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Effect of κ-Carrageenan and Guar Gum as a Substitute for Inorganic Polyphosphate on Pork Sausages

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Abstract Guar gum and κ-carrageenan were investigated as a substitutes for phosphate in pork meat processing. Emulsion-type pork sausages were prepared in which 0.5% phosphate was used for the control, and either κ-carrageenan or guar gum were added at levels of 0.1 or 0.5% for comparison. The hydrocolloid compounds significantly enhanced water holding capacity and cooking loss. However, hardness, cohesiveness, and chewiness were not well-maintained when compared to the control; this was attributable to the altered water distribution as well as enhanced water holding capacity of the sausages by the addition of κ-carrageenan and guar gum. Furthermore, the phosphate-free sausages had similar storage stability as the phosphate-added sausage. Overall, the results suggest that κ-carrageenan or guar gum can be used in place of phosphate in conventional processing to successfully prepare phosphate-free pork sausages.

Keywords: phosphate, guar gum, κ-carrageenan, sausage

Introduction

Today, many consumers manage their health based on food choices. In fact, an 81% of consumers choose certain foods because they contain functional health ingredients or impart health benefits, such as antioxidant, cholesterol-lowering properties, etc (1,2). In meat processing, curing agent are used to improve process quality, as well as for color and flavor enhancement. Sodium chloride is a primary curing agent that plays a key role in maintaining water holding capacity (WHC) by extracting salt soluble proteins. Public health authorities, however, have recommended that individuals reduce dietary sodium intake due to the risk of developing hypertension by excessive levels in the diet (3,4). In addition, there have been attempts to reduce or eliminate all the chemical additives used in meat processing such as nitrite, sorbate, and phosphate, and to ultimately develop phosphate-free meat products (5). Phosphate, which is recognized as safe food additive, was found to increase bone disease risk in humans by influencing ionic balance (6,7). Consequently, there is increasing demand for replacement of select meat product ingredients with natural compounds; however, at the present time, no effeorts have been made to develop sodium- and phosphate- free meat products. Most studies examining sodium and phosphate replacement can be classified into two categories. Firstly, non-meat protein sources have been used as functional replacements to sodium ions by enhancing water binding capacity (8-10). These include isolated soy proteins, whey proteins, and casein. Secondly, high molecular weight hydrocolloids, including gums, pectin, cellulose, and starches were investigated for their ability to improve the final quality of low sodium or phosphate products (10-18). By replacing the functions of sodium and phosphate, Such substitutes were supposed to improve the WHC and binding properties of meat products.

Carrageenans (from red seaweed) are mixtures of several related galactans having sulfated groups. They are used extensively in food processing as thickeners or gelling agents (19) as well as in low-fat meat products as fat replacers (10,11,20,21). Guar gum functions as an economical thickener in numerous food products based on its high viscosity and solubility (19). The introduction of these hydrocolloids offers health benefits as well as affects on the physicochemical characteristics of final products (19).

Previously, using a model system, it was found that κ-carrageenan and guar gum could be used as phosphate substitutes in pork meat process (22). Therefore, the present study investigated the applicability of these compounds on a product level, in which we prepared emulsion-type pork sausages with the 2 candidate compounds and compared their characteristics to a control sausage.

Materials and Methods

Preparation of pork sausages Certified organic grade vacuum packed, refrigerated lean pork, and frozen pork backfat were obtained from Doorae Food Ltd. (Goesan, Korea). An emulsion-type pork sausage was prepared as a basic formulation with the following: lean pork (52.1%), pork backfat (22.3%), ice (18.6%), potato starch (2.2%), sodium chloride (1.5%), glucose (0.6%), and seasoning (2.7%). All the ingredients were kindly donated from Doorae Food. The κ-carrageenan (Daehung Co., Ltd., Seoul, Korea), guar gum (G4129; Sigma-Aldrich Co., St. Louis, MO, USA),

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Table 1. Experimental groups for phosphate replacement in pork sausages

Group	Treatments[1)]
T1	Control with 0.5% phosphate
T2	0.1% κ-carrageenan[2)]
T3	0.5% κ-carrageenan[2)]
T4	0.1% guar gum[2)]
T5	0.5% guar gum[2)]

[1)]NaCl (1.5%) was added to all treatments.
[2)]κ-Carrageenan and guar gum was added instead of phosphate.

and sodium tripolyphosphate (Samchun Pure Chemical Co., Ltd., Seoul, Korea) for sausage preparation were purchased as food grade. The experimental groups were summarized in Table 1. The basic sausage formulation was mixed using either phosphate or a phosphate replacer (κ-carrageenan or gum guar) in a silent cutter until the temperature of the mixture reached 12-15°C. Total emulsification time was about 3 min. The mixed emulsions were stuffed into a collagen casing (2.0 cm of diameter, Nippi Collagen Industry, Shizuoka, Japan), dried (25 min), smoked (55°C for 13 min) by sawdust, and then cooked to an internal temperature of 72°C in a smokehouse (Bastramat 1500; Bayha & Strackbein Gmbh, Amsberg, Germany). The cooked sausages were cooled with water spray and kept at 4°C for 12 hr before vacuum packaging. The samples were stored at 4°C until analysis.

