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Effect of linseed supplementation and slaughter age on meat quality of grazing crossbred Galician x Burguete foals

Running title: Effect of diet and slaughter age on foal meat quality

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ABSTRACT

BACKGROUND: The aim of this study was to assess the effect of finishing diet (control concentrate *vs*. linseed concentrate) and slaughter age (13 vs. 26 months) on meat and nutritional quality of foal meat. For this study, 46 foals from crossing Galicia Mountain x Burguete breeds were used.

RESULTS: The obtained results showed that slaughter age had influence on chemical composition and colour parameters. Foals slaughtered at the age of 13 months had lower content of intramuscular fat and higher cholesterol contents than those slaughtered at 26 months of age. Regarding colour parameters, older foals showed the highest values of redness and lowest myoglobin contents. Finishing diet had a low effect on the majority of parameters evaluated. Foals fed with linseed presented lower shear force values than those fed with control concentrate. Fatty acid and amino acid contents were hardly influenced by finishing diet, whereas slaughter age effect had a high impact on fatty acid profile. Older animals showed lowest SFA values and n-6/n-3 ratio.

CONCLUSION: Older animals presented the best meat quality. Diet had low effect in meat quality and could be related to the short time during finishing diet was administered and the low amount of linseed in the experimental diet.

Keywords: Horsemeat, Linseed, Chemical composition, Fatty acids, Amino acids, Nutritional value

INTRODUCTION

Horsemeat consumption is not comparable to other kind of meats such as beef, chicken or pork, which are more important in the human diet.[1] Despite this fact, horsemeat has become more popular in recent years and it has great potential as an alternative meat.[2] Nowadays, consumers are more health conscious and demand high quality food products.[3] Therefore, horsemeat can be considered a good substitute for traditional meat (chicken, pork, sheep or beef) because of its nutritional characteristics. This meat is characterized by low fat,[1,4] low cholesterol content,[5-7] high iron content and vitamins of B group.[8] This meat also has a favourable dietetic fatty acid profile, with a high content of unsaturated fatty acids, n-3 polyunsaturated fatty acids, and low n-6/n-3 ratio [9-12] and provides a large amount of essential amino acids.[4,7,9]

As other meats, horsemeat properties are influenced by livestock production system [1,7]. Recent studies displayed that horsemeat quality can be influenced by breed and crossbreed,[10,13] finishing diet [4,14] or age/live weight [1,9,14] among others. Therefore, the aim of this work was to study for the first time the influence of finishing diet with linseed (control concentrate *vs.* linseed concentrate) and slaughter age (13 *vs.* 26 months) on chemical composition, physicochemical properties (pH, colour and textural parameters) and nutritional value (fatty acid and amino acid content) of crossbred Galician x Burguete foals meat.

MATERIALS AND METHODS

Experimental design and animal management

The experiment took place in farm Marco da Curra, A Coruña (Galicia - north-west of Spain), located at 650 m of altitude, in a hard winter weather. A herd of mix-breed Galician Mountain (GM) mares were crossed with a Burguete (BU) stallion. Mating took place naturally in the field. For this study, forty-six foals (16 males and 30 females), from crossing GM×BU, were used. Births took place from April to July in two different years, 2013 and 2014. In order to carry out the experiment, foals were distributed in two groups: twenty-four foals, 10 males and 14 females born

in 2013 (group 1; 26 months old foals; 784±17 days old at slaughter; 443±62 kg of live weight; 242±24 kg of carcass weight) and twenty-two foals, 6 males and 16 females born in 2014 (group 2; 13 months old foals; 403±19 days old at slaughter; 339±53 kg of live weight; 179±26 kg of carcass weight).

All foals were reared with their mothers and were allowed to suck freely at pasture. They were weaned at 6-7 months-old and after weaning foals were fed alone on pasture following a rotational grazing both in seeded and natural fields, being pasture the main part of the diet. The herd was changed from a plot to another when the height of pasture was approximately 10 cm in the spring and 5 cm in the winter. The vegetation was composed by seeded (*Lolium perenne* and *Trifolium repens*) and natural fields (*Agrostis* spp., *Lotus corniculatus*, *Holcus lanatus*, *Bromus mollis*, *Pseudoarrenatherum longifolium*, etc.). All foals were finished at pasture and during a period of 104 days (on average) were fed with two types of supplementation. Twelve foals (5 males and 7 females) from 26 months old foals and 11 from 13 months old foals (3 males and 8 females) were supplemented on pasture with 2 kg of conventional concentrate per foal/day. Twelve foals (5 males and 7 females) from 26 months old foals and 11 foals (3 males and 8 females) from 13 months old foals were supplemented on pasture with 2 kg of linseed-rich concentrate per foal/day. Table 1 shows the chemical composition of the supplementary concentrates. Supplementation was increased gradually from 300 g per foal/d to 2 kg in order to avoid colics that usually appear with a sudden change in the diet, but foals quickly adapted to supplementation and only in ten days they were eating 2 kg.

