# **AUTHOR QUERY FORM**

	Journal: JFOE	Please e-mail or fax your responses and any corrections to:
	Article Number: 6167	E-mail: corrections.esch@elsevier.sps.co.in Fax: +31 2048 52799
ELSEVIER		

Dear Author,

Please check your proof carefully and mark all corrections at the appropriate place in the proof (e.g., by using on-screen annotation in the PDF file) or compile them in a separate list.

For correction or revision of any artwork, please consult <u>http://www.elsevier.com/artworkinstructions.</u>

We were unable to process your file(s) fully electronically and have proceeded by



Scanning (parts of) your article



Rekeying (parts of) your article



Any queries or remarks that have arisen during the processing of your manuscript are listed below and highlighted by flags in the proof. Click on the 'Q' link to go to the location in the proof.

Location in article	Query / Remark: <u>click on the Q link to go</u> Please insert your reply or correction at the corresponding line in the proof
<u>Q1</u>	Please note that reference Ruiz et al. (2002) is cited in text but not listed. Kindly check.
<u>Q2</u>	Please note that Fig. 3B and 3C has been changed to Fig. 2B and 2C. Kindly check.
<u>Q3</u>	This section comprises references that occur in the reference list but not in the body of the text. Please position each reference in the text or, alternatively, delete it. Any reference not dealt with will be retained in this section.
<u>Q4</u>	Please note that as Refs. García and Carrapiso (2001) and García and Carrapiso (2001) were identical, the latter has been removed from reference list and ensuing references have been renumbered.
Q1	Reference Ruiz et al. (2002) has been included in the present version of the manuscript (above Sandler and Karo reference, line 491)
Q3	Boletin del Estado (2004) reference has to be deleted (lines 423-427). Norma UNE (1979) had been named in the manuscript (line 208) but not included as a reference. In the present version this reference has been included (line 208).
Q2 Q4	OK
Q5	A space has to be inserted between "." and "Then" (line 236)
<b>Q</b> 6	In the title of Table 2, insert "of" instead of "for"
Q7	MRI instead of "Magnetic Resonance Imaging"

Thank you for your assistance.

**ARTICLE IN PRESS** 

Journal of Food Engineering xxx (2010) xxx-xxx

Contents lists available at ScienceDirect

# ELSEVIER



26

27

28

29

30

31

32

33

34

35

36

37

38

60

61

62

63

64

65

66

67

68

69

70

71

72

73

74

75

76

77

78

79

80

81

82

83

84

journal homepage: www.elsevier.com/locate/jfoodeng

Journal of Food Engineering

# Sensory traits prediction in dry-cured hams from fresh product via MRI and lipid composition

Trinidad Pérez-Palacios<sup>a,\*</sup>, Teresa Antequera<sup>a</sup>, Rubén Molano<sup>b</sup>, Pablo G. Rodríguez<sup>c</sup>, Ramón Palacios<sup>d</sup>

<sup>a</sup> Tecnología de los Alimentos, Facultad Veterinaria, Universidad de Extremadura, Av. Universidad s/n, 10071 Cáceres, Spain

<sup>b</sup> Departamento de Matemáticas, Escuela Politécnica, Universidad de Extremadura, Av. Universidad s/n, 10071 Cáceres, Spain

<sup>c</sup> Departamento de Ingeniería de Sistemas Informáticos y Telemáticos, Escuela Politécnica, Universidad de Extremadura, Av. Universidad s/n, 10071 Cáceres, Spain

<sup>d</sup> Servicio de Radiología, Hospital Universitario Infanta Cristina, Ctra. de Portugal s/n, 06800 Badajoz, Spain

# ARTICLE INFO

23	
12	Article history:
13	Received 18 February 2010
14	Received in revised form 16 June 2010
15	Accepted 20 June 2010

- 16 Available online xxxx
- 17 Keywords:
- 18 Sensory traits
- 19 Iberian ham
- 20 MRI
- 21 Computational texture features
- 22 Lipid composition
- 23 Multiple Linear Regression 24

# ABSTRACT

The aim of this paper is to describe a methodology that can predict Iberian dry-cured ham sensory traits from raw material characteristics, lipid composition and Magnetic Resonance Imaging-based analysis, by using Multiple Linear Regression statistics. Thus, <u>18</u> sensory traits are tried to be defined from <u>10</u> fatty acids and <u>17</u> computational texture features. Dependence linearity within each group of independent variables is determined. Then, Multiple Linear Regression (MLR) is applied, obtaining allowable statistical coefficients (adjusted coefficient of determination,  $\overline{R^2} > 0.750$  and *p*-value < 0.05) for five sensory traits defined from fatty acids (fat hardness, lean hardness, flavour intensity, brightness and juiciness), and four traits from computational texture features (marbling, odour intensity, flavour intensity and redness). Finally, prediction analysis is validated with a display of statistical data ( $R^2_{LOO}$  and <u>p</u>-value<sub>LOO</sub>). Therefore, some sensory traits in Iberian dry-cured hams can be predicted from fatty acids and computational texture characteristics in fresh products.

© 2010 Elsevier Ltd. All rights reserved.

39

6

8

9

# 40 1. Introduction

The Iberian pig is an autochthonous porcine breed developed 41 traditionally in the south-west of Spain. Hams from Iberian pigs 42 fattened on acorn and grass in an extensive rearing system are 43 more highly appreciate by consumers, in Spain as well as in many 44 45 other countries (M.A.R.M., 2009), because of their exceptional sensory attributes (García et al., 1996). Juiciness and flavour intensity 46 47 are two sensory traits that better explained the acceptability of dry-cured ham (Ruiz et al., 2002). The curing process for Iberian 48 01 ham includes three major phases: a first period (salting/post-salt-49 ing) in which low temperature is combined with high relative 50 humidity to allow for salt diffusion within the hams. A second 51 stage at moderately raised temperatures (26-28 °C) and progres-52 sively lowered relative humidity (to 40%) in order to achieve ade-53 quate drying of the hams. Finally, hams are left to mature for 12-54 55 15 months in a cellar (temperatures ranging between 10 and  $22^{\circ}C$ with relative humidity of about 70%). 56

The composition of the fresh Iberian hams, especially lipid-related ones, which are characterized by high levels of intramuscular fat and monounsaturated fatty acids (MUFA) (Cava et al., 2000), has

\* Corresponding author. Fax: +34 927 257110.

0260-8774/\$ - see front matter © 2010 Elsevier Ltd. All rights reserved. doi:10.1016/j.jfoodeng.2010.06.015

a great influence on dry-cured sensory characteristics (Andrés et al., 1999, 2004).

Several factors, including genetics, crossbreeding and rearing system influence the fatty acid profile of Iberian hams (Tejeda et al., 2002; Andrés et al., 2007), having an effect on sensory characteristics (Carrapiso et al., 2003). In fact, some studies have shown the relationship between some fatty acids of raw material and a number of sensory traits in dry-cured hams (Carrapiso et al., 2003; Ruiz-Carrascal et al., 2000). In addition, the proportions of major fatty acids (FA) (palmitic (C16:0), stearic (C18:0), oleic (C18:1 n – 9) and linoleic (C18:2 n – 6) acids) from subcutaneous fat have been used to classify Iberian pigs as a function of their feeding background during the fattening period (Pérez-Palacios et al., 2009).

There is an increased interest in non-destructive methods for analysing meat products, a scope where Magnetic Resonance Imaging (MRI) can be integrated as a non-destructive, non-invasive, non-radiant and innocuous technique. The combination of MRI and image analysis can present decisive capabilities for the characterisation of muscle structures. Different MRI applications have demonstrated the feasibility of this technique for studying meat and meat products. The calculation of intramuscular fat levels in the Iberian ham *Biceps femoris* and *Semimembranosus* muscles can also result from MRI applications (Ávila et al., 2005). The implementation of active contours in MRI can be used to explore

E-mail address: triny@unex.es (T. Pérez-Palacios).

165

181

198

#### T. Pérez-Palacios et al./Journal of Food Engineering xxx (2010) xxx-xxx

85 the Biceps femoris and Semimembranosus muscles in Iberian hams 86 (Caro et al., 2001; Antequera et al., 2007). Other studies have relied 87 on MRI and computational texture features to classify Iberian 88 products as a function of cross-breeding (Cernadas et al., 2001), 89 feeding background (Pérez-Palacios et al., 2010) and in terms of 90 intramuscular fat content and certain sensory attributes (Antequ-91 era et al., 2003). The relationship between sensory traits and some computational texture features has also been demonstrated 92 93 (Pérez-Palacios et al., 2010).

94 Since the 1950s the sensory evaluation science has experienced 95 big developments (Schutz, 1998). Data obtained by means of sensory analysis are as reliable as those from a physical-chemical 96 97 determination (García and Carrapiso, 2001). Sensory analysis measures those features influencing consumers' acceptance, such as 98 99 appearance, texture, odour, taste and flavour. Quantitative-100 descriptive analyses are most widely used to assess the sensory 101 quality of Iberian dry-cured hams (Ruiz et al., 1998; Andrés et al., 102 2004).

103 Some sensory traits of meats have been predicted by using dif-104 ferent statistical analyses. Olivares et al. (2009) carried out a 105 regression analysis for estimating the relationships between main 106 fatty acid proportion in subcutaneous backfat and liver and vitamin A concentration. Partial least squares regression predictive 107 models have been used to predict colour, marbling and wavelet 108 surface texture of beef (Jackman et al., 2008). Other studies based 109 110 on image analysis have succeeded to predict lamb tenderness 111 (Chandraratne et al., 2006) and pork marbling level (Qiao et al., 112 2007) by using different statistical analysis. Taking a step forward, 113 it may be stated that the use of mathematical models for predicting 114 sensory traits in Iberian dry-cured hams as a function of some 115 characteristics in the raw material can turn out to be a highly sig-116 nificant process within this scientific and technological backdrop. Thus it may be assessed the final quality of a long process dry-117 cured product (around 2 years), which has high acceptability and 118 119 price, before starting the processing.

The objective of the present study is to predict sensory traits in
Iberian dry-cured hams from specific characteristics in the raw
material, lipid composition and computational texture features,
using Multiple Linear Regression (MLR).

