Effect of finishing diet and duration on the sensory quality and volatile profile of lamb meat

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Abstract

Animal production factors diet can affect the sensory quality of lamb meat. The study investigated the effect of diet composition and duration of consumption on theproximate analysis, volatile profile and sensory quality of lamb meat. Ninety-nine male Texel × Scottish Blackface lambs were raised at pasture for 10 months before being assigned in groups of 11 to one of the following treatments: 100% Silage (S) for 36 (S36), 54 (S54) or 72 (S72) days; 50% Silage 50% - 50% Concentrate (SC) for 36 (SC36), 54 (SC54) or 72 (SC72) days; 100% Concentrate (C) for 36 (C36) or 54 (C54) or 72 (C72) days. A trained sensory panel found *Intensity of Lamb Aroma*, *Dry Aftertaste* and *Astringent Aftertaste* to be higher in meat from lambs on the concentrate diet. Discriminant analysis showed that the volatile profile enabled discrimination of lamb based on dietary treatment but the volatile differences were insufficient to impact highly on sensory quality. Muscle from animals in the S54 group had higher *Manure/Faecal Aroma* and *Woolly Aroma* than the SC54 and C54 groups, possibly related to higher levels of indole and skatole. Further research is required to establish if these small differences would influence consumer acceptability.

Keywords: Animalfeed, Silage, Concentrate, Discriminant analysis, Palatability, SPME/GC/MS

1 Introduction

The main feedstuffs consumed by sheep for meat production are derived from cereal grains and pasture (either grazed or ensiled grass), with combinations of both feed sources often in use over the lifetime of animal (Almela et al., 2010). The growth rates of sheep receiving solely grass-based diets are lower and ultimate carcass weights may also be lower (Murphy, Loerch, McClure, & Solomon, 1994; Priolo, Micol, Agabriel, Prache, & Dransfield, 2002); thus, grain-based concentrates, which are more energy dense, are often used to shorten the time to slaughter, increase dressing percentage, and improve carcass quality (De Brito, Ponnampalam, & Hopkins, 2017; Jaborek, Zerby, Moeller, & Fluharty, 2017) .

In addition to the effects of diet on production parameters (De Brito et al., 2017), dietary constituents may also have a considerable effect on meat quality (Kitessa et al., 2009). There are differences in the consumer acceptability of meat from grain-fed and grass-fed sheep (Font i Furnols et al., 2006; Sanudo et al., 2007) attributable to, among other factors, variation in the level of intramuscular fat (IMF) and subcutaneous fat and their fatty acid composition (Howes, Bekhit, Burritt, & Campbell, 2015). Consumer assessment of lamb meat is influenced by the taste and/or aroma deriving from volatile compounds, which are known to be affected by the relative proportions of fatty acids in the meat (Ponnampalam, Sinclair, Egan, Ferrier, & Leury, 2002). With regard to flavour specifically, the extent to which flavour intensity is altered depends on the types of both forage and grain consumed (Duckett & Kuber, 2001). Meat from sheep receiving primarily grass-based diets (pasture or grass silage) is reported to have a pastoral (grassy) flavour (Young, Lane, Priolo, & Fraser, 2003). In this context, nutritional strategies may be used to modulate the sensory quality of lamb ultimately affecting consumer preference (Almela et al., 2010); There are other instances too, in which nutritional interventions could be useful. For example, in a previous

78 study (Gkarane et al., 2017), we reported less favourable sensory attributes in lamb from
79 rams compared to castrates; in this instance a modification to the diet might be useful in
80 overcoming undesirable sensory attributes. The objective of the current study was to test the
81 hypothesis that different proportions and durations of feeding cereal concentrate and silage-
82 based diets would affect the sensory quality and volatile profile of lamb meat from rams.
83

84 **2 Materials and methods**

85 **2.1 Animal husbandry, slaughter and sampling**

86 All animal procedures used in this study were conducted under experimental license
87 from the Irish Health Products Regulatory Authority (HPRA) in accordance with the
88 European Union (Protection of Animals used for Scientific Purposes) Regulations 2012 (S.I.
89 No. 543/ 2012). Ninety-nine ram lambs (Texel × Scottish Blackface) were sourced from Irish
90 farms in March 2015. Lambs were raised at pasture from birth (March 2015) and were
91 weaned at 130 d of age after which they were transported to the Teagasc Sheep Research
92 Centre, Athenry, Co. Galway, Ireland (Claffey et al., 2018). Lambs were maintained at
93 pasture until selected for commencement of an intensive indoor finishing period. Lambs were
94 allocated to the following nine dietary treatments consisting of three grass silage:concentrate
95 ratios (100:0 (S), 50:50 (DM basis) (SC), 0:100 (C)) with each diet being fed for three pre-
96 slaughter feeding durations (36, 54 and 72 d) to give the following dietary treatments: S36,
97 S54, S72, SC36, SC54, SC72, C36, C54, C72. The grass silage was predominantly *Lolium*
98 *perenne L.* and the concentrate diet consisted of 30% maize, 30% barley, 16.5% soya hulls
99 and 15.5% soybean meal. In line with commercial practice, lambs were selected for treatment
100 based on initial live weight and predicted growth rate on the assigned diets to yield lambs
101 with similar weights at slaughter. Thus, the lightest lambs were assigned to the C72 treatment
102 and the heaviest to the S36 treatment. For the indoor finishing period (36, 54 or 72 d) lambs

were individually penned in metal floor feeding pens (182 cm × 122 cm). At the end of the finishing period, lambs were transported to a commercial abattoir (Gillivan's, Moate, Co. Westmeath, Ireland) for slaughter. The mean ages in days (±SD) of the animals at slaughter were 252 (±6.4), 260 (±3.7), 273 (±6.0), 248 (±3.8), 254 (±4.8), 271 (±5.3), 248 (±6.1), 258 (±5.0), 266 (±4.3) for the S36, S54, S72, SC36, SC54, SC72, C36, C54, C72 treatments, respectively. After slaughter, carcasses were chilled overnight and transported to Teagasc Food Research Centre, Ashtown, Dublin 15, Ireland, for dissection. Ultimate pH (pHu) of *M. longissimus thoracis et lumborum* (LTL) was measured at 25 h post slaughter at the 13th rib using a SympHony SP70P hand-held pH meter (VWR, Dublin, Ireland). Both LTL muscles were excised from each carcass, cut into 2.5 cm thick steaks, vacuum packed, aged for 8 d at 4 °C and frozen at -20 °C until required for analysis.

2.2 Compositional analysis

Samples of LTL were thawed overnight at 4 °C and homogenized using a Kenwood CH180 Compact Mini Chopper (Kenwood, Hampshire, UK). Moisture and intramuscular fat (IMF) contents were determined using the SMART Trac Rapid Fat Analyzer (CEM Corporation, NC, USA) according to AOAC Methods 985.14 and 985.26 (AOAC, 1990), respectively. Protein concentration was determined using a LECO FP328 (LECO Corp., MI, USA) protein analyzer based on the Dumas method and according to AOAC method 992.15 (AOAC, 1990). Ash was determined following incineration of samples overnight in a furnace at 540 °C.

2.3 Reagents and fibres for volatile analysis

Volatile standards, the alkane mixture (C7 - C30), methanol (for preparation of stock solutions of the standards), and sodium sulfate were supplied by Sigma-Aldrich Ireland Ltd

(Arklow, Co. Wicklow, Ireland). The volatile standards hexanoic acid and α-terpineol were supplied from VWR International Ltd (Blanchardstown, Dublin 15, Ireland) while 1-pentadecanol was supplied from Fisher Scientific Ireland Ltd (Blanchardstown, Dublin 15, Ireland). Solid phase microextraction (SPME) fibres (50/30 μm CAR/DVB/PDMS fibre; 1 cm length) were supplied by Agilent Technology (Part Number: SU57298U; Unit 3, Euro Business House, Cork, Ireland). All reagents and chemicals were of chromatographic quality.

2.4 Sample preparation and volatile analysis

Before analysis LTL samples were thawed by immersion of frozen vacuum packed samples in water at room temperature for 20 min. Thawed steaks were grilled with the fat attached, using a clamshell grill until an internal temperature of 70 °C was reached (monitored using a hand-held digital thermometer; Eurolec, Dublin, Ireland). Subcutaneous fat was removed and 7 g from the core was weighed and homogenized with 7 g Na_2SO_4 using a Kenwood CH180 Compact Mini Chopper (Kenwood, Hampshire, UK). A 5 ± 0.05 g sample of the mixture was placed in a 20 ml glass headspace vial sealed with a polytetrafluoroethylene (PTFE)-faced silicone septum (VWR, Dublin, Ireland). The vial containing the sample was equilibrated in a water bath set at 90 ± 2 ºC for 20 min and the fibre was exposed to the headspace over the sample for a further 20 min. These SPME conditions (adopted based on maximizing the number of compounds detected, the total peak area and the detection of BCFAs) were considered optimum as previously described in Gkarane et al. (2018). After adsorption, the fibre (50/30 μm CAR/DVB/PDMS) was removed from the vial and immediately inserted into the injection port of the GC. Analysis of the volatile compounds was carried out using a Varian 3800 GC coupled to a Varian Saturn 2000 ion trap mass spectrometer (Varian Chromatography Systems, Walnut Creek, CA, USA). Volatile extraction, adsorption and injection were performed manually. The injector,

operating in splitless mode, was set at 250 °C and the desorption time was 8 min. Helium was used as carrier gas with a constant flow rate of 1.0 ml/min. Volatile compounds were separated using an Agilent ZB-5MS column (30 m length, 0.25 mm internal diameter, 0.25 μm film thickness) (Agilent Technologies, Palo Alto, CA, USA). The GC oven temperature was programmed as follows: 40°C for 5 min, increasing to 230 °C at 4 °C/min and holding for 5 min, with a total acquisition time of 57.5 min. The GC/MS transfer line was heated at 280°C. Acquisition was performed in electron impact (EI) mode (70 eV) at 10 microscans/s, scanning the mass range 33–230 m/z. Saturated n-alkanes (C7 - C30) injected directly (1 μl) onto the column were run under the same GC-MS conditions (at split ratio of 1:50) to obtain linear retention index (LRI) values for the volatile compounds detected. Compounds were identified by comparing their mass spectra with those of spectra from the NIST/EPA/NIH Mass Spectral Database (Version 2.0 g, 2011), those of authenticated standards and linear retention indices matching those of published values (Gkarane et al., 2018). Individual animals were considered as experimental units and one meat sample from each animal was subjected to analysis using a randomized block design to avoid experimental bias. Integration of the peak areas of the volatile compounds used specific ion identification for each molecule (to deal with co-elution of some compounds). An external standard (bromobenzene (10 ppm)) was run daily under the same SPME and GC-MS conditions as the samples. For volatile analysis, the peak area (PA) of each volatile was first normalised against bromobenzene before adding a constant (+1) and being logarithmically transformed to achieve a normal distribution. The amount of each volatile was expressed as logarithmically transformed PA for that compound.

