota occurring at realistic levels of pathogen contamination. The *overall error*, related with the difference between naturally occurring and artificially contaminating pathogens, remains to be investigated for ground pork meat naturally contaminated with *S. enterica* or *L. monocytogenes*.

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Conflict of Interest Statement

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Acknowledgments

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Go to:

Footnotes

[1] http://www.combase.cc

[2] http://www.namif.org/content/process-lethality-spreadsheet

[3] https://browser.combase.cc/DMFit.aspx

[4] http://www.meathaccp.wisc.edu/therm

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Inactivation of Shiga Toxin–Producing O157:H7 and Non-O157:H7 Shiga Toxin–Producing *Escherichia coli* in Brine-Injected, Gas-Grilled Steaks†‡

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ABSTRACT

We quantified translocation of *Escherichia coli* O157:H7 (ECOH) and non-O157:H7 verocytotoxigenic *E. coli* (STEC) into beef subprimals after brine injection and subsequently monitored their viability after cooking steaks cut therefrom. Beef subprimals were inoculated on the lean side with ca. 6.0 log CFU/g of a five-strain cocktail of rifampin-resistant ECOH or kanamycin-resistant STEC, and then passed once through an automatic brine-injector tenderizer, with the lean side facing upward. Brine solutions (9.9% ± 0.3% over fresh weight) consisted of 3.3% (wt/vol) of sodium tripolyphosphate and 3.3% (wt/vol) of sodium chloride, prepared both with (Lac^+, pH = 6.76) and without (Lac^-, pH = 8.02) a 25% (vol/vol) solution of a 60% potassium lactate–sodium diacetate syrup. For all samples injected with Lac^- or Lac^+ brine, levels of ECOH or STEC recovered from the topmost 1 cm (i.e., segment 1) of a core sample obtained from tenderized subprimals ranged from ca. 4.7 to 6.3 log CFU/g; however, it was possible to recover ECOH or STEC from all six segments of all cores tested. Next, brine-injected steaks from tenderized subprimals were cooked on a commercial open-flame gas grill to internal endpoint temperatures of either 37.8°C (100°F), 48.8°C (120°F), 60°C (140°F), or 71.1°C (160°F). Regardless of brine formulation or temperature, cooking achieved reductions (expressed as log CFU per gram) of 0.3 to 4.1 of ECOH and 0.5 to 3.6 of STEC. However, fortuitous survivors were recovered even at 71.1°C (160°F) for ECOH and for STEC. Thus, ECOH and STEC behaved similarly, relative to translocation and thermal destruction: Tenderization via brine injection transferred both pathogens throughout subprimals and cooking highly contaminated, brine-injected steaks on a commercial gas grill at 71.1°C (160°F) did not kill all cells due, primarily, to nonuniform heating (i.e., cold spots) within the meat.

Over the past 30 years undercooked ground beef has quite arguably been the food vehicle most commonly attributable to illness from verocytotoxigenic *Escherichia coli*; however, since the 1990s, among meat products, mechanically and/or chemically tenderized beef (i.e., nonintact beef) has also been more commonly associated with human illness *(2, 3, 8, 9, 11, 20, 31, 40, 42)*. Illnesses attributed to contamination of foods, especially meat, with ECOH are well documented *(27, 33)*. In contrast, of some 14 outbreaks attributed to non-O157:H7 verocytotoxigenic *E. coli* (STEC) since 1990, only 5 were associated with a food vehicle, and none involved beef *(27)*. That being said, it is noteworthy that in August 2010, a Pennsylvania slaughtering and processing facility recalled some 8,500 lb (3,855.5 kg) of ground beef because of possible contamination with serotype O26 STEC *(26)* and its association with a cluster of illnesses in Maine and New York, thus making this the first reported outbreak attributed to a non-O157 serotype of *E. coli* in beef.

A wealth of general information has been published on diarrheagenic *E. coli (4, 29, 33)*, and considerable information exits for characterization and control of ECOH in foods *(5)*, including in tenderized–enhanced beef *(2, 3, 38)*, but there have been far fewer such studies published for STEC *(6, 7, 27)*. As is true for ECOH, any cells of STEC that might be present on the surface of whole-muscle meats could potentially be transferred into deeper tissue by tenderization. To date, a few studies have addressed and/or quantified internalization of ECOH, but not STEC, from the surface into the interior of beef subprimals after blade tenderization or chemical injection and/or monitored their subsequent viability after storage *(12, 25, 39, 45)*. Several investigators have also quantified thermal destruction of

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ECOH, but not STEC, in ground beef *(4, 17, 18, 28, 34)*, and fewer studies have been published on thermal inactivation of ECOH in mechanically or chemically tenderized beef *(13, 22, 32, 37, 39, 45)*. However, there have been relatively few, if any, publications on the comparative translocation of ECOH and STEC into blade- or chemically tenderized steaks and/or their fates after proper cooking.

Careful scrutiny of the available literature reveals that among the handful of illness-related recalls linked to nonintact beef, the incriminated products were most often linked with marinated or brine-injected products *(1, 31)*. Considering that about 18% of beef products sold at retail are mechanically tenderized–enhanced *(2)*, and that such products might be perceived by some individuals as being more like steaks (i.e., "intact") than like ground beef (i.e., "nonintact") and thus may not be properly cooked, there could be a potential threat to public health from undercooked tenderized–enhanced beef, especially since both Schmidt et al. *(36)* and Cox et al. *(10)* reported that between 40 and 58% of consumers ordered their steaks medium rare (60 to 62.8°C) to rare (54.4 to 57.2°C). Thus, a greater understanding of how beef is processed, that being tenderized versus injected versus marinated versus tumbled, as well as how it should be cooked, will lead to a more focused, comprehensive, and meaningful comparative risk assessment of intact and nonintact beef. Sufficient data have not been published, however, to conclusively state whether there is a greater risk from ECOH compared with STEC in nonintact beef products, and/or whether the method used for enhancement, namely injection versus mechanical tenderization, appreciably affects the safety of nonintact beef. Thus, the objective of this research was to comparatively and comprehensively fill data voids related to the translocation of ECOH and STEC into beef subprimals after enhancement via chemical injection and to quantify the subsequent lethality of Shiga toxin–producing cells of *E. coli* within steaks prepared from injected–inoculated subprimals after cooking on a commercial open-flame gas grill.

MATERIALS AND METHODS

Bacterial strains. The five rifampin-resistant (100 μg/ml; Sigma Chemical Co., St. Louis, MO) strains of ECOH (USDA-FSIS 011-82, ATCC 43888, ATCC 43889, ATCC 43890, and USDA-FSIS 45756) and the five kanamycin-resistant (100 μg/ml; Sigma Chemical Co.) strains of STEC (B395 [serotype O111:H7], CDC 96-3285 [serotype O45], CDC 90-3128 [serotype O103:H2], CDC 97-3068 [serotype O121], and 83-75 [serotype O145:HNM]) used in this study were confirmed, cultured, and maintained as described previously *(22, 25)*. Of note, the kanamycin-resistant STEC strains were generated specifically for the purposes of the present study, whereas the rifampin-resistant ECOH strains were generated specifically for/in our previous study *(22)*.

Inoculation and tenderization of subprimals. Vacuum-packaged top butt beef subprimals (U.S. Department of Agriculture Institutional Meat Purchase Specifications no. 184; ca. 7 to 9 kg [15 to 20 lb] each) were obtained from a local wholesale distributor and stored at 4°C for up 7 days. Each subprimal was inoculated essentially as described previously *(22, 25)*. In brief, each subprimal was inoculated by pipetting 10 ml of either the ECOH or STEC bacterial suspensions over the lean-side surface of the subprimal to a target concentration of ca. 6.0 log CFU/g. The opening of each bag was then sealed with tape, and the inoculated subprimals were stored with the inoculated surface facing down for at least 30 min at 4°C to allow the weight of the subprimal to distribute the inoculum over the surface and to promote attachment of the cells to the meat. Next, one set of subprimals was passed once through an automatic brine injector–tenderizer (Koch/Gunther Injectamatic PI-21, Koch Equipment, Kansas City, MO), with the lean side facing upward. Another set of inoculated subprimals not chemically injected served as positive controls. Brine solutions were formulated as follows: (i) 3.3% (wt/vol) of sodium tripolyphosphate (Brifisol STP New, B.K. Giulini Corp., Simi Valley, CA) and 3.3% (wt/vol) of sodium chloride (Culinox 999 food-grade salt, Morton International, Inc., Chicago, IL) (Lac^-), or (ii) 3.3% of sodium tripolyphosphate (Brifisol STP New), 3.3% (wt/vol) of sodium chloride (Culinox 999), and 25% (vol/vol) of a 60% solution consisting of 56% potassium lactate and 4% sodium diacetate on a dry-solids basis (wt/wt; UltraLac KL-564, Hawkins, Inc., Minneapolis, MN) (Lac^+). After injection to a target level of ca. 10% over total weight, up to six core samples were obtained from each of the subprimals and cut into five or six consecutive segments, starting from the inoculated surface: Segments 1 to 4 comprised the top 4 cm, and segments 5 and 6 comprised the deepest 4 to 8 cm (Fig. 1A and 1B). Two trials were conducted for each pathogen cocktail, with a single trial consisting of two tenderized subprimals and two nontenderized subprimals (positive controls). For some experiments, tenderized subprimals were vacuum sealed and held at 4°C for up to 15 days to determine the effect of brine and refrigerated storage on the fate of ECOH and STEC. For the translocation matrix, 1 inoculation level × 2 brine formulations × 6 core samples per formulation × 2 trials per formulation × 2 pathogen types × 2 sampling days were tested, for a sum of 96 core samples tested.

Cooking of chemically tenderized steaks. Vacuum-packaged top butt beef subprimals were inoculated (ca. 6.0 log CFU/g) with either ECOH or STEC and chemically injected as described above. Steaks were cut from each inoculated, tenderized beef subprimal to a thickness of ca. 2.54 cm (1 in.) and stored for 0 or 15 days at 4°C. The thickness of the steak was selected based on our related publication *(25)*, wherein we reported that the thickness of steaks (2.54 versus 3.18 cm) did not significantly affect the extent of thermal inactivation of ECOH or STEC in blade-tenderized beef, and also because most people prefer steaks of medium thickness, that being 2.54 cm. Next, chemically injected steaks were cooked on a commercial open-flame gas grill (model XXE-4, Bakers Pride, New Rochelle, NY) to instantaneous internal endpoint temperatures of either 37.8°C (100°F), 48.8°C (120°F), 60°C (140°F), or 71.1°C (160°F). Beefsteaks were flipped at the approximate midpoint between the initial and target endpoint temperature. Two calibrated, stainless steel thermocouple probes (type T, model HQTQIN-116-18, Omega Engineering, Inc., Stamford, CT) were inserted into the approximate geometric center of each steak and used to measure the internal temperature of the beefsteaks during cooking; two additional type T thermocouples were used to monitor the temperature of the surface of the grill and the surrounding air, respectively. Steaks were removed from the grill when both thermocouples within a steak reached the target end temperature. The temperature of the steaks, the surface of the grill, and the ambient air ca. 30 cm above the grill grates were continuously monitored with an eight-channel thermocouple data logger (model OM-CP-OCTTEMP, Omega

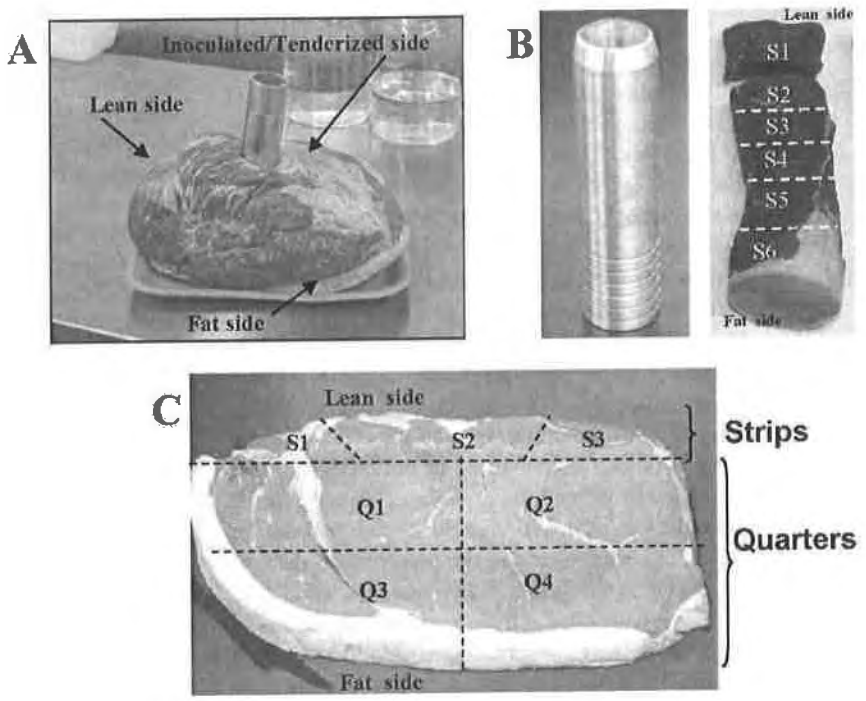

FIGURE 1. *(A) Coring of a beef subprimal. (B) Core apparatus and segmentation of a core sample into six consecutive segments. (C) Segmentation of a brine-injected steak into strips and quarters.*

Engineering, Inc.) at 5-s intervals. Inoculated subprimals that were not injected or cooked served as positive controls. To quantify thermal destruction, as shown in Figure 1C, both cooked and uncooked steaks were portioned into three strips (S1, S2, and S3), each about 1 to 2 cm in depth, and the remaining portion of the steak was cut into four approximately equal quarters (Q1, Q2, Q3, and Q4). Upon removal of a steak from the grill, a calibrated, handheld digital thermometer (model AccuTuff 340, Atkins Technical, Inc., Gainesville, FL) was used to obtain up to eight additional temperature readings from the strips, quarters, and geometric center of each steak. More specifically, when both thermocouples within a steak achieved the desired target temperature, the steak was removed from the grill and placed on a polystyrene foam packaging tray (Koch Supplies, Kansas City, MO), and temperature readings were taken from lean or fat portions of each strip and quarter, as well as from the approximate geometric center, of each steak. Three steaks were individually cooked at each target temperature, and three steaks were not cooked (positive controls). Each of the two trials consisted of 1 inoculation level × 2 brine formulations × 4 cooking temperatures × 3 steaks per temperature × 2 trials per formulation × 2 pathogen types × 2 sampling days, for a total of 192 steaks cooked.

Microbiological analyses. To quantify translocation, each of the five or six segments cut from core samples obtained from tenderized subprimals was weighed separately, diluted in 0.1% peptone water (Difco, BD, Sparks, MD), and macerated for 30 s by using a blender, as described previously *(25)*. The slurry was serially diluted in 0.1% peptone water and surface plated onto sorbitol MacConkey agar (Difco, BD) plates plus rifampin (100 μg/ml [SMACR]; Sigma Chemical Co.) or sorbitol MacConkey agar (Difco, BD) plates plus kanamycin (100 μg/ml [SMACK]; Sigma Chemical Co.) for ECOH and STEC, respectively, as described elsewhere *(22, 25)*. Plates were incubated at 37°C for 24 h, and surviving cells were enumerated. When negative for the pathogen by direct plating, samples were enriched as described before *(22, 25)*. The strips and quarters were weighed separately, macerated in a blender, and subsequently plated, with and without prior dilution in sterile 0.1% peptone water, onto SMACR and SMACK for ECOH and STEC, respectively, essentially as described previously *(22)*. Plates were incubated at 37°C for 24 h. When negative for the pathogen by direct plating, samples were enriched as done before *(25)*.

Statistical analyses. For phase I of the study, as performed previously *(22, 25)*, transfer of ECOH and STEC cells into the deeper tissues of subprimals via chemical tenderization was expressed (in percent) as the number of cells (CFU per gram) recovered separately from each of the five or six segments obtained from chemically tenderized subprimal cores, divided separately by the number of cells (CFU per gram) recovered from segment 1 of the cores obtained from the nontenderized, positive-control subprimals. The means and standard deviations for the levels of the pathogen recovered from each of the five or six segments and the cumulative totals recovered from core samples were calculated with the statistical function option provided with Excel 2003 software (Microsoft Corp., Redmond, WA). Analysis of variance (ANOVA) was used to determine the effects and interactions of the factors on the log translocation values. Differences in translocation observed for each brine formulation, storage day, sample type, and/or combinations thereof were considered significant by using the least significant difference (LSD) technique at a significance level of $P \leq 0.05$. For phase II of this study, the SAS system (version 9.2, SAS Institute Inc., Cary, NC) was used to determine statistically significant differences among pathogen viability during storage of subprimals or steaks, cooking temperatures, and sample types (i.e., strips versus quarters). Means and standard deviations in the cooking experiments were calculated from individual sets of data for each of the two separate trials at each of the four temperatures tested by using triplicate samples at each time interval. ANOVA was used to determine the effects and interactions of the factors on the log reduction values. Differences in lethality observed for each temperature, sample type, and/or combinations thereof were considered significant, using the LSD technique, with $P \leq 0.05$.

RESULTS

Translocation and distribution of ECOH and STEC in beef subprimals after tenderization by chemical injection. The brine formulations tested contained salt and phosphate, both with (Lac^+ = pH 6.76 ± 0.07) and without (Lac^- = pH 8.02 ± 0.25) lactate and diacetate. Brine was delivered at 9.92% ± 0.33% over the fresh, green weight of subprimals. The results validated that tenderization by chemical injection transfers cells of *E. coli* throughout the interior of beef subprimals, with the majority of the cells of ECOH (3.0 to 93.3%) and STEC (25.5 to 82.2%) remaining in the topmost 1 cm (Table 1). These results are in agreement with our prior work on blade tenderization *(23, 24)*, wherein we also reported that the majority of cells of ECOH remained in the topmost 1 cm after tenderization. In general, there were no discernible ($P \geq 0.05$) differences in pathogen viability or in translocation of ECOH or STEC cells related to the presence or absence of lactate-diacetate in the brine, either within a couple of hours after injection or after refrigerated storage for up to 15 days. Although, there was no significant ($P \geq 0.05$) effect of refrigerated storage on pathogen viability in chemically injected steaks, there were generally lower numbers of both ECOH and STEC remaining after 15 days of refrigerated storage compared with starting levels.

Regardless of brine formulation or storage time, in general, there were no significant ($P \geq 0.05$) differences in the levels of ECOH or STEC recovered from segment 1 of the tenderized subprimals compared with levels of these pathogens recovered from segment 1 of the core samples obtained from nontenderized, positive-control subprimals. Levels of ECOH or STEC (Table 1) recovered from segment 1 ranged from about 4.7 to 6.3 and 5.5 to 6.2 CFU/g, respectively. For subprimals injected with Lac^+ or Lac^- brine, the percentages of cells of ECOH or STEC in segment 2 were ca. 5.6- to 23.2-fold or 7.3- to 15.3-fold lower, respectively, than the percentages of cells recovered from segment 1. A significant ($P \leq 0.05$) linear decrease in pathogen levels was observed from segments 2 through 6, but it was possible to recover cells of ECOH and STEC from all six segments of all cores tested. Total levels of ECOH and STEC transferred into all six segments ranged from 4.1 to >100% and 30.6 to 99.6%, respectively. Levels of ECOH or STEC recovered from all six segments of all cores tested ranged from about 5.1 to 6.4 and 5.6 to 6.2 CFU/g, respectively. No appreciable difference between ECOH and STEC in overall translocation was observed, but lesser levels of ECOH and STEC were internalized into the deeper interior tissues of the meat (segments 2 through 6), compared with the surface (segment 1). Experiments are in progress to evaluate additional brine formulations for potential effects on ECOH and STEC during subsequent storage and/or cooking of nonintact beef.

Thermal inactivation of ECOH and STEC in chemically tenderized beefsteaks after cooking on a gas grill. The average come-up times required to reach target internal temperatures of 37.8, 48.9, 60.0, and 71.1°C in brine-injected steaks from tenderized subprimals were ca. 4.7 ± 0.7, 6.3 ± 0.9, 11.0 ± 1.20, and 17.4 ± 2.5 min, respectively. Likewise, the average grill and air temperatures (total of 14,108 readings) were ca. 193.1 ± 18.8°C and 98.1 ± 12.2°C, respectively. Regardless of brine formulation or storage time, as expected, the level of inactivation for ECOH and STEC increased significantly ($P \leq 0.05$) with increasing cooking temperatures between 37.8 and 71.1°C. In addition, regardless of brine formulation, storage time, or cooking temperatures, there were no statistical ($P \geq 0.05$) differences in lethality between ECOH and STEC. In general, for a given formulation and given storage time, regardless of the cooking temperature, no statistical ($P \geq 0.05$) differences were observed among the three strips or among the four quarters of steaks with respect to the extent of thermal inactivation of ECOH or STEC (data not shown). For a given cooking temperature and storage time, with the exception of strips (topmost 1 cm; S1 plus S2 plus S3) from steaks cooked on day 0 to a target internal temperature of 71.1°C, brine formulation did not ($P \geq 0.05$) appreciably affect lethality of ECOH for strips (S1 plus S2 plus S3), or for quarters (Q1 plus Q2 plus Q3 plus Q4), or for total steaks (all strips plus all quarters) (Table 2). Similarly, for a given cooking temperature and storage time or formulation, with the exception of quarters from steaks injected with Lac^+ brine that were stored at 4°C for 15 days and cooked at 60.0°C, no statistical differences ($P \geq 0.05$) in the extent of thermal inactivation of STEC were observed for strips (S1 plus S2 plus S3), for quarters (Q1 plus Q2 plus Q3 plus Q4), or for the summation of both strips and quarters for steaks injected with Lac^+ or Lac^- brine that were subsequently stored refrigerated for 2 weeks and then cooked (Table 3). In addition, for a given cooking temperature and formulation, although there were generally lower numbers of ECOH (Table 2) and STEC (Table 3) remaining after 15 days of refrigerated storage compared with starting levels, no significant ($P \geq 0.05$) effect of storage on lethality of ECOH and STEC was observed for strips (S1 plus S2 plus S3), for quarters (Q1 plus Q2 plus Q3 plus Q4), or for total steaks (all strips plus all quarters) that were stored for up to 15 days at 4°C.

Storage of steaks injected with Lac^+ and Lac^- brine for 15 days at 4°C reduced the levels of ECOH by 0.7 and 1.1 log CFU/g, respectively, whereas the levels of STEC increased slightly by 0.1 and 0.3 log CFU/g. In addition, regardless of storage time, brine formulation, or cooking temperatures, average total reductions ranged from 0.3 to 4.1 log CFU/g for ECOH and from 0.5 to 3.6 log CFU/g for STEC. Although appreciably more cells of ECOH and STEC were recovered from steaks cooked to lower target internal temperatures (37.8 or 48.9°C) compared with those that were cooked to higher target internal temperatures (60.0 or 71.1°C), it was possible to recover cells of ECOH and STEC either by direct plating or by enrichment at all temperatures tested (Tables 4 and 5). It was possible to recover fortuitous survivors from chemically injected steaks after cooking, most likely because of the existence of cold spots (nonhomogeneous heating) within strips or quarters of some steaks. Evidence in support of this contention was

TABLE 1. *Recovery of ECOH and STEC (ca. 6.0 log CFU/g) from segmented core samples from chemically injected subprimals*

		ECOH				STEC			
		Day 0		Day 15		Day 0		Day 15	
Brine formulation	Segment no.	Log CFU/g recovered	% transfer[a]	Log CFU/g recovered	% transfer	Log CFU/g recovered	% transfer	Log CFU/g recovered	% transfer
Lac^-	Control[b]	6.51 ± 0.37 A[c]		6.28 ± 2.12 A		6.31 ± 0.34 A		5.78 ± 0.41 A	
	1	5.78 ± 0.41 A	18.79	4.70 ± 1.04 B	3.04	6.19 ± 0.38 A	76.87	5.70 ± 0.47 A	82.20
	2	4.42 ± 0.37 B	0.81	4.01 ± 1.37 BC	0.54	5.02 ± 0.60 B	5.13	4.81 ± 0.80 B	10.66
	3	3.81 ± 0.46 BC	0.20	3.42 ± 1.03 CD	0.14	4.09 ± 0.53 BC	0.62	4.04 ± 0.59 BC	1.79
	4	3.34 ± 0.53 C	0.07	2.87 ± 0.37 D	0.04	3.33 ± 0.65 C	0.11	3.57 ± 0.61 C	0.61
	5	4.84 ± 1.19 B	2.16	2.97 ± 0.77 D	0.05	4.64 ± 0.94 C	2.14	3.59 ± 0.57 C	0.64
	6	4.30 ± 0.94 B	0.62	3.71 ± 1.28 CD	0.27	4.11 ± 0.64 BC	0.64	4.37 ± 0.68 BC	3.70
Total[d]		5.86	22.64	5.08	4.08	6.24	85.51	5.78	99.62
Lac^+	Control	6.58 ± 0.31 A		5.98 ± 0.77 A		6.32 ± 0.33 A		6.11 ± 1.33 A	
	1	6.32 ± 0.81 AB	54.55	5.92 ± 0.38 A	93.25	5.74 ± 0.41 A	26.39	5.52 ± 0.77 A	25.53
	2	5.53 ± 1.29 B	8.85	4.89 ± 0.74 B	8.10	4.55 ± 1.70 B	1.72	4.65 ± 1.06 B	3.47
	3	4.39 ± 0.97 CD	0.64	4.37 ± 1.14 BC	2.48	4.17 ± 1.39 BC	0.72	3.76 ± 1.12 BC	0.44
	4	3.77 ± 0.55 CD	0.16	4.10 ± 0.92 CD	1.33	3.59 ± 0.52 C	0.19	3.08 ± 0.55 C	0.09
	5	3.61 ± 0.75 D	0.11	3.53 ± 1.12 D	0.36	3.74 ± 0.64 C	0.26	3.51 ± 0.64 C	0.25
	6	4.42 ± 0.71 C	0.69	4.38 ± 0.72 BC	2.54	4.42 ± 0.88 D	1.28	4.16 ± 0.81 B	1.13
Total		6.40	64.98	6.01	108.06[e]	5.80	30.56	5.60	30.91

[a] Percent transfer was calculated as (CFU per gram of tenderized subprimal core segment divided by CFU per gram of segment 1 of nontenderized subprimal core) × 100.

[b] Control samples are segment 1 of nontenderized subprimal cores.

[c] For a given formulation and storage day, means with different letters within columns are significantly ($P \leq 0.05$) different by the LSD test.

[d] Total level of ECOH or STEC (log CFU per gram or percent) transferred into all six segments of a core sample.

[e] Total percent exceeded 100% because of sampling variability of control (nontenderized) treatment.

