Effect of including carob pulp in the diet of fattening pigs on the fatty acid composition and oxidative stability of pork

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Abstract

The effect of feeding pigs with carob pulp on meat qualitywas investigated. Nine pigswere finished on a conventional concentrate-based diet (control), while two groups received a diet comprising of the same ingredients with the inclusion of 8% or 15% carob pulp (Carob 8% and Carob 15%, respectively). Feeding carob-containing diets reduced the concentration of saturated fatty acids in themuscle, increased the concentration of monounsaturated fatty acids in meat (P b 0.01) and of n-3 polyunsaturated fatty acids (PUFAs) and reduced the n-6/n-3 PUFA ratio (P b 0.001). The meat underwent slow oxidative deterioration over 9 days of storage. However, the Carob 15% treatment increased meat susceptibility to lipid oxidation across storage (P= 0.03), while the dietary treatment did not affect meat colour stability. In conclusion, feeding pigs with carob pulp could represent a strategy, in the Mediterranean areas, to naturally improve meat nutritional value and to promote the exploitation of this local feed resource.

Keywords: Carob pulp, Pork, Fatty acids, Lipid oxidation,Colour stability.

1. Introduction

Carob tree (Ceratonia siliqua L.) is native to the Mediterranean areas and mainly grown in Italy, Spain, Portugal, Greece and Morocco (FAO, 2011). Traditionally, carob fruits have been used in human and animal nutrition. Nowadays, carob fruit constituents found applications in food, pharmaceutical and cosmetic industries, with special interest being devoted to carob gum which is produced from the seeds (Dakia, Blecker, Robert, Wathelet, & Paquot, 2008). Therefore, as the seeds are considered the most valuable part of the fruits, carob pulp can be considered, in some instances, as a by-product resulting from the processing procedure of the pods (Vekiari, Ouzounidoua, Ozturk, & Görkc, 2011) and could find valuable application as local and cheap feed resource for livestock nutrition in the areas of production. Carob pulp contains high levels of sugars, particularly lowmolecular weight carbohydrates, such as sucrose, thus representing a potentially good source of energy in animal diets (Marakis, 1996). On the other hand, carob pulp is characterized by a rather lowprotein and fat content (Avallone, Plessi, Baraldi, & Monzani, 1997). Nevertheless, carob pulp has a favourable fatty acid composition due to the presence of essential fatty acids, such as linoleic and alpha-linolenic acids (Ayaz et al., 2009) and might represent a natural source of desirable fatty acids in the diets of concentrate-fed animals. Studies so far conducted to evaluate the possibility of feeding carob pulp to livestock have mainly focused on ruminants (Priolo, Waghorn, Lanza, Biondi, & Pennisi, 2000; Silanikove et al., 2006) and highlighted that the main limitation to the inclusion of high levels of carob pulp in the diet is its high content of condensed tannins. Highly polymerized condensed tannins are plant secondary compounds, belonging to the heterogeneous group of phenolic compounds, which are receiving considerable attention in animal nutrition. On one hand, tannins can form complexes with proteins and carbohydrates and, when present at high levels in the diet, can act as antinutritional factors for ruminants and monogastric animals (Jezierny, Mosenthin, & Bauer, 2010; Makkar, 2003). On the other hand, the dietary administration of tannins to pigs has been shown to exert positive effects on the functionality of the gastrointestinal tract (Biagi, Cipollini, Paulicks, & Roth, 2010).

Furthermore, tannins are known as potent antioxidants (Hagerman et al., 1998) and, although their efficacy in vivo is still under debate, the dietary administration of tannin-rich feeds to animals is being explored as a strategy to provide natural antioxidants in the diet to improve meat quality traits such as ì oxidative stability (Vasta & Luciano, 2011). Some studies have elucidated the chemical composition of the phenolic compounds occurring in carob pods and have demonstrated that most of these compounds possess strong antioxidant capacity (Kumazawa et al., 2002; Owen et al., 2003; Papagiannopoulos, Wollseifen, Mellenthin, Haber, & Galensa, 2004). Bastida et al. (2009) were able to extend the storage stability of pork meat by using carob extract as a functional ingredient in meat preparation.

