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Ewe's Diet (Pasture vs Grain-Based Feed) Affects Volatile Profile of Cooked Meat from Light Lamb

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The effects of ewe's diet during gestation and lactation on the volatile compounds profile in cooked meat from light lamb were compared. Two lamb groups from ewes that had been fed pasture (PA) or grain-based concentrate (FE) were tested. Cooked loin mixed with saliva was analyzed by solid phase microextraction, gas chromatography, and mass spectrometry. The fiber coating used was divinylbenzene-carboxen-polydimethylsiloxane. The volatiles detected and quantified were aldehydes, alcohols, ketones, phenols, indole, and sulfur compounds. The ewe's diet strongly affected the volatile compounds profile of the cooked meat. The total volatiles concentration was higher in PA (409 mg kg^{-1}) than in FE (201 mg kg^{-1}). The major volatiles in PA were phenol, 4-methylphenol, and hexanoic acid, while the major volatile in FE was 3-hydroxy-2-butanone. No branched C8−C9 fatty acids responsible for mutton flavor were detected in either group. The findings suggest that nutritional strategies can be use during gestation and lactation to modify the aroma of light lamb meat in the light of consumer preferences.

KEYWORDS: Mutton; pastoral; feed; flavor; nutritional; gestation; lactation

INTRODUCTION

Aroma is the sensory attribute that best identifies cooked lamb meat, where it strongly contributes to its acceptance (*1*). The chemistry of the aroma of cooked meat is very complex and depends on the formation of volatile compounds detectable by olfactory receptors. Specific mutton aroma has been related with the presence of branched volatile fatty acid with 8 and 9 carbons, more specifically 4-methyloctanoic, 4-ethyloctanoic, and 4-methylnonanoic (*2*−*4*), formed by ruminal fermentation of carbohydrates. These fatty acids accumulate in the fat of adult sheep (*5*) and are quite stable and sufficiently volatile to be detected by olfactory receptors (*6*, *7*). Sheep meat is also characterized by high levels of volatile derivates of cysteine, whose principal source is in sheep's wool (*8*), while the composition of aldehydes, alcohols, ketones, phenols, indoles, and other volatile compounds depends both on the cooking method and on genetic and husbandry factors, especially diet.

The cooking method is crucial in the formation of volatile compounds of meat. Meat heating generates more than 1000 volatile compounds and precursors from the Maillard reaction and lipid degradation, with sulfur and carbonyl compounds predominating (*9*). These compounds may interact to form other secondary volatiles. The Maillard reaction intensifies at temperatures above 140 °C, takes place in aqueous solution, and is favored by meat dehydration. Roasting at 150−200 °C decreases the rate of alkane, alkene, and furan formation but increases the rate of formation of aldehydes, ketones, and pyrrols. Sulfur compounds, such as triazoles and pyridines, are more associated to roasted meat, while some thiols are more related to meat cooked at low temperature (70−100 °C) (*10*).

Lamb diet refers to the time of gestation, lactation, and fattening. During gestation and lactation, precursors or aromatic compounds are transferred from the ewe to the lamb (*11*). In the case of light lamb, slaughtered with a carcass weight of less than 13 kg, the fattening period is short, so that the mother's diet may have a special impact on the volatiles profile of cooked meat, which would explain the differences in aroma detectable by consumers. Basically there are two types of diet for sheep: those based on grain feeding and those based on traditional pasture, although both feeding methods can be combined. A grain-based feeding is used in intensive rearing, providing more carbohydrates and higher levels of oleic and linoleic acids. Grain diet increases the levels in meat of branched fatty acids, aldehydes, ketones, and lactones, which are derived from both acids, providing more intense mutton aroma, although with certain sweet notes (*12*, *13*). Occasionally, such diets are complemented with local agricultural wastes. Herbal diets, on the other hand, are characteristic of extensive rearing practices in which the sheep graze. Such diets provide more proteins and more linolenic acid and increase the levels of phenols, indoles, and terpenes in the meat, along with aldehydes and ketones, which are derived from the linolenic acid (*14*−*17*). The so-called "pastoral" flavor (*18*) is characterized by descriptors such as "grassy", "animal", "faecal", "sheep", and "milky" (*14*, *19*) and may be disliked by some consumers (*20*).

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Studies consulted on volatiles in cooked sheep meat (*21*, *22*) mention heavy lambs and adult animals, which give a fatty meat with an intense flavor of mutton. However, there are no data available on the volatiles of light lamb as traditionally consumed in the Mediterranean regions of Europe. The aim of this study was to determine whether the diet of ewes during gestation and lactation affects the volatiles profile of cooked light lamb. All the lambs were fattened with a feed concentrate to delimit the effect of ewe diet on the volatiles of the meat. Ascertaining of the volatiles of cooked lamb was a preliminary step to determining whether it is possible to modulate the aroma of the cooked meat of light lamb through the feeding of ewes.

MATERIALS AND METHODS

Reagents. Pure reference standards for pentanal, 4-methyl-2-pentanone, hexanal, 1-penten-3-ol, heptanal, 1-pentanol, octanal, 3-hydroxy-2-butanone, (*E*)-2-heptenal, 1-hexanol, nonanal, 1-octen-3-ol, 1-heptanol, acetic acid, 2-ethyl-1-hexanol, benzaldehyde, (*E*)-2-nonenal, propanoic acid, 1-octanol, (*E*)-2-octen-1-ol, (*E*)-2-decenal, nonanol, (*E*,*E*)-2,4-decadienal, hexanoic acid, phenol, octanoic acid, 4-methylphenol, nonanoic acid, piperonal, 1-butanol, dimethyl-sulphone, and 1-dodecanol were supplied by Acros Organics (Geel, Belgium). (*E*)-2-Octenal, (*E*)-2-undecenal, indol, 4-methyl-octanoic acid, 4-methyl-nonanoic acid, and 4-ethyl-octanoic acid were supplied by Sigma-Aldrich (St. Louis, MO).