TABLE 2. *Levels of ECOH recovered from nonintact steaks inoculated with ca. 6.0 log CFU/g before and after cooking*

Cooking temp (°C)	Storage (days)	ECOH level (log CFU/g ± SD)					
		Strips (S1 plus S2 plus S3)		Quarters (Q1 plus Q2 plus Q3 plus Q4)		Total steak (all strips plus all quarters)[a]	
		Lac$^-$	Lac$^+$	Lac$^-$	Lac$^+$	Lac$^-$	Lac$^+$
Uncooked	0	6.36 ± 0.24 A[b]	6.25 ± 0.26 A	5.24 ± 0.01 A	5.25 ± 0.10 A	6.40 ± 0.22 A	6.30 ± 0.24 A
	15	5.25 ± 0.14 A	5.46 ± 0.41 A	4.26 ± 0.02 A	4.75 ± 0.46 A	5.30 ± 0.13 A	5.60 ± 0.24 A
37.8	0	5.11 ± 0.04 AB	5.24 ± 0.20 AB	4.37 ± 0.36 AB	4.45 ± 0.71 AB	5.19 ± 0.03 AB	5.32 ± 0.28 AB
	15	4.92 ± 0.38 A	4.97 ± 0.03 A	3.88 ± 0.22 AB	4.31 ± 0.28 AB	4.96 ± 0.36 A	5.06 ± 0.03 A
48.9	0	4.89 ± 0.23 B	4.30 ± 0.56 BC	3.85 ± 0.74 BC	3.79 ± 0.16 B	4.94 ± 0.28 B	4.44 ± 0.46 BC
	15	4.14 ± 1.81 AB	4.29 ± 0.06 AB	3.06 ± 1.72 ABC	3.52 ± 0.13 AB	4.17 ± 1.80 AB	4.36 ± 0.07 AB
60.0	0	4.24 ± 0.40 B	4.19 ± 0.27 BC	2.76 ± 1.03 CD	3.69 ± 0.48 B	4.26 ± 0.42 B	4.32 ± 0.32 BC
	15	2.91 ± 1.23 BC	3.06 ± 1.61 BC	2.84 ± 0.63 BC	3.15 ± 0.11 B	3.55 ± 0.35 BC	3.67 ± 0.81 BC
71.1	0	1.47 ± 0.07 C	3.32 ± 0.29 C	2.09 ± 0.78 D	1.93 ± 0.48 B	2.25 ± 0.59 C	3.34 ± 0.30 C
	15	2.66 ± 1.12 C	2.48 ± 1.42 C	2.07 ± 0.87 C	1.64 ± 0.37 B	2.77 ± 1.07 C	2.61 ± 1.25 C

[a] ECOH levels reported are the summation of total CFU from all strips plus all quarters and represents the results from two trials and 42 pieces of meat.

[b] For a given formulation and storage time, temperature means with different letters within a column are significantly ($P \leq 0.05$) different by the LSD test.

obtained by taking up to eight independent temperature readings from each steak immediately after it was removed from the grill (Table 6). The results revealed that, although on average the target endpoint temperatures were achieved or exceeded, the range in temperature for a given target endpoint temperature varied considerably. Of note, for 71.1°C (160°F), the recommended minimum internal instantaneous cooking temperature *(41, 43)*, the temperatures within steaks, that being for individual strips and/or quarters, ranged from 48.3 to 102.2°C (119 to 216°F).

DISCUSSION

Historically, strains of O157:H7 are the most commonly recognized serotype of *E. coli* associated with foodborne illness. In recent years, however, non-O157 Shiga toxin–producing strains have also been linked to outbreaks and cases worldwide *(7, 27)*. Our group and other investigators validated that mechanical tenderization of beef forces cells of Shiga toxin–producing *E. coli* into the deeper tissue of the meat *(12, 15, 16, 25)*. Of particular note, colleagues at Kansas State University (Manhattan) reported that 3 to 4% of surface-inoculated ECOH were transferred into the approximate geometric center of beef subprimals by blade tenderization *(32, 39)*. Other investigators also confirmed that tenderization transfers cells into the interior of meat, but with decreasing levels correlated with the depth to which the blade penetrates the meat *(38)*. In addition, Gill and colleagues *(14)* subsequently reported that injection in combination with mechanical tenderization increased contamination of beef primal cuts with *Listeria innocua* by 1,000-fold. The results herein for chemical injection are in

TABLE 3. *Levels of STEC recovered from nonintact steaks inoculated with ca. 6.0 log CFU/g before and after cooking*

Cooking temp (°C)	Storage (days)	STEC level (log CFU/g ± SD)					
		Strips (S1 plus S2 plus S3)		Quarters (Q1 plus Q2 plus Q3 plus Q4)		Total steak (all strips plus all quarters)[a]	
		Lac$^-$	Lac$^+$	Lac$^-$	Lac$^+$	Lac$^-$	Lac$^+$
Uncooked	0	5.71 ± 0.18 A[b]	5.94 ± 0.19 A	4.70 ± 0.34 A	4.97 ± 0.22 A	5.77 ± 0.19 A	5.99 ± 0.15 A
	15	6.02 ± 0.09 A	6.04 ± 0.14 A	4.86 ± 0.43 A	5.01 ± 0.10 A	6.06 ± 0.12 A	6.09 ± 0.12 A
37.8	0	4.95 ± 0.28 AB	5.43 ± 0.14 AB	3.83 ± 0.86 AB	4.37 ± 0.27 AB	4.99 ± 0.32 AB	5.46 ± 0.15 AB
	15	4.67 ± 0.25 AB	4.60 ± 0.27 B	4.21 ± 0.67 AB	3.30 ± 0.11 B	4.82 ± 0.36 AB	4.61 ± 0.26 B
48.9	0	4.42 ± 0.46 AB	4.49 ± 0.89 B	3.61 ± 0.25 AB	4.22 ± 1.06 AB	4.48 ± 0.43 AB	4.68 ± 0.95 BC
	15	4.21 ± 0.07 BC	3.92 ± 0.16 BC	4.09 ± 0.70 ABC	3.42 ± 0.27 B	4.51 ± 0.34 BC	4.04 ± 0.19 BC
60.0	0	4.05 ± 0.48 BC	4.07 ± 1.55 B	3.03 ± 0.65 BC	3.38 ± 0.99 B	4.09 ± 0.50 B	4.18 ± 1.45 BC
	15	3.55 ± 0.19 BC	2.38 ± 0.06 D	2.99 ± 0.54 BC	1.68 ± 0.42 B	3.66 ± 0.22 BC	2.46 ± 0.53 D
71.1	0	2.71 ± 1.41 C	2.63 ± 0.44 C	2.01 ± 0.82 C	1.79 ± 0.43 B	2.81 ± 1.26 C	2.69 ± 0.43 C
	15	2.83 ± 1.01 C	2.81 ± 1.19 CD	2.85 ± 0.22 C	2.37 ± 1.31 BC	3.31 ± 0.34 C	2.94 ± 1.20 CD

[a] STEC levels reported are the summation of total CFU from all strips plus all quarters and represents the results from two trials and 42 pieces of meat.

[b] For a given formulation and storage time, temperature means with different letters within a column are significantly ($P \leq 0.05$) different by the LSD test.

TABLE 4. *Postenrichment recovery rates for ECOH from cooked steak portions failing to yield the pathogen by direct plating*

Brine formulation	Temp (°C)	Storage (days)	Strips (S1 plus S2 plus S3)[a]	Quarters (Q1 plus Q2 plus Q3 plus Q4)[b]
Lac^-	37.8	0	18/18 direct plating[c] 0/0 enrichment[e]	24/24 direct plating[d] 0/0 enrichment[f]
		15	18/18 direct plating 0/0 enrichment	23/24 direct plating 1/1 enrichment
	48.9	0	18/18 direct plating 0/0 enrichment	23/24 direct plating 1/1 enrichment
		15	12/18 direct plating 6/6 enrichment	17/24 direct plating 6/7 enrichment
	60.0	0	16/18 direct plating 1/2 enrichment	17/24 direct plating 6/7 enrichment
		15	10/18 direct plating 6/8 enrichment	14/24 direct plating 9/10 enrichment
	71.1	0	8/18 direct plating 5/10 enrichment	5/24 direct plating 6/19 enrichment
		15	6/18 direct plating 4/12 enrichment	7/24 direct plating 6/17 enrichment
Lac^+	37.8	0	18/18 direct plating[c] 0/0 enrichment[e]	24/24 direct plating[d] 0/0 enrichment[f]
		15	18/18 direct plating 0/0 enrichment	24/24 direct plating 0/0 enrichment
	48.9	0	17/18 direct plating 1/1 enrichment	22/24 direct plating 2/2 enrichment
		15	16/18 direct plating 2/2 enrichment	22/24 direct plating 1/2 enrichment
	60.0	0	15/18 direct plating 2/3 enrichment	20/24 direct plating 4/4 enrichment
		15	13/18 direct plating 1/5 enrichment	18/24 direct plating 3/6 enrichment
	71.1	0	11/18 direct plating 4/7 enrichment	7/24 direct plating 14/17 enrichment
		15	9/18 direct plating 4/9 enrichment	7/24 direct plating 2/17 enrichment

[a] Enrichment and direct plating results for a composite of strips 1, 2, and/or 3 (summation of 3 steaks × 3 strips × 2 trials; 18 strips total per each temperature) obtained from cooked steaks.

[b] Enrichment and direct plating results for a composite of quarters 1, 2, 3, and/or 4 (summation of 3 steaks × 4 quarters × 2 trials; 24 quarters total per each temperature) obtained from cooked steaks.

[c] Number of strip composite samples from which ECOH were recovered by direct plating/total number of composite samples direct plated.

[d] Number of quarter composite samples from which ECOH were recovered by direct plating/total number of composite samples direct plated.

[e] Number of strip composite samples from which ECOH were recovered by enrichment/total number of composite samples enriched.

[f] Number of quarter composite samples from which ECOH were recovered by enrichment/total number of composite samples enriched.

agreement with the above-mentioned studies, in that most cells (3.0 to 93.3%) remained in the topmost 1 cm of beef subprimals after tenderization, and that both pathogens were transferred throughout the subprimal in decreasing order into the lower segments, that being segments 2 through 6. In general, we observed an increase in percent recovery in segment 6 compared with segments 3, 4, or 5. Although we have no data to support this contention, it is possible that in addition to the physical impingement or transfer of cells into the interior of the subprimals by the blades, any back pressure and/or vacuum created by the withdrawal of the blades from subprimals during tenderization could force additional cells into the deepest tissue of the meat, that being segment 6. Further studies are warranted to verify how and why more cells are recovered from segment 6 compared with segments 3, 4, and 5, and to confirm if this observation is reproducible and/or statistically relevant. Regardless, our data also revealed, for the first time, that in general, there were no discernible differences in the extent or levels of translocation between ECOH and STEC after chemical injection and/or in their viability during subsequent refrigerated storage of nonintact beef subprimals. The brine formulations used in the present study, which contained salt and phosphate, both with and without lactate and diacetate, were selected based on discussions with collaborators in the meat industry to be representative of what several commercial processors were using at the time this study was initiated, including a processor that supplied a major/global retail chain. It would be of value to evaluate other formulations and to test different salts, such as calcium, in

TABLE 5. *Postenrichment recovery rates for STEC from cooked steak portions failing to yield the pathogen by direct plating*

Brine formulation	Temp (°C)	Storage (days)	Strips (S1 plus S2 plus S3)[a]	Quarters (Q1 plus Q2 plus Q3 plus Q4)[b]
Lac$^-$	37.8	0	17/18 direct plating[c] 1/1 enrichment[e]	24/24 direct plating[d] 0/0 enrichment[f]
		15	18/18 direct plating 0/0 enrichment	24/24 direct plating 0/0 enrichment
	48.9	0	16/18 direct plating 1/2 enrichment	22/24 direct plating 2/2 enrichment
		15	17/18 direct plating 1/1 enrichment	20/24 direct plating 2/4 enrichment
	60.0	0	14/18 direct plating 4/4 enrichment	14/24 direct plating 2/10 enrichment
		15	13/18 direct plating 1/5 enrichment	12/24 direct plating 2/12 enrichment
	71.1	0	13/18 direct plating 1/5 enrichment	9/24 direct plating 7/15 enrichment
		15	9/18 direct plating 1/9 enrichment	7/24 direct plating 0/17 enrichment
Lac$^+$	37.8	0	18/18 direct plating[c] 0/0 enrichment[e]	24/24 direct plating[d] 0/0 enrichment[f]
		15	17/18 direct plating 1/1 enrichment	23/24 direct plating 1/1 enrichment
	48.9	0	18/18 direct plating 0/0 enrichment	24/24 direct plating 0/0 enrichment
		15	16/18 direct plating 1/2 enrichment	21/24 direct plating 0/3 enrichment
	60.0	0	18/18 direct plating 0/0 enrichment	18/24 direct plating 4/6 enrichment
		15	11/18 direct plating 1/7 enrichment	13/24 direct plating 5/11 enrichment
	71.1	0	9/18 direct plating 3/9 enrichment	6/24 direct plating 8/18 enrichment
		15	12/18 direct plating 0/6 enrichment	8/24 direct plating 6/16 enrichment

[a] Enrichment and direct plating results for a composite of strips 1, 2, and/or 3 (summation of 3 steaks × 3 strips × 2 trials; 18 strips total per each temperature) obtained from cooked steaks.
[b] Enrichment and direct plating results for a composite of quarters 1, 2, 3, and/or 4 (summation of 3 steaks × 4 quarters × 2 trials; 24 quarters total per each temperature) obtained from cooked steaks.
[c] Number of strip composite samples from which STEC were recovered by direct plating/total number of composite samples direct plated.
[d] Number of quarter composite samples from which STEC were recovered by direct plating/total number of composite samples direct plated.
[e] Number of strip composite samples from which STEC were recovered by enrichment/total number of composite samples enriched.
[f] Number of quarter composite samples from which STEC were recovered by enrichment/total number of composite samples enriched.

combination with other antimicrobials, including organic acids, in the brine used for injection to better tenderize and possibly protect nonintact products, with respect to spoilage and pathogenic microbes. To this end, Yoon et al. *(45)* reported that brines containing selected organic acids (e.g., acetic, citric) when used in combination with chemical tenderizers (e.g., calcium chloride) generated greater thermal destruction of ECOH during subsequent cooking of tenderized and enhanced nonintact raw beef. As noted by Shen et al. *(37)*, the choice of cooking appliance also affected thermal inactivation of ECOH in their model nonintact beef system.

Given the apparent rise in the United States in illnesses linked to verocytotoxigenic *E. coli* displaying serotypes other than ECOH *(35)*, considerable efforts have been directed to obtain information on the behavior of STEC in foods to facilitate the development of appropriate control strategies. The limited data collected thus far suggest that certain STEC might behave similarly to ECOH at the physiological level when challenged by food-relevant conditions of temperature, pH, salt, and water content *(27)*. As summarized by Mathusa et al. *(27)*, desiccation resistance on paper disks and in dry foods was not serotype dependent for comparisons among O157, O26, and O111 strains; there were no significant differences on beef tissue surfaces between ECOH and STEC in response to acidified sodium chlorite (1,000 ppm), octanoic acid (9,000 ppm), and peracetic acid (200 ppm), and in general, STEC displayed similar heat resistance (in apple juice) to ECOH. Our data are in general agreement with the above-mentioned studies with both ECOH and STEC showing similar reductions (0.3 to 4.1 log CFU/g) after cooking injected

TABLE 6. *Average temperature and range indentified for end target temperatures after cooking brine-injected beefsteaks on a gas grill*

Brine formulation	Target cooking temp (°C)[a]	Storage (days)	Avg (range) temp achieved (°C)[b]	
			ECOH	STEC
Lac$^-$	37.8	0	47.2 (32.2–61.1)	48.9 (31.7–70.0)
		15	47.2 (23.9–58.9)	52.8 (40.0–77.2)
	48.9	0	58.3 (27.2–81.1)	58.3 (37.8–76.7)
		15	57.2 (33.3–72.2)	57.2 (43.9–76.7)
	60.0	0	66.1 (43.3–91.1)	69.4 (49.4–97.2)
		15	68.3 (48.3–80.0)	69.4 (55.6–82.2)
	71.1	0	73.9 (63.9–88.9)	77.2 (61.1–89.4)
		15	73.3 (48.3–91.6)	76.1 (65.0–95.0)
Lac$^+$	37.8	0	45.5 (25.0–72.2)	46.7 (28.9–67.2)
		15	49.6 (34.4–72.2)	51.5 (37.8–71.1)
	48.9	0	54.4 (27.2–70.0)	58.3 (31.1–77.7)
		15	59.6 (35–73.3)	56.7 (35.0–80.5)
	60.0	0	62.4 (42.2–78.3)	66.1 (43.9–83.9)
		15	69.3 (48.9–83.9)	70.0 (52.2–82.2)
	71.1	0	77.2 (64.4–87.8)	80.5 (62.7–88.9)
		15	76.8 (59.4–89.4)	80.0 (59.4–102.2)

[a] The target cooking temperature was the temperature achieved by two independent, internal thermocouples within each steak.
[b] Values are the average of eight independent temperature readings within each steak after removing steaks from the grill (two trials, three steaks per trial, and 8 readings per steak for a total of 48 readings).

steaks on a gas grill. In related studies, we observed no discernible differences in thermal resistance between ECOH and STEC after cooking blade-tenderized steaks on a gas grill (data not shown). Moreover, in general, higher temperatures generated greater lethality (>2.5 log CFU/g), and there were no apparent differences in lethality based on thickness (1.0 versus 1.5 in. [2.5 to 3.8 cm]) of blade-tenderized steaks in our related studies (data not shown). Shen and colleagues *(37)* reported *E. coli* reductions of 1.1 to 4.2 log CFU/g after broiling or roasting of a simulated restructured beef product containing sodium chloride and sodium tripolyphosphate, whereas researchers at Kansas State University reported *E. coli* reductions of 3.0 to 6.0 log CFU/g *(39)* in blade-tenderized beefsteaks after cooking on a gas grill and an electric skillet. In related studies on ground beef, other investigators reported *E. coli* reductions of 1.5 to 5.5 log CFU/g after cooking to 60 or 68.3°C *(17, 18)*. Such differences among studies could be attributed, at least in part, to differences in strains, cooking methods–appliances, types of meat, and/or plating media. Regardless, federal agencies have specified cooking parameters deemed adequate for assuring the safety of red meat and poultry products *(41, 43)*. The existing literature and our findings suggest that interventions effective against ECOH (or even *Salmonella*) would be equally as effective toward STEC *(27)*. These findings will assist in the development of comparative risk assessments of intact and nonintact beef products.

In the present study, fortuitous survivors were recovered from chemically injected steaks after cooking. It must be stated, however, that non-ecologically relevant levels of ECOH and STEC were surface inoculated onto beef subprimals and, as such, cooking these highly contaminated steaks on a gas grill, even when the recommended temperature of 71.1°C (160°F) was achieved, was not sufficient to kill all cells of either of these pathogen cocktails. Fortuitous survivors were most likely observed because not all portions of the steak achieved the target end temperature, due to a reduction in heat penetration from the insulating effects of fat or connective tissue, or the added moisture from injection, and/or from the intrinsic variability in temperature at the cooking surface. As discussed, even when the target end temperature was achieved as recorded by two independent thermocouples inserted into the same steak, the observed range of temperatures, as subsequently measured postcooking by using a handheld temperature monitor, varied considerably despite the fact that the overall average temperatures substantially exceeded the intended target temperatures. This could be significant from the public health perspective, as it is likely that most people will take only a single measurement of temperature, if any, to determine doneness. Our findings are of immediate and appreciable relevance because we evaluated conditions likely practiced by consumers, and because we tenderized and cooked steaks by using commercial apparatuses rather than small-scale, laboratory-controlled conditions, and/or a model meat system to simulate tenderization and/or a water bath to simulate cooking. Given the nonhomogeneous nature of steaks and the related physics–kinetics associated with cooking, it is likely that not all portions of the meat achieved the target temperature; however, this would result in significant reductions in pathogen numbers (e.g., 2.5 to 5.0 log), albeit while allowing for the recovery of fortuitous survivors, as has been reported elsewhere *(13, 24, 37, 45)*. Thus, it may be necessary to evaluate slightly higher endpoint cooking temperatures, with or without a holding time, to ensure total elimination of ECOH and STEC. Alternatively, given that the risk might never be totally eliminated, and the extremely low prevalence or levels of ECOH and STEC likely to be encountered outside the

laboratory setting *(3, 19, 44)*, a 1.0- to 2.0-log reduction achieved by cooking could still have an appreciable and positive effect on public health. Future efforts should be directed to generate *D*-values in synthetic media or model meat systems for the individual strains composing these pathogen cocktails.

Although the National Advisory Committee on Microbiological Criteria for Foods *(30)* concluded that blade-tenderized, nonintact beefsteaks do not pose a greater risk to public health from ECOH than do intact beefsteaks, if the meat is oven broiled and cooked to an internal temperature of ≥60°C (140°F), the process of tenderization does indeed transfer pathogens that might be present on the surface of the meat, albeit at low occurrences and levels *(3, 19, 44)*, to the interior of the product. It should be noted that there are currently no requirements for such products to be labeled as "nonintact" and, moreover, based on the absence of an identifier on the label and/or due to difficulty with visually discerning differences between products that have been pierced and those that have not, there is growing concern that consumers and/or retail establishments would not know that such products are nonintact and, as such, might require longer cooking times and/or higher temperatures to prevent foodborne illness. As mentioned, this risk is compounded by the fact that consumers frequently order steaks cooked to less than a medium degree of doneness (<60°C [<140°F]) *(10, 21, 36)*, and that ca. 18% of beef sold at retail is mechanically tenderized and/or enhanced *(2)*. Regardless, our data validate that ECOH and STEC behave similarly with respect to translocation and thermal inactivation within chemically enhanced subprimals and steaks. Our findings also establish that proper cooking appreciably reduces the levels of Shiga toxin–producing *E. coli* in chemically tenderized meat, but does not eliminate the pathogen, due to nonuniform heating within steaks. Further research is warranted to develop interventions to treat subprimals prior to tenderization and/or to develop brines for injection that may lessen the prevalence and levels of ECOH and/or STEC during subsequent storage and cooking. Regardless, the data herein are useful to estimate the comparative risk between intact and nonintact meats and to assist in the validation of targeted interventions and the development of potential labeling requirements for such products.

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Cooking Inactivation of Shiga Toxin Producing *Escherichia coli* (STEC) in Non-intact Beefs--a mini-Review

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Abstract

Escherichia coli O157:H7 and Non-O157 shiga toxin producing *E. coli* (STEC) serogroups O26, O45, O103, O111, O121, or O145 are both considered adulterated in non-intact beefs. Forty to fifty-eight percent of U.S. consumers prefer to order beef steaks of medium rare to rare status. From 2000 to 2007, undercooked non-intact beef products have been involved in several outbreaks in the United States due to contamination with *E. coli* O157:H7.Traditional cooking practices of non-intact beef steaks contaminated with Non-O157 STEC may result in the same food safety risks as shown in *E. coli* O157:H7 through consumption of undercooked contaminated products. This mini-review focuses on the research advances in recent 10 years to evaluate the effectiveness of cooking inactivation of *E. coli* O157:H7 and Non-O157 STEC contaminated in ground beef, mechanical tenderized beef, and moisture enhanced non-intact beef.

Introduction

Non-intact beef products include ground beef, mechanically or chemically tenderized beef cuts, restructured entrees, and meat products that have been injected with brining solutions for enhancement of flavor and/or tenderness [1]. *Escherichia coli* O157:H7 or Non O157 Shiga toxin-producing *E. coli* (STEC) may be translocated from the meat surface to internal tissue by mechanical tenderization or needle injection of solutions, or entrapped in the tissue during restructuring. These processing procedures could protect the pathogens from lethal heating effects, especially if the products are undercooked [2,3].

E. coli O157:H7 or Non-O157 STEC can generate shiga-like toxin and cause severe hemolytic uremic syndrome in infected human bodies, and 10 cells of infection may cause a healthy person dead [4]. Since 1999, *E. coli* O157:H7 has been considered as an adulterant of raw non-intact beef products [1]. On November 2011, U.S. Department of Agriculture, Food Safety and Inspection Service (USDA-FSIS) announced that, as of June 2012, non-intact beef products would also be considered adulterated if they were contaminated with Non-O157 STEC serogroups O26, O45, O103, O111, O121, or O145 [5]. In an early report from Centers for Disease Control and Prevention (CDC), 62,000 cases of symptomatic *E. coli* O157:H7 were estimated to occur annually in the United States, resulting in approximately 1,800 hospitalizations and 52 deaths [6]. However, CDC's new data [7] generated in 2011, revealed that *E. coli* O157:H7 and Non-O157 STEC caused annual foodborne illnesses ranging from 73,000 to 97,000 and 37,000 to 169,000, respectively, which increased dramatically since their report [6] of 1999.

Most recently, USDA-FSIS's "Risk Profile of Pathogenic Non-O157 STEC" states that traditional cooking of non-intact steaks contaminated with Non-O157 STEC may result in the same food safety risks as shown in *E. coli* O157:H7 through consumption of undercooked contaminated products [8]. A recent study reported that, Non-O157 STEC with 9 serogroups was isolated from 300 out of 4133 ground beefs collected from 18 commercial ground beef produces [9]. This high isolation rate indicates that non-intact beefs, if contaminated or undercooked, could be a source of foodborne illness associated with Non-O157 STEC. Recent estimates indicate that Non-O157 STEC is responsible for 112,752 foodborne illnesses in the U.S. annually [7]. According to the survey, 40-58% of U.S. consumers ordered beef steaks at medium rare (60 to 62.8°C) to rare (54.4 to 57.2°C), which could be a potential threat to public health [10].

Thus, comprehensive studies of demonstrating *E. coli* O157:H7 and Non-O157 STEC lethality during common cooking practices for non-intact beef products lead a great understanding and meaningful comparative risk assessment of non-intact beef safety. This mini-review discusses the regulatory cooking recommendations and research progress related to thermal inactivation of STEC in non-intact beefs.

Regulatory cooking recommendations

The USDA-FSIS indicated that ground beef cooked to an internal temperature of at least 160°F (71.1°C) [11], and non-intact blade-tenderized beef steaks oven-broiled to an internal temperature of 140°F (60°C) or above would not present a great risk to the consumers [2,3]. Recently, they suggested that the microbiological safety of mechanically tenderized beef steaks could be assured by cooking to an internal temperature of 71°C, or cooking to internal temperature of 63°C with rest time of 3 min [12]. They also recommended that the starting temperature of the cooking oven should be at 350°F (176°C) or no lower than 325°F (162.8°C) when cooking corned beef [13]. The North Carolina State's Division of Environmental Health in the Department of Environment and Natural Resources stated that cooking all muscle beef such as steaks and roasts to an internal temperature of 155°F (68.3°C), as measured by a food thermometer at home, will eliminate the bacteria inside the meat [14]. In 2002, a

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comparative risk assessment for non-intact and intact beef steaks conducted by USDA-FSIS [2,3] indicated that there were insufficient data to determine whether the traditional cooking methods are adequate to destroy *E. coli* O157:H7 translocated during the blade tenderization process. Because proper cooking renders foods microbial safe, it is important to develop effective cooking protocols for non-intact beef products.