Slaughter of the animals

Foals were slaughtered and dressed in an authorized abattoir, 50 km away from the farm, in a commercial plant. The foals were stunned in the frontal region with a captive-bolt, according to current EU regulations.[15] The *longissimus thoracis* (LT) muscle was cut into five 2.5 cm thick steaks. The first three steaks were used to determine pH, colour, proximate composition, cholesterol

and fatty acid and amino acid profile. The fourth and fifth steaks were used to determine water holding capacity and texture parameter.

Chemical composition and physicochemical analysis

The pH, chemical composition, colour, and the relative content of myoglobin (MYO), metmyoglobin (MET) and oximyoglobin (OX) were measured according to Pateiro *et al.*[16] Water holding capacity (calculated as cooking loss) and Warner Bratzler test were also performed following procedure described by Pateiro *et al.*[16] All the parameters were measured at 24 h *post-mortem*.

Analysis of cholesterol

For determination of total cholesterol, saponification, extraction and identification were performed in following the procedure described by Domínguez *et al.*[9] The content of total cholesterol in foal meat was calculated, in duplicate for each muscle sample, based on the external standard technique, from a standard curve of peak area *vs.* concentration.

Analysis of fatty acid methyl esters (FAME)

Total fat was extracted from 10 g of ground meat sample, according to Bligh and Dyer method.[17] Total fatty acids were quantified according to Domínguez *et al.*[9] procedure. Fifty milligrams of fat was used to determine the fatty acid profile. For the fatty acids transesterification, 4ml of a sodium methoxide (2%) solution was added to the fat samples, vortexed every 5min during the 15min at room temperature, then 4ml of a H_2SO4 solution (in methanol at 33%) was added, vortexed for a few seconds and vortexed again before adding 2ml of distilled water. The organic phase (containing fatty acid methyl esters) was extracted with 2.5ml of hexane.

Separation and quantification of the FAMEs was carried out using a gas chromatograph (GC-Agilent 6890N; Agilent Technologies Spain, S.L., Madrid, Spain), equipped with a flame ionization detector and an automatic sample injector HP 7683, and using a Supelco SPTM-2560 fused silica

capillary column (100m, 0.25mmi.d., 0.2µm film thickness; Supelco Inc., Bellafonte, PA, USA), following the chromatographic conditions described by Domínguez *et al.*[9]

Individual FAMEs were identified by comparing their retention times with those of authenticated standards, and the results were expressed as g/ 100g of total fatty acids identified. The atherogenic index (AI) and thrombogenic index (TI) were calculated according to Ulbricht and Southgate,[18] whereas the hypocholesterolemic/Hypercholesterolemic ratio (h/H) was calculated according to Santos-Silva *et al*.[19]

Protein amino acid profile

The hydrolysis of the protein, derivatization, and identification of hydrolysed was carried out following the procedure described by Domínguez *et al*.[20]

Statistical analysis

A total of 46 foals were analysed for different parameters. The effect of sex was not taken into account after verifying that did not significantly affect any of the parameters studied. After that, the effect of diet, slaughter age and the interaction between diet x slaughter age on chemical composition, physicochemical parameters, cholesterol, fatty acids and amino acids content was examined using analysis of variance (ANOVA) with the General Lineal Model (GLM) procedure, where these parameters was set as dependent variables and sex, finishing diet and slaughter age as fixed effect. Correlations between variables were determined by Pearson's linear correlation coefficient ($P<0.05$). The values were given in terms of mean values and standard error of the mean (SEM). All statistical analysis was performed using IBM SPSS Statistics 19 software.[21]

RESULTS

Chemical composition and physicochemical properties

The effect of finishing diet and slaughter age on chemical composition and physicochemical parameters of foal meat are shown in Table 2. The interaction between the main effects (D x A) had effect on six of the studied parameters. The pH values were significantly affected by finishing

feeding, being within an acceptable range (below 6). Regarding chemical composition, the slaughter age affected moisture, intramuscular fat (IMF) and cholesterol content, while feeding affected the ash amount. The mean moisture content of foals showed values close to 73%, significantly higher (P<0.001) in animals slaughtered at earliest age (74.2 *vs.* 72.4%). Significant differences (P<0.001) were also found in IMF content, since the highest values were observed in oldest animals (1.72 *vs.* 0.56%). Cholesterol contents also displayed significant differences (P<0.001) between slaughter ages, showing the highest values in animals slaughter at 13 months old (50.9 *vs.* 39.6 mg/100 g wet tissue). On the other hand, finishing diet affected only ashes content, with lower values in meat from foals fed with linseed-rich concentrate (1.23 *vs.* 1.32%, P<0.05).

