

Effects of Meat Type, Storage Time and Temperature on Various Physical, Chemical and Microbiological Characteristics of Ground Pork

DONNA S. BENTLEY, JAMES O. REAGAN*, NELSON A. COX and J. STAN BAILEY¹

The University of Georgia, Department of Animal & Dairy Science, Athens, Georgia 30602

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ABSTRACT

Whole-hog sausage was prepared from hot- and cold-boned pork raw materials to determine the effects of meat type, storage temperature and length of storage on various processing and bacteriological characteristics. Samples were stored at -1 and 4°C for 0, 28 and 56 d. Various physical, chemical and microbiological properties of the sausage were evaluated. Thiobarbituric acid (TBA) values were not affected by meat type (pre or postrigor). Hunter-Color values varied significantly among the meat types and storage temperatures. Total bacterial counts varied significantly among the hot- and cold-boned pork sausage samples (day 0). Cold-boned sausage stored at -1°C had lower plate counts of the various treatments for days 28 and 56. *Pseudomonas* was the predominant organism found in hot- and cold-boned sausage samples. Hot-boned sausage exhibited a more diverse bacterial population than did cold-boned sausage. More gram-positive organisms were found in hot-boned sausage samples. Cold-boned sausage had a lower total bacterial count at day 0 and maintained lower counts and therefore a longer shelf life throughout the study when held at -1°C.

Production of whole-hog sausage has increased tremendously within the past few years. Pork sausage is a popular meat item in homes, restaurants and other institutions. Whole-hog sausage is superior to pork sausage made from pork trimmings in many respects. One important characteristic is that of greater processing efficiencies. By using the entire carcass for sausage production, there is less waste, shorter holding periods for carcasses and more efficient chill systems for the product.

Hot-boned raw materials have been reported to possess many superior sausage making characteristics, which include increased energy saving, and reduced cooler space, labor and materials used (6,9).

The overall quality of pork sausage is greatly affected by the method of processing, initial microbial levels, length of storage and various formulated ingredients.

Length of time in storage was observed to correspond with cooking yields, whereas longer storage periods were related to higher yields (15). Lin et al. (13,14) reported that prerigor pork sausage stored at 2°C had significantly lower TBA values, higher pH values, higher juiciness and overall acceptance scores and less cooking losses than postrigor pork sausage. Effects of various processing methods for pork products have been discussed by Siegel et al. (16) and Theno et al. (19).

The objective of this study was to investigate the effects of meat type (prerigor, postrigor), storage temperature (-1°C, 4°C) and length of storage (0, 28, 56 d) on various processing and bacteriological characteristics of hot-boned pork sausage.

MATERIALS AND METHODS

Hot-boned raw sausage materials were obtained from a packer sow weighing approximately 227 kg. One side of the carcass was hot-boned within 1 h postmortem and coarse ground using a standard kidney plate (4.48 cm). The meat (45.5-kg batch) was mixed for 1 min in a Butcher Boy Meat Mixer and samples were obtained for fat determinations using the modified Babcock procedure. Batches were formulated to contain $25 \pm 1\%$ fat. Appropriate amounts of fresh pork sausage seasoning (Formula 64T, A. C. Legg Packing Co., Inc.) were added to the hot-boned materials and mixed for an additional 2 min to assure even distribution of the seasoning. After mixing, each batch was fine ground (0.3-cm plate) and stuffed into commercial opaque sausage bags (E. G. Luber Engineering and Sales Co.), tied with a twist lock tab and placed into a -3°C brine chiller. The temperature of the product before stuffing was 30°C. The internal temperature of several bags of sausage was monitored on a multipoint recorder until it reached 2°C. The bags were then removed from the chiller, boxed and randomly assigned to one of the two storage temperatures (-1°C or 4°C) for varied lengths of storage (0, 28, or 56 d). After 2 h of equilibration in the cooler the internal temperature of the packages was 0.5°C.

The remaining side of the carcass (chilled 24 h) was cold-boned and processed into sausage using the previously described procedure. The temperature of the raw materials at the time of stuffing was 15.5°C. The packaged samples were placed

¹USDA-ARS, Richard B. Russell Research Center, Athens, Georgia 30613.

in the brine chiller and chilled to an internal temperature of 2°C. After a 2-h equilibration period in the appropriate coolers the temperature of the packages was 1°C.

Random pH readings of the raw materials were taken at the time of boning using a Corning (Model 610A Expand) portable pH meter. An average of the measurements of the hot- and cold-boned pork was taken and pH values were recorded as 6.5 and 5.9, respectively.

At the end of each storage period, samples of each meat type were obtained for determinations of internal color, TBA analysis, bacterial density and identification of bacterial isolates randomly selected from total plate counts.