Some studies assessed the effect of feeding phenolic-rich plant extracts or tannin-rich feeds, such as chestnut, to pigs on the performance, antioxidant status and meat and fat quality traits (Bermúdez, Franco, Franco, Carballo, & Lorenzo, 2012; Frankič & Salobir, 2011; Pugliese et al., 2013; Rossi et al., 2013). However, due to the high variability of the phenolic compound profiles between different natural sources, it is not possible to directly extrapolate hypotheses fromthe above studies on the possible effects that feeding pigs with carob could exert on meat quality. Very limited information is available on the effect of feeding carob pulp to monogastric animals on meat quality, despite the fact that there is evidence of the use of carob pulp in pig feeding since the New Testament (Luke, 15, 16). To the best of our knowledge, only one study investigated the effects of the dietary administration of carob pulp to growing pigs on somemeat quality traits including intramuscular fatty acid composition (Kotrotsios, Christaki, Bonos, & Floru-Paneri, 2012), while information on the effect of dietary carob pulp on meat oxidative stability has not yet been provided.

Therefore, the objective of the present investigation was to evaluate the effect of the inclusion of two different levels of carob pulp in the finishing diet of pigs on the intramuscular fatty acid composition and oxidative stability of meat.

2. Materials and methods

2.1. Animals and diets

96 Twenty-seven Pietrain × Large White barrows, born at the end of January 2012, and weaned at 5
97 weeks of age (7 kg live weight, approximately) were used. Pigswere fedwith commercial starter
98 concentrates until 60 days of age and, subsequently, animals were grown on commercial grower
99 concentrates until 180 days of age. Then, pigs were randomly assigned to one of three experimental
100 treatments (with 9 animals in each group). One group (control) was fed with a commercial finishing
101 concentrate-based diet. The other two groups received a diet comprising of the same ingredients as
102 the control diet with 8% or 15% carob pulp was also included (groups Carob 8% and Carob 15%,
103 respectively).
104 The composition of the experimental diets is described in Table 1. During the experimental feeding
105 phase (120 days), animals received 2 kg of concentrate (as fed)/day/head and had ad libitumaccess
106 to water. All animals were slaughtered in a commercial abattoir at 300 days of age. Animals were
107 electrically stunned and exsanguinated, the carcass weight was recorded and the muscle longissimus
108 thoracis et lumborum (LTL; approximately 400 g) was removed from each carcass and immediately
109 transported and refrigerated to the laboratory. The experimental protocol was approved by the
110 Animal Welfare Committee of the University of Catania and animals were handled by specialized
111 personnel following the guidelines of the European Parliament and Council (2010/63/EU
112 Directive).
113 *2.2. Analyses of feedstuffs*
114 Samples of the experimental diets and of carob pulp, collected during the trial, were analysed for
115 neutral detergent fibre (NDF) according to Van Soest, Robertson, and Lewis (1991). Furthermore,
116 according to AOAC (1995), feedstuffs were also analysed for ash, crude protein and crude fat (ether
117 extract). Following the procedure described by Makkar, Blümmel, Borowy, and Becker (1993),
118 total phenolic compounds were extracted from the feed samples using aqueous acetone (70% v/v),
119 analysed by means of the Folin–Ciocalteu reagent and expressed as tannic acid equivalents. The
120 fatty acid composition of the feedstuffs was analysed by gas-chromatography using the method
121 described by Sukhija and Palmquist (1988) and was expressed as g/100 g of total fatty acids.