Regarding colour parameters, redness (a*) and chroma (C*) were significantly (P<0.01) affected by the both studied factors, lightness (L*) and hue (h*) were significantly (P<0.001) influenced by slaughter age, and yellowness (b*) was significantly affected by finishing diet (P<0.01). The values found for a* were higher in oldest animals (13.5 vs. 11.7, P<0.001) and fed with linseed-rich concentrate (13.3 vs. 11.9, P<0.001).

Table 2 also shows the results found for pigment forms. Myoglobin was significantly affected by the both studied factors (P<0.05), with highest values at earliest age (41.7 vs. 33.4%) and in animals fed with control concentrate (39.9 vs. 35.7%). In addition, it was found that myoglobin and IMF content were correlated to a* values (r = -0.820 and r = 0.458, P<0.01, respectively) and metmyoglobin with C* and h* (r = 0.355 and r = -0.932, P<0.05, respectively).

On the other hand, statistical analysis showed significant differences (P<0.05) on cooking loss by D x A effect and finishing diet. Animals fed with control concentrate showed the highest values (23.0 vs. 21.5%). With regards to texture, finishing feeding presented a significant effect (P<0.01) on the evaluated parameters. The lowest shear force values were observed in animals fed with linseed-rich concentrate (33.0 vs. 41.8 N). Finally, cooking loss was positively correlated to shear force (r = 0.362, P<0.05).

Fatty acid profile

The effect of finishing diet and slaughter age on fatty acid composition of foal meat is presented in Table 3. There were no interaction between finishing feeding and slaughter age. The finishing diet had a slight effect on intramuscular fatty acid profile. Animals fed with control concentrate showed that saturated fatty acids (SFA) were the predominant fatty acids (34.74 g/100 g of FAME), followed by monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) with similar values (around 32 g/100 g of FAME). In contrast, the major fatty acids in animals fed with linseed were the MUFA (35.79 g/100 g of FAME), followed by SFA (33.75 g/100 g of FAME) and PUFA (30.47 g/100 g of FAME). Only a few significant differences were found in fatty acids between finishing diets (7 out of 28 fatty acids). The linseed group compared to animals finished with control concentrate, had significantly ($P<0.05$) higher amounts of C18:1n-9 and C20:1n-9, and significantly ($P<0.05$) lower contents of C16:0, C20:3n-6, C20:5n-3, C22:5n-3 and C22:6n-3. These differences made the content of SFA be greater in animals from control concentrate than from treatment with linseed (34.78 vs. 33.79 g/100 g of FAME), while MUFA did not show differences between treatments. With regard to PUFA concentration, finishing diet did not significantly affect its contents ($P=0.210$). In this way, total n-6 and n-3 PUFA were not showed significant differences between diets. The contents of n-6 PUFA were higher in foals fed with control concentrate, and n-3 PUFA content was similar between diets (around 14 g/100 g of FAME). Finally, regarding nutritional indices, the finishing diet only affect the h/H ratio.

On the other hand, the slaughter age affected the fatty acid profile. In 13 months-old animals, PUFA were the most abundant fatty acids, followed by SFA and MUFA. By contrast, in foals slaughtered at 26 months the prevalence fatty acids were the MUFA, followed by SFA and PUFA. Twenty-three out of 28 fatty acids were influenced by slaughter age. The concentration of SFA and PUFA decreased as slaughter age increased ($r= -0.60$ and $r= -0.61$ $P<0.01$, respectively). Nevertheless, MUFA amount increased from 28.75 g/100 g of FAME in animals slaughtered at 13

months to 39.30 g/100 g of FAME in foals slaughtered at 26 months old. The main difference in SFA was related to the higher C18:0 content in young animals compared to older foals. The higher MUFA amount in 26 months old foals could be explained by their higher (P<0.001) C16:1n-7 and C18:1n-9 amounts. In contrast, the higher PUFA content in 13 months old animals was mainly due to the higher of n-6 PUFA amounts, while n-3 PUFA content did not present significant differences between slaughter age groups. Young foals presented significant (P<0.001) higher C18:2n-6, C18:3n-6, C20:2n-6, C20:3n-6 and C20:4n-6 amounts than those obtained from old animals. Despite the above differences, in nutritional indices only the n-6/n-3 ratio displayed significant (P<0.001) differences, with higher values in young compared with older animals. Finally, the AI, TI indexes and h/H ratio were not affected by slaughter age.