# 124 2. Material and methods

# 125 2.1. Experimental design

126 This study is based on testing done with 10 castrated male Ibe-127 rian pigs feeding on acorn and grass outdoors. After the fattening 128 period, all the pigs, weighing an average of 160 kg, were slaugh-129 tered by electrical stunning and exsanguination at a local slaugh-130 terhouse. The right and left raw hams were taken from each 131 animal. MR images were then retrieved for all the hams (n = 20), and the next step involved the dissection of the Biceps femoris mus-132 cles in the left limbs  $(n_1 = 10)$  to analyze the lipid composition of 133 the raw hams, while the right hams  $(n_2 = 10)$  began a ripening per-134 iod by following the procedure described in Antequera et al. 135 (2007), where  $n = n_1 + n_2$ . Hams were rubbed with salt, containing 136 about 1% potassium nitrate, and placed in piles of salt at 3 °C and 137 85% relative humidity for 1 day/kg weight (salting). After salting 138 the hams were washed to remove salt from the surface and hung 139 140 at low temperature  $(4 \circ C \pm 1)$  and the relative humidity was progressively lowered to 75% over 80 days, to allow diffusion of salt 141 142 into the hams (post-salting). The hams were then taken to a natu-143 ral dryer at temperatures varying from 4 to 28 °C and 70% to 50% 144 relative humidity during 130 days (drying). Next, the hams were 145 left to mature for 14 months in a cellar at 10-25 °C and relative 146 humidity of 65–80% (cellar). Once the process ended, the Biceps

*femoris* muscles began to be dissected for their submission to the sensory analysis. 148

# 2.2. MRI acquisition 149

Magnetic resonance sequences enable the exploration of Biceps 150 femoris muscles in raw Iberian hams via image analysis. MRI infor-151 mation, stored on a database acquired at the "Infanta Cristina" Uni-152 versity Hospital (Badajoz, Spain), were obtained by using an MRI 153 (Philips Gyroscan NT Intera 1.5 T) scanner. The "body" antenna 154 was used according to sequences of T1 with the following param-155 eters: 120,× 85 mm for field-of view (FOV), 20 ms for echo time 156 (TE), 500 ms for repetition time (TR), 2 mm of thickness for slices, 157 90° for flip angle (a spin echo sequence),  $0.23 \times 0.20$  mm for pixel 158 resolution, and 60 as the number of slices for each ham. The MRI 159 acquisition was done at 20 °C and it took 28 min for each ham. 160 There were overall a total of 1800 images on the database. All 161 the images were in DICOM format, with a  $512 \times 512$  resolution, 162 and were converted into GIF with the same resolution and 256 grey 163 levels. 164

# 2.3. Computer-aided MRI analysis

A software application (Fig. 1) described in Pérez-Palacios et al. 166 (2010) was used for the analysis of MRI. It is freely available on the 167 page of the GIM research group, http://gim.unex.es. The Biceps 168 femoris muscle was detected by using active contours (Caro et al., 169 2001). Then, the maximum rectangular area in the muscle, called 170 Region of Interest (ROI) was automatically selected, and after-171 wards, the ROIs were analysed by applying computational texture 172 analysis. Seventeen computational texture features were calcu-173 lated: Energy, Entropy, Haralicks Correlation (HC), Inverse Differ-174 ence Moment (IDM), Inertia, Cluster Shade (CS), Cluster 175 Prominence (CP), Small Number Emphasis (SNE), Long Number 176 Emphasis (LNE), Number Nonuniformity (NNU), Second Moment 177 (SM), Entropy (ENT), Long Run Emphasis (LRE), Short Run Empha-178 sis (SRE), Grey Level Nonuniforminty (GLNU), Run Length Nonuni-179 formity (RLN) and Run Percentage (RPC). 180

# 2.4. Fatty acid methyl ester preparation and analysis

Lipids were extracted with chloroform:methanol (2:1, v/v)182 according to the method described in Pérez-Palacios et al. (2008). 183 Fatty acid methyl esters (FAMEs) obtained from lipid tissues were 184 assembled by transesterification in the presence of sodium metal 185 (0.1 N) and sulphuric acid within methanol (Sandler and Karo, 186 1992). FAMEs were analysed by gas chromatography, using a Hew-187 lett-Packard HP-5890-II gas chromatograph, equipped with an on-188 column injector and a flame ionization detector (FID). Separation 189 was done on a polyethylenglycol capillary column (60 m long, 190 0.32 mm id, 0.25 mm film thickness) (Supelcowax-10; Supelco, 191 Bellafonte, PA, USA) maintained at 230 °C for 60 min. The injector 192 and detector temperatures were kept at 230 °C. The carrier gas 193 was nitrogen, at a flow rate of 0.8 ml/min rate. The individual com-194 pounds were then identified as a result of the comparison made 195 between their retention times and standard retentions (Sigma, St. 196 Louis, MO, USA). 197

# 2.5. Sensory analysis

The dry-cured hams in this experiment were assessed by 199 trained staff in a panel of 14 members. Eighteen sensory attributes 200 were analysed in dry-cured Iberian hams (Ruiz et al., 1998), 201 grouped in terms of subcutaneous fat appearance and texture (yellow and pink colour, hardness and oiliness), lean appearance and texture (red colour, brightness, marbling, hardness, juiciness and 204

# **ARTICLE IN PRESS**

3

247

250

251

252

253

254

255

256

257

258

259

260

261

262

263

265

266

267

268

269

270

271

272

273

274

275



Fig. 1. Window from software application for the analysis of Magnetic Resonance Imaging.

205 pastiness), taste (salty, sweet and bitter), aroma (odour intensity) 206 and flavour (intensity, cured, rancid and persistence). The analyses 207 were conducted in tasting rooms with the conditions specified in 208 () 3 UNE regulation, i.e., all the sessions were performed at room tem-209 perature in a sensory room equipped with white fluorescent lighting. The software used to record scores in the sensory sessions was 210 the FIZZ Network (version 2.20: Biosystemes, France). The hams 211 were cut into 1.5 mm thick slices, with a slicing machine. Slices 212 213 were then served on plates to panellists. The panel sessions were held at mid-morning, about 4 h after breakfast. Panellists, who 214 were also able to have about 200 ml of water at room temperature, 215 evaluated the different sensory traits by means of a quantitative-216 217 descriptive analysis in a non-structured scale 0-10. Three samples randomly presented to the panellist were analysed in each session, 218 219 where we recorded the panel average of each sample.