Proximate analysis Moisture, protein, lipid, and ash were assayed according to the AOAC methods (23).

pH Using a homogenizer (Nihonseiki, Tokyo, Japan), 10 g of samples were homogenized in 100 mL of distilled water for 30 sec at 7,000 rpm. The pH levles of the homogenate was determined using a pH meter (Mettler Delta 340; Mettler-Toledo Ltd., Beaumont Leys Leicester, UK) (13).

Water holding capacities (WHC) The centrifugation method described by Laakkonen *et al.* (24) was used to measure WHC. Sausages samples (0.5±0.05 g) from each treatment were placed in centrifugation tube with filter units, heated for 20 min at 80°C, and then cooled for 10 min. Samples were centrifuged at 2,000×g for 10 min (4°C) and WHC was calculated as the change of sample weight.

Cooking loss Prepared sausages (25 g) from each treatment were cooked for 2 min at 100°C and then cooled down to room temperature. Cooking loss was calculated by weight difference of samples (10).

Textural properties Sausages samples were cut into 1×2×1 cm (width×length×height) pieces and were then evaluated by a mastication test using a rheometer (Compac-100; Sun Scientific Co., Tokyo, Japan) under the following operational conditions: circle-type adaptor, adaptor area of 0.79 cm^2; table speed of 60 mm/min, and compression at 50% (25). The textural properties were expressed as hardness, cohesiveness, springiness, and chewiness using the Rheology Data System (Ver 2.01, Sun Scientific Co.).

Microbiological stability A 10 g sample of each type of sausage was mixed with 90 mL of 0.1% peptone solution and homogenized by a Stomach blender (model 400; Seward, London, UK). The homogenate was serially diluted and spread on the plate count agar (PCA, Difco Laboratories, Detroit, MI, USA). Plates were incubated at 37°C for 48 hr and then microbial count were performed. The levels are reported as colony forming unit (CFU) per gram of sample.

Lipid oxidation A thiobarbituric acid (TBA) test was used to determine the degree of lipid oxidation of sausage samples during storage at 4°C (26). The samples (10 g) was homogenized with 15 mL of cold 10% perchloric acid and 25 mL of distilled water by Stomach Lab blender (model 400; Seward) for 10 sec at 1,000 rpm, and the homogenate was filtered with filter paper (No. 1; Whatman International Ltd., Kent, UK). The filtrate (5 mL) were mixed, vortexed with 5 mL of 0.02 M Thiobarbituric acid (TBA) solution and then kept in under dark-cold condition for 16 hr. The upper layer of each sampe mixture was read spectorphotometrically at 529 nm and lipid oxidation was expressed as mg malonaldehyde per kg sample.

Volatile basic nitrogen (VBN) value To investigate the influence on the protein deterioration, VBN was determined by the method of Conway (27,28) using a Conway unit (Shibata Co., Ltd., Tokyo, Japan). VBN was expressed in terms of mg VBN per 100 g sample.

Sensory evaluation Well-trained in-house tasting panelists (n=5) evaluated sensory attributes of juiciness, cohesiveness, chewiness, and overall acceptability, using on a 5-point scale: 1, very poor; 2, poor; 3, common; 4, good; 5, very good (13). The samples were evaluated independently by the panelists 3 different times.

Statistical analyses All data were analyzed using the ANOVA procedure of SAS (SAS Institute Inc., Cary, NC, USA) and the significance was defined at $p<0.05$.

Results and Discussion

Basic quality of pork sausages The proximate compositions of phosphate-free pork sausages are shown in Table 2. No significant difference were observed among the groups. Table 3 summarizes the treatment effects on the pH, WHC, and cooking loss. pH is one of the major factors affecting the WHC of meat (29). The κ-carrageenan (T2, T3) and guar gum (T4, T5) additions significantly lowered the pH of the sausages by 0.1-0.2 unit as compared to the phosphate control group (T1). In contrast, Park *et al.* (22) reported that these compounds offered no significant changes in model system. Since a major role of phosphate in meat processing is to enhance protein binding by increasing pH, examinations of WHC, and cooking loss were required to confirm the effects of the phosphate substitutes. The guar gum groups (T4, T5) exhibited highest WHC with lowest cooking loss values as compared to the other groups. Also, WHC increased in proportion to the κ-carrageenan concentration, and at 0.5% level (T3) showed similar values to those of guar gum groups. In

