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EVALUATION OF THE SENSORY ATTRIBUTES ALONG RABBIT LOIN BY A TRAINED PANEL

MARTÍNEZ-ÁLVARO M. AND HERNÁNDEZ P.

Institute for Animal Science and Technology, Universitat Politècnica de València, 46022 València, Spain.

Abstract: The aim of this study was to evaluate and quantify variation in sensory attributes along the *Longissimus dorsi* (LD) muscle in rabbits. A descriptive analysis was performed by a panel of 8 assessors previously trained in the evaluation of rabbit meat. Reference standards used in training for the evaluation of rabbit meat are also described. Sensory attributes rabbit and liver odour, rabbit and liver flavour, toughness, juiciness and fibrousness were assessed in 56 rabbits from a divergent selection experiment for intramuscular fat (28 slaughtered at 9 wk and 28 slaughtered at 13 wk). Immediately after cooking, loins were cut lengthwise into 4 equidistant pieces from caudal to cranial end (LD_1, LD_2, LD_3 and LD_4). Assessors were able to detect and quantify a longitudinal sensory variation in muscle LD. Caudal extreme LD_1 was tougher and more fibrous than LD_2, LD_3 and LD_4, and less juicy than LD_3 and LD_4. The greatest variation was found between caudal and cranial ends, with LD_1 being 9% tougher (P=0.99), 11% more fibrous (P=1.00) and 12% less juicy (P=0.99) than LD_4. Assessors found few variations along LD muscle in flavour and odour attributes. Location LD_3 showed 9% greater rabbit odour (P=0.99) and flavour (P=0.97) than LD_4, and 8% greater rabbit odour than LD_2 (P=0.97). Our results highlight the importance of randomisation within muscle location in sensory studies on rabbit LD muscle, as there is considerable sensory variation along this muscle.

Key Words: loin, panel training, rabbit, sensory evaluation, within muscle variation.

INTRODUCTION

The *Longissimus dorsi* muscle (LD) shows a lengthwise variation in its metabolism and chemical composition in most livestock species, including rabbits (Vigneron *et al.,* 1976 in rabbits and Faucitano *et al.,* 2004 in pigs). These differences could influence its sensory properties.

Most sensory studies in rabbit meat are performed in LD muscle (Gondret *et al.,* 1998; Hernández *et al.,* 2005 and Gašperlin *et al.,* 2006). Rabbit loin is often cut lengthwise into 4 pieces for its sensory evaluation (see for example Gondret *et al.,* 1998 and Hernández *et al.,* 2005), and the variation in the composition along this muscle should be taken into account when randomising sample presentation. However, to our knowledge there is no previous work in rabbits evaluating and quantifying the sensory variation along LD muscle. In pigs, Hansen *et al.*, (2004) observed that sensory hardness gradually varied from the caudal to the cranial location in LD, being harder at the caudal location.

A quantitative descriptive analysis by a panel of trained assessors is a good way to objectively describe and compare the sensory properties of food products (Lawless and Heymann, 2010). Selection and training of assessors based on normative indications (AENOR, 2014) are necessary to obtain reproducible assessments with good discriminatory ability. There are some studies describing reference standards used in the training for meat evaluation; for example, Gasperi *et al.* (2005) in lambs or Gorraiz *et al.* (2000) in ruminants. To our knowledge, there is no information on reference standards used in the training of assessors in rabbit meat.

The aim of this study was to evaluate and quantify the sensory variation along LD muscle in rabbits. Additionally, a description of the reference standards used in the training of assessors for the evaluation of rabbit meat is included.

Correspondence: M. Martínez-Álvaro, mamaral9@upv.es. Received *July 2017* - Accepted *November 2017.*
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MATERIAL AND METHODS

Animals

This study was performed with 56 rabbits from the sixth generation of a divergent selection experiment for intramuscular fat (IMF). More details of this experiment can be found in Martínez-Álvaro *et al.* (2016a and b). Litters were homogenised at birth up to 9 kits per litter. Rabbits were reared collectively from weaning to slaughter and fed *ad libitum* with a commercial diet. They were under a constant photoperiod of 16:8 h and controlled ventilation.

Twenty-eight rabbits (14 from the line selected for high IMF and 14 from the line selected for low IMF) were slaughtered at 9 wk of age and another 28 (14 per line) were slaughtered at 13 wk of age. Live weights of the animals were 1647 g at 9 wk and 2596 g at 13 wk on average. Animals were slaughtered using electrical stunning and exsanguination. After slaughter, carcasses were chilled for 24 h at 4°C. From each animal, both LD muscles were excised. Right muscles were vacuum packed and stored at –20°C until sensory analysis, whereas left muscles were reserved for other analyses (Martínez-Álvaro *et al.,* 2016b). All experimental procedures involving animals were approved by the Universitat Politècnica de València Research Ethics Committee, according to Council Directives 98/58/EC and 2010/63/EU.

