

Original article

Fructooligosaccharides as a fat replacer in fermented cooked sausages

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Summary Fermented cooked sausages with a 50% reduction in pork back fat and addition of 0%, 3%, 6% or 9% of fructooligosaccharides (FOS) were produced and studied during manufacturing and storage. Their production was monitored by physicochemical (pH, water activity, weight loss, proximate composition, colour and texture profiles) and microbiological analysis (aerobic mesophiles, lactic acid bacteria, and total and faecal coliforms). During storage, it was evaluated the sensory properties, stability to lipid oxidation and microbiological safety. The final fat content of the control was 27.54%. F0, F3, F6 and F9 treatments had final fat contents of 17.63%, 17.55%, 17.91% and 17.59%, respectively, representing an approximately 40% reduction in the fat content. The simple reduction in pork back fat without fat substitute adversely affected the technological and sensory properties of the fermented cooked sausages, but the addition of FOS suppressed the defects caused by the fat reduction. The content of FOS did not changed during storage, indicating that this functional prebiotic compound can be used for developing of reduced fat fermented meat products.

Keywords Fermentation, meat products, prebiotics.

Introduction

Several studies have linked high fat consumption with an increased incidence of cardiovascular disease, obesity and certain cancers (Jiménez-Colmenero, 1996; Hooper *et al.*, 2001; Rothstein, 2006; Zhang *et al.*, 2010). Public health agencies have recommended reducing fat consumption, especially saturated fat, which has considerably increased the demand for meat products with reduced fat in many countries. Thus, it has been essential for the meat industry to reduce the fat content of meat products to satisfy the dietary and organoleptic demands of consumers (Sandrou & Arvanitoyannis, 2000).

Fermented sausages are characterised by high levels of fat. Generally, 15–20% fat is added during their preparation, but, because of the dehydration that occurs during manufacturing, the fat content of the final product can reach values of more than 40% (Wirth, 1989). Reducing the fat content of fermented sausages, despite its nutritional significance, may result in sensory issues because the lipolytic reactions that occur during

maturation produce compounds that benefit the flavour and aroma of fermented sausages (Ordóñez *et al.*, 1999). In addition, fat also improves the tenderness, juiciness and overall palatability (Wirth, 1989). Reducing fat also adversely affects the technological quality of fermented sausages because fat controls the release of moisture from the inner layers of the product. This process is crucial for efficient fermentation (Wirth, 1989). Moreover, a decrease in fat content may increase weight loss during manufacturing (Papadima & Bloukas, 1999; Muguerza *et al.*, 2002).

Recently, the search for fat substitutes has been the focus of many studies promoting the health appeal of meat products. Various dietary fibres have been used as fat substitutes in meat products (Chang & Carpenter, 1997; Hughes *et al.*, 1997; Griguelmo *et al.*, 1999; Pappa *et al.*, 2000; Mendoza *et al.*, 2001; García *et al.*, 2002). This strategy adds another important nutritional benefit because an increase in dietary fibre consumption is considered to be a method to reduce the risk of obesity and cardiovascular disease and the onset of colon cancer (Beecher, 1999; Desmedt & Jacobs, 2001).

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Fructooligosaccharides (FOS) are soluble dietary fibres that stimulate the growth of bifidobacteria in the digestive tract (Yun, 1996; Arvanitoyannis & Van Houwelingen-Koukialiaroglou, 2005). Because of the prebiotic properties of FOS, FOS consumption may contribute to a reduction in total serum cholesterol and an improvement of the immune system (Sgarbieri & Pacheco, 1999; Arvanitoyannis & Van Houwelingen-Koukialiaroglou, 2005; Chakraborty *et al.*, 2006; Dou *et al.*, 2009). These fibres have a neutral taste and are stable over wide pH and temperature ranges. Therefore, they have great potential for applications in the meat industry. FOS have successfully been used in some meat products (Cáceres *et al.*, 2004; Salazar *et al.*, 2009).

However, the use of FOS as a fat substitute in fermented cooked sausages has not been studied until now, and its stability during storage has not been evaluated. Therefore, the objective of this study was to evaluate the use of FOS as a fat substitute in fermented cooked sausages. Their physicochemical, microbiological and sensory quality parameters were evaluated during manufacturing and storage.

Materials and methods

Formulation and processing

Five treatments were developed to determine the influence of reducing the fat content and adding FOS on the quality of fermented cooked sausages (Table 1). The raw materials were grounded using a 8-mm disc and mixed with the following ingredients: sodium chloride (2.5%), glucose (1.0%), sodium nitrite (0.015%), white pepper (0.2%), garlic powder (0.3%), nutmeg (0.02%), sodium erythorbate (0.025%) and a Bactoferm TPS-X (Chr. Hansen, São Paulo, SP, Brazil) starter culture composed of *Staphylococcus xylosum* and *Pediococcus pentosaceus* (0.25%). In the treatments with reduced fat, 0 (F0), 3 (F3), 6 (F6) or 9% (F9) FOS (NutraFlora®; Corn Products Brazil, São Paulo, Brazil) were added;

Table 1 Formulation of fermented cooked sausage treatments

| | Treatments (%) | | | | |
|---------------|----------------|------|------|------|------|
| | Control | F0 | F3 | F6 | F9 |
| Pork | 65 | 72.5 | 72.5 | 72.5 | 72.5 |
| Beef | 20 | 20 | 20 | 20 | 20 |
| Pork back fat | 15 | 7.5 | 7.5 | 7.5 | 7.5 |
| FOS | 0 | 0 | 3 | 6 | 9 |

Pork (moisture: 73.51% ± 0.65; protein: 22.73% ± 0.73; fat: 4.32 ± 0.35);

Beef (moisture: 70.95% ± 0.52; protein: 23.16% ± 0.22; fat: 4.45 ± 0.39);

Pork back fat (moisture: 13.39% ± 0.68; protein: 8.02% ± 0.21; fat:

78.50 ± 0.41).

FOS, fructooligosaccharides.

