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Cooking effect on fatty acid profile of pork breakfast sausages enriched in conjugated linoleic acid by dietary supplementation or direct addition

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ABSTRACT

The effectiveness of increasing CLA in pork products through animal dietary supplementation or direct addition in the product formulation has been studied, and the effect of grilling on dry matter and fat contents and fatty acid composition has been analysed. Sausages made with meat and back fat from pigs with CLA dietary supplementation had the highest saturated fatty acid content. Sausages from dietary supplementation and direct addition had CLA levels between 6% and 7% of total fatty acids. Moisture and fat contents decreased and increased respectively after cooking for the three sausage types (control, dietary supplementation, direct addition). Grilling had little effect on fatty acid levels, especially for sausages with direct addition in the product formulation. In general, saturated fatty acids increased and poly-unsaturated fatty acids decreased due to the increase of C16:0 and to the decrease of C18:2 $n-6c$ and C18:3 $n-3$ fatty acids. Added CLA, both from animal dietary supplementation or direct addition, remained at similar levels in cooked sausages to those found in raw sausages.

1. Introduction

The relationship between diet, health and lifestyle is now a key focal point for consumers, researchers and policy makers alike as we witness an increase in obesity and the rise of diet-related chronic diseases (Swinburn, 2009). A key objective in the European Technology Platform on Food for Live focuses on ensuring that the healthy choice is an easy choice for consumers. The meat industry is addressing this demand by adopting strategies to produce meat products which have more beneficial ingredient profiles in a value-added manner. One such strategy includes the incorporation of functional lipids into existing meat products to provide a healthier version of an existing product.

Conjugated linoleic acid (CLA) is a collective term for a group of octadecenoic acids that are geometric and positional isomers of linoleic acid (C18:2) (Pariza, Park, & Cook, 2001). CLA has been shown to have a variety of biological effects (Hur, Park, & Joo, 2007). Several health benefits, such as anticancer, anti-oxidation, anti-atherosclerosis and improving immuno-responses (Belury, Nickel, Bird, & Wu, 1996; Lee, Kritchevsky, & Pariza, 1994; Miller, Stanton, & Devery, 2001; Pariza & Hargraves, 1985; Park et al., 1999) have been reported for CLA. These substances have been found in the meat and milk of ruminants, where they are mainly formed by biohydrogenation of grass derived fatty acids. Pork contains only small amounts of CLA because pig is a mono-gastric animal (Chin, Liu, Storkson, Pariza, & Ha, 1992).

Interest in dietary supplementation with CLA for pigs increased during the last decade due to its potential to improve productive, carcass and meat quality traits and, at the same time, for obtaining meat and meat products enriched in CLA (Marco et al., 2009; Martín, Antequera, Muriel, Andrés, & Ruiz, 2008a; Schmid, Collomb, Sieber, & Bee, 2006). A second approach for increasing CLA in meat products is its direct addition as an ingredient during the manufacturing process (Hah et al., 2006; Martín, Ruiz, Kivikari, & Puolanne, 2008b). In addition to the healthy benefits of CLA, its addition into products provides a strategy for partial replacement of saturated fatty acids in the diet by unsaturated fatty acids (Martín et al., 2008a).

While strategies can be enacted to improve the ingredient profile of foodstuffs the cooking method can have an impact on the levels of the beneficial ingredient in the product which is ready to consume. Some studies focused on the influence of processing and cooking on CLA content in meat products that naturally contain CLA such as beef (Ma, Wierzbicki, Field, & Clandinin, 1999; Shantha, Crum, & Decker, 1994) or lamb meat (Badiani et al., 2004). However, little work has been presented which assesses the impact of cooking on the fatty acid profiles of CLA supplemented meat products.

The aim of the present study was to study the effects of grilling on the chemical and fatty acid composition of pork products enriched in CLA through animal dietary supplementation or through direct addition in the product formulation. In addition this study

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aims to compare the useful of CLA dietary supplementation or direct addition into breakfast sausages.

2. Material and methods

2.1. Treatments

Sausages were manufactured according to three different protocols (control, dietary supplementation and direct addition in the formulation) using Boston Butt (*Musculus infraspinatus*, *M. supraspinatus*, *M. subscapularis* and *M. serratus ventralis*) and back fat removed from the pigs 24 h after slaughter. The CLA supplement used for both, diet and formulation supplementation, was Luta-CLA 60 (BASF, Germany), which consists of 56% (w/w) of the two main CLA isomers (*t*9, *c*11 and *c*10, *t*12), dissolved in a base of linoleic acid.

Dietary supplementation with CLA was carried as reported in Marco et al. (2009). Ten female pigs of approximately 40 kg live weight were selected and their diet was supplemented with 2.0% of CLA (3.57% of total oil added per tonne of feeding). Pigs were fed *ad libitum* from hopper feeders. The feeding trial lasted for 8 weeks, after which the animals were slaughtered having reached live weights of approximately 95 kg.

Pork breakfast sausages with CLA added in the formulation were prepared with meat and back fat from pigs with non-CLA enriched diets and live weights of approximately 95 kg, where 2% Luta-CLA (1.12% of CLA), substituting the same weight of back fat, was added to the mixture during the manufacturing process.

As a control, another treatment was prepared using meat and back fat from pigs fed with a non-CLA enriched diet and with no added CLA.