## 220 2.6. Statistical prediction

In order to predict the sensory traits of Iberian dry-cured hams 221 as a function of the fatty acid composition and the computational 222 223 texture features of the raw material, three sets of parameters were 224 considered: computational texture features, fatty acids and sensory traits, containing 17, 10 and 18 variables, respectively. An 225 individual ham was the experimental unit for analysis of all data: 226 227 **O7** n = 20 for the Magnetic Resonance Imaging analysis in fresh hams,  $n_1$  = 10 for the fatty acid analysis in fresh hams and  $n_2$  = 10 for the 228 sensory analysis in dry-cured hams. 229

230 A correlation matrix was used to study the linear dependence within each independent variables set (fatty acids and computa-231 tional texture features), and therefore to reduce the number of 232 233 them. The Pearson correlation coefficient, r > 0.700 serves as proof 234 of the existence of such a linear dependence. If two variables are 235 highly correlated, the information provided is the same (Shiranita 5et al., 2000), Then, Multiple Linear Regression (MLR) was applied 236 ( 237 to determine the independent variables (computational texture 238 features or fatty acids) which define a dependent variable (sensory 239 traits). Suitable relationships yield the adjusted coefficient of determination,  $R^2 > 0.750$  and p-value < 0.05. The choice of  $R^2$  240 rather than the coefficient of determination ( $R^2$ ), is due to the fact 241 that  $R^2$  is an estimation of the goodness of the model fit in the population, and any estimated model from a sample fits that sample better than <u>a</u> population. Thus,  $R^2$  overestimates the goodness of fit. In turn,  $R^2$  balances this optimism by considering the sample size and number of variables (Eq. (1)). 246

$$\overline{R^2} = R^2 - \frac{p \cdot (1 - R^2)}{n - p - 1} = 1 - \frac{n - 1}{n - p - 1} \cdot (1 - R^2)$$
<sup>(1)</sup>

where p is the number of independent variables and n the number of cases. In our case, n = 10.

In MLR,  $R^2 \ge 0.800$  is a good indicator of model accuracy (Jackman et al., 2009; Shiranita et al., 2000). Therefore, as  $\overline{R^2} \le R^2$ ,  $\overline{R^2} \ge 0.750$  in our model indicates high expectations in relation to how actual precision can be obtained by this method.

*p*-Value contrasts the null hypothesis (denoted as  $H_0$ ) that the population value is zero (contrast regression) (Eq. (2)), and allows us to decide if there is a significant linear relationship between the dependent variable and all independent variables. A result of *p*-value ≤ 0.05, with the significance level  $\alpha$  = 0.05, will indicate that the hyperplane defined by the regression equation provides a good fit for the cluster of dots.

$$\begin{cases} H_0 : R^2 = 0\\ H_1 : R^2 \neq 0 \end{cases}$$

$$\tag{2}$$

where  $H_1$  is the alternative hypothesis.

Finally, the statistical analysis was validated by using the Leave-One-Out Cross Validation method (LOOCV). It consists in applying MLR without considering the data from one of the 10 studied hams, calculating again the adjusted coefficient of determination  $(R^2_{LOO})$  and *p*-value<sub>LOO</sub>. Thus, MLR was applied 10 times, without considering one ham in each MLR analysis. For validating the statistical analysis the  $R^2_{LOO}$  and *p*-value<sub>LOO</sub> should be close to the values previously established,  $R^2_{LOO} > 0.750$  and *p*-value<sub>LOO</sub> < 0.05. It is thus possible to know if the model maintains the quality statis-

# JFOE 6167 14 July 2010

# **ARTICLE IN PRESS**

4

280

281

282

283

284

285

286

287

288

289

290

T. Pérez-Palacios et al. / Journal of Food Engineering xxx (2010) xxx-xxx

tics, regardless of the presence or absence of some of the hamsstudied.

Analyses were done by using the SPSS package (v.15.0).

# 279 3. Results and discussion

Table 1 shows the average percentage of fatty acids in fresh lberian hams  $(n_1 = 10)$  for the present study. The major fatty acids were oleic (C18:1 n = 9), palmitic (C16:0), stearic (C18:0) and linoleic (C18:2 n = 6) acids (56.23%, 20.60%, 8.06% and 6.41%, respectively), followed by myristic (C14:0), arachidonic (C20:4 n = 6) and  $\alpha$ -linolenic acid (C18:3 n = 3) (0.97%, 0.82% and 0.46%, respectively), while the  $\alpha$ -eicosatridecanoic (C20:3 n = 3),  $\gamma$ -eicosatridecanoic (C20:3 n = 6) and  $\gamma$ -linolenic (C18:3 n = 6) acids showed smaller proportions (0.13%, 0.11% and 0.02%, respectively). The values for the computational texture features obtained after analysing the MRI from fresh Iberian hams are shown in Table 2.

In relation to the sensory analysis of dry-cured hams, Fig. 2 shows mean scores for appearance and texture in subcutaneous fat (Fig. 2A), appearance and texture in lean (Fig. 2B) and odour, taste, and flavour (Fig. 2C), with a display of high scores for fat oil-

#### Table 1

Fatty acid composition (expressed as percentage of fatty acid methylesters ± standard error of the mean) of intramuscular lipids of *Biceps femoris* from fresh Iberian hams.

Fatty acids	
Myristic acid (C14:0)	0.97 ± 0.13
Palmitic acid (C16:0)	$20.60 \pm 0.89$
Stearic acid (C18:0)	8.06 ± 1.80
Oleic acid (C18:1 <i>n</i> – 9)	56.23 ± 1.75
Linoleic acid (C18:2 $n - 6$ )	6.41 ± 0.62
$\gamma$ -Linolenic acid (C18:3 $n - 6$ )	$0.02 \pm 0.003$
$\alpha$ -Linolenic acid (C18:3 $n - 3$ )	0.46 ± 0.19
$\gamma$ -Eicostridecanoic acid (C20:3 $n - 6$ )	0.11 ± 0.02
$\alpha$ -Eicostridecanoic acid (C20:3 $n$ – 3)	$0.13 \pm 0.09$
Arachidonic acid (C20:4 $n - 6$ )	$0.82 \pm 0.26$

# Table 2

Q6 Values of MRI-based texture characteristics for *Biceps femoris* from fresh Iberian hams.

Computational texture features	
Energy	$(9 \pm 1) \times 10^{-4}$
Entropy	$3.20 \pm 0.07$
Haralicks Correlation	$(7 \pm 0.5)  imes 10^{-4}$
Inverse Difference Moment	0.06 ± 0.003
Intertia	607.70 ± 75.28
Cluster Shade	$(-35 \pm 29) \times 10^3$
Cluster Prominence	$(91\pm46)\times10^5$
SNE <sup>a</sup>	3.31 ± 0.81
LNE <sup>b</sup>	8.12 ± 2.50
NNU <sup>c</sup>	$(13.9 \pm 5.8) \times 10^3$
SM <sup>d</sup>	393.11 ± 197.39
ENT <sup>e</sup>	$-7.67 \pm 2.80$
LRE <sup>f</sup>	$1.10 \pm 0.003$
SRE <sup>g</sup>	$0.97 \pm 0.003$
GLNU <sup>h</sup>	132.25 ± 45.46
RLNU <sup>i</sup>	$(4.2 \pm 1.0) \times 10^3$
RPC <sup>j</sup>	$0.96 \pm 0.002$
d CNIE: amall sumbar amabasia	

<sup>a</sup> SNE: small number emphasis.

- <sup>b</sup> LNE: large number emphasis.
- <sup>c</sup> NNU: number nonuniformity.
- <sup>d</sup> SM: second moment.
- e ENT: entropy.
- f LRE: long run emphasis.
- <sup>g</sup> SRE: short run emphasis.
- <sup>h</sup> GLNU: gray level nonuniformity.
- <sup>i</sup> RLNU: run length nonuniformity.
- <sup>j</sup> RPC: run percentage.

iness (4.33), lean juiciness (4.64), redness (5.31), and flavour intensity (5.71). These results agree with those recorded in other works that also analyse Iberian dry-cured hams from pigs fattened with natural resources (acorn and grass) outdoors (Pérez-Palacios et al., 2009; Carrapiso et al., 2003). Q2 299

# 3.1. Independent variables selection

Α

300

A correlation matrix was built for both computational texture 301 features and fatty acids sets. After analysing the Pearson correlation coefficient and because two variables are highly correlated 303 when r > 0.700, the following variables were selected: myristic 304 (C14:0), palmitic (C16:0), stearic (C18:0), oleic (C18:1  $\eta - 9$ ), linoleic (C18:2  $\eta - 6$ ),  $\gamma$ -linolenic (C18:3  $\eta - 6$ ),  $\alpha$ -linolenic (C18:3 306 n - 3) and arachidonic (C20:4  $\eta - 6$ ) acids, with the only acids left 307



**Fig. 2.** Sensory analysis of Iberian dry-cured hams. (A) Appearance and texture of ham subcutaneous fat; (B) appearance and texture of lean and (C) odour, taste and flavour.

5

312

313

314

315

316

317

318

319

320

321

322

323

324

325

326

327

328

329

330

331

332

333

334

335

336

337 338

339

340

341

342

343

344

345

346

347

348

349

350

351

352

353

354

355

356

357

358

# Table 3

Multiple Linear Regression statistical data between Iberian dry-cured hams sensory traits and fatty acids in raw material.

	Multiple Linear Regression	
	$\overline{R^2}$	<i>p</i> -Value
Fat hardness	0.981	0.002
Lean hardness	0.877	0.004
Flavour intensity	0.889	0.010
Brightness	0.805	0.005
Juiciness	0.787	0.016

## Table 4

Multiple Linear Regression statistical data between lberian dry-cured hams sensory traits and computational texture features in raw material.

	Multiple Linear Regression	
	$\overline{R^2}$	<i>p</i> -Value
Marbling	0.988	<0.001
Odour intensity	0.777	0.001
Flavour intensity	0.807	0.005
Redness	0.779	0.002

Table 5

Statistical data for validating fat hardness prediction as a function of fatty acids.

n <sup>a</sup>	$\overline{R^2}_{LOO}$	p-Value <sub>LOO</sub>
Ham 1	0.978	0.0165
Ham 2	0.975	0.0184
Ham 3	0.961	0.0293
Ham 4	0.925	0.0155
Ham 5	0.978	0.0165
Ham 6	0.988	0.0091
Ham 7	0.995	0.0036
Ham 8	0.981	0.0143
Ham 9	0.982	0.0132
Ham 10	0.969	0.0233

<sup>a</sup> *n*: ham excluded in each analysis for calculating the statistical data considering the other nine hams.