Table 2 Processing conditions for the fermented cooked sausages

| | Temperature (°C) | Relative humidity (%) | Air movement (m s ⁻¹) |
|-----------------------|------------------|-----------------------|-----------------------------------|
| Fermentation (24 h) | 28 ± 0.1 | 85–90 | 0.5 |
| Heat processing (min) | | | |
| 60 | 50 ± 0.1 | 85–90 | – |
| 30 | 60 ± 0.1 | 85–90 | – |
| * | 70 ± 0.1 | 85–90 | – |
| Ripening (144 h) | 15 ± 0.1 | 65–75 | 0.5 |
| Storage (60 days) | 4 ± 0.1 | 70–75 | – |

*Until all the treatments reached an internal temperature of 62 °C.

the FOS used were composed of 33.8% 1-kestose, 49.7% nystose and 12.5% fructofuranosyl nystose.

After mixing, the mass of meat was grounded using a 3-mm disc and stuffed in collagen casings of 50 mm diameter. The pieces were placed into a maturation chamber at 28 ± 0.1 °C with a relative humidity between 85% and 90%, until all the treatments reached a pH of 5.2 or less (Table 2). For heat processing, the pieces were placed in the smokehouse and initially heated at 50 °C for an hour, 60 °C for 30 min, then at 70 °C until an internal temperature of 62 °C was attained. Following heat processing, the fermented cooked sausages were cooled to 20 °C and placed in a 15 °C drying room (relative humidity 65–75%), until all of the treatments reached water activity values of less than 0.92. Next, the casings were removed, and the samples to be held at 4 °C (±1 °C) with a relative humidity 70–75% for 60-day evaluations were packaged in low density (Unipac/Univac B320) barrier pouches, using a vacuum sealer (Minivac CU18; Selovac, São Paulo, SP, Brazil), and the absolute pressure of the vacuum was 98.7 kPa. The pouches were 90 µm with an oxygen permeability of 40 cm³ m⁻² day⁻¹ atm⁻¹ at 77% relative humidity (RH), 23 °C.

Analysis performed during manufacturing

Physicochemical analysis

The pH was measured using a pH MA-130 meter (Mettler Toledo Indústria and Comércio Ltda, SP, Brazil). Water activity (Aw) was measured using a Decagon Aqualab apparatus (Decagon Devices Inc., Pullman, WA, USA). The pH and Aw were determined before the stuffing process and 24, 36, 72 and 168 h after production using three pieces per treatment, and each analysis was performed in triplicate. The weight loss was determined by the weight difference among ten sausages just after the stuffing process and after the end of sausage production. The proximate composition (moisture, protein, fat and ash contents) was determined before the stuffing process and at the end of process

(168 h) according to the Association of Official Analytical Chemists (AOAC, 2005). All tests were performed in triplicate using three sausage samples per treatment.

Microbiological analysis

The microbiological characteristics during manufacturing (at 0, 24, 36 and 168 h) were evaluated in duplicate using three pieces per treatment, following the methodology proposed by Downes & Ito (2001). Sample portions (25 g) were homogenised in 225 mL of 0.1% peptone water (Oxoid Unipath Ltd., Basingstoke, Hampshire, UK), and serial dilutions were used for the microbiological analysis. Mesophilic aerobic bacteria were quantified using standard plate-count agar (Oxoid) at 35 °C for 48 h. Lactic acid bacteria (LAB) were quantified using De Man, Rogosa and Sharpe (MRS) agar (Oxoid) at 37 °C for 48 h. The total coliforms were quantified in crystal agar neutro-bile violet-red (Oxoid) at 37 °C for 24 h. Faecal coliforms were quantified in EC broth (Oxoid) at 45 °C for 48 h.

Analysis performed during storage

Physicochemical analysis

The degree of lipid oxidation was evaluated by measuring 2-thiobarbituric reactive substances (TBARS) values according to Bruna *et al.* (2001), using trichloroacetic acid instead of perchloric acid as solvent. Because of the presence of FOS in the samples, the incubation temperature was reduced to 40 °C, and the time was increased to 90 min, as proposed by Wang *et al.* (2002). Results were expressed as µg of malondialdehyde (MDA) g⁻¹ sample. The TBARS index was assessed immediately after completing manufacturing (day 0) and after 15, 30, 45 and 60 days of storage using three pieces per treatment.

The texture profile analysis (TPA) parameters were determined after completing manufacturing (day 0) and after 15, 30, 45 and 60 days of storage, using a TA-TX2 Texture Analyzer (Stable Micro Systems Ltd., Surrey, UK) with a load cell of 10 kg. Fifteen cylinders per batch were used to evaluate the texture. The samples, approximately 2 cm thick and 2 cm in diameter, were axially compressed into two consecutive cycles of 20% of compression, with a 30-mm diameter probe, at a constant speed of 1 mm s⁻¹. The TPA parameters of hardness (the peak force during the first compression cycle), springiness (the height that the food recovers during the time that elapses between the end of the first bite and the start of the second bite), cohesiveness (the ratio of the positive force area during the second compression portion to the positive force area during the first compression) and adhesiveness (negative force area for the first compression, representing the work necessary to pull the probe away from the sample) were determined.

The colour determination was performed after completing manufacturing (day 0) and after 15, 30, 45, and

60 days of storage, with a Hunter Lab colorimeter (Colourquest-II; Hunter Associates Laboratory Inc., Reston, Virginia, USA) using a 10-mm port size, illuminant D₆₅ and a 10° standard observer. CIELAB L*, a* and b* values were determined as indicators of lightness, redness and yellowness. Colour variables were measured at four points on the central part of the cut surface of three slices of the five sausages.

The FOS concentration was determined before (day 0) and after 60 days of storage, according to the methodology proposed by Horwitz (2000). The analyses were performed in triplicate using three pieces per treatment.

Microbiological analysis

Mesophilic aerobic bacteria, lactic acid bacteria and total and faecal coliforms were determined during storage (at 0, 15, 45 and 60 days) according to the methodology described by Downes & Ito (2001). Three pieces per treatment were used, and the analyses were performed in duplicate.

Consumer study

This study protocol was approved by the Ethics in Research Committee of the University of Campinas (SP, Brazil) under number 122/2010. The consumer study was conducted by 60 untrained panellists recruited among students, faculty and staff members from the university campus whose ages ranged from 18 to 60 years. They were asked to express their opinion of the colour, aroma, taste, texture and overall acceptability of the product. All data were recorded on a questionnaire designed to indicate the degree of likeability for each sample using a non-structured scoring scale of nine centimetres (0 = disliked extremely and 9 = liked extremely) (Meilgaard *et al.*, 1999). The samples were evaluated immediately after manufacturing was complete (day 0) and after 15, 30 or 45 days in storage. Samples were evaluated by each consumer in a monadic order and were presented to the panellists balancing the first-order and carry-over effects according to Macfie *et al.* (1989).