2.5 Lamb meat preparation for sensory analysis

176 The LTL muscle from the left side of each carcass was used for sensory analysis. On
177 the day of sensory testing, packaged frozen steaks were thawed by immersion in water at
178 room temperature for 45 min. Steaks were grilled, with subcutaneous fat attached, to an
179 internal temperature of 70 °C, using a clamshell grill. On reaching 70 °C (monitored using a
180 hand-held digital thermometer (Eurolec, Dublin, Ireland)) the steaks were removed from the
181 grill, wrapped with aluminium foil and allowed to rest for 3 min. Each steak was unwrapped
182 and following removal of the subcutaneous fat, cut into 8 pieces of approximately 2 cm^3.
183 Samples were re-wrapped with foil, assigned a random three-digit code, held in an oven set at
184 60 °C and served to the panellists within 20 min.
185

186 **2.6 Panel training**

187 Staff at Teagasc Food Research Centre, Ashtown, participated as sensory panellists in
188 16 training sessions prior to participating in sensory testing. Training sessions included: lamb
189 meat tasting to generate descriptors for aroma, flavour, texture/mouthfeel, taste and aftertaste;
190 spiking sessions using lamb flavour/aroma compounds; and training using physical and
191 chemical reference standards. A detailed procedure for the panel training is described in
192 Gkarane et al. (2017).
193

194 **2.7 Quantitative descriptive analysis**

195 Quantitative descriptive analysis (QDA) was performed on one day per week over 8
196 weeks with two sensory sessions per day (morning and afternoon). In each session, six
197 samples were assessed using a balanced and randomized design. Panellists were asked to rate
198 38 attributes (generated during the training) for each sample, by marking a point on a 100
199 mm unstructured line scale. Unsalted crackers and water at room temperature were given to

panellists to cleanse the palate between samples. The sensory attribute definitions, agreed during the training sessions (Gkarane et al., 2017), were available to each panellist during tasting. Panellist evaluations were recorded using Compusense 5 (v4.4, Compusense Inc., Guelph, Ontario, Canada).

2.8 Statistical analyses

Proximate and sensory analysis data were tested for the normality of the residuals for each variable. In the case of non-normal distribution, data were transformed using the Box-Cox transformation. All data were analysed using a mixed model with diet, duration and diet x duration as fixed effects (SAS (v9.4)). For the sensory data, the sensory analysis session and carcass weight were considered as random effects. Analysis was conducted in the MIXED procedure of SAS (v9.4). All data were presented as least square means.

Principal component analysis (PCA) of the sensory and volatile data for the nine treatments was performed using XLSTAT®statistical software (Version 19.01.41647; Addinsoft, Paris, France). Associations between sensory attributes and diets, and volatile compounds and diets were also investigated using Discriminant Analysis (DA) performed using XLSTAT®statistical software (Version 19.01.41647; Addinsoft, Paris, France).

3. Results and discussion

3.1 Proximate analysis

There was no difference in muscle fat content among dietary treatments or finishing periods (Table 1). Other authors have reported that lambs receiving concentrate diets generally have higher growth rates (Fraser & Rowarth, 1996) and IMF than lambs receiving pasture-based diets (De Brito et al., 2017). However, Crouse et al. (1978) found no difference

224 in fat thickness or percentage carcass fat of lambs fed low, medium or high energy diets and
225 slaughtered at constant weights. Similarly, Aurousseau et al. (2007) detected no differences
226 in the lipid content of *M. longissimus thoracis* of lambs raised and finished on pasture only,
227 raised on grass and finished in stalls for 22 or 41 d, or raised and finished indoors (in stalls)
228 on concentrates and hay only. They attributed the lack of differences between treatments to
229 similarity in energy expenditure between animals and a higher rate of gain from good quality
230 grass.

231 For protein, there was a diet × duration interaction whereby the muscle from the S
232 group had lower protein content than that of the SC and C groups at 54 d and 72 d, but there
233 were no differences due to diets at 36 d (Supplementary Table S1). The lower protein content
234 of the lamb muscle from the S group may be explained by the fact that concentrate diets have
235 higher dry matter and crude protein content than silage-based diets (Warren et al., 2008);
236 however, this was more noticeable when the feeding duration increased to 54 and 72 days. In
237 addition, there were differences due to duration in the C group, whereby the muscle of the 54
238 d and 72 d groups had higher protein content than the 36 d group. In general, concentrate-
239 based diets favour the production of propionate leading to increased insulin secretion and
240 stimulation of protein and fat synthesis in muscle (Weekes, 1986). Muscle from lambs
241 receiving the experimental diets for 36 and 54 d duration had higher muscle ash content ($P < .05$)
242 than lambs fed for the 72 d duration, although there was a diet × duration interaction
243 whereby the SC group at 54 d had higher ash content than the S and C groups.

244

245 **3.2 Effect of diet on the sensory and volatile profiles of lamb meat**

246 In general, a limited effect of the different dietary treatments on the 38 sensory
247 descriptors was noted (only seven were significantly affected; $P < .05$) (Table 2). For three of
248 these (*Animal/Farm Smell, Woolly Aroma* and *Fattiness*) there were diet × duration

interactions which are discussed in the next section (3.3). *Intensity of Lamb Aroma*, *Dry Aftertaste* and *Astringent Aftertaste* scored higher ($P < .05$) in the C group compared to the S and SC groups. *Farmyard Flavour* scored lower ($P < .05$) in the SC group compared to the C group, but was similar to S group. Although significant effects on sensory descriptors were few, lamb from animals fed the SC group received lower scores ($P = .015–0.078$) for attributes that may be considered hedonically negative by some consumers (i.e. *Animal/Farm Smell*, *Woolly Aroma, Manure/Faecal Aroma*, *Off-flavours*) (Table 2) although no consumer evaluation was performed in this study. Similar conclusions regarding lamb meat assessed by European consumers was reported by Font i Furnols et al. (2009) where meat from lambs fed concentrate or a mixture of pasture and concentrate was more acceptable compared to meat from lambs at pasture. Specifically, the meat from lambs fed a mixture of pasture (6% of live weight, LW) and concentrate (1.2% of LW) was the most acceptable. Arsenos et al. (2002) showed that meat from lambs fed lucerne hay with low and medium levels of concentrate was preferred more than meat from lambs fed high levels of concentrates. Other studies have reported bigger differences when comparing grass-based systems with concentrate-based system (Priolo et al., 2002; Resconi, Campo, Furnols, Montossi, & Sanudo, 2009), with concentrate-fed lambs having more intense lamb odour and/or flavour than grass or forage-fed lambs but also higher acceptability (Borton, Loerch, McClure, & Wulf, 2005; Resconi et al., 2009; Schreurs, Lane, Tavendale, Barry, & McNabb, 2008).

The volatile analysis showed that only ten volatile compounds were significantly ($P <.05$) affected by diet (Table 3), seven of which showed diet × duration interactions (described in section 3.3). The SC and C groups had higher ($P < .05$) values for dimethyl sulphide (formed through Strecker degradation of methionine (Bailey, Rourke, Gutheil, & Wang, 1992)), than the S diet. Levels of hexanal (a compound that derives from oxidation of linoleic acid in muscle (C18:2n-6) (Elmore et al., 2005)), increased gradually with increasing

dietary concentrate although only the C and S groups were significantly different from each other ($P < .05$). This could be due to the higher proportion (%) of C18:2n-6 in the C group compared to the other groups (Rowe, Macedo, Visentainer, Souza, & Matsushita, 1999). Muscle from lambs fed the S diet had higher values ($P < .05$) for skatole than the SC and C diets. Skatole (which has a "faecal/manure aroma") derives from the degradation of dietary tryptophan and since lush pasture is a source of more readily degradable protein than cereal concentrates, it is also a possible source of tryptophan (Tavendale, Lane, Schreurs, Fraser, & Meagher, 2006). In addition, pasture-based diets have a high ratio of protein to readily fermentable carbohydrate (Schreurs et al., 2008; Young et al., 2003). This may explain the higher levels of skatole in muscle from animals on the S group compared the other groups. Priolo et al. (2004) reported differences in p-cymene and eight sesquiterpenes among lambs fed either on grass or on concentrates for different periods while Resconi et al. (2010) found that lambs fed only on pasture had lower levels of carbonyl compounds (alkanals, alkadienals, ketones, strecker aldehydes) than those fed on grass with a concentrate supplement, or only with concentrate.

Multivariate analysis techniques were applied to investigate potential differences between groups and associations with the sensory and volatile data. Following discriminant analysis of the sensory data, the first component (F1) explained 58.87% of the variation and the second component (F2) explained 41.13% of the variation (Fig. 1). The centroids of the dietary treatments were placed in different quadrants (Fig. 1a), revealing some associations with some sensory attributes (Fig. 1b). The factor loadings of the sensory attributes that were considered significant were higher than 0.30. In general, the overlapping of the groups confirmed that the sensory profile of the lambs fed on different diets was similar. Also, the P values from Wilk's Lambda test, Pillai's trace test and Roy's greatest root test showed that the mean vectors only approached significance (range $P = .06$-0.10). Nevertheless, the C group

(centroid located in the upper right quadrant) was more associated with *Dry Aftertaste* and *Astringent Aftertaste.* The S group (centroid located in the upper left quadrant) was more associated with *Fattiness.* For the SC group (centroid in the bottom left quadrant), although visually it was associated with *Juiciness, Intensity of Roast Meat Aroma* and *Intensity of Roast Meat Flavour,* the factor loadings of these attributes were ≤0.30. However, it is clear that the SC group was not associated with attributes that may be viewed as undesirable (i.e. *Manure/Faecal Aroma, Animal/Farm Smell, Off-flavours, Farmyard Flavour;* factor loadings >0.30 for F2).