Human Saliva. Saliva from four panellists (two male and two female, nonsmoking, and 30−50 years old) were collected. Two hours before collection, the panellists would clean teeth without using dental toothpaste or mouthwash, and they could not eat any food. At the time of collection, participants rinsed their mouths with cold water, repeated twice, and 3 mL of saliva were collected from each panellist. Saliva from all panellists was mixed and homogenized into a glass bottle and then kept in a water bath at 37 °C until analysis of lamb samples (for 0−4 h). The same panellists were used for all sessions of saliva collection.

Animals and Diets. Two groups of six light lambs (Segureña sheep breed) from two exploitations in SE Spain were denominated PA (pasture) and FE (grain-based feeding). The PA ewes and lambs were raised semiextensively, the mothers grazing during the autumn and winter in pastures of the Segura Mountains, where the dominant vegetation comprises several trees as *Pinus halepensis, Pinus nigra*, or *Quercus ilex*, brushes of *Rosmarinus officinalis, Juniperus oxycedrus, Juniperus thurifera, Juniperus phoenicea, Quercus coccifera, Rhamnus lycioides*, and *Thymus vulgaris*, and meadows of gramineae containing *Helictotrichon filifolium, Carex halleriana, Festuca capillifolia, Stipa tenacísima, Pipthaterum miliaceum*, and *Avenula murcica*. The FE ewes and lambs were intensively raised, and the ewes were feed with grain-based concentrate, complemented with barley, maize, orange peel, and straw.

The lambs (both PA and FE) were fed ewe milk during the first three weeks of life before transferring to a mixed feeding regime based on ewe milk and a growth feed provided ad libitum until weaning at 40 days of age. Both groups of lambs were then given a concentrated fattening feed provided ad libitum until they reached 60 days of age and 25 kg live weight. The raw materials used to feed the sheep and lambs are listed in **Table 1**.

Sampling. The lambs were slaughtered in a local abattoir in accordance with EU regulations. Mean carcass weight was 12 kg. The carcasses were aged at 2 °C for 24 h before professional butchers extracted the loin (Longissimus dorsi-lumborum muscle), which was deboned, vacuum packed, and frozen at −18 °C. For the analyses, the loins were brought to −5 °C and cut with a slicing machine (V220/380, Mobba, Badalona, Spain), providing 1.5 cm thick rounds of variable diameter, according to the muscle section. Once defrosted, the meat was cooked on a double electric hot plate (Media Liscia, Silanos, Milan, Italy) at 150 °C until reaching an internal temperature of 72 °C for 2 min, as controlled by an SA880 SSX digital thermometer (Oregon Scientific, Alcobendas, Spain). The surface temperature of meat did not reach 100 °C. Cooking was made at low temperature because dietary changes of volatile compounds maybe are more detectable in the cooked meat where thermal degradation is low. After cooking, the samples were cooled at room temperature (22 °C), remaining wrapped in aluminum foil to minimize volatile loss. They were then vacuum packed and immediately frozen at −80 °C to prevent any degradation before chromatographic analysis. The frozen samples were stored for up 30 days before analysis.

Table 1. Vegetables Used for Ewe and Lamb Feeding[a]

	g 100 g^{-1}
Grass (PA): *Helictotrichon filifolium, Carex halleriana, Festuca capillifolia, Stipa tenacisima, Pipthaterum miliaceum,* and *Avenula murcica*	
Brushes (PA): *Rosmarinus officinalis, Juniperus oxycedrus, Juniperus thurifera, Juniperus phoenicea, Quercus coccifera, Rhamnus lycioides*, and *Thymus sp.*	
Feed for Ewes (FE)	
wheat bran	25.2
sunflower seed extraction flour	14.6
soybean hulls	11.8
carob flour	3.3
rice cylinder	0.3
sugar cane molasses	2.6
barley	3.9
corn	3.9
orange peel	3.9
straw	21.1
Growing Feed for Lambs (PA and FE)	
barley	37.5
toasted soya extraction flour	20.6
wheat	22.4
cake corn germ extraction	5.3
corn gluten feed	4.5
distilled from wheat	4.4
corn gluten	3.4
crude palm oil	2.3
soybean hulls	1.9
calcium salts of fatty acids	0.5
Fattening Feed for Lambs (PA and FE)	
barley	13.8
cake corn germ extraction	5.3
corn gluten feed	4.5
distilled from wheat	4.4
corn gluten	3.4
crude palm oil	2.3
soybean hulls	1.2

[a] Mineral supplements (calcium carbonate and mine salt) are not included.

Solid-Phase Microextraction. To simulate the release of the cooked lamb meat volatile compounds into the human mouth, samples of ground meat (1 g) together with human saliva (1.5 mL) were transferred into a 20 mL hermetic sealed vial. The mixture was homogenized in an ultrasonic bath (J.P. Selecta, SA, Barcelona, Spain) at 37 °C for 5 min. After this, and in order to achieve equilibrium between each sample and its headspace before SPME analysis, vials were kept in a water bath at 37 °C for 15 min.

The headspace volatile constituents were isolated using an SPME device (Supelco Inc., Bellefonte, PA) containing a fused-silica fiber coated with a (50/30 μm thick, coating phase), 2 cm length layer of DVB-CAR-PDMS (divinylbenzene-carboxen-polydimethylsiloxane). The fiber was exposed above the sample headspace for 45 min (time needed to achieve the equilibrium among the three phases that conform the system). This equilibrium time was calculated according to Steffen and Pawliszyn (*23*). For this determination four analytical standards, characteristic of the cooked lamb meat aroma, and at the same time representatives for the polarity and volatility of the overall group of components studied, were chosen. As shown in **Figure 1**, the equilibrium time is identified as the point at which the adsorption of the components on the fiber coating reached a plateau.

During this exposure time, the samples were held in a water bath at 37 °C. After 45 min of extraction time, the SPME device was removed from the meat sample vial and inserted directly into the injection port of the GC-MS. Before the extraction, the fiber was conditioned by heating it in the gas chromatograph injection port at 270 °C for 1 h. All the HS-SPME sample analysis were prepared and analyzed in triplicate.

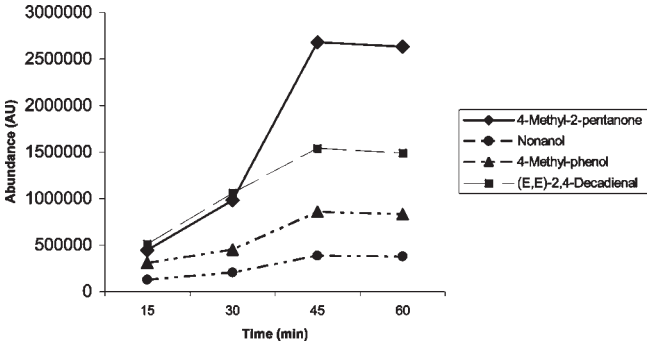

Figure 1. Determination of the equilibrium time for divinylbenzene-carboxen-polydimethylsiloxane fiber coating (area units vs minutes).

Gas Chromatography−Mass Spectrometry of SPME Samples. Once the analytes were adsorbed onto the coating, they were subjected to analysis by capillary gas chromatography−mass spectrometry as follows. An Agilent Technologies 6890 N gas chromatograph (GC) (Palo Alto, CA) equipped with a 30 m × 0.25 mm HP-5 (cross-linked phenyl-methyl siloxane) column with 0.25 μm film thickness, and a DB-Wax 52 CB (polyethyleneglycol-Carbowax) 30 m × 0.32 mm i.d. with 1.0 μm film thickness was used. Both stationary phases were supplied by Agilent Technologies (Palo Alto, CA). Helium was used as the carrier gas (constant pressure, β-ionone eluting at 36.62 min for HP-5MS column and 47.02 min for DB-Wax column). The analytes were desorbed at 250 °C for 10 min in the injection port of the GC. To improve the recovery of the highly volatile components, a splitless injection was applied during the first 0.6 min of the injection. After this time, the split ratio was set to 50:1. The GC was linked to an Agilent model 5972 inert mass spectrometry detector. For both stationary phases, the initial oven temperature was set at 40 °C, then increased at 2.5 °C/min to 150 °C, and finally raised to 250 °C at a rate of 10 °C/min; the transfer line to the mass selective detector was kept at 280 °C. The mass spectrometer was operated in electron impact ionization mode with an ionizing energy of 70 eV, scanning from m/z 50 to 500 at 3.21 scan/s. The quadrupole and ion source temperatures were set at 150 and 230 °C, respectively. The electron multiplier voltage was maintained at 1300 V. Analysis was performed with selected ion monitoring mode (SIM) considering target ion (T) and two qualifiers (Q1 and Q2).

The individual peaks were identified by retention times and retention indices (relative to C6−C17 *n*-alkanes), compared with those of known compounds, and by comparison of mass spectra using the NBS75K library (U.S. National Bureau of Standards, 2002) and spectra obtained from the standard. **Table 2** shows the compounds along with their retention index, and the target and qualifier ions considered for their positive identifications.

Quantification. For the purpose of quantifying the isolated components, linear regression models were determined using standard dilution techniques for all the compounds identified. Samples of meat from the lambs under study were used as matrix for the standard dilution analysis. Whereas both the matrix as the qualitative and quantitative differences detected among the two lamb meat volatile profiles can affect the behavior of the volatile components, two different lineal regression equations, one for each type of lamb under study, were calculated (**Table 3**). Calibration curves were determined by applying the same conditions to the standard dilutions as to the analytical samples.

Statistical Analysis. Data were analyzed using Statistix for Windows 2.0 program (Analytical Software, Tallahassee FL). An analysis of variance was used to investigate the effect of the ewe diet on the dependent variables. The least-squares means (LSM) and the significance of the treatment was calculated using type IV sum of squares. The Scheffe Means Test was used to compare the LSM, which were considered to be statistically different when $P < 0.05$.

RESULTS AND DISCUSSION

Table 4 shows the effect of the ewe diet on the volatile compounds of the resulting cooked light lamb meat. The volatile compounds detected and quantified in the headspace were aldehydes, alcohols, acids, ketones, phenolic, indole, and sulfur compounds.