Statistical analysis

The data were evaluated with a variance analysis (ANOVA). The averages were compared by Tukey's test at a confidence level of 5% ($P \leq 0.05$) using the SPSS statistical package (SPSS Inc., Chicago, IL, USA).

Results and discussion

Analysis performed during manufacturing

Physicochemical analysis

The physicochemical characteristics of fermented cooked sausages with reduced fat content and added

Table 3 Physicochemical characteristics of the fermented cooked sausages with reduced fat content and added fructooligosaccharides during manufacturing

| Hours | Control | F0 | F3 | F6 | F9 |
|-----------------|----------------------------|----------------------------|----------------------------|----------------------------|---------------------------|
| pH | | | | | |
| 0 | 6.20 ± 0.07 ^{ab} | 6.24 ± 0.02 ^a | 6.11 ± 0.03 ^b | 6.26 ± 0.10 ^a | 6.02 ± 0.03 ^c |
| 24 | 4.81 ± 0.04 ^a | 4.73 ± 0.02 ^c | 4.79 ± 0.02 ^a | 4.74 ± 0.02 ^{bc} | 4.77 ± 0.02 ^{ab} |
| 36 | 4.88 ± 0.04 ^{bc} | 4.97 ± 0.01 ^a | 4.86 ± 0.01 ^c | 4.90 ± 0.01 ^b | 4.88 ± 0.03 ^{bc} |
| 168 | 4.76 ± 0.02 ^c | 4.88 ± 0.02 ^a | 4.73 ± 0.02 ^{cd} | 4.72 ± 0.01 ^d | 4.81 ± 0.04 ^b |
| Aw | | | | | |
| 0 | 0.978 ± 0.01 ^a | 0.972 ± 0.00 ^a | 0.964 ± 0.01 ^a | 0.969 ± 0.01 ^a | 0.962 ± 0.01 ^a |
| 24 | 0.978 ± 0.01 ^a | 0.972 ± 0.01 ^a | 0.965 ± 0.00 ^{ab} | 0.960 ± 0.00 ^b | 0.960 ± 0.01 ^b |
| 36 | 0.951 ± 0.00 ^{ab} | 0.952 ± 0.00 ^a | 0.948 ± 0.00 ^{ab} | 0.951 ± 0.00 ^{ab} | 0.945 ± 0.00 ^b |
| 72 | 0.9371 ± 0.01 ^a | 0.930 ± 0.00 ^{ab} | 0.935 ± 0.00 ^a | 0.918 ± 0.00 ^b | 0.936 ± 0.00 ^a |
| 168 | 0.903 ± 0.00 ^a | 0.902 ± 0.00 ^a | 0.902 ± 0.00 ^a | 0.897 ± 0.00 ^b | 0.898 ± 0.00 ^b |
| Weight loss (%) | | | | | |
| 168 | 38.04 ± 1.03 ^b | 40.05 ± 0.44 ^a | 36.42 ± 1.19 ^{bc} | 36.32 ± 1.78 ^{bc} | 35.12 ± 0.83 ^c |
| Moisture (%) | | | | | |
| 0 | 62.05 ± 0.10 ^b | 64.38 ± 0.22 ^a | 61.49 ± 0.99 ^b | 60.44 ± 0.12 ^c | 59.03 ± 0.18 ^d |
| 168 | 41.06 ± 0.12 ^b | 42.91 ± 0.22 ^a | 40.59 ± 0.11 ^c | 40.64 ± 0.29 ^c | 40.11 ± 0.14 ^c |
| Protein (%) | | | | | |
| 0 | 20.27 ± 0.27 ^a | 20.36 ± 0.89 ^a | 19.31 ± 0.37 ^a | 19.41 ± 0.42 ^a | 17.74 ± 0.59 ^a |
| 168 | 32.54 ± 0.34 ^a | 29.26 ± 1.56 ^b | 28.93 ± 0.40 ^b | 30.04 ± 0.87 ^b | 28.23 ± 0.43 ^b |
| Fat (%) | | | | | |
| 0 | 19.42 ± 0.29 ^a | 11.87 ± 0.15 ^{cd} | 13.23 ± 0.35 ^b | 12.37 ± 0.22 ^c | 11.21 ± 0.30 ^d |
| 168 | 27.54 ± 1.00 ^a | 17.63 ± 0.29 ^b | 17.55 ± 0.24 ^b | 16.91 ± 0.20 ^b | 16.59 ± 0.42 ^b |
| Ash (%) | | | | | |
| 0 | 3.12 ± 0.25 ^a | 3.22 ± 0.05 ^a | 3.21 ± 0.04 ^a | 2.97 ± 0.03 ^a | 2.95 ± 0.05 ^a |
| 168 | 5.24 ± 0.05 ^b | 5.64 ± 0.16 ^a | 5.16 ± 0.07 ^b | 4.91 ± 0.08 ^c | 4.74 ± 0.05 ^c |

*Values represent the mean (±standard deviation). Means accompanied by the same letter on the same line do not present a statistically significant difference ($P > 0.05$) according to Tukey's test. The following treatments were used: Control (15% fat); F0 (7.5% fat); F3 (7.5% fat + 3% FOS); F6 (7.5% fat + 6% FOS); F9 (7.5% fat + 9% FOS).

FOS, fructooligosaccharides.

FOS are shown in Table 3. Changes in both pH and Aw were occurred, as expected for this type of meat product (Vural, 1998; Campagnol *et al.*, 2011). The initial pH ranged from 6.02 to 6.26, and after 24 h of fermentation, the pH was <5.00 in all of the treatments. This acidification, which is caused by the accumulation of lactic acid from lactic acid bacteria, is very important for the microbiological safety of the product because it causes reduction or inhibition of numerous food-degrading and pathogenic microorganisms (Leroy *et al.*, 2006). After manufacturing (168 h), the pH values ranged from 4.72 to 4.88, and the Aw values decreased from approximately 0.97 at time 0 to approximately 0.90 after processing (168 h). Reducing the fat did not significantly influence the final Aw values; this was also observed by Mendoza *et al.* (2001), García *et al.* (2002) and Olivares *et al.* (2010). However, adding 6% or 9% FOS resulted in significantly lower Aw values ($P \leq 0.05$).