Table 2. Proximate analysis of pork sausages with different formulation to replace phosphate

Group[1)]	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
T1	58.77±0.81[2)]	19.98±0.76	18.94±0.41	2.28±0.32
T2	58.25±0.53	20.20±0.76	19.46±0.50	2.07±0.10
T3	58.49±0.04	21.60±1.25	17.59±1.29	2.31±0.08
T4	57.92±0.33	18.58±1.71	21.29±1.84	2.18±0.09
T5	58.05±1.04	20.47±1.20	19.13±0.24	2.32±0.24

[1)]See Table 1.
[2)]The experiments was done by triplicate and the values were not significantly different ($p<0.05$).

addition, cooking loss was lowered in κ-carrageenan or guar gum treated groups (T2-T5) compared to control ($p<0.05$). According to the results, the differences in pH caused by substituting κ-carrageenan and guar gum for phosphate didn't influence on the basic quality of sausages, but may have enhanced quality.

Textural properties Table 4 shows the added hydrocolloids' effects on the textural properties of pork sausage. In general, the control (T1) presented higher values than the κ-carrageenan (T2, T3) and guar gum (T4, T5) groups. The treatment groups (T2-T5) showed lower shear force and hardness than the control ($p<0.05$), but no differences were observed by the hydrocolloid concentration. Cohesiveness and chewiness were also significantly lower in the treatment groups, whereas springiness was not affected by the elimination of phosphate from the formulation. The guar gum added groups (T4, T5) showed the lowest textural parameters; this is in contrast to their offering of the highest WHC values and can be explained by the basic properties of guar gum. Although guar gum is widely used in food formulations, partly due to its high solubility over broad temperature range as well as high pH-temperature stability, it is recognized as unsuitable to help gelation (19). Thus, it was expected that guar gum's main function would be to maintain WHC in the pork sausages, which would lower the hardness arrtibutable to higher water content.

Table 3. Influence of κ-carrageenan and guar gum on water holding capacity (WHC) and cooking loss of pork sausages

Group[1)]	pH	WHC (%)	Cooking loss (%)
T1	5.99±0.10$^{a2)}$	78.08±4.13^{ab}	2.63±0.43^{a}
T2	5.80±0.02^{bc}	72.93±5.41^{b}	1.34±0.16^{b}
T3	5.72±0.01^{c}	78.01±3.76^{ab}	1.28±0.17^{b}
T4	5.84±0.02^{bc}	80.72±5.44^{a}	1.21±0.14^{b}
T5	5.86±0.04^{b}	80.66±4.09^{a}	1.31±0.11^{b}

[1)]See Table 1.
[2)]The experiments was done by triplicate; $^{a-c}$different letters in a column indicate significant difference ($p<0.05$).

Sensory evaluation For the most part, the sensory attributes of products will follow patterns that are similar to the textural analysis results. And the altered distribution of water of sausages by adding the hydrocolloids would supposedly influence their sensory properties. Overall, the control offered better sensory characteristics than the other groups; however, the groups containing the hydrocolloids had higher juiciness scores than the control due to increased WHC (Table 5).

Storage stability of pork sausages Table 6 presents the microbial analysis results for the sausages during storage at 4°C. The additions of κ-carrageenan and guar gum did not

Table 4. Influence of κ-carrageenan and guar gum on textural properties of pork sausages

Group[1)]	Shear force (kg)	Hardness (kg)	Cohesiveness (%)	Springiness (%)	Chewiness
T1	0.37±0.06$^{a2)}$	0.54±0.07^{a}	0.37±0.07^{a}	0.67±0.03^{a}	0.15±0.02^{a}
T2	0.24±0.06^{b}	0.24±0.05^{bc}	0.24±0.03^{bc}	0.71±0.07^{a}	0.04±0.005^{bc}
T3	0.20±0.02^{b}	0.24±0.08^{bc}	0.23±0.01^{bc}	0.63±0.03^{a}	0.03±0.009^{c}
T4	0.22±0.03^{b}	0.19±0.01^{c}	0.24±0.06^{bc}	0.66±0.13^{a}	0.03±0.01^{c}
T5	0.19±0.04^{b}	0.21±0.03^{c}	0.19±0.09^{c}	0.54±0.22^{a}	0.02±0.02^{c}

[1)]See Table 1.
[2)]The experiments was done by triplicate; $^{a-c}$different letters in a column indicate significant difference ($p<0.05$).