Table 2. Kovats Index (KI), Primary Ions (T), and Secondary Ions (Q1 and Q2) Used for Identifying Volatile Compounds

compd	KI[a]	KI[b]	T	Q1	Q2
pentanal	960	780	58	44	86
4-methyl-2-pentanone	985	792	58	43	100
hexanal	1075	816	72	56	82
1-butanol	1166	774	56	57	43
1-penten-3-ol	1173	777	57	41	85
heptanal	1207	886	86	70	44
1-pentanol	1284	802	55	42	70
octanal	1328	991	100	110	84
3-hydroxy-2-butanone	1329	783	88	73	45
(*E*)-2-heptenal	1361	939	83	55	70
1-hexanol	1392	861	69	56	84
nonanal	1427	1112	98	114	124
(*E*)-2-octenal	1457	1056	83	97	108
1-octen-3-ol	1476	966	72	57	85
1-heptanol	1479	957	83	70	56
acetic acid	1486	768	60	45	43
2-ethyl-1-hexanol	1507	1024	70	57	98
benzaldehyde	1535	941	77	106	105
(*E*)-2-nonenal	1543	1176	83	70	96
propanoic acid	1556	791	57	74	55
1-octanol	1562	1079	70	56	84
(*E*)-2-octen-1-ol	1609	1072	68	57	81
(*E*)-2-decenal	1634	1283	70	83	98
nonanol	1651	1193	69	57	87
E-2-undecenal	1744	1381	97	83	111
(*E*,*E*) 2,4-decadienal	1814	1336	81	67	95
hexanoic acid	1876	1024	60	73	87
dimethyl sulphone	1933	904	79	94	81
1-dodecanol	1986	1501	97	111	125
phenol	2025	979	94	66	63
octanoic acid	2062	1230	60	73	85
4-methyl-phenol	2080	1088	107	77	90
nonanoic acid	2131	1322	60	73	98
indole	2313	1319	117	90	63
piperonal	2361	1325	149	121	63

[a] DB-WAX column. [b] HP-5 column.

As can be seen, the ewe diet strongly affected the volatile profile of the cooked meat. The mean concentration of total volatile compounds detected by SPME-GC-MS was considerably higher in PA (409 mg kg^{-1}) than in FE (201 mg kg^{-1}) meat. Significant differences in mean values were observed in 23 of the 35 compounds detected and quantified, although high standard deviations were found. Meat is a variable material, especially fat. Factors strongly affecting the formation of volatiles in cooked meat, such as fat composition or intramuscular fat, may vary with each individual, even in lambs reared together under the same conditions (*24*). High standard deviations were also recorded in other similar studies (*21*, *25*, *26*).

The total quantity of aldehydes was higher in PA than in FE, a result that was inconsistent with previous studies. The aliphatic aldehydes, including hexanal, nonenal, and decenal, which are normally predominant in the aroma of cooked meat, were higher in PA. Grain-based diets, richer in oleic and linoleic acids, increase the levels of certain aldehydes, such as hexanal, 2-heptenal, and 2−4-decadienal (*12*, *13*). Compared with grain-based diets, grasses are richer in linolenic acid, the precursor of various unsaturated aldehydes, e.g. 4-hepatanal, 2,4-heptadienal, and 2,6-nonadienal (*27*). The supplementation of pasture diet with barley and soya increased unsaturated aliphatic aldehydes and ketones but did not affect the saturated aliphatic aldehydes of cooked (70 °C) mutton, analyzed by SPME-GC-MS (*28*). Piperonal had not been reported in cooked lamb. Piperonyl alcohol and other

Table 3. Linear Regression Equations Used for Quantifying Volatile Compounds in Cooked Lamb: Quantification Factors (QF) and Regression Coefficients (R^2)

	PA		FE	
compd	QF	R^2	QF	R^2
pentanal	1.10^{-6}	0.928	1.10^{-6}	0.928
4-methyl-2-pentanone	3.10^{-7}	0.903	3.10^{-7}	0.903
hexanal	5.10^{-7}	0.955	5.10^{-7}	0.955
1-butanol			5.10^{-7}	0.835
1-penten-3-ol	1.10^{-7}	0.831	1.10^{-7}	0.831
heptanal	5.10^{-7}	0.938	$1.1.10^{-6}$	0.878
1-pentanol	1.10^{-7}	0.995	1.10^{-7}	0.995
3-hydroxy-2-butanone			$8.7.10^{-5}$	0.973
octanal	1.10^{-6}	0.965		
(*E*)-2-heptenal	$4.2.10^{-6}$	0.933	$4.2.10^{-6}$	0.933
1-hexanol	1.10^{-7}	0.951	1.10^{-7}	0.951
nonanal	$2.5.10^{-7}$	0.975	3.10^{-7}	0.947
(*E*)-2-octenal	5.10^{-7}	0.945	3.10^{-7}	0.962
1-octen-3-ol	1.10^{-7}	0.990	1.10^{-7}	0.990
1-heptanol	3.10^{-7}	0.941	5.10^{-7}	0.903
acetic acid	4.10^{-6}	0.920	$2.7.10^{-6}$	0.929
2-ethyl-1-hexanol	$2.5.10^{-7}$	0.977	$2.5.10^{-7}$	0.977
benzaldehyde	2.10^{-8}	0.868	2.10^{-8}	0.868
(*E*)-2-nonenal	$2.6.10^{-5}$	0.910	5.10^{-6}	0.956
propanoic acid	$2.8.10^{-5}$	0.820	$2.8.10^{-5}$	0.820
1-octanol	2.10^{-7}	0.917	2.10^{-7}	0.917
(*E*)-2-octen-1-ol	1.10^{-6}	0.986	1.10^{-6}	0.986
(*E*)-2-decenal	$5.1.10^{-5}$	0.925		
nonanol	3.10^{-7}	0.965	$1.6.10^{-8}$	0.862
(*E*)-2-undecenal	$2.9.10^{-5}$	0.992		
(*E*,*E*) 2,4-decadienal	$3.5.10^{-6}$	0.838	$4.7.10^{-6}$	0.804
hexanoic acid	$3.7.10^{-5}$	0.978	$3.7.10^{-5}$	0.978
dimethyl-sulphone			$1.3.10^{-4}$	0.951
1-dodecanol			1.10^{-6}	0.981
phenol	$3.1.10^{-6}$	0.912	$2.5.10^{-7}$	0.904
octanoic acid	$4.1.10^{-6}$	0.951	$3.3.10^{-5}$	0.943
4-methyl-phenol	1.10^{-4}	0.941	$2.5.10^{-7}$	0.876
nonanoic acid	$1.6.10^{-5}$	0.912	$1.6.10^{-5}$	0.912
indol	$1.6.10^{-7}$	0.932	$1.6.10^{-7}$	0.932
piperonal	2.10^{-8}	0.868	2.10^{-8}	0.868