122

123 *2.3. Analyses of meat samples*

124 Fresh LTL samples were divided into two 200-g portions. One portion was immediately vacuum-
125 packaged and stored at−30 °C pending analysis of intramuscular fatty acid composition. The
126 remaining portion was vacuum-packaged and stored at 4 °C. After 24 h of refrigerated storage, bags
127 were opened and the ultimate pH of LTL was measured using an Orion 9106 pH-meter equipped
128 with a penetrating electrode (OrionResearch Incorporated, Boston, MA). Then, each muscle was
129 divided into 3 sub-samples (2 cm thickness) using a knife. The sub-samples were placed in
130 polystyrene trays, covered with PVC film and stored in the dark at 4 °C. Colour and lipid oxidation
131 measurements were performed after 2 h of blooming (day 0) and after 5 and 9 days of storage, using
132 one sub-sample for each day of storage. All the analyseswere performed as described below.

133

134 *2.3.1. Intramuscular fatty acid composition*

135 Intramuscular lipids were extracted according to the method used by Folch, Lees, and Stanley
136 (1957). Briefly, 5 g of LTL was blended with extraction solvent chloroform/methanol (2:1, v/v)
137 twice, filtered, placed in separator funnels and mixed with saline solution (0.88% KCl). After
138 separation into two phases, the chloroform lipid fraction was collected and washed with distilled
139 water/methanol (1:1, v/v). After a further filtration and evaporation by means of a rotary evaporator,
140 lipid extracts were transferred to test tubes for subsequent gas chromatographic analysis. Duplicates
141 of 100 mg of lipid extract were methylated adding 1 ml of hexane and 0.05 ml of 2 N methanolic
142 KOH. Nonanoic acid (C9:0) was used as an internal standard. Gas chromatographic analysiswas
143 performed using a Varianmodel Star 3400 CX instrument equipped with a CP 88 capillary column
144 (length 100 m, internal diameter 0.25mm, film thickness 0.25 μm). Operating conditions were: a
145 helium flow rate of 0.7 ml/min, a FID detector set at 260 °C, a split–splitless injector at 220 °C with
146 an injection rate of 120 ml/min, and an injection volume of 1 μl. The temperature programme of the

147 column was: 4 min at 140 °C and a subsequent increase to 220 °C at 4 °C/min. Retention time and
148 area of each peak were computed using the Varian Star 3.4.1. software. The individual fatty acid
149 peaks were identified by comparison of retention times with those of known mixtures of standard
150 fatty acids (37 component FAME mix, 18919–1 AMP, Supelco, Bellefonte, PA) run under the same
151 operating conditions. Fatty acids were expressed as g/100 g of total fatty acids.
152
153 *2.3.2. Colour stability and lipid oxidation measurements*
154 Meat colour stability was evaluated by measuring the colour descriptors L* (lightness), a*
155 (redness), b* (yellowness), C* (saturation) and H* (hue angle) in the CIE L*,a*, and b* colour
156 spaces.Measurements were performed using a Minolta CM 2022 spectrophotometer (d/8° geometry;
157 Minolta Co. Ltd. Osaka, Japan) set to operate in the specular components excluded (SCE) mode and
158 to measure with the illuminant A and 10° standard observer. The reflectance spectra from 400 to
159 700nmwavelengthwere also recorded for calculation ofmetmyoglobin (MMb) formation according
160 to Krzywicki (1979). All measurements were taken in duplicate directly on the meat surface and the
161 mean values were calculated.
162 Lipid oxidation was determined by measuring the 2-thiobarbituric acid reactive substances
163 (TBARS) according to the method described by Siu and Draper (1978). Meat samples (2.5 g) were
164 homogenized with 12.5 ml of distilledwater using a Heidolph Diax 900 tissue homogenizer
165 (Heidolph ElektroGmbH & Co. KG, Kelheim, Germany) operating at 9500 rpm. During the
166 homogenization, samples were put in a water/ ice bath. Subsequently, 12.5 ml of 10% (w/v)
167 trichloroacetic acid was added to precipitate proteins and then the samples were vortexed.
168 Using a Whatman No. 1 filter paper, the homogenates were filtered and 4 ml of filtratewere added
169 to 1 ml of 0.06Maqueous thiobarbituric acid into pyrex-glass tubes. The tubeswere incubated in
170 awater bath at 80 °C for 90 min and the absorbance of each samplewas read at 532 nm using a
171 Shimadzu UV/vis spectrophotometer (UV-1601; Shimadzu Corporation, Milan, Italy). The

assaywas calibratedwith solutions of known concentration of TEP (1,1,3,3,-tetraethoxypropane) in distilled water. Results were expressed as mg of malonaldehyde (MDA)/kg of meat.

