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Effect of feeding on the composition of *longissmus* muscle of Hungarian Grey and Holstein Friesian bulls

III. Amino acid composition and mineral content

Abstract

In this experiment the effect of the extensive diet (grass/grass silage and concentrate linseed supplemented) *versus* intensive diet (maize silage and concentrate) on the amino acid composition and mineral content of Hungarian Grey and Hungarian Holstein Friesian young bulls was analysed. In addition the relationships between fatty acid profile and amino acid composition as well as mineral content of *longissimus* muscle were investigated. The extensive diet caused some changes in the proportion of some amino acids including isoleucine, leucine, phenylalanine, threonine, valine, cysteine, glycine, proline, serine and ammonia. The breed influenced only the histidine concentration of *longissimus* muscle. In extensive groups arginine to glycine and arginine to leucine ratios were higher compared to intensive fed bulls. The mineral composition of *longissimus* muscle was mainly influenced by diet and there was a breed x diet interaction, whereas breed differences were detected for calcium (Ca) and sodium (Na). The phosphorus (P) and the iron (Fe) content in *longissimus* muscle of extensively fed animals were significantly higher, whereas Na and copper (Cu) content were lower. The beef of Hungarian Grey bulls contained less Ca and Na compared to Holstein counterparts. Cysteine and leucine concentration were positively correlated to intramuscular fat content and negatively with *n*-3 and *n*-6 fatty acids (FA). Among minerals the P and Fe content correlated negatively to intramuscular fat content. The closest correlations were determined between P and linolenic acid (C18:3*n*-3) as well as *n*-3 fatty acids (r=0.81). The lower *n*-6 and *n*-3 fatty acid ratio in extensive groups occured simultaneously with higher proline, valine, phenylalanine, P, and with lower glycine, cystine, leucine and sodium proportion in *longissimus* muscle.

Key Words: amino acids, minerals, beef, Hungarian Grey bulls, Holstein Friesian bulls

Zusammenfassung

Titel der Arbeit: **Der Einfluss der Fütterung auf die Zusammensetzung des *Musculus longissimus dorsi* von Jungbullen der Rassen Ungarisches Grauvieh und Holstein Friesian. 3. Mitt. Aminosäurezusammensetzung und Mineralstoffgehalt**

In diesem Versuch wurde die Wirkung einer extensiven Fütterung (Gras/Grassilage und ein Konzentrat mit einem Zusatz von Leinsamenschrot) und einer intensiven Fütterung (Maissilage und Konzentrat) auf den Aminosäure- und Mineralstoffgehalt von Ungarischem Grauvieh and Ungarischen Holstein Jungbullen analysiert. Außerdem wurde der Zusammenhang zwischen Fettsäurezusammensetzung und Aminosäurezusammensetzung sowie dem Mineralstoffgehalt des *Musculus longissimus dorsi* festgestellt. Die extensive Fütterung verursachte Änderungen im Aminosäureanteil einschließlich Isoleucin, Leucin, Phenylalanin, Threonin, Valin, Cystein, Glycin, Prolin, Serin und Ammoniak. Die Rasse beeinflusste nur die Konzentration des Histidins im *Musculus longissimus dorsi*. Im Muskel der extensiv gefütterten Bullen wurden höhere Arginin/Glycin - und Arginin/Leucin - Quotienten gemessen. Der Mineralstoffgehalt im *Musculus longissimus dorsi* wurde von der Fütterung beeinflusst, während ein Einfluss der Rasse für den Ca- und Na-Gehalt festgestellt wurde. Der Gehalt von P und Fe in *Musculus longissimus dorsi* von extensiv gefütterten Tieren war signifikant höher, während der Na- und Cu-Gehalt geringer war. Das Fleisch von Ungarischem Grauvieh Bullen wies eine geringere Konzentration von Ca und Na im Vergleich zu Holstein Friesian Tieren auf. Cystein und Leucin korrelierten positiv mit dem intramuskulären Fettgehalt und negativ mit den *n*-3 and *n*-6 Fettsäuren. Von den Mineralstoffen korrelieren P und Fe negativ mit dem intramuskulären Fettgehalt. Die höchsten Korrelationen wurden zwischen dem Phosphor-Gehalt und der Linolensäure sowie der Summe der *n*-3 Fettsäuren (r=0,81) ermittelt. Das Rindfleisch der extensiven Gruppen wies das niedrigste Verhältnis von *n*-6 und *n*-3 Fettsäuren auf und hatte gleichzeitig höhere Prolin, Valin, Phenylalanin, Phosphor-Gehalte sowie niedrigere Glycin, Cystein, Leucin and Natrium-Konzentrationen auf.

Schlüsselwörter: Aminosäure, Mineralstoffe, Rindfleisch, Ungarisches Grauvieh, Holstein Friesen Bullen

Introduction

The beef consumption has been decreased during the last decades due to negative health images. Many studies (reviewed by SCOLLAN et al., 2006; ENDER et al., 2000; HOLLO et al., 2007) deal with the opportunities of enhancing the beneficial fatty acids [increasing the *n*-3 polyunsaturated fatty acids and conjugated linoleic acids (CLA) and reducing saturated fatty acids (SFA)] in beef, but little is known about the alteration of other nutritional ingredients (amino acid composition and mineral content) in beef.

Free amino acids and dipeptides are occurring in low concentrations in muscle. Muscle aminopeptidases contribute to the generation of free amino acids post mortem. The concentration of each amino acid is important for its contribution to taste (KATO et al., 1989). A negative relation was observed (SZŰCS et al., 1985) between the arginine and histidine content of the beef and its tastiness. According to FEIDT et al. (1991) the release of free amino acids in bovine meat is strongly muscle type dependent. Besides the muscle type the concentration of free amino acids and peptides in meat may also be affected by post mortem handling of meat (FELTON et al., 2000). A possible effect of feeding high or low indispensable amino acids can influence the amino acid structure and simultaneously the growing performance of bulls (KLUTH et al., 2000). Contrary to other species, there is very little known according to the influence of diet on the amino acid content in beef. However, investigations of the factors controlling the amino acid content of muscles are necessary in order to improve the quality and safety of meat.

Beef contains besides the high biological valuable protein some important minerals (NOUR and THONNEY, 1988; BIESALSKI, 2005; WILLIAMSON et al., 2005). The minerals in the human body are essential in neurological and digestive systems and the heart. The mineral content of beef was reported in some studies (GILLET et al., 1967; SIM and WELLINGTON, 1976; DOORNENBAL and MURRAY, 1981; BARGE et al., 2005). These focused mainly on the effect of age, breed, gender and type of muscle, only few studies analysed the effect of diet on the mineral content of beef (ONO et al., 1986; GIUFFRIDA-MENDOZA et al., 2007). The effect of dietary concentration of elements and their chemical form were researched by STANDISH et al. (1969) KESSLER et al. (2003). LOMBARDI-BOCCIA et al. (2005) analysed the influence of cooking processes on the concentration of trace elements (iron, zinc, copper) in different meat also in case of the beef.

The objectives of this study are to examine the effect of intensive *versus* extensive feeding on the amino acid composition and mineral content of *longissimus* muscle of Hungarian Grey and Holstein Friesian bulls and to determine the relationship between the altered fatty acid composition and the amino acid composition and mineral content.

Materials and methods

In total 40 Hungarian Grey (HG) and Holstein Friