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Influence of the salting time on physico-chemical parameters, lipolysis and proteolysis of dry-cured foal "cecina"

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ABSTRACT

The present study deals with the effect of the salting time [0.1 (short time, ST), 0.2 (medium time, MT) or 0.3 (long time, LT) days of salting/kg] on the main technological characteristics of a traditional Spanish dry-cured "cecina" from foal meat. The effects of length of salting time on chemical composition, instrumental colour, lipolysis, proteolysis and textural parameters of dry-cured foal "cecina" were studied.

Almost all physico-chemical parameters were significantly ($P < 0.05$) affected by salting time except pH, intramuscular fat and TBARs index. Batches salted for shorter time showed a greater release ($P < 0.001$) of total free fatty acids (FFA) (21.4 g/100 g of fat). Total free amino acids (FAA) content was also significantly ($P < 0.001$) affected by salting time, since dry-cured foal "cecina" with shorter salting time had higher total FAA content (3099 mg/100 g DM).

1. Introduction

Sodium chloride (NaCl) is the most important ingredient in the manufacturing process of dry-cured foal "cecina" for its contribution to the water-holding capacity, prevention of microbial growth, reduction of water activity, facilitating the solubilisation of certain proteins and conferring a typical salty taste (Lorenzo, 2014). Moreover, salt affects some chemical and biochemical reactions such as proteolysis, lipolysis and lipid oxidation which contribute to the development of texture and typical flavour of dry-cured lacón (Lorenzo, 2014).

On the basis of the scientific information and following health recommendations, the meat industry is trying to develop low-salt products because meat products represent a relatively relevant part of the dietary sodium intake (Armenteros, Aristoy, Barat, & Toldrá, 2009). Due to the increased knowledge about the links between sodium intake and coronary heart diseases, consumers' demand for low-salt meat products with the same quality than normal ones has increased (Ruusunen & Puolanne, 2005). Sodium reduction in meat products is possible but difficult to achieve due to the numerous technological properties of NaCl, especially in the meat industry. In fact, NaCl is an essential ingredient in processed meat products, contributing to the water-holding capacity, colour, fat-holding properties, flavour and texture. Moreover, salt decreases water activity (a_w) and this significantly affects the shelf-life (Taomina, 2010). In addition, it is known that salt has an impact on physicochemical properties, such as colour, water activity and moisture and chloride content (Andrés, Ventanas, Ventanas, Cava, & Ruiz, 2005), proteolysis (Martín, Antequera, Ventanas, Benítez-Donos, & Córdoba, 2001), lipolysis (Andrés, Cava, Martin, Ventanas, & Ruiz, 2005) and sensory characteristics (Purriños et al., 2011), which are involved in the development of the typical flavour of dry-cured meat products (Purriños, Franco, Carballo, & Lorenzo, 2012; Wang, Jin, Zhang, Ahn, & Zhang, 2012).

Equine meat is characterized by low fat, low cholesterol content, and high levels of Fe-heme (Lorenzo, Pateiro, & Franco, 2013). Horse meat is characterized by high levels of unsaturated fatty acids (above 55%); polyunsaturated fatty acids (PUFA), predominantly the essential $n-6$ (linoleic acid) and $n-3$ (alpha-linolenic acid) PUFA and monounsaturated fatty acids (MUFA), primarily oleic acid (Lorenzo, 2013). So, horsemeat could play a major role in this context as a red meat alternative to beef and be claimed as "dietetic" meat (Lorenzo et al., 2014). On the other hand, dry-cured foal "cecina" is a new meat product and up to now, the scientific literature about dry-curing and ripening of foal meat is scarce (Lorenzo, 2014; Lorenzo & Franco, 2012; Lorenzo, Montes, Purriños, & Franco, 2012; Lorenzo, Temperán, Bermúdez, Cobas, & Purriños, 2012). The objective of this study was to evaluate how salting time affects physico-chemical properties, proteolysis and lipolysis of dry-cured foal "cecina".

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2. Material and methods

2.1. Animal management and manufacture of dry-cured foal cecina

For this study, thirty foals from crossing Galician Mountain × Hispano-Bretón (GM × HB) were used. Foals were obtained from an experimental herd of Agricultural Research Centre of Mabegondo (Marco da Curra, A Coruña, Spain). All foals were slaughtered at the age of eighteen months. Immediately after killing, carcasses were chilled at 4 °C in a cold chamber for 24 h. At this point, the knuckle (*quadriceps femoris* composed by: *vastus lateralis*, *vastus intermedius*, *vastus medialis* and *rectus femoris* muscles) was excised from the both sides of each carcass. A total of sixty knuckles with an average weight of 2.58 ± 0.53 kg, were used. Samples were randomly distributed in three groups (20 per treatment). Raw knuckles were salted with an excess of coarse salt. A heap was formed consisting of alternating layers of knuckles and layers of salt. In this way, the knuckles were totally covered with salt. Twenty foal "cecina" was salted 0.1 days/kg (short time; ST), twenty foal "cecina" was salted 0.2 days/kg (medium time; MT) and twenty foal "cecina" was salted 0.3 days/kg (long time; LT) in a salting room with a temperature between 2 and 5 °C and relative humidity (RH) between 80 and 90%. After the salting stage the knuckles were taken from the heap, brushed, washed, and transferred to a post-salting room where they stayed for 40 days at 2–5 °C and around 85–90% RH. After the post-salting stage, the knuckles were smoked using oak wood for 5 days at 12–15 °C and 65–75% RH in a smoke house. After the smoked process the knuckles were transferred to a room at 14–16 °C and 74–78% RH where a drying-curing process took place for 75 days. The air convection in the drying room was intermittent and the air velocity around the knuckles when the fan was running ranged between 0.3 and 0.6 m/s. Samples were analyzed at the end of dry-curing process. A total of each sixty units of dry-cured cecina (20 per treatment) were divided in two. The first half was used for colour measurement on cut surface, for obtaining pieces of 1 × 1 × 2.5 cm for WB and Texture Profile Analysis and for pH measurement. The second half was ground for chemical composition, free fatty acids and free amino acid analysis.

2.2. Analytical methods

2.2.1. pH, water activity, TBARs values and colour parameters

The pH of samples was measured using a digital pH-metre (HI 99163, Hanna Instruments, Eibar, Spain) equipped with a glass probe for penetration (blade for meat, 35 mm). Colour measurements were carried out using a CM-600d colorimeter (Minolta Chroma Meter Measuring Head, Osaka, Japan). Each piece was cut and the colour on the cut surface was measured three times for each analytical point. CIELAB space: lightness, (L^*); redness, (a^*); yellowness, (b^*) were obtained. Chroma (C^*) and Hue (h_{ab}) were calculated from a^* and b^* values according to formula:

$$C^* = \sqrt{(a^*)^2 + (b^*)^2} \quad \text{and} \quad h_{ab} = \arctan\left(\frac{b^*}{a^*}\right)$$

Lipid oxidation was assessed in triplicate by the 2-thiobarbituric acid (TBARs) method of Vyncke (1975) with the modification that samples were incubated at 96 °C for 40 min in a forced oven (Memmert UFP 600, Schwabach, Germany). Thiobarbituric acid reactive substances (TBARs) values were calculated from a standard curve performed with 1,1,3,3-tetraethoxypropane and expressed as mg MDA/kg sample. Water activity was determinated using a Fastlab (Gbx, Romans sur Isére Cédex, France) water activity metre, previously calibrated with sodium chloride and potassium sulphate.

2.2.2. Chemical composition

Moisture, fat, protein (Kjeldahl $N \times 6.25$) and ash were quantified according to the ISO recommended standards 1442:1997 (ISO, 1997), 1443:1973 (ISO, 1973), 937:1978 (ISO, 1978), and 936:1998 (ISO, 1998), respectively. Total chlorides were quantified according to the Carpentier–Vohlard official method (ISO 1841-1:1996).

2.2.3. WB and texture analysis profile

Seven meat pieces of 1 × 1 × 2.5 cm (height × width × length) were removed parallel to the muscle fibre direction and were completely cut using a Warner-Bratzler (WB) shear blade with a triangular slot cutting edge (1 mm thick). Maximum shear force, shear firmness and total necessary work performed to cut the sample were obtained. Textural profile analysis (TPA) test measured in a texture Analyzer (TA.XT.plus of Stable Micro Systems, Vienna Court, UK) by compressing to 60% with a compression probe of 19.85 cm^2 of surface contact at a compression speed of 3.33 mm/s and recording speed was also 3.33 mm/s. Hardness (kg), cohesiveness, springiness (mm) and chewiness (kg × mm) were obtained using the computer software (Texture Exponent 32 (version 1.0.0.68), Stable Micro Systems, Vienna Court, UK).

2.2.4. Free fatty acid

Total intramuscular lipids were extracted from 50 g of ground sample, according to Folch, Lees, and Sloane-Stanley (1957) procedure. Free fatty acids were separated from 20 mg of the extracted lipids using NH_2-aminopropyl mini-columns (Sep-Pak Vac 1 cc, 100 mg, Waters, Milford, MA) as described by Kaluzny, Duncan, Merritt, and Epps (1985). Free fatty acids were transesterified following the method described by Shehata, de Man, and Alexander (1970) with some modifications; 4 mL of a sodium methoxide (2%) solution were added to the fraction, vortexed every 5 min during 20 min at room temperature, then 4 mL of a H_2SO_4 solution (in methanol at 50%), vortexed a few seconds and vortexed again before adding 2 mL of distilled water. Organic phase (containing fatty acids methyl esters) was extracted two times with 1 mL of hexane and the solvent was evaporated until dryness under N_2. The residue was resuspended in 0.5 mL of hexane and transferred to a vial. The FAMEs were stored at −80 °C until chromatographic analysis. Separation and quantification of FAMEs was determined following Domínguez and Lorenzo (2014).

2.2.5. Free amino acids

The extraction and the derivatization of free amino acids were performed, as described by Alonso, Álvarez, and Zapico (1994). The identification and quantification of amino acids were carried out used a HPLC, Alliance 2695 model (Waters, Milford, USA) and a UV/Visible Waters 996 photodiode array detector (Waters Milford, USA) using the conditions described by Alonso, Álvarez, and Zapico (1994) with some minor modifications. Empower 2™ advanced software (Waters, Milford, USA) was used to control system operation and results management. The column used was a reversed phase Kinetex 5 μm C18 4.6 mm ID × 250 mm from Phenomenex (Torrance, CA). The temperature of the column was controlled to 50 °C with a column heater (Spectra Physics 8792). The wavelength of the detector was at 254 nm. Standards of the 21 individual amino acids were supplied by Sigma Chemical Co. (St Louis, MO).

2.3. Statistical analysis

For the statistical analysis of the results of physico-chemical properties, lipolysis and proteolysis an analysis of variance

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