Amino acid profile

The effect of finishing diet and slaughter age on amino acid content of foal meat is summarized in Table 4. Only glycine show a significant interaction between finishing feeding x slaughter age (P<0.05). All samples exhibited the following profile: the major amino acid was glutamic acid (around 3.50 g/100 g of meat) followed by aspartic acid and lysine with similar values (around 2.10 g/100 g of meat) and leucine (around 1.95 g/100 g of meat). The essential amino acids that presented the highest concentration were lysine, leucine and arginine, representing together about 50% of total essential amino acids, while the methionine and cysteine showed the lowest essential amino acids values (between 0.10 and 0.20 g/100 g of meat). Glutamic acid, aspartic acid and alanine were the most abundant amino acids found in the non-essential fraction, representing together around 67% of the total non-essential amino acids, whereas the lowest values were found out in tyrosine (around 0.65 g/100 g of meat) and serine (around 0.80 g/100 g of meat), each representing 7% of total non-essential amino acids. Finally, the values of essential/non-essential ratio ranged from 1.11 to 1.14.

Only a few significant differences were found in amino acids contents between finishing diets and slaughter age effects. The finishing diet affected only one out of 17 amino acids. Animals finished with control concentrate presented the lowest total non-essential amino acids values ($P<0.05$). Foals fed with conventional concentrate presented lower values ($P<0.05$) of glycine than those fed with linseed enriched-concentrate. On the other hand, the total essential amino acids did not show significant differences between diets. Regarding the effect of slaughter age, only 2 out of 17 amino acids presented significant differences between groups, with young animals showing higher ($P<0.05$) tyrosine and phenylalanine amounts than the animals slaughtered at 26 months.

DISCUSSION

Chemical composition and physicochemical properties

Recent studies noticed that horsemeat quality can be influenced by several factors as age [2] and finishing diet.[4] According to the aforementioned authors, slaughter age is one of the factors that influenced IMF content and therefore, the quality of foal meat. In this study, IMF values were lower than those obtained by other authors in horsemeat,[12-14] who found values in most cases above 2.5%. This is probably due to use only a supplementation of 2 kg as finishing diet, while the studies mentioned included indoors fattening periods with *ad libitum* supply. However, these outcomes are in agreement with those reported by Sarriés and Beriain[14] who found higher IMF content in oldest foals. On the other hand, the mean moisture contents were higher than those observed in foals slaughter at 15–24 months of age with mean values close to 72%,[13,14,22] and lower than those found by other authors in young foals.[1] As expected, moisture was negatively correlated with IMF contents ($r = -0.893$, $P<0.01$), since the highest values of IMF were found in oldest animals with lowest moisture contents. Regarding protein and ash contents, both percentages were similar to those noticed in different researches.[6,13] The cholesterol contents were slightly lower than the values found in the literature (55-64 mg/100 g).[2] Significant differences ($P<0.01$) found between slaughter ages could be related to IMF contents. IMF is one of the parameters that can affect the amount of

cholesterol, in fact both parameters were negatively correlated (r = -0.517, P<0.01). Thereby, samples with high IMF contents present proportionately less membrane polar lipids and therefore lower amounts of the cholesterol associated with these membranes.[23]

The values of pH were similar to those reported by other authors in horsemeat.[1,10,13,14] Regarding colour parameters, the L* values decreased with slaughter age which contrast with the results found by in other studies.[14] Horsemeat is characterized by high myoglobin content and its high ability to combine with oxygen, which impairs red colour stability get reduced.[8] Roth *et al.* [24] stated that the L* value is affected by pigment concentration, whereas a* value is affected by oxidation state of pigment (reduced, oxygenated and oxidized) and to a lesser extent by pigment concentration. These results are in agreement with those reported by Sarriés and Beriain who observed that a* values increased with the age of animal.[14] This fact could be related to old animals were grazing for a longer period and they had more physical activity. Slaughter age also had high influence on C* and metmyoglobin displaying the same behaviour like a*. Older animals presented the highest values for these parameters. On the other hand, cooking losses and tenderness are considered important quality indicators and they have an impact on consumer acceptability.[25] In contrast to other studies in foal meat,[1,10,22] feeding showed significant differences on cooking loss. Our values were slightly higher than those found by other authors in foals slaughtered at 15 months,[4,10] but lower than those reported by Lanza *et al.*[22] According to tenderness classification proposed by Belew *et al*,[26] meat from animals fed with linseed would be classified as "tender" (31.4 N < WB shear force < 38.3 N), while meat from foals fed with control concentrate could be considered as "intermediate" (38.3 N < WB shear force < 45.1 N). Texture parameters confirmed that horsemeat is a tender meat compared with other species.[2] No significant effects of slaughter age were found on shear force values. This finding is in agreement with the results noticed by Franco *et al.* [1] who did not find any significant differences between meat from 9 to 12 months old foals (26.2 *vs*. 27.3 N).