piperonyl compounds are used as insecticide for rice, wheat, fruit tree, vegetables, etc. Piperonyl compounds have been detected in milk, blood, and several animal tissues from small ruminants (*29*).

Compared with the aldehydes, the total quantity of alcohols detected was low. For example, 1-dodecanol and 1-butanol were only detected in FE. 1-Butanol is one of the volatile alcohols in fat that permit herbal diets and grain-based diets to be distinguished in sheep meat (*27*). In contrast, the volatile fatty acid concentration found in PA (76 mg kg^{-1}) and FE (58 mg kg^{-1}) was quite high. The mean hexanoic and nonanoic acid content was higher in PA, while FE showed higher levels of octanoic and propanoic acids. A grain-based diet increases the deposition of 8- and 10-carbon fatty acids, which might have favored their transmission to the lamb (*14*, *21*). However, none of the branched fatty acids characteristic of ovine fat, such as methyl-octanoic, ethyl-octanoic, and methyl-nonanoic acids, was detected, despite their similar molecular size and volatility to those which were detected. Concentrations of 0.7, 8, and 10 mg kg^{-1} have been described in the raw fat of heavy lambs, rams, and ewes, respectively (*5*). Cooking hardly affected the concentration of 4-ethyloctanoic acid of sheep meat extracts (*6*). Regardless of the ewe diet, the release of branched volatile C8−C9 fatty acids responsible for the typical aroma of mutton would be irrelevant in light lamb meat.

FE meat showed a high concentration of 3-hydroxy-2-butanone, a ketone with an odor described as lardy, sweet, caramel, or creamy. This ketone is an intermediate in the butanodiol cycle of glucose fermentation (*30*) and is found in some vegetable used to feed animals (*31*). Therefore, the FE ewe diet based on cereals, which are rich in glucose, complemented with sugar cane molasses rich in sucrose and glucose (sugars easily fermentable by digestive bacteria), seems to have favored the transmission of 3-hydroxy-2-butanone to the lamb. There was probably an intense fermentation of glucose during ewe digestion, and excessive absorbed 3-hydroxy-2-butanone did not metabolize in other compounds. Therefore, there was significant transmission of 2-hidroxy-3-butanone from mother to lamb through the blood (gestation) and the milk (lactation). In contrast, 3-hydroxy-2-butanone was not detected in PA, even though the PA diet probably contained grain from wild grass species. This indicates that the fermentation of glucose was irrelevant to produce effective transmission of 3-hydroxy-2-butanone in PA. In addition, both groups of lambs (PA and FE) were fattened after weaning using the same feed

Table 4. Effects of Ewe Diet (Pasture vs Grain-Based Feeding) on Cooked Light Lamb Meat Volatile Profile (Expressed as mg kg^{-1})

