Viability of *Escherichia coli* O157:H7 during Manufacturing and Storage of a Fermented, Semidry Soudjouk-Style Sausage†

MEHMET CALICIOLGU,1 NANCY G. FAITH,2 DENNIS R. BUEGE,1 AND JOHN B. LUCHANSKY3*

1Meat Science and Muscle Biology Laboratory and 2Food Research Institute, University of Wisconsin–Madison, Madison, Wisconsin 53706; and 3Microbial Food Safety Research Unit, Eastern Regional Research Center, U.S. Department of Agriculture, Agricultural Research Service, 600 East Mermaid Lane, Wyndmoor, Pennsylvania 19038, USA

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**ABSTRACT**

Soudjouk-style batter was inoculated with a five-strain mixture of *Escherichia coli* O157:H7 at about 7.6 log_{10} CFU/g in each of two trials. The sticks were fermented and dried at 22°C and 50% relative humidity (RH) for 3 days and then at 9°C and 40% RH for 18 h. After being flattened to about 1.25 cm, the sticks were conditioned at 38°C and 70% RH or at 22°C and 50% RH for about 3 days. After the latter conditioning treatment, sticks either were cooked to an internal temperature of 63°C or received no heat treatment. Final mean pH values after conditioning at 22°C and 50% RH for soudjouk manufactured with a starter culture and dextrose (1.0%) and for soudjouk manufactured without a starter culture were about 4.9 and 6.0, respectively. For soudjouk produced with a starter culture, pathogen numbers were reduced by 4.53 and 0.88 log_{10} CFU/g after conditioning at 38°C and 70% RH and at 22°C and 50% RH, respectively. For soudjouk produced via natural fermentation, pathogen numbers were reduced by 1.39 and 0.09 log_{10} CFU/g after conditioning at 38°C and 70% RH and at 22°C and 50% RH, respectively. Cooking reduced pathogen numbers to below the levels detectable by direct plating (<1.0 log_{10} CFU/g) and by enrichment for soudjouk produced with a starter culture and also reduced pathogen numbers by 6.28 log_{10} CFU/g for soudjouk produced via natural fermentation. However, cooking also resulted in an unacceptable product. In general, the reduction in pathogen numbers achieved by storage at ambient temperature (25°C) was greater than that achieved by storage at cooler temperatures (4 and 15°C), particularly for soudjouk prepared with a starter culture (for which a final pH value of 4.8 and a 6.4-log reduction were obtained after 21 days at 25°C) rather than for that prepared without a starter culture (for which a final pH value of 6.1 and a 2.6-log reduction were obtained after 21 days at 25°C). These results indicate that naturally fermented old-country-type sausage may allow the survival of *E. coli* O157:H7 in the absence of controlled fermentation, postfermentation cooking, and/or an ambient-storage processing step.

Ethnic fermented meats such as chorizo, salchichon, landjager, and soudjouk are becoming more available in the United States as the immigrant population and the demand for such products continue to increase. Several types and brands of ethnic sausage are available via the Internet and/or from local manufacturers producing relatively small batches of these products. Many of these sausages are often fermented without a starter culture and/or without the addition of a carbohydrate, resulting in an overall quality that can vary widely from batch to batch. The lower acid content (pH > 5.0), variable drying times and conditions, and frequent handling during the manufacture of ethnic sausages are of concern because of the presence and ability of pathogens, notably *Escherichia coli* O157:H7, to remain viable in meats manufactured in this manner (5, 7). Soudjouk is one example of an ethnic sausage that is growing in popularity and is produced via a traditional process involving the natural fermentation of all ruminant meat without additional cooking or smoking (2–4, 10–12). Soudjouk is a spicy hot, semidry sausage common in the Middle East and in Balkan countries as well as among immigrants to the United States originating from these geographical areas. To date, little has been published with regard to the survival of *E. coli* O157:H7 in a small-batch “old country” sausage such as soudjouk. The present study expands on our previous research with soudjouk-style sausage (2) and monitors the effect of a different manufacturing process and postfermentation cooking regimen on the viability of *E. coli* O157:H7. These data are warranted because of the various processes used by small-scale manufacturers and the limited resources of such manufacturers to conduct such studies to determine lethality and assure safety.

**MATERIALS AND METHODS**

**Bacteria.** Acid-adapted stationary-phase cells of a five-strain mixture of *E. coli* O157:H7 were prepared as previously described (2) except that strain EC505B was replaced with strain EC205B, a beef isolate from the University of Wisconsin Food Research Institute. The commercial *Pedicoccus acidilactici* starter culture (Saga 75, Quest International, Rochester, Minn.) was maintained and propagated according to the manufacturer’s instructions. The starter culture was prepared by adding 2.8 ml of thawed Saga 75 brought to 50 ml with sterile water to each 30-lb (13.6-kg) portion of raw meat.

**Manufacture of sausage.** A diagram of the manufacture, cooking, and storage of soudjouk-style sausage is provided in Fig.
FIGURE 1. Flow diagram depicting the soudjouk-manufacturing process evaluated.

The raw batter was prepared in 60-lb (27.2-kg) batches composed of coarsely ground beef with a target fat content of about 20%. Nonmeat ingredients included 2% salt and 4.7% spice mix (house blend, Kayseri Basterma, Inc., Poughkeepsie, N.Y.). Neither nitrate nor nitrite was used. The meat block was inoculated with E. coli O157:H7, mixed with the other nonmeat ingredients, and held at 4°C for 24 h. The resulting batter was divided into two equal portions, of which one received 1.0% dextrose (A. E. Staley, Decatur, Ill.) and starter culture (ca. 7.0 log_{10} CFU/g) and the other received no starter culture. The batter was ground through a 1/8-in. (0.32-cm) plate using a commercial grinder (model 84142, Hobart Manufacturing Co., Troy, Ohio) and then stuffed into 6.5-horsepower (4.9-kilowatt) motorized commercial stuffer equipped with a 3.5-in. (8.8-cm) stuffing tube using a commercially available sausage stuffer (model 8501, Hobart Manufacturing Co., Troy, Ohio). 