Research Progress of Thermal Inactivation of *E. coli* O157:H7 and Non-O157 STEC

Previous studies showed clearly that the lethality of STECs during and after cooking depends on multiple factors including cooked internal temperatures, cooking appliances, products' thickness, ingredients and the location of the bacteria within the meat. Besides, *E. coli* O157:H7 and Non-O157 STEC behave similarly when exposed to thermal treatments. The efficacy of thermal inactivation of STECs in non-intact beefs relies on cooked internal target temperatures. An early research from Sporing [15] showed that oven-broiling 3.2 cm thick beef steaks to the internal temperature of 60 to 76.7°C resulted in a 6.5 log reduction of *E. coli* O157:H7. Luchansky *et al.* [16] reported that cooking blade-tenderized beef subprimal steaks to internal temperature ranging from 48.8 to 60°C on a commercial open-flame gas grill resulted in 2.6 to 4.6 log CFU/g reductions of surfaced inoculated *E. coli* O157:H7. Recently, Luchansky et al. [17] reported that cooking brine-injected steaks on a commercial open-flame gas grill to internal temperatures of 37.8°C (100°F) to 71.1°C (160°F) achieved reductions of 0.3 to 4.1log CFU/g of *E. coli* O157:H7 and 0.5 to 3.6 log CFU/g of Non-O157 STEC among different brine formulation or internal temperatures. In a related study, they also found that cooking refrigerated, frozen, or freeze-thawed ground beef patties on commercial gas or electric grills to internal temperatures of 71.1 and 76.6°C reduced 5.1-7.0 log CFU/g of *E. coli* O157:H7 and Non-O157 STEC [18].

Non-intact beefs are typically prepared in commercial restaurants or consumer homes using a variety of cooking methods including broiling, grilling, frying and microwaving [19,15,2,3]. The survey of food intakes by individuals conducted by the USDA indicated that approximately 30% of consumers grill or broil steaks, while 40% of them fry steaks [20]. According to the guidelines of cooking by the American Meat Science Association, cooking methods include roasting, broiling and pan-broiling. Roasting is a method transmitting heat to the meat by convection in a closed preheated oven; broiling cooks directly through radiant heat from one direction; and pan-broiling is used for cooking thin patties by direct heat of conduction [21]. The terminologies of the cooking methods were also described by other names in different research studies, which are shown in Table 1.

Various cooking methods affected thermal inactivation efficacy of STEC in non-intact beefs. In a study to compare a double-side grill with a single side grill simulating consumer-style, Rhee et al. [22] revealed that *E. coli* O157:H7 reductions increased as one-turnover-single-sided grill (4.7 log CFU/g) <multi-turnover-single-sided grill (5.6 log CFU/g) < double-sided grill (6.9 log CFU/g) when ground beef was cooked to internal temperature of 71.1°C. Studies [15,23,24,25] from various research institutions suggested that broiling was the best cooking method regarding thermal inactivation of *E. coli*O157:H7 in non-intact beefs. Sporing [15] reported approximately 3 to 5, 4 to 6 and 5 to 6 log CFU/g reductions of *E. coli* O157:H7 on beef steaks cooked to the internal temperature of 65.6°C using electric skillet frying, gas grilling and oven broiling, respectively; they also found that cooking effectiveness on pathogen inactivation increased in the order of broiling > grilling > frying. Ortega-Valenzuela et al. [23] reported that broiling was more effective in eliminating *E. coli* O157:H7 compared with gas grilling when the same internal temperature was reached. Similar results were also obtained by Mukherjee *et al.* [24]. Moreover, Yoon et al. [25] confirmed that broiling was more effective in reducing *E. coli* O157:H7 than grilling and pan-frying in tenderized ground beef patties when cooked to 60 or 65°C.The possible explanation is that broiling causes more even distribution of heat surrounding the samples, takes longer time to reach the final internal temperature, and allows the temperature of the product near the surface to increase more significantly than grilling or frying [15]. In a recent study conducted by the author, thermal inactivation of *E. coli* O157:H7 in salt and sodium-tripolyphosphate moisture enhanced reconstructed beefs increased in the order of double pan-broiling ≤ pan-broiling<roasting when the internal temperature reached 65°C [26]. The author's another study found that cooking appliance with high starting temperatures of 204-260°C resulted in greater reductions (3.3-5.5 log CFU/g) than those obtained at low starting temperature of 149°C (1.5-2.4 log CFU/g) [27]. In the same project, Adler *et al.* [28] found that *E. coli* O157:H7 survivals caused by pan broiling increased with increasing depth of the contamination, whereas roasting in a kitchen oven showed similar pathogen survivals regardless of the depth.

Table 1: The conundrum of terminologies of cooking methods of various studies.

Heat transferring	What is stated by other studies	What is stated by AMSA Appliances
Conduction	Grilling (23; 24; 25)	Double pan-broiling George Foreman® grill (26) (26)
Conduction	Frying (15; 23; 24; 25)	Pan-broiling Presto® electric skillet (26; 27; 34)Sanyo® grill (26;27;34)
Convection	Broiling (15; 23; 24; 25)	Roasting Oster® toaster oven (26; 27)Magic Chef ® kitchen oven (26;27)

In addition to the cooking methods, the thickness of non-intact beef is another factor that influences the heat transfer through the product and consequently the elimination rates of pathogens inside the steaks. Sporing [15] reported that thick steaks (3.2 cm) compared to thin ones (1.3 cm) had more reduction of *E. coli* O157:H7 when cooked to the same internal temperature by broiling in a standard kitchen oven. The study also found that the thick (4.0 cm) moisture enhanced non-intact beefs resulted in greater inactivation of *E. coli* O157:H7 than their thinner (1.5 cm or 2.5 cm) counterparts when cooked to an internal temperature of 65°C [26]. In a similar study, Adler *et al.* [28] observed that pan-broiling or roasting blade-tenderized beef steaks with a thickness of 2.4 cm resulted in an additional 1 log CFU/g reduction of *E. coli* O157:H7 compared to the 1.2 cm thick samples. This might be attributed to overheating of the thicker products due to the longer cooking time that was taken to cook them to the same internal temperature. However, Luchansky's study [29] found that regardless of temperature or thickness, cooking 2.54 and 3.81 cm thick mechanical tenderized steaks on a commercial open-flame gas grill to internal temperatures of 48.9-71.1°C reached

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2.0-4.1 log (*E. coli* O157:H7) and 1.5-4.5 log (Non O157-STEC) reductions, respectively. They pointed out that *E. coli* O157:H7 and Non O157-STEC behaved similarly during heat processing, and some unexpected survivors were due to uneven heating of the products [29].

The above major factors influencing thermal inaction of the pathogen in non-intact beefs were concluded in table 2. Additional studies showed that other factors including beef ingredients, stress adapted STEC cells, cooked beef searing time and turning over times influenced STEC reduction during thermal process. Four studies conducted by Muherjee and Yoon [30,31,32,33] indicated that lethality rate of internalized *E. coli* O157:H7 in marinated, tenderized, and reconstructed beef patties increased with adding organic acids (lactic, citric and acetic acid) in beef mixture. Muherjee *et al.* [30] found that potassium lactate protected *E. coli* O157:H7 in beefs during thermal processing. Their study showed that addition of cetylpyridiniumchlorite (CPC) to the brine solution of moisture enhanced reconstructed beefs resulted in the lowest number of *E. coli* O157:H7 survivors when the samples were cooked to 65°C [34]. The same study also found that acid stress-adapted cells were resistant to heat treatments, however, cold or desiccation stress-adapted cells were more sensitive to heat as compared with unstress-adapted E. coli O157:H7 cells [34]. In a study of investigating searing time of cooked non-intact beef, Porto-Fett *et al.* [35] suggested that cooking beef prime rib to internal temperatures of 48.9, 60.0, or 71.1°C with following hold at 60.0°C for ≥ 8 h could achieve a 5.0-log reduction of *E. coli* O157:H7. In another study of investigating the effect of steaks' turning over times for STEC control, Gill *et al.* [36] observed that turning over mechanically tenderized steaks more than twice during grilling to 63°C killed all contaminated *E. coli* O157:H7, which achieved better pathogen inactivation effect than those cooking to 71°C with only once turning over.

Other technologies regarding inactivation of *E. coli* O157:H7 and Non-O157 STEC

In addition to thermal treatments, other technologies including application of surface trimming, commercial antimicrobials and probiotics (e.g. lactic acid bacteria) during non-intact beef processing were reported to effectively reduce *E. coli* O157:H7and Non-O157 STEC populations. In a study of evaluating the efficacy of water-wash with beef surface trimming to inactivate *E. coli* O157:H7 on vacuum packaged beef trimmings, Lemmons *et al.* [37] reported that full surface trimming without or with water-wash reduced the pathogen populations below detective limit. A study conducted by Echeverry *et al.* [38] found that spraying lactic acid bacteria, acidified sodium chlorite and lactic acid on mechanically tenderized and brine enhanced beef reduced internalized *E. coli* O157:H7 by 1.2 to >2.2 log, and 0.8 to 3.0 log, respectively. In a similar study, Echeverry *et al.* [39] reported that lactic acid bacteria and lactic acid reduced internal *E. coli* O157:H7 up to 3.0 log and acidified sodium chlorite reduced the pathogen ranged from 1.4 to 2.3 login mechanically tenderized and brine enhanced beef under a simulated purveyor setting. A series studies of Adler *et al.* [40], Byelashov *et al.* [41], and Ko *et al.* [42] found that adding cetylpyridinium chloride (0.5-5.5%) in non-intact beef bring solutions showed immediate or additional reduction of *E. coli* O157:H7 by 1 to > 3.5 log/unit. In the same research group, Geornaras *et al.* [43] reported that acidified sodium chlorite, peroxyacetic acid, or sodium metasilicate reduced *E. coli* O157:H7 or Non-O157 STEC on non-intact beef trimmings by 0.7 to 1.0, 0.6 to 1.0, and 1.3 to 1.5 log CFU/cm², respectively. In a related study, Fouladkhah *et al.* [44] found that dipping beef trimmings in 5% lactic acid of 25 or 55°C reduced *E. coli* O157:H7 and Non-O157 STEC by 0.5 to 0.9 and 1.0 to 1.4 log CFU/cm², respectively. Wolf *et al.* [45] also reported that lactic acid treatment reduced *E. coli* O157:H7 on beef trimmings by 0.91 to 1.41 and Non-O157 STEC by 0.48 to 0.82 log CFU/cm². A most recent study of Jadeja and Hung [46] suggested that washing beef trimming in electrolyzed oxidative water with free chlorine of 50 ppm reduced *E. coli* O157:H7 and Non-O157 STEC by 0.44 to 1.54log CFU/cm².These studies clearly showed that applying commercial antimicrobials during non-intact beef processing reached averagely 0.5 to 3.0 log reduction of *E. coli* O157:H7 or Non-O157 STEC, which is lower than those of thermal treatments.

Table 2: Major thermal processing factors influencing cooking inactivation of pathogens in non-intact beefs.

Factors	Influence	References
Cooked internal temperatures	The higher, the more reduction	[15-18]
Cooking methods	Broiling (roasting) is the best	[15,23-26]
Thickness of beef	The thicker, the more reduction	[15,26,28]

In conclusion, research results generated from these studies quantified the efficacies of cooking intervention as post-harvest STEC control practices applied to non-intact beef preparation at retail, foodservices, and home. They provided useful information regarding developing educational material and extension fact sheets, such as a cooking guidance manual about proper production and storage of non-intact beef for industry personnel, foodservice, and consumers, and choosing appropriate cooking approaches and degree of doneness when cooking beef products. These findings should also be useful for U.S. Department of Agriculture, Food Safety and Inspection Service USDA-FSIS to update risk assessments of *E. coli* O157 and Non-O157 STEC on non-intact and intact beef products.

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Consumer Attitudes toward Food, Nutrition & Health

A Trended Survey

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  International Food Information Council Foundation

- Tony Flood

  Director, Food Safety Communications International
  Food Information Council Foundation

- Robert "Bob" Gravani, PhD

  Professor of Food Science, Cornell University

- Diane Van

  Manager, USDA Meat and Poultry Hotline

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Agenda

- **Brief Background**
- **Presentation of Findings**
- **Pathogens that Cause Foodborne Illness**
- **Practical Tips for Consumers**
- **Questions and Answers**
  - Submit your questions via email to foodandhealth@ific.org
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Primarily supported by the broad-based food, beverage and agricultural industries.

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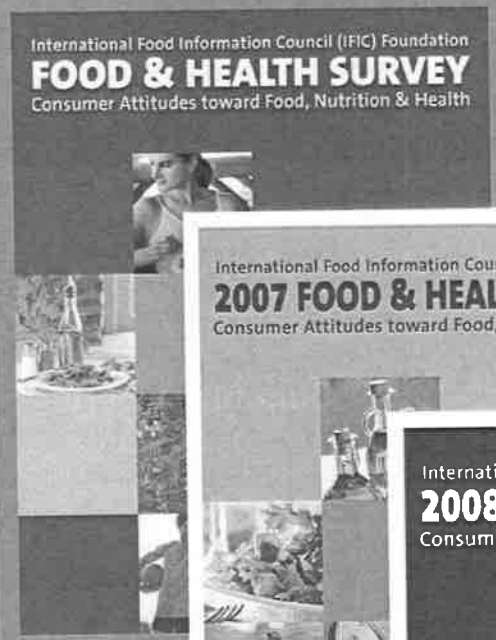

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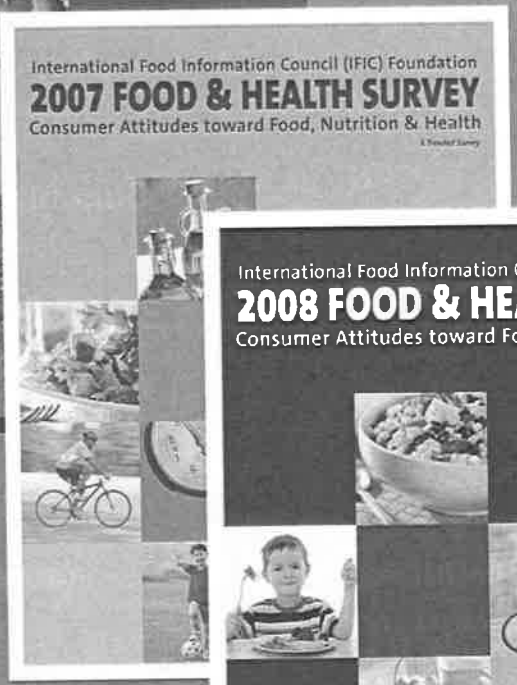

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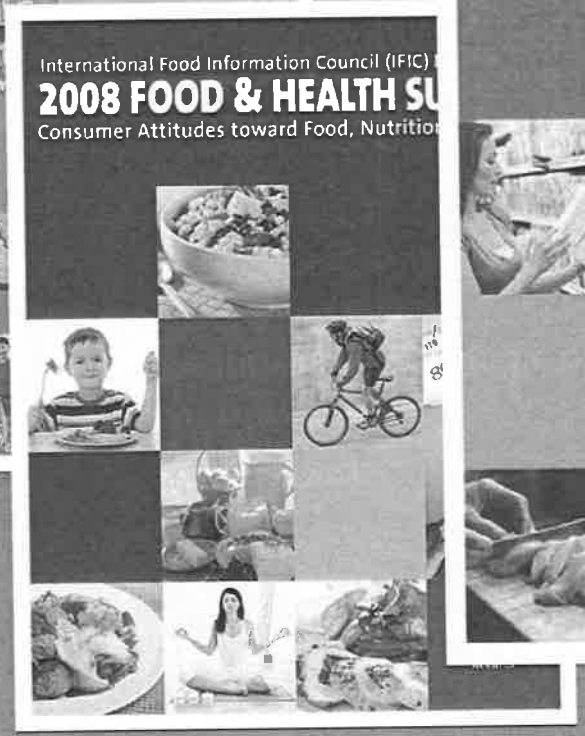

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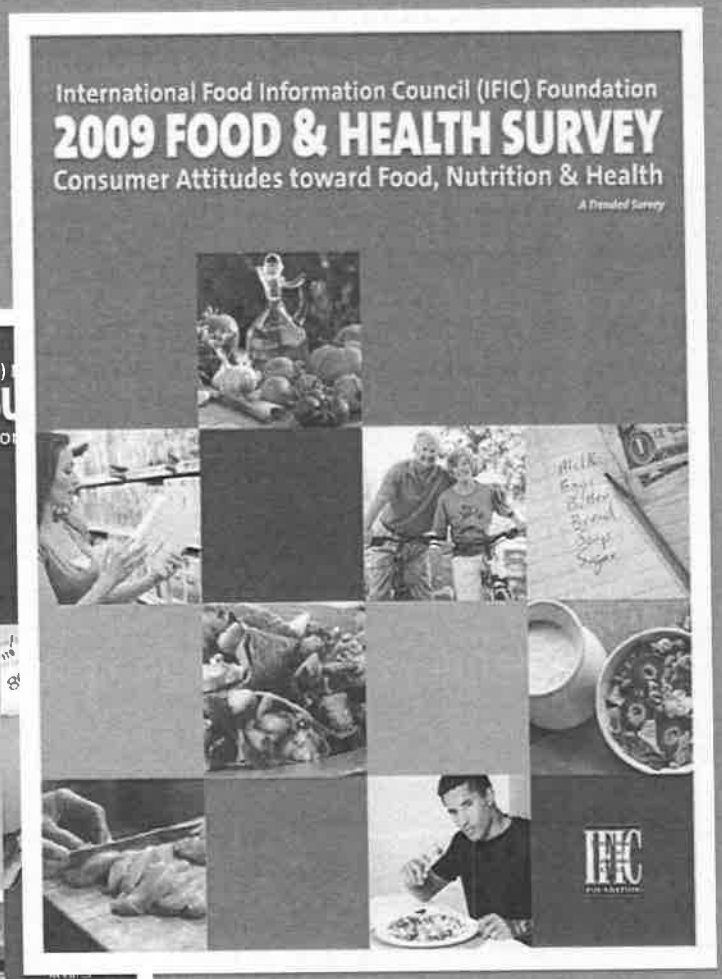

2009

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Methodology

Methodology	**Web Survey**
Population	**Representative Sample of Americans Aged 18+**
Data Collection Period	**February 19-March 11, 2009**
Sample Size (Error)	**n=1,064 (± 3.0 for 2009) (± 4.4 among 2009, 2008, 2007, 2006)**
Data Weighting*	**Data Weighted on Age, Gender, Income, Education and Race**

*Weighting is a widely accepted statistical technique that is used to ensure that the distribution of the sample reflects that of the population on key demographics. With any data collection method, even when the outgoing sample is balanced to the Census, some populations are more likely than others to respond.

▲ / ▼ Significant increase/decrease from year indicated

Taste Still the Number One Factor Influencing Purchases; Price Increases to an All-Time High

□ Some Impact □ Great Impact

How much of an impact do the following have on your decision to buy foods and beverages?
(n=1064)

▲ / ▼ Significant increase/decrease from year indicated

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Sources of Information Guiding Food Safety and Health Practices

Food label	61%
Friends/family	42%
Health professional	33%
Grocery store, drug store, or specialty store	28%
Magazine article	28%
Internet article	27%
TV news program	24%
Product or manufacturer communications	13%
Newspaper	13%
Health association	9%
Dietitian	8%
Government official/agency	5%
Radio news program	3%
Blog or social networking site	1%
Other	4%

What three sources of information do you use most often to guide your food, nutrition, and food safety practices? *Select three.* (n=1064)

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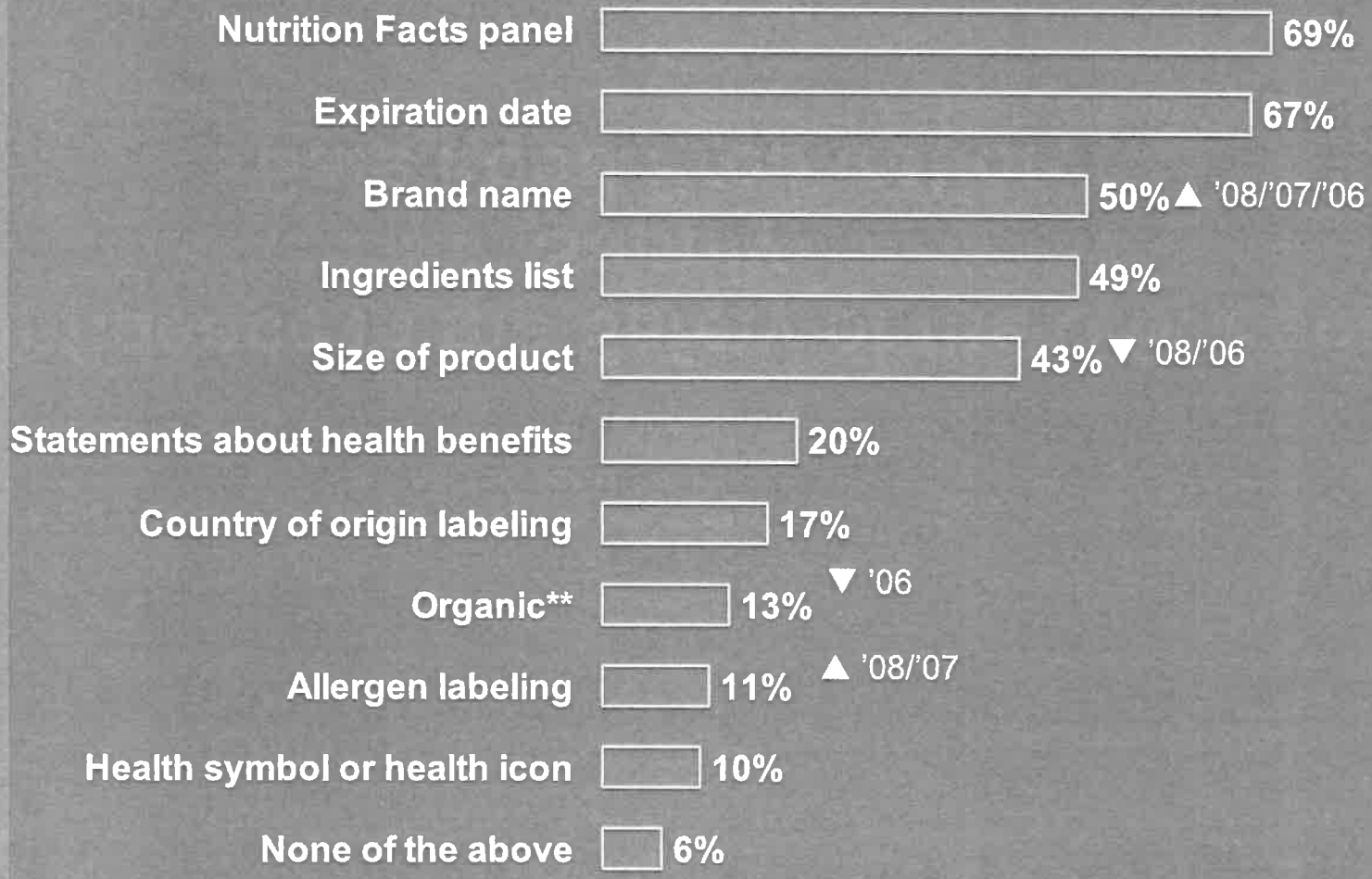
Information Used on the Food and Beverage Package



What information do you look for on the food or beverage package when deciding to purchase or eat a food or beverage? *Select all that apply.* (n=1064)

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FOOD SAFETY

Confidence in the Safety of the U.S. Food Supply
Safe Food Preparation

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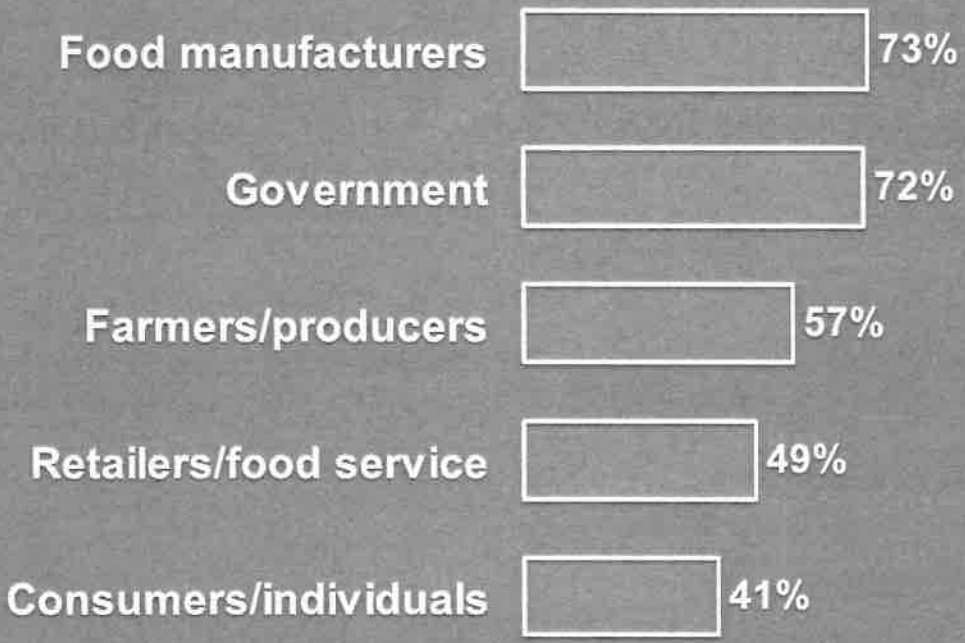
Perceived Responsibility for the Safety of the U.S. Food Supply



In general, who do you believe is responsible for food safety in the U.S.? *Select all that apply.* (n=1064)

NOTE: "Other" response (one percent) not shown

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Person-on-the-Mall Speaks....

