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Fatty acid composition of intramuscular fat and odour-active compounds of lamb commercialized in northern Spain

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ABSTRACT

Muscle fatty acid composition and odour-active compounds released during cooking were characterized in lamb chops (*Longissimus thoracis et lumborum*, $n = 48$) collected at retail level in northern Spain. Lamb samples were classified in two groups according to their 10 *t*/11 *t*-18:1 ratio: ≤1 (10 *t*-non-shifted, $n = 21$) and > 1 (10 *t*-shifted, $n = 28$). Higher n-3 polyunsaturated fatty acid, vaccenic (11 *t*-18:1) and rumenic acid (9*c*,11 *t*-18:2), and *iso*-branched chain fatty acid contents were found in non-shifted lamb samples while n-6 polyunsaturated fatty acid, internal methyl-branched chain fatty acid, and 10 *t*-18:1 contents were greater in shifted samples. Regardless the fatty acid profile differences between lamb sample groups, odour-active compound profile was very similar and mostly affected by the cooking conditions. Overall, the main odour-active compounds of cooked lamb were described as "green", "meaty", "roasted", and "fatty" being methyl pyrazine, methional, dimethyl pyrazine, and dimethyl trisulphide the main odour-active compounds. Aldehydes and alcohols were the most abundant volatiles in all samples, and they were mostly originated from the oxidation of unsaturated fatty acids during cooking.

1. Introduction

In Mediterranean countries, suckling (< 7 kg carcass weight) and light (5–13 kg) lamb production have been prioritized for many years (Sañudo, Sanchez, & Alfonso, 1998). Particularly in Spain, suckling lamb production is concentrated in northern regions and the consumer prefers the sensory characteristics of suckling and light lambs, in terms of texture and flavour, compared to older and heavier lambs (Mediano, Mitxeo, Villalba, Beristain, & de Elizagarate, 2009; Sañudo, Sanchez, et al., 1998; Sañudo, Nute, et al., 1998). These specific characteristics could be related to the impact of production system, primarily age at slaughter and feeding regime, in the nutritional and sensory properties of lamb but also in the ewe's milk, being this the only feed source of suckling lambs (Almela et al., 2010; Sañudo, Sanchez, et al., 1998; Sañudo, Nute, et al., 1998). At retail level, however, due to the seasonality of this type of product (suckling or light lamb), other sheep meats are introduced in the market which are most of the times obtained from older animals, produced under different systems, and usually sold at lower prices, subjecting local producers into a price competition (Mediano et al., 2009).

From a nutritional point of view, fatty acid (FA) composition of lamb is particularly affected by changes in the production system that can modify rumen environment and biohydrogenation processes of dietary unsaturated FAs. Thus, a high proportion of forage in the diet promotes the regular biohydrogenation pathway that accumulates n-3 polyunsaturated FAs (PUFA) and vaccenic acid (11 *t*-18:1) in milk and meat. In contrast, a high proportion of concentrate in the diet provokes a modification in the regular biohydrogenation pathway towards the accumulation of 10 *t*- instead of 11 *t*-18:1 and a higher n-6/n-3 ratio in milk and meat (Bessa, Alves, & Santos-Silva, 2015; Shingfield & Griinari, 2007).

From a sensory point of view, volatile compounds responsible of the characteristic lamb flavour are generated by chemical reactions such as lipid oxidation during lamb cooking. Therefore, the FA profile of the meat associated to a production system could affect the sensory quality of lamb. Most of the carbonyl volatile compounds are generated from the oxidation of unsaturated FAs and can be powerful odorant compounds (Vasta, D'Alessandro, Priolo, Petrotos, & Martemucci, 2012). Different compounds have been identified depending on unsaturated FA substrate; meats with high content of α-linolenic acid (18:3n-3) release compounds such as 1-penten-3-ol and 2-ethyl-furan while meats with high content of linoleic acid (18:2n-6) release other compounds

