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Short communication

Feasibility of use of fatty acid and triacylglycerol profiles for the authentication of commercial labelling in Iberian dry-cured sausages

Alberto Horcada [a,*], Víctor M. Fernández-Cabanás [a], Oliva Polvillo [b], Baltasar Botella [c], M. Dolores Cubiles [d], Rafael Pino [d], Mónica Narváez-Rivas [e], Manuel León-Camacho [f], Rafael Rodríguez Acuña [c]

[a] Departamento de Ciencias Agroforestales, Universidad de Sevilla, Spain
[b] Servicio General de Investigación Agraria (CITIUS), Universidad de Sevilla, Spain
[c] Instituto Andaluz de Tecnología (IAT), Seville, Spain
[d] Departamento de Estadística e Investigación Operativa, Universidad de Sevilla, Spain
[e] Department of Biochemistry, University of Turku, FI-20014 Turku, Finland
[f] Departamento de Caracterización y Calidad de alimentos, Instituto de la grasa (CSIC) de Sevilla, Spain

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ABSTRACT

In the present study, fatty acid and triacylglycerol profiles were used to evaluate the possibility of authenticating Iberian dry-cured sausages according to their label specifications. 42 Commercial brand '*chorizo*' and 39 commercial brand '*salchichón*' sausages from Iberian pigs were purchased. 36 Samples were labelled *Bellota* and 45 bore the generic *Ibérico* label. In the market, *Bellota* is considered to be a better class than the generic *Ibérico* since products with the *Bellota* label are manufactured with high quality fat obtained from extensively reared pigs fed on acorns and pasture. Analyses of fatty acids and triacylglycerols were carried out by gas chromatography and a flame ion detector. A CP-SIL 88 column (highly substituted cyanopropyl phase; 50 m × 0.25 mm i.d., 0.2 μm film thickness) (Varian, Palo Alto, USA) was used for fatty acid analysis and a fused silica capillary DB-17HT column (50% phenyl–50% methylpolysiloxane; 30 m × 0.25 mm i.d., 0.15 μm film thickness) was used for triacylglycerols. Twelve fatty acids and 16 triacylglycerols were identified. Various discriminant models (linear quadratic discriminant analyses, logistic regression and support vector machines) were trained to predict the sample class (*Bellota* or *Ibérico*). These models included fatty acids and triacylglycerols separately and combined fatty acid and triacylglycerol profiles. The number of correctly classified samples according to discriminant analyses can be considered low (lower than 65%). The greatest discriminant rate was obtained when triacylglycerol profiles were included in the model, whilst using a combination of fatty acid and triacylglycerol profiles did not improve the rate of correct assignation. The values that represent the reliability of prediction of the samples according to the label specification were higher for the *Ibérico* class than for the *Bellota* class. In fact, quadratic and Support Vector Machine discriminate analyses were not able to assign the *Bellota* class (0%) when combined fatty acids and triacylglycerols were included in the model. The use of fatty acid and triacylglycerol profiles to discriminate Iberian dry-cured sausages in the market according to their labelling information is unclear. In order to ensure the genuineness of Iberian dry-cured sausages in the market, identification of fatty acid and triacylglycerol profiles should be combined with the application of quality standard traceability techniques.

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1. Introduction

Dry-cured sausage production is a major industry in the Spanish economy. The most widely consumed types are '*chorizo*' and '*salchichón*'. Both products are manufactured from a mixture of chopped meat (pork and beef), lard, sugars, authorised additives (nitrate, nitrite, and antioxidants), starter cultures and spices. '*Chorizo*' is characterised by the fact that it includes Spanish paprika and garlic in its formulation, while '*salchichón*' includes black pepper [1,2]. Differences in the formulation mixture, origin of product, process of fermentation and ripening determine the quality of the product. A high-value product is one which includes meat and back fat from Iberian pigs [3].

Iberian dry-cured sausages are meat products manufactured according to traditional methods in the south and west of Spain and regulated by a generic Spanish quality standard for all raw and dry-cured sausage products [1]. Among these products, sausages

* Corresponding author. Tel.: +34 954 48 64 48; fax: +34 954 48 64 36.
E-mail address: albertohi@us.es (A. Horcada).

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made from the Iberian breed of pig are considered a high-quality food product because of the intrinsic characteristics of the breed and the raw material used in their feeding. Various studies have demonstrated that diet composition during final fattening processes in pigs has an influence on the quality of Iberian pig products [4,5]. According to traditional extensive production systems, Iberian pigs are raised outside in the Mediterranean silvopastoral system known as '*Dehesa*'. This system includes a final fattening phase called '*montanera*', where pigs feed on natural resources, mainly acorns and pasture in extensive conditions [6,7]. However, when acorns are not sufficient, the final fattening diet of pigs is supplemented with commercial feed specifically for swine production. Spain has legislation on the differentiation of ham or subcutaneous fat of Iberian pigs according to their feeding regime [8]. However, there is no specific legislation regarding the differentiation of Iberian dry-cured sausages according to their origin. Hence, dry-cured sausages manufactured with lard obtained from Iberian pigs fattened in '*montanera*' are usually labelled as *Bellota*, whilst those containing fat from animals fed with compound feeds are generically called *Ibérico*. Therefore, a method for authentication of Iberian dry cured products in the marketplace is required.

The sensorial and nutritional attributes of products from animals reared extensively in '*montanera*' mean that they are considered high quality products, while those from animals fed on concentrate are of less quality [9]. Consequently, Iberian pig products labelled as having undergone '*montanera*' fattening have a higher price and this, in turn, increases the possibility of frauds involving other generic Iberian products. In order to discriminate both products, different analytical methods can be found in scientific literature.