Sensory analysis

Panel training. Eight people unrelated to the experiment and regular consumers of rabbit meat were trained in the sensory evaluation of rabbit meat following the recommendations in the UNE-EN-ISO 8586:2014 standard (AENOR, 2014). Sensory attributes considered in the training are described in Table 1. These sensory attributes are commonly used in the evaluation of rabbit meat (see for example Hernández *et al.,* 2000*,* 2005; Gašperlin *et al.,* 2006 and María *et al.,* 2008) or were suggested by the assessors in a preliminary session evaluating rabbit meat. Sensory attributes were trained individually and discussed in joint evaluations during 6 sessions of approximately 1 h each, distributed in 2 wk. Training was performed to ensure the assessors agreed on the definitions of sensory attributes and were able to detect inherent variation present in rabbit meat.

Table 2 shows the composition of the reference standards for low, medium and high intensities of rabbit and liver odour and flavour. Reference samples were prepared using 9 wk rabbit meat from the hind leg as a meat matrix. They were chosen according to the studies of Gasperi *et al.* (2005) in lamb and Maughan *et al.* (2012) in beef, with some modifications to adapt them to rabbit meat. Low intensity standards were prepared soaking the meat in water 24 h to extract water-soluble odour components (firstly proposed in beef by Carmack *et al.,* 1995). Medium and high standards were elaborated adding different amounts of rabbit perirenal fat or rabbit liver to the meat matrix, as described in Table 2. Ingredients were minced to obtain homogeneous reference samples. Reference samples of 50 g were vacuum packed and cooked at 80°C for 20 min by immersion in a water bath

Table 1: Sensory attributes and definitions used to evaluate rabbit meat.

Attribute	Definition
Rabbit odour	Intensity of characteristic odour of rabbit meat.
Liver odour	Characteristic odour of organs and blood of animals.
Rabbit flavour	The combination of taste, odour and tactile stimuli perceived retronasally during chewing – referring to the characteristic flavour of rabbit meat.
Liver flavour	The combination of taste, odour and tactile stimuli perceived retronasally during chewing – referring to the characteristic flavour of organs and blood of animals.
Toughness	Force required to bite the meat sample with molar teeth during the initial chewing.
Juiciness	Moisture perceived during chewing, from the moisture released by the sample and from the secreted saliva.
Fibrousness	Number and thickness of fibres perceived during chewing.

Table 2: Composition of the reference standards for different intensities of rabbit and liver odour and flavour attributes proposed in the training for evaluation of rabbit meat.

	Intensity of attribute		
Attribute	Low	Medium	High
Rabbit odour and flavour	50 g of m	40 g of m+10 g of pf	30 g of m+20 g of pf
Liver odour and flavour	50 g of m	40 g of m+10 g of l	30 g of m+20 g of l

m: meat matrix obtained from hind leg of 9 wk rabbits. In low intensity standards, m was soaked 24 h in water to extract water-soluble odour components; pf: rabbit perirenal fat; l: rabbit liver.

with automatic temperature control (HS-B20, IKA Labortechnik, Staufen, Germany). Samples were served hot using heating equipment. Odour and flavour attributes were immediately tested by the assessors after opening the vacuum packed plastic bags.

Table 3 describes the reference standards used for low, medium and high intensities of toughness, juiciness and fibrousness attributes. These standards were chosen by adapting reference standards published in other species (in beef: Carmack *et al.*, 1995 and Braghieri *et al.*, 2012; in lamb: Gasperi *et al.*, 2005) to rabbit meat. Nine wk rabbit hind legs were deboned and used as low reference standards for toughness and fibrousness and as the high reference standard for juiciness. Loins from 9 wk rabbits were used as medium reference standards for the 3 attributes; and loins from adult rabbits (with 30 wk of age or longer) were used as high reference standards for toughness and fibrousness and as the low reference standard for juiciness. Samples were vacuum packed and cooked at 80°C by immersion in a water bath with automatic temperature control during different times (see Table 3). After cooking, loins and hind legs were cut into 4 pieces, wrapped in aluminium foil and served hot to assessors using heating equipment.

