

# Physicochemical properties of low sodium frankfurter with added walnut: effect of transglutaminase combined with caseinate, KCl and dietary fibre as salt replacers

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## Abstract

This study compares the effects of combinations of microbial transglutaminase (TGase) and various non-meat ingredients (caseinate, KCl and wheat fibre) used as salt replacers, with the effects of NaCl on the physicochemical properties (cooking loss, emulsion stability, texture and colour) of frankfurters with added walnuts. The combination of TGase with caseinate, KCl or fibre led to harder, springier and chewier ( $P < 0.05$ ) frankfurters with better water- and fat-binding properties (emulsion stability and cooking loss) ( $P < 0.05$ ) than those made with TGase only. Ranking of ingredient efficiency in combination with TGase showed that caseinate > KCl > fibre. Frankfurters with caseinate presented the highest lightness and the lowest redness values. Frankfurter with NaCl had a harder, springier and chewier gel/emulsion network with lower cooking loss than those NaCl free.  
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**Keywords:** Frankfurters; Walnut; Low sodium; Transglutaminase; Caseinate; KCl; Wheat fibre

## 1. Introduction

Cardiovascular diseases are the principal cause of death in developed countries, and diet is one of the major factors in their incidence. Epidemiological studies show that frequent consumption of nuts in general, and walnuts in particular, correlates inversely with myocardial infarction or death by vascular ischaemic disease (Fraser, Sabaté, Beeson, & Strahan, 1992; Iwamoto et al., 2000; Sabaté, 1993). This effect has been associated with the peculiar blend of nutrients and phytochemical compounds found in walnuts: high-biological-value proteins, vegetable fibre, monounsaturated (oleic) and polyunsaturated (linoleic and linolenic) fatty acids and micronutrients such as folic acid, magne-

sium, liposoluble vitamins (especially  $\gamma$ -tocopherol), and other antioxidants (phytosterols and polyphenols). All these combine in complex ways to exert beneficial effects on serum lipid profiles and other risk factors that can cause or exacerbate cardiovascular diseases.

Since not many people can be persuaded to consume walnuts in their pure state every day over a long period, it has been suggested that walnut intake could be promoted by including it as an ingredient in frequently consumed foods (Diehl, 2002), such as meat derivatives. With that end in view, restructured beef steak and frankfurters with added walnuts (and salts) have been formulated, resulting in products with acceptable physicochemical and sensory properties (Carballo, Ayo, & Jiménez Colmenero, 2003; Cofrades et al., 2004; Jiménez Colmenero, Serrano, Ayo, Cofrades, & Carballo, 2003).

Sodium intakes in most developed countries are greatly in excess of physiological requirements. High salt

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intake has been related to high blood pressure, one of the three major risk factors for cardiovascular diseases (Antonios & MacGregor, 1997). Since over 20–30% of dietary salt comes from meat and meat products (Wirth, 1991), there is growing interest among consumers and processors in reducing the use of salt (minimizing sodium) in meat processing. A number of binding agents have been used to overcome property problems in low-salt products, including problems associated with water- and fat-binding properties and texture (Collins, 1997; Monahan & Troy, 1997).

Addition of transglutaminase has been proposed as a means of inducing gelification of muscle protein that reduces or eliminates the need to add NaCl (Kuraishi et al., 1997; Nielsen, Petersen, & Moller, 1995). Microbial transglutaminase (TGase) has been used in the preparation of a variety of meat products, among them cooked gel/emulsion systems, but this has generally been in combination with salt and various food proteins and polysaccharides (Jarmoluk & Pietrasik, 2003; Kerry, O'Donnell, Brown, Kerry, & Buckley, 1999; Kuraishi et al., 1997; Muguruma et al., 2003; Pietrasik & Li-Chan, 2002). There have been few studies that analysed the effect of TGase on muscle systems in the absence of NaCl. In those cases, the exclusion of salt led to products with poor physicochemical properties (Kerry et al., 1999; Pietrasik & Li-Chan, 2002; Tellez-Luis, Uresti, Ramirez, & Vazquez, 2002). Combinations of TGase with suitable non-meat ingredients are needed to overcome those problems in NaCl-free meat products. Kerry et al. (1999) reported that TGase in combination with non-meat proteins (among them caseinate) could be used successfully in beef and poultry patties as direct replacers of NaCl and tripolyphosphate salts. O'Kennedy (2000) concluded that caseinate and TGase in meat dispersions with no added salts could rival salt-containing meat systems as regards achieving low cook losses.