# Table 6

Statistical data for validating marbling prediction as function of computational texture features.

n <sup>a</sup>	$\overline{R^2}_{LOO}$	<i>p</i> -Value <sub>LOO</sub>
Ham 1	0.987	0.0000
Ham 2	0.988	0.0001
Ham 3	0.995	0.0001
Ham 4	0.987	0.0000
Ham 5	0.988	0.0001
Ham 6	0.978	0.0001
Ham 7	0.988	0.0004
Ham 8	0.988	0.0001
Ham 9	0.989	0.0001
Ham 10	0.984	0.0002

<sup>a</sup> *n*: ham excluded in each analysis for calculating the statistical data considering the other nine hams.

out from the fatty acid set as follows: y-eicosatridecanoic (C20:3 308 n-6) and  $\alpha$ -eicosatridecanoic (C20:3  $\underline{n}-3$ ) acids, and Energy, 309 HC, Inertia, CS, LNE, SRE and GLNU in the computational texture features set. 311

# 3.2. Dry-cured ham sensory traits predicted from fatty acids and computational texture features of fresh product

Tables 3 and 4 show statistical data of MLR after correlation of dry-cured ham sensory traits (n = 17) and fatty acids (n = 8) and computational texture features (n = 7) in fresh material. Five sensory traits (fat hardness, lean hardness, flavour intensity, brightness and juiciness) could be defined by fatty acids (Table 3), and four (marbling, odour intensity, flavour intensity and redness) could be defined by computational texture features (Table 4). For all these sensory traits,  $R^2$  and p-value were found within the expected range, with outstanding values in some cases, such us fat hardness ( $R^2 = 0.981$  and p-value = 0.002) and marbling ( $R^2 = 0.988$  and p-value < 0.001), and others were found close to the permitted limit in other cases, for example odour intensity ( $R^2 = 0.777$ ) and redness ( $R^2 = 0.779$ ).

The LOOCV method was determined to validate the MLR analysis. As explained above in Section 2, the method consists in applying MLR by skipping the data from one of the 10 hams. Fat hardness and marbling, defined by fatty acid and computational texture features, respectively, have been taken as examples. Tables 5 and 6 show statistical data  $(R^2_{1,00})$  and p-value<sub>1,00</sub>) for validating fat hardness and marbling prediction, respectively.  $R^2_{LOO}$  values were higher than 0.750 in the 10 analyses, ranging between 0.925 and 0.995 in fat hardness, and between 0.978 and 0.995 in marbling. Results for p-value<sub>LOO</sub> were lower than 0.03 in the 10 analyses of both variables. Statistical data for validating the rest of the sensory traits were also found within the established limits. Therefore, it can be indicated that some sensory traits in Iberian dry-cured hams can be predicted from fatty acids and computational texture characteristics in fresh material with statistical accuracy.

Table 7 shows the prediction equations of the Iberian dry-cured hams sensory traits as a function of fatty acids (Eqs. A-E) and computational texture features (Eqs. F-I).

Although the prediction of sensory traits in Iberian dry-cured hams has remained an area of scientific neglect until now the influence of fatty acid composition on many sensory characteristics has been demonstrated. The most obvious correlations appear among stearic (C18:0) and oleic (C18:1 n - 9) acids, and brightness and oiliness. Palmitic (C16:0), stearic (C18:0) and oleic (C18:1 n - 9) acids were observed to be closely related to solid fat content (Davenel et al., 1999; Ruiz-Carrascal et al., 2000; Carrapiso et al., 2003). Juiciness, sweetness, fat hardness and cured aroma have also shown a relationship with fatty acid composition, whereas odour intensity, fibrousnesses, bitterness, aroma intensity, persistence and toasted aroma were not correlated to these chemical components (Carrapiso et al., 2003). The results in this paper also

#### Table 7

Prediction equations of dry-cured ham sensory traits from characteristics of raw material, fatty acids (A-E) and computational texture features (F-I).

Prediction equations	
A	Fat hardness: 81.568 + 10.623C14:0 + 2.028C16:0 - 0.602C18:1n - 9 - 2.449C18:2n - 6 + 1.939C18:3n - 3 + 2.624C20:4n - 3 + 2.644C20:4n - 3 + 2.644C0:4n - 3
В	Lean hardness: 5.782–5.007C14:0 + 2.028C16:0 + 1.366C18:2n – 6 – 7.073C18:3n – 3 + 180.003C18:3n – 6
С	Flavour intensity: 47.300–0.541C16:0–0.213C18:0 – 0.453C18:1n – 9 – 3.699C18:3n – 3 – 1.874C20:4n – 6
D	Brightness: -25.707 + 0.579C16:0 + 0.262C18:1n - 9 + 2.497C18:3n - 3
E	Juiciness: 10.598–0.412C18:0 – 1.300C18:2n – 6 – 7.197C18:3n – 3 – 3.096C20:4n – 6
F	Redness: 8.134 – 8896.878Energy + 0.006GLNU
G	Marbling: 1473.140 – 6872.589Energy + 0.028Inertia + 0.100LNE – 1506.477SRE
Н	Odour intensity: 4.797 – 0.005GLNU
I	Flavour intensity: -4.041 + 3157.723HC + 0.014Inertia - 0.000012CS