2.4. Statistical analysis

Data of intramuscular fatty acids were analysed using a GLM to test the effect of the dietary treatment (diet: control, Carob 8% and Carob 15%). Results of meat colour stability descriptors (L*, a*, b*, C*, H* andMMb) and of lipid oxidation (TBARS values) were analysed using a GLM procedure for repeated measures. The fixed factors in the model were: the dietary treatment (Diet), the time of storage (time; days 0, 5, 9) and their interaction (diet × time), while individual animal was included as a random factor.

Multiple comparisons of the means were performed using the Tukey's adjustment. The analysis was carried out using the statistical software Minitab, version 16 (Minitab Inc., State College, PA).

3. Results and discussion

3.1. Animal performance, intramuscular fat concentration and fatty acid composition

The dietary treatments did not result in differences in the main performance parameters. Indeed, as shown in Table 2, the final weight of the animals, as well as carcass weight and dressing percentage were comparable between treatments. Very limited information has been provided on the effects of supplementing carob in the finishing diet of pigs on animal performance and meat quality. However, it was demonstrated that the inclusion of up to 12% of carob pulp in pig diets slightly reduced nutrient digestibility (Kotrotsios, Christaki, Bonos, & Florou- Paneri, 2010), but with no detrimental effect on animal growth performance (Kotrotsios et al., 2012). The comparable nutrient composition and energy level between the three experimental diets can explain, in the present study, the lack of difference in the measured performance parameters between treatments. Also, protein and energy levels were comparable between the diets, which could be one of the reasons explaining

198 the fact the concentration of intramuscular fatwas comparable between treatments and was overall
199 acceptable (average value: 2.86 g/100 g muscle; Table 3). Indeed, it has been shown that differences
200 in the energy and proteins in the diets for finishing pigs can result in differences in the
201 concentration of intramuscular fat (Teye et al., 2006).
202 With regard to the concentration of the individual fatty acids, we found that oleic acid (cis-9 C18:1),
203 α-linolenic acid (C18:3 n-3), total n-3 poly-unsaturated fatty acids (n-3 PUFA) and total
204 monounsaturated fatty acids (MUFA) were affected by the dietary treatment ($P \leq 0.001$) and were
205 higher in muscle from pigs fed the diets supplemented with carob pulp, regardless of the level of
206 inclusion (Table 3).
207 Conversely, the meat from animals fed with Carob 8% and Carob 15% diets contained lower
208 percentage of myristic (C14:0), palmitic (C16:0), palmit-oleic (cis-9 C16:1) and linoleic (C18:2 n-
209 6) acids and of the total saturated fatty acids (SFA) and n-6 PUFA ($P < 0.05$) compared to meat
210 from animals fed control diet. The concentration of polyunsaturated fatty acids (PUFAs) tended to
211 be higher in muscle frompigs fed the control diet compared to the intramuscular fat frompigs fed the
212 diets supplemented with 8% and 15% of carob pulp ($P= 0.054$), due to the higher concentration of
213 n-6 PUFA in muscle from pigs fed with the control diet. Therefore, our results showed that the
214 dietary administration of either 8% and 15% carob pulp induced modifications in the acid profile of
215 pork toward a preferential increase of n-3 PUFA rather than of total PUFA,while decreasing the
216 concentration of n-6 PUFA and saturated fatty acids.
217 Few studies evaluated the fatty acid composition of meat from animals fed with carob pulp and very
218 limited information on monogastric animals is available. According to Kotrotsios et al. (2012),
219 including carob in the diet of pigs had no significant effect on fatty acid profile of meat, except for a
220 tendency to increase the levels of PUFA. Additionally, other differences between our study and that
221 described by Kotrotsios et al. (2012) do not allow to make straightforward comparisons.
222 For example, we slaughtered animals at 300 days of age, while Kotrotsios et al. (2012) used much
223 younger animals (180 days). Also, we used carob pulp instead of whole carob pods as in the case of