Hitherto untried combinations of TGase and non-meat ingredients should be explored to assess their potential as binding agents. A number of different non-meat ingredients have been utilized as salt (NaCl) replacers, among them other chloride salts. KCl is the only salt with generally recognized as safe (GRAS) status as a replacement of NaCl (Collins, 1997). Potassium chloride has been used to substitute NaCl in meat products without loss of functionality. However, off-flavour problems limit its use to levels around 1% (Collins, 1997). As non-meat ingredients, dietary fibres are desirable not only for their nutritional properties but also for their functional and technological properties. Fibres can be used in cooked meat products (e.g. meat emulsions, patés) to increase the cooking yield thanks to their water- and fat-binding properties, and to improve texture (Thebaudin, Lefebvre, Harrington, & Bourgeois, 1997).

No reports have been found on the binding and textural characteristics of low sodium (no added NaCl)

meat emulsions with added walnut using combinations of TGase and other non-meat ingredients as salt replacers. The objective of this study was therefore to compare the effects of combinations of TGase and various non-meat ingredients (caseinate, KCl and fibre) as salt replacers with the effects of NaCl on the physicochemical properties (cooking loss, emulsion stability, texture and colour) of frankfurters with added walnuts.

## 2. Materials and methods

### 2.1. Preparation of meat and additives

Post-rigor pork meat (mixture of *M. biceps femoris*, *M. semimembranosus*, *M. semitendinosus*, *M. gracilis* and *M. adductor*) (5 kg) was trimmed of visible fat and connective tissue and passed through a grinder with a 0.6 cm plate. Lots of approx. 800 g were vacuum packed, frozen and stored at  $-20^{\circ}\text{C}$  until use, which took place within one week. Additives used for preparation of meat batters included sodium chloride, sodium polyphosphate (SPP), sodium nitrite, potassium chloride (Panreac Quimica, S.A. Barcelona, Spain), sodium caseinate (ANVISA, Madrid, Spain), wheat fibre (97% dietary fibre of which: 94.5% insoluble dietary fibre and 2.5% soluble dietary fibre) (Vitacel WF200, Campi y Jové, S.A., Barcelona, Spain) and microbial transglutaminase (ACTIVA WM, Ajinomoto Europe Sales GmbH, Hamburg, Germany). The enzyme was used in a mixture containing 1% transglutaminase and 99% maltodextrin, with a standard transglutaminase activity of approximately 100 units/g. Walnut paste (approx. 12  $\mu\text{m}$  of particle size) was supplied by La Morella Nuts, S.A. (Tarragona, Spain).

### 2.2. Preparation of meat batters

Meat packages were thawed (approx. 20 h at  $3 \pm 2^{\circ}\text{C}$ , up to between  $-1$  and  $-2^{\circ}\text{C}$ ). Five different meat batters were formulated (Table 1) as follows: raw meat was homogenized and ground for 1 min in a chilled cutter ( $2^{\circ}\text{C}$ ) (Stephan Universal Machine UM5, Stephan u. Söhne GmbH & Co., Hameln, Germany). NaCl, KCl, wheat fibre or caseinate (according to formulation) was added to the meat along with sodium nitrite and SPP that had been previously dissolved in water and chilled ( $2^{\circ}\text{C}$ ), and the whole was mixed again for 1 min. After that, the walnut was also added and the batters homogenized for 1 min. Finally, TGase was added and the whole mixed again for 2 min under vacuum ( $2^{\circ}\text{C}$ , 610 mm Hg). Mixing time was standardized to 5 min and the final temperature was below  $10^{\circ}\text{C}$  in all cases.