Color measurements

A Macbeth colorimeter (Model MC-1010S) was used to determine the color characteristics of the internal surfaces of the samples. The data are reported in the Hunter "L" "a" "b" Color System with "L" (lightness), "a" (redness) and "b" (yellowness) being used to correlate color changes to other sausage properties. A cross section of the sample, covered with polyethylene film, was used for internal color measurement. Three replicates were taken from each sample (bag) and measured for color values.

TBA values

Thiobarbituric acid (TBA) values were determined by the distillation method of Tarladgis et al. (18). The values were reported as mg of malonaldehyde per 1000 g of sample. All analyses were performed in duplicate.

Microbial determinations

Random samples (10 g) were obtained from the raw sausage materials at the end of each storage period. Duplicate samples from each treatment were blended in a Waring Blender with 90 ml of physiological saline (0.85%) solution. Following appropriate dilutions, duplicate pour plates of tryptic soy agar (Difco) were made and incubated at 8°C for 7-10 d to determine psychrotrophic counts. Plate counts for each sample were reported as log₁₀ per g.

Microbial isolation and identification

After incubation, isolates to be identified were randomly selected using a numerical grid (4) and a random numbers table (17). A total of 300 colonies was picked and streaked to produce isolated colony forming units on plates of tryptic soy agar (Difco). All cultures used were grown at 25°C for 18-24 h. Once the purity of the culture was confirmed by visual examination of the colonies, isolates were tested for gram reaction, morphology, motility, catalase and oxidase reaction to categorize them so that further tests could be made to identify each isolate.

The characteristics used to identify each isolate as determined by Bergey's Manual of Systematic Bacteriology (11) were as follows: (a) *Pseudomonas fluorescens*, gram-negative, catalase-positive rods, oxidase-positive, motile, slow growth at 37°C, no growth at 41°C, no reduction of nitrite, no fermentation of dextrose, fluorescent pigment produced, (b) non-pigmented *Pseudomonas*, gram-negative, catalase-positive rods, motile, oxidase-positive, aerobic acid production from dextrose, no anaerobic fermentation of dextrose, no pigment produced, (c) *Enterobacteriaceae*, gram-negative, oxidase-negative, glucose-positive rods, were identified by the Micro-ID, API-20E or AutoMicrobic system (GNI Card), (d) *Acinetobacter*, gram-

negative, oxidase-negative rods were identified by the AutoMicrobic system (GNI Card) and (e) *Aeromonas*, gram-negative, oxidase and catalase-positive rods with rounded ends were identified by the AutoMicrobic system (GNI Card).

Micro-ID (The General Diagnostics Division of Warner-Lambert Company, Morris Plains, New Jersey), a self-contained identification unit containing reagent-impregnated paper discs for 15 biochemical tests requiring only 4 h of incubation at 37°C, was used to identify *Enterobacteriaceae* to species. The Micro-ID system has been evaluated by Cox et al. (5) and found to accurately identify foodborne *Enterobacteriaceae*.

The API-20E system, (Analytab Products, Inc., Plainview, New York), includes a series of 20 plastic cupules affixed to a plastic strip; each cupule contains a dehydrated substrate for a different biochemical test and requires 18-24 h of incubation at 37°C. The API-20E strip was used in the identification of *Enterobacteriaceae* and has previously been evaluated with food isolates and found to be accurate (3).

The AutoMicrobic system (AMS; Vitek Systems, Inc., Hazelwood, MO) consists of a diluent-dispenser, filling module, reader-incubator module with a data terminal, previously described in detail (1). The AMS gram-negative identification card (GNI) was used to identify gram-negative isolates and the gram-positive identification card (GPI) was used to identify gram-positive isolates. The GNI and GPI are plastic cards with 30 wells which contain 29 biochemical broths and one growth control broth.

Statistical analysis

Analysis of data was performed according to the Statistical Analysis System of Barr and Goodnight (2). Duncan's multiple range test (8) was used to indicate significant differences between particular variables. Significant differences were accepted at the 5% level of probability.

RESULTS AND DISCUSSION

Overall mean values for moisture and fat content stratified by meat type are presented in Table 1. Percentage moisture varied significantly between the hot- and cold-boned treatments. However, the practical significance of the observed difference is questionable, as the variation could be attributed to analyzing means instead of individual measurements. No significant differences were observed in fat content between the two meat types.

TBA values and color measurement values stratified by meat type are shown in Table 2. TBA values were not significantly different among treatments. Significant differences in "L" (lightness), "a" (redness) and "b" (yellowness) were found between the pre- and posttrigor sausage samples with all three measurements being higher for the cold-boned materials.