224 the study of Kotrotsios et al. (2012). Furthermore, while soybean oil was used in our study, the
225 source of vegetal fat used by Kotrotsios et al. (2012)was not specified, and this information could
226 be useful to understand differences in the deposition of PUFA in the muscle (Teye et al., 2006).
227 Finally, Kotrotsios et al. (2012) did not provide the fatty acid composition of the experimental diets,
228 which could largely account for the intramuscular fatty acid composition. Indeed, in the case of
229 monogastric animals, such as pigs, differences in the fatty acid composition of meat can directly
230 reflect differences in the fatty acid profile of the diets (Wood et al., 2008). Some of the results
231 observed in the present study may be partially explained by the fatty acid composition of the
232 experimental diets. For example, the higher levels of n-3 PUFA and the lower n-6/n-3 PUFA ratio
233 in the muscle from pigs fed with the carobsupplemented diets is consistent with the fatty acid
234 composition of the diets, whereby those containing carob among the ingredients increased the levels
235 of n-3 PUFA,while decreasing the n-6 PUFA compared to the control diet, as shown in Table 1.
236 Although, in the present study, the proportion of other ingredients besides carob differed between
237 the diets in order to balance for energy and proteins, some of these results could be attributed to the
238 inclusion of carob pulp. In our experimental diets, carob pulp replaced barley and wheat middlings,
239 while the proportion of corn was increased. However, it is unlikely that the higher levels of corn in
240 the carob-containing diets could be responsible for an increase in n-3 PUFA in the muscle, as corn
241 is recognized as a poor source of alpha-linolenic acid (Raes,De Smet, & Demeyer, 2004). Rather, it
242 has been shown that that carob pod constituents contain remarkable levels of essential fatty
243 acids,with a rather lown-6/n-3 PUFA ratio (Ayaz et al., 2009; Vekiari et al., 2011). Furthermore, it
244 has been also found that the inclusion of carob pulp in a concentrate-based diet for lambs increased
245 its content of n-3 PUFA (Vasta et al., 2007). Overall, froma nutritional meat quality perspective, the
246 main findings of this study were related to an improvement of meat fatty acid composition
247 consequent to the dietary administration of carob pulp. Indeed, the lower levels of SFA and higher
248 levels of MUFA in muscle from carob-fed animals meet the recommendation of lowering the intake
249 SFA and increasing that of unsaturated fatty acids to decrease the risk of cardiovascular diseases