The discriminant analysis plot of the volatile data (Fig. 2) showed that the three groups (S, SC and C) were clearly separated. The first component (F1) explained 73.04% of the variation and the second component (F2) explained 26.96 % of the variation. The factor loadings of the volatile compounds that were considered significant were equal or higher than 0.30. The P values from Wilk's Lambda test, Pillai's trace test and Roy's greatest root test (P <.001) indicate that at least one of the groups was different from another, whereby according to the Fisher distances test the C group differed from the S group (P <.001) and from the SC group (P =.001). For F1, the S and SC groups (both placed on the left side of the plot) were separated from the C group. The compounds that contributed to this separation were 2,5-dimethylpyrazine, 2,6-dimethylpyrazine, 2-ethyl-3,6-dimethylpyrazine, (*E*,*Z*)-2,6-nonadienal, pentadecane, hexadecane, and pentadecanol (factor loadings ≥0.3 for F1, data not shown). The slight overlap of the S and SC groups indicated that their volatile profile had some similarities. The results are in accordance with Vasta et al. (2011) who, through discriminant analysis, showed that the volatile profile of meat from animals fed silage-based diets was different from those on a concentrate-based diet suggesting that this could be due to the presence of compounds in silage-based diets arising from bacterial fermentation of herbage that makes the "volatile fingerprint" different. The second component separated the SC from

the C and S groups and the compounds that contributed to the variation were dimethyl sulfide and indole (factor loadings ≥0.3 for F2). The differences in the volatile profile (Fig. 2) show that both S and SC groups differed from the C group; however, the differences were not reflected in the sensory quality to a large extent as few differences were detected (Table 2). The explanation could be that, while the volatile analysis showed 10 compounds to be significantly affected, only seven (dimethylsulfide, hexanal, 2,6-nonadienal, indole, skatole, 2,5-dimethylpyrazine and 2-ethyl-3,6-dimethylpyrazine) have low odour thresholds and have been reported to be odour-active in previous lamb meat flavour studies (Gkarane et al., 2018). Furthermore, only three out of the seven compounds (dimethylsulfide, hexanal and skatole) had a "clear" diet effect since the others had an interaction with duration. These compounds, even if present at concentrations above the odour threshold, may not be adequate to elicit significant sensory differences among diets which could explain the similarity in the sensory profiles of the lambs on different diets. Another hypothesis is that the panellist's sensitivity was insufficient to detect the differences in the aroma or that even if they detected them they didn't score them very differently on the magnitude scale of 0-100. Thus, while the discriminant analysis separated the lamb based on diets, it seems that there are limitations that should be considered regarding the compounds that could ultimately influence flavour.

The fact that only few effects of dietary treatment on the volatile and sensory profiles of lamb were noted in the present study is surprising given that differences in the fatty acid profile of the lambs due to the different dietary treatments were present (unpublished results). For example, the C18:3 content was higher ($P < .001$) and the C18:2 content lower ($P < .001$) in LTL from the S treatment compared to the C treatment while LTL from the SC treatment had intermediate values (unpublished results). However, the lack of differences in IMF in this study could explain the lack of differences in the volatile profile due to diets. According to Vasta, D'Alessandro, Priolo, Petrotos, and Martemucci (2012) andFrank, Kaczmarska,

Paterson, Piyasiri, and Warner (2017) most of the odour-impact volatiles in meat systems are lypophilic and their accumulation in animal tissue is correlated with the level of intramuscular fat deposition. Furthermore, differences in flavour volatiles and/or fatty acid composition following diet modification do not always have a major effect on sensory quality as reported by Kitessa et al. (2009) and Muir, Deaker, and Bown (1998). It is also important to recognise that the volatiles extracted by a static method headspace such as SPME may not be representative of the headspace volatiles (considering that many factors (Jelen, Majcher, & Dziadas, 2012) influence the extracted compounds). Finally, the compounds detected by SPME may not be perceived by trained panellists and the perception of trained panellists can't be equated to the perception of consumers (Munoz, 1998).

3.3 Effect of finishing duration on the sensory and volatile profiles of lamb meat

Sensory analysis showed that only two attributes (*Animal/Farm Smell* and *Woolly Aroma*) were affected by finishing duration, both of which had a diet × duration interaction which will be described later in this section (Table 2). A recent study (Guerrero et al., 2018) also reported that feeding duration (30, 50 or 70 d) had a minor impact on sensory attributes of dry cured ham from culled ewes.

The volatile analysis showed that seven volatile compounds were affected ($P < .05$) by the finishing duration, regardless of the finishing diet (Table 3). For four compounds (octanal, nonanal, 1-octanol, and nonanoic acid) the 54 d group had higher levels ($P < .05$) than the other two groups (36 d and 72 d) which did not differ from each other. For two compounds (dodecanal and tridecanal) the 54 d group was higher ($P < .05$) than the 72 d group but both were similar to the 36 d group. For one compound (2-pentylfuran) values for the 54 and 72 d groups were both higher ($P < .05$) than the 36 d group. This quadratic pattern (i.e. an increase to 54 d and a decrease thereafter) could be attributed to a number of factors including the

374 different average daily gains and feed conversion efficiencies of the lambs. In the current
375 study the average live weights (and ages) of lambs assigned to the experimental diets (S, SC
376 and C) were 41.9 ± 2.4 kg (214 ± 5 d), 39.0 ± 5.2 kg (204 ± 5d) and 38.9 ± 5.9 kg (197 ± 8 d)
377 for the 36, 54 and 72 d groups, respectively. These differences in maturity and associated
378 differences in average daily gain (ADG) and feed conversion efficiency (FCE), on
379 assignment to the experimental diets, may have contributed to the minor differences in
380 sensory character and volatiles. Similarly Arsenos et al. (2002) reported that lambs
381 slaughtered at similar target slaughter weights may have differences in degree of maturity
382 which may impact on meat quality and consumer acceptability.

383 Studies indicate that regardless of diet there is a limit to daily intake in ruminants
384 (Allison, 1985; Caton & Dhuyvetter, 1997) after a defined period on a diet. A multitude of
385 factors can affect feed palatability in ruminants and, thus, voluntary feed intake and rate of
386 passage through the gut, including interactions between environmental conditions, animal
387 requirements (physiological or metabolic demands), physical characteristics of the diet
388 (composition, digestibility, energy density) and amount of protein which bypasses the rumen,
389 efficiency of microbial growth and extent of methane loss (Baumont, 1996; Caton &
390 Dhuyvetter, 1997; Decruyenaere, Buldgen, & Stilmant, 2009; Okine, Mathison, Kaske,
391 Kennelly, & Christopherson, 1998). These factors may in turn be influenced by feeding
392 duration with an ultimate effect on the lamb's metabolism and meat quality.

393 There were some interactions between diet and duration with respect to their effects
394 on sensory and volatile profiles. The sensory analysis showed differences among groups at 54
395 d, whereby *Manure/Faecal Aroma* scores for the S group were higher ($P < .05$) than the
396 scores for the SC and C groups, but there were no differences among groups at the other two
397 feeding durations (Supplementary Table S2). In the S group specifically, scores of
398 *Manure/Faecal Aroma* and *Woolly Aroma* for 54 d were higher ($P < .05$) than for 36 d and 72

d ($P < .05$) whereas for *Animal /Farm Smell* scores for 54 d were higher ($P < .05$) than 36 d but similar ($P > .05$) to 72 d. For *Fattiness/Greasiness*, scores from the S group were higher ($P < .05$) than the scores of SC and C groups only at 72 d.

There were ten significant ($P < .05$) diet × duration interactions in the volatile analysis (Supplementary Table S3). For (*Z*)-4-heptenal there were no differences due to duration in the S group; however, in the SC group the 54 d value was higher ($P < .05$) than the 72 d value, neither of which differed ($P > .05$) from the 36 d value, while in the C group the 36 d value was higher ($P < .05$) than both the 54 and 72 d values. In addition, there were differences due to diet in the 72 d period, with S group having higher values than the C group and similar to the values of the SC group. For (*E,Z*)-2,6-nonadienal there were differences due to duration only in the SC and C groups, whereby the 36 and 54 d values, which did not differ, were higher ($P < .05$) than the 72 d values. A difference due to diet was found only for the 72 d group, whereby the S group had higher ($P < .05$) values than both the SC and C groups which did not differ. These two aldehydes derive from linolenic acid (C18:3n-3) (Elmore et al., 2005), associated with grass-based diets (Enser et al., 1998), which could explain why levels were lower with inclusion of concentrates for the longer (i.e. 72 d) finishing duration.

For 1-pentadecanol, the S and SC groups had higher ($P < .05$) values at 36 and 54 d, which did not differ, compared to the C group; at 72 d values decreased ($P < .05$) from S to SC to C group. Long-chain fatty alcohols, like pentadecanol, derive from wax ester hydrolysis and are considered as diet biomarkers; notable differences in the alcohol content of wax are found mainly among grasses and legumes (Kelman, Bugalho, & Dove, 2003), which could explain the higher levels in muscle from the S and SC groups compared with the C group, regardless of the finishing duration. For 2-heptanone, differences due to diet were observed; thus, at 36 d values were lower ($P < .05$) in the S group than either the SC or C groups, which did not differ. This compound was generally present at higher levels (although

not significant) in muscle from the SC and C groups at all finishing durations, probably because it derives from C18:2n-6 (Elmore et al., 2005), which is associated with grain-based diets (Enser et al., 1998). Differences in 2-heptanone due to feeding duration were significant only in the S group, whereby values at 36 d were lower (P <.05) than either 54 or 72 d, which did not differ.

Indole was detected at each duration of feeding in the S group, but only detected at 54 d in the SC group and at 72 d in the C group (Supplementary Table S3). The frequency of detection was higher in muscles from the S group since it derives from tryptophan degradation in the rumen mainly of grass-fed lambs and has been identified with pastoral flavours (Schreurs et al., 2008). The higher scores for *Woolly Aroma* and *Manure/Faecal Aroma* in muscle from the S54 group could be due to the higher levels of indole and skatole (faecal, mothball-like aroma) compared to SC54 and C54 groups (although for skatole the diet × duration effect approached significance (P <.1)).