Sensory evaluation of the samples. A quantitative descriptive analysis was performed (Lawless and Heymann, 2010). LD muscles were thawed at 4°C for 24 h in their vacuum-packed plastic bags and cooked at 80°C for 1h by immersion in a water bath with automatic temperature control. This cooking procedure was previously described by Ariño *et al.* (2007). Internal temperature of control samples was 72.9±0.3°C, controlled by a penetration thermistor probe (Checktemp 1 Digital Thermometer H198509; Hanna Instruments Deutschland GmbH, Vöhringen, Germany). Immediately after cooking, loins were unpacked and cut lengthwise into four equidistant pieces from caudal to cranial end (LD_1=7th to 5th lumbar vertebra; LD_2=5th to 3rd lumbar vertebra; LD_3=3rd to 1st lumbar vertebra and LD_4=1st lumbar to 9th thoracic vertebra). Samples were wrapped in aluminium foil and served hot using heating equipment.

Each of the 8 assessors evaluated 4 samples per session (one per each LD location) during 7 sessions, following a complete block design (Stell and Torrie, 1980). The samples were evaluated using a 10-cm unstructured continuous line, as recommended by the UNE-EN-ISO 4121:2006 standard (AENOR, 2006). Distances (cm) from the left extreme to the evaluation mark were registered. Assessors tasted the samples with 3-digit blinding codes under red coloured lights to minimise bias, and the order of presentation was randomised according to the line, slaughter age, and LD location (Macfie *et al.,* 1989). Assessors were asked not to smoke, eat or drink anything except water for 1 h before the evaluation sessions, and were provided with water and unsalted bread for cleansing

Table 3: Reference standards for different intensities of toughness, fibrousness and juiciness attributes proposed in the training for evaluation of rabbit meat.

	Intensity of attribute		
Attribute	Low	Medium	High
Toughness	9 wk hind leg meat cooked 20 min	9 wk loin cooked 60 min	adult loin cooked 60 min
Juiciness	adult loin cooked 60 min	9 wk loin cooked 60 min	9 wk hind leg meat cooked 20 min
Fibrousness	9 wk hind leg meat cooked 20 min	9 wk loin cooked 60 min	adult loin cooked 60 min

All rabbit samples were cooked at 80°C by immersion in a water bath; Adult rabbits were 30 wk of age or longer.

Table 4: Descriptive statistics for sensory attributes of *Longissimus dorsi* muscle in rabbits.

Attribute	Mean	SD	CV(×100)
Rabbit odour	4.53	0.98	21.6
Liver odour	1.67	0.97	58.0
Rabbit flavour	4.15	0.97	23.4
Liver flavour	2.17	1.00	46.2
Toughness	4.50	0.90	20.0
Juiciness	3.60	0.87	24.2
Fibrousness	4.50	0.90	20.1

SD: standard deviation; CV: coefficient of variation.

their palate between samples. Evaluations were carried out in a standard laboratory in accordance with the UNE 8589:2010 standard (AENOR, 2010).

Statistical analysis

To correct the assessor effect, sensory data were standardised subtracting the mean and dividing by the standard deviation of each assessor, as recommended by Næs *et al.* (2010). Then, the mean of the model was added to standardised data. Muscle location effect was evaluated with a model including the fixed effects of LD location, line, age, sex and session. Residuals were assumed to be independently normally distributed. A Bayesian analysis was performed. Bounded flat priors were assumed for all unknowns. Marginal posterior distributions were estimated using Gibbs Sampling and convergence was tested for each chain using the Z criterion of Geweke, and Monte Carlo sampling errors were computed using time-series procedures (Sorensen and Gianola, 2002). Chains of 60 000 samples with a burn-in period of 10 000 were used and one sample in 10 was saved to avoid high correlations between consecutive samples. The rabbit program developed in the Institute for Animal Science and Technology (Valencia, Spain) was used to solve the model.

In sensory analyses, it is difficult to interpret what a relevant difference is, so instead of assessing the differences between LD locations, we analysed the ratios between levels. The features of the marginal posterior distributions calculated were: median of the ratio between LD locations, the standard deviation, and the probability (*P*) of the ratio >1 when the median is greater than one or <1 when the median is lower than one. More details of these features can be found in Blasco (2017).

RESULTS AND DISCUSSION

Table 4 shows descriptive parameters of sensory attributes in LD muscle. Rabbit odour and flavour were higher on average and more variable than liver odour and flavour. Our scores are in line with other sensory studies in rabbits in the same muscle (Gondret *et al.,* 1998; Hernández *et al.,* 2000; Gašperlin *et al.,* 2006 and María *et al.,* 2008).

Table 5 shows ratios between LD_1, LD_2, LD_3 and LD_4 locations for sensory attributes. Assessors were able to detect a variation in sensory texture properties along LD muscle. Caudal extreme LD_1 was tougher and more fibrous than LD_2,

Table 5: Features of the marginal posterior distributions of the ratios between *Longissimus dorsi* (LD) muscle locations[1] for sensory attributes in rabbits.