Microbiological analyses. For each of two trials, three samples (raw batter or soudjouk sticks) were collected prior to stuffing, after drying, after conditioning, after cooking, and/or during storage for 7, 14, and 21 days at 4, 15, and 25°C. Samples were processed for direct enumeration or enrichment of the pathogen as previously described (6). It should be noted that raw materials were examined and found to be free of indigenous E. coli O157:H7.

RESULTS

Microbiological analyses of soudjouk sticks fermented naturally. In sticks (pH ca. 6.0) fermented without a starter culture, pathogen numbers decreased by <1.4 log_{10} CFU/g after conditioning at 38°C and 70% relative humidity (RH) or at 22°C and 50% RH (Table 1). Although subsequent cooking of the sticks conditioned at 22°C and 50% RH to an internal temperature of 63°C provided a reduction of 6.3 log_{10} CFU of the pathogen per g, according to an empirical evaluation by an untrained panel, it also severely affected product texture (i.e., made the product more greasy) and appearance (i.e., produced an off-color). When uncooked soudjouk that had been conditioned at 22°C and 50% RH was stored at 25°C for 21 days, a reduction of 2.6 log_{10} CFU/g was obtained, whereas storage at 4 or 15°C resulted in a reduction of ≤1.0 log_{10} CFU/g at the end of the storage period.

Microbiological analyses of soudjouk sticks fermented via a starter culture. In sticks with 1.0% added dextrose (pH ca. 4.9) fermented with Saga 75, pathogen numbers decreased by 4.53 and 0.88 log_{10} CFU/g after conditioning at 38°C and 70% RH and at 22°C and 50% RH, respectively (Table 1). Pathogen numbers dropped to below the level detectable by enrichment plating after cooking, but the resulting product was unacceptable. When uncooked soudjouk sticks that had been conditioned at 22°C and 50% RH were stored for 21 days, reductions of 6.4 log_{10} CFU/g at 25°C, 1.61 log_{10} CFU/g at 15°C, and 1.34 log_{10} CFU/g at 4°C were obtained.

DISCUSSION

Several studies have established the viability of Escherichia coli O157:H7 in fermented meats (5, 7), including soudjouk (2, 3), that typically rely solely on natural fermentation and drying for both quality and safety attributes. In addition to underscoring the importance of controlling the fermentation time, temperature, and end point, these studies have confirmed that factors such as the presence and levels of salt, nitrite, and smoke; the casing size; the postfermentation heating regimen; and the synthetic media used to enumerate this pathogen can in large measure influence lethality estimations for a given process. It is important for both large and small manufacturers to control these parameters in order to satisfy regulatory guidelines for the assessment or validation of the safety of fermented meat products with regard to E. coli O157:H7 (1, 8, 9).

In brief, the regulatory guidelines proffered by the U.S. Department of Agriculture’s Food Safety and Inspection Service require the attainment of a 5-log_{10} reduction in pathogen numbers (options 1, 2, and 4), a hold-and-test program for finished product (option 3), or raw batter testing and a 2-log_{10} reduction in pathogen numbers (option 5) for controlling E. coli O157:H7 in dry and semidry fermented sausage.
primary objective of the present study was to evaluate a process to identify steps that generated either a 2- or a 5-log reduction of *E. coli* O157:H7 for soudjouk-style sausage in order to assist smaller manufacturers who may lack the facilities and/or the resources to do so independently.

We evaluated a manufacturing process for soudjouk-style sausage used by a small manufacturer in the United States. In general, natural fermentation and drying was not as effective as the use of a starter culture in reducing levels of *E. coli* O157:H7. Although our findings do not constitute a process validation per se because two, rather than three, trials were performed, a 5-log reduction of *E. coli* O157:H7 was achieved by cooking. However, since cooking also resulted in an unacceptable product, future studies may be directed at modifying the cooking time and temperature to achieve an appreciable reduction of the pathogen and still produce a desirable product. A 5-log reduction was also achieved through storage of the finished product at the ambient temperature for 2 to 3 weeks for sticks produced using a starter culture. It was also possible to achieve about a 4.5-log reduction of the pathogen by shifting the temperature and humidity from 22°C and 50% RH to 38°C and 70% RH during conditioning for sticks produced using a starter culture. These data indicate that increasing the temperature from 22 to 38°C for the second half of the fermentation process (i.e., conditioning) and holding the product for an additional day or so may be a viable alternative to cooking. Collectively, these results are in good agreement with our previous study, wherein we validated an *E. coli* O157:H7 reduction of 5 log_{10} CFU/g in Turkish soudjouk via (i) controlled fermentation to a pH of ≤4.6 and subsequent cooking to an internal temperature of 54.4°C; (ii) controlled fermentation, no postfermentation cooking, and vacuum-packaged storage at 21°C for at least 14 days; or (iii) natural fermentation to a pH of ≤5.4, cooking to an internal temperature of 54.4°C, holding for 60 min, and storage at 21°C for at least 14 days.

Our findings confirm that the manufacture of soudjouk-style sausage without a starter culture and/or without a postfermentation cooking step and/or without extended postproduction storage at an ambient temperature may not assure the safety of the product with regard to the guidelines of U.S. Department of Agriculture’s Food Safety and Inspection Service. Nevertheless, our results provide a framework for small-scale producers of “old world” sausage to modify their current manufacturing processes to enhance product safety with regard to *E. coli* O157:H7.

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