The weight loss of the fermented cooked sausages ranged from 35.12% to 40.05% (Table 3). Although the weight loss was close to the 30–40% levels that were recommended by Rust (1994) and are ideal for this type

of meat product, some differences were observed between the control and the reduced fat treatments. The treatment with a 50% reduction in pork back fat without added FOS (F0) exhibited significantly more weight loss than the control, which was observed in several studies with similar goals (Papadima & Bloukas, 1999; Mendoza *et al.*, 2001; García *et al.*, 2002; Muguierza *et al.*, 2002; Olivares *et al.*, 2010). Conversely, the weight loss for treatments with a 50% pork back fat reduction and 3%, 6% or 9% added FOS (F3, F6 and F9) did not significantly differ from the control.

The initial and final moisture, protein and ash contents of all of the treatments were consistent with other studies in the literature for this type of meat product (Fernández *et al.*, 1995; Mendoza *et al.*, 2001) (Table 3). The final fat content of the control was 27.54%, which was similar to traditional fermented sausages (Fernández *et al.*, 1995; Mendoza *et al.*, 2001). Conversely, the F0, F3, F6 and F9 treatments had final fat contents of 17.63%, 17.55%, 17.91% and 17.59%, respectively, representing an approximately 40% reduction in the fat content. This provides healthier nutritional properties for the reformulated products when

Table 4 Microbiological characteristics (log CFU g⁻¹) of the fermented cooked sausages with reduced fat content and added fructooligosaccharides during manufacturing

| Hours | Control | F0 | F3 | F6 | F9 |
|-----------------------------|---------------------------|--------------------------|---------------------------|---------------------------|---------------------------|
| Mesophilic aerobic bacteria | | | | | |
| 0 | 6.77 ± 0.07 ^{a*} | 6.77 ± 0.09 ^a | 6.78 ± 0.06 ^a | 6.90 ± 0.13 ^a | 6.85 ± 0.10 ^a |
| 24 | 8.68 ± 0.06 ^a | 8.55 ± 0.14 ^a | 8.55 ± 0.06 ^a | 8.65 ± 0.07 ^a | 8.66 ± 0.04 ^a |
| 36 | 6.09 ± 0.11 ^a | 5.89 ± 0.14 ^a | 5.30 ± 0.14 ^b | 5.87 ± 1.24 ^{ab} | 5.26 ± 0.93 ^b |
| 168 | 5.76 ± 0.07 ^b | 5.87 ± 0.13 ^a | 5.60 ± 0.05 ^c | 5.77 ± 0.02 ^b | 5.94 ± 0.04 ^a |
| Lactic acid bacteria | | | | | |
| 0 | 6.65 ± 0.10 ^c | 6.66 ± 0.06 ^c | 7.04 ± 0.05 ^a | 6.88 ± 0.08 ^b | 6.94 ± 0.00 ^{ab} |
| 24 | 8.88 ± 0.04 ^a | 8.43 ± 0.13 ^d | 8.54 ± 0.02 ^{cd} | 8.68 ± 0.08 ^{bc} | 8.70 ± 0.04 ^b |
| 36 | 6.04 ± 0.05 ^a | 6.00 ± 0.03 ^a | 5.16 ± 0.02 ^c | 5.83 ± 0.02 ^b | 6.08 ± 0.00 ^a |
| 168 | 5.78 ± 0.07 ^b | 5.79 ± 0.13 ^b | 5.60 ± 0.05 ^c | 5.77 ± 0.02 ^b | 5.98 ± 0.04 ^a |
| Total coliforms | | | | | |
| 0 | 4.53 ± 0.23 ^a | 4.75 ± 0.04 ^a | 4.72 ± 0.47 ^a | 4.94 ± 0.46 ^a | 4.89 ± 0.04 ^a |
| 24 | 3.86 ± 0.03 ^a | 3.99 ± 0.12 ^a | 3.63 ± 0.02 ^a | 3.73 ± 0.09 ^a | 3.87 ± 0.12 ^a |
| 36 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |
| 168 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |

*Values represent the mean (±standard deviation). Means accompanied by the same letter on the same line do not present a statistically significant difference ($P > 0.05$) according to Tukey's test. The following treatments were used: Control (15% fat); F0 (7.5% fat); F3 (7.5% fat + 3% FOS); F6 (7.5% fat + 6% FOS); F9 (7.5% fat + 9% FOS). FOS, fructooligosaccharides.

compared to the control treatment, which was representative of commercial samples sold for consumption.

Microbiological analysis

The microbiological characteristics of fermented cooked sausages with reduced fat content and added FOS are shown in Table 4. The shifts in the microbiota in all of the treatments showed patterns that were similar to those of fermented sausages (Ordóñez *et al.*, 1999; García *et al.*, 2002), which demonstrates that a reduction in pork back fat and the addition of FOS do not influence microbial growth during manufacturing. The counts of aerobic mesophilic bacteria and lactic acid bacteria were similar throughout the manufacturing process. After 24 h of fermentation, these organisms reached counts of more than 8.0 logs of CFU g⁻¹; after cooking, there was a reduction of approximately 2 logs in all of the treatments. The initial values of the total coliforms were low, and faecal coliforms were not detected, demonstrating good hygiene and sanitary quality of the raw material in addition to an adequate technological process (Table 2). The total coliforms were eliminated in all of the treatments after 36 h of manufacturing. This was attributed to the acidification of the product and the cooking process (Lisazo *et al.*, 1999; Gonzáles-Fernández *et al.*, 2006).

Analysis performed during storage

Physicochemical analysis

2-thiobarbituric reactive substances values were measured over the 60 days of storage as indicators of the

extent of lipid oxidation (Table 5). This phenomenon is a major cause of reduced shelf life in fermented meat products (Li *et al.*, 2001), and TBARS values of more than 1.00 µg of malonaldehyde g⁻¹ of sample can be harmful to consumer health (Baka *et al.*, 2011). In this study, the TBARS values increased during storage in all of the treatments ($P \leq 0.05$). The formulations with reduced fat content (F0, F3, F6 and F9) had similar TBARS values throughout the storage period, which demonstrates that the effect of adding FOS on lipid oxidation is not significant. The control formulation showed higher TBARS values compared to treatments with reduced fat at the end of manufacturing (day 0) and after 45 and 60 days of storage ($P \leq 0.05$). Increased lipid oxidation in fermented sausages with increased fat levels was also reported by Soyer & Ertas (2007), Liaros *et al.* (2009) and Olivares *et al.* (2011).