Fatty acid profile

The content and composition of specific fatty acids in meats are important factors to assess its nutritional quality. Several studies have shown that animal diet influences the fatty acid composition of meat. Thus, the supplementation of animal diets with PUFA would be an effective approach to promote the enrichment of meat with n-3 PUFA. In linseed group, the main fatty acid were MUFA, whereas foals finished with control concentrate displayed that SFA concentration was the predominant, which agrees with data previously published by other authors in horse [22,27] and in donkey meat.[28] In contrast, Lorenzo *et al.* [6,7] noticed PUFA as the main fatty acids in foals fed only with pasture. The fact that linseed group presented higher amounts of MUFA could be related with the high amounts of C18:1n-9 in the linseed experimental diet (Table 1).

The effect of feeding with linseed has been studied by several authors in beef and lamb meat,[29,30] however there are no information about feeding with linseed on horsemeat fatty acids. These authors found increased proportions of C18:3n-3, C20:5n-3 and C22:5n-3 in animals fed with linseed diet. As the same way, Andrés *et al.* [29] and Urrutia *et al.* [31] also noticed that lambs fed with linseed diet increases the C18:3n-3 and n-3 PUFA contents. In addition, the highest n-3 PUFA content in the other studies in animals fed with linseed diet was associated with the increase of C18:3n-3 content.[32] However, in our study, there was not a significant difference in C18:3n-3 content between diets, but the differences were significant ($P<0.05$) for C20:5n-3, C22:5n-3 and C22:6n-3, being higher in animals fed with linseed diet. However, the linseed concentration (Table 1) could be not high enough to find differences in fatty acid profile. Taking in mind that the daily dry matter intake of a horse ranges between 1.5-3% of body weight,[33] the 2 kg of supplementation represented a little part of the diet, which was not enough to produce differences in C18:3n-3 content. Another possible explanation to the low effect of diet in fatty acids could be that the short time during finishing diet was administered was not enough long (last 3.5 months) and the low amount of linseed (5%) in the concentrate to produce changes in fatty acids profile. Moreover, the

main part of diet was pasture, being the same for both groups, and being also a rich natural supply of C18:3n-3. It would probably be easier to find differences in intensive feeding systems with *ad libitum* supply. On the other hand, the lowest values of C18:2n-6, n-6 PUFA and the highest values of MUFA in linseed group were consistent with the results obtained by Urrutia *et al.* [31] in lamb meat fed with linseed.

As commented above, the slaughter age had high influence on fatty acid profile. Old animals presented the lowest amounts of SFA and PUFA and the highest of MUFA. These outcomes are in agreement with those reported by Sarriés *et al,*[34] in foals slaughtered at 16 or 24 months. Moreover, De Palo *et al.* [27] also observed that animals slaughtered at 18 months presented higher amounts of MUFA than animals slaughtered at 11 months. Our results were mainly due to the differences in C18:0, C16:1n7 and C18:1n9. According to Lorenzo *et al.* [2] and Sarriés *et al.* [34], the differences between these fatty acid amounts could be linked to a higher desaturase activity in old foals. In contrast, Domínguez *et al*.,[9] Lorenzo *et al.* [11] and Polidori *et al.* [28] did not find differences in SFA, MUFA and PUFA amounts between foals slaughtered at 8 and 12 months. This seems to confirm that only 3.5 months is not sufficient to observe differences in the fatty acid profile of horsemeat.

The lower amounts of PUFA in older than in young animals were due to the lower contents of total n-6 PUFA (mainly C18:2n-6 and C20:4n-6 content). These results are in agreement with data previously published by Sarriés *et al*.[34] The decreased proportion of C18:2n-6 in old foals may support the hypothesis by Kazala *et al.*,[35] who assumed that the majority of this fatty acid is presented in the membrane phospholipids, which may be diluted with the increasing triacylglycerol arising from increasing total lipid in the tissue. The contents of C20:3n-3, C20:5n-3, C22:5n-3 and C22:6n-3 decrease with slaughter age, whereas C18:3n-3 and total n-3 PUFA did not vary according to slaughter age. The differences in long-chain n-3 PUFA could be related to a difference in the "elongase" activity as increase slaughter age. Lush pastures have high C18:3n-3 proportion (50-75%) and its content in horse tissues are directly related to the dietary intake of the animal.[36]

Therefore, the fact that C18:3n-3 content did not decrease in foal tissue as increase the slaughter age could be due to the fact that pasture was the main part of the diet in animals from both groups. The same behaviour was observed in foals slaughtered at 9 and 15 months,[6,11] where C18:3n-3 and total n-3 PUFA did not vary according to slaughter age. The values of C18:3n-3 found by these authors increase in foal tissue as increase the slaughter age (8.90 *vs.* 13.15 g/100 g of FAME).[6,11] In contrast with our results, Domínguez *et al.* [9] and Polidori *et al.* [28] found that foals slaughtered at 12 months had higher C18:2n-6 and lower C18:3n-3 contents than animals slaughtered at 8 months of age.