421

422

423

424

425

426

427

428 429

430

431

432

433

434

435

436

437 438

439

440

441 442

443

444

445

446

447

448

449

450 451

452

453

454

455

456

457

458

459

460

461 462

463

464

465

466

467

468

469

470 471

472

473

474

475

476

477

478

479

480

481

482

483

484

485

486

487

488

489

490

491

492

493

494

495

496

497

498

499

6

T. Pérez-Palacios et al./Journal of Food Engineering xxx (2010) xxx-xxx

359 demonstrated the presence of these fatty acids within the predic-360 tion equation of the sensory traits being predicted. It can also be 361 observed that the linolenic acid (C18:3 n - 3) was present in the 362 five sensory trait prediction equations from fatty acids (Eqs. A-E). This fatty acid is not a major one, with only 0.46% (Table 1), 363 however it was considered as an important lipid component for 364 365 classifying Iberian hams in terms of the pigs' feeding backgrounds (Pérez-Palacios et al., 2009). 366

Other studies have focused on reaching an approximate solu-367 368 tion for the "semantic gap" existing between the computational features and some biological terms (Reyes et al., 2008; Jian et al., 369 370 2009), and yet, no significant solution has been found. MRI techniques involving T1 allow for the detection of hydrogen and other 371 features like fat fluidity and water retention, which lengthen the T1 372 373 relaxation time (Lufkin, 1998). For this reason, samples with high 374 water amount have a longer T1 time (Wehrli, 2002). Thus, chemi-375 cal components (volatile compounds, amino acids, dipeptides and 376 so on) related to sensory traits (odour and flavour intensity) may modify T1 sequences and lead to differing MRI. Pérez-Palacios 377 et al. (2010) have recently shown the relationship between sensory 378 379 traits and computational texture features in dry-cured hams.

Iberian dry-cured hams sensory trait prediction can therefore
 be achieved from fatty acids and computational textures features
 in fresh material, using Multiple Linear Regression. The use of com putational texture features for predicting some sensory traits
 seems to prove to be a significant step forward, especially because
 Magnetic Resonance Imaging-based analysis is non-destructive.

# 386 4. Uncited references

# 387 Q3 Boletín Oficial del Estado (2004) and Norma UNE (1979),

# 388 Acknowledgments

Trinidad Pérez Palacios wishes to thank the "Junta de Extrema-389 dura" for her pre-doctoral grant. The authors wish to acknowledge 390 the funding received for this research from both the Junta de 391 392 Extremadura (Regional Government Board - Research Projects 393 3PR05B027 and PDT08A021; Consejería de Economía, Comercio e 394 Innovación and FEDER- economic support for research groups: 395 GRU09148 and GRU09025) and from the Spanish Government (Na-396 tional Research Plan) and the European Union (FEDER funds) by 397 means of the grant reference TIN2008-03063. We also wish to thank the "Hermanos Roa" company from Villar del Rey (Badajoz), 398 as well as the "Infanta Cristina" University Hospital Radiology Ser-399 vice, for their direct contribution and support. 400

# 401 References

420

- 402 Q4 Andrés, A.I., Ruiz, J., Ventanas, J., Tejeda, J.F., Mayoral, A.I., 1999. Muscle fibre types
   403 in Iberian pigs: influence of crossbreeding with Doroc breed and rearing
   404 conditions. Annales of Zootechnie 48, 397–405.
   405 Andrés A.I. Cava, B. Ventanas, I. Thovar, V. Ruiz, J. 2004. Sensory characteristics of
- Andrés, A.I., Cava, R., Ventanas, J., Thovar, V., Ruiz, J., 2004. Sensory characteristics of Iberian ham: influence of salt content and processing conditions. Meat Science 68, 45–51.
   Andrés A.I. Cava, R. Ventanas, L. Muriel, F. Ruiz, L. 2007. Effect of salt content and Andrés A.I. Cava, R. Ventanas, L. Muriel, F. Ruiz, L. 2007. Effect of salt content and
- Andrés, A.I., Cava, R., Ventanas, J., Muriel, E., Ruiz, J., 2007. Effect of salt content and processing conditions on volatile compounds formation throughout the ripening of Iberian ham. European Food Research and Technology 225, 677– 684.
- Antequera, T., Muriel, E., Rodríguez, P.G., Cernadas, E., Ruiz, J., 2003. Magnetic resonance imaging as a predictive tool for sensory characteristics and intramuscular fat content of dry-cured loin. Journal of the Science of Food and Agriculture 83, 268–274.
- Antequera, T., Caro, A., Rodríguez, G.P., Pérez, T., 2007. Monitoring the ripening process of Iberian ham by computer vision on magnetic resonance imaging. Meat Science 76, 561–567.
   Ávila, M.M., Durán, M.L., Caro, A., Antequera, T., Gallardo, R., 2005. Thresholding
  - Ávila, M.M., Durán, M.L., Caro, A., Antequera, T., Gallardo, R., 2005. Thresholding methods on MRI to evaluate intramuscular fat level on Iberian ham. Lectures

Notes in Computer Science (LNCS 3523): Pattern Recognition and Image Analysis 697, 704.

