Commercial manufacture of raw ripened fermented sausages formulated with mechanically separated chicken meat

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Introduction

Both in the traditional and modern manufacture of raw ripened fermented sausages, successful processing personnel use only the highest quality of meats available. Deviation from this practice leads inevitably to production problems which affect not only the organoleptic quality of the product but also its safety (Acton and Dick, 1976; Terrell et al. 1978; Klettner and Baumgartner, 1980). One key to the production of safe sausages is the effective and rapid initial acidification of the meat mix. This can be brought about by use of the chemical acidulant glucono-delta lactone (GdL) or by the addition of lactic acid bacterial starter cultures. Either or both of these agents allow certain additional latitude in production methods although their use does not completely insure that pathogenic bacteria present in the meat will be eliminated during the curing-ripening period (Goepfert and Chung, 1970; Baran and Stevenson, 1975; Kovacs and Takas, 1979). Other factors also significantly influence both the efficacy of desired acidulation and pathogen survival. The interaction of available carbohydrate, NaCl, curing salts and temperatures of fermentation and drying are the more of important of these (Klettner and Baumgartner, 1980; Liepe, 1983).

In order to improve product selling price - production cost ratios a number of alternatives in manufacturing have been examined. For example, Joseph et al. (1978a,b) added mechanically deboned beef or soy protein fibre to dry sausage formulations. The inclusion of poultry meats in semi-dry and dry sausages has been the subject of a number of studies (Keller and Acton, 1974; Acton and Dick, 1975; Baran and Stevenson, 1975; Dhillon and Maurier, 1975; McMahon and Dawson, 1976; Raccach and Baker, 1979). In some of these products poultry meats were used at levels > 50% of the total (Acton and Dick, 1975). However in most of these studies sausages were cooked at ≥60°C before drying although Baran and Stevenson (1975) heat-treated to 46°C.

Since both Salmonella and Staphylococcus occur regularly in sampled poultry at levels usually higher than in pork and beef (McKinley and Avens 1981; Swaminathan et al. 1978; Bentley and Pettit 1982, Davidson et al. 1985) the formulation of sausages with mechanically separated chicken meat might be expected to contain these organisms in numbers higher than would be otherwise present.

The purpose of the present work was to evaluate whether raw ripened dry sausage could be formulated with mechanically separated chicken meat (MSCM) under industrial conditions to yield an organoleptically acceptable product without additional risk from bacterial pathogens.

Materials and Methods

Sausage preparation Manufacture of sausages was conducted in 50 kg batches in an inspected meat plant by regular production staff. The meat mixture consisted of pork, beef and beef back fat in the proportions of 0.4, 0.35 and 0.25, respectively. The spice-adjunct mixture contained 3% NaCl, 0.02% Nitrate, 0.02% nitrite, 0.05% erythorbate, 0.3% glucose, 0.4% lactose, 0.4% white pepper, 0.4% sodium caseinate and 0.4% modified wheat flour. Experimental batches were formulated with 0.44% GdL or one of two types of starter culture; Trumark LT II (0.1%) which consisted of Pediococcus pentosaceus plus Staphylococcus carnosus or a 50:50 mixture of Duploferment 66 with Duploferment "Spezial" (yielded 70% Staphylococcus carnosus plus 30% Lactobacillus plantarum). The latter starter was added at a rate of 0.05%. After lean meats were chopped, fat was added and this was followed by starter or GdL addition. Sausages were stuffed in 60 mm diameter fibrous casings and weighed about 0.5 kg each. Sausages were held 8 h at 10°C (70% RH) and placed in a smokehouse at 22°C (92% RH) for 17 h. The temperature was reduced to 20°C (90% RH) and after 10 h the RH was reduced to 88% for an additional 14 h. Temperature was then reduced to 18°C for 10.5 h and over the next week gradual reductions in RH were conducted until 84% RH was reached. Sausages were smoked and dried an additional 20 days at 16°C and 76% RH.