For 2,5-dimethyl pyrazine, values at 36 d in the S group were higher (P <.0.5) than in the C group, neither of which differed from the SC group; there were no statistical significant differences (P >.05) due to dietary treatment at the other durations of feeding despite the fact that the trend was similar (Supplementary Table S3). Differences due to feeding duration were significant only in the S group, whereby values at 36 d were higher (P <.05) than at 72 d, neither of which differed from 54 d. For 2-ethyl-3,6-dimethylpyrazine, values at 54 d in the S group and SC groups were higher (P <.0.5) than those of the C group, while there were no differences (P >.05) due to dietary treatment at the other durations of feeding (although a similar trend was observed). Similar to 2,5-dimethylpyrazine, the S group, had higher (P <.05) values at 36 d than 72 d, neither of which differed from values at 54 d. Muscle from animals fed the S and SC diets had numerically higher levels for some pyrazines than the C diet. This could be due to a possibly higher content of specific amino acids (e.g. cysteine,

449 glycine), that contribute to the Maillard reaction, in muscle from animals fed silage-based
450 diets as reported by other authors (Farmer, 1994). Koutsidis et al. (2008) reported a
451 significant effect of the diet (grass silage *vs* concentrate) on the concentration of free amino
452 acids (which can participate in the Maillard reaction) in bovine muscle, with animals fed
453 grass silage having higher levels than animals fed a concentrate diet. In addition, Tai and Ho
454 (1997) found that an oxidized cysteine/glucose reaction model produced more pyrazines and
455 furans as opposed to a non-oxidized cysteine/glucose reaction model that produced more
456 sulphur compounds; thus, differences in susceptibility of muscle to oxidation may contribute
457 to differences in pyrazine formation.

458 For pentadecane there were differences among diets at all feeding durations whereby
459 the S group and SC groups, which did not differ, had higher ($P < .05$) values than the C group
460 (Supplementary Table S3). For hexadecane, differences among diets were found for all
461 finishing durations whereby at 36 and 54 d the S group had higher levels ($P < .05$) than the C
462 group but both were similar ($P > .05$) to the SC group, while at 72 d the S and SC groups,
463 which did not differ, had higher values than the C group. Hydrocarbons like pentadecane and
464 hexadecane, are lipid oxidation compounds, and have been characterised as tracers of a
465 pasture diet in lamb (Sivadier, Ratel, & Engel, 2010); this could explain why levels were
466 lower with concentrate feeding at all durations. For 4-methyloctanoic acid differences among
467 diets were detected only at the 36 d of feeding duration with the S and SC groups, which did
468 not differ, having higher values ($P < .05$) than the C group.

469 In general, the majority of the aroma and flavour attribute scores as well as volatile
470 compounds followed a quadratic pattern, i.e. values increased from 36 to 54 d and decreased
471 from 54 to 72 d, mainly in S and SC groups (Supplementary Table S2 and Table S3). The
472 PCA plot (Supplementary Fig. S1) for all nine groups (using only the aroma and flavour
473 attributes and selected volatiles) explained 46.55% of the variance, whereby the first

474 component separated the three groups of 54 days duration (located on the right side of the
475 plot) from the other six groups (left side of the plot). The plot showed that S54 group was
476 characterised by the attributes "*Manure/Faecal Aroma*" and "*Rancid Aroma*", clustered with
477 skatole, indole, p-cresol, 4-heptenal, 2-nonenal, 2,6-nonadienal 2,5-dimethylpyrazine and 2-
478 ethyl-3,6-dimethylpyrazine (factor loadings 0.6-0.8 on PC2). Previous studies have shown
479 that phenols and indoles (associated with animal-like odours) as well as 4-heptenal (Young,
480 Berdagué, Viallon, Rousset-Akrim, & Theriez, 1997; Young et al., 2003) and pyrazines
481 (Bueno et al., 2013) have low odour thresholds and may be causally involved in lamb meat
482 aromas perceived by trained panellists. The SC54 group was characterised by "*Animal/Farm*
483 *Smell*", "*Woolly Aroma*" and "*Sweaty Aroma*" (factor loadings 0.6-0.8 on PC1) (this
484 association is more meaningful when comparing SC54 group with SC36 or SC72 groups; See
485 supplementary Table S2). The compounds which may have contributed to these attributes
486 (Factor loadings 0.6-0.9 on PC1) were mainly lipid oxidation compounds (heptanal, 1-
487 hexanol, 1-heptanol, octanal, 2-octenal, 1-octanol, nonanal, decanal, 2-decenal, 2,4-
488 decadienal, 2-octen-1-ol) and other compounds e.g. α-terpineol, 2-pentylfuran, nonanoic acid,
489 benzaldehyde and phenylacetaldehyde, dimethyldisulfide and dimethyltrisulfide.
490 The results of the PCA plot could also be explained in part by the numerically higher
491 (although not significant) proportions of C18:3n-3, eicosapentaenoic acid (EPA; C20:5n-3)
492 and n-3 fatty acids in the S54 group compared to S36 and S72 groups, the higher proportion
493 of arachidonic acid (C20:4n-6) of the SC54 compared to SC36 and SC72 groups and the
494 higher level of PUFA of C54 group compared to C36 and C72 group (unpublished results).
495

496 **4. Conclusion**

497 When lambs receive different proportions of silage and concentrates for durations up
498 to approximately ten weeks pre-slaughter, effects on the sensory quality (and flavour

volatiles) of lamb meat are relatively few. Some sensory attributes with potentially negative connotations (*Animal/Farm Smell*, *Manure/Faecal Aroma*, *Farmyard Flavour*, *Off-flavours*) appear to be lower when a mixed diet of silage and concentrate is fed. With regard to the duration of feeding, a diet composed of silage only, fed for an intermediate period, appears to be associated with less desirable sensory aroma attributes (*Manure/Faecal Aroma*, *Rancid Aroma*) which could be due to indoles or lipid oxidation compounds. Further research is required to establish if these small differences would influence consumer acceptability

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Conflict of interest

The authors have no conflicts of interest to declare.

Authors contributions

V. Gkarane conducted the experimental work, collected and statistically analysed the data and drafted the manuscript. N. Brunton contributed to the method development for volatile analysis and interpretation of the volatile results. P. Allen contributed to the study design and

sensory analysis. R. Gravador contributed to the sensory analysis and conducted the proximate analysis. N. Claffey contributed to the animal management and sample collection. M. Diskin contributed to the study design and animal management oversight. A. Fahey contributed to the study design and the univariate statistical analysis. L. Farmer contributed to the method development for volatile analysis and manuscript revision. A. Moloney contributed to the study design. M. Alcalde contributed to the sensory analysis. P. Murphy contributed to the multivariate statistical analysis. F. Monahan had overall responsibility for the project, contributed to the study design, method development and interpretation of the results. All authors read the manuscript and contributed to manuscript revisions.

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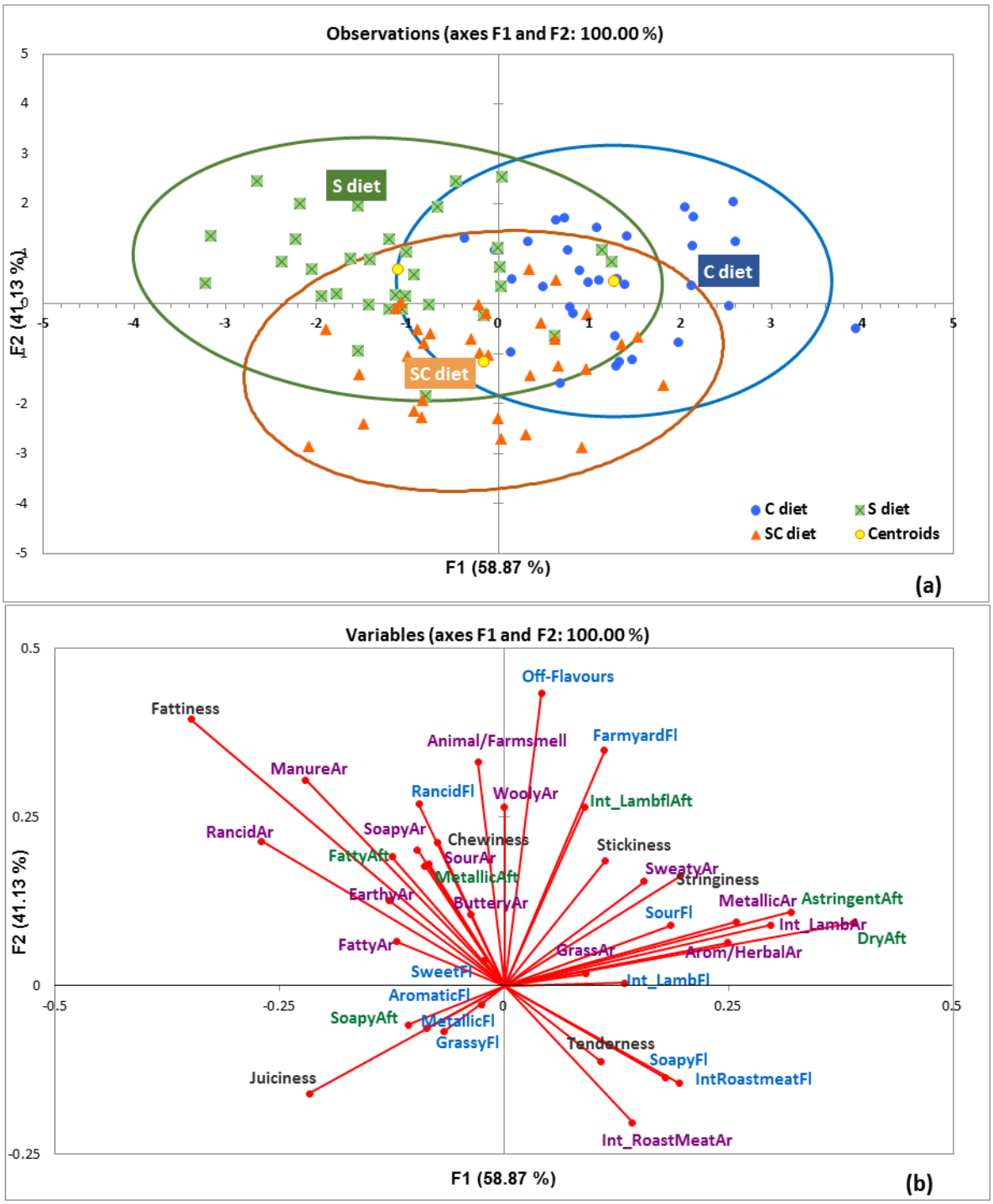

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679 Fig. 1 Discriminant analysis (DA) plot (a) and variable loadings plot (b) of the sensory attributes of
680 lamb meat as affected by dietary treatments. Plot (**a**): S diet refers to the silage diet; SC diet refers to
681 the diet consisting of 50:50 silage:concentrate; C diet refers to the concentrate diet. Plot (**b**): "Ar",
682 "Fl" and "Aft" refer to Aroma, Flavour and Aftertaste attributes, respectively.