2.2. Sausage manufacture

Sausages were manufactured with the following formulation (w/w), 44.2% of lean meat, 18.7% of back fat, 2.5% seasoning, 7.0% rusk and 27.5% water. Prior to manufacture, back fat was chopped whilst frozen for 1 min at chopping speed 2 and bowl speed 2 (2, 2) using a bowl chopper (Fatosa C-35-2Z, Fatosa S.A., Sabadell, Spain) and then refrigerated. Diced lean meat, seasoning, overnight-hydrated rusk and ⅓ of the ice water were introduced into the bowl chopper and blended at speed (1, 1) for 20 s. The chopped fat was then added to the bowl along with the remaining ice water. All ingredients were then chopped for 2 min at speed (1, 1) and the mix was stuffed into collagen casings of 16 to the lb. The process was made by triplicate with meat and back fat from each animal (batch). For each batch, the sausages were then vacuum packed in bags containing six sausages, and stored at −20 °C.

Prior to cooking, raw samples were taken from all the treatments for subsequent analysis. Sausages were grilled for 30 min at 200 °C using a domestic oven grill (B-AH51-7 SIEMENS-Electrogerate, GmbH Germany), minced (R301 Ultra Robot Coupe, Robot Coupe UK Ltd., Middlesex, UK), vacuum packed and frozen for subsequent analysis. All results are expressed as the mean of six replicates of each treatment.

2.3. Analysis of fat and dry matter contents

Fat and dry matter contents were analysed with Smart Trac (CEM SMART Trac™ Fat and Moisture Analyzer, CEM Corporation, Matthews, USA), using a combination of microwave drying technology and Nuclear Magnetic Resonance (NMR). Minced samples (two replicates of 3 g) were dried in the microwave to calculate their dry matter content using the difference of weight before and after drying. Dried samples were placed in the NMR analyzer to calculate their fat content.

2.4. Fatty acid analysis

The total fatty acids were extracted, methylated and analysed with an adaptation of the method described by Aldai, Osoro, Barron, and Nájera (2006), which has been reported to be highly effective for poly-unsaturated analysis (Juárez et al., 2008). Separation and quantification of the fatty acid methyl esters was carried out using a gas chromatograph (GC, Varian Star 3400CX, Varian Associates Inc., California, USA) equipped with a flame ionisation detector automatic sample injector, and using a BPX-70 capillary column (120 m, 0.25 mm i.d., 0.2 μm film thickness, SGE, Australia). Tricosanoic acid methyl ester (C23:0 ME) at 10 mg/ml was used as an internal standard. Individual fatty acid methyl esters were identified by comparing their retention times with those of authenticated standards from Sigma (Sigma Chemical Co. Ltd., Poole, UK). Fatty acids were expressed as a percentage of total fatty acids identified and grouped as follows: saturated (SFA), monounsaturated (MUFA) and poly-unsaturated (PUFA) fatty acids. PUFA/SFA ratio and total CLA and Δ9-desaturase activities were calculated.

2.5. Statistical analysis

Statistical analyses were performed using Statistica 7.0 for Windows (StatSoft Inc., 2006). The effects of the different treatments (control, CLA dietary supplementation and CLA addition in the formulation) and cooking process as well as the interaction between them were studied using analysis of variance (multifactor ANOVA).

3. Results and discussion

In general, heat is applied to meat products in different ways to improve its hygienic quality by inactivation of pathogenic microorganisms to enhance its flavour and taste, and increase shelf life (Bognàr, 1998; Pokorny', 1999). During cooking, physicochemical reactions modify the food nutritional value. Cooking induces water loss in the food, increasing its lipid content, while only some fat is lost (García-Arias, Álvarez Pontes, García-Linares, García-Fernández, & Sánchez-Muniz, 2003; Yarmand & Homayouni, 2009).

In all the types of sausages, fat ($P < 0.01$) and dry matter ($P < 0.001$) contents increased after cooking with no interaction ($P > 0.05$) between cooking and treatment observed (Table 1). If expressed on dry matter basis, fat content of control, diet and formulation sausages decreased ($P < 0.001$) from 55.7%, 57.2% and 54.3% to 49.9%, 52.2% and 50.0% respectively, due to cooking losses. However, this was accompanied, by higher decreases in moisture content in all treatments following cooking, resulting in an apparent increase of fat content. This has been reported by other authors: for example Baggio and Bragagnolo (2006) in sausages, meat balls and hamburgers and by Dreeling, Allen, and Butler (2000) in beefburgers. Sheard, Wood, Nute, and Ball (1998) noted a similar effect in pork loin chops.

Fatty acid profiles of the sausages (Tables 2 and 3) were in line with those profiles reported elsewhere for pork products (Baggio & Bragagnolo, 2006; Lauridsen, Mu, & Henckel, 2005; Lo Fiego, Macchioni, Santoro, Pastorelli, & Corino, 2005; Pereira, Tarley, Matsushita, & Souza, 2000). The interaction between the studied factors (cooking and treatment) was significant ($P < 0.05$) for SFA and PUFA indices and for PUFA/SFA ratio (Table 2), as well as for several individual fatty acids (Table 3). Therefore cooking impacted in different ways depending on the type of sausage.

When the fatty acid profiles of the different types of sausages are compared, the levels of SFA of sausages from pigs with CLA dietary supplementation showed higher levels ($P < 0.001$) in comparison to those from the other two types. In this context, Dugan,