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such as heptanal and (*E*,*E*)-2,4-decadienal (Elmore, Campo, Enser, & Mottram, 2002). During cooking, carbonyl compounds originated from lipid oxidation can also react with amino compounds (*i.e.*, non-enzymatic browning reactions) where strong meaty odours derived from N-heterocyclic compounds like alkyl pyrazines and sulfur-containing compounds like 2-methyl-3-furanthiol are formed (Elmore & Mottram, 2006; Farmer, Hagan, & Paraskevas, 1999). The volatile profile of cooked lamb could be affected not only by ante-mortem factors (*i.e.*, breed, feeding), but also by other post-mortem factors related to the analysis (*i.e.*, sample preparation, cooking method, extraction method; Vasta & Priolo, 2006; Vasta, Ratel, & Engel, 2007).

Most of the scientific literature related to ovine volatile compounds has been focused on heavier sheep carcasses produced in countries like UK, Australia or New Zealand. However, studies focused on suckling and light lambs are limited, probably because their production is localized in certain regions. Therefore, the objective of this study was to characterize the nutritional and sensory quality of lamb commercialized in northern Spain and collected in two time periods (peak consumption periods). Previously, very few differences were observed between peak consumption periods and type of retail stores due to the considerable variability among subcutaneous fat samples (Bravo-Lamas, Barron, Kramer, Etaio, & Aldai, 2016). However, using the 10 *t*/11 *t* FA ratio of the tissue, two lamb groups with very characteristic FA profile were differentiated which were related to the main ruminal biohydrogenation pathway and the type of *trans*-18:1 isomer deposition in the tissues. According to Bessa et al. (2015), the 10 *t*/11 *t* FA ratio varies in a continuous range from very low values (*i.e.*, 0.1) to very high values (*i.e.*, 20) (Aldai, de Renobales, Barron, & Kramer, 2013). This ratio is generally below 0.3 in animals fed forage based diets even if supplemented with oil (Aldai et al., 2011; Rosa et al., 2014), and increased ratios are obtained when dietary forages are replaced by starchy feedstuffs. At present, there is not an established value of 10 *t*/11 *t* FA ratio above which 10 *t*-shift occurred, but according to Bessa et al. (2015), intuitively, 1 could be considered as the threshold. From here, '10 *t*-shifted' samples were called the ones with 10 *t*/11 *t* FA ratio over 1 and '10 *t*-non-shifted' samples were called the ones with 10 *t*/11 *t* FA ratio below 1.

In the present study, the same two groups of lambs (10 *t*-shifted and 10 *t*-non-shifted) have been investigated where muscle FA composition and odour-active volatile compounds were characterized, and the relationship between unsaturated FAs and the volatile compounds generated by lipid oxidation in cooked lamb was studied.

2. Materials and methods

2.1. Lamb samples

Details about the retail collection of lamb chops and available information on the commercial category of the animals were previously reported in Bravo-Lamas et al. (2016). We recognized that important background information (*i.e.*, breed, diet, age, weight at slaughter, etc) was not available due to the nature of the study (commercial survey carried out in different retail stores). Briefly, lamb samples ($n = 48$) from the Basque Country and Navarre regions (northern Spain) were purchased in two times of the year close to peak consumption periods (May and December 2013; $n = 24$ in each period) from 24 different retail stores including small butcher-shops and medium-large grocery stores. From each store and sampling period 10 to 15 chops, from the same animal, were purchased. In the laboratory, *Longissimus thoracis et lumborum* (LTL) muscle was separated and split in two portions for volatile compound and FA determinations. Samples for volatile compound analysis were vacuum packed and stored at −80 °C, while those for FA analysis were freeze-dried, ground, vacuum packed and stored at −80 °C. The average proximate chemical composition of LTL samples was (% of fresh weight): 75.7 ± 0.6% of water, 19.9 ± 0.5% of protein, and 2.76 ± 0.74% of fat (average ± standard deviation). For comparison purposes and as stated in the objective of the study, from the 48 lamb samples collected two groups were considered according to the 10 *t*/11 *t* FA ratio: ratio ≤ 1 ($n = 21$; 10 *t*-non-shifted samples), and ratio > 1 ($n = 27$; 10 *t*-shifted samples). Even though 10 *t*-18:1 was the predominant *trans* isomer, the large number of lambs with a low 10 *t*/11 *t* FA ratio could be linked to the local Latxa breed managed under semi-extensive conditions. From May on, flocks usually spend most of the time grazing with a residual concentrate contribution in some cases. In winter, however, due to grass shortage, animals are usually managed indoors and concentrate-fed. The variability in 10 *t*- and 11 *t*-18:1 content of the lamb samples was widely discussed in the previous work (Bravo-Lamas et al., 2016). However, despite the large intra-group variability for the content of 10 *t*- and 11 *t*-18:1, mean 10 *t*/11 *t* FA ratio values were 0.22 ± 0.14 (ranging from 0.11 to 0.49) for 10 t/11 *t* < 1 and 6.61 ± 6.46 (ranging from 1.17 to 25.90) for 10 t/11 *t* > 1 groups.