Microbial analyses Frozen samples were analyzed for total bacteria (APT, 32°C, 48 h) lactic acid bacteria, LAB (mrs, 32°C anaerobic 48 h), micrococci (MSA containing 3% NaCl, 32°C, 48 h aerobic), pediococci (MRS, 45°C, anaerobic, 48 h) lactobacilli, MRS containing 2% lactose plus 0.1M arginine and 25 mg/l phenol red but glucose and beef extract were not included in this medium (32°C, anaerobic, 48 h), coliforms (VRB with overlay, 35°C, 24 h aerobic), Staphylococcus aureus was counted on BP (Baird-Parker Agar) using spread-plates (35°C, 48 h aerobic) followed by coagulase confirmation. Salmonella

(presence/absence) were examined by the Canadian Health Protection Branch method (MFA-10) using Nutrient Broth, tetrathionate and selenite cysteine enrichments followed by examination of growth on Brilliant Green Sulfa Agar and Bismuth Sulfite Agar. Staphylococci and Salmonella were analyzed using fresh (unfrozen) samples. The identity of Salmonella was confirmed using the micro ID identification kit (Warner-Lambert Co., Morris Plains N.J., USA). Complete cross section slices were taken from sausages for bacterial analyses (11 g treated with a Stomacher 400 and diluted in 0.1% w/v peptone). All media used were obtained from Difo laboratories Ltd., Detroit, USA.

Chemical analyses Nitrate, nitrite, moisture, protein and fat analyses were conducted as described in AOAC (1984). Thiobarbituric acid was measured after Tarladgis et al. (1960) and water activity was assessed using a Novasina model TH2/TH1 (Nova Sina, Zurich) hygrometer. Mesurements of pH were made using electronic metering systems (Fisher Scientific, Ottawa, Canada: Knick Electronische Messgerate, Berlin).

Sensory and instrumental analysis Ten panelists trained in the evaluation of sausage quality evaluated treatments in four separate sessions. Panelists were asked to compare thickness of the outer layer, surface oiliness, greasiness, toughness and smoked flavour using a 15 cm descriptive analysis line scale. A randomized block design was utilized with all six treatments evaluated being present at each session.

A universal food rheometer (interfaced with a personal computer) was used to analyze cohesiveness and firmness of eight replicate cores for each treatment.

An analysis of variance was completed on the sensory and instrumental data and significant differences at the 5% level noted. Correlations between instrumental and sensory data were calculated.

Results and Discussion

Results from preliminary experiments indicated that levels of MSCM ≥20% yielded products of poor organoleptic quality. Subsequent work therefore centered on the use of up to 15% MSCM.

The rate of change in pH from the initial value of sausage meat mixes (pH 5.9 - 6.0) is presented in Table 1. The most rapid and dramatic drop in pH occurred within the first day in GdL treatments, and is similar to that reported in other work (Baumgartner et al. 1980; Gerigk and Gossling, 1981; Metaxopoulos et al. 1981b; Paneras and Bloukas, 1984; Petaja et al 1985). After day 1, the most rapid drop in pH occurred in samples to which Trumark starter had been added. The subsequent acidification of GdL-treated samples proceeded more slowly than in the other treatments. The pH decline in Duploferment samples took place slower than in the other treatments, taking 6 days to reach a value of 5.0, although this might not be considered abnormal in view of the low temperature used for fermentation (22°C). The use of mechanically separated meat did not have a significant influence upon either the initial or final pH, and values obtained were considered acceptable (Diebel et al. 1961; Debevere et al. 1975; Sirvio et al. 1977; Baumgartner et al. 1980) for this type of product.

Total and lactic bacteria in starter-inoculated treatments were close to their maximum numbers at day 4 and within two more days had reached peak levels. Lactic bacteria (MRS, 32°C) dominated the organisms recovered on APT agar in all treatments (Figs. 1, 2 and 3). In Duploferment and GdL treatments lactic bacteria were mainly lactobacilli. Results from other studies of raw ripened (≤24°C) sausages from a number of countries also indicated that lactobacilli were the dominant bacterial component of the microflora within a week of the completion of fermentation (Nurmi, 1966; Debevere et al. 1975; Niskanen and Nurmi, 1976; Hofmann and Scharner, 1980; Gerigk and Gossling, 1981; Metaxopoulos, et al. 1981b; Simonetti et al. 1983; Brankova et al. 1984a,b; Paneras and Bloukas, 1984; Gokalp and Ockerman, 1985) whether or not starter cultures were used.