The batters were stuffed into 20 mm diameter Nojax cellulose casings (Viscase S.A., Bagnold Cedex, France) and hand-linked. Frankfurters were heat processed in

Table 1  
Formulation (g) of the different samples<sup>a</sup>

Samples	Meat	Walnut	NaCl	TGase	Caseinate	KCl	Fibre	Water
Control	700	200	25	0	0	0	0	150
MTG	700	200	0	7	0	0	0	150
MTG/SC	700	200	0	7	20	0	0	150
MTG/KCl	700	200	0	7	0	10	0	150
MTG/F	700	200	0	7	0	0	20	150

<sup>a</sup> All samples also contain 0.18% SPP and 0.012% NaNO<sub>3</sub>.

water baths as follows: 40 °C for 15 min, then 70 °C to a final internal temperature of 70 °C. The frankfurters were cooled in ice water to a core temperature of 6–10 °C and the casings were removed. The internal temperature was measured using thermocouples connected to a temperature recorder (Yokogawa Hokushin Electric YEW, Mod. 3087, Tokyo, Japan). Frankfurters were vacuum packed (Cryovac® BB4L, oxygen permeability 30 cm<sup>3</sup> m<sup>-2</sup> 24 h<sup>-1</sup> at 23 °C, 0% RH and 1 bar) and stored at 2 °C until analysis (48 h).

### 2.3. Proximate analysis, pH, and sodium and potassium content

Moisture and ash contents of the batters were determined (AOAC, 1984) in quadruplicate. Fat content was evaluated (in duplicate) according to Bligh and Dyer (1959). Protein content was measured in quadruplicate by a Nitrogen Determinator LECO FP-2000 (Leco Corporation, St Joseph, MI). The pH of the raw products was determined in duplicate using a pH meter (Radiometer PHM 93, Copenhagen, Denmark) on a homogenate of 10 g sample in 100 ml distilled water.

Sodium and potassium contents were evaluated (in duplicate) by atomic absorption spectrometer (Perkin-Elmer, 5100, Norwalk, CT, USA), as described by AMAAS (1982).

### 2.4. Emulsion stability and cooking loss

Emulsion stability of the different batters was determined (in quadruplicate) according to the procedure described by Jiménez Colmenero, Carballo, and Solas (1995) with some modifications. Approximately 50 g (exactly weighed) of meat batter was placed in tubes, which were then centrifuged (5 min, 2632 g, at 2 °C) and heated (40 °C/15 min followed by 20 min/70 °C). The tubes were then left to stand upside down (for 40 min) to release the exudate. Total fluid released was expressed as % of sample weight. The fat released was the weight on heating the exudate for 16 h at 103 °C. Water released was the difference between total fluid released and fat released. Water and fat released were expressed as a % of sample weight.

Cooking loss of frankfurters was calculated as weight loss during heat processing and expressed as % of initial

sample weight. Determinations were carried out in quadruplicate.

### 2.5. Surface colour

Surface colour Cielab values (lightness, *L\**; redness, *a\** and yellowness, *b\**) of frankfurters were evaluated on a HunterLab model D25-9 system (D45/2°) (Hunter Associates Laboratory Inc., Reston, VA). Eight replicates of the analysis were performed for each formulation.