250 (Simopoulos, 2002). Also, nutritional guidelines recommend decreasing the PUFA n-6/n-3 ratio in
251 food, which should not exceed a threshold value of 4 (Simopoulos, 2002; UK Department of
252 Health, 1994). In our experiment we found that the inclusion of 15% of carob in the diet resulted in
253 a PUFA n-6/n-3 ratio of 4.44 compared to the meat from animals given the control diet in which the
254 ratio was above 13.
255
256 *3.2. Lipid oxidation and colour stability*
257 Oxidative phenomena are considered one of the main causes of deterioration of meat (Morrissey,
258 Sheehy, Galvin, Kerry, & Buckley, 1998) and are responsible for the production of off-flavours in
259 meat, due to the lipid oxidation, and for the discolouration of meat, due to the oxidation of
260 myoglobin. In the present study, lipid oxidation increased during the 9 days of storage period ($P < 0.001$; Table 4). Furthermore, an effect of the dietary treatment ($P < 0.05$) revealed that meat from
261 0.001; Table 4). Furthermore, an effect of the dietary treatment ($P < 0.05$) revealed that meat from
262 animals fed with the Carob 15% diet overall experienced a more pronounced lipid peroxidation
263 across time of storage compared to meat from animals in the other treatments. It was reported that
264 PUFA, and especially highly unsaturated PUFA, in the intramuscular fat is particularly susceptible
265 to the initiation and propagation of lipid oxidation (Morrissey et al., 1998;Wood et al., 2003).
266 Therefore, the concentration of the readily oxidisable PUFA in the intramuscular fat can provide
267 information about the effects of the diet on meat oxidative stability (Luciano et al., 2011, 2013).
268 From this standpoint, the highest percentage of highly unsaturated n-3 PUFA in meat from animals
269 fed with the Carob 15% diet, could have contributed to a higher susceptibility of muscle toward
270 lipid oxidation. A possible antioxidant effect of dietary phenolic compounds from carob in
271 improving meat resistance to lipid oxidation could have been expected. Nevertheless, the in vivo
272 antioxidant properties of dietary phenolic compounds against meat oxidative deterioration are still a
273 controversial issue (Vasta & Luciano, 2011). Carob contains high levels of phenolic compounds,
274 most of which are potentially strong antioxidants, but are in the form of nonextractable condensed
275 tannins (Silanikove et al., 2006). Research on the possible bioavailability of condensed tannins in

276 the animal organism seems to suggest that these compounds are poorly degraded into the
277 gastrointestinal tract and, therefore, not readily bioavailable (Abia & Fry, 2001; López-Andrés et
278 al., 2013).
279 However, it should also be stressed that the mean TBARS values found in the present studies over
280 the 9 days of storage were rather low and ranged from 0.07 to 0.52 mg MDA/kg of meat. These
281 values overall indicate that meat underwent low oxidative deterioration, as the threshold TBARS
282 value for the sensory detection of rancid flavours has been reported to be between 0.5 and 1 mg
283 MDA/kg of meat (Lanari, Schaefer, & Scheller, 1995; Rossi et al., 2013).
284 Meat colour is the main sensory attribute affecting consumer's decision to purchase, as the red
285 colour is associated with freshness (Morrissey et al., 1998). The oxidation of myoglobin and the
286 consequent accumulation of metmyoglobin are the primary factors explaining the changes in meat
287 colour coordinates in pork (Lindahl, Lundström, & Tornberg, 2001). In particular, the decrease in
288 meat redness (a*) value and the increase in hue angle (H*) value are used to describe meat colour
289 deterioration for their positive relationwith metmyoglobin concentration in meat. As expected, hue
290 angle (H*) and metmyoglobin values increased, while the redness (a*) and saturation (C*) values
291 decreased across the 9 days of storage, regardless of the dietary treatment. However, the dietary
292 treatment did not affect any of the above colour descriptors. No studies are available on the effect of
293 feeding pigs with diets supplementedwith carob onmeat colour stability. Regarding the effects of
294 polyphenol-rich diets on pork colour stability, O'Grady, Carpenter, Lynch, O'Brien, and Kerry
295 (2008) found no effect of including grape seed extract and bearberry in the diets of pigs on meat
296 colour stability descriptors. As discussed above for lipid oxidation, it should be observed that, in the
297 experimental conditions adopted in the present study, the colour stability parameters were subjected
298 to slight changes across storage duration. In the present study, lightness was the only colour
299 descriptor for which an effect, in tendency, of the dietary treatment was observed ($P = 0.09$). Meat
300 from animals in the Carob 15% group tended to have higher L* values compared to the control
301 treatment ($P = 0.07$), while the Carob 8% treatment resulted in higher L* values compared to the

control treatment (P = 0.03). The lack of significance of the diet × time interaction highlights that the effect of the dietary treatment observed in lightness was not dependent on the time of storage. It is not easy to propose a plausible explanation for this result, considering also that meat pH measured after 24 h post-mortem was not affected by the dietary treatment (average value: 5.48; data not shown). However this finding is in agreement with previous studies which demonstrated that feeding lambs with carob pulp increased meat lightness (Priolo et al., 2000).