Boletín Oficial del Estado, 2004. Orden PRE/3844/2004, de 18 de noviembre, por la que se establecen los métodos oficiales de toma de muestras en canales de

Q3 cerdos ibéricos y el método de análisis para la determinación de la composición

- de ácidos grasos de los lípidos totales del tejido adiposo subcutáneo de cerdos ibéricos.
- Caro, A., Rodríguez, P.G., Cernadas, E., Durán, M.L., Villa, D., 2001. Applying active contours to muscle recognition in Iberian ham MRI. In: IASTED International Conference Signal Processing, Pattern Recognition and Applications, Rhodes, Greece.
- Carrapiso, A.I., Bonilla, F., García, C., 2003. Effect of crossbreeding and rearing system on sensory characteristics of Iberian ham. Meat Science 65, 623–629.
- Cava, R., Ventanas, J., Ruiz, J., Andrés, A.I., Antequera, T., 2000. Sensory characteristics of Iberian ham: influence of rearing system and muscle location. Food Science and Technology International 6, 235–242.
- Cernadas, E., Antequera, T., Rodríguez, P.G., Durán, M.L., Gallardo, R., Villa, D., 2001. Magnetic resonance imaging to classify loin from Iberian pigs. In: Webb, G., Belton, P.S., Gil, A.M., Delgadillo, I. (Eds.), Magnetic Resonance in Food Science. Royal Society of Chemistry, Cambridge, UK, pp. 239–245.
- Chandraratne, M.R., Samarasinghe, S., Kulasiri, D., Bickerstaffe, R., 2006. Prediction of lamb tenderness using image surface texture features. Journal of Food Engineering 77, 492–499.
- Davenel, A., Riaublanc, A., Marchal, P., Gandemer, G., 1999. Quality of pig adipose tissue: relationship between solid fat content and lipid composition. Meat Science 51, 73–79.
- García, C., Carrapiso, A.I., 2001. La calidad sensorial del jamón Ibérico y su evaluación: la cala y la cata del jamón. In: Ventanas, J. (Ed.), Tecnología del jamón Ibérico. Mundi Prensa, Madrid, pp. 391–418.
- García, C., Ventanas, J., Antequera, T., Ruiz, J., Cava, R., Alvarez, P., 1996. Measuring sensorial quality of Iberian ham by Rash model. Journal of Food Quality 19, 397–412.
- Jackman, P., Sun, D.W., Du, D.J., Allen, P., Downey, G., 2008. Prediction of beef eating quality from colour, marbling and wavelet texture features. Meat Science 80, 1273–1281.
- Jackman, P., Sun, D.W., Du, D.J., Allen, P., 2009. Prediction of beef eating qualities from colour, marbling and wavelet surface texture features using homogenous carcass treatment. Pattern Recognition 42, 751–763.

Jian, M., Gou, H., Liu, L., 2009. Texture image classification using visual perceptual texture features and gabor wavelet features. Journal of Computers 4, 763–770. Lufkin, R.B., 1998. The MRI Manual. Mosby-Year Book, St Louis, Missouri.

- M.A.R.M., 2009. Ministerio de Medio Ambiente, Medio Rural y Marino. Dirección General de Industrias y Mercados Alimentarios. Datos de las Denominaciones de Origen Protegidas (D.O.P.). Años, 2002–2006. (http://www.marm.es).
- Norma UNE, 1979. Una norma europea 87004. Pruebas especiales de calidad sensorial.
- Olivares, A., Daza, A., Rey, A.I., López-Bote, C.J., 2009. Dietary vitamin A concentration alters fatty acid composition in pigs. Meat Science 81, 295–299.
- Pérez-Palacios, T., Ruiz, R., Martin, D., Muriel, E., Antequera, T., 2008. Comparison of different methods for total lipid quantification. Food Chemistry 110, 1025– 1029.
- Pérez-Palacios, T., Ruiz, J., Tejeda, J.F., Antequera, T., 2009. Subcutaneous and intramuscular lipid traits as tools for classifying Iberian pigs as a function of their feeding background. Meat Science 81, 632–640.
- Pérez-Palacios, T., Antequera, T., Durán, M., Caro, A., Rodríguez, G.P., Ruiz, J., 2010. MRI-based analysis, lipid composition and sensory traits for studying Iberian dry-cured hams from pigs fed with different diets. Food Research International 43, 248–254.
- Qiao, J., Ngadi, M.O., Wang, N., Gariépy, C., Prasher, S.O., 2007. Pork quality and marbling level assessment using a hyperspectral imaging system. Journal of Food Engineering 83, 10–16.
- Reyes, C., Durán, M.L., Alonso, T., Rodríguez, P.G., Caro, A., 2008. Behaviour of texture features in a CBIR system. Lecture Notes in Artificial Intelligence. Hybrid Artificial Intelligence Systems 5271, 425–432.
- Ruiz, J., Ventanas, J., Cava, R., Timon, M.L., García, C., 1998. Sensory characteristics of Iberian ham: influence of processing time and slice location. Food Research International 31, 53–58.
- Ruiz-Carrascal, J., Ventanas, J., Cava, R., Andrés, A.I., García, C., 2000. Texture and appearance of dry-cured ham as affected by fat content and fatty acid composition. Food Research International 33, 91–95.
- PSandler, S.R., Karo, W., 1992. Source Book of Advances Organic Laboratory Preparations. Academic Press, San Diego.
- Schutz, H.G., 1998. Evolution of sensory science discipline. Food Technology 52, 42–46.
- Shiranita, K., Hayashi, K., Otsubo, A., Miyajima, T., Takiyama, R., 2000. Grading meat quality by image processing. Pattern Recognition 33, 97–104.
- Tejeda, J.F., Gandemer, G., Antequera, T., Viau, M., Garcia, C., 2002. Lipid traits of muscles as related to genotype and fattening diet in Iberian pigs: total intramuscular lipids and triacylglycerols. Meat Science 60, 357–363.
- Wehrli, F.W., 2002. Contrast development and manipulation in MR imaging. In: Atlas, S.W. (Ed.), Magnetic Resonance Imaging of the Brain and Spine. Raven Press, New York, pp. 33–58.