### 2.6. Texture profile analysis (TPA)

Texture profile analysis was performed in a TA-XT2 Texture Analyser (Texture Technologies Corp., Scarsdale, NY) as described by Bourne (1978). Eight frankfurter cores (diam. = 20 mm, height = 25 mm) were axially compressed to 30% of their original height. Force–time deformation curves were derived with a 50 N load cell applied at a crosshead speed of 0.8 mm/s. Attributes were calculated as follows: hardness (Hd) = peak force (N) required for first compression; cohesiveness (Ch) = ratio of active work done under the second compression curve to that done under the first compression curve (dimensionless); springiness (Sp) = distance (mm) the sample recovers after the first compression; chewiness (Cw) = Hd × Ch × Sp (N × mm). Measurement of samples was carried out at room temperature.

### 2.7. Statistical analysis

Data were analysed using Statgraphics Plus 2.1 (STSC Inc., Rockville, MD) for one-way ANOVA. Least squares difference was used for comparison of mean values among treatments, and to identify significant differences (*P* < 0.05) among treatment.

## 3. Results

### 3.1. Proximate analysis, pH, and sodium and potassium content

Proximate values differed little among all the samples, but there were some significant differences derived from

the type of formulation (Table 2). MTG and MTG/SC samples had the highest ( $P < 0.05$ ) moisture and protein contents respectively. Ash levels were higher ( $P < 0.05$ ) in the control (with NaCl) and MTG/KCl samples (Table 2). The fat content, which was similar ( $P > 0.05$ ) in all samples, was mainly (about 85%) attributable to walnut, since meat raw material contained less than 3% fat.

Sodium and potassium contents differed according to the formulation (Table 2). The control sample (with NaCl) contained the highest amount ( $P < 0.05$ ) of Na (828 mg/100 g), which is close to normal content for products with these characteristics (Collins, 1997). In MTG, MTG/F and MTG/KCl samples, on the other hand, Na values were close to 100 mg/100 g, which is about 85–90% less than the control. Potassium content was highest ( $P < 0.05$ ) in MTG/KCl sample (Table 2).

The samples containing NaCl and KCl (control and MTG/KCl) presented the same pH, which was higher ( $P < 0.05$ ) than in all other samples (MTG, MTG/F and MTG/SC) (Table 2).

### 3.2. Emulsion stability and cooking loss

The emulsion stability of different batters was affected by formulation (Table 3). Total fluid release was highest ( $P < 0.05$ ) in batter with TGase alone (MTG sample) and lowest ( $P < 0.05$ ) in MTG/SC and control samples. Fat release was highest ( $P < 0.05$ ) in MTG sample, while there was significant difference among all the other samples (Table 3). As in total fluid release, the water release, ranged between 2.05% and 10.69%, were lowest ( $P < 0.05$ ) in MTG/SC and control (NaCl) samples and highest ( $P < 0.05$ ) in MTG sample. Batter prepared with TGase alone showed poor fat- and water-binding properties, which were improved by any of the added compounds. The best improvement was observed in sample with caseinate (MTG/SC), where the total fluid released was 80% less than in MTG sample (Table 3). The positive effect of combinations of TGase and casein on emulsifying properties has been reported by Muguruma et al. (2003).

Cooking loss was significantly lower in the control sample (with NaCl) and highest in the MTG sample (Table 3). All the ingredients added along with TGase

(caseinate, KCl and fibre) reduced ( $P < 0.05$ ) cooking loss, although none did so as effectively as NaCl (control sample).

### 3.3. Texture

Texture profile analysis (Table 4) indicated that Hd, Sp and Cw were highest ( $P < 0.05$ ) in the control and lowest ( $P < 0.05$ ) in the sample containing only TGase (MTG). The combination of TGase with the various non-meat ingredients increased ( $P < 0.05$ ) Hd, Sp and Cw; the effect was most pronounced in the sample with caseinate, followed by sample with KCl (Table 4). Cohesiveness was highest in sample MTG/KCl, followed by the control; the other samples did not differ significantly from one another (Table 4).