Overall, it could be of interest to study the effect of dietary carob pulp on pork oxidative stability using different packaging conditions, such as modified atmosphere packaging, able to extend the monitoring period and to observe possible effects of the dietary treatment over an extended storage or display duration.

4. Conclusions

The results obtained in this investigation suggest that the inclusion of carob pulp into concentrate-based finishing diet for pigs could be an efficient and economical feeding strategy in the Mediterranean area. These results demonstrated that up to 15% of carob pulp can be included in the diet with no adverse effects on the growth performance. Furthermore, the diet in which carob pulp was included among the ingredients improved the nutritional value of pork by increasing the concentration ofmonounsaturated fatty acids and of n-3 polyunsaturated fatty acids,while lowering the percentage of saturated fatty acids. No effect of the dietary treatment was found on meat lipid oxidation and colour stability over aerobic refrigerated storage; therefore, a possible antioxidant effect of phenolic compounds present in carob pulp against meat oxidative deterioration was not observed.Nevertheless, considering that, in our experimental conditions, meat experienced slight oxidative processes, it would be of interest to investigate the possible antioxidant effect of dietary carob pulp on the oxidative stability of pork using other storage conditions, such as modified atmosphere packaging.

Conflict of interest

The authors declare that there are no conflicts of interest.

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Table 1
Ingredient and chemical composition of the experimental concentrates.

	Control	Carob 8%	Carob 15%
Ingredients (%)			
Corn	23.5	30	36
Barley	34.7	22.5	12.5
Soya bean meal	10	13	16
Faba bean	11	9	6.6
Wheat middlings	15	11.7	8
Carob pulp	0	8	15
Soybean oil	3	3	3
Premix[a]	2.8	2.8	2.9
Chemical composition			
Calculated digestible energy (MJ/kg)	13.43	13.18	13.10
Dry matter (DM)[b]	90.3	89.1	90.1
Ash[c]	9.9	7.2	5.5
Crude protein (CP)[c]	16.3	16.9	18.4
Neutral detergent fibre (NDF)[c]	26	20.9	19.4
Ether extract (EE)[c]	6.49	5.49	5.24
Total phenolic compounds[d]	2.76	2.90	3.16
Fatty acid composition (% of total fatty acids)			
C12:0	0.33	0.81	0.72
C14:0	0.75	0.52	0.42
C16:0	11.17	13.64	12.50
C16:1	0.38	0.75	0.53
C18:0	2.66	3.47	3.06
cis-9 C18:1	16.26	13.27	14.75
cis-9, *cis*-12 C18:2	41.58	35.21	34.18
cis-6, cis-9, cis-12 C18:3 n-6	1.44	0.55	0.50
cis-9, cis-12, cis-15 C18:3 n-3	25.45	31.77	33.33

[a] Included: calcium carbonate, sodium chloride, total phosphorus (dicalcium phosphate, calcium phosphate), vitamin premix, lysine, methionine, threonine.
[b] Expressed as g/100 g of fresh weight.
[c] Expressed as g/100 g of DM.
[d] Expressed as mg of tannic acid equivalents/g of DM.

Table 2
Animal weight and slaughter performances.

Item	Control	Carob 8%	Carob 15%	SEM	*P* value
No of pigs	9	9	9		
Final weight, kg	130.56	132.23	131.22	1.180	0.855
Carcass weight, kg	106.77	109.22	108.10	0.986	0.617
Dressing, %	81.77	82.60	82.41	0.355	0.627

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Table 3
Effect of the dietary treatments on the intramuscular fatty acid composition of LTL.