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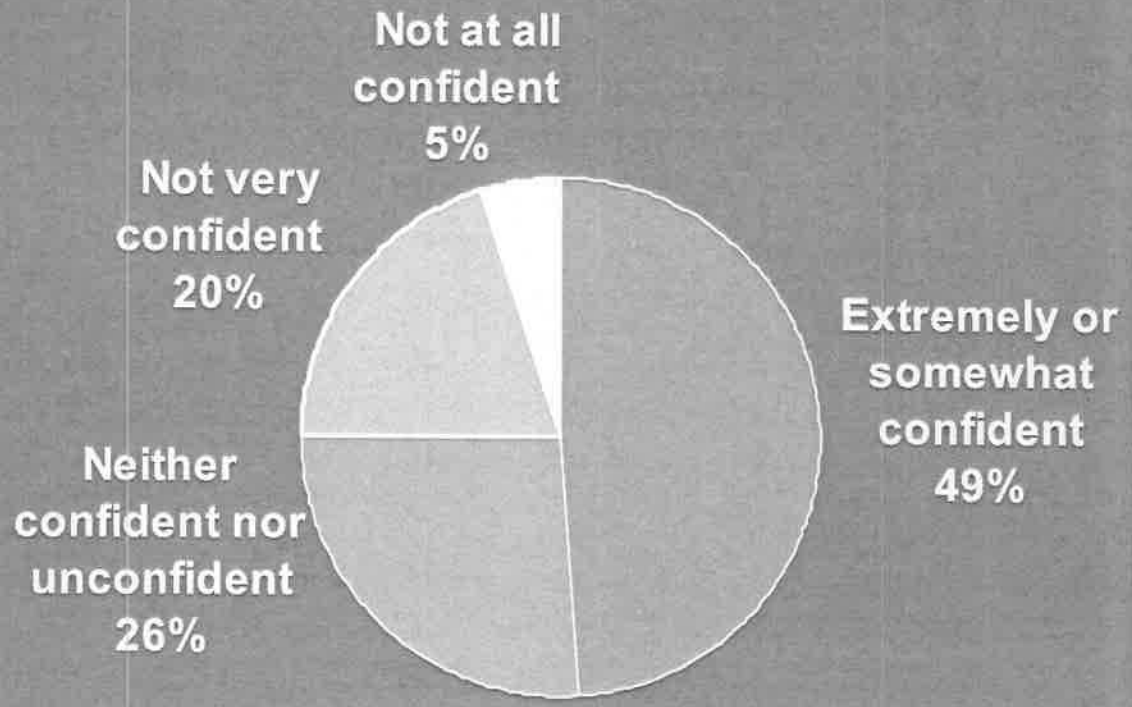
Nearly Half of Consumes are Confident in the Safety of the U.S. Food Supply.



To what extent, if at all, are you confident in the safety of the U.S. food supply? (n=1064)

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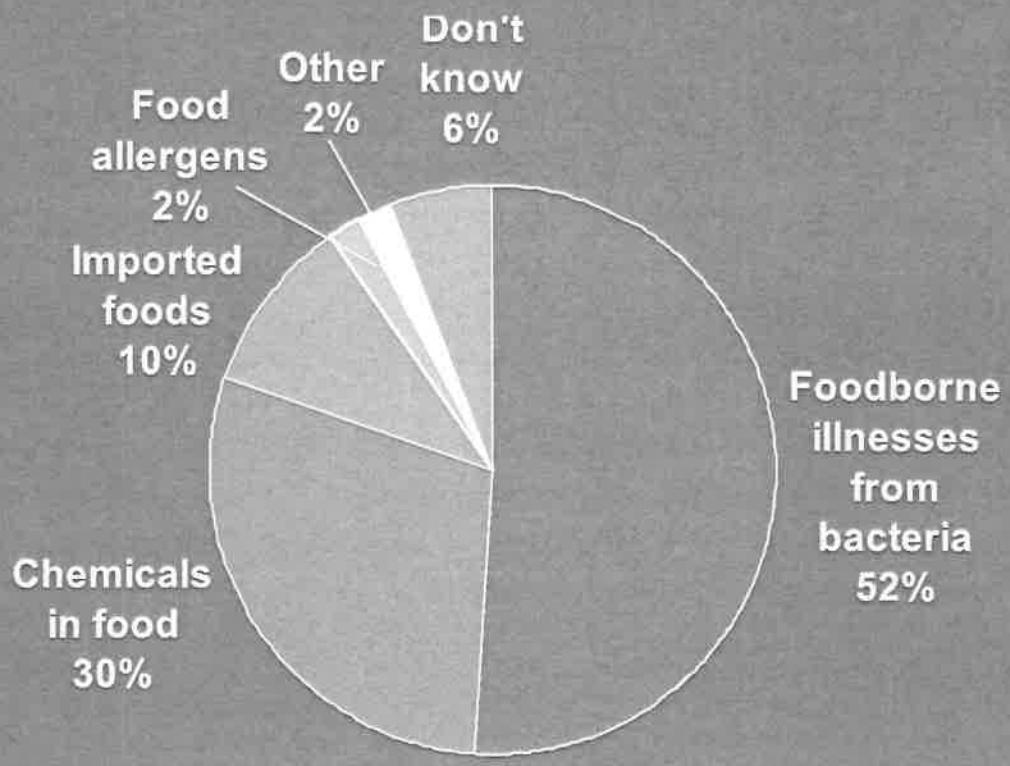
Foodborne Illness is Perceived as the Most Important Food Safety Issue.



What, in your opinion, is the most important food safety issue today? *Select one.* (n=1064)*

*Total does not add to 100% due to rounding

▲ / ▼ Significant increase/decrease from year indicated

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Safe Food Preparation




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Fewer Americans are Taking Food Safety Precautions to Reduce Their Risk

Wash my hands with soap and water	87% ▼ '08
Wash cutting board(s) with soap and water or bleach	77% ▼ '08
Cook to required temperature (such as 165 degrees F for poultry	71% ▼ '08
Properly store leftovers within 2 hours of serving	69% ▼ '08
Separate raw meat, poultry and seafood from ready-to-eat food products	63% ▼ '08
Use different or freshly cleaned cutting boards for each product (such as raw meat/poultry/produce)	50%
Use a food thermometer to check the doneness of meat and poultry items	25%
None of the above	5%

Which of the following actions do you perform regularly when cooking, preparing, and consuming food products? *Select all that apply.* (n=1064)

▲ / ▼ Significant increase/decrease from year indicated

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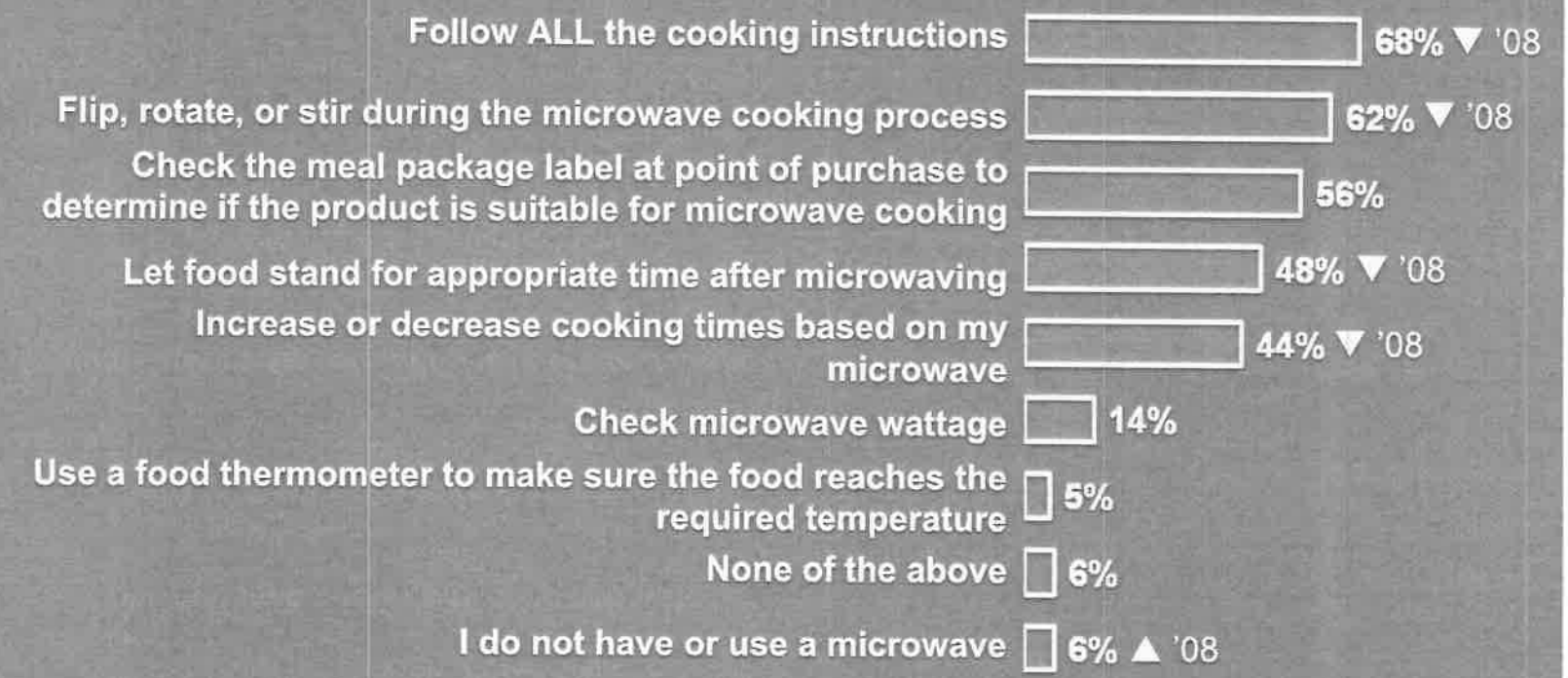
Fewer Consumers Adhering to Some Safe Microwave Cooking Practices

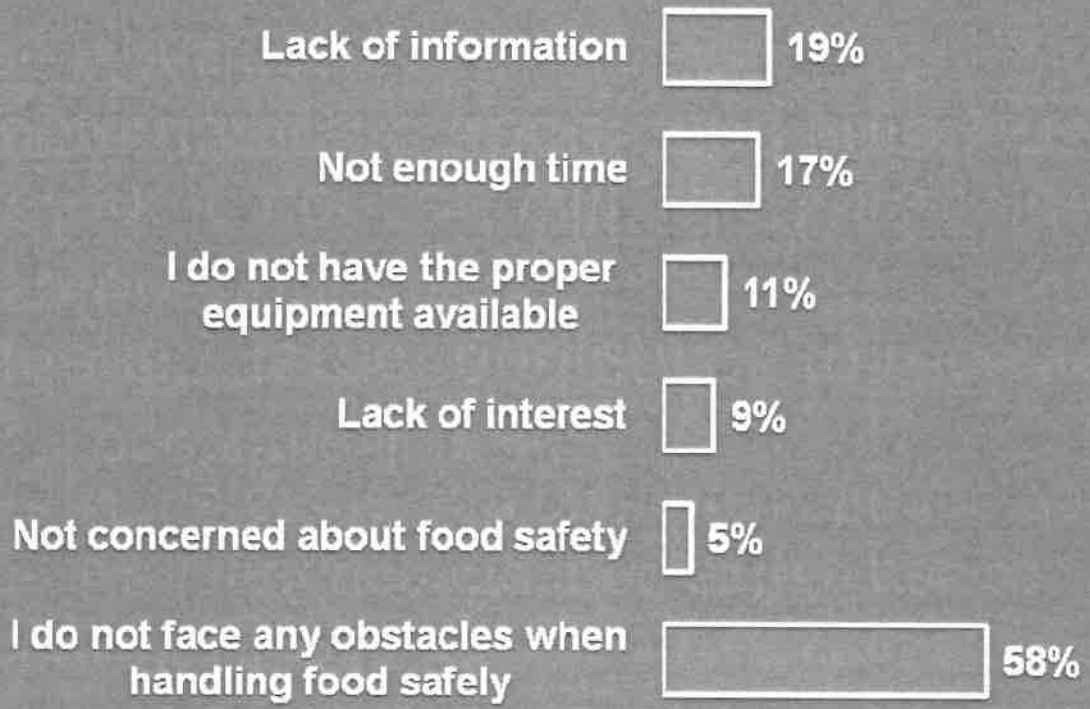


Which of the following actions do you perform regularly when preparing microwavable meals (e.g., frozen meals, pre-packaged meals that contain cooking instructions) at home? *Select all that apply.* (n=1064)

▲ / ▼ Significant increase/decrease from year indicated

2009 Food & Health Survey

The Majority of Americans do not Perceive Any Obstacles to Handling Food Safely.



What obstacles, if any, do you face when handling food safely? *Select all that apply.* (n=1064)

NOTE: "Other" response (one percent) not shown

▲ / ▼ Significant increase/decrease from year indicated

2009 Food & Health Survey

International Food Information Council Foundation

A Call to Action

- Increase awareness about general food safety practices
  - Re-introduce the food safety basics
  - Understand and communicate the risks of foodborne illness
- Identify target audiences to receive food safety education
- Work in concert with all stakeholders to communicate about food safety risks
  - Consumers
  - Industry
  - Government
  - Academics
  - Retailers

Please dial in to the audio portion: **(800) 658-3095**
Access Code: **964856914#**

Please submit your questions via email to
foodandhealth@ific.org

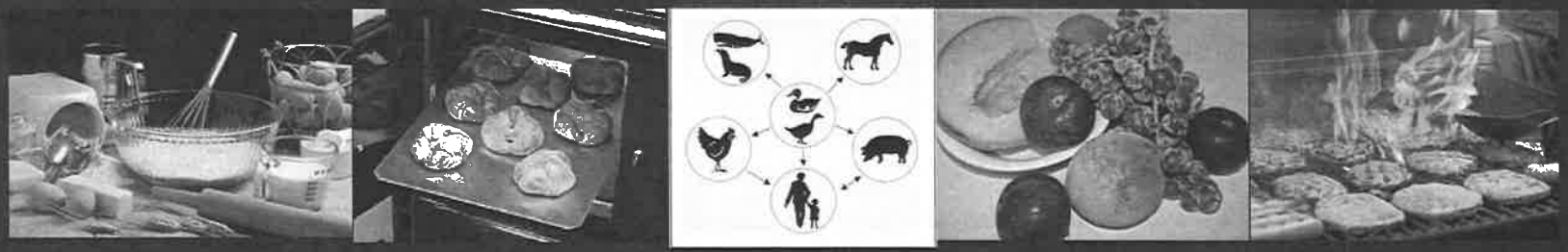
Risks of Foodborne Illness

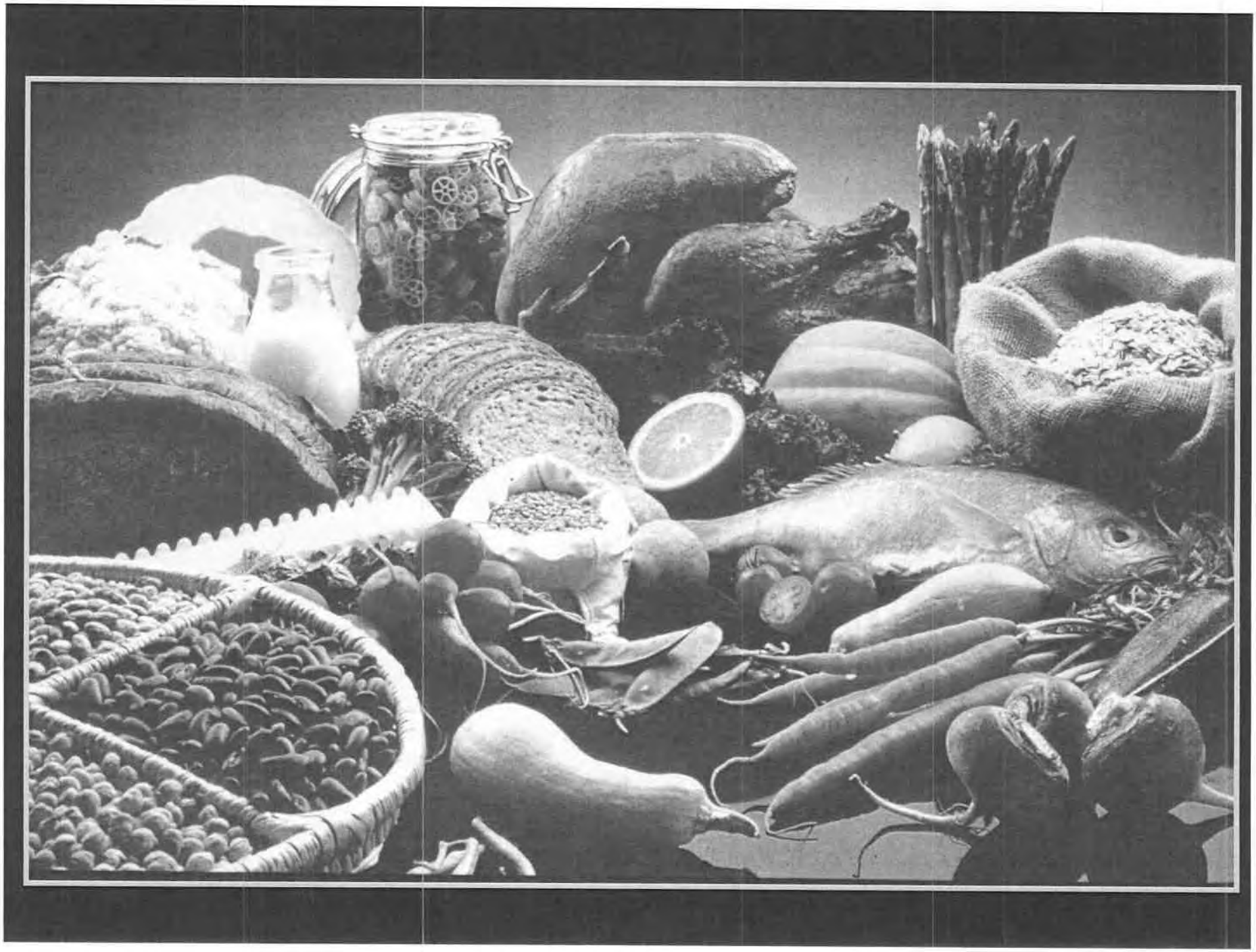
By

Robert B. Gravani, Ph.D.

Department of Food Science

Cornell University





Today's consumers are more concerned about food quality and safety than ever before !

Foodborne Illness in the U.S.

Every year foodborne illnesses result in an estimated:

- **76 Million cases of illness**
- **325,000 people hospitalized**
- **5,000 needless deaths each year**
- **Economic losses ~ $10 - $83 Billion**

Sources: CDC & Food Code

Foodborne Illness in the U.S.

What do these numbers really mean?

- 1 in 4 Americans will develop foodborne illness
- 1 in 1000 people will be hospitalized due to the symptoms of foodborne illnesses

Foodborne Outbreaks, 2007

Confirmed Etiology	No. Outbreaks	No. Cases
Bacterial	257	6,410
Chemical	34	141
Parasitic	5	65
Viral	199	6,120

CDC
SAFER • HEALTHIER • PEOPLE™

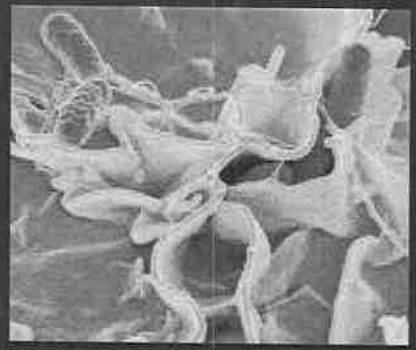
Foodborne Outbreaks, 2007

All Etiologies	No. Outbreaks	No. Cases
Total Confirmed	507	13,138
Total Suspected	226	2,944
Unknown	362	5,079
Grand Total	1097	21,183

CDC
SAFER • HEALTHIER • PEOPLE™

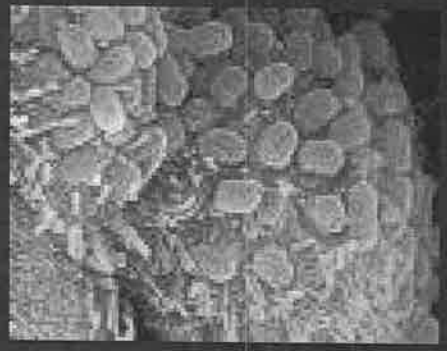
Bacterial Pathogens of concern

* *Salmonella*



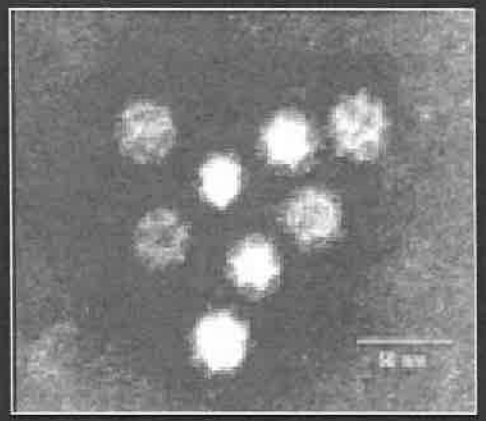
* *Listeria*

* *E. coli* O157:H7

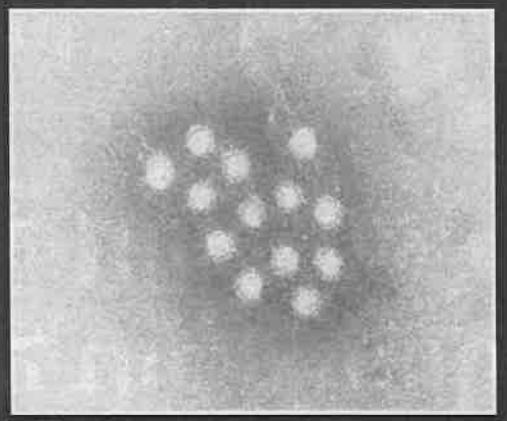


Viral Foodborne Pathogens

* *Norovirus*



* *Hepatitis A*

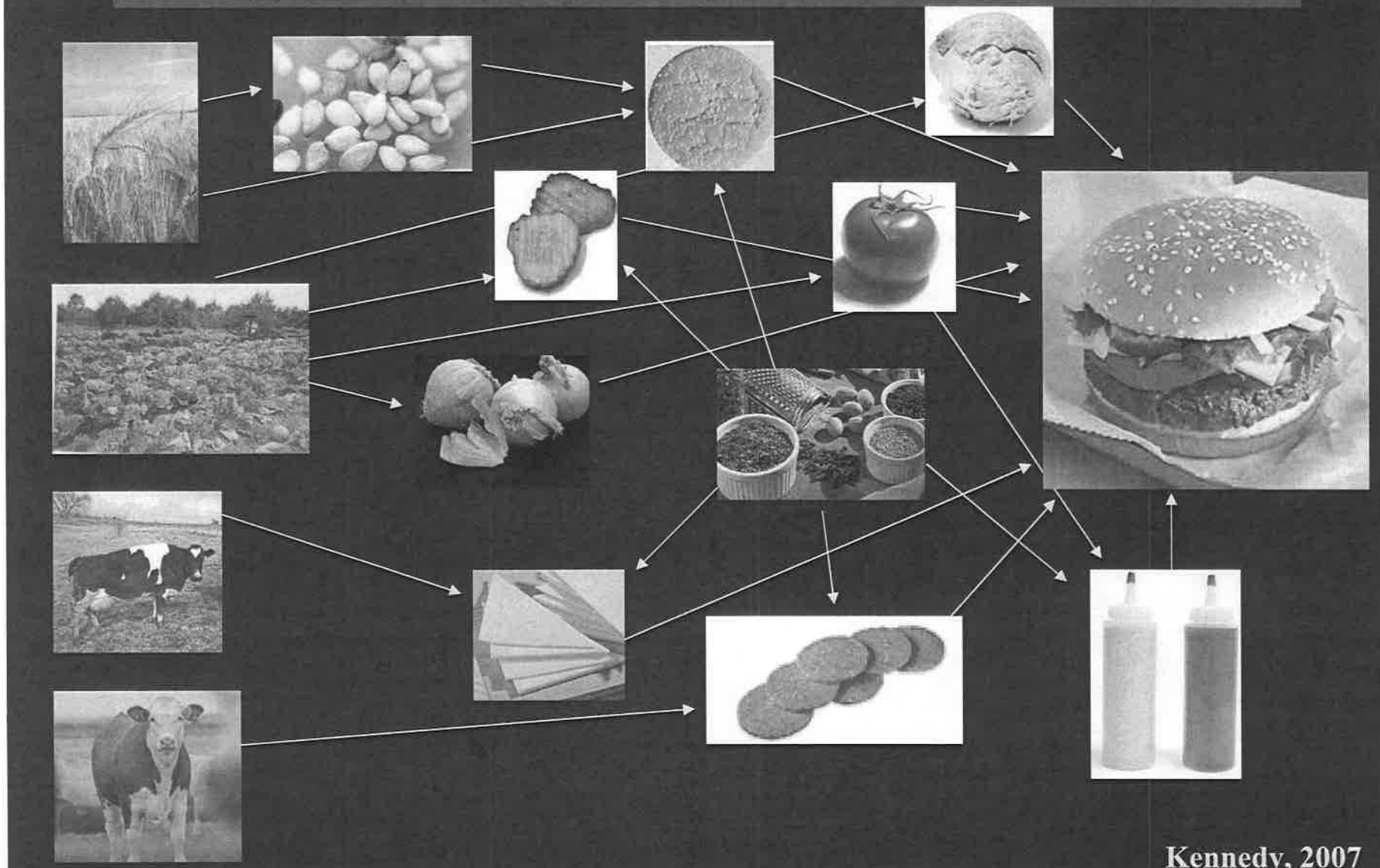


Factors Affecting the Number of Foodborne Illnesses

- Increases in elderly and chronically ill persons
- Changes in consumer practices: awareness of hazards, risks and hygiene

- Changes in eating habits
- International travel
- Globalization of the food industry

Supply Chain Complexity



Kennedy, 2007

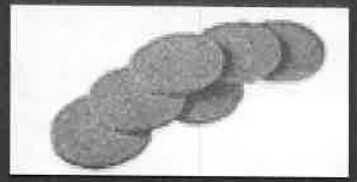
Cheeseburger Supply Chain

bleached wheat flour
malted barley flour
thiamine
riboflavin
Niacin
folic acid
reduced iron
Water
corn syrup
sesame seeds
soybean oil
Yeast
Salt
calcium sulfate
calcium carbonate
calcium silicate
soy flour

baking soda
wheat gluten
calcium propionate
enzymes
mono- and diglycerides
diacetyl tartaric acid esters
ethanol
sorbitol
polysorbate 20
potassium propionate
sodium stearoyl lactylate
corn starch
ammonium chloride
ammonium sulfate
calcium peroxide
ascorbic acid
azodicarbonamide