### 3.4. Colour

$L^*$  values were most affected by addition of caseinate (MTG/SC). In the other samples lightness ranged from 66.3 to 66.8 (Table 5). Redness values were lower in samples with TGase (MTG, MTG/SC, MTG/KCl and MTG/F) than in the control;  $a^*$  values were lowest ( $P < 0.05$ ) in the product containing caseinate (Table 5). Yellowness values of the control, MTG/C and MTG/KCl samples were similar ( $P > 0.05$ ), and  $b^*$  values were highest ( $P < 0.05$ ) in sample MTG. In quantitative terms, the variations induced in colour were slight.

## 4. Discussion

A functional food can be a natural food, a food to which a component has been added, or a food from which a component has been removed by technological or biotechnological means in order to affect beneficially one or more target functions in the body in a way that is relevant to either an improved state of health and well-being and/or reduction of risk of disease (Diplock et al., 1999). This study analysed products designed to incorporate specific characteristics as regards composition. To that end, animal fat was kept to a minimum, and walnut was added to promote high levels of bioactive

Table 2  
Proximate analysis (%), sodium and potassium contents (mg/100 g) and pH of different frankfurters<sup>a</sup>

Samples	Protein	Moisture	Fat	Ash	Sodium	Potassium	pH
Control	16.5 <sup>a</sup>	63.3 <sup>a</sup>	14.9 <sup>a</sup>	3.4 <sup>a</sup>	828.1 <sup>a</sup>	270.9 <sup>a</sup>	6.00 <sup>a</sup>
MTG	17.0 <sup>ab</sup>	65.0 <sup>b</sup>	15.4 <sup>a</sup>	1.2 <sup>bd</sup>	94.3 <sup>b</sup>	263.4 <sup>a</sup>	5.82 <sup>b</sup>
MTG/SC	17.9 <sup>b</sup>	63.0 <sup>a</sup>	15.2 <sup>a</sup>	1.2 <sup>d</sup>	131.1 <sup>c</sup>	233.3 <sup>a</sup>	5.85 <sup>b</sup>
MTG/KCl	16.5 <sup>a</sup>	63.9 <sup>c</sup>	15.8 <sup>a</sup>	2.0 <sup>c</sup>	95.3 <sup>b</sup>	633.3 <sup>b</sup>	6.00 <sup>a</sup>
MTG/F	16.1 <sup>a</sup>	63.9 <sup>c</sup>	15.4 <sup>a</sup>	1.2 <sup>b</sup>	88.4 <sup>b</sup>	265.1 <sup>a</sup>	5.85 <sup>b</sup>
SEM	0.26	0.18	0.32	0.01	2.32	17.18	0.01

<sup>a</sup> For sample denomination see Table 1. Means with different letters in the same column are significantly different ( $P < 0.05$ ). SEM: standard error of the mean.

Table 3  
Emulsion stability of batters and cooking loss of frankfurters<sup>a</sup>

Samples	Total fluid released (%)	Fat released (%)	Water released (%)	Cooking loss (%)
Control	3.77 <sup>a</sup>	0.39 <sup>a</sup>	3.38 <sup>a</sup>	2.0 <sup>a</sup>
MTG	11.59 <sup>b</sup>	0.90 <sup>b</sup>	10.69 <sup>b</sup>	13.2 <sup>b</sup>
MTG/SC	2.18 <sup>c</sup>	0.13 <sup>a</sup>	2.05 <sup>c</sup>	5.5 <sup>c</sup>
MTG/KCl	5.83 <sup>d</sup>	0.51 <sup>a</sup>	5.32 <sup>d</sup>	9.7 <sup>d</sup>
MTG/F	7.19 <sup>e</sup>	0.50 <sup>a</sup>	6.69 <sup>e</sup>	8.9 <sup>d</sup>
SEM	0.36	0.04	0.33	0.16

<sup>a</sup> For sample denomination see Table 1. Means with different letters in the same column are significantly different ( $P < 0.05$ ). SEM: standard error of the mean.