	PA diet[a]	FE diet		
compd[b]	M ± SD	M ± SD	F	P[c]
aldehydes	*45.6 ± 39.7*	*3.51 ± 1.34*	*6.72*	*
pentanal	3.72 ± 3.21	0.22 ± 0.20	7.14	*
hexanal	14.1 ± 12.1	1.15 ± 0.46	6.79	*
heptanal	0.51 ± 0.58	0.03 ± 0.05	4.07	NS
octanal	0.50 ± 0.54	nd	5.11	*
(*E*)-2-heptenal	2.24 ± 1.22	0.56 ± 0.31	10.53	**
nonanal	1.00 ± 0.65	0.29 ± 0.19	6.61	*
(*E*)-2-octenal	0.03 ± 0.02	0.04 ± 0.03	0.03	NS
benzaldehyde	0.02 ± 0.00	0.01 ± 0.00	16.94	**
(*E*)-2-nonenal	10.2 ± 9.93	0.19 ± 0.22	6.04	*
(*E*)-2-decenal	9.19 ± 11.1	n.d.	4.11	NS
(*E*)-2-undecenal	1.32 ± 1.32	n.d.	5.94	*
(*E*,*E*) 2.4-decadienal	2.73 ± 2.44	0.86 ± 0.34	3.44	NS
piperonal	0.08 ± 0.04	0.15 ± 0.07	4.77	NS
alcohols	*6.34 ± 4.70*	*3.61 ± 1.17*	*1.91*	*NS*
1-butanol	nd	0.21 ± 0.11	20.37	***
1-penten-3-ol	0.19 ± 0.09	0.10 ± 0.04	3.94	NS
1-pentanol	1.13 ± 1.00	0.76 ± 0.22	0.79	NS
1-hexanol	2.72 ± 2.48	1.19 ± 0.48	2.19	NS
1-octen-3-ol	0.75 ± 0.46	0.27 ± 0.11	6.00	*
1-heptanol	0.08 ± 0.06	0.04 ± 0.02	3.18	NS
2-ethyl-1-hexanol	0.24 ± 0.12	0.29 ± 0.17	0.36	NS
1-octanol	0.50 ± 0.40	0.21 ± 0.07	3.11	NS
(*E*)-2-octen-1-ol	0.49 ± 0.26	0.10 ± 0.04	12.70	**
nonanol	0.21 ± 0.17	0.01 ± 0.00	8.90	**
1-dodecanol	nd	0.39 ± 0.26	13.49	**
ketones	*0.50 ± 0.14*	*78.1 ± 26.7*	*36.16*	***
4-methyl-2-pentanone	0.50 ± 0.14	0.27 ± 0.12	8.80	**
3-hydroxy-2-butanone	nd	77.8 ± 26.8	50.47	***
acids	*76.1 ± 22.8*	*57.8 ± 40.0*	*0.94*	*NS*
acetic	17.0 ± 5.60	16.16 ± 7.05	0.05	NS
propanoic	15.2 ± 4.46	21.0 ± 24.5	0.33	NS
hexanoic	37.8 ± 17.7	13.98 ± 6.79	9.47	**
octanoic	0.96 ± 0.30	4.02 ± 1.97	14.14	**
nonanoic	5.15 ± 2.28	2.66 ± 1.14	5.66	*
phenols	*279.0 ± 66.0*	*31.0 ± 9.16*	*86.16*	***
phenol	209.7 ± 72.7	26.0 ± 7.13	37.90	***
4-methyl-phenol	67.7 ± 22.7	0.21 ± 0.10	53.15	***
indole	1.68 ± 1.15	4.80 ± 3.06	5.44	*
dimethyl-sulphone	n.d.	21.9 ± 20.5	6.83	*

[a] PA: pasture; FE grain-based feed. [b] M: mean; SD: standard deviation; F: ANOVA F-Statistic; P: probability values. [c] Level of significance: *** $P \leq$ <0.001; ** $P \leq 0.01$; * $P \leq 0.05$; NS $P > 0.05$. nd: not detected.

(see **Table 1**), including cereals, but their 3-hydroxy-2-butanone contents were different. Digestive fermentations in light lambs are less intense than that in adult ruminants, such as pregnant ewes. Experimental grain-based diets provided detectable levels of 2-hydroxy-3-butanone in grilled lamb analyzed by Tenax TA-GC-MS (*21*).

On the other hand, PA presented a higher phenolic compounds content than FE. Phenol and 4-methylphenol showed mean concentration of 210 and 68 mg kg^{-1} in PA, compared with the 26 and 0.2 mg kg^{-1} recorded in FE. The herbal diet consumed by PA ewes clearly increased the level of volatile phenols in the cooked lamb. A pastoral flavor has been related with the deposition in fat of methylphenols, isopropylphenols, and other phenolic compounds (*5*). A wide variety of alkyphenols are present in plants, and their presence in meat is related with the ruminant fermentation of lignins and the ingestion of diets rich in diterpenes and tyrosine. Two other phenolic compounds present in forage are coumaric and ferulic acids. The transmission of phenolic compounds from mother to lamb during gestation and lactation has been demonstrated (*11*). As the quantity of dietary polyphenols increases in pregnant ewes, so the polyphenol concentration increases in the subsequent lamb muscle (*32*). This suggests that dietary phenolic precursors, as lignin and others, would be transformed by digestive fermentations, metabolized by pregnant ewe, and finally, phenolic volatiles or their precursors would be transmitted and eventually accumulated in the lamb meat. As raw lamb contains more phenolic precursors, there is greater possibility that volatile phenols will be detected in the cooked meat by SPME. No phenol or methyl-phenol were found in grilled meat from lamb fed on grain-based diets (*21*).

Low levels of the indole compounds associated with pastoral flavor were found. Skatole was not detected, while indole concentrations were lower than 5 mg kg^{-1}. Indole compounds are generated by the digestive fermentation of proteins through microbial decarboxylation and the desamination of tryptophan. However, its presence in fat as well may be due to the absorption of faecal gases by animals raised indoors (*33*). This would perhaps explain why the concentration of indole was slightly higher in FE than in PA. FE meat showed a considerable concentration of dimethyl-sulphone, a compound used as a source of sulfur in animal feed supplements. There were no volatile sulfur compounds, such as 4,6-dimethyl-oxatiane, 3,5-dimethyl-1,2,4-trithiolane, or 2,4,6-dimethyl perhydro-1,3,5-dithiazine (*8*), or volatile nitrogen compounds such as 2-penthyl pyridine, which have been reported in sheep meat by other authors (*4*).