Finally, regarding the nutritional indices, only the n-6/n-3 ratio was affected by slaughter age, with lower n-6/n-3 ratio in older animals. Despite these differences, all groups of animals were within the nutritional recommendations for human diet (n-6/n-3 <4), according to the UK Department of Health.[37] Our results (1-1.43) were similar to those previously reported by Franco and Lorenzo,[4] Lorenzo *et al.* [6] and Franco *et al.*[10] In contrast, higher values were reported in literature (4.09 to 6.69;[22] 4.34 to 4.54;[27] 8.0 to 15.56[34]). The AI, TI and h/H values obtained in the present study showed a better fatty acid profile (from nutritional point of view) than those previously reported by other authors in foal meat.[11,34]

Amino acid profile

The amino acid profile obtained in this research agree with those previously reported by other authors in foal meat.[4,5,28] The results showed that the inclusion of linseed in the diet and the increase in slaughter age had very low effect in the amino acid contents. In the same line, Domínguez *et al.* [9] and Polidori *et al.* [28] also did not find high differences in amino acid content between animals slaughtered at 8 or 12 months. In addition, Lorenzo *et al.* [7] also found no differences in amino acids content between foals fed with different amounts of concentrate during the finishing diet. Arginine was included in the essential amino acids profile according to Hoffman *et al.*[38]. Finally, the values of essential/non-essential ratio (1.11-1.14) agree with those previously reported by other authors.[5-8]

However, Franco and Lorenzo,[4] Badiani *et al.* [8] and Franco *et al.* [10] found lower values (0.8-0.9) than those obtained in the present study.

CONCLUSIONS

The obtained results show that slaughter age had a great influence on meat quality of foals, while finishing diet had a minor effect on the majority of parameters evaluated. In spite of the difference between quality parameters values, fatty acids profile from animals slaughtered at both ages (13 and 26 months) are in agreement with the range of values proposed by the UK Department of Health. On the other hand, the fact that diet had low effect in meat quality could be related to the short time during finishing diet was administered and the low amount of linseed in the experimental diet.

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Table 1. Composition of supplementary concentrates

	Finishing diet	
	Control	**Linseed**
Chemical composition (%)		
Dry matter	86.9	89.0
Fat	3.29	4.78
Protein	12.4	13.0
Ash	4.99	5.44
Crude fibre	5.1	6.6
Fatty acids (%)		
C14:0	0.18	0.30
C16:0	15.78	14.38
C16:1n-7	0.18	0.14
C18:0	2.10	2.36
C18:1n-9	31.28	33.66
C18:2n-6	45.02	38.61
C20:0	0.39	0.27
C18:3n-3	2.99	8.41
C22:0	0.24	0.15
C24:0	0.17	0.17
SFA	18.86	17.63
MUFA	31.46	33.80
PUFA	48.01	47.02

Control diet: oat flour (30.03%); cornmeal (15.47%); barley flour (30.10%); soybean meal (9.99%); bran (7.95%); glycerol (3.96%); vitamins + salt + calcium carbonate + calcium phosphate (2.52%)
Linseed diet: oat flour (45.95%); cornmeal (14.54%); barley flour (13.07%); soybean meal (9.06%); bran (5.92%); linseed (5.01%); glycerol (3.96%); vitamins + salt + calcium carbonate + calcium phosphate (2.50%)

Table 2. Effect of finishing diet and slaughter age on chemical composition, colour parameters, water holding capacity and texture parameters of *longissimus thoracis*