Item	Control	Carob 8%	Carob 15%	SEM	*P* value
No. of pigs	9	9	9	–	–
Intramuscular fat (mg/100 g of LTL)	2328	3283	2967	291	0.410
Individual fatty acids (g/100 g of total fatty acids)					
C12:0	0.10	0.06	0.05	0.010	0.084
C14:0	0.96^{a}	0.56^{b}	0.57^{b}	0.071	0.023
C16:0	27.34^{a}	20.42^{b}	18.50^{b}	1.030	< 0.001
cis-9 C16:1	3.24^{a}	1.91^{ab}	1.63^{b}	0.271	0.028
C17:0	0.48	0.41	0.36	0.028	0.234
C18:0	9.08	10.33	11.14	0.391	0.092
cis-9 C18:1	26.82^{b}	36.36^{a}	38.28^{a}	1.350	< 0.001
cis-9, *cis*-12 C18:2 n-6	21.49^{a}	18.12^{ab}	16.53^{b}	0.702	0.007
cis-9, *cis*-12, *cis*-15 C18:3 n-3	0.89^{c}	2.12^{b}	3.18^{a}	0.225	< 0.001
C20:2 n-6	0.24	0.37	0.51	0.055	0.147
C20:3 n-3 (ETA)	0.28	0.34	0.46	0.035	0.105
C20:4 n-6 (AA)	2.92	1.98	1.50	0.261	0.071
C20:5 n-3 (EPA)	0.41	0.26	0.20	0.063	0.376
C22:5 n-3 (DPA)	0.19	0.27	0.22	0.027	0.513
Classes of fatty acids (g/100 g of total fatty acids)					
SFA	37.96^{a}	31.78^{b}	30.61^{b}	1.050	0.005
MUFA	30.06^{b}	38.27^{a}	39.91^{a}	1.250	0.001
PUFA	26.44	23.46	22.60	0.694	0.054
n-6 PUFA	24.42^{a}	20.10^{b}	18.03^{b}	0.807	0.001
n-3 PUFA	1.78^{c}	2.99^{b}	4.06^{a}	0.248	< 0.001
PUFA/SFA	0.70	0.74	0.74	0.028	0.815
n-6/n-3	13.74^{a}	6.72^{b}	4.44^{b}	1.750	< 0.001

^{a,b}Mean values within a row with unlike superscript letters are significantly different ($P < 0.05$).

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ble 4

ect of the dietary treatment and time of refrigerated storage on meat colour stability and lipid oxidation during 9 days of refrigerated storage.

tem	Dietary treatment (diet)			Time of storage (time)			SEM	*P* value		
	Control	Carob 8%	Carob 15%	0	5	9		Diet	Time	Diet × time
TBARS (mg MDA/Kg of meat)	0.25^{b}	0.24^{b}	0.37^{a}	0.07^{z}	0.27^{y}	0.52^{x}	0.027	0.030	<0.001	0.113
C* values	52.50^{y}	54.62^{x}	54.30^{x}	50.60^{z}	54.08^{y}	56.83^{x}	0.446	0.090	<0.001	0.455
a* values	6.83	6.73	6.98	7.79^{x}	7.18^{x}	5.59^{y}	0.219	0.934	<0.001	0.429
b* values	7.88	7.68	8.12	7.85	8.30	7.53	0.260	0.877	0.342	0.448
C* values	10.52	10.29	10.75	11.08^{x}	11.05^{x}	9.43^{y}	0.318	0.907	0.012	0.441
H* values	48.64	47.37	49.09	45.19^{y}	48.36^{xy}	51.55^{x}	0.763	0.703	0.001	0.644
Metmyoglobin %	26.43	22.66	24.85	18.31^{y}	23.48^{xy}	32.15^{x}	1.590	0.696	0.001	0.980

Within row, different superscripts indicate differences between dietary treatments ($P < 0.05$) tested using the Tukey's adjustment for multiple comparisons.

Within row, different superscripts indicate differences between days of storage ($P < 0.05$) tested using the Tukey's adjustment for multiple comparisons.