Table 4  
Texture profile analysis of different frankfurters<sup>a</sup>

Samples	Hardness (N)	Springiness (mm)	Cohesiveness (dimensionless)	Chewiness (N × mm)
Control	20.78 <sup>a</sup>	5.15 <sup>a</sup>	0.49 <sup>a</sup>	52.00 <sup>a</sup>
MTG	10.01 <sup>b</sup>	4.25 <sup>b</sup>	0.46 <sup>b</sup>	19.18 <sup>b</sup>
MTG/SC	17.23 <sup>c</sup>	4.68 <sup>c</sup>	0.45 <sup>b</sup>	36.21 <sup>c</sup>
MTG/KCl	15.55 <sup>d</sup>	4.90 <sup>d</sup>	0.52 <sup>c</sup>	39.38 <sup>c</sup>
MTG/F	14.13 <sup>e</sup>	4.44 <sup>e</sup>	0.45 <sup>b</sup>	28.16 <sup>d</sup>
SEM	0.25	0.06	0.008	0.80

<sup>a</sup> For sample denomination see Table 1. Means with different letters in the same column are significantly different ( $P < 0.05$ ). SEM: standard error of the mean.

Table 5  
Colour parameters of different samples<sup>a</sup>

Samples	Lightness $L^*$	Redness $a^*$	Yellowness $b^*$
Control	66.7 <sup>a</sup>	3.5 <sup>a</sup>	9.9 <sup>a</sup>
MTG	66.3 <sup>b</sup>	3.2 <sup>b</sup>	10.7 <sup>b</sup>
MTG/CS	68.7 <sup>c</sup>	2.3 <sup>d</sup>	9.9 <sup>a</sup>
MTG/KCl	66.8 <sup>a</sup>	2.7 <sup>c</sup>	10.0 <sup>a</sup>
MTG/F	66.6 <sup>ab</sup>	2.8 <sup>c</sup>	10.2 <sup>c</sup>
SEM	0.13	0.04	0.08

<sup>a</sup> For sample denomination see Table 1. Means with different letters in the same column are significantly different ( $P < 0.05$ ). SEM: standard error of the mean.

components that have been associated with major health benefits, particularly in connection with cardiovascular risk (Fraser et al., 1992; Iwamoto et al., 2000; Sabaté, 1993). Sodium levels were likewise considerably reduced (Table 2), the aim being to reduce the dietary sodium from meat products, with clear benefits to some risk factors for cardiovascular diseases (Antonios & MacGregor, 1997). Addition of fibre to frequently consumed foods like the meat products prepared in this study (Table 1) should help increase the daily fibre intake, which at present is lower than recommended (Backers & Noli, 1997). At the same time, there is evidence to suggest that potassium has the opposite effect to sodium: that is, the higher the potassium intake the lower is the blood pressure (Antonios & MacGregor, 1997). Therefore, these frankfurters should meet the requirements for consideration as a functional food.

The physicochemical characteristics of different frankfurters (Tables 2–5) were related to the formulation (Table 1). The good properties (Tables 3–5) of the

control sample were to be expected since NaCl solubilizes meat proteins, and this increases the number of locations in the polypeptide chains capable of interacting during heating. The result is a stable, elastic and rigid protein gel matrix with good water- and fat-binding properties (Carballo, Mota, Barreto, & Jiménez Colmenero, 1995).

In response to consumer demand for a reduction in dietary sodium intake, a variety of approaches to replacement or substitution of sodium chloride are available for meat processing, among them TGase-based binding systems. Recent research has shown that TGase acts by cross-linking meat proteins; it can therefore be used in meat processing to improve rheological properties while reducing, or even eliminating, the need for salts (Kuraishi et al., 1997). Soluble myofibrillar proteins are a good substrate for cross-linking reactions with TGase (Kuraishi et al., 1997); however, in the MTG sample the levels were low (due to low ionic strength caused by lack of NaCl) (Table 1), and hence