	Control		Linseed		SEM	P-value		
	13 months (n=11)	26 months (n=12)	13 months (n=11)	26 months (n=12)		D	A	DxA
Chemical composition								
pH	5.61^{ab}	5.66^{b}	5.56^{a}	5.58^{a}	0.01	0.005	0.135	0.525
Moisture	74.5^{b}	72.6^{a}	73.9^{b}	72.1^{a}	0.21	0.113	<0.001	0.791
Intramuscular fat	0.37^{a}	1.82^{b}	0.74^{a}	1.61^{b}	0.13	0.642	<0.001	0.111
Protein	22.5	22.7	22.6	23.2	0.14	0.363	0.128	0.449
Ash	1.26^{ab}	1.38^{c}	1.30^{bc}	1.17^{a}	0.02	0.010	0.970	<0.001
Cholesterol (mg/100g wet tissue)	50.7^{b}	42.8^{a}	51.1^{b}	36.3^{a}	1.43	0.192	<0.001	0.134
Colour parameters								
Lightness (L*)	38.6^{d}	34.4^{a}	37.1^{c}	35.8^{b}	0.30	0.857	<0.001	<0.001
Redness (a*)	11.2^{a}	12.6^{b}	12.2^{b}	14.3^{c}	0.23	<0.001	<0.001	0.282
Yellowness (b*)	10.6^{b}	9.09^{a}	10.4^{b}	10.9^{b}	0.17	0.006	0.107	0.001
Chroma (C*)	15.4^{a}	15.6^{a}	16.0^{a}	18.0^{b}	0.25	<0.001	0.010	0.025
Hue (h*)	43.4^{d}	35.7^{a}	40.4^{c}	37.3^{b}	0.51	0.168	<0.001	<0.001
Pigment form								
Myoglobin (%)	42.4^{b}	37.4^{b}	40.9^{b}	30.5^{a}	1.16	0.035	<0.001	0.165
Metmyoglobin (%)	14.1^{a}	19.8^{c}	15.6^{b}	19.8^{c}	0.45	0.177	<0.001	0.137
Oxymioglobin (%)	43.5^{a}	42.8^{a}	43.5^{a}	49.8^{b}	0.99	0.065	0.136	0.066
WHC								
Cooking loss (%)	23.3^{b}	22.7^{b}	20.2^{a}	22.7^{b}	0.38	0.030	0.178	0.032
Texture parameters								
Firmness (N s^{-1})	10.8^{b}	11.3^{b}	7.65^{a}	9.50^{ab}	0.52	0.006	0.174	0.435
Total work (N mm)	179^{b}	228^{c}	144^{a}	196^{b}	7.12	0.004	<0.001	0.892
Shear force (N)	43.2^{b}	40.5^{b}	29.3^{a}	36.7^{ab}	1.60	0.004	0.416	0.091

SEM: standard error of the mean

D: finishing diet; A: slaughter age; D x A: finishing diet x slaughter age

$^{a-d}$ Means in the same row with different letters differ significantly (P<0.05)

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457 **Table 3.** Effect of finishing diet and slaughter age on fatty acid profile (g/100 g of total fatty acids) of *longissimus thoracis*

	Finishing diet		Slaughter age		SEM	P-value		
	Control (n=23)	Linseed (n=23)	13 months (n=22)	26 months (n=24)		*D*	*A*	*DxA*
Fatty acids								
C10:0	0.05	0.05	0.06	0.05	0.01	0.845	0.139	0.291
C12:0	0.18	0.17	0.25	0.11	0.02	0.575	<0.001	0.868
C14:0	2.13	2.14	1.95	2.31	0.07	0.966	0.011	0.828
C14:1n-5	0.49	0.45	0.48	0.45	0.02	0.217	0.286	0.381
C15:0	0.17	0.16	0.18	0.15	0.00	0.161	<0.001	0.832
C16:0	25.5	24.5	24.8	25.1	0.18	0.013	0.368	0.470
C16:1n-7	4.10	3.96	2.78	5.18	0.29	0.793	<0.001	0.568
C17:0	0.34	0.35	0.38	0.31	0.01	0.831	<0.001	0.872
C17:1n-7	0.44	0.49	0.47	0.47	0.01	0.053	0.997	0.889
C18:0	6.12	6.09	7.21	5.10	0.21	0.896	<0.001	0.825
9*t*-C18:1	0.08	0.08	0.07	0.09	0.00	0.150	<0.001	0.458
11*t*-C18:1	0.04	0.04	0.05	0.03	0.00	0.591	<0.001	0.834
C18:1n-9	25.5	28.6	23.0	30.8	0.89	0.018	<0.001	0.348
C18:1n-7	1.71	1.70	1.61	1.80	0.03	0.744	0.002	0.936
C18:2n-6	15.9	14.1	18.4	12.0	0.73	0.102	<0.001	0.426
C20:0	0.09	0.08	0.11	0.07	0.01	0.329	0.001	0.871
C18:3n-6	0.13	0.12	0.14	0.11	0.00	0.153	<0.001	0.285
C20:1n-9	0.29	0.33	0.27	0.35	0.01	0.025	<0.001	0.890
C18:3n-3	11.1	11.8	11.1	11.9	0.35	0.337	0.273	0.560
C20:2n-6	0.28	0.26	0.32	0.23	0.01	0.330	<0.001	0.141
C22:0	0.05	0.04	0.08	0.01	0.01	0.281	<0.001	0.065
C20:3n-6	0.44	0.36	0.52	0.28	0.03	0.017	<0.001	0.375
C20:3n-3	0.52	0.49	0.57	0.45	0.02	0.205	<0.001	0.428
C20:4n-6	1.48	1.20	1.73	0.99	0.09	0.042	<0.001	0.387
C23:0	0.11	0.10	0.13	0.08	0.01	0.227	<0.001	0.871
C20:5n-3	0.73	0.58	0.97	0.37	0.06	0.038	<0.001	0.199