683

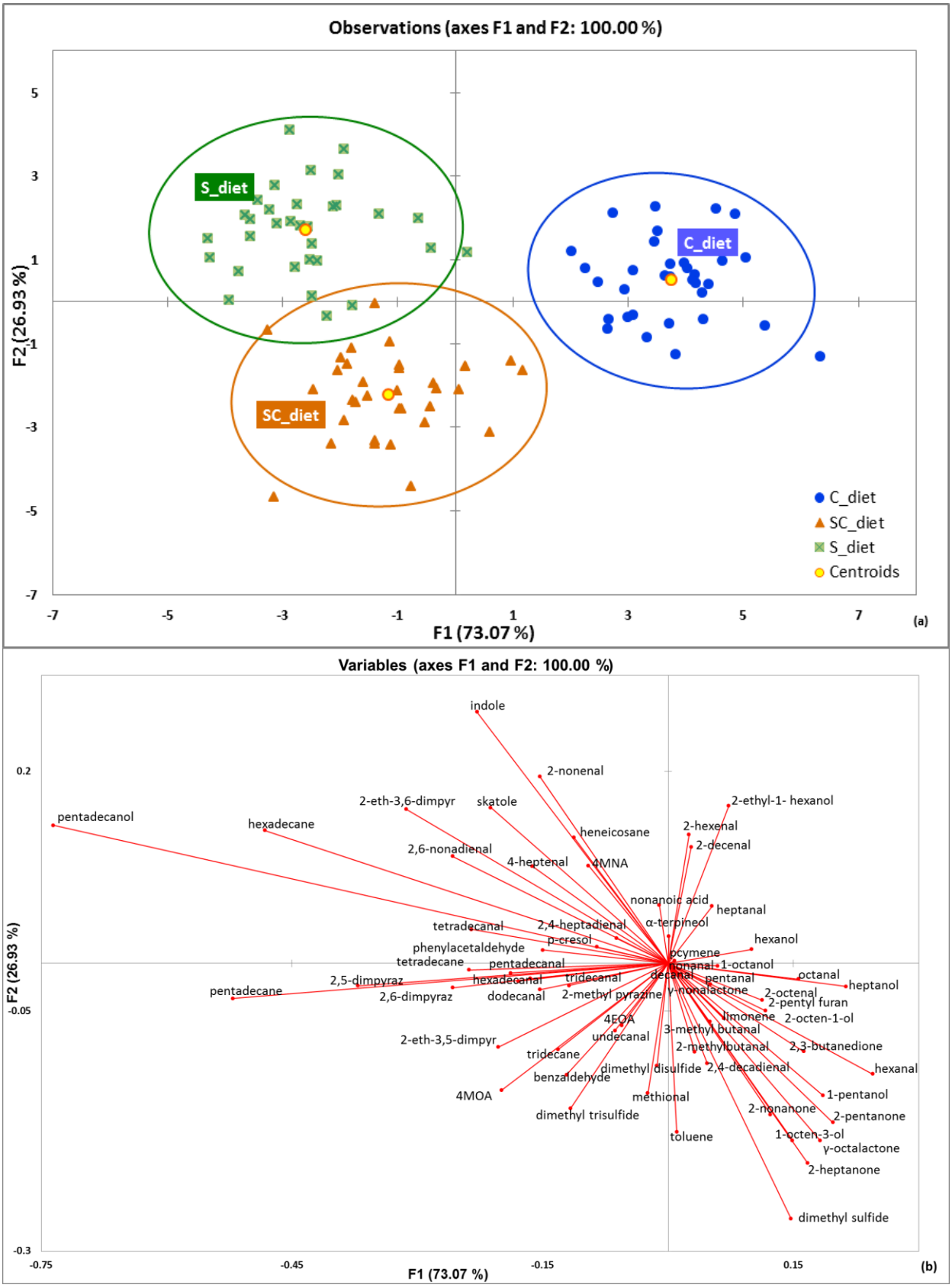

Fig. 2 Discriminant analysis (DA) plot (a) and variable loadings plot (b) of the volatile compounds as affected by dietary treatment. Plot (**a**): S diet refers to the silage diet; SC diet refers to the diet consisting of 50:50 silage:concentrate; C diet refers to the concentrate diet. Plot (**b**) 4-MOA, 4-methyloctanoic acid in muscle; 4-MNA, 4-methylnonanoic acid in muscle, 4-EOA; 4-ethyloctanoic acid in muscle; 2-eth-3,6-dimpyr, 2-ethyl-3,6-dimethylpyrazine; 2-eth-3,5-dimpyr, 2-ethyl-3,5-dimethylpyrazine; 2,5-dimpyraz, 2,5-dimethylpyrazine; 2,6-dimpyraz, 2,6-dimethylpyrazine.

692 Table 1 Least square mean values for proximate analysis and ultimate pH (pHu) in *longissimus thoracis et lumborum* (LTL) muscle fed three different diets
693 (100% Silage (S); 50% Silage: 50% Concentrate (SC); 100% Concentrate (C)) for three durations of feeding (36, 54, 72 days).

	Diet			Feeding duration			SEM	Significance		
	S	SC	C	36	54	72		Diet	Duration	Diet x Duration
Moisture	73.9[b]	73.1[a]	73.3[ab]	73.5	73.6	73.2	0.14	0.041		
Protein	21.1[a]	21.9[b]	22.0[b]	21.5	21.6	21.8	0.10	<0.001		0.001
Fat	3.71	3.94	3.79	3.77	3.78	3.88	0.13			
Ash	1.05	1.09	1.05	1.10[b]	1.08[b]	1.01[a]	0.01		0.001	0.023
pHu	5.73	5.71	5.78	5.69	5.78	5.76	0.02			

694
695 a,b Within row, means assigned different superscripts indicate differences among diets (S *vs* SC *vs* C) or durations (36 *vs* 54 *vs* 72 days).
696

697 Table 2 Least square mean scores for sensory attributes in grilled LTL muscle as affected by diet (100% Silage (S); 50% Silage: 50% Concentrate (SC);
698 100% Concentrate (C)) and duration of feeding (36, 54, 72 days).

	Diet			Duration			SEM	Significance[1]		
Sensory	Silage (S) 100%	50% (S) - 50% (C)	Concentrate (C) 100%	36	54	72		Diet	Duration	Diet × Duration
Aroma										
Intensity of roast meat aroma	41.3	44.8	43.8	44.4	41.7	43.8	0.93			
Intensity of lamb aroma	39.8[a]	40.1[a]	43.1[b]	40.4	40.5	42.0	0.71	0.036		
Grassy	7.5	7.6	7.9	7.8	7.3	8.0	0.28			
Aromatic/herbal	11.8	12.0	13.3	13.2	11.3	12.7	0.38			
Metallic/bloody	14.0	14.0	15.3	14.2	14.0	15.1	0.34			
Animal/farm Smell	15.5[b]	12.8[a]	15.1[b]	12.1[a]	16.6[b]	14.7[b]	0.60	0.039	0.007	0.032
Woolly	14.3[b]	12.5[a]	14.4[b]	11.9[a]	16.2[b]	13.0[b]	0.56	0.045	0.007	0.038
Buttery	7.0	6.8	7.1	6.8	7.3	6.8	0.30			
Fatty	8.2	8.0	7.9	7.6	8.2	8.3	0.33			
Rancid	8.0	6.2	6.3	6.6	7.5	6.4	0.43			
Manure/faecal	9.8	6.8	7.7	6.9	10.1	7.4	0.55			0.016
Sour	7.8[b]	6.6[a]	7.2[ab]	6.0	8.7	7.0	0.43	0.078*		
Sweaty	14.9	14.5	16.2	14.0	16.5	15.1	0.49			
Soapy	3.7	3.2	3.4	3.5	3.3	3.5	0.16			
Earthy	10.5	9.9	10.0	9.8	10.2	10.4	0.27			
Flavour										
Intensity of roast meat flavour	36.9	39.3	39.4	39.5	37.0	39.0	0.78			
Intensity of lamb flavour	42.9	43.5	42.9	43.9	42.8	44.1	0.70			
Grassy	8.3	8.4	8.0	7.8	8.2	8.6	0.24			
Metallic/bloody	20.2	20.6	19.8	20.3	20.7	19.6	0.49			
Aromatic/herbal	9.4	9.3	9.2	8.8	9.1	10.0	0.27			

Soapy	5.2	6.2	6.3	5.3	6.6	5.9	0.28			
Rancid	8.5	6.8	7.8	7.0	8.5	7.6	0.41			
Farmyard	8.9[ab]	7.3[a]	9.9[b]	8.3	8.9	8.9	0.47	0.015		
Sour	7.9	8.2	9.5	9.4	8.3	7.9	0.45			
Sweet	11.4	11.3	11.4	10.8	11.2	12.2	0.39			
Off-flavours	19.6[b]	15.8[a]	19.7[b]	18.9	18.7	17.5	0.67	0.066*		
Texture										
Tenderness	54.4	58.0	57.5	56.7	57.7	55.6	1.57			
Juiciness	48.4	49.1	45.7	47.8	46.1	49.3	0.81			
Chewiness	51.9	46.7	49.5	49.8	47.6	50.7	1.47			
Fattiness/greasiness	30.7	25.5	26.4	27.1	28.4	27.2	0.65	0.003		0.044
Stringiness/fibrousness	33.8	32.5	37.7	36.7	34.1	33.3	1.29			
Stickiness	26.8	25.7	27.9	27.2	27.2	25.9	0.63			
Aftertaste										
Intensity of lamb aftertaste	34.1	32.9	34.7	34.6	33.5	33.7	0.43			
Soapy	9.3	9.5	8.9	8.6	9.4	9.6	0.31			
Metallic/ bloody	20.8	19.1	19.9	19.6	19.4	20.7	0.49			
Fatty/ greasy	17.7	15.9	16.5	16.7	17.4	15.9	0.48			
Dry	11.3[a]	11.8[a]	13.5[b]	12.6	12.2	11.7	0.34	0.009		
Astringent	7.2[a]	7.6[a]	9.3[b]	7.6	8.3	8.2	0.35	0.030		

a,b within row, different superscripts indicate differences among diets (S *vs* SC *vs* C) or durations (36 *vs* 54 *vs* 72 days).