TABLE 1. Mean values for moisture and fat content stratified by meat type^a.

Meat type ^b	Moisture (%)	Fat (%)
HB	57.5 ^c	24.4 ^c
CB	56.2 ^d	25.4 ^c

^aOnly data for day 0 included.

^bMeat type: HB = hot-boned, CB = cold-boned.

^{c,d}Means in the same column bearing different superscripts differ significantly (P<.05).

TABLE 2. Mean values for various objective parameters stratified by meat type^a.

Meat type ^b	TBA Value (mg malonaldehyde per kg sample)	Color measurements ^c		
		"L"	"a"	"b"
HB	0.1993 ^d	52.9 ^e	+ 10.3 ^e	+ 12.2 ^e
CB	0.2765 ^d	55.8 ^d	+ 13.0 ^d	+ 16.0 ^d

^aOnly data from Day 0 included

^bMeat type: HB = hot-boned, CB = cold-boned.

^c"L" (lightness), "a" (redness), "b" (yellowness) are based on the Hunter - "L" "a" "b" Color System.

^{d,e}Means in the same column bearing different superscripts differ significantly ($P < .05$).

TABLE 3. Mean values for various objective parameters stratified by meat type and storage temperature.

Treatment ^a	TBA Value (mg malonaldehyde per kg sample)	Color Measurements ^b		
		"L"	"a"	"b"
HL	0.1201 ^d	52.4 ^d	+ 11.8 ^c	+ 14.4 ^d
HH	0.1108 ^d	52.2 ^d	+ 8.5 ^d	+ 12.7 ^e
CL	0.2430 ^c	57.2 ^c	+ 6.7 ^e	+ 15.5 ^c
CH	0.1541 ^d	56.6 ^c	+ 9.1 ^d	+ 15.4 ^c

^aTreatment: HL = Hot-boned; low storage temperature (-1°C)

HH = Hot-boned; high storage temperature (4°C)

CL = Cold-boned; low storage temperature (-1°C)

CH = Cold-boned; high storage temperature (4°C)

^b"L" (lightness), "a" (redness), "b" (yellowness) are based on the Hunter- "L" "a" "b" Color System.

^{c,d,e}Means in the same column bearing different superscripts differ significantly ($P < .05$).

TABLE 4. Mean values for various objective parameters stratified by time in storage.

Time in storage (d)	TBA Value (mg malonaldehyde per kg sample)	Color Measurements ^a		
		"L"	"a"	"b"
28	0.1047 ^c	54.5 ^b	+ 8.9 ^b	+ 14.3 ^c
56	0.2092 ^b	54.7 ^b	+ 9.1 ^b	+ 14.7 ^b

^a"L" (lightness), "a" (redness), "b" (yellowness) are based on the Hunter-"L" "a" "b" Color System.

^{b,c}Means in the same column bearing different superscripts differ significantly ($P < .05$).

Table 3 presents overall mean values for various objective parameters including TBA values and color measurements stratified by meat type and storage temperature. These data include measurements from the day 28 and

day 56 samples. TBA values among treatments were similar with the exception of the cold-boned sausage samples stored at the low storage temperature (-1°C), which had a significantly higher TBA measurement. All values were considered to be within an acceptable range. A significant difference for "L" (lightness) measurements was found between hot- and cold-boned sausage samples. The cold-boned samples were lighter in color, which supports the values found for day 0 samples (Table 2). The "a" (redness) values varied significantly among treatments with the hot-boned sample stored at -1°C having the highest value. The "b" (yellowness) values showed a significant difference among treatments with the cold-boned samples having the higher overall values.

Overall mean values for TBA and color measurements stratified by time in storage are presented in Table 4. The extent of oxidative rancidity developed during storage of pork sausage samples was estimated with the TBA test. As expected, TBA values were observed to increase with time in storage. However, even after 56 d of cooler storage the TBA values showed low amounts of rancidity occurring. "L" and "a" color values showed no significant differences over time in storage. However, the "b" color value was significantly higher for the day 56 samples. Since the "b" values differed by only .4 unit the difference could be considered questionable.

At day 0, total bacterial counts for hot-boned sausage samples were significantly higher than for cold-boned sausage samples (Fig. 1). There were no differences in the total bacterial counts of the hot-boned sausage held at either -1.0 or 4.0°C. However, cold-boned sausage held at -1°C had significantly lower counts than did cold-boned samples held at 4°C (Fig. 1). Previous work by Lin et al. (13,14) showed that prerigor sausage samples had slightly higher values for psychrotrophic bacterial counts, but the differences were not significant. In comparison, Lee et al. (12) and Fung et al. (10) stated that in beef, hot-boned meat had higher total bacterial counts when compared to conventionally processed beef. Total plate count numbers increased significantly with time in storage (Fig. 1). Davidson et al. (7) reported that psychrotrophic counts for hot-processed pork sausage (after 15-20 d in storage) showed significantly greater counts.