Milk
Water
sodium citrate
sodium phosphate
artificial color
acetic acid
Enzymes

milkfat
cream
salt
sorbic acid
cheese culture
soy lecithin
starch



USDA inspected beef

lettuce

dehydrated onions

<u>Grill Seasonings</u>
Salt
Pepper
cottonseed oil
soybean oil

<u>Special Sauce</u>
Soybean oil
distilled vinegar
egg yolks
sugar
corn syrup
spice extractives
xanthan gum
prop. glycol alginate
potassium sorbate
garlic powder
caramel color
Turmeric
EDTA

pickles
water
HF corn syrup
onion powder
spice
salt
mustard flour
sodium benzoate
mustard bran
hydrolyzed proteins
paprika
calcium disodium

Cucumbers
water
Vinegar
Salt
calcium chloride
Alum
Natura lflavorings
polysorbate 80
turmeric

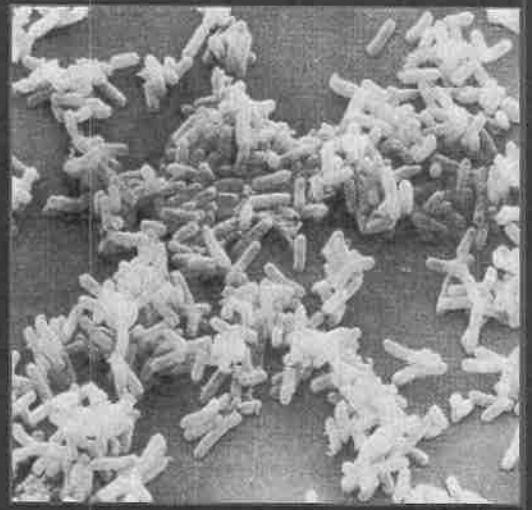
Kennedy, 2007

Factors Affecting the Number of Foodborne Illnesses

- Changes in food processing technologies
- Improved diagnostics and medical advances
- Increases in food prepared outside the home: multiple handling of food
- Changes in microorganisms: Microbial Evolution

Changes in Microorganisms

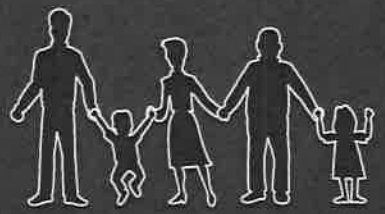
- Fewer cells needed to cause illness & some of the illness are more severe



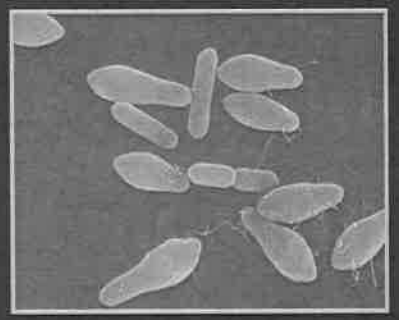
- Severe complications can result
- Adaptation to environmental conditions

The Cycle of Infection

HOSTS



PATHOGENS



ENVIRONMENT

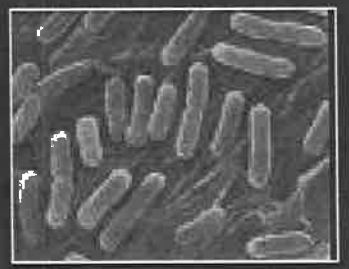
Factors Increasing the Risk of Foodborne Infection or the Severity of Illness

- ✓ Microbial
- ✓ Host
- ✓ Environmental

CAST, 1994

Microbial Factors

- Type and strain of pathogen ingested
  - * Some pathogens & strains are more virulent than others



- Quantity of pathogens ingested
  - * High numbers ingested may increase severity of illness &/or shorten onset time

CAST, 1994

Host Factors

- Age less than 5 years
  - * Lack of developed immune system
  - * Smaller infective dose-by-weight required

- Age greater than 50 or 60 years (depending on pathogen)
  - * Immune systems failing; weakened by chronic illnesses

CAST, 1994

Host Factors

- Pregnancy
  - * Altered immunity during pregnancy
- Hospitalized persons
  - * Individuals already debilitated by illness
- Concomitant Infections

CAST, 1994

Host Factors

- Immuno-compromised individuals… including those on chemotherapy or radiation therapy; recipients of organ transplants taking immuno-compromising drugs; persons with leukemia, AIDS, or other illnesses

CAST, 1994

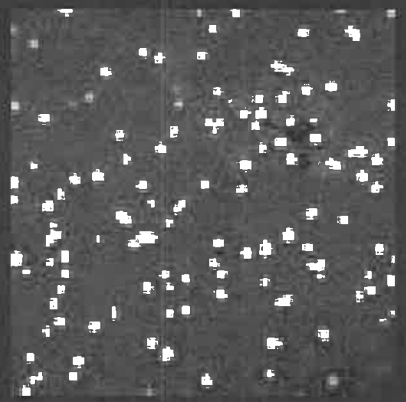
Persons with AIDS and Late-Stage HIV

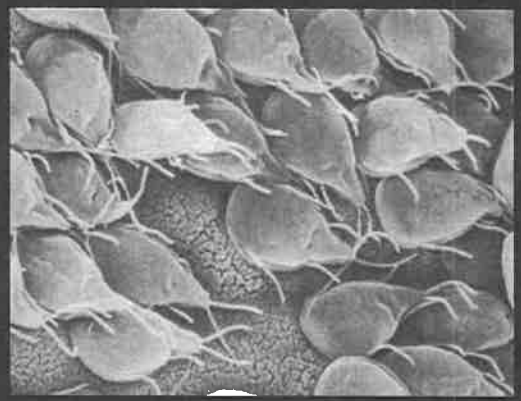
- Salmonellosis
  - * 20 times higher risk
  - * 6 times more likely to develop life-threatening blood disease
- Listeriosis
  - * 200-300 times higher risk
  - * 25% of cases end in death

FDA

Environmental Factors

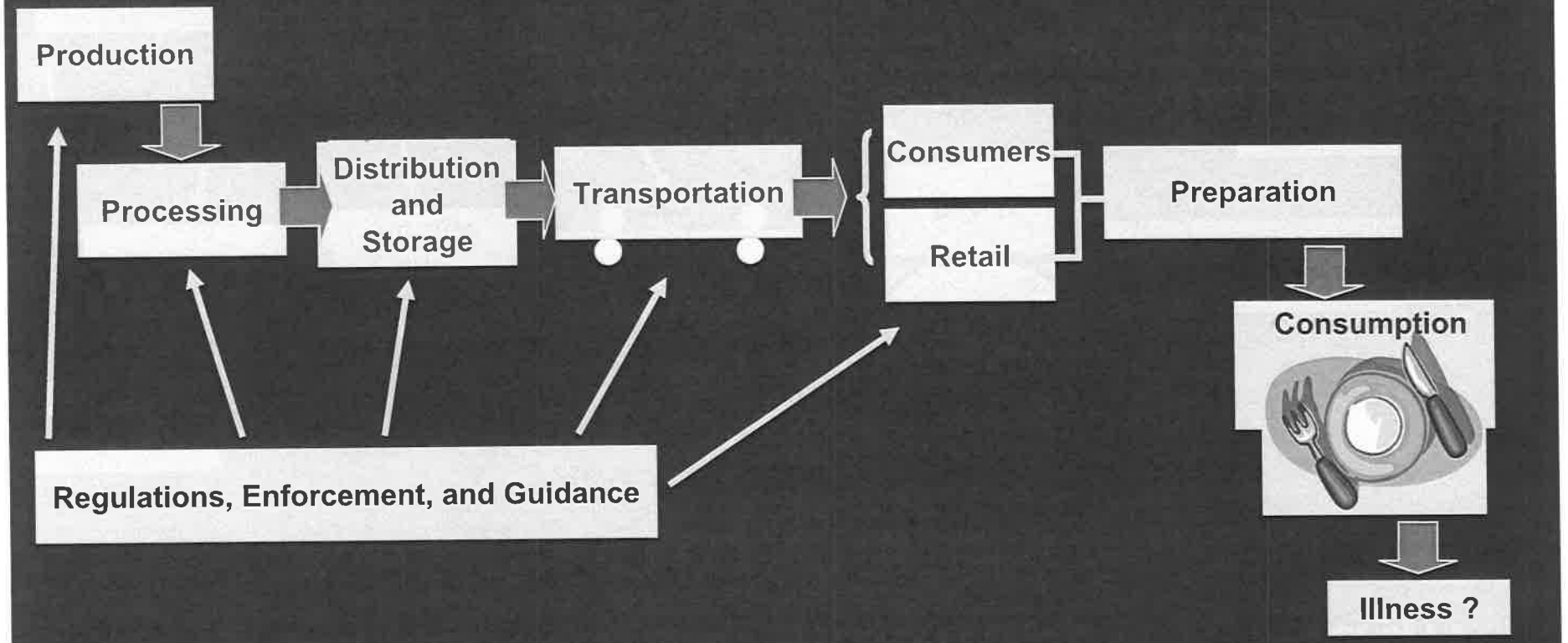
- Geographic location
  - * Exposure to virulent strains of pathogens
  - * Varied distribution of organisms in water, food & soil





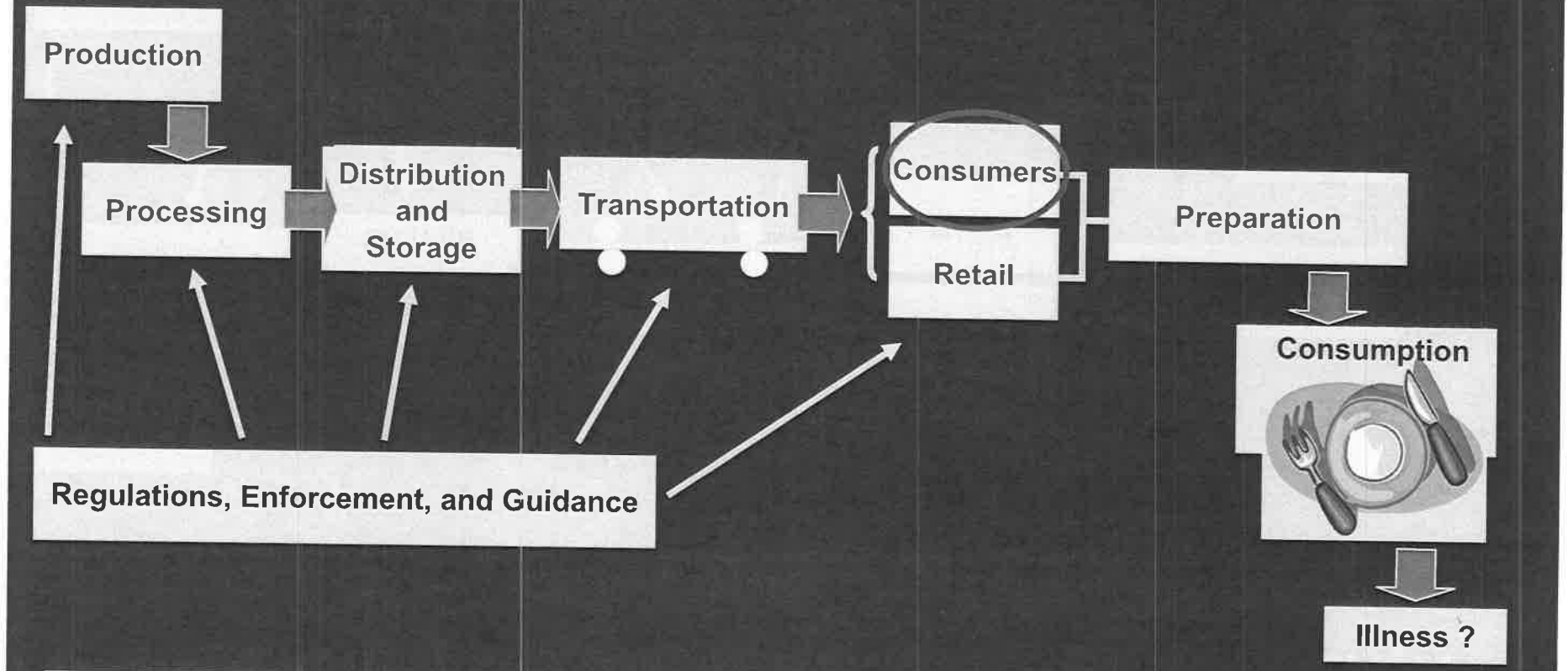
CAST, 1994

Reducing Foodborne Illness through Prevention

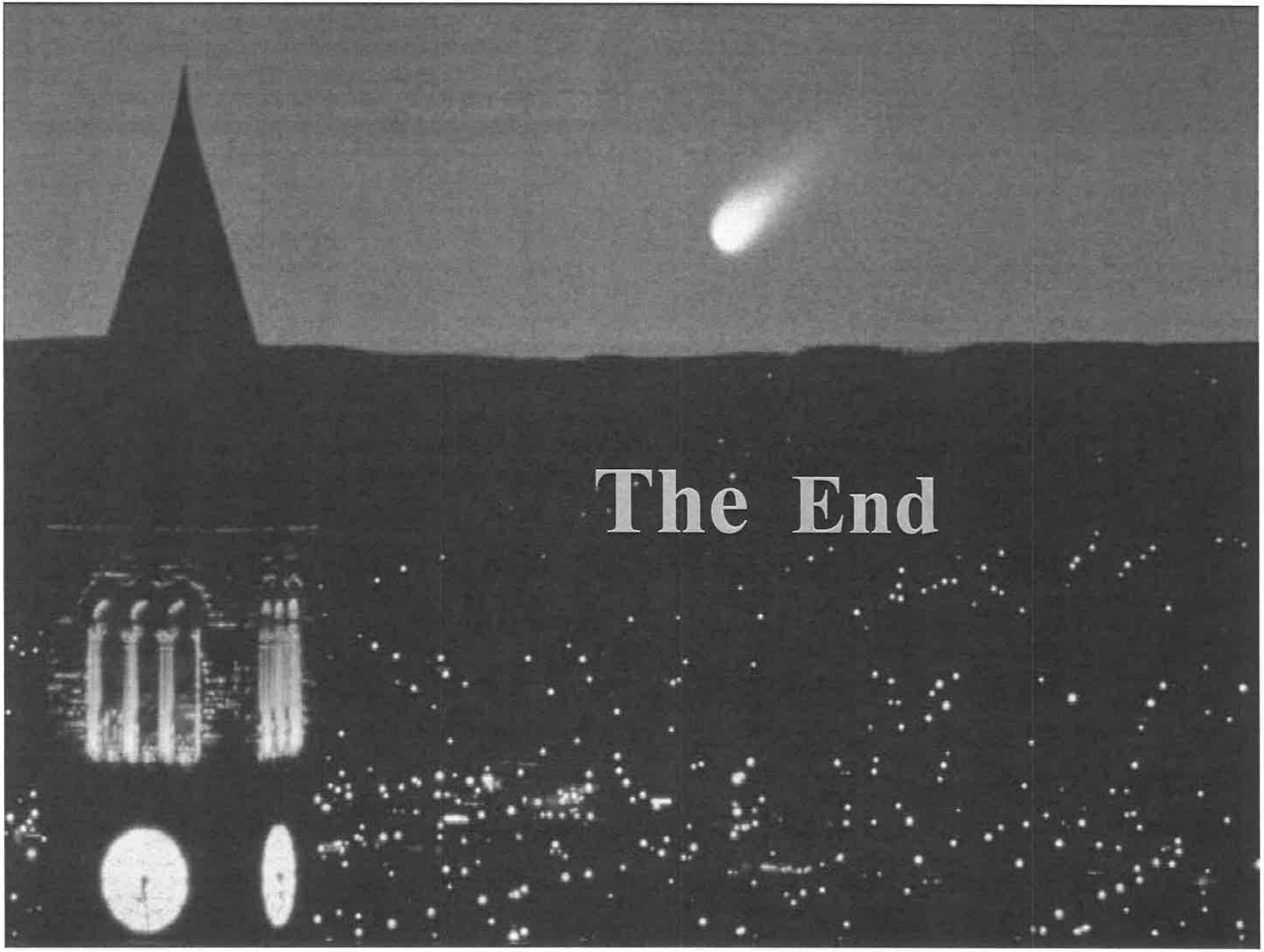


Adapted from CDC

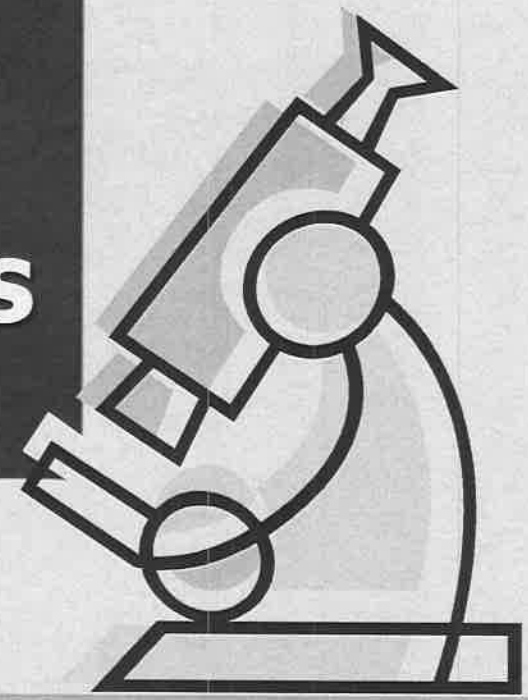
Reducing Foodborne Illness through Prevention



Adapted from CDC


The End

Food Safety Thrives When You Focus on



USDA

Diane Van, Manager
USDA Meat and Poultry Hotline

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Access Code: **964856914#**
Please submit your questions via email to
foodandhealth@ific.org

Three major causes of foodborne illness in the United States are

-- Not cooking food to a safe temperature.
-- Holding food at an unsafe temperature.
-- Lack of hand washing.

The mission of USDA's Food Safety and Inspection Service (FSIS) is to protect the health of Americans and prevent foodborne illness.

Food Safety Thrives When You Focus on Five

1. Purchasing

2. Cooking

3. Holding

4. Separating

5. Cleaning

Please dial in to the audio portion: **(800) 658-3095**
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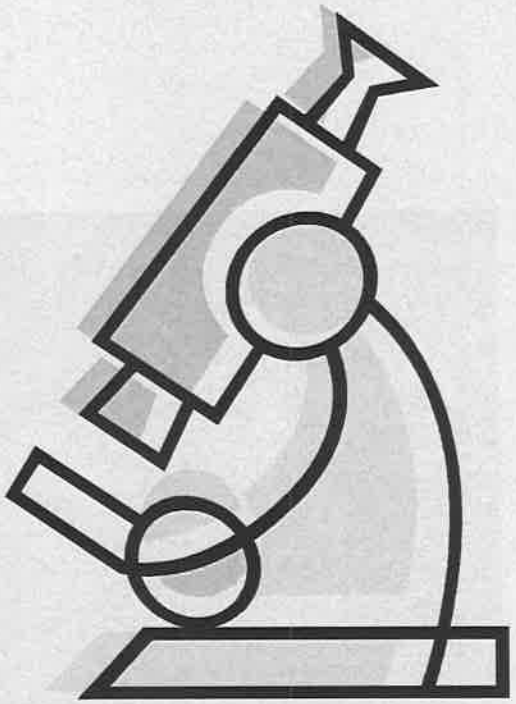


Purchasing

1 Food Safety Thrives When You Focus on Five

Buy Refrigerated Perishables

Make sure meat, poultry and seafood products -- whether raw, pre-packaged, or from the deli -- are refrigerated when purchased.

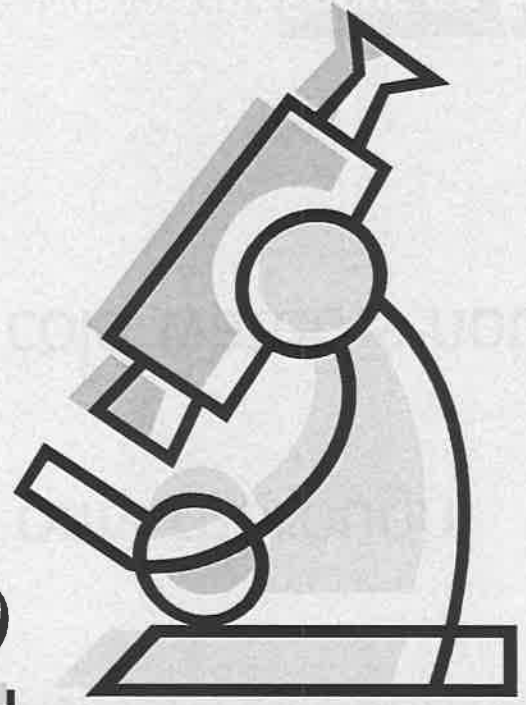
Purchase meat, poultry, seafood and other perishable products last. Keep packages of raw meat, poultry and seafood separate from other foods,
particularly foods that will be eaten without further cooking.

No Dents

Canned goods should be free of dents, cracks or bulging lids.

Go Directly Home

Plan to drive directly home from the grocery store. You may want to take a cooler with ice for perishables. Always refrigerate perishable food within 2 hours. Refrigerate within 1 hour when the temperature is above 90 °F.



Cooking

2 Food Safety Thrives When You Focus on Five

S.M.I.T.

Always cook to

"Safe Minimum Internal Temperatures."

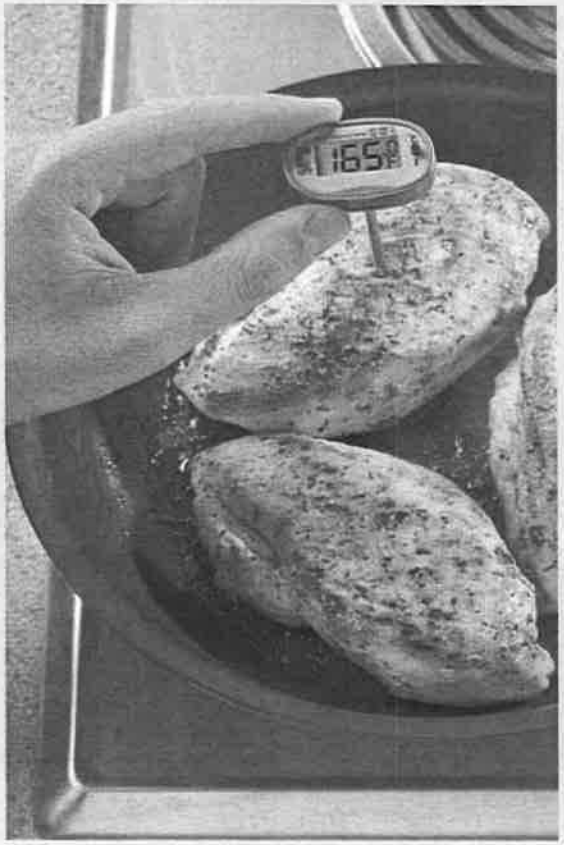
If harmful bacteria are present, only thorough cooking will destroy them.

Freezing or rinsing the foods in cold water is not sufficient to destroy bacteria.

Use a Food Thermometer

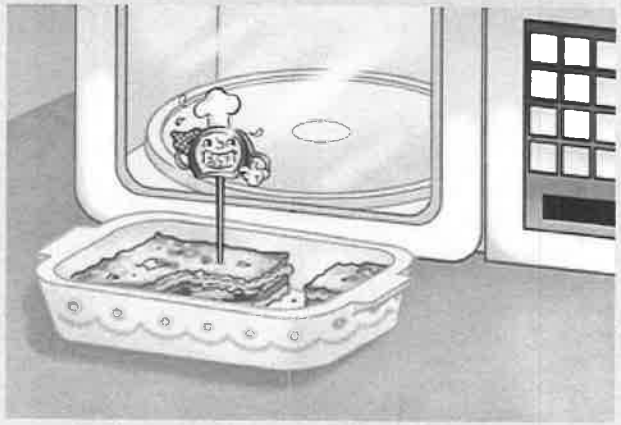
to determine if your meat, poultry or casserole has reached a safe minimum internal temperature. Check the product in several spots to assure that a safe temperature has been reached.

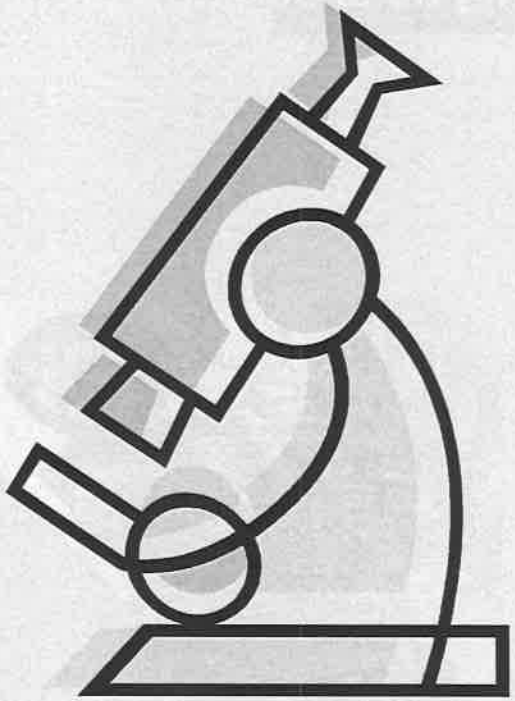
Color is not a reliable indicator of safely cooked foods.



When Microwaving Foods

- Carefully follow manufacturers instructions.
- Know your microwave wattage. Wattage makes a difference in the amount of time needed to cook food to a safe internal temperature.
- Use microwave-safe containers, cover, rotate, and allow for the standing time, which contributes to thorough cooking.



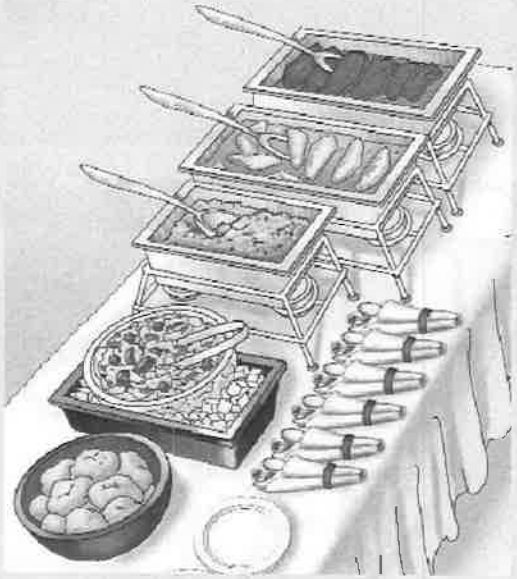


3 Holding

Holding

Hold hot foods at or above 140 °F and cold foods 40 °F or below.

Keep hot food hot with chafing dishes, slow cookers, and warming trays.



Keep Food Cold

- Nest dishes in beds of ice.
- Or use a series of small serving trays and replace them often. Replace empty platters — don't refill them.
- Discard food that has been sitting out and may have been handled by many people.

Don't Leave Food Out

Never leave foods, raw or cooked, at room temperature longer than 2 hours.