Chemical analyses based on gas chromatography to identify the fatty acid profile of the subcutaneous fat of Iberian pigs have been a controversial criterion established for classifying animals according to their feeding system [8–10]. Recently, triacylglycerol composition has been proposed as a method to authenticate Iberian pig fat [11,12] as it has been widely used for oil [13]. In fact, in the study reported by Viera-Alcaide et al. [14] on establishing a method for analysing the triacylglycerol fraction of '*montanera*' and other subcutaneous fat samples by means of gas chromatography and flame ionisation detection, it was concluded that triacylglycerol contents could discriminate between animals from '*montanera*' and concentrate feeding regimes. While determination of fatty acid and triacylglycerol profiles associated with feeding regime has been reported for Iberian subcutaneous fat [15,16] and for Iberian ham [17,18], little information can be found regarding the profiles of Iberian sausages. In fact, generic standards of quality in relation to fatty acid profiles of Iberian ham or subcutaneous fat of Iberian pigs have been regulated [19,8], but there is no generic standard for the classification of dry-cured sausages according to the raw material source.

Since the determination of fatty acid and triacylglycerol profiles has become a feasible system to identify the origin of Iberian pork products in the case of ham and subcutaneous fat, the aim of this work is to explore the feasibility of using these analytical procedures to authenticate Iberian dry-cured sausages labelled as *Bellota* and generic *Ibérico* in the market. The information obtained could be applied to industrial processing of Iberian dry-cured products in Spain, and contribute to the development of the pig industry as well as to new standards for the control of Iberian dry-cured sausages in the market.

2. Experimental

2.1. Dry-cured sausage samples

A total of 81 Iberian dry-cured sausages (42 '*chorizo*' and 39 '*salchichón*'), belonging to 42 commercial brands were purchased. Dry-cured sausages were acquired from commercial stores located in southern and western Spain (Extremadura, Andalucía and Castilla y León). According to the specifications on the label, 18 samples were labelled as *Bellota chorizo* and 24 as *Ibérico chorizo*. In the case of the '*salchichón*' sausages, 18 were labelled as *Bellota* and 21 as *Ibérico*. Products were manufactured in geographical areas recognised by the Spanish quality standard for the production of *Ibérico* products. The mixes used in the manufacture of these sausages are made up of minced lean pork and beef (50–60%), Iberian pork back fat (20–30%), common salt, Spanish paprika (only in '*chorizo*'), sugar, black pepper (only in '*salchichón*'), and garlic (as well as other authorised additives). Differences between *Bellota* and *Ibérico* labels refer to the origin of the fat used in the sausage manufacturing process. A *Bellota* label should indicate that the fat comes from pigs that have been fed on acorns and pastures during their final fattening period. The *Ibérico* label indicates that the fat included in sausage has been obtained from Iberian pigs which were raised on concentrate feeds.

2.2. Fat extraction procedure

All samples were acquired packed under vacuum and were chopped just before laboratory analysis. According to the method proposed by Folch et al. [20], the extraction of sausage fat was performed as follows: 250–400 g of dry-cured sausages were chopped and minced in a domestic mincer (Illico, Moulinex DJE241, France). After that, 2 g from each sample were accurately weighed into an 80 mL tube to which 30 mL of trichloromethane: methanol (2:1) was added. The mixture was homogenised using a laboratory grinder (Ultraturrax T8 IKA-Werke GMBH & Co.KG, Staufen, Germany) for 1 min at 15,500 rpm. Then, 5 mL of potassium chloride (0.88% in distilled water) was added and the mixture was homogenised again using a laboratory grinder for 1 min at 15,500 rpm. The resulting homogenate was filtered and the liquid phase was collected. After allowing it to settle, the aqueous layer was removed, and 1.5 g of anhydrous sodium sulphate was added. The organic phase was then filtered through a Whatman no. 1 filter paper and the solvent was removed at 35 °C. Two duplicates of each sample were made. Fat samples were stored at −20 °C until analysis.

2.3. Fatty acid analysis

At present there is no Spanish legislation on the fatty acid composition of Iberian dry-cured sausages, in fact, there is no official method for dry-cured sausages fat analysis. Therefore, an adaptation of the chromatographic method used for determining the fatty acid composition of total lipids from subcutaneous adipose tissue has been used. Fatty acid methyl esters (FAMEs) were analysed following the method proposed by Viera-Alcaide et al. [14] and De Pedro et al. [21]. In a 10 mL screw-top test tube, approximately 0.1 g of the extracted fat (melted 1.5 min in a microwave at 360 W) was weighed. 8 mL of hexane was added and the mixture was homogenised by shaking. 0.4 mL of 2 N methanolic potassium hydroxide was added, the cap fitted with a PTFE-joint was put on, and the tube was shaken vigorously for 30 s. After leaving it to stratify until the hexane layer became clear, 1.0 mL of this phase was injected in the gas chromatograph. Separation of FAMEs was carried out using a Varian 3800 (Varian Co, Palo Alto, CA USA) gas chromatograph equipped with a flame ionisation detector (FID) and a CP-SIL 88 (highly substituted cyanopropyl phase) silica capillary column of 50 m × 0.25 mm ID, coated with a 0.2 μm film thickness of stationary phase (Varian, Palo Alto, USA). The oven temperature was kept at 170 °C for 17 min and was then raised to 190 °C at a rate of 5.0 °C min^{-1} and held isothermally for 9 min. The injector and detector temperature was kept at 250 °C. Helium was used as the carrier (1 mL min^{-1}