there was insufficient protein aggregation to form a strong protein network. This would explain why the resulting product had poorer water and fat binding properties (Table 3) and a softer texture than the control sample (Table 4). Several researchers have indicated that it is not feasible to obtain muscle products using TGase alone as binding agent (O'Kennedy, 2000; Pietrasik & Li-Chan, 2002; Tellez-Luis et al., 2002). As a result, TGase is generally used in combination with salt and/or various food proteins and polysaccharides in raw (Jiménez Colmenero et al., 2003; Kuraishi et al., 1997; O'Kennedy, 2000) and cooked (Kerry et al., 1999; Kilic, 2003; Muguruma et al., 2003; Pietrasik & Li-Chan, 2002) meat products.

Walnut contains mainly albumin, globulin, prolamin and glutelin (Sze-Tao & Sathe, 2000), which are obviously incorporated in the meat systems as formulated. A wide range of food proteins could be cross-linked by TGase, among them plant proteins (with structural properties similar to those of walnut proteins). Some of them (globulins, gluten), either on their own or as part of a meat extender (oat, soybean), provide a good substrate for the TGase reaction, forming polymers that significantly improve some technological properties of great utility in comminuted meats (Kurth & Rogers, 1984; Muguruma et al., 2003; Siu, Ma, Mock, & Mine, 2002). Potential cross-linkings among myofibrillar proteins, walnut proteins and TGase did not adequately overcome the problem of reproducing the effect of NaCl on physicochemical characteristics of frankfurters (Tables 3–5). There are two factors that could affect the reaction of TGase with the meat and walnut proteins present in the formulated products. One is the limited capacity to interact of some of these native vegetable proteins, which TGase will not cross-link unless they are denatured (Nielsen, 1995; O'Kennedy, 2000); the high denaturation temperature of some of these proteins (results not published) prevents the protein from undergoing sufficient structural change under thermal treatment (see Section 2), thereby limiting its interaction with the proteins of the muscle system (Feng & Xiong, 2002). The other is that protein aggregation in meat batter containing TGase occurs at a higher temperature than in the corresponding control (Muguruma et al., 2003), so that it might be best to raise final temperatures. In light of these considerations, it would seem necessary to study the thermal denaturation conditions of walnut proteins and their effect on protein interactions (work in progress), and to establish the best heat processing conditions accordingly.

Various non-meat ingredients have been assayed in combination with TGase to impart suitable physicochemical characteristics to a number of meat products (Jarmoluk & Pietrasik, 2003; Kilic, 2003; Muguruma et al., 2003; O'Kennedy, 2000). It has been suggested that a TGase/caseinate system could be used to produce

restructured meat in the raw state without addition of salts (Kuraishi et al., 1997); however, in cooked meats it has generally been used along with NaCl (Kilic, 2003; O'Kennedy, 2000; Pietrasik & Jarmoluk, 2003). There have been few studies in which combinations of MTG/caseinate have been used in the absence of NaCl (Kerry et al., 1999; O'Kennedy, 2000) to reduce sodium levels. Caseinate has proven to be a good substrate for TGase (Kuraishi et al., 1997), facilitating cross-linking between meat protein and caseinate molecules (Kurth & Rogers, 1984), which promotes the formation of a much more stable gel matrix during heating. This leads to a smaller release of water and fat and hence lower cooking loss (Table 3), higher emulsion stability (Table 3) and stronger gel structures (Table 4) than are achieved by the action of the enzyme alone (MTG sample). Ours results are consistent with those reported by Kerry et al. (1999) in beef and poultry patties. The effect of caseinates may also be facilitated by their role as meat extenders, and by the high protein content of MTG/CS frankfurters as compared to the MTG sample (Table 2). Also, it has been reported that when casein was treated with TGase for 30 min at 40 °C (similar to the procedure in this experiment), the cross-linking of TGase-catalysed protein improved thermal stability (Muguruma et al., 2003). This may result in some changes in thermal protein denaturation and aggregation patterns, which could affect gel characteristics. Frankfurters with caseinate presented the highest lightness and the lowest redness (Table 5); several other authors have reported that caseinate addition to frankfurters decreased the redness of the product (Pietrasik & Jarmoluk, 2003).