A 50% reduction in pork back fat (F0) caused a significant increase in hardness (Table 5). Similar results have been reported by several authors for fermented sausages with reduced fat contents (Mendoza *et al.*, 2001; García *et al.*, 2002; Salazar *et al.*, 2009). The addition of 3%, 6% or 9% FOS reduced the hardness, and these values were not statistically significant different from the control. This was also observed by Cáceres *et al.* (2004) and Salazar *et al.* (2009) and may be attributed to the ability of FOS to form gels that provide a softness similar to that provided by fat. The treatments with reduced fat had significantly higher cohesiveness and chewiness values than the controls, which demonstrate that the addition of FOS has little effect on these parameters. This may be explained by stronger bonds

Table 5 Physicochemical characteristics of the fermented cooked sausages with reduced fat content and added fructooligosaccharides during storage

| Days | Control | F0 | F3 | F6 | F9 |
|---|---------------------------------|---------------------------------|--------------------------------|--------------------------------|--------------------------------|
| 2-thiobarbituric reactive substances ($\mu\text{g MDA g}^{-1}$ sample) | | | | | |
| 0 | 0.487 \pm 0.07 ^{aC} | 0.286 \pm 0.01 ^{bD} | 0.329 \pm 0.01 ^{bD} | 0.295 \pm 0.02 ^{bC} | 0.277 \pm 0.02 ^{bC} |
| 15 | 0.510 \pm 0.04 ^{aC} | 0.428 \pm 0.09 ^{abC} | 0.372 \pm 0.01 ^{bC} | 0.335 \pm 0.03 ^{bC} | 0.330 \pm 0.02 ^{bC} |
| 30 | 0.536 \pm 0.01 ^{aBC} | 0.490 \pm 0.02 ^{aB} | 0.458 \pm 0.01 ^{aB} | 0.441 \pm 0.06 ^{aB} | 0.470 \pm 0.07 ^{aB} |
| 45 | 0.632 \pm 0.04 ^{aB} | 0.517 \pm 0.04 ^{bB} | 0.509 \pm 0.01 ^{bB} | 0.498 \pm 0.02 ^{bB} | 0.535 \pm 0.01 ^{bB} |
| 60 | 1.710 \pm 0.02 ^{aA} | 1.250 \pm 0.15 ^{bA} | 1.074 \pm 0.07 ^{bA} | 1.120 \pm 0.15 ^{bA} | 1.017 \pm 0.01 ^{bA} |
| Hardness (N) | | | | | |
| 0 | 51.84 \pm 0.88 ^b | 59.00 \pm 2.03 ^a | 52.37 \pm 1.51 ^b | 52.25 \pm 1.24 ^b | 51.99 \pm 1.18 ^b |
| 15 | 52.82 \pm 1.46 ^b | 68.00 \pm 1.37 ^a | 56.32 \pm 1.83 ^b | 52.01 \pm 0.86 ^b | 55.93 \pm 1.25 ^b |
| 30 | 51.83 \pm 1.10 ^b | 59.98 \pm 0.83 ^a | 50.50 \pm 0.78 ^b | 51.85 \pm 1.62 ^b | 50.18 \pm 0.88 ^b |
| 45 | 54.11 \pm 0.88 ^b | 70.06 \pm 1.19 ^a | 58.35 \pm 1.6 ^b | 54.65 \pm 1.41 ^b | 56.77 \pm 1.94 ^b |
| 60 | 58.11 \pm 0.89 ^b | 70.14 \pm 0.86 ^a | 59.28 \pm 1.59 ^b | 57.24 \pm 1.65 ^b | 59.08 \pm 1.60 ^b |
| Cohesiveness | | | | | |
| 0 | 0.64 \pm 0.05 ^b | 0.69 \pm 0.04 ^a | 0.68 \pm 0.05 ^a | 0.68 \pm 0.00 ^a | 0.67 \pm 0.00 ^a |
| 15 | 0.64 \pm 0.05 ^b | 0.68 \pm 0.05 ^a | 0.70 \pm 0.01 ^a | 0.68 \pm 0.00 ^a | 0.67 \pm 0.05 ^a |
| 30 | 0.65 \pm 0.01 ^b | 0.68 \pm 0.00 ^a | 0.69 \pm 0.01 ^a | 0.70 \pm 0.00 ^a | 0.68 \pm 0.01 ^a |
| 45 | 0.64 \pm 0.01 ^b | 0.69 \pm 0.02 ^a | 0.69 \pm 0.01 ^a | 0.68 \pm 0.00 ^a | 0.68 \pm 0.01 ^a |
| 60 | 0.63 \pm 0.01 ^b | 0.69 \pm 0.00 ^a | 0.73 \pm 0.01 ^a | 0.69 \pm 0.00 ^a | 0.69 \pm 0.00 ^a |
| Chewiness (N) | | | | | |
| 0 | 21.56 \pm 0.32 ^b | 30.46 \pm 0.99 ^a | 29.58 \pm 0.84 ^a | 29.11 \pm 0.76 ^a | 29.60 \pm 0.59 ^a |
| 15 | 27.79 \pm 0.62 ^b | 35.21 \pm 0.74 ^a | 32.91 \pm 0.95 ^a | 35.96 \pm 0.49 ^a | 34.07 \pm 0.87 ^a |
| 30 | 22.24 \pm 0.98 ^b | 31.88 \pm 0.64 ^a | 31.85 \pm 0.31 ^a | 31.40 \pm 1.00 ^a | 32.44 \pm 0.51 ^a |