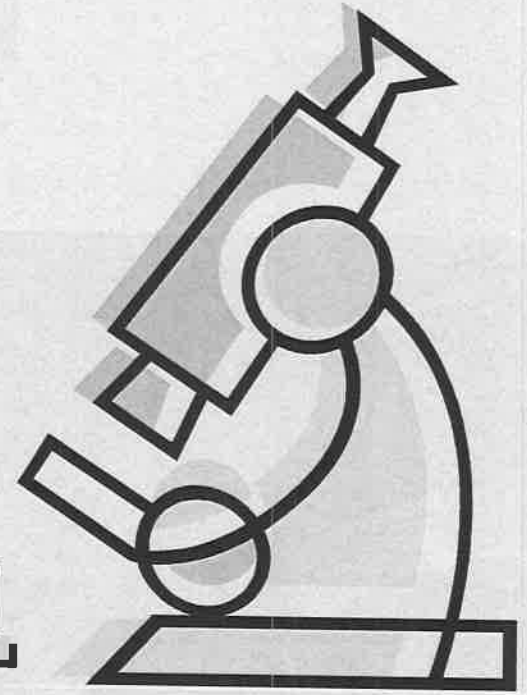
On a hot day with temperatures above **90 °F**, this decreases to 1 hour.

Reheat to 165 °F

Do not use chafing dishes, slow cookers, and warming trays for reheating. They're just for maintaining the reheated temperature.

Make sure to reheat foods to at least 165 °F.

Bring soups, sauces and gravies to a rolling boil.



4 Separating

Food Safety Thrives When You

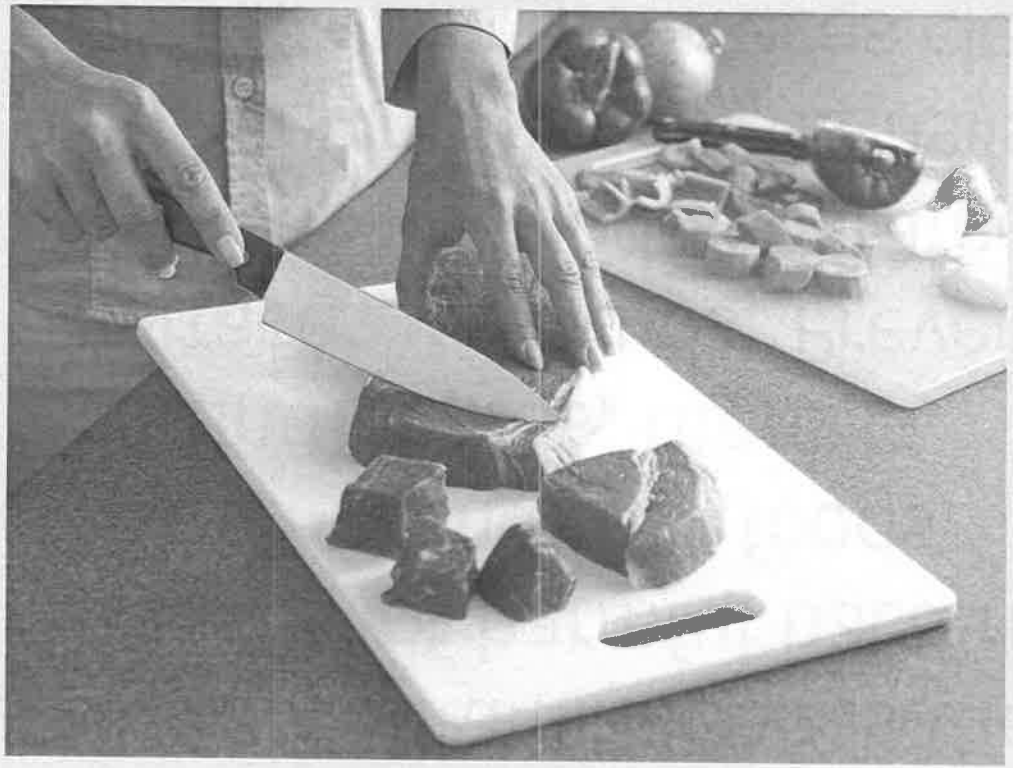
4

Focus on Five

Cross-contamination

Cross-contamination is the transfer of harmful bacteria to a food from other foods, cutting boards, utensils, surfaces, or hands. Prevent it by keeping food separated and by keeping hands, utensils, and food handling surfaces clean.

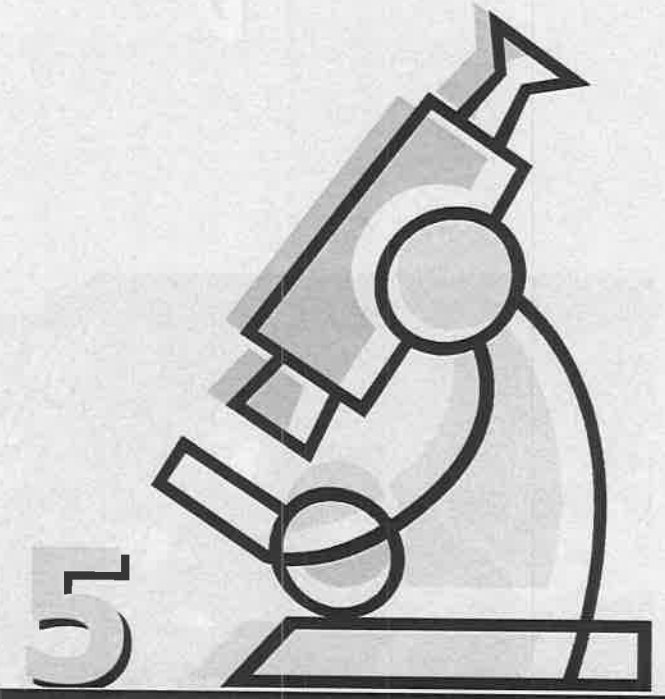
Cross-contamination



Use one cutting board for fresh produce — and a separate one for raw meat, poultry, and seafood.

Separate

Separate foods in your grocery cart. Keep raw meat, poultry, seafood, and their juices away from other food to further prevent the possibility of cross-contamination.



Cleaning

Food Safety Thrives When You Focus on Five

5

Cleaning

Always wash hands with soap and warm water for 20 seconds

- Before beginning food preparation,
- After handling food, and
- After using the bathroom, changing diapers, or touching pets.

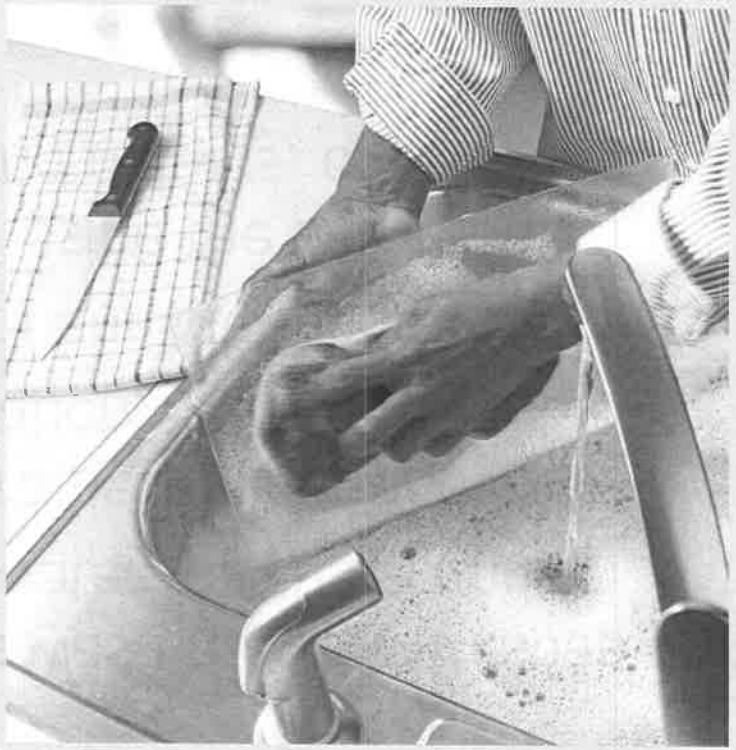
Not washing hands is a major cause of foodborne illness.

Wash

- Wash counters, equipment, utensils, and cutting boards with soap and hot water immediately after use.

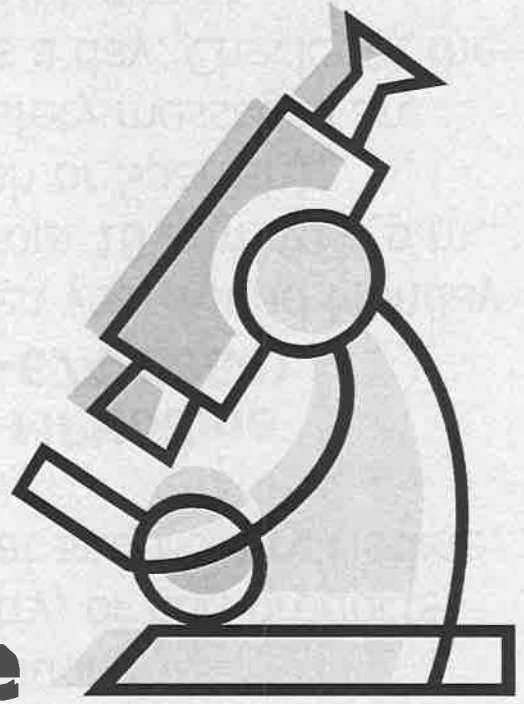
- Sanitize with a chlorine solution of 1 tablespoon of unscented, liquid chlorine bleach in 1 gallon of water.

- Let the solution stand on the board after washing, or follow the instructions on sanitizing products.



Protect Your Family

- PURCHASE
- COOK
- CHILL
- SEPARATE
- CLEAN



Food Safety Thrives When You Focus on Five

1	2	3	4	5

Call the USDA Meat & Poultry Hotline

If you have a question about meat, poultry, or egg products, call the USDA Meat and Poultry Hotline toll free at

1-888-MPHotline
(1-888-674-6854)

The Hotline is open year-round Monday through Friday from 10 a.m. to 4 p.m. ET (English or Spanish). Recorded food safety messages are available 24 hours a day. Check out the FSIS Web site at

www.fsis.usda.gov.

Send E-mail questions to **MPHotline.fsis@usda.gov.**

Food Safety Thrives When You Focus on Five

Ask Karen!

FSIS' automated response system can provide food safety information 24/7.

Also, live "chat" now available.

Look for the Spanish version of "Ask Karen" in 2010.

AskKaren.gov

Diane Van, Manager
USDA Meat and Poultry Hotline




Questions and Answers

Please submit your questions via email to
foodandhealth@ific.org




Upcoming Web Casts:

November: ***Food & Health Survey Series***
Part 4: **"Americans' Approach to Dieting and Weight Management Strategies"**

Food & Health Survey Report Available on IFIC.org and Foundation Publications Store!

The Executive Summary is available at
http://www.ific.org/research/foodandhealthsurvey.cfm

Full Report is available for purchase at
http://www.ificpubs.org/servlet/Detail?no=47

Data Tables and Comparative Topline are available for purchase at
http://www.ificpubs.org/servlet/Detail?no=48

Twitter hashtag: #foodsafety

CPE Credit

- Registered Dietitians can download a Certificate of Completion for ONE CPE credit at:

http://www.ific.org/adacpe/2009fandhsurveycpe.cfm

International Food Information Council (IFIC) Foundation

2009 FOOD & HEALTH SURVEY

Consumer Attitudes toward Food, Nutrition & Health

A Trended Survey

For more information:
International Food Information Council Foundation
1100 Connecticut Avenue NW, Suite 430
Washington, DC 20036
Web: http://ific.org

For inquiries: foodandhealth@ific.org

INTERNATIONAL
FOOD INFORMATION
COUNCIL FOUNDATION

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Research Note

Frequency of Inadequate Chicken Cross-Contamination Prevention and Cooking Practices in Restaurants†

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ABSTRACT

This study was conducted by the Environmental Health Specialists Network (EHS-Net) of the Centers for Disease Control and Prevention. The purpose was to examine restaurant chicken preparation and cooking practices and kitchen managers' food safety knowledge concerning chicken. EHS-Net members interviewed managers about chicken preparation practices in 448 restaurants. The study revealed that many restaurants were not following U.S. Food and Drug Administration Food Code guidance concerning cross-contamination prevention and proper cooking and that managers lacked basic food safety knowledge about chicken. Forty percent of managers said that they never, rarely, or only sometimes designated certain cutting boards for raw meat (including chicken). One-third of managers said that they did not wash and rinse surfaces before sanitizing them. Over half of managers said that thermometers were not used to determine the final cook temperature of chicken. Only 43% of managers knew the temperature to which raw chicken needed to be cooked for it to be safe to eat. These findings indicate that restaurant chicken preparation and cooking practices and manager food safety knowledge need improvement. Findings from this study could be used by food safety programs and the restaurant industry to target training and intervention efforts to improve chicken preparation and cooking practices and knowledge concerning safe chicken preparation.

Poultry is the fourth most common commodity associated with foodborne illness and the number one commodity associated with deaths from foodborne illness in the United States *(7)*. These facts indicate that poultry is a significant food safety problem in the United States.

Surveillance data indicate that during 1998 through 2008, 61% (376) of foodborne illness outbreaks linked with poultry were also linked with restaurants or delicatessens *(1)*. Additional data indicate that eating chicken outside the home (e.g., in a restaurant) is an important risk factor for foodborne illness. Case-control studies have revealed that consumption of chicken outside the home is linked with *Salmonella* Enteritidis *(4, 5)* and *Campylobacter jejuni (2, 3)* infections. These findings indicate that improvement of restaurant chicken preparation and cooking practices is needed, because proper preparation and cooking can help prevent foodborne illness and outbreaks.

Foodborne illness linked with chicken can be caused through cross-contamination from raw chicken to ready-to-eat (RTE) foods or the environment, such as food contact surfaces and equipment. Cross-contamination often occurs during raw chicken preparation. For example, a restaurant outbreak investigation revealed that cross-contamination from raw chicken to chopped cilantro and a cutting board used for cooked chicken led to an outbreak of *Salmonella* infections among restaurant customers *(8)*. Contaminated chicken can also cause foodborne illness when the chicken is not cooked to a temperature high enough to kill foodborne pathogens on or in the chicken. For example, a restaurant outbreak investigation revealed that undercooked chicken liver pâté caused a *Campylobacter* infection outbreak among restaurant customers *(6)*.

The U.S. Food and Drug Administration (FDA) Food Code provides the basis for state and local food codes that regulate retail food establishments in the United States. This Code contains regulatory guidance aimed at preventing cross-contamination in restaurants. This guidance includes the following prescriptions: properly clean food contact surfaces (includes washing, rinsing, and sanitizing), minimize bare hand contact with food that is not in RTE form (e.g., raw chicken), and separate raw animal foods (e.g., raw chicken) from other foods (e.g., RTE foods) *(10)*. The FDA Food Code also contains guidance aimed at ensuring that

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raw meat and poultry are cooked to a high enough temperature to kill foodborne pathogens. Specifically, the Food Code states that raw chicken should be cooked to 165°F (73.9°C) or above for at least 15 s and that final cook temperatures should be routinely measured with a thermometer to ensure that the correct temperature is reached *(10, 11)*. The Food Code also states that persons in charge (e.g., managers) of retail establishments should be knowledgeable about various food safety topics, including adequate temperatures for safe cooking of potentially hazardous foods such as raw chicken *(12)*.

Information about how chicken is prepared in restaurants and about manager knowledge of safe chicken preparation are essential to the development of effective interventions. However, relatively little information exists on these topics. This study was undertaken to fill the data gap. Specifically, the study was conducted to assess the frequency of chicken preparation practices linked with cross-contamination prevention and proper cooking and to assess manager knowledge of safe chicken preparation. Where appropriate, we discuss results of the study in the context of the FDA Food Code.

MATERIALS AND METHODS

This study was conducted by the Environmental Health Specialists Network (EHS-Net), a network of environmental health specialists and epidemiologists focused on the investigation of environmental factors that contribute to foodborne illness. EHS-Net is a collaborative project of the Centers for Disease Control and Prevention (CDC), the FDA, the U.S. Department of Agriculture (USDA), and state and local health departments. In 2008, when this study was conducted, the state and local EHS-Net sites were in California, Connecticut, Georgia, Iowa, Minnesota, New York, Oregon, Rhode Island, and Tennessee.

The study protocol was cleared by the CDC Institutional Review Board and the appropriate review boards at the EHS-Net sites. All data collectors (EHS-Net environmental health specialists) participated in training designed to increase data collection consistency.

Data collectors obtained data from approximately 50 restaurants at each EHS-Net site. Restaurants were defined as establishments that prepare and serve food or beverages to customers but that are not institutions, food carts, mobile food units, temporary food stands, supermarkets, restaurants in supermarkets, or caterers. Restaurants that did not cook raw chicken (i.e., chicken that has not undergone a kill step and needs further cooking to reduce pathogens to a level unable to cause adverse health outcomes) were excluded from the study.

To request participation in the study, data collectors called restaurants randomly selected from lists of restaurants located in predefined geographical areas of each of the EHS-Net sites. When the manager agreed to participate in the study, the data collector arranged for an on-site interview with a kitchen manager and an observation of chicken preparation. Only one restaurant from any given regional or national chain was included for each EHS-Net site. For example, if chain A had three restaurants at an EHS-Net site, only one of those would be eligible to participate at that site. Only English-speaking managers were interviewed. Data collection was anonymous, i.e., no data that could identify individual restaurants or managers were collected.

Once on site, data collectors interviewed the manager about restaurant characteristics and raw chicken preparation and cooking practices. Preparation questions focused on cross-contamination prevention. Topics included policies and practices concerning cleaning of food contact surfaces used to prepare raw chicken, use of disposable (single use) gloves when preparing raw chicken to minimize bare hand contact, use of cutting boards designated for only raw chicken to separate chicken from other foods, and washing or rinsing raw chicken before preparation. For some questions, managers were asked to rate the frequency with which they engaged in specific practices on a 5-point Likert scale ("never," "rarely," "sometimes," "often," and "always").

TABLE 1. *Data on restaurant demographics obtained from interviews with 448 kitchen managers*

Interview item	No. of responses	% of total
Restaurant ownership		
Independent	299	66.7
Chain	149	33.3
Menu description		
American	268	59.8
Italian	49	10.9
Mexican	46	10.3
Asian	46	10.3
Other	39	8.7

Cooking questions focused on the use of thermometers to determine final cook temperatures and whether thermometers were calibrated (i.e., checked for accuracy). The interview also included several food safety knowledge questions. Data collectors also collected observational data on chicken preparation and cooking; these data are not presented here.

We conducted univariate analyses to obtain descriptive statistics on the variables of interest. Data from Likert scale questions were collapsed into two categories ("never," "rarely," and "sometimes" in one and "often" and "always" in the other). Analysis was conducted with SPSS 19 statistical software (SPSS Inc., Chicago, IL).

RESULTS

Restaurant characteristics. Kitchen managers in 448 restaurants agreed to participate in the study. Seventy-four percent of contacted managers agreed to participate (this percentage is based on data from eight of the nine EHS-Net sites; participation rate data were unavailable for one site). Most managers said that their restaurant was independently owned and served an American menu (Table 1). The median number of meals served daily in these restaurants was 200 (25th percentile, 100 meals; 75th percentile, 400 meals).

Cross-contamination prevention. More than 90% of managers said that their restaurant had a cleaning policy regarding food contact surfaces used to prepare raw chicken (Table 2). When asked to describe their cleaning policies, more than 80% of managers said that they washed, rinsed, and sanitized food contact surfaces, as recommended by the FDA. However, some managers also described cleaning policies that did not meet FDA recommendations. These managers said that they washed and rinsed surfaces but did not sanitize them (10%), that they used only sanitizer (e.g.,

TABLE 2. *Data on restaurant chicken cross-contamination prevention practices obtained from interviews with 448 kitchen managers*[a]

Interview item	No. of responses	% of *N*
Is there a cleaning policy regarding food contact surfaces that have been used to prepare raw chicken? ($N = 412$)[b]		
No	38	9.2
Yes	374	90.8
What is the policy for how food contact surfaces are cleaned and/or sanitized after they have been used for raw chicken? ($N = 374$)[c]		
Wash, rinse, sanitize	312	83.4
Wash, rinse, do not sanitize	38	10.2
Only sanitize	17	3.8
Wipe surfaces with towel stored in sanitizer solution	107	28.6
Wipe surfaces with towel not stored in sanitizer solution	15	4.0
Other[d]	45	12.0
How often are single-use (disposable) gloves used during the preparation of raw chicken? ($N = 446$)		
Never, rarely, sometimes	126	25.3
Often, always	320	71.7
How often do you designate certain cutting boards for raw meat only? ($N = 424$)		
Never, rarely, sometimes	171	40.3
Often, always	253	59.7
How often is chicken washed or rinsed before preparation? ($N = 444$)		
Never, rarely, sometimes	258	58.1
Often, always	186	41.9

[a] Number of total responses differs for each question because of missing data or skip patterns in the interview.
[b] This question was answered only by managers of restaurants in which raw chicken was prepared in some way before cooking.
[c] Participants were able to provide multiple responses to the question; thus, the number of responses add to more than 374 and the percentages add to more than 100.
[d] Examples of this response are "bleach, wash, rinse" and "rinse and sanitize."

sanitizer spray bottle) (4%), and that they only wiped surfaces using a towel that was (29%) or was not (4%) stored in sanitizer solution. One-quarter of managers said that disposable gloves were never, rarely, or only sometimes used during the preparation of raw chicken. Forty percent said that they never, rarely, or only sometimes designated certain cutting boards for raw meat only (including chicken). Forty-two percent said that raw chicken was often or always washed or rinsed before preparation.

Cooking. Forty-six percent of managers said that food workers used the FDA-recommended method of taking temperatures with a thermometer to determine when chicken had reached its final cook temperature (Table 3). However,

TABLE 3. *Data on restaurant chicken cooking practices obtained from interviews with 448 kitchen managers*[a]

Interview item	No. of responses	% of *N*
How do food workers determine when chicken has reached its final cook temperature? ($N = 448$)[b]		
Chicken's appearance	211	47.1
Chicken's feel or touch	124	27.7
Timer	128	28.6
Experience, skill	70	15.6
Thermometer	205	45.8
Other[c]	53	11.8
How often are thermometers calibrated? ($N = 202$)		
At least once per day	56	27.7
At least once per week	44	21.8
At least once per month	30	14.9
Less than once per month	3	1.5
Never	23	11.4
Other[d]	12	5.9
Unsure	34	16.8

[a] Number of total responses differs for each question because of missing data or skip patterns in the interview.
[b] Participants were able to provide multiple responses to the question: thus, the number of responses add to more than 448 and the percentages add to more than 100.
[c] Examples of this response are "cook till the chicken floats in the fryer" and "sauce gets thick."
[d] Examples of this response are "sometimes" and "when needed."

54% of managers also reported other methods to determine final cook temperature. These methods included chicken's appearance (47%), chicken's feel or touch (28%), a timer (29%), and experience and skill (46%). Of the managers who reported using a thermometer, 28% said thermometers were never calibrated or were calibrated less often than once per week. Seventeen percent of managers did not know how often their thermometers were calibrated.

Manager knowledge. When asked to what temperature raw chicken needed to be cooked for it to be safe to eat, 43.3% (194) of managers correctly reported the temperature recommended by the FDA (165°F [73.9°C]). One-quarter (24.6%, 110) of managers provided a temperature that was below 165°F (median, 155°F [68.3°C]; minimum, 90°F [32.2°C]; maximum, 160°F [71.1°C]), and 24.8% (111) provided a temperature that was above 165°F (median, 180°F [82.2°C]; minimum, 168°F [75.6°C]; maximum, 500°F [260°C]). Only 7.4% (33) of managers said that they did not know the answer to this question. Managers were also asked which two pathogens among *Salmonella*, *Campylobacter*, *Staphylococcus aureus*, and *Escherichia coli* O157:H7 were most likely to contaminate their kitchens during raw chicken preparation. One correct response, *Salmonella*, was given by 86.4% (387) of managers, and the other correct response, *Campylobacter*, was given by 15.2% (68). Only 13.6% (61) correctly reported that

both *Salmonella* and *Campylobacter* were likely to cause contamination. Almost 17% (16.5%, 74) chose *S. aureus*, and 56.9% (255) chose *E. coli* O157:H7; these pathogens are not typically associated with chicken. Almost 10% (9.6%, 43) of managers said that they did not know the answer to the question.

DISCUSSION

These results suggest that some restaurants are not engaging in practices that could reduce the potential for pathogen cross-contamination from raw chicken to the environment or food. Although most managers said that their restaurants followed FDA-recommended policies for cleaning surfaces used to prepare raw chicken, which require washing, rinsing, and sanitizing, some managers also described policies that did not follow this FDA guidance, such as only wiping surfaces with a towel. These findings are concerning, because it is difficult to prevent cross-contamination when food contact surfaces are not adequately washed, rinsed, and sanitized.

Some managers said that they did not often use disposable gloves during preparation of raw chicken or that they did not designate specific cutting boards for use only with raw meat. Although cross-contamination from raw chicken can be prevented without the use of disposable gloves or designated cutting boards, implementing these preventive practices can make it easier to prevent cross-contamination and consequently reduce foodborne illness risk.

Over half of the managers said that they rinsed or washed raw chicken. This process may reduce pathogens on the chicken but can also create cross-contamination (e.g., through rinse water splashing onto other food or the environment). USDA regulations allow poultry processing facilities to rinse poultry as long as the process does not create cross-contamination *(9)*. Currently, no regulations or guidelines have been published for retail food establishments regarding rinsing raw chicken. Rinsing of raw chicken may be appropriate for restaurants if they adequately address the cross-contamination risk associated with this practice. In this study, we did not assess restaurant practices concerning cross-contamination prevention during rinsing of raw chicken. Because our results suggest that this practice is widespread, future food safety research and intervention efforts should address this topic.

The findings from this study also suggest that some restaurants are not engaging in practices that could help ensure adequate cooking of chicken. The use of thermometers to determine the final cook temperature of chicken is a crucial step for ensuring that chicken is safe to eat. The fact that some managers reported using methods other than thermometers (e.g., chicken's appearance) to determine adequate cooking is a matter of concern. However, taking the temperature of every piece of chicken cooked is not always necessary. When a restaurant has established standard operating procedures that have been tested and verified to ensure that they consistently address food safety hazards, the restaurant can rely on those procedures *(13)*. Some of the restaurants in our study may have had such procedures in place related to cooking chicken; we did not collect data on this topic. A relatively small percentage of managers said that the thermometers they used to determine the final cook temperature of chicken were not often or never calibrated. However, even this small percentage is concerning because uncalibrated thermometers may give inaccurate temperature readings, leading to undercooked chicken.