Attribute	LD_1/LD_2			LD_1/LD_3			LD_1/LD_4			LD_2/LD_3			LD_2/LD_4			LD_3/LD_4		
	M	SD	*P*	M	SD	*P*	M	SD	*P*	M	SD	*P*	M	SD	*P*	M	SD	*P*
Rabbit odour	1.05	0.04	0.90	0.98	0.04	0.74	1.06	0.04	0.93	0.93	0.04	0.97	1.00	0.04	0.56	1.09	0.04	0.99
Liver odour	0.96	0.11	0.63	0.91	0.11	0.81	0.86	0.10	0.92	0.94	0.11	0.71	0.89	0.10	0.85	0.95	0.10	0.70
Rabbit flavour	0.99	0.04	0.56	0.98	0.04	0.70	1.06	0.05	0.90	0.99	0.04	0.63	1.07	0.05	0.93	1.09	0.05	0.97
Liver flavour	0.92	0.09	0.81	0.89	0.08	0.91	0.87	0.08	0.94	0.96	0.08	0.68	0.94	0.08	0.75	0.98	0.08	0.59
Toughness	1.06	0.04	0.95	1.07	0.04	0.97	1.09	0.04	0.99	1.01	0.04	0.61	1.03	0.04	0.76	1.02	0.04	0.64
Juiciness	1.00	0.05	0.53	0.93	0.04	0.94	0.89	0.04	0.99	0.94	0.04	0.92	0.89	0.04	0.99	0.95	0.04	0.86
Fibrousness	1.07	0.04	0.95	1.10	0.04	0.99	1.11	0.04	1.00	1.04	0.04	0.82	1.04	0.04	0.85	1.00	0.04	0.55

LD locations: *Longissimus dorsi* muscle cut lengthwise into four equidistant pieces from caudal to cranial end (LD_1=seventh to fifth lumbar vertebra; LD_2=fifth to third lumbar vertebra; LD_3=third to first lumbar vertebra and LD_4=first lumbar to ninth thoracic vertebra); M: median of the marginal posterior distribution of the ratio; SD: standard deviation of the marginal posterior distribution of the ratio; *P*: probability of the ratio >1 when the median is higher than 1 and probability of the ratio <1 when the median is lower than 1.

LD_3 and LD_4, and less juicy than LD_3 and LD_4. The greatest variation was found between caudal and cranial ends, with LD_1 being 9% tougher (P=0.99), 11% more fibrous (P=1.00) and 12% less juicy (P=0.99) than LD_4. Locations LD_2, LD_3 and LD_4 showed hardly any sensory texture variation between them, except for juiciness, which was 12% greater in LD_4 than in LD_2 (P=0.99).

Assessors found few variations along LD muscle in flavour and odour attributes (Table 5). Location LD_3 showed 9% greater rabbit odour (P=0.99) and flavour (P=0.97) than LD_4, and 8% greater rabbit odour than LD_2 (P=0.97). Location LD_4 showed 15% greater liver flavour than LD_1 and 7% lower rabbit flavour than LD_2, but in both cases, the probability of the ratio being different from 1 was moderate (P=0.94 and 0.93, respectively; Table 5).

Sensory variations observed between LD locations may be caused by morphological or metabolic differences within the muscle. Vigneron *et al.* (1976) described an increase in the number of fibres from cranial to caudal loin ends in rabbits. These authors also found an increase in the proportion of intermediate fast-twitch fibres αR (intermediate diameter) in comparison to slow-twitch fibres βR (small diameter) from cranial to caudal loin ends. Their results could explain the greater fibrousness that we found in caudal end in comparison to cranial. A longitudinal variation in LD texture traits has been previously reported in pigs (Hansen *et al.,* 2004), lambs (Shackelford *et al.,* 2004) and beef (Wheeler *et al.,* 2007). In pigs, toughness increased from cranial to caudal end (Hansen *et al.,* 2004), as observed in our study in rabbits. In beef, cranial and caudal ends showed lower instrumental toughness than the middle of the muscle (Wheeler *et al.,* 2007). In lambs, there was a significant interaction between the animal genotype and the loin location, with callipyge lambs being tougher in anterior location and non-callipyge lambs tougher in posterior location (Shackelford *et al.,* 2004).

Our results show that there is a considerable variation in sensory attributes along the LD muscle in rabbits, particularly in texture properties of toughness, juiciness and fibrousness, which varied around 10% between caudal and cranial extremes. Variations in liver and rabbit odour and flavour were also found, but they were lower. Randomisation within muscle location in sensory evaluation studies of rabbit LD muscle is necessary to correct this variability.

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