| 45 | 28.67 \pm 0.91 ^b | 37.98 \pm 0.93 ^a | 35.64 \pm 0.99 ^a | 33.96 \pm 0.85 ^a | 36.18 \pm 1.12 ^a |
| 60 | 29.25 \pm 0.52 ^b | 41.58 \pm 0.71 ^a | 45.11 \pm 1.75 ^a | 43.94 \pm 1.24 ^a | 42.43 \pm 1.35 ^a |
| Adhesiveness (Ns) | | | | | |
| 0 | -0.10 \pm 0.17 ^a | -0.35 \pm 0.13 ^a | -0.24 \pm 0.86 ^a | -0.22 \pm 0.57 ^a | -0.27 \pm 0.07 ^a |
| 15 | -0.25 \pm 0.06 ^a | -0.38 \pm 0.09 ^a | -0.46 \pm 0.09 ^a | -0.16 \pm 0.03 ^a | -0.20 \pm 0.03 ^a |
| 30 | -0.37 \pm 0.10 ^a | -0.62 \pm 0.13 ^a | -0.39 \pm 0.86 ^a | -0.40 \pm 0.11 ^a | -0.69 \pm 0.09 ^a |
| 45 | -0.33 \pm 0.11 ^a | -0.47 \pm 0.14 ^a | -0.47 \pm 0.13 ^a | -0.54 \pm 0.12 ^a | -0.36 \pm 0.14 ^a |
| 60 | -0.26 \pm 0.09 ^a | -0.51 \pm 0.10 ^a | -0.71 \pm 0.15 ^a | -0.35 \pm 0.09 ^a | -0.66 \pm 0.15 ^a |
| <i>L</i> * | | | | | |
| 0 | 52.84 \pm 0.29 ^a | 51.14 \pm 0.28 ^{ab} | 52.94 \pm 0.38 ^{bc} | 49.25 \pm 0.55 ^{cd} | 48.94 \pm 0.48 ^d |
| 15 | 51.66 \pm 0.27 ^a | 46.92 \pm 0.16 ^c | 49.37 \pm 0.46 ^b | 49.48 \pm 0.47 ^b | 46.16 \pm 0.21 ^c |
| 30 | 52.84 \pm 0.29 ^a | 51.14 \pm 0.28 ^{ab} | 50.94 \pm 0.38 ^{bc} | 49.25 \pm 0.55 ^{cd} | 483.94 \pm 0.48 ^d |
| 45 | 52.52 \pm 0.21 ^a | 49.71 \pm 0.31 ^b | 47.66 \pm 0.42 ^c | 49.88 \pm 0.38 ^b | 45.22 \pm 0.18 ^d |
| 60 | 52.11 \pm 0.50 ^a | 50.00 \pm 0.39 ^b | 47.80 \pm 0.19 ^c | 48.02 \pm 0.2 ^c | 47.57 \pm 0.21 ^c |
| <i>a</i> * | | | | | |
| 0 | 12.90 \pm 0.25 ^a | 13.62 \pm 0.18 ^a | 13.63 \pm 0.22 ^a | 13.44 \pm 0.26 ^a | 12.80 \pm 0.17 ^a |
| 15 | 12.78 \pm 0.10 ^c | 14.25 \pm 0.15 ^a | 13.19 \pm 0.30 ^{bc} | 13.87 \pm 0.23 ^{ab} | 13.69 \pm 0.15 ^{ab} |
| 30 | 12.90 \pm 0.25 ^a | 13.62 \pm 0.18 ^a | 13.63 \pm 0.22 ^a | 13.44 \pm 0.26 ^a | 12.80 \pm 0.17 ^a |
| 45 | 12.78 \pm 0.23 ^c | 14.00 \pm 0.09 ^{ab} | 14.72 \pm 0.13 ^a | 14.11 \pm 0.18 ^{ab} | 13.92 \pm 0.11 ^b |
| 60 | 12.65 \pm 0.75 ^b | 13.86 \pm 0.25 ^{ab} | 14.66 \pm 0.17 ^a | 13.91 \pm 0.17 ^{ab} | 13.34 \pm 0.20 ^{ab} |
| <i>b</i> * | | | | | |
| 0 | 9.16 \pm 0.20 ^a | 9.32 \pm 0.21 ^a | 8.15 \pm 0.34 ^a | 9.17 \pm 0.42 ^a | 8.21 \pm 0.19 ^a |
| 15 | 9.18 \pm 0.20 ^a | 9.36 \pm 0.21 ^a | 8.20 \pm 0.34 ^a | 9.20 \pm 0.42 ^a | 8.27 \pm 0.10 ^a |
| 30 | 8.69 \pm 0.31 ^a | 8.93 \pm 0.14 ^a | 8.97 \pm 0.17 ^a | 8.41 \pm 0.28 ^{ab} | 7.64 \pm 0.11 ^b |
| 45 | 9.24 \pm 0.07 ^a | 8.34 \pm 0.15 ^b | 8.36 \pm 0.13 ^b | 8.16 \pm 0.11 ^b | 8.33 \pm 0.20 ^b |
| 60 | 9.77 \pm 0.22 ^a | 9.00 \pm 0.29 ^b | 9.44 \pm 0.10 ^{ab} | 8.82 \pm 0.10 ^b | 8.77 \pm 0.09 ^b |
| FOS (g%) | | | | | |
| 0 | ND | ND | 3.15 \pm 0.49 ^{cA} | 6.15 \pm 0.02 ^{bA} | 9.09 \pm 0.87 ^{aA} |
| 60 | ND | ND | 3.36 \pm 0.30 ^{cA} | 6.29 \pm 0.30 ^{bA} | 9.50 \pm 0.16 ^{aA} |

Values represent the mean (\pm standard deviation). Means in the same row with the same lowercase letters did not present a statistically significant difference ($P > 0.05$) according to Tukey's test. Means in the same column with the same capital letters did not present a statistically significant difference ($P > 0.05$) according to Tukey's test. The following treatments were used: Control (15% fat); F0 (7.5% fat); F3 (7.5% fat + 3% FOS); F6 (7.5% fat + 6% FOS); F9 (7.5% fat + 9% FOS).

FOS, fructooligosaccharides.

among the meat particles caused by the reduction in fat, which favours increased cohesiveness and chewiness (Jiménez-Colmenero, 2000).