[1]Probability of significance for the main effects of diet, duration and diet x duration tested using the MIXED model (P <.05).

*P <.1.

Table 3 Least square mean values for logarithmically transformed peak areas of aroma compounds detected in the headspace of grilled *longissimus thoracis et lumborum* (LTL) muscle fed three different diets (100% Silage (S); 50% Silage: 50% Concentrate (SC); 100% Concentrate (C)) for three durations (36, 54, 72 days).

Volatile compound	LRI[1]	Ions Used[2]	Method of Identification[3]	Diet			Feeding Duration (days)			SEM	Significance		
				S	SC	C	36	54	72		Diet	Duration	Diet × Duration
Sulphur compounds													
Dimethyl sulfide		63,62,61	NIST, Std, LRI	1.97^{a}	2.90^{b}	2.70^{b}	2.73	2.63	2.22	0.151	0.029		
Dimethyl disulfide	719	94,79	NIST, Std, LRI	2.34	2.71	2.41	2.42	2.89	2.16	0.176			
Dimethyl trisulfide	963	126	NIST, Std, LRI	4.27	4.36	4.21	4.25	4.39	4.21	0.037			
Aldehydes													
2-Methylbutanal		39,41,57	NIST, Std, LRI	4.17	4.36	4.29	4.40	4.28	4.15	0.095			
3-Methylbutanal		41,43,58	NIST, Std, LRI	4.33	4.47	4.47	4.57	4.47	4.24	0.097			
Pentanal		43,44,58	NIST, Std, LRI	4.26	4.32	4.36	4.39	4.42	4.13	0.082			
(*E*)-2-Hexenal	849	39,41,55	NIST, Std, LRI	3.01	2.65	2.97	3.12	2.70	2.82	0.139			
Hexanal	800	39,41,56	NIST, Std, LRI	5.26^{a}	5.37^{ab}	5.45^{b}	5.32	5.43	5.33	0.031	0.044		
Methional	905	48,104	NIST, Std, LRI	3.82	4.13	3.86	4.13	3.93	3.76	0.117			
(*E*,*E*)-2,4-Heptadienal	1008	81,53	NIST, Std, LRI	2.20	2.04	1.91	2.23	2.25	1.66	0.194			
(*Z*)-4-Heptenal	898	67,39,55	NIST, Std, LRI	4.08	4.00	3.96	4.08^{b}	4.06^{b}	3.90^{a}	0.029		0.013	0.007
Heptanal	900	39,41,70	NIST, Std, LRI	5.33	5.30	5.35	5.32	5.40	5.26	0.025			
(*E*)-2-Octenal	1056	39,55,83	NIST, Std, LRI	4.40	4.44	4.48	4.40	4.51	4.41	0.027			
Octanal	1002	41,67,69	NIST, Std, LRI	5.41	5.44	5.50	5.42^{a}	5.55^{b}	5.39^{a}	0.024		0.012	
(*E*,*Z*)-2,6-Nonadienal	1150	41,69,70	NIST, Std, LRI	4.10^{b}	4.01^{b}	3.93^{a}	4.06	4.09	3.89	0.028	0.026	0.007	0.003
(E)-2-Nonenal	1158	29,41,55	NIST, Std, LRI	4.99	4.86	4.86	4.92	4.96	4.82	0.028			
Nonanal	1101	69,81,57	NIST, Std, LRI	6.02	6.03	6.03	6.01^{a}	6.13^{b}	5.95^{a}	0.023		0.006	
(*E*,*E*)-2,4-Decadienal	1315	81,67	NIST, Std, LRI	3.98	4.05	4.03	3.97	4.09	4.01	0.031			
(*E*)-2-Decenal	1260	39,81,55	NIST, Std, LRI	4.47	4.40	4.46	4.42	4.53	4.37	0.029			
Decanal	1204	41,67,55	NIST, Std, LRI	4.83	4.83	4.84	4.82	4.89	4.79	0.021			
Undecanal	1306	41,67,81	NIST, Std, LRI	3.90	4.04	3.78	3.66	3.97	4.09	0.126			
Dodecanal	1406	41,67,81	NIST, Std, LRI	4.43	4.43	4.36	4.39^{a}	4.48^{b}	4.35^{a}	0.022		0.044	
Tridecanal	1510	41,67,81	NIST, LRI	4.48	4.47	4.40	4.46^{a}	4.52^{b}	4.37^{a}	0.024		0.042	

Tetradecanal	1607	41,67,81	NIST, LRI	4.97	4.93	4.85	4.90	4.97	4.87	0.023				
Pentadecanal	1705	41,67,81	NIST, LRI	5.07	5.05	4.96	5.02	5.09	4.97	0.026				
Hexadecanal	1818	41,67,81	NIST, LRI	5.67	5.65	5.55	5.62	5.66	5.59	0.030				
Alcohols														
1-Pentanol	809	41,55,70	NIST, Std, LRI	3.24	3.73	3.88	3.67	3.63	3.55	0.132				
1-Hexanol	868	41,56,39	NIST, Std, LRI	4.35	4.35	4.40	4.35	4.44	4.31	0.026				
1-Heptanol	969	41,55,70	NIST, Std, LRI	4.48	4.52	4.60	4.51	4.60	4.49	0.025				
1-Octen-3-ol	980	43,57,69	NIST, Std, LRI	4.81	4.93	4.93	4.84	4.94	4.89	0.027				
2-Octen-1-ol	1066	41,57,67	NIST, Std, LRI	3.97	4.00	4.01	3.93	4.05	4.01	0.024				
2-Ethyl-1-hexanol	1027	41,55,57	NIST, Std, LRI	4.43	4.27	4.46	4.27	4.48	4.41	0.053				
1-Octanol	1069	41,55,69	NIST, Std, LRI	5.11	5.12	5.14	5.10^{ab}	5.22^{b}	5.04^{a}	0.023		0.008		
α-Terpineol	1191	93,59,121	NIST, Std, LRI	4.91	4.89	4.91	4.87	4.95	4.88	0.036				
1-Pentadecanol	1766	69,83,97	NIST, Std, LRI	5.57^{c}	5.35^{b}	5.00^{a}	5.31	5.31	5.29	0.034	<0.001		<0.001	
Ketones														
2-Pentanone		43,71,86	NIST, Std, LRI	0.70	1.42	1.59	1.24	1.45	1.02	0.166				
2,3-Butanedione		43	NIST, Std, LRI	2.59	3.13	3.42	2.99	3.35	2.80	0.202				
2-Heptanone	887	43,58	NIST, Std, LRI	3.73	4.04	4.03	3.74	4.07	3.99	0.062				0.029
2-Nonanone	1089	43,58	NIST, Std, LRI	3.91	4.01	4.00	3.92	4.04	3.95	0.027				
γ-Octalactone	1251	85,57	NIST, Std, LRI	1.11	1.82	1.89	1.15	1.87	1.80	0.153				
γ-Nonalactone	1356	85,29	NIST, Std, LRI	3.06	3.12	3.13	2.91	3.22	3.18	0.089				
Terpenes														
p-cymene	1020	119,91	NIST, Std, LRI	2.81	2.80	2.82	2.51	3.06	2.87	0.122				
Limonene	1024	67,68,93	NIST, Std, LRI	4.27	4.31	4.32	4.25	4.38	4.27	0.029				
Phenols														
p-Cresol	1071	107,108	NIST, Std, LRI	3.32	3.19	2.99	3.46	3.17	2.88	0.171				
Indoles														
Indole	1287	117,89	NIST, Std, LRI	0.64^{b}	0.09^{a}	0.07^{a}	0.09	0.54	0.17	0.086	0.006		0.021	
Skatole (3-methyl indole)	1379	130,131	NIST, Std, LRI	1.11^{b}	0.51^{ab}	0.34^{a}	0.59	1.05	0.32	0.137	0.048	0.080*	0.063*	
Pyrazines														
2-Methyl pyrazine	822	94,67	NIST, Std, LRI	1.09	1.07	0.65	0.94	1.23	0.64	0.183				
2,5-Dimethyl pyrazine	909	108,42	NIST, Std, LRI	2.44^{b}	2.14^{b}	0.68^{a}	2.27	1.55	1.45	0.222	0.002		0.022	