Various different non-meat ingredients have been used as salt (NaCl) replacers, among them other chloride salts like KCl. Substitution of the potassium for the sodium ion scarcely modifies the TGase activity (95 versus 90) (Ajinomoto, commercial information), but as far as we know there have been no studies to assess the combined effect of TGase and KCl on physicochemical characteristics of meat emulsions. When compared with the product containing only TGase (MTG sample), the results for the MTG/KCl sample are consistent with higher solubility of the protein (higher ionic strength due to KCl); this would facilitate interaction between salt soluble protein and enzyme and enhance the ability of proteins to form a network. This means that as heating proceeded, a more stable, elastic and rigid protein gel matrix was formed (Tables 3 and 4). Different ratios of salt-soluble proteins may have been involved in the network formation process due to different salt (Na and K) and chloride levels (ionic strength), giving better physicochemical properties in the control meat system than in those prepared with TGase and KCl (MTG/KCl sample) (Tables 3 and 4).

Dietary fibres are not only desirable for their nutritional properties but also for their functional and tech-

nological properties. Dietary fibres from different sources have been studied alone or combined with other ingredients for formulation of different meat products, with a view to increasing cooking yields thanks to their water- and fat-binding properties, and to improving texture (Thebaudin et al., 1997). The wheat fibre used in this experiment was almost 95% insoluble. Insoluble fibre particularly favours water-binding properties and fat absorption capacity (Backers & Noli, 1997; Thebaudin et al., 1997), thus helping to reduce cooking loss and to improve emulsion stability (Table 3). Water may be bound to insoluble polysaccharides by capillary effects or by hydrogen bonds, ionic bonds and/or hydrophobic interactions, and by surface tension in the pores of the matrix (Thebaudin et al., 1997). Insoluble fibres can influence food texture owing to their water-binding ability and swelling properties (Thebaudin et al., 1997). Insoluble fibre can enhance the consistency of meat products (Table 4) through the formation of an insoluble three-dimensional network (Backers & Noli, 1997) capable of modifying rheological properties of the continuous phase of emulsions. There have been conflicting results regarding the effect of dietary fibre on physicochemical characteristics of meat emulsions, but various studies have shown that it improves water- and fat-binding properties (Cofrades, Guerra, Carballo, Fernández-Martín, & Jiménez Colmenero, 2000; Grigelmo-Miguel, Abadías-Serós, & Martín-Belloso, 1999; Hughes, Cofrades, & Troy, 1997; Thebaudin et al., 1997) and hardening (Cofrades et al., 2000; Todd, Cunningham, Claus, & Schwenke, 1989; Troutt et al., 1992; Claus & Hunt, 1991) of cooked products (Tables 3 and 4). However, despite the technological benefits of fibre, its use in combination with TGase (MTG/F sample) led to gel/emulsion structures with poorer physicochemical characteristics than the control (Tables 3–5).

Consumer demand for functional foods has increased. Frankfurters formulated with walnut and free of NaCl are one answer to this demand. The properties of our meat products showed that TGase in combination with ingredients (caseinate, KCl and fibre) could be used to achieve physicochemical characteristics proximate to those of products with normal NaCl levels. We would conclude that it is not feasible to produce a salt-free frankfurter with added walnut using TGase alone. The combination of TGase with caseinate, KCl or fibre led to a firmer gel network with better water- and fat-binding properties than in samples made with TGase only. Ranking of ingredient efficiency in combination with TGase showed caseinate > KCl > fibre. These results suggest that given the right combination of TGase with several of these components (used together) in a meat emulsion with no added NaCl, it should be possible to produce a functional frankfurter that rivals salt-containing meat systems in physicochemical properties.

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