A 50% reduction in fat and the addition of 3%, 6% or 9% FOS did not change the values of a^* or b^* at the end of manufacturing (day 0) (Table 5). The control formulation showed lower a^* values and higher b^* values compared to treatments with reduced fat after 45 and 60 days of storage ($P \leq 0.05$). This can be correlated with an increase in the lipid oxidation as described earlier (Table 5). The addition of FOS significantly decreased the lightness (L^*), an observation also noted by Salazar *et al.* (2009). This decrease in lightness is because of the increased turbidity caused by the shrinkage of the gel formed by the FOS, which is a result of the dehydration that occurred during the processing of the product (Matuszek, 2001).

The results of the quantification of FOS are shown in Table 5. According to Brazilian legislation (Anvisa, 1998), an F3 treatment can be considered as a 'source of fibre' because it contains more than 3 g of fibre per 100 g, and F6 and F9 treatments can be considered products with 'high fibre content' because they contain more than 6 g of fibre per 100 g. There is no consensus among researchers concerning the minimum FOS consumption that is needed to stimulate the growth of probiotic microorganisms so as to be considered a prebiotic. According to some studies, a daily intake of 5 g of FOS is sufficient to stimulate the growth of bifidobacteria (Roberfroid & Slavin, 2000; Shin *et al.*, 2000; Rao, 2001; Manning & Gibson, 2004; Sangeethaa *et al.*, 2005). On the basis of these studies, we can

conclude that the consumption of one 50 g serving of fermented cooked sausages that contain 3 (F3), 6 (F6) or 9% (F9) FOS would supply approximately 30%, 60% or 90% of the FOS, respectively, that are recommended for achieving prebiotic effects. There was no significant difference between the levels of FOS observed for the products at time zero and after 60 days of refrigerated storage (4 ± 1 °C), indicating that prebiotic ingredients exhibit excellent stability.

Microbiological analysis

The microbiological characteristics during the storage period are presented in Table 6. During the 60 days of storage, no coliform (faecal or total) bacteria were detected. The counts of aerobic mesophilic and lactic acid bacteria declined during storage in all of the treatments. This is consistent with the results of Bozkurt & Erkmén (2002, 2004, 2007), who observed increases in aerobic mesophilic and lactic acid bacteria during the production and decreases in these bacteria during storage.

Consumer study

The results of the consumer study are presented in Table 7. Although the storage period had been established for 60 days, a sensory evaluation was not performed after 60 days as a result of high TBARS values (Table 3). Over a 45-day storage period, there were no differences in the aroma or taste between the controls and the F0, F3, F6 and F9 treatments ($P > 0.05$). The colour attribute values did not differ among the treatments at the beginning of storage (day

Table 6 Microbiological characteristics (log CFU g⁻¹) of the fermented cooked sausages with reduced fat content and added fructooligosaccharides during storage.

| Days | Control | F0 | F3 | F6 | F9 |
|-----------------------------|---------------------------|--------------------------|--------------------------|---------------------------|---------------------------|
| Mesophilic aerobic bacteria | | | | | |
| 15 | 5.94 ± 0.02 ^{a*} | 5.48 ± 0.04 ^b | 5.30 ± 0.03 ^d | 5.35 ± 0.04 ^{cd} | 5.41 ± 0.06 ^{bc} |
| 30 | 4.67 ± 0.01 ^b | 5.00 ± 0.08 ^a | 4.20 ± 0.15 ^c | 4.00 ± 0.00 ^d | 5.17 ± 0.10 ^a |
| 45 | 3.03 ± 0.21 ^c | 5.00 ± 0.11 ^a | 4.04 ± 0.16 ^b | 2.68 ± 0.20 ^d | 3.00 ± 0.10 ^{cd} |
| 60 | 2.80 ± 0.36 ^b | 3.56 ± 0.22 ^a | 2.82 ± 0.07 ^b | 2.29 ± 0.14 ^c | 2.34 ± 0.08 ^c |
| Lactic acid bacteria | | | | | |
| 15 | 5.62 ± 0.09 ^b | 5.76 ± 0.03 ^a | 5.50 ± 0.03 ^c | 5.38 ± 0.04 ^d | 5.27 ± 0.06 ^e |
| 30 | 4.89 ± 0.09 ^b | 4.75 ± 0.08 ^b | 4.29 ± 0.16 ^c | 4.08 ± 0.13 ^d | 5.22 ± 0.02 ^a |
| 45 | 3.19 ± 0.05 ^b | 4.17 ± 0.03 ^a | 3.17 ± 0.07 ^b | 2.80 ± 0.04 ^c | 2.95 ± 0.03 ^b |
| 60 | 2.78 ± 0.17 ^b | 3.30 ± 0.01 ^a | 2.70 ± 0.01 ^b | 2.08 ± 0.13 ^c | 2.19 ± 0.20 ^c |
| Total coliforms | | | | | |
| 15 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |
| 30 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |
| 45 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |
| 60 | <1.00 | <1.00 | <1.00 | <1.00 | <1.00 |

*Values represent the mean (± standard deviation). Means accompanied by the same letter on the same line do not present a statistically significant difference ($P > 0.05$) according to Tukey's test. The following treatments were used: Control (15% fat); F0 (7.5% fat); F3 (7.5% fat + 3% FOS); F6 (7.5% fat + 6% FOS); F9 (7.5% fat + 9% FOS).

FOS, fructooligosaccharides.

Table 7 Consumer acceptability ($n = 60$) of colour, aroma, taste, texture and overall acceptability of the fermented cooked sausages with reduced fat content and added fructooligosaccharides during storage.