2,6-Dimethyl pyrazine	909	108,42	NIST, Std, LRI	4.27[b]	4.11[ab]	3.09[a]	3.58	4.05	3.85	0.216	0.055*		
2-Ethyl-3,5-dimethyl-pyrazine	1071	135,134	NIST, Std, LRI	4.86	4.93	4.54	4.83	4.81	4.69	0.084			
2-Ethyl-3,6-dimethyl-pyrazine	1083	135,136	NIST, Std, LRI	2.69[b]	1.66[ab]	1.03[a]	2.45[b]	1.92[ab]	1.01[a]	0.213	0.003	0.012	0.003
Benzenoid compounds													
Benzaldehyde	957	105,77	NIST, Std, LRI	6.19	6.25	6.12	6.15	6.29	6.12	0.034			
Phenyl acetaldehyde	1039	91,92	NIST, Std, LRI	4.97	4.94	4.86	4.90	4.99	4.89	0.033			
Toluene	748	91,92	NIST, Std, LRI	4.68	4.82	4.73	4.72	4.83	4.67	0.038			
Furans													
2-Pentylfuran	987	81,138,53	NIST, Std, LRI	4.03	4.25	4.43	3.66[a]	4.46[b]	4.60[b]	0.139		0.010	
Hydrocarbons													
Tridecane		41,57,71	NIST, Std, LRI	4.52	4.55	4.46	4.49	4.57	4.47	0.028			
Tetradecane		41,57,71	NIST, Std, LRI	4.57	4.54	4.44	4.48	4.60	4.47	0.025			
Pentadecane		41,57,71	NIST, Std, LRI	4.88[b]	4.84[b]	4.62[a]	4.77	4.82	4.75	0.023	<0.001		0.000
Hexadecane		41,57,71	NIST, Std, LRI	4.57[b]	4.46[b]	4.33[a]	4.47	4.49	4.41	0.021	<0.001		0.000
Heneicosane		41,57,71	NIST, Std, LRI	0.98	0.59	0.58	0.52	0.96	0.67	0.122			
BCFAs													
4-Methyloctanoic acid	1232	55,57,73	NIST, Std, LRI	1.31	1.58	0.76	1.18	1.44	1.04	0.157			0.048
4-Ethyloctanoic acid	1313	55,57,71	NIST, Std, LRI	1.94	2.12	1.81	2.34	1.42	2.11	0.168			
4-Methylnonanoic acid	1323	55,57,71	NIST, Std, LRI	1.26	0.89	0.86	0.98	0.76	1.27	0.149			
Organic acids													
Nonanoic acid	1275	60	NIST, Std, LRI	3.74	3.67	3.71	3.58[a]	3.89[b]	3.65[ab]	0.051		0.029	

[a,b] Within row, means assigned different superscripts indicate differences among diets (S vs SC vs C) or durations (36 vs 54 vs 72 days).

[1] Linear retention indices (LRI) calculated from the n-alkanes (C7-C30) run under the same GC-MS conditions as LTL muscle samples.

[2] Specific ions used for volatile identification and peak area integration.

[3] Method of identification: NIST (NIST library), Std (authentic standard) and LRI.

* P <.1

Supplementary Table S1 Least square mean values for proximate analysis and ultimate pH (pHu) in *longissimus thoracis et lumborum* (LTL) muscle fed three different diets (100% Silage (S); 50% Silage: 50% Concentrate (SC); 100% Concentrate(C)) for three durations of feeding (36, 54, 72 days).

	100 % S			SC			100% C				p-values		
	Feeding duration			Feeding duration			Feeding duration						
	36	54	72	36	54	72	36	54	72	SEM	Diet	Duration	Diet x Duration
Moisture	73.4	74.3	74.1	73.4	72.9	72.9	73.7	73.6	72.8	0.14	0.045		
Protein	21.3	20.9^{x}	21.1^{x}	21.9	21.7^{y}	22.1^{y}	21.4^{a}	22.2^{by}	22.3^{by}	0.10	<0.001		0.001
Fat	4.04	3.48	3.6	3.51	4.32	3.98	3.75	3.54	4.07	0.13			
Ash	1.09	1.05^{x}	1.02	1.14	1.10^{y}	1.03	1.08	1.08^{xy}	1.00	0.01		0.001	0.023
pH_u	5.61	5.84	5.75	5.68	5.75	5.69	5.77	5.74	5.82	0.02			

[a,b,c] Within row, means assigned different superscripts indicate differences between durations within each diet (i.e. S36 *vs* S54 *vs* S72, SC36 *vs* SC54 *vs* SC72 and C36 *vs* C54 *vs* C72) .

[x,y,z] Within row, means assigned different superscripts indicate differences between diets with finishing duration 36 or 54 or 72 days (i.e. S36 *vs* SC36 *vs* C36 or S54 *vs* SC54 *vs* C54 or S72 *vs* SC72 *vs* C72).

721 Supplementary Table S2 Least square mean scores for sensory attributes in grilled LTL muscle as affected by (100% Silage (S); 50% Silage: 50%

722 Concentrate (SC); 100% Concentrate (C)) and duration of feeding (36, 54, 72 days).

723

	100 % S			50%S:50%C			100% C			SEM	Significance[1]		
	Feeding duration			Feeding duration			Feeding duration						
	36	54	72	36	54	72	36	54	72		Diet	Duration	Diet x Duration
Aroma													
Intensity of roast meat aroma	43.6	41.6	38.9	46.3	43.3	44.8	43.9	39.6	47.9	0.93			
Intensity of lamb aroma	39.9	39.7	39.9	40.8	39.4	40.1	41.0	42.1	46.1	0.71	0.036		
Grassy	7.4	6.6	8.6	8.3	6.5	8.0	7.6	8.7	7.4	0.28			
Aromatic/herbal	13.2	9.2	12.9	12.5	12.0	11.5	13.2	12.8	13.8	0.38			
Metallic/bloody	14.4	13.0	14.5	13.3	13.5	15.3	14.9	15.5	15.6	0.34			
Animal/farm Smell	12.7[a]	18.9[b]	14.9[ab]	10.7	14.4	13.3	13.2	16.3	15.8	0.60	0.039	0.007	0.032
Woolly	12.7[a]	17.3[b]	13.0[a]	11.4	14.6	11.5	12.0	16.6	14.6	0.56	0.045	0.007	0.038
Buttery	6.2	8.0	6.8	6.4	7.1	7.0	8.0	6.8	6.5	0.30			
Fatty	7.5	8.9	8.4	7.7	8.9	7.5	7.8	6.8	9.1	0.33			
Rancid	8.0	8.6	6.5	5.8	7.4	5.6	5.9	6.5	6.3	0.43			
Manure/faecal	8.5[a]	14.1[by]	7.0[a]	5.4	7.3[x]	7.9	7.4	8.4[x]	7.3	0.55			0.016
Sour	7.0	10.2	6.3	5.6	7.6	6.8	5.9	7.8	8.1	0.43	0.078*		
Sweaty	14.3	16.5	14.1	13.1	16.0	14.3	14.8	16.9	16.9	0.49			
Soapy	4.1	3.3	3.6	2.9	7.6	6.8	3.4	7.8	8.1	0.16			
Earthy	10.6	9.9	11.0	9.5	10.4	9.8	9.0	10.3	10.6	0.27			
Flavour													
Intensity of roast meat flavour	37.4	36.7	36.7	39.9	36.8	41.2	41.8	37.5	38.9	0.78			

Intensity of lamb flavour	43.5	42.1	43.1	44.4	41.5	44.5	43.8	44.8	44.7	0.70		
Grassy	7.8	8.1	9.0	8.1	8.1	9.1	7.7	8.2	8.3	0.24		
Metallic/bloody	21.8	19.3	19.5	19.8	21.5	20.5	18.9	21.2	19.3	0.49		
Aromatic/herbal	9.1	8.6	10.4	8.5	9.1	10.3	8.6	9.6	9.3	0.27		
Soapy	4.9	5.3	5.4	5.4	7.2	6.2	5.3	7.2	6.4	0.28		
Rancid	8.2	8.5	8.6	5.5	7.8	7.1	7.2	9.2	7.0	0.41		
Farmyard	7.9	10.5	8.4	7.4	7.0	7.6	10.4	9.0	10.4	0.47	0.015	
Sour	7.8	8.2	7.7	9.1	7.8	7.7	11.4	8.9	8.3	0.45		
Sweet	11.9	10.4	12.0	10.7	10.9	12.4	9.6	12.3	12.3	0.39		
Off-flavours	19.9	21.2	17.8	15.7	15.5	16.2	21.2	19.3	18.5	0.67	0.066*	
Texture												
Tenderness	58.4	59.1	45.6	54.9	56.0	63.0	56.7	57.8	58.1	1.57		
Juiciness	49.8	45.1	50.3	50.6	46.5	50.2	43.0	46.6	47.3	0.81		
Chewiness	49.1	48.3	58.3	51.9	44.6	43.7	48.4	50.0	50.1	1.47		
Fattiness/greasiness	29.6	31.1	31.5[y]	25.4	26.1	25.1[x]	26.0	28.1	25.0[x]	0.65	0.003	0.044
Stringiness/ fibrousness	34.2	30.5	36.6	36.7	32.8	28.1	39.1	38.9	35.0	1.29		
Stickiness	27.7	27.2	25.4	26.6	23.9	26.7	27.5	30.5	25.7	0.63		
Aftertaste												
Intensity of lamb aftertaste	34.9	33.1	34.1	33.8	32.6	32.3	34.8	34.7	34.7	0.43		
Soapy	8.3	9.8	9.8	9.0	9.2	10.2	8.7	9.2	8.8	0.31		
Metallic/ bloody	21.8	18.0	22.4	18.3	19.6	19.3	18.0	20.7	20.7	0.49		
Fatty/ greasy	16.8	18.3	17.8	16.2	16.2	15.4	16.9	18.2	14.2	0.48		
Dry	11.7	12.2	9.9	12.0	11.6	11.9	14.6	12.7	13.4	0.34	0.009	

Astringent	6.3	8.1	7.3	7.0	8.0	8.0	9.7	8.8	9.3	0.35	0.030

[1]Probability of significance for the main effects of diet, duration and diet x duration tested using the MIXED model ($P < 0.05$)

[a,b,c] within row, different superscripts indicate differences between durations within each diet (i.e. S36 *vs* S54 *vs* S72, SC36 *vs* SC54 *vs* SC72 and C36 *vs* C54 *vs* C72) ($P < 0.05$).

[x,y,z] within row, different superscripts indicate differences among diets with finishing duration 36 or 54 or 72 days (i.e. S36 *vs* SC36 *vs* C36 or S54 *vs* SC54 *vs* C54 or S72 *vs* SC72 *vs* C72) ($P < 0.05$).

Supplementary Table S3 Least square mean values for logarithmically transformed peak areas of aroma compounds detected in the headspace of grilled *longissimus thoracis et lumborum* (LTL) muscle fed three different diets (100% Silage (S); 50% Silage: 50% Concentrate (SC); 100% Concentrate (C)) for three durations of feeding (36, 54, 72 days).