C22:5n-3	1.48	1.21	1.81	0.92	0.09	0.028	<0.001	0.551
C22:6n-3	0.38	0.29	0.48	0.21	0.03	0.011	<0.001	0.229
SFA	34.7	33.8	35.2	33.4	0.23	0.007	<0.001	0.554
MUFA	32.7	35.8	28.8	39.3	1.16	0.070	<0.001	0.394
PUFA	32.5	30.5	36.0	27.3	1.06	0.210	<0.001	0.331
n-3	14.2	14.4	14.9	13.8	0.36	0.904	0.123	0.415
n-6	18.3	16.1	21.1	13.6	0.85	0.087	<0.001	0.408
Nutritional indices								
n-6/n-3	1.28	1.13	1.43	1.00	0.05	0.101	<0.001	0.900
AI	0.52	0.50	0.51	0.52	0.01	0.121	0.444	0.596
TI	0.49	0.47	0.48	0.48	0.01	0.223	0.830	0.306
h/H	2.15	2.26	2.24	2.18	0.03	0.049	0.284	0.522

SEM: standard error of the mean
D: finishing diet, A: slaughter age, D x A: finishing diet x slaughter age
SFA: Saturated fatty acids =∑ (C10:0+ C12:0+ C14:0+ C15:0+ C16:0+ C17:0+ C18:0+ C20:0+ C22:0+ C23:0)
MUFA: Monounsaturated fatty acids =∑ (C14:1n-5+ C16:1n-7+ C17:1n-7+ 9*t*-C18:1+ 7*t*-C18:1+ C18:1n-9+ C18:1n-7+ C20:1n-9)
PUFA: Polyunsaturated fatty acids =∑ (C18:2n-6+ C18:3n-6+ C18:3n-3+ C20:2n-6+ C20:3n-6+ C20:3n-3+ C20:4n-6+ C20:5n-3+ C22:5n-3+ C22:6n-3)
h/H: ratio hypocholesterolemic/hypercholesterolemic fatty acids =[Σ(C18:1n-9, C18:1n-7, C18:2n-6, C18:3n-3, C20:3n-6, C20:4n-6)/Σ (C14:0 and C16:0)]
AI: Atherogenic index = [C12:0+ (4*C14:0)+ C16:0]/[(∑MUFA)+ (∑PUFA)]
IT: Thrombogenic index = [C14:0+ C16:0+ C18:0] / [(0.5*∑MUFA)+ (0.5*n-6)+ (3*n-3)+ (n-3/n-6)]

Table 4. Effect of finishing diet and slaughter age on amino acid content (g/100 g meat) of *longissimus thoracis*

	Finishing diet		Slaughter age		SEM	P-value		
	Control (n=23)	Linseed (n=23)	13 months (n=22)	26 months (n=24)		*D*	*A*	*DxA*
Amino acids								
Non-essential								
Aspartic acid	2.08	2.18	2.15	2.11	0.03	0.113	0.499	0.573
Serine	0.80	0.81	0.83	0.77	0.02	0.885	0.070	0.677
Glutamic acid	3.38	3.57	3.52	3.44	0.05	0.069	0.439	0.459
Glycine	0.93	1.02	1.02	0.94	0.01	0.025	0.062	0.026
Alanine	1.26	1.34	1.34	1.27	0.02	0.058	0.113	0.051
Proline	0.85	1.01	0.95	0.91	0.05	0.116	0.662	0.818
Tyrosine	0.67	0.67	0.71	0.63	0.02	0.972	0.015	0.651
Total non-essential	9.97	10.6	10.5	10.1	0.14	0.037	0.132	0.251
Essential								
Histidine	1.08	1.14	1.13	1.10	0.02	0.115	0.366	0.103
Arginine	1.77	1.85	1.87	1.75	0.04	0.265	0.124	0.025
Threonine	0.99	1.04	1.05	0.99	0.03	0.277	0.153	0.052
Cysteine	0.12	0.14	0.15	0.11	0.01	0.315	0.078	0.386
Valine	1.12	1.15	1.17	1.11	0.02	0.249	0.062	0.109
Methionine	0.17	0.20	0.21	0.17	0.01	0.315	0.112	0.627
Lysine	2.06	2.14	2.10	2.10	0.03	0.184	0.986	0.977
Isoleucine	1.12	1.14	1.15	1.11	0.02	0.595	0.234	0.524
Leucine	1.94	1.99	1.98	1.94	0.03	0.370	0.444	0.665
Phenylalanine	0.95	1.00	1.04	0.92	0.03	0.177	0.010	0.096
Total essential	11.3	11.8	11.9	11.3	0.16	0.153	0.101	0.122
Essential/Non-essential ratio	1.14	1.11	1.13	1.12	0.01	0.096	0.627	0.359

SEM: standard error of the mean
D: finishing diet
A: slaughter age
D x A: finishing diet x slaughter age

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