Findings from this study suggest that manager knowledge concerning the temperature to which raw chicken needed to be cooked for it to be safe to eat is lacking. One-quarter of managers thought that the temperature to which raw chicken needed to be cooked was lower than the FDA-recommended minimum temperature. When managers do not know this minimum temperature, it seems unlikely that this standard will be met. Another one-quarter of managers thought that the temperature to which raw chicken needed to be cooked was higher than the FDA-recommended temperature. Although cooking chicken to a temperature higher than the recommended minimum temperature is safe, the fact that many managers believed that chicken needed to be cooked to these higher temperatures demonstrates these managers' lack of current food safety knowledge about chicken.

Most managers knew that *Salmonella* was commonly associated with raw chicken. However, few knew that *Campylobacter* was also commonly associated with raw chicken. Chicken contamination prevention practices do not generally vary by pathogen; basic steps to address the risk of pathogen contamination would be effective for both *Campylobacter* and *Salmonella*. However, simple knowledge of pathogens associated with food is not enough to ensure safe food; steps must be taken to prevent exposure to these pathogens. Our data suggest that manager knowledge of pathogens associated with food is not necessarily associated with effective prevention practices.

A limitation of our study is that the data were collected through self-report methods and thus may be susceptible to a bias toward overreporting socially desirable behaviors, such as preparing chicken properly. Data were collected from English-speaking managers only; thus, our data may not represent the proportion of kitchen managers who are not English speakers.

The data reported here indicate that knowledge and practices associated with restaurant chicken preparation and cooking need improvement. Findings from this study could be used by food safety programs and the restaurant industry to target training and intervention efforts to improve chicken preparation and cooking practices. Training and intervention efforts could focus on the more worrisome findings from this study, such as inadequate cleaning and sanitizing practices and lack of knowledge concerning the recommended final cook temperature of chicken. Intervention efforts could also focus on identifying and addressing the barriers to safe chicken preparation and cooking practices.

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Consumer-Reported Handling of Raw Poultry Products at Home: Results from a National Survey

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ABSTRACT

Salmonella and *Campylobacter* cause an estimated combined total of 1.8 million foodborne infections each year in the United States. Most cases of salmonellosis and campylobacteriosis are associated with eating raw or undercooked poultry or with cross-contamination. Between 1998 and 2008, 20% of *Salmonella* and 16% *of Campylobacter* foodborne disease outbreaks were associated with food prepared inside the home. A nationally representative Web survey of U.S. adult grocery shoppers (n = 1,504) was conducted to estimate the percentage of consumers who follow recommended food safety practices when handling raw poultry at home. The survey results identified areas of low adherence to current recommended food safety practices: not washing raw poultry before cooking, proper refrigerator storage of raw poultry, use of a food thermometer to determine doneness, and proper thawing of raw poultry in cold water. Nearly 70% of consumers reported washing or rinsing raw poultry before cooking it, a potentially unsafe practice because "splashing" of contaminated water may lead to the transfer of pathogens to other foods and other kitchen surfaces. Only 17.5% of consumers reported correctly storing raw poultry in the refrigerator. Sixty-two percent of consumers own a food thermometer, and of these, 26% or fewer reported using one to check the internal temperature of smaller cuts of poultry and ground poultry. Only 11% of consumers who thaw raw poultry in cold water reported doing so correctly. The study results, coupled with other research findings, will inform the development of science-based consumer education materials that can help reduce foodborne illness from *Salmonella* and *Campylobacter*.

Of the estimated 48 million cases of foodborne infections each year in the United States, nontyphoidal *Salmonella* spp. and *Campylobacter* spp., respectively, cause 1 million and 0.8 million foodborne infections *(45, 46)*. In addition, *Salmonella* and *Campylobacter* contribute, respectively, to 35 and 15% of all hospitalizations and 28 and 6% of all deaths related to foodborne infections *(45, 46)*. Poor food handling practices in the home could be responsible for approximately one-fifth of all foodborne disease in the United States *(21)*.

Most cases of salmonellosis and campylobacteriosis are associated with eating raw or undercooked poultry, cross-contamination, and abusive storage and handling conditions of poultry across the farm-to-table continuum *(8, 9, 54)*. *Salmonella* and *Campylobacter* can be easily transferred from raw poultry to hands and kitchen surfaces *(1, 8, 10–12, 14, 16, 19, 20, 23, 33, 34, 37, 39, 40, 43, 58, 59)* and can persist on food contact surfaces for extensive periods of time, which may lead to increased risks of cross-contamination among food handlers, food contact surfaces, and ready-to-eat foods *(15, 23)*.

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To estimate the weighted percentage of consumers who follow recommended food safety practices when handling raw poultry, a nationally representative Web survey of U.S. adult grocery shoppers was conducted. Findings from this study will inform the development of science-based consumer education materials that can help reduce foodborne illness from *Salmonella* and *Campylobacter*.

MATERIALS AND METHODS

The survey administration and analysis procedures are described below. RTI International's Committee for the Protection of Human Subjects (RTI International, Research Triangle Park, NC), which serves as RTI's Institutional Review Board, reviewed and approved the study protocol.

Sample. The survey sample was selected from KnowledgePanel, a Web-enabled panel developed and maintained by GfK Custom Research (New York, NY). The panel is constructed using a probabilistic address-based sampling method that uses probability-based sampling of addresses from the U.S. Postal Service's Delivery Sequence File. The Web-enabled panel is statistically representative of the U.S. population *(49)*. The address-based sampling approach provides coverage of 97% of all U.S. households, including Internet and non-Internet households and cell phone–only households. Individual panelists who do not have a computer or Internet access are provided free basic laptops with

Internet access in exchange for participating on the panel. All new panel members complete an initial survey that collects information on demographic characteristics to create member profiles, which can be used for sample selection and weighting.

At the time of sample selection, approximately 50,000 panel members were actively participating in the Web-enabled panel. A sample of 4,531 adult panel members who had primary or shared responsibility for their household grocery shopping was randomly selected to receive the survey.

Survey procedures and response. The survey was e-mailed to a random sample of panel members aged 18 years and older who had primary or shared responsibility for their household grocery shopping. Selected panel members were eligible to complete the survey if they did at least half of the household grocery shopping and had prepared raw poultry and eggs in the past 30 days. Data were collected over a 14-day field period in September 2013. On average, respondents completed the survey in 14 min. To maximize response rate, two e-mail reminders were sent to nonrespondents. Of the 4,531 sampled panelists, 2,686 individuals responded to the survey, for a completion rate of 59.2%. Of these individuals, 1,504 were eligible and completed the survey, for a qualification rate of 56.0%. A subset of respondents ($n = 406$) who prepared raw ground poultry in the past 3 months completed the series of questions on raw ground poultry.

Questionnaire. The questionnaire asked respondents three series of questions to collect information on reported behavior the last time they purchased and prepared (i) raw poultry, whole or parts (e.g., whole turkey or chicken, turkey or chicken breasts, or other chicken parts, such as legs or thighs); (ii) raw ground poultry; and (iii) shell eggs. These questions were based on the four core safe food handling practices established by the Be Food Safe campaign of the U.S. Department of Agriculture (USDA) and the Partnership for Food Safety Education (PFSE)—clean, separate, cook, and chill (www.befoodsafe.org)—and on the USDA's cold storage recommendations *(55)*. When possible, we used or modified questions from existing surveys to develop the instrument. For example, several questions were taken from the 2010 Food Safety Survey *(57)*. For most questions, respondents were asked to select an item from a list of responses to avoid cuing respondents to the "correct" response. To encourage respondents to report their actual behavior rather than their usual behavior and to help minimize biases associated with self-reporting and social desirability response, the questionnaire asked respondents about preparation and handling practices for the last time they purchased or handled raw poultry, raw ground poultry, or shell eggs. This article presents the survey results regarding consumer handling of raw poultry (whole or parts) and raw ground poultry.

Because this survey was administered 1 month after a press release received extensive national media attention and announced "... an average of 90 percent of the [U.S.] population washes chicken" *(17)*, we asked respondents if they had read or heard anything about washing raw poultry in the past few weeks.

Prior to survey administration, the survey instrument was evaluated with six adults using cognitive interviewing techniques *(60)*. After participants completed the survey, an interviewer asked each participant to provide his response and explanation for his response choice and asked whether the question or response items were confusing or difficult to understand. Subsequently, the survey instrument was refined based on the results from the cognitive interviews. Additionally, with a sample of 30 panel members from the study target population, GfK Custom Research conducted a pretest of the survey instrument. The pretest verified that the survey was functioning correctly and that respondents understood questions and response items (through open-ended questions) and estimated the survey incidence-eligibility rate, consent-cooperation rate, and the median survey length.

Analysis. The sample was independently weighted using standard poststratification weighting procedures that adjust both for any survey nonresponse and for noncoverage, resulting in demographic distributions that align with demographic benchmarks from the most recent (March 2013) Current Population Survey. The final weights were trimmed and scaled to sum to the total U.S. population of adult grocery shoppers; thus, the weighted survey results are representative of the U.S. population of adult grocery shoppers. The responses to the survey questions were analyzed to estimate the weighted percentage of adult U.S. household grocery shoppers (i.e., consumers) who followed recommended practices for raw poultry (whole or parts) and raw ground poultry. The analysis was conducted using SAS software, version 9.3 (SAS Institute Inc., Cary, NC).

RESULTS

Table 1 provides the respondents' demographic characteristics. Of the 1,504 respondents, 67.0% were female. The majority of respondents were white, non-Hispanic (70.5%), and 45 years old or older (57.5%) and had at least some college education (63.5%). Less than one-third of respondents had an individual in the household at risk for foodborne illness (e.g., adults aged 60 years or older; pregnant women; children aged 5 years or younger; or individuals diagnosed with diabetes, kidney disease, or another condition that weakens the immune system).

Table 2 presents the weighted percentage of U.S. consumers who reported following recommended food safety practices the last time they handled and prepared raw poultry (whole or parts) and raw ground poultry.

Clean. The majority of consumers who prepared raw poultry (87.8%) and who prepared raw ground poultry (90.0%) reported washing their hands after handling the raw product. The majority of consumers (68.7%) rinsed or washed raw poultry before they cooked it, a potentially unsafe practice because "splashing" of contaminated water may lead to the transfer of pathogens to other foods and other kitchen surfaces *(18)*. Twenty-two percent of consumers reported that they had read or heard something about washing raw poultry in the weeks prior to the survey. Based on what they read or heard, 57.2% of consumers reported they will continue to wash raw poultry before they cook it, 11.1% of consumers reported they stopped washing raw poultry before they cook it, 7.3% of consumers reported they plan to stop washing raw poultry before they cook it, and 18.4% of consumers reported they will continue to *not* wash raw poultry before they cook it.

Separate. While grocery shopping, the majority of consumers keep raw poultry and raw ground poultry separate from other foods; 76.3 and 69.1% of consumers reported placing raw poultry and raw ground poultry, respectively, in a separate plastic bag before putting it in their shopping cart or making an effort to keep it from

TABLE 1. *Demographic characteristics of U.S. adult grocery shoppers who handle raw poultry*

	n	Weighted %
Gender		
Female	1,003	67.0
Male	501	33.0
Age (yr)		
18–29	129	14.4
30–44	365	28.2
45–59	517	31.6
60+	493	25.9
Education		
Less than high school	72	7.8
High school	411	28.6
Some college	482	31.6
Bachelor's degree or higher	539	31.9
Race/ethnicity		
White, non-Hispanic	1,178	70.5
Black, non-Hispanic	131	10.7
Hispanic	98	11.4
2+ races, non-Hispanic	45	3.5
Other, non-Hispanic	52	3.9
Annual household income		
Less than $35,000	373	28.0
$35,000 or more	1,131	72.0
Marital status		
Married	939	58.8
Divorced	184	11.8
Never married	175	14.8
Living with partner	113	8.9
Widowed	64	3.6
Separated	29	2.1
MSA status[a]		
Metro	1,263	84.1
Nonmetro	241	15.9
At-risk individual in household[b]		
60 years or older	593	33.5
Pregnant	20	2.0
5 years of age or younger	152	12.4
Diagnosed with diabetes or kidney disease	196	12.4
Diagnosed with condition that weakens the immune system	46	3.0

[a] MSA, metropolitan statistical area.
[b] Respondents could select multiple responses.

touching other foods in their cart. In addition, 70.3 and 61.8% of consumers, respectively, reported that raw poultry or raw ground poultry was separated from other foods in their grocery bags at checkout.

Once home, however, only about 17% of consumers correctly stored raw poultry or raw ground poultry on the bottom shelf of their refrigerators in a sealed container or plastic bag. After using a cutting board or other surface to prepare raw poultry at home, 94.4% of consumers followed practices to prevent cross-contamination by either washing the cutting board or other surface with soap or bleach-disinfectant or using a different cutting board for preparing the next food product. Similarly, 97.1% of consumers either washed dishes or utensils (e.g., knife) with soap or bleach-disinfectant or used separate utensils and dishes for preparing the next food product after preparing raw poultry at home.

Cook. Sixty-two percent of consumers reported owning a food thermometer. Of food thermometer owners, the majority reported using a food thermometer the last time they cooked to check the doneness (i.e., internal temperature) of whole turkeys (73.2%) and whole chickens (56.7%). However, fewer consumers (12 to 26%) used one to measure the internal temperature of smaller cuts of poultry and ground poultry. Regarding leftovers, about 35% of consumers reported reheating leftovers until steaming hot or using a food thermometer to check the internal temperature.

Chill. According to USDA cold storage recommendations *(55)*, consumers should cook, freeze, or discard raw poultry within 1 to 2 days and freeze or discard cooked poultry within 3 to 4 days to prevent bacteria growth and, ultimately, foodborne illness. The majority of consumers safely store raw poultry (70.6 to 94.4%) and cooked poultry (90.5 to 92.8%) per USDA cold storage recommendations. About three-quarters of consumers safely thaw raw poultry either in the refrigerator, microwave, or cold water. However, of those who thaw raw poultry in cold water (n = 199), fewer than 11% put the raw poultry in a sealed container or plastic bag, submerge it in cold water, and change the water every 30 min per the USDA's recommendation. After raw poultry has been thawed in cold water, the USDA recommends cooking it immediately; about 12% of consumers do not follow this recommendation. About 90% of consumers safely refrigerate leftovers containing poultry within 2 h as recommended by the PFSE, although it is not known whether they store leftovers in shallow containers for rapid cooling as also recommended by the PFSE.

DISCUSSION

Research studies have confirmed the presence of *Salmonella*, *Campylobacter*, and other foodborne pathogens in household kitchens *(26, 30, 44)*. Although half of Americans think it is "not very common" for people in the United States to get foodborne illness because of the way food is prepared in their home *(57)*, food safety experts estimate that the home is the primary location where foodborne disease outbreaks occur *(4–7, 27, 29, 38, 48)*. Of the 11,627 reported foodborne disease outbreaks occurring in a single place of food preparation between 1998 and 2008, 1,058 were associated with food prepared in a private home *(21)*. During the same 10 years, 20% of *Salmonella* and 16% *of Campylobacter* foodborne disease outbreaks were associated with food prepared inside the home *(21)*.

Although most respondents reported washing their hands with soap and warm water after handling raw poultry, it is not known if they followed recommended hand-washing procedures, such as washing hands for at least 20 s.

TABLE 2. *Weighted percentage of U.S. consumers who follow recommended safe food handling practices for raw poultry*[a]

Recommendation	Follow safe practices					
	Raw poultry (whole and parts)			Raw ground poultry		
	n	Weighted %	SE	*n*	Weighted %	SE
Clean						
Wash hands with soap and water after handling raw poultry	1,294	87.8	1.0	406	90.0	1.7
Do not wash or rinse raw poultry	484	31.3	1.4	NA	NA	NA
Separate						
Separate raw poultry from other food in shopping cart	1,092	76.3	1.3	317	69.1	2.6
Separate raw poultry from other food in shopping bags at checkout	1,020	70.3	1.4	292	61.8	2.9
Place raw poultry in sealed container or plastic bag on bottom shelf of refrigerator	102	17.5	1.8	34	17.6	3.3
Properly wash or use separate cutting board for preparing next food	1,144	94.4	0.8	NA	NA	NA
Properly wash or use separate dishes or utensils for preparing next food	1,355	97.1	0.6	NA	NA	NA
Cook						
Own food thermometer	987	62.0	1.6	NA	NA	NA
If owned, use a food thermometer to measure internal temperature of cooked poultry *(all foods cooked by respondents)*						
Whole turkeys[b]	95	9.8	1.1	NA	NA	NA
Whole chickens[b]	572	73.2	1.9	NA	NA	NA
Turkey breasts[b]	375	56.7	2.3	NA	NA	NA
Chicken breasts or other[b]	244	42.6	2.6	NA	NA	NA
Meatloaf or similar dish containing ground poultry[b]	228	26.3	1.8	NA	NA	NA
Patties made with ground poultry[b]	NA	NA	NA	159	22.8	1.9
Reheat leftovers to 165°F or until steaming hot	NA	NA	NA	77	11.7	1.5
Chill	420	34.7	1.7	137	36.1	3.0
Cook, freeze, or discard raw poultry within 1 to 2 days (according to USDA's cold storage chart)	418	79.5	2.3	120	70.6	4.5
Freeze or discard cooked poultry within 3 to 4 days (according to USDA's cold storage chart)	1,181	92.8	0.8	349	90.5	1.8
Thaw raw poultry in the refrigerator, microwave, or cold water	648	75.3	1.7	198	74.31	3.3
To thaw raw poultry in cold water, place in a sealed container or plastic bag, submerge in cold water, and change the water every 30 min	25	11.0	2.4	3	6.3	4.2
If raw poultry is thawed in cold water or microwave, cook it immediately	259	88.1	2.7	67	89.5	5.7
If raw poultry is thawed in refrigerator, cook it within 1 to 2 days	394	94.4	1.2	124	93.0	2.1
Refrigerate leftovers within 2 h	1,160	88.4	1.1	361	90.4	2.1

[a] *n*, number of respondents; SE, standard error; NA, not applicable.
[b] Estimated percentage was calculated among respondents who were eligible and answered the question; for example, only respondents who cook meatloaf or similar dishes containing ground poultry answered whether they used a food thermometer to determine doneness the last time they cooked the food.

Likewise, most respondents reported following recommended practices to avoid cross-contamination when using cutting boards and utensils; however, it is not known how well these items were washed. Adherence to these recommendations is better assessed through observational studies.

The USDA recommends that consumers do not wash or rinse raw poultry *(51)* because "splashing" of contaminated water may lead to the transfer of pathogens to other foods and other kitchen surfaces *(18)*. In addition, because washing poultry does not remove pathogens, it is not a necessary practice. Everis and Betts *(18)* found that when poultry is washed or rinsed, "splashing" of contaminated water can travel up to 28 in. (71.12 cm) on either side of the sink and 20 in. (50.8 cm) in front of the sink. Additionally, they found significantly higher levels of bacteria on the surface of wet raw poultry (i.e., placed under running tap water for 10 s and held over the sink for 5 s to drain) compared with dry raw poultry, and wet raw poultry transfers significantly higher levels of bacteria to contact surfaces compared with dry poultry *(18)*. Additional peer-reviewed research is needed to support the recommendation to not wash or rinse raw poultry to avoid cross-contamination. A recent study was conducted by researchers at Drexel University, who found that about 90% of respondents surveyed in the Philadelphia area wash their chicken before cooking it *(17)* (unpublished data). The study concluded that Philadelphians most likely wash their poultry because recipes have historically called for washing poultry and because they want to "rinse the slime off of just-opened chicken. . . ."

Few consumers correctly stored raw poultry products in the refrigerator. According to the PFSE (www.befoodsafe.org), raw poultry should be placed in a sealed container or

plastic bag on the bottom shelf of the refrigerator to prevent raw juices from contaminating other foods because products left in their original packaging could potentially leak. Although about half of the survey respondents placed the raw poultry on the bottom shelf of the refrigerator as recommended, most did not place it in a sealed container or bag. Research addressing U.S. consumer cold storage practices is scarce. Koppel and colleagues *(32)* found that Asian consumers who stored raw poultry in the refrigerator were more likely to store it on the top or middle shelf of the refrigerator.

According to the USDA, poultry must be cooked to a safe minimal internal temperature of 165°F (73.9°C) to destroy bacteria. Using a food thermometer is the only reliable way to ensure that the internal temperature has been reached and any foodborne bacteria destroyed. Although the majority of consumers who own a food thermometer reported using it to determine doneness of whole turkeys or chickens, fewer than 26% of consumers reported using it the last time they cooked smaller cuts of poultry or ground poultry; however, it is not known whether the product was cooked to the recommended internal temperature of 165°F (73.9°C). Failure to use a food thermometer is a potentially unsafe practice, given that 70% of chicken pieces that were judged by consumers as "done" had not reached safe internal cooking temperatures according to a summary of food safety literature *(3, 30)*. Similarly, the 2010 Food Safety Survey *(57)*, a random-digit-dialing telephone survey of 4,568 English- or Spanish-speaking, noninstitutionalized U.S. adults, reported that 28% of consumers who own a food thermometer always or often use one to check the internal temperature of smaller cuts of poultry. Thus, the percentage of consumers using a food thermometer to check the doneness of smaller cuts of poultry seems to have remained fairly constant over the past 3 years. According to several consumer focus group studies conducted for the USDA Food Safety and Inspection Service *(31, 52, 53, 56)*, consumers generally rely on sight (e.g., the color of the meat or juices) and other senses (e.g., touch, taste) to determine the doneness of smaller cuts of poultry instead of using a food thermometer. In these focus groups, many participants thought food thermometers were unnecessary to determine whether meat and poultry was "cooked thoroughly" because they had been "cooking for years without once getting food poisoning."

Although most consumers reported safely thawing raw poultry (in the refrigerator, microwave, or cold water), as recommended by the USDA, few consumers who thaw raw poultry in cold water correctly follow the recommendation to place raw poultry in an airtight packaging or a leak-proof bag, submerge it in cold water, and change the water every 30 min to be sure it stays cold. A study conducted in The Netherlands with a small sample size found a difference between consumer knowledge and behavior related to freezing and defrosting raw meat or poultry *(13)*. For example, the study found that respondents knew that meat should be placed in an airtight package, but not all respondents actually used an airtight package *(13)*.

The survey results identified areas of low adherence to currently recommended practices, based on self-reported data, including not washing raw poultry, proper refrigerator storage of raw poultry, use of a food thermometer to determine doneness, and proper thawing of raw poultry in cold water. Additional research is needed to better understand why adherence to these recommendations is low. Possible reasons include (i) lack of awareness of the recommended practice(s), as may be the case for not washing raw poultry, or (ii) perceived barriers to following recommended practice(s), as may be the case with not using a food thermometer because it is inconvenient. Understanding the barriers to following recommended practices can help inform the development of educational materials to address these barriers *(61)*. Additionally, it is important that educational messages motivate consumers by highlighting the benefits of following recommended practices; for example, with regard to food thermometer usage, pointing out that using a food thermometer can help consumers avoid overcooking their food.

This study had a few limitations. Our study used self-reported behaviors, which may overstate actual behavior because of self-reporting and social desirability bias *(16, 19, 24, 28, 41–43, 58)*. In their review of consumer food safety practices in the home, Redmond and Griffith *(42)* compared the correspondence between self-reported practices and observed behavior. In this article, they reported that a study conducted by Anderson and colleagues *(2)* found that 87% of participants reported washing their hands all or most of the time before food preparation but that, when observed, only 45% attempted to wash their hands before preparing food.

When completing surveys, people tend to report their usual behavior rather than their exact behavior *(25)*. For example, Lessler and colleagues *(35)* found that when asked how many times they visited the dentist last year, people tended to report the usual pattern of two annual visits, rather than the number of times they actually visited the dentist. Likewise, when reporting dietary intake over a period of days or weeks, people tended to report what they think they usually eat, rather than recalling what they actually ate *(47)*. To help minimize self-reporting bias, we asked respondents to consider what they actually did the last time they prepared raw poultry or raw ground poultry at home; thus, we were more likely to elicit respondents' actual behavior instead of their knowledge of recommended food safety practices or their usual practice. Another concern when conducting surveys is social desirability bias, the tendency of respondents to report what they perceive to be the acceptable or "correct" behavior, which can overstate actual behavior *(36)*. Thus, asking about the last time the respondent prepared the product can help minimize social desirability bias as well. This approach is used in the National Longitudinal Study of Adolescent Health *(50)* to collect information on sensitive behaviors, such as sexual practices and drug use.

As with any survey, nonresponse may result in nonresponse bias. For this study, the extent of nonresponse bias was not assessed, although previous research conducted using KnowledgePanel found no association between the

type of sample (nonrespondents versus respondents) and the survey responses *(22)*.

Based on the survey findings, we conclude that education is needed to improve consumer handling practices for raw poultry to decrease illness attributed to *Salmonella* and *Campylobacter* in raw poultry products. In particular, most consumers are not following these practices currently recommended by the USDA and the PFSE: not washing raw poultry before cooking, properly storing raw poultry in the refrigerator, using a food thermometer to determine doneness of poultry (including small cuts), and properly thawing raw poultry in cold water. To elicit behavior change, food safety messages need to motivate consumers to follow recommended practices. Findings from this study will inform the development of a multifaceted consumer intervention that includes an interactive Web site, game, and mobile application and extension educational curriculum.

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Evaluation of Safe Food-Handling Instructions on Raw Meat and Poultry Products

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ABSTRACT

Every year in the United States, millions of people become ill, thousands of people die, and substantial economic costs are incurred from foodborne diseases. As a measure to prevent foodborne diseases, since July 1994, the U.S. Department of Agriculture has required that safe food-handling labels be placed on retail packages of raw or partially cooked meat and poultry products. Through selected states' Behavioral Risk Factor Surveillance System (BRFSS) interviews, survey data were collected to determine the proportion of adults aware of the label and adults who reported changing their raw meat-handling practices because of the label. Fifty-one percent of the 14,262 respondents reported that they had seen the label. Of these, 79% remembered reading the label, and 37% of persons who reported that they had seen and read the label reported changing their raw meat preparation methods because of the label. Women were more likely than men to have read the label, as were persons who are at least 30 years of age compared to younger adults ($P < 0.05$). Both label awareness and risky food-handling behaviors increased with education and income, suggesting that safe food-handling labels have limited influence on consumer practices. Our results also suggest that the labels might be more effective in discouraging cross-contamination than in promoting thorough cooking practices. We suggest that the label is only one component among many food safety education programs that are needed to inform consumers about proper food-handling and preparation practices and to motivate persons who have risky food-handling and preparation behaviors to change these behaviors.