| Days | Control | F0 | F3 | F6 | F9 |
|------------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|
| Colour | | | | | |
| 0 | 5.75 ± 1.87 ^{aA*} | 6.53 ± 1.34 ^{aA} | 6.05 ± 1.80 ^{aA} | 6.28 ± 1.86 ^{aA} | 6.53 ± 1.71 ^{aA} |
| 15 | 5.50 ± 1.85 ^{aAB} | 6.17 ± 1.82 ^{aA} | 6.02 ± 1.66 ^{aA} | 6.35 ± 1.66 ^{aA} | 6.34 ± 1.61 ^{aA} |
| 30 | 5.86 ± 1.89 ^{bA} | 6.34 ± 1.62 ^{abA} | 6.70 ± 1.58 ^{abA} | 6.61 ± 1.69 ^{abA} | 6.86 ± 1.58 ^{abA} |
| 45 | 4.66 ± 1.85 ^{bB} | 6.10 ± 1.62 ^{aA} | 5.93 ± 1.78 ^{aA} | 6.07 ± 1.62 ^{aA} | 6.19 ± 1.80 ^{aA} |
| Aroma | | | | | |
| 0 | 5.81 ± 1.97 ^{aA} | 5.78 ± 2.00 ^{aA} | 5.61 ± 1.71 ^{aA} | 5.43 ± 2.09 ^{aA} | 5.80 ± 1.88 ^{aA} |
| 15 | 5.85 ± 1.96 ^{aA} | 6.48 ± 1.97 ^{aA} | 6.03 ± 1.61 ^{aA} | 6.32 ± 1.74 ^{aA} | 6.21 ± 1.77 ^{aA} |
| 30 | 5.89 ± 1.89 ^{aA} | 6.09 ± 1.71 ^{aA} | 6.18 ± 2.05 ^{aA} | 6.30 ± 1.82 ^{aA} | 6.04 ± 2.03 ^{aA} |
| 45 | 5.29 ± 1.97 ^{aA} | 5.74 ± 1.96 ^{aA} | 5.72 ± 1.87 ^{aA} | 5.59 ± 1.82 ^{aA} | 5.82 ± 2.02 ^{aA} |
| Taste | | | | | |
| 0 | 5.87 ± 2.11 ^{aA} | 6.35 ± 1.89 ^{aA} | 5.88 ± 2.10 ^{aA} | 6.10 ± 2.15 ^{aA} | 6.04 ± 2.08 ^{aA} |
| 15 | 6.32 ± 1.90 ^{aA} | 6.59 ± 1.78 ^{aA} | 6.45 ± 1.75 ^{aA} | 6.40 ± 1.83 ^{aA} | 6.68 ± 1.51 ^{aA} |
| 30 | 5.96 ± 2.14 ^{aA} | 6.43 ± 1.52 ^{aA} | 6.48 ± 1.65 ^{aA} | 6.16 ± 1.97 ^{aA} | 6.28 ± 1.90 ^{aA} |
| 45 | 5.44 ± 2.07 ^{aA} | 6.22 ± 1.88 ^{aA} | 6.33 ± 1.60 ^{aA} | 5.98 ± 1.65 ^{aA} | 6.23 ± 1.79 ^{aA} |
| Texture | | | | | |
| 0 | 5.64 ± 2.29 ^{aAB} | 5.58 ± 1.84 ^{bA} | 6.55 ± 1.61 ^{aA} | 6.24 ± 1.90 ^{aA} | 6.50 ± 1.73 ^{aA} |
| 15 | 6.43 ± 1.94 ^{aA} | 5.48 ± 1.87 ^{bA} | 6.63 ± 1.63 ^{aA} | 6.63 ± 1.65 ^{aA} | 6.89 ± 1.37 ^{aA} |
| 30 | 6.24 ± 2.16 ^{aA} | 5.62 ± 1.45 ^{bA} | 6.59 ± 1.63 ^{aA} | 6.62 ± 1.84 ^{aA} | 6.53 ± 2.06 ^{aA} |
| 45 | 6.27 ± 2.61 ^{aA} | 5.19 ± 1.75 ^{bA} | 6.59 ± 1.77 ^{aA} | 6.58 ± 1.60 ^{aA} | 6.42 ± 1.74 ^{aA} |
| Overall acceptability | | | | | |
| 0 | 6.14 ± 1.96 ^{aA} | 5.44 ± 1.58 ^{bA} | 6.10 ± 1.86 ^{aA} | 6.22 ± 1.85 ^{aA} | 6.15 ± 1.91 ^{aA} |
| 15 | 6.22 ± 1.90 ^{aA} | 5.63 ± 1.74 ^{bA} | 6.53 ± 1.49 ^{aA} | 6.65 ± 1.53 ^{aA} | 6.71 ± 1.55 ^{aA} |
| 30 | 5.90 ± 1.94 ^{abA} | 5.47 ± 1.61 ^{bA} | 6.42 ± 1.79 ^{aA} | 6.52 ± 1.78 ^{aA} | 6.21 ± 2.06 ^{aA} |
| 45 | 5.91 ± 1.84 ^{ab} | 5.36 ± 1.63 ^{bA} | 6.47 ± 1.39 ^{aA} | 6.04 ± 1.63 ^{abA} | 6.19 ± 1.60 ^{aA} |

*Values represent the mean (±standard deviation). Means in the same row with the same lowercase letters did not present a statistically significant difference ($P > 0.05$) according to Tukey's test. Means in the same column with the same capital letters did not present a statistically significant difference ($P > 0.05$) according to Tukey's test. The following treatments were used: Control (15% fat); F0 (7.5% fat); F3 (7.5% fat + 3% FOS); F6 (7.5% fat + 6% FOS); F9 (7.5% fat + 9% FOS). FOS, fructooligosaccharides.

0) ($P > 0.05$). This indicates that the panellists did not notice or consider the decrease in lightness (L^*) to be important in the treatments with added FOS (Table 5). After 45 days of storage, the control showed a reduction in its colour values, which differed significantly from the F0, F3, F6 and F9 treatments. This may be related to increased lipid oxidation (Table 5). The texture did not differ significantly between the treatments with added FOS (F3, F6, and F9) and the control; however, the treatment with a 50% reduction in fat and no added FOS (F0) exhibited lower texture values ($P \leq 0.05$). This may be attributed to the increased hardness observed in the instrumental texture profile analysis (Table 5) and may be responsible for the decreased overall acceptability of the F0 treatment ($P \leq 0.05$). Mendoza *et al.* (2001) were in agreement with these results when they assessed fermented sausages with a similar fat content to those in this study. They observed a significant decrease in the overall acceptability when the fat content was reduced by 50%. There was no significant difference in the overall acceptability between the control and the F3, F6 and

F9 treatments during the 45 days of storage. These results demonstrate that the use of FOS as a fat substitute helps maintain the sensory quality of the product during storage.

Conclusions

A 50% reduction in pork back fat decreases the technological and sensory quality of fermented cooked sausages. FOS are not degraded during storage, and their use at 3%, 6% or 9% levels both enriches the product with prebiotic fibres and reduces the technological and sensory defects caused by the reduction in fat. Thus, the use of FOS has been shown to be a good alternative for the development of healthier products with acceptable sensorial qualities.

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