The volatile composition of three lamb samples were not performed due to the small amount of LTL muscle of these samples and, therefore, 19 samples from 10 *t*-non-shifted and 26 samples from 10 *t*-shifted groups were analyzed.

2.2. Fatty acid analysis

Total lipids were extracted from 1.5 g of freeze-dried meat with chloroform-methanol (2:1, *v*/v; Folch, Lees, & Stanley, 1957). Lipid aliquots of 10 mg were methylated separately using acidic (methanolic hydrochloric acid 3 N, Supelco, Bellefonte, PA, USA) and base (sodium methoxide, 0.5 N methanolic base, Supelco) catalysis (Kramer, Hernandez, Cruz-Hernandez, Kraft, & Dugan, 2008). Prior to methylation, 1 mL of internal standard was added (1 mg/mL of equal amounts of esterified 13:0 and 23:0; Nu-Check Prep Inc., Elysian, MN, USA). The obtained FA methyl esters were analyzed using a 7890A gas chromatograph with flame ionization detector (Agilent Technologies, Madrid, Spain) and coupled to a 7693 automatic injector (Agilent Technologies). FA methyl ester separation was performed as previously described in Aldai et al. (2012) following specific chromatographic conditions reported by Kramer et al. (2008) for the SP-2560 column (100 m, 0.25 mm i.d., and 0.20 μm film thickness; Supelco) and by Delmonte et al. (2011) for the SLB-IL111 ionic liquid column (100 m, 0.25 mm i.d., and 0.20 μm film thickness; Supelco).

Identification of FA methyl esters using specific reference standards was detailed in Bravo-Lamas et al. (2016). When confirmation of the chemical structure was required, FA methyl ester fractionation with silver-ion solid phase extraction cartridges was performed (Belaunzaran, Bravo-Lamas, Kramer, & Aldai, 2014; Kramer et al., 2008). The content of FAs in lamb samples was expressed as mg per g of fat.

2.3. Odour-active compound analysis

After thawing the LTL samples from the same store overnight at 4 °C (from the same animal and sampling period), there were manually cut into pieces of 4 cm^3 (2 × 2 × 1) approximately using a thick knife. Each piece was wrapped in aluminium foil, and all were introduced in a bigger aluminium package. One LTL muscle piece was connected to a thermocouple in order to monitor the internal cooking temperature. Aluminium package was closed and cooked in an oven (SelfCooking Center Series, AGBs Rational AG, Germany) to an internal muscle temperature of 80 °C. Then, cooked muscle pieces were cut into smaller pieces using a knife and 20 g were weighed in a screw topper flask that was sealed with a Dreschel head. The flask was placed in a waterbath at 85 °C from which volatiles were extracted by a dynamic headspace using nitrogen at 50 mL/min flow rate for 30 min and collected in a Tenax TA trap (SGC Europe Ltd., Milton Keynes, UK). Then, volatiles were desorbed at 260 °C for 7.5 min and concentrated in a cold trap (10 °C) using an UNITY 1 thermal desorption equipment coupled to an