Foodborne infections cause millions of illnesses and contribute to thousands of deaths each year in the United States *(4)*. A variety of pathogenic bacteria (e.g., *Escherichia coli* O157:H7, *Salmonella, Campylobacter*) are found in meat and poultry *(12)*. Food preparers can reduce the risk of foodborne disease by proper handling and thorough cooking of meat and poultry.

Since July 1994, the U.S. Department of Agriculture has required that safe food-handling instructions be printed on retail packages of raw meat and poultry *(6)*. These labels contain messages about time and temperature controls (e.g., cooking raw meat or poultry thoroughly, thawing these food items, and storage of leftovers) and avoiding cross-contamination of meat products and other foods (e.g., keeping meat and poultry separate from other foods and washing hands, utensils, and cutting boards after contact with raw meat or poultry). Spanish-language safe food-handling labels are currently required on raw meat and poultry products in Puerto Rico and are optional in the rest of the nation (Fig. 1). In the present survey, we examine the extent of consumers' label awareness and the influence of the label on consumer preparation practices for raw meat and poultry.

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METHODS

In 1995, questions about the safe food-handling labels on raw meat or poultry were added to the Behavioral Risk Factor Surveillance System (BRFSS) interviews in Colorado, Florida, Missouri, New York, and Tennessee. In 1996, these questions were also added to the BRFSS interviews in Indiana and New Jersey. Survey respondents were asked if they had seen label information about safe handling of raw meat or poultry; if they had seen the label, if they remembered reading anything in the label about safe handling of raw meat and poultry; and if the label information on raw meat or poultry changed the way they prepared these products. Respondents were also asked about their usual practices after handling raw meat or chicken, their usual practices after using a cutting surface for cutting raw meat or chicken, and if they had eaten pink hamburgers in the past 12 months. The BRFSS is a state-based system that surveys noninstitutionalized adults about their health behaviors and practices, using random digit dialing techniques to obtain disproportionate stratified random samples or three-stage cluster samples based on the Waksburg method *(14)*. All interviews were conducted in English.

Excluded from analyses were respondents who reported that they did not handle undercooked meat or chicken. Persons who responded that they usually "continue cooking" or they "rinse and/or wipe their hands then continue cooking" after handling raw meat or poultry were classified as persons who usually did not wash their hands after handling raw meat or chicken. Persons who

FIGURE 1. *Safe food-handling label in English (top) and Spanish (bottom).*

responded that they usually "continue using the cutting surface as is" or "rinse and/or wipe the cutting surface then continue cooking" after contact with raw meat or chicken were classified as persons who did not usually wash their cutting board after using it to cut raw meat or chicken. Persons who reported that they ate one or more "hamburgers that were still pink in the middle" in the previous 12 months were classified as pink hamburger consumers. Excluded from analyses of specific questions were respondents who stated that they "didn't know" or were "not sure" or refused to answer these questions.

Statistical Analytical Software (SAS, version 6.12) and software for Survey Data Analysis (SUDAAN, version 7.0) were used to produce weighted estimates of the prevalence of consumers' awareness of labels and the impact of these labels on consumer behavior. Estimated prevalences were based on factors that adjusted for the respondent's probability of selection and age-, race-, and sex-specific population distributions of the 1990 U.S. Census data for each state's population *(7, 8)*. Logistic regression analysis was used to evaluate the association between recognition of the safe food-handling label and safe food-handling and preparation practices. Odds ratios were calculated, after controlling for age, sex, education, and yearly salary. Age was stratified into three groups: 18 to 29 years, 30 to 59 years, and 60 years and older. Education was stratified into three categories: less than 12th grade, high school graduate, college graduate or higher. Yearly salary was divided into four strata: <$15,000; $15,000 to $34,999; $35,000 to $49,999; and >$50,000.

The variable for residential area was created by matching the county of the respondent's residence with Economic Research Service rural–urban continuum codes (i.e., Beale codes) *(3)*. Urban areas were central counties of metropolitan areas and other counties in metropolitan areas with populations of one million or more people. Suburban or small town areas were fringe counties of metropolitan areas and counties with urban populations of 20,000 people or more that were not adjacent to metropolitan areas. Rural areas were counties with urban population of 2,500 to 19,999 not adjacent to metropolitan areas or completely rural counties (total urban population less than 2,500 persons) *(10)*. The formula used to estimate the response rate was developed by the Council of American Survey Research Organizations *(2)*.

To test the null hypothesis that the prevalence of seeing label information on raw meat or poultry did not vary by increasing age or education, a Wald chi-square test was performed at a 0.05 significance level. To test the null hypotheses that there were no differences in prevalences among specified levels of a variable for seeing the label, for reading the label, and for changing raw meat preparation methods because of the label, *t* tests were performed at a 0.05 significance level *(11)*.

RESULTS

Overall, 17,246 respondents completed the questionnaire (2,461 in Colorado; 3,335 in Florida; 2,212 in Indiana; 1,572 in Missouri; 3,149 in New Jersey; 2,477 in New York; 2,040 in Tennessee). The median Council of American Survey Research Organizations response rate was 60%. A total of 2,281 respondents were excluded from analysis because they reported that they did not handle undercooked meat or chicken. Among the remaining 14,262 respondents, 51% stated that they had seen the safe food-handling label information (Table 1). Among persons who saw the label information, 79% reported reading the label information and 37% of these reported that they changed their raw meat preparation methods because of the label information.

Label awareness. Awareness of safe food-handling labels varied by state (Table 1). The proportion of respondents who remembered seeing the label information on uncooked meat or poultry was lowest in New York (42%) and highest in Missouri (59%).

The proportion of respondents who reported seeing safe food-handling labels also varied by demographic group. Seeing label information on raw meat or poultry was less commonly reported among men than women, and among Asians or Pacific Islanders than other racial or ethnic groups. Prevalence of seeing the label increased with increasing age ($P < 0.001$) and increasing education ($P < 0.001$). The proportion of respondents who reported seeing the label was lowest among persons in households earning less than $15,000 per year, followed by those in households with incomes of $50,000 or more per year.

Reading of label. Among those persons who remembered seeing the label information, the proportion of respondents who recalled reading the label information varied from 74% in Indiana to 84% in Tennessee (Table 1). Reading the label information was less common among men than women and less common among Hispanics than other racial or ethnic groups. The proportion of respondents who remembered reading the label information on uncooked meat or poultry was lowest in 18- to 29-year-olds and highest in 30- to 59-year-olds. The percentage of persons who reported reading the label was significantly lower among

TABLE 1. *Frequencies of seeing and reading label information and self-reported change in raw meat preparation methods because of label information on packages of raw meat or poultry, by demographic characteristic, 1995 and 1996 BRFSS*[a]

Demographic characteristic	Saw label (n = 14,262) (%)	Read label[b] (n = 7,339) (%)	Changed raw meat preparation methods because of label, among persons who reported that they have seen and read label (n = 5,738) (%)
Total	51	79	37
State			
Colorado[c]	52	75	31
Florida	57[d]	78	34
Indiana	50	74	33
Missouri	59[d]	76	28
New Jersey	50	80[d]	36
New York	42[d]	81[d]	43[d]
Tennessee	54	84[d]	44[d]
Sex			
Male[c]	43	71	37
Female	56[d]	83[d]	37
Age (in years)			
18–29[c]	46	73	41
30–59	52[d]	81[d]	36[d]
≥60	52[d]	79[d]	34[d]
Race/ethnicity			
White[c]	52	80	34
Black	46[d]	76	48[d]
Asian	30[d]	77	32
Hispanic	48[d]	69[d]	51[d]
Education			
<Grade 12[c]	43	76	49
High school grad	52[d]	78	39[d]
College or more	52[d]	80	33[d]
Yearly salary			
<$15,000[c]	46	78	42
$15,000–34,999	52[d]	77	37[d]
$35,000–49,999	54[d]	84[d]	36[d]
≥$50,000	15[d]	79	35[d]
Residential area			
Urban[c]	50	79	37
Suburban/small town	54[d]	78	32[d]
Rural	54	79	35

[a] Excluding persons who reported that they did not handle raw meat or chicken.
[b] Among persons who reported that they have seen label.
[c] Referent group.
[d] Significantly different from referent group, $P < 0.05$.

persons with less than a high school education. The percentage of persons who reported reading the label was significantly lower among households earning less than $15,000 per year than for those among households earning more than $35,000 per year.

Self-reported change in behavior. Among those persons who recalled seeing the new label, the proportion who reported that the labels changed their preparation methods for raw meat or poultry varied by demographic group (Table 1). The lowest prevalence of reported change in raw meat preparation practices after seeing these labels was found among respondents of Asian race. The proportion of persons who reported changing raw meat or poultry preparation practices decreased with increasing age, increasing education, and increasing income ($P < 0.05$).

Among respondents who reported changing their raw meat preparation practices because of the label, 15% reported that they did not wash their hands or cutting board with soap and water after contact with raw meat or poultry, and 19% reported eating pink hamburger in the 12 months before interview. The prevalence of these risky food-handling and consumption practices was similar among respondents who reported not changing their raw meat preparation practices because of the label (14% did not wash their hands after contact with raw meat or poultry, 13% did not wash their cutting board after contact with raw meat or poultry, and 21% reported eating pink hamburger in the 12 months before interview).

Association between label awareness and proper food-handling and preparation practices. After adjusting for age, sex, completed education, and yearly salary, persons who reported that they had seen the label were significantly more likely than those who had not seen it to report usually washing their hands after handling raw meat or chicken (adjusted odds ratio = 1.4; 95% confidence interval, 1.3 to 1.6) and to report washing cutting boards after contact with raw meat or chicken (odds ratio = 1.5; 95% confidence interval, 1.4 to 1.7). However, there was not a statistically significant association between seeing the label and not eating pink hamburgers in the past 12 months (odds ratio = 0.9; 95% confidence interval, 0.8 to 1.0).

DISCUSSION

In our large survey, conducted during the first years after nationwide implementation of safe food-handling labels on all retail packages of raw meat and poultry, approximately half of respondents noticed the safe food-handling labels. The majority of persons who had seen the label reported that they remembered reading the label messages. More than one-third of these persons reported that they had changed their meat preparation practices because of the label. Our survey, among the general population, found a lower level of consumer awareness of the label, however, than surveys among Oregon food preparers (83%) and U.S. primary food shoppers (59%) *(5, 15)*. The higher rate of consumer awareness of the label in other studies compared with our study might be related to the sample frame. Our survey was of the general population, and other studies were focused on label awareness in food preparers *(15)* and primary food shoppers *(5)*. The food preparers and shoppers surveyed in other studies were, perhaps, more likely to see and read safe food-handling labels *(16)*. As in other studies, we found that women were more likely than men

to be aware of the label *(15)* and that awareness increased with age. We suspect that persons who have practical knowledge of food-handling techniques might be more likely to read label information than persons who lack such knowledge. The sex and age characteristics of persons most aware of the label support the idea that label awareness might be related to frequency of food preparation. This might be because food preparers are particularly likely to notice and have an interest in information on the label *(13)*. If this is the case, the utility of labeling might be limited for the general population and especially for persons without frequent food-handling experience.

Seeing the label was least common among persons of Asian race, perhaps reflecting the marginal effectiveness of labels written in English among persons who speak English as a second language. Hispanics, however, reported considerably greater label recognition, suggesting that current efforts to provide labels written in Spanish might be worthwhile. According to a study of product warning message awareness among less-acculturated, Spanish-speaking Hispanics in San Francisco, Hispanics were less likely to be aware of warnings, particularly those written only in English *(9)*.

In this survey, 37% of respondents who said that they saw and read the label information on raw meat and poultry responded that they changed their raw meat and poultry preparation methods because of the label. We found a lower level of behavior change than a survey among supermarket shoppers (43%) *(5)*. This might be related to the sample frame, because supermarket shoppers could be more receptive to information on food safety labels and more likely to modify their raw meat and poultry preparation habits. While label awareness increased with education and annual income, high-risk food-handling, preparation, and consumption behaviors were more common among those with higher education and income, which is consistent with previous surveys *(1, 17)*. This suggests that safe food-handling labels have only limited influence on consumer's practices. Our results also suggest that the labels may be more effective in discouraging cross-contamination than in promoting thorough cooking practices.

Another possible reason for the lower rate of behavior change in our sample is that a higher proportion of our respondents had safe food-handling practices before they saw the label and therefore would respond that they did not change their raw meat preparation habits because of the label. We were not able to assess directly the respondent's food-handling practices at the time they saw the label. However, the proportion of persons who reported that they had risky food-handling and preparation habits (i.e., not washing their hands or cutting boards after contact with raw meat or poultry or eating pink hamburgers in the 12 months before interview) did not differ among persons who reported changing their raw meat or poultry preparation methods because of the label and among those who reported not changing their raw meat or poultry preparation habits because of the label. These findings suggest that not all persons who said that they did not change their raw meat or poultry preparation habits gave this response because they already had safe food handling and preparation practices. These findings also suggest that even among the persons who report changing their raw meat preparation practices because of the label, the label did not successfully prevent these persons from having risky food-handling behaviors, several of which are addressed by the label's messages. Persons might not have changed their raw meat or poultry preparation practices because they might have preferences for raw or undercooked foods or they might not fully understand the hazards of food contamination *(13)*.

In summary, the safe food-handling label was recognized by half of this survey's respondents. While the label is an inexpensive and reasonable method to distribute food safety information to the consumer, additional food safety education programs are needed to educate consumers about proper food-handling and preparation practices. In particular, efforts are needed to motivate persons who have risky food-handling and preparation behaviors to change these behaviors.

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A SUMMARY OF RESEARCH INFORMATION ON WHY INTERNAL MEAT COLOR SHOULD NOT BE USED AS AN INDICATION OF MEAT DONENESS

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Abstract

This article is a review of research studies of differences in cooked meat color. These studies indicate that the visual assessment of cooked meat color should not be used to evaluate doneness of meat and poultry products. To accurately assure the safety of raw ground meat products, these food items must be heated to temperatures for times necessary to ensure destruction of pathogenic microorganisms such as *Salmonella* spp. and *Escherichia coli* O157:H7, as recommended by the FDA Food Codes *(8, 9)*.

Consumers are incorrectly given the impression, through display of an illustration of a bimetallic coil thermometer on packages of raw meat and poultry products, that this is the device they should use to measure temperature of these products when they are cooked. However, the only accurate way to measure the temperature of food products, such as thin meat patties, fish, and poultry items, is the use of thin tipped thermistors or thermocouple thermometers.

Introduction

After serious outbreaks of *E. coli* O157:H7 associated with inadequately cooked ground beef patties *(2, 3, 4, 5, 20)*, government regulatory agencies and various trade associations have issued guidelines and have made suggestions regarding the cooking of ground raw meat patties, particularly ground beef patties. In 1993, the FDA issued interim guidelines to its 1976 Model Food Code *(10)* to suggest that beef patties be cooked to 68.3C (155F). The Food Marketing Institute and the American Meat Institute, in cooperation with the National Live Stock and Meat Board, USDA, and FDA, published guidelines *(11, 12)* for both consumers and the foodservice industry regarding safe handling and preparation of ground beef. These guidelines recommended that both consumers and foodservice units cook ground beef patties until there is no pink color and the juices are clear.

In order to assure destruction of *E. coli* O157:H7, it was also recommended that consumers cook ground beef patties to an internal endpoint temperature of 71.1C (160F) and that foodservice operations cook ground beef patties to a temperature of 68.3C (155F) (with a 16-second holding). The USDA began enforcing these temperatures and holding times *(21)* in the fall of 1993. These cooking times and temperatures were derived from a research study conducted by Goodfellow and Brown *(14)* in 1978. Their research effort determined D-values (time at a specific temperature to reduce a population of microorganisms by 1 log value or by a factor of 10 to 1) for a mixture of 5 strains of *Salmonella* spp. This was accomplished by measuring the destruction of *Salmonella* spp. in ground beef in test tubes held in a water bath at fixed temperatures.

The FDA 1995 and 1997 Food Codes *(8, 9)* recommend that comminuted raw meat, poultry, and fish products be cooked until all parts reach a temperature of 68C (155F) for 15 seconds. Recommendations are also given for cooking these products at alternative temperatures and times [e.g., 63C (145F) for 3 minutes or 66C (150F) for 1 minute]. If these time and temperature recommendations are followed, destruction of *E. coli* O157:H7 and other vegetative pathogens is assured. However, the recommendation to cook ground beef patties until there is no pink color and the juices run clear is erroneous. Some ground beef patties may appear brown before they have reached a desired temperature, and some ground beef

patties may remain pink at temperatures well above 71.1C (160F).

Factors Influencing Meat Color

The red color in meat is due mainly to a protein pigment called ***myoglobin*** and, to a lesser extent, hemoglobin. Red meats such as mutton and beef contain more myoglobin than do lamb, veal, and pork. There is less myoglobin in turkey and chicken breasts than in the legs and thighs. Meat that has only a small amount of myoglobin is called "light" or "white" meat; "dark" or "red" meat is significantly higher in this pigment protein.

Myoglobin molecules contain an iron porphyrin compound called heme. In the center of heme is a molecule of iron. When the iron is in the ferrous state, myoglobin can bind oxygen. In living muscle, this is very important, because this enables myoglobin to bind or take oxygen from hemoglobin of blood. However, when the animal dies, the supply of oxygen in cell tissue is depleted, and the color of meat (muscle) turns to the purplish red color of myoglobin. When fresh meat is cut and myoglobin is again exposed to oxygen in the air, the iron in the ferrous state again binds oxygen and meat color is a desirable cherry red color due to the formation of oxymyoglobin. After a period of time, or due to environmental conditions, the iron in the interior of this pigment is oxidized. It is no longer able to bind oxygen, and a brown-colored pigment (metmyoglobin) is formed *(6, 7, 23)*. The meat is usually still wholesome and safe to consume when it is cooked; however, it usually signifies lack of freshness and loses customer appeal.

Cured Meat Color

A change also occurs in the pigment myoglobin when meats are "cured". Sodium nitrite, salt, and heat are used during the curing process. The nitrite combines with myoglobin to form the stable pink color of cured meats, nitroso-myoglobin. This cured meat pigment is unstable in the presence of light, and photo-induced fading will occur. The addition of nitrites to cured meats (e.g., sausages and ham) also prevents the growth of *Clostridium botulinum (6, 23)*.

Cooked Meat Color

It is usual for the color of red meats to change to a grayish-brown color when meats are cooked and heated sufficiently. This is due to the formation of the pigment (denatured globin hemichrome). Because heat brings about this change in color, the color of cooked meat, especially tender cuts of beef, has been used, and in many instances, is still used as an index of doneness. Consumers expect fresh meats to be red, and "well-cooked" meats to be gray-brown. Consumers also interpret red or pink color in cooked meat as indicating the "amount of doneness". Many consumers prefer beef to be cooked so that it is rare (red) in the interior. Others prefer their beef cooked to a "well-done" visual appearance (gray-brown color with no evidence of red). Consumers are quite concerned if pork is pink in color, because they assume that the meat has not been heated or cooked long enough to destroy any possible trichinae. Consumers are also concerned if poultry is pink, as this indicates to them that the product has not been cooked sufficiently.

When meat processors experience problems with cooked meat products, they first check to assure that the desired internal temperature has been reached. If the pink color persists in products reaching desired cooking temperatures, processors often suspect that nitrite, nitrate, or nitric oxide contamination has resulted in a pink, cured pigment reaction. It is now known that three distinct pigments may commonly cause red or pink color in cooked meats. They are: 1) undenatured myoglobin and oxymyoglobin (the red pigments of fresh meat), 2) nitrosyl hemochrome (the pink pigment of cured meat), and 3) reduced globin hemochromes of well-cooked meats. Occasionally, exposure of cooked meat to carbon monoxide causes formation of globin carbon monoxide hemochrome (i.e., turkey cooked in a gas oven, the pink ring in Texas barbecued beef cooked slowly in heavy smoke). This undesired red or pink color may be present immediately after cooking, or it may gradually develop during distribution or retail display. Initial

pH, cooking temperature, cooking method, processing procedures, packaging, and microbial growth (growth of *Pseudomonas* in cooked brats and *Clostridium* spp. in vacuum packaged beef) have all been shown to influence pink or red color in cooked meats *(6, 7)*.

The following research studies and reviews provide documentation that the use of visual methods for assessing meat doneness is inaccurate and should not be used to assure the safety of cooked meat and poultry products.

1. In 1989, Mendenhall *(19)* evaluated color differences in cooked ground beef patties produced from beef components from three purveyors. The pH values of the ground beef patties ranged from 5.6 to 6.2. The patties were cooked to 71C (160F). The internal color, as described by a sensory panel, varied from gray in patties within the normal pH range (5.3 to 5.7) of muscle to slightly red in patties with a pH of 6.2. Red to pink cooked color was most intense inside those patties with the highest pH and the greatest concentration of total pigments. Bull meat exhibited a much higher pH and a greater number of total pigments.

2. Ground beef patties containing either 4 or 20% fat were cooked by an electric grill alone or in combination with an overhead broiler unit to be (visually) either medium or well-done. (The visual standards used in this study were photographic standards developed by a committee of the American Meat Science Association in 1983 and were typically identified as assessment of doneness used by the foodservice and restaurant industries). About 20% of patties cooked to a visual medium stage of doneness did not reach recommended internal temperatures and holding times required for food safety *(1)*.

3. Ground beef patties from various sources of beef were cooked to 55 to 77C (131 to 170.6F) *(15)*. It was reported that internal patty color became less red as endpoint temperature increased. However, some patties cooked to lower temperatures turned brown prematurely. The authors concluded that visual evaluation of internal patty color was not an accurate indicator of patty doneness. Some patties cooked as low as 66C (150.8F) appeared as brown as those cooked to 71C (159.8F). Raw material source had little effect on internal patty color. Expressed juice became less red and more yellow with increasing endpoint temperature. Expressible juice never ran "clear." The authors suggested that a more appropriate guideline for cooking ground beef patties, would be "Cook until juices lack redness."

4. Hunt *(16)* reported that persistent pink color of cooked meat has been associated with elevated pH in meat; environmental exposure to gases such as nitrous oxide, carbon monoxide, or ammonia; and exposure to other meat additives, particularly acids that might increase the pH.

"Premature browning" (the development of a cooked appearance at temperatures much lower than typical temperatures) was found not to be associated with fat level or compaction. Some research indicates that premature browning is associated with longer lengths of frozen storage. Some authorities speculate that the oxidative state of the meat may be important to development of premature browning. The author cautioned that premature browning of ground beef patties may compromise the safety of food service establishments, such as nursing homes and school lunch kitchens, and consumer home kitchens that use internal color as an indicator of doneness. Numerous incidents of premature browning were noted in ground meat cooked to 55C (131F).

5. At the 1995 Meat Industry Research Conference in Chicago, Lavelle et al. *(17)* reported that premature browning of ground beef patties occurs when the pigments are either in an oxymyoglobin or metmyoglobin form prior to cooking. If the pigments are in a deoxymyoglobin state prior to cooking, internal cooked color can be used as an indicator of pathogenic microorganism destruction. Otherwise, either a lack of redness in the expressible juice color or specific time-temperature cooking is more appropriate indicators of patty doneness.

6. Visual and instrumental determinations were made on the color of beef patties that were cooked fresh, frozen, or after thawing *(23)*. The internal color of patties cooked to 71C (159.8F) within 12 hours of thawing at 7C (45F) remained red-pink. Only after thawing for 18 hours or longer did cooking to 71C (159.8F) result in a well-done appearance. The color of patties thawed while vacuum packaged and then cooked was more red than the color of non-vacuum-packaged patties after cooking. Spectral analysis of the raw product indicated that the effects of thawing and packaging on cooked color were linked to the level of metmyoglobin (metMb); higher levels of metMb resulted in less red patty color after cooking. In two other trials, the metMb level was varied by storage and/or processing conditions. Differences in the metMb level before freezing seemed to decrease during freezing and thawing. Differences in metMb before processing did not significantly affect cooked meat color. Patties cooked from the frozen state were less red than those cooked directly after processing. After 24 hours of thawing, patties cooked to 71C (159.8F) were brown, irrespective of metMb level. Premature browning [i.e., the appearance of patties being well done at temperatures lower than 71C (159.8F)] only occurred in the thawed patties. After 24 hours of thawing, the patties appeared well done at 65C (149F). It was concluded that handling, other than internal temperature, strongly influences cooked beef patty color. Therefore, the color of cooked beef patties should not be used as an indicator of internal temperature.

7. Van Laack et al. *(22)* reported a cooking study for seventeen commercially prepared patty formulations. All patties were cooked to an internal temperature of 71C (159.8F). Pink cooked color occurred in 8 of the products and was due to incomplete denaturation of myoglobin. Although there was some relation between pH and cooked color, other factors seemed to be involved. When products were reanalyzed after 1-year storage at -27C (-16.6F), 16 products were red/pink when cooked to 71C (159.8F). This increase in redness could not be explained. Cooking to internal temperatures between 81 to 87C (177.8 to 188.6F) was necessary for complete disappearance of red/pink color. Premature browning, where a product looks well done at temperatures lower than 71C (159.8F), occurred in one formulation. Color differences before and after storage could not be explained by changes in reducing capacity or metMb content.

8. In 1997, Van Laack et al. *(24)*, reported evaluating the color of cooked patties prepared from beef with a pH of 5.7 (normal) or pH 6.2 (dark cutting) beef with either 0, 25, 50, or 75% lean, finely-textured beef. Patties were cooked to 68.3C (155F), 71C (159.8F), or until >75% were "well done". Inclusion of lean, finely textured beef in normal patties (pH 5.7) did not affect visual color scores or myoglobin denaturation. To appear well done, dark cutting beef (pH 6.2) patties without lean, finely textured beef content had to be cooked to 89C (192F), whereas dark cutting beef patties with 75% lean, finely textured beef had to be cooked to 83C (181F). The authors postulated that possibly, conditions during production of lean, finely textured beef are responsible for the increased heat sensitivity of myoglobin from lean, finely textured beef. This study is another demonstration that visual indication of doneness (i.e., meat color) is not a reliable indicator of internal meat temperature.

9. Warren et al. *(26)* reported that premature browning of ground beef is the development of a brown or cooked appearance at temperatures lower than those typically associated with well-done meat. Some ground beef patties are known to develop a premature brown color when cooked to 55C (131F). This occurrence has been noted in patties produced from older animals. Premature browning has also appeared to be related to length of frozen storage. However, these authors concluded that frozen storage was not the primary cause. They proposed that the patties that browned prematurely were more oxidized and had higher redox potentials and less total reducing activity than ground beef patties with normal cooked color.

10. To study premature browning in ground beef *(27)*, chemical properties were measured on raw and cooked patties [55, 65 and 75C (131, 149, and 167F)] that developed normal and premature brown color when cooked to 55C (131F). Normal color patties were visually and instrumentally redder at all