Starter micrococci were initially dominant in Duploferment treatments (Fig. 1) but they did not grow during sausage maturation in any of the test batches. Nurmi, (1966) pointed out that micrococci do not have to grow in order to contribute to sausage quality. Nonetheless, micrococci have been reported to initially increase in numbers during fermentations at 24°C (Debevere et al. 1975; Sirvio et al. 1977; Gerigk and Gossling, 1981; Holley, 1986) reaching maxima of 10^5-10^7/g although weak growth has also been noted (Metaxopoulos et al. 1981a). In contrast, Simonetti et al. (1981) reported micrococci to reach levels of 10^9/g in Italian salami fermented for two days. It is unlikely that the micrococci of Duploferment and Trumark treatments peaked at levels higher than those noted in Figs. 1 and 2 since their growth on conventional media (Hugh and Leifson's carbohydrate and MSA) was extremely slow at 22°C.

In Trumark treatments, which were rapidly acidulated by the added pediococci, micrococci numbers decreased rapidly. This result was predictable on the basis of previous observations where micrococci were reported to be acid sensitive (Klettner and Baumgartner, 1980). Micrococci remained at a fairly low but stable level through the ripening process in GdL treatments. Pediococci grew initially in all batches but they did not form a significant portion of the microflora in Duploferment treatments (Fig. 1). In contrast, these organisms dominated in Trumark treatments (Fig. 2) where they were the major starter organism added. Pediococci formed the

second largest population of bacteria in GdL treatments (Fig. 3). Pediococci were also recovered as lactic bacteria on MRS at 32°C. Modification of MRS by the addition of arginine and phenol red (MRS-B) permitted easier discrimination between lactobacilli and pediococci in Trumark-treated samples. Pediococci colonies were surrounded by pink zones (arginine positive) and lactobacilli by yellow zones. Most heterofermentative lactobacilli are also arginine positive but microscopic inspection revealed that colonies developing at 45°C were exclusively pediococci and these equalled recoveries at 32°C on the same medium. Deibel et al. (1961) also observed that lactobacilli found in sausages did not grow on MRS at 45°C. The numbers of homofermentative lactobacilli (arginine negative) were initially low ($\log_{10}$ 4.0/g) in Trumark-treated samples (Fig. 2) but slowly reached a maximum of log $_{10}$ 6.29 by day 18 and subsequently their numbers were stable. These organisms are desirable and probably dominated throughout Duploferment (Fig. 1) and GdL (Fig. 3) treated batches although specific analysis for their presence in these treatments was not made. In GdL-treated samples about 80% of the organisms recovered on MRS at 32°C were lactobacilli as determined by microscopic and catalase (neg.) testing.

The numbers of Staphylococcus aureus in all treatments were at or below the detection limit (≤100/g meat up to day 4) and were ≤10/g at day 14. The use of high quality meats and low temperatures for fermentation are believed responsible for this result. (Daly et al. 1973; Smith et al. 1983; Bacus, 1984; Marcy et al. 1985).

In contrast, approximately 43% of samples were naturally contaminated with Salmonella but their presence and disappearance was unrelated to the use of MSCM in the formulations. All samples were free from Salmonella contamination by day 14.

The concentrations of salt, nitrate and nitrite used have been shown to be ineffective against both staphyloccocci and Salmonella in freshly prepared meat batters (Baran and Stevenson, 1975; Genigeorgis, 1976; Bacus, 1984; Collins-Thompson et al. 1984). However, several studies have shown the effectiveness of bacterial starter cultures in the prevention of Salmonella growth at temperatures similar to those used here (Sirvio et al. 1977; Brankova et al. 1984a) and at higher ripening temperatures (Goepfert and Chung, 1970; Baran and Stevenson, 1975; Smith et al. 1975; Masters et al. 1981).

Initial numbers of coliforms were ≤2000 cells/g and this rapidly decreased so that by day 4 only 4 samples contained numbers ≤500/g and these decreased to ≤10/g by day 14. These results are similar to those observed elsewhere (Debevere et al. 1975; Paneras and Bloukas, 1984).