Table 5. Sensory evaluation of pork sausages prepared with κ-carrageenan and guar gum

Group[1)]	Juiciness	Cohesiveness	Chewiness	Total acceptability
T1	2.77±0.50$^{a2)}$	3.38±0.69^{a}	3.38±0.69^{a}	3.27±0.66^{a}
T2	3.16±0.28^{a}	2.33±0.57^{b}	2.16±0.28^{bc}	2.16±0.76^{bc}
T3	3.00±0.00^{a}	2.33±0.28^{b}	2.33±0.28^{b}	2.16±0.76^{bc}
T4	3.16±0.28^{a}	1.83±1.04^{bc}	1.83±1.04^{bc}	2.00±0.86^{bc}
T5	2.83±0.76^{a}	1.83±1.04^{bc}	1.83±1.44^{bc}	1.83±1.04^{c}

[1)]See Table 1.
[2)]Five-membered panel evaluated the sensory attribute by triplicate; $^{a-c}$different letters in a column indicate significant difference ($p<0.05$).

Table 6. Changes of total microbial population changes (CFU/g) in pork sausages prepared with κ-carrageenan and guar gum during storage at 4°C

Group[1)]	0 day	3 day	7 day
T1	2.86±0.10[2)]	2.49±0.16	2.38±0.55
T2	2.38±0.12	2.03±0.05	2.92±0.10
T3	2.69±0.08	3.23±0.01	3.14±0.01
T4	2.61±0.02	3.09±0.07	2.65±0.06
T5	2.51±0.04	2.61±0.01	2.81±0.21

[1)]See Table 1.
[2)]The experiments was done by triplicate and the values were not significantly different ($p<0.05$).

Table 7. 2-Thiobarbituric acid (TBA) values of pork sausages prepared with κ-carrageenan and guar gum during storage at 4°C (mg malonaldehyde/kg sample)

Group[1)]	0 day	3 day	7 day
T1	0.21±0.07$^{c2)}$	0.21±0.05^{d}	0.18±0.07^{e}
T2	0.32±0.01^{b}	0.30±0.01^{c}	0.25±0.01^{d}
T3	0.25±0.01^{c}	0.30±0.02^{c}	0.33±0.01^{c}
T4	0.35±0.01^{b}	0.37±0.01^{b}	0.37±0.01^{b}
T5	0.61±0.03^{a}	0.62±0.02^{a}	0.63±0.06^{a}

[1)]See Table 1.
[2)]The experiments was done by triplicate; $^{a-c}$different letters in a column indicate significant difference ($p<0.05$).

Table 8. Volatile basic nitrogen (VBN) values of pork sausages prepared with κ-carrageenan and guar gum during storage at 4°C (mg VBN/100 g sample)

Group[1)]	0 day	3 day	7 day
T1	7.39±1.49$^{bc2)}$	7.80±1.44^{a}	9.06±1.51^{ah}
T2	7.73±1.25^{ab}	6.95±0.88^{a}	8.78±0.96^{ab}
T3	8.19±0.96^{ab}	7.77±0.41^{a}	8.32±1.66^{ab}
T4	8.96±0.58^{ab}	7.13±1.71^{a}	10.80±1.45^{a}
T5	9.38±0.77^{a}	9.88±2.47^{a}	9.79±0.79^{ab}

[1)]See Table 1.
[2)]The experiments was done by triplicate; $^{a-c}$different letters in a column indicate significant difference ($p<0.05$).

affect the microbial stability of the sausages, and there were no significant changes in the samples over 7 days at 4°C.

The TBA values indicating the degree of lipid oxidation are shown in Table 7. The values within each group did not increase significantly during storage, suggesting that the rate of lipid oxidation was not influenced by the presence of phosphate, κ-carrageenan, or guar gum. The higher initial TBA values of the samples reflect possible increases of lipid oxidation during meat processing by changes in water distribution, and thereby, increased possibility of reactants. However, all the samples presented TBA values under a level for safe eating.

Increased VBN values, which result from protein decomposition during storage, can be an index of freshness for meat products. The VBN values of the samples slightly increased after 3 days, but it did change significantly within groups, similar to the TBA values (Table 8). The treatment groups (T2-T5) showed higher initial levels as compared to the control (T1), and did not increase over the storage period. However, in the control, VBN increased on day 7, but there was no significant difference. The values of all samples in each group did not exceed 20 mg/100 g of sample, which is described as the necessary level to detect meat spoilage (30).

In conclusion, carrageean and guar gum were successfully applied in the manufacture of emulsion-type pork sausages, substituting phosphate without changing storage stability. Despite improving the basic characteristics of the sausages, such as WHC and cooking loss, there was reduced textural preference for the samples containing carrageenan and guar gum, raising the need for successive subtle asjustments to the process, so that ultimately a 100% phosphate-free sausage can be developed in the in future.

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