The volatile profile found in the cooked light lamb differed substantially from those mentioned in the literature. The feeding method and raw meat composition would have been responsible for these differences. Diet, digestive activity, and nutritional absorption are different in young lamb that do not graze during the fattening period than in suckling lamb, heavy lamb, or mutton. No ewe feeding studies in connection whith the volatile compounds of cooked lamb are available in the consulted literature. Most of previous studies tested pasture vs concentrate diets in heavy lamb or mutton in the fattening stage. It was reported that heated sheep fat (subcutaneous and perirenal) volatiles are affected by diet (*12*, *22*, *27*, *34*), but few studies have analyzed cooked lamb meat. The dietary manipulation of fatty acids modified the volatile profile of grilled meat from heavy lambs (*21*). In another experiment, the supplementation of a pasture diet with barley and soya modified the amount of aliphatic aldehydes and ketones in the cooked mutton (*28*). On the other hand, hexanal, phenol, dimethyl-sulphone, or 2-hydroxy-3-butanone were not reported in cooked (70 °C) meat of suckling lamb fed on maternal milk, analyzed by steam-distillation-extraction "SDE" GC-MS (*25*).

Methodological differences, such as the cooking procedure, the addition of saliva, and the volatile extraction method, may have contributed to the differences observed in the results with respect to other research. The formation of volatile from Maillard compounds needs roast or grill procedures (*10*). The addition of saliva to reproduce the real conditions in which flavor is developed, which may affect the release of volatiles from the cooked meat to the headspace (*35*). Substantial variations as a function of the extraction method (dynamic headspace entrainment on Tenax TA, SDE, or SPME) were reported in the volatile compounds of grilled goat meat (*26*). The SPME fiber type also affects the determination of volatiled in meat. For example, a divinylbenzene-carboxen-polydimethylsiloxane fiber presented better reproducibility and extracted the volatile compounds of cooked beef more efficiently, than a carboxen-polydimethylsiloxane fiber (*36*). Other factors may also affect the detection and quantification of volatiles, such as their concentration in the headspace, their chemical affinity and competition with other volatiles for the fiber, functional group, presence of aromatic radicals, or steric properties. Interactions between different groups of carbonyl volatiles in the polymeric coating of the fiber might help explain the different levels recorded in the PA (predomination of aldehydes) and FE (predomination of ketones) cooked lamb.

Irrespective of the experimental conditions, the literature indicates that diet clearly affects volatile formation in cooked sheep meat. Thus, the diet of ewes during gestation and lactation seems to play an important part in the volatile profile of light lamb and, consequently, in its aroma. Therefore, nutritional strategies may be used to modulate the aroma of lamb meat as a function of consumer preference. Future studies will be necessary to throw more light on the sensory contribution of the volatile compounds present in cooked light lamb.

LITERATURE CITED

(1) Young, O. A.; Braggins, T. J. Sheepmeat odour and flavour. In *Flavour of Meat, Meat Products and Seafoods* 2nd ed.; Blackie Academic & Professional Eds.: London, 2004; Vol. 1, pp 102−123.

(2) Hornstein, I.; Crowe, P. F. Meat flavor: lamb. *J. Agric. Food Chem.* **1963**, *11*, 147–149.

(3) Brennand, C. P.; Lindsay, R. C. Distribution of volatile branched-chain fatty acids in various lamb tissues. *Meat Sci.* **1992**, *31*, 411–421.

(4) Sutherland, M. M.; Ames, J. M. Free fatty acids composition of the adipose tissue of intact and castrated lambs slaughtered at 12 and 30 weeks of age. *J. Agric. Food Chem.* **1996**, *44*, 3113–3116.

(5) Ha, J. K.; Lindsay, R. C. Distribution of volatile branched-chain fatty acids in perinephric fats of various red meat species. *Lebensm. Wiss. Technol.* **1990**, *23*, 433–440.

(6) Rota, V.; Schieberle, P. Changes in key odorants of sheep meat induced by cooking. In *ACS Symposium Series*; American Chemical Society; Washington, DC, 2006; Vol. 920, pp 73−83.

(7) Brennand, C. P.; Ha, J. K.; Lindsay, R. C. Aroma properties and thresholds of some branched-chain and other minor volatile fatty acids occurring in milkfat and meat lipids. *J. Sensory Stud.* **1989**, *4*, 105–120.

(8) Cramer, D. A. Chemical compounds implicated in lamb flavour. *Food Technol.* **1983**, *37* (5), 249–257.

(9) Pegg, R. B.; Shahidi, F.. Heat effects on meat. Flavour development. In *Eycyclopedia of Meat Sciences*, 1st ed.; Academic Press: Oxford, 2004; Vol. 1, pp 570−578.

(10) MacLeod, G.; Seyyedain-Ardebeli, M. Natural and simulated meat flavour (with particular reference to beef). *CRC Crit. Rev. Food Sci. Nutr.* **1986**, *14*, 309–437.

(11) Fresno-Bermejo, L. Administración de etomidato-lipuro en ovejas gestantes a término. Estudio del paso placentario y efectos cardiovasculares, respiratorios y del equilibrio ácido-base, en madres y sus fetos. Ph.D. Thesis, Universitat Autónoma de Barcelona, Barcelona, Spain, 2007.

(12) Sebastián, I.; Viallon, C.; Berge, P.; Dransfield, E.; Berdagué, J. L. Analysis of the volatile fraction and the flavour characteristics of lamb: relationships with the type of feeding. *Sci. Aliments* **2003**, *23*, 497–511.

(13) Suzuky, J.; Bailey, M. E. Direct sampling capilary GLC analysis of flavor volatiles from ovine fat. *J. Agric. Food. Chem.* **1985**, *33* (3), 343–347.

(14) Young, O. A.; Lane, G. A.; Priolo, A.; Fraser, K. Pastoral and species flavour in lambs raised on pasture, lucerne or maize. *J. Sci. Food Agric.* **2003**, *83*, 93–104.