	LRI	100% S			50%S:50%C			100% C			SEM	Significance		
		Feeding Duration			Feeding Duration			Feeding Duration						
		36	54	72	36	54	72	36	54	72		Diet	Duration	Diet × Duration
Sulphur compounds														
Dimethyl sulfide		2.45	1.89	1.57	2.63	3.12	2.95	3.10	2.87	2.14	0.151	0.029		
Dimethyl disulfide	719	2.67	2.39	1.97	2.60	3.55	2.00	1.99	2.73	2.50	0.176			
Dimethyl trisulfide	963	4.33	4.26	4.22	4.28	4.56	4.25	4.14	4.34	4.15	0.037			
Aldehydes														
2-Methylbutanal		4.42	4.13	3.96	4.40	4.22	4.47	4.38	4.48	4.01	0.097			
3-Methylbutanal		4.60	4.37	4.02	4.53	4.36	4.53	4.58	4.67	4.16	0.095			
Pentanal		4.33	4.50	3.95	4.41	4.15	4.39	4.43	4.60	4.06	0.082			
(*E*)-2-Hexenal	849	3.48	2.66	2.91	2.67	2.53	2.76	3.22	2.91	2.78	0.139			
Hexanal	800	5.17	5.31	5.31	5.34	5.47	5.31	5.46	5.50	5.39	0.031	0.044		
Methional	905	4.05	3.93	3.49	4.30	3.94	4.16	4.04	3.93	3.63	0.117			
(*E*,*E*)-2,4-Heptadienal	1008	1.70	2.19	2.71	2.94	2.14	1.03	2.06	2.41	1.25	0.194			
(*Z*)-4-Heptenal	898	4.03	4.12	4.09^{y}	4.05^{ab}	4.12^{b}	3.83^{xya}	4.17^{b}	3.94^{a}	3.78^{xa}	0.029		0.013	0.007
Heptanal	900	5.26	5.40	5.32	5.31	5.43	5.18	5.38	5.37	5.29	0.025			
(*E*)-2-Octenal	1056	4.36	4.43	4.41	4.44	4.54	4.33	4.39	4.55	4.48	0.027			
Octanal	1002	5.36	5.50	5.38	5.40	5.60	5.32	5.48	5.55	5.46	0.024		0.012	
(*E*,*Z*)-2,6-Nonadienal	1150	4.07	4.13	4.11^{y}	4.06^{b}	4.14^{b}	3.81^{xa}	4.03^{b}	3.99^{b}	3.76^{xa}	0.028	0.026	0.007	0.003

(*E*)-2-Nonenal	1158	4.96	5.05	4.96	4.89	4.97	4.71	4.92	4.87	4.80	0.028			
Nonanal	1101	5.99	6.10	5.99	6.00	6.20	5.89	6.03	6.09	5.97	0.023		0.006	
(*E*,*E*)-2,4-Decadienal	1315	3.93	4.02	3.99	4.07	4.12	3.98	3.91	4.13	4.05	0.031			
(*E*)-2-Decenal	1260	4.44	4.57	4.39	4.41	4.53	4.27	4.42	4.50	4.47	0.029			
Decanal	1204	4.77	4.90	4.81	4.84	4.91	4.75	4.84	4.86	4.80	0.021			
Undecanal	1306	3.51	3.52	4.23	3.95	3.98	4.18	3.52	3.97	3.84	0.126			
Dodecanal	1406	4.43	4.48	4.40	4.43	4.52	4.34	4.31	4.46	4.33	0.022		0.044	
Tridecanal	1510	4.51	4.51	4.43	4.47	4.59	4.36	4.40	4.46	4.33	0.024		0.042	
Tetradecanal	1607	4.99	4.98	4.95	4.93	5.02	4.84	4.80	4.92	4.83	0.023			
Pentadecanal	1705	5.11	5.09	5.01	5.06	5.14	4.95	4.90	5.03	4.95	0.026			
Hexadecanal	1818	5.73	5.62	5.66	5.66	5.72	5.57	5.45	5.65	5.56	0.030			
Alcohols														
1-Pentanol	809	3.91^{x}	2.50	3.31	3.38	4.20^{y}	3.60	3.73	4.19^{y}	3.73	0.132			
1-Hexanol	868	4.27	4.42	4.36	4.36	4.46	4.24	4.42	4.44	4.34	0.026			
1-Heptanol	969	4.43	4.54	4.47	4.52	4.63	4.40	4.57	4.63	4.60	0.025			
1-Octen-3-ol	980	4.77	4.79	4.87	4.92	5.03	4.85	4.83	5.02	4.94	0.027			
2-Octen-1-ol	1066	3.93	3.95	4.03	3.93	4.13	3.96	3.94	4.07	4.03	0.024			
2-Ethyl-1-hexanol	1027	4.36	4.47	4.46	3.98	4.50	4.33	4.48	4.46	4.44	0.053			
1-Octanol	1069	5.06	5.20	5.06	5.12	5.27	4.96	5.13	5.18	5.11	0.023		0.008	
α-Terpineol	1191	4.89	4.93	4.92	4.87	4.97	4.83	4.86	4.96	4.89	0.036			
1-Pentadecanol	1766	5.52^{y}	5.53^{y}	5.67^{z}	5.32^{y}	5.45^{y}	5.27^{y}	5.08^{x}	4.97^{x}	4.94^{x}	0.03	<0.001		<0.001
Ketones														
2-Pentanone		0.64	1.18	0.28	1.21	1.51	1.55	1.88	1.65	1.23	0.166			
2,3-Butanedione		2.50	3.43	1.85	2.34	3.15	3.91	4.15	3.48	2.63	0.202			
2-Heptanone	887	3.24^{xa}	3.96^{b}	3.98^{b}	4.01^{y}	4.13	3.98	3.96^{y}	4.11	4.02	0.062			0.029
2-Nonanone	1089	3.84	3.93	3.95	3.99	4.10	3.93	3.95	4.08	3.98	0.027			

γ-Octalactone	1251	0.83	1.44	1.07	1.17	2.53	1.77	1.47	1.65	2.56	0.153			
γ-Nonalactone	1356	2.73	3.43	3.02	2.99	3.13	3.26	3.01	3.11	3.27	0.089			
Terpenes														
p-cymene	1020	2.72	2.45	3.24	2.37	3.38	2.67	2.43	3.35	2.69	0.122			
Limonene	1024	4.24	4.31	4.25	4.29	4.40	4.24	4.21	4.45	4.30	0.029			
Phenols														
p-Cresol	1071	3.84	3.15	2.98	3.27	3.36	2.95	3.27	3.00	2.71	0.171			
Indoles														
Indole	1287	0.27	1.36^{y}	0.28	0.00	0.26^{x}	0.00	0.00	0.00^{x}	0.22	0.086	0.006		0.021
Skatole (3-methyl indole)	1379	0.94	2.00	0.38	0.59	0.65	0.31	0.24	0.50	0.27	0.137	0.048	0.080*	0.063*
Pyrazines														
2-Methyl pyrazine	822	1.21	1.28	0.79	0.80	1.67	0.75	0.80	0.75	0.39	0.183			
2,5-Dimethyl pyrazine	909	3.60^{yb}	2.09^{ab}	1.65^{a}	2.43^{xy}	2.08	1.91	0.79^{x}	0.48	0.78	0.222	0.002		0.022
2,6-Dimethyl pyrazine	909	5.10	3.66	4.07	3.31	5.05	3.96	2.34	3.44	3.51	0.216	0.055*		
2-Ethyl-3,5-dimethyl-pyrazine	1071	3.43	3.13	1.50	1.99	2.25	0.75	1.94	0.38	0.76	0.213			
2-Ethyl-3,6-dimethyl-pyrazine	1083	5.06^{b}	4.56^{yab}	4.97^{a}	4.87	5.09^{y}	4.83	4.55	4.80	4.27^{x}	0.084	0.003	0.012	0.003
Benzenoid compounds														
Benzaldehyde	957	6.20	6.18	6.18	6.21	6.41	6.12	6.05	6.27	6.05	0.034			
Phenyl acetaldehyde	1039	4.98	4.97	4.97	4.92	5.04	4.85	4.79	4.96	4.84	0.033			

Toluene	748	4.69	4.69	4.65	4.77	4.95	4.72	4.69	4.85	4.65	0.038			
Etherocyclic compounds														
2-Pentylfuran	987	3.72	3.79	4.59	3.41	4.79	4.56	3.84	4.79	4.66	0.139		0.010	
Hydrocarbons														
Tridecane		4.48	4.49	4.59	4.59	4.65	4.42	4.40	4.58	4.39	0.028			
Tetradecane		4.57	4.60	4.53	4.53	4.64	4.46	4.36	4.55	4.41	0.025			
Pentadecane		4.89^{y}	4.89^{y}	4.87^{y}	4.81^{y}	4.92^{y}	4.79^{y}	4.62^{x}	4.66^{x}	4.59^{x}	0.023	<0.001		0.000
Hexadecane		4.60^{y}	4.56^{y}	4.56^{y}	4.48^{xy}	4.52^{xy}	4.39^{y}	4.34^{x}	4.39^{x}	4.28^{x}	0.021	<0.001		0.000
Heneicosane		1.03	0.92	0.99	0.53	0.73	0.50	0.00	1.23	0.51	0.122			
BCFAs														
4-Methyl octanoic acid	1232	1.45^{y}	1.95	0.54	2.08^{y}	1.19	1.48	0.00^{x}	1.18	1.11	0.157			0.048
4-Ethyl octanoic acid	1313	2.21	1.20	2.42	2.98	1.91	1.47	1.81	1.16	2.45	0.168			
4-Methyl nonanoic acid	1323	0.98	1.46	1.35	0.81	0.24	1.61	1.15	0.59	0.85	0.149			
Organic acids														
Nonanoic acid	1275	3.66	3.90	3.66	3.38	3.96	3.68	3.70	3.82	3.60	0.051		0.029	

[a,b,c] Within row, means assigned different superscripts indicate differences between durations within each diet (i.e. S36 *vs* S54 *vs* S72, SC36 *vs* SC54 *vs* SC72 and C36 *vs* C54 *vs* C72).

[x,y,z] Within row, means assigned different superscripts indicate differences among diets at finishing durations of 36 or 54 or 72 days (i.e. S36 *vs* SC36 *vs* C36 or S54 *vs* SC54 *vs* C54 or S72 *vs* SC72 *vs* C72).

* P <.1.

742