Physicochemical characteristics of manufactured sausages are shown in Table 2. At the end of ripening, Duploferment samples had lost less moisture and had slightly higher a_w values than either Trumark or GdL samples. It is notable that these differences in a_w and moisture were related to the type of acidulant and were not related to the use of MSCM. Water activity, protein and fat content in the mature sausages were found appropriate for these products (Acton and Dick, 1976; Acton, 1978; Terrell et al. 1978, Vansteenkiste and Van Hoof, 1979, Baumgartner et al. 1980).

Thiobarbituric acid values were taken as indicators of the development of oxidative rancidity (Townsend et al. 1978; Ockerman and Kuo, 1982). These values increased only slightly during sausage maturation (Table 2) and final values did not indicate the development of rancidity during sausage maturation (Wisniewski and Maurier, 1979; Joseph et al. 1978a; Salminen et al. 1985).

Initial nitrate levels varied from 174 to 381 ppm but these levels were reduced by day 28 to<100 ppm in Duploferment treatments. Highest values of nitrate at day 28 (which were similar to initial values) were recorded in Trumark treatments where acidification by pediococci was believed responsible for inhibition of the co-starter micrococci which normally would reduce nitrate. Nitrite levels ranged from 4.2 ppm initially to an average level of 2.1 ppm by day 28.

Sensory and instrumental analysis

An in-plant screening of manufactured products believed by the sausage maker to be inadequate was used to eliminate some treatments from sensory testing. Treatments examined organoleptically contained three levels of mechanically separated chicken meat (0, 10 or 15%) and were ripened with Trumark LT II or GdL.

While there was essentially no "case hardening" of the Trumark samples, GdL-treated samples exhibited a hardened outer edge which was significantly more pronounced in 10 and 15% MSCM formulated samples. There was no effect of MSCM on surface oiliness but samples containing MSCM in greater quantities were less tough. No significant interactions nor treatment effects were seen when greasiness was evaluated.

No differences in smoked aroma among treatments were attributible to the acidulant used but the majority of panelists felt that GdL-acidulated samples had a more intense smoked flavour. The 10 and 15% MSCM containing sausages did not differ from each other with respect to cohesiveness but the unamended treatments were significantly more cohesive than those to which MSCM was added. The firmness of control and the 10% MSCM samples were not different but 15% MSCM treatments were less firm than the others.

Conclusion

The addition of MSCM up to 15% did not present additional microbial risk in the consumption of raw ripened sausages. The use of 10% MSCM was not detectable organoleptically by a trained group of panelists when a bacterial starter culture was used in the fermentation. Nitrate was not reduced significantly in Trumark-treated samples and could have been eliminated from the formulation without a significant effect on colour.

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Table 1. Changes in meat pH during ripening of uncooked dry sausage.

%w/w) MSCM	Acidulant	Days Maturation 1	2	3	4	5	6	9	28
0	A	5.67	5.73	5.60	5.32	5.17	5.14	5.13	5.05
10	A	5.79	5.83	5.68	5.47	5.18	5.09	5.19	5.14
15	A	5.79	5.80	5.77	5.54	5.30	5.15	5.12	5.04
0	B	5.65	5.08	5.13	5.01	4.94	4.99	5.03	4.92
10	B	5.61	5.05	5.05	5.02	4.99	4.97	5.06	4.92
15	B	5.65	5.04	5.08	5.03	4.97	5.01	5.07	4.98
0	GdL	5.33	5.33	5.26	5.19	5.01	4.96	4.98	4.92
10	GdL	5.34	5.44	5.44	5.27	5.11	4.99	5.01	5.20
15	GdL	5.36	5.43	5.46	5.32	5.19	5.13	5.08	4.92

A = Duploferment starter culture
B = Trumark LT II starter culture
GdL = glucono-delta lactone

Table 2. Physicochemical characteristics of raw ripened sausages amended with mechanically separated chicken meat (MSCM)