(15) Rousset-Akrim, S.; Young, O. A.; Berdagué, J. L. Diet and growth effects in panel assessment of sheepmeat odour and flavour. *Meat Sci.* **1997**, *45*, 169–181.

(16) Prache, S.; Cornu, A.; Berdagué, J. L.; Priolo, A. Traceability of animal feeding diet in the meat and milk of small ruminants. *Small Ruminant Res.* **2005**, *59*, 157–168.

(17) Ha, J. K.; Lindsay, R. C. Volatile alkylphenols and thiophenol in species-related characterizing flavors of red meats. *J. Food Sci.* **1991**, *56*, 1197–1202.

(18) Schreurs, N. M.; Lane, G. A.; Tavendale, M. H.; Barry, T. N.; McNabb, W. C. Pastoral flavour in meat products from ruminants fed fresh forages and its amelioration by forage condensed tannins. *Anim. Feed Sci. Technol.* **2008**, *146*, 193–221.

(19) Priolo, A.; Micol, D.; Agabriel, J. Effects of grass feeding systems on ruminant meat colour and flavour: a review. *Anim. Res.* **2001**, *50*, 185–200.

(20) Prescott, J.; Young, O.; O'Neil, L. The impact of variations in flavour compounds on meat acceptability: a comparation of Japanese and New Zealand consumers. *Food Qual. Preference* **2001**, *12*, 257–264.

(21) Elmore, J. S.; Sarah, L. C.; Enser, M.; Mottram, D. S.; Sinclair, L. A.; Wilkinson, R. G.; Wood, J. D. Dietary manipulation of fatty acid composition in lamb meat and its effect on the volatile aroma compounds of grilled lamb. *Meat Sci.* **2005**, *69*, 233–242.

(22) Young, O. A.; Berdagué, J. L.; Viallon, C.; Rousset-Akrim, S.; Theriez, M. Fat-borne volatiles and sheep meat odour. *Meat Sci.* **1997**, *45*, 169–181.

(23) Steffen, A.; Pawliszyn, J. Analysis of flavour volatiles using headspace solid-phase microextraction. *J. Agric. Food Chem.* **1996**, *44*, 2187–2193.

(24) Nieto, G.; Díaz, P.; Bañón, S.; Garrido, M. D. Dietary administration of ewe diets with a distillate from rosemary leaves (*Rosmarinus officinalis L.*): influence on lamb meat quality. *Meat Sci.* **2010**, *84*, 23–29.

(25) Osorio, M. T.; Zumalacárregui, J. M.; Cabeza, E. A.; Figueira, A.; Mateo, J. Effect of rearing system on some quality traits and volatile compounds of suckling lamb meat. *Small Ruminant Res.* **2008**, *78*, 1–12.

(26) Madruga, M. S.; Elmore, J. S.; Dodson, A. T.; Mottram, D. Volatile flavour profile of goat meat extracted by three widely used techniques. *Food Chem.* **2009**, *115*, 1081–1087.

(27) Vasta, V.; Priolo, A. Ruminant fat volatiles as affected by diet. A review. *Meat Sci.* **2006**, *73*, 218–228.

(28) Resconi, V. C.; Campo, M. A.; Escudero, A.; Ferreira, V.; Montossi, F.; Sañudo, S. Influencia del sistema de alimentación sobre el aroma de la carne de cerdo. Estudio sensorial y químico. *Eurocarne* **2009**, *181*, 48–58.

(29) FAO-WHO. Piperonyl butoxide. In *Pestice Residues in Food—2001. Evaluation Part I—Residues*. Food and Agriculture Organization of the United Nations and World Health Organization: Rome, Italy, 2002, pp 607−706.

(30) Celinska, E.; Grajek, W. Biotechnological production of 2,3-butanediol—current state and prospects. *Biotechnol. Adv.* **2009**, *27*, 715–725.

(31) Xiao, Z. J.; Liu, P. H.; Qin, J.Y. y; Xu, P. Statistical optimization of mediumcomponents for enhanced acetoin production from molasses and soybean mealhydrolysate. *Appl. Microbiol. Biotechnol.* **2007**, *74*, 61–68.

(32) Moñino, M. I.; Martínez, C.; Sotomayor, J, A.; Lafuente, A.; Jordán, M .J,. Polyphenolic transmission to segureño lamb meat from ewes dietary supplemented with the distillate from rosemary (*Rosmarinus officinalis*) leaves. *J. Agric. Food Chem.* **2008**, *56*, 3363–3367.

(33) Hansen, L. L.; Larsen, A. E.; Jensen, B. B.; Hansen-Møller, J.; Barton-Gade, P. Influence of stocking rate and faeces deposition in the pen at different temperatures on Skatole concentration (boar taint) in subcutaneous fat. *Anim. Prod.* **1994**, *59*, 99–110.

(34) Sivadier, G.; Ratel, J.; Engel, E. Persistence of pasture feeding volatile biomarkers in lamb fats. *Food Chem.* **2010**, *118*, 418–425.

(35) Flores, M.; Olivares, A. Release of aroma compounds from dry-fermented sausages as affected by antioxidant and saliva addition. *Eur. Food Res. Technol.* **2008**, *1*, 283–290.

(36) Machiels, D.; Istasse, L. Evaluation of two commercial solid-phase miroextraction fibres for the analysis of target aroma compounds in cooked beef meat. *Talanta* **2003**, *61*, 529–537.

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