The percentage of isolates recovered from hot and cold boned pork sausage are shown in Tables 5 and 6. The bacterial populations of both hot- and cold-boned sausage through day 28, were predominately *Pseudomonas* (Tables 5 and 6). Sausage held at 4°C had a more diverse bacterial population than did sausage held at -1°C (Tables 5 and 6). Hot-boned sausage exhibited a more varied group of microorganisms, with the percentage of pseudomonads gradually decreasing over time of storage (Table 5). In comparison, within the cold-boned sausage samples there was a less diverse group of organisms and when held at -1°C the bacterial population of the cold-boned sausage was almost 100% *Pseudomonas* (Table 6). In addition, more gram-positive organisms were found on the hot-boned than on the cold-boned sausage.

TABLE 5. Percentage of isolates recovered from hot-boned pork sausage.

Bacteria	Days of storage at temp				
	0	28		56	
		-1°C	4°C	-1°C	4°C
<i>Acinetobacter calcoaceticus</i>	---	16.0	4.0	---	4.0
<i>Aeromonas hydrophila</i>	2.0	---	---	---	---
<i>Enterobacter agglomerans</i>	2.0	---	4.0	---	---
<i>Hafnia alvei</i>	---	---	8.0	---	---
<i>Pseudomonas</i> spp	62.0	76.0	44.0	28.0	8.0
<i>Serratia liquefaciens</i>	14.0	---	20.0	16.0	68.0
Gram (-) rod					
Oxidase (+)	12.0	---	---	12.0	12.0
Gram (-) rod					
Oxidase (-)	8.0	8.0	8.0	---	---
Gram (+) rod					
Catalase (+)	---	---	12.0	32.0	8.0
Gram (+) rod					
Catalase (-)	---	---	---	12.0	---

TABLE 6. Percentage of isolates recovered from cold-boned pork sausage.

Bacteria	Days of storage at temp.				
	0	28		56	
		-1°C	4°C	-1°C	4°C
<i>Acinetobacter calcoaceticus</i>	12.0	---	4.0	---	---
<i>Citrobacter freundii</i>	4.0	---	---	---	---
<i>Escherichia coli</i>	4.0	---	---	---	---
<i>Pseudomonas</i> spp	48.0	100.0	72.0	84.0	40.0
<i>Serratia liquefaciens</i>	4.0	---	4.0	---	44.0
Gram (-) rod					
Oxidase (+)	8.0	---	---	16.0	12.0
Gram (-) rod					
Oxidase (-)	18.0	---	20.0	---	---
Gram (+) rod					
Catalase (+)	2.0	---	---	---	4.0

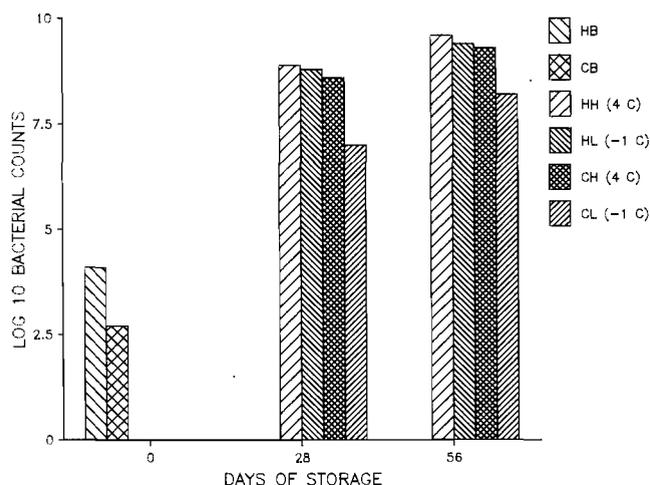


Figure 1. Total bacterial counts (log₁₀/g) of pork sausage stratified by meat type (HB or H = hot-boned; CB or C = cold-boned), storage temperature (L = -1°C; H = 4°C) and time in storage (days).

CONCLUSIONS

Whole-hog sausage production by means of accelerated processing appears to be a feasible method of sausage manufacture in that the desirable sausage making characteristics of hot-boned raw materials can be maintained for an extended time when stored at 2°C or less. However, the microbiological properties and quality of hot-boned pork sausage in the present study deteriorated over increased storage periods resulting in a decreased shelflife. The results of this study suggest the need for additional work to investigate the feasibility of whole-hog sausage production from a microbiological standpoint. Further studies in this area should include a detailed examination of microbial isolates for a comparison with this work.

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