Acidulant[a]	%(w/w) MSCM	Water Activity Day 14	Water Activity Day 28	Day 28 % (w/w) Protein	Moisture	Fat	TBA[b]
A	0	0.919	0.880	18.5	33.5	43.5	0.72
A	10	0.924	0.886	20.3	30.2	44.1	0.87
A	15	0.925	0.880	17.7	28.5	45.1	1.35
B	0	0.915	0.884	20.3	26.2	45.6	0.45
B	10	0.918	0.840	20.0	24.2	43.6	1.17
B	15	0.921	0.844	19.5	29.6	43.3	0.95
GdL	0	0.915	0.843	19.9	28.3	44.6	0.76
GdL	10	0.913	0.847	18.9	26.9	44.7	0.96
GdL	15	0.915	0.861	19.2	26.7	45.4	0.93

a) as in Table 1
b) mg malonaldehyde/kg sample

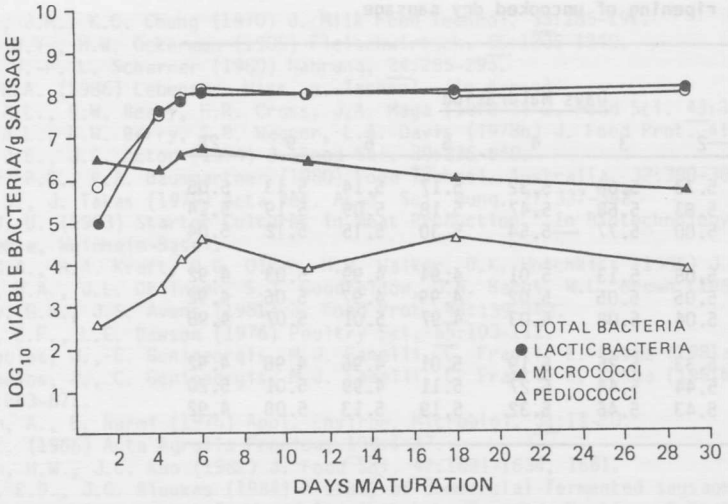

FIGURE 1. Viable bacteria in sausages inoculated with Duploferment bacterial starter culture.

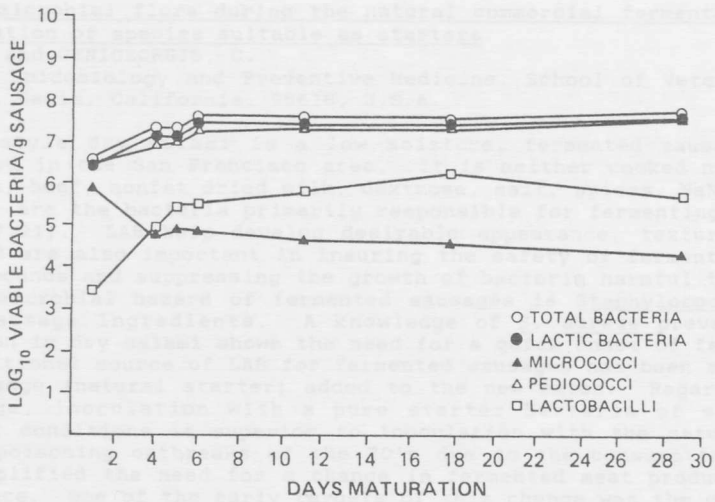

FIGURE 2. Viable bacteria in ripening sausages inoculated with Trumark LT-II starter culture.

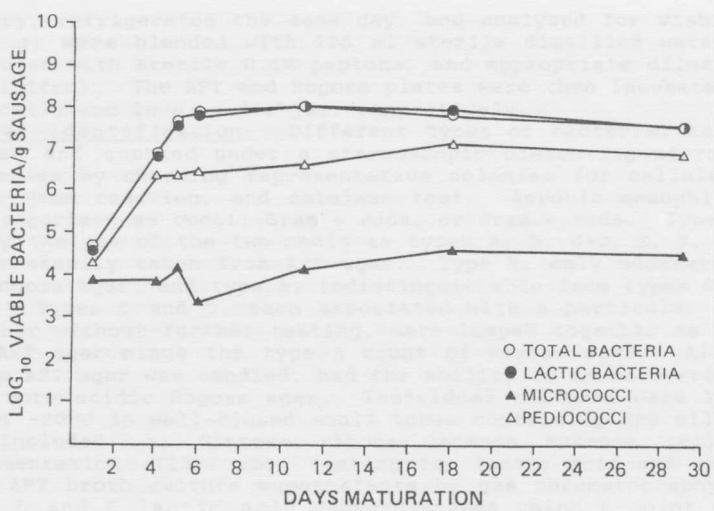

FIGURE 3. Viable bacteria in ripening sausages acidulated with glucono-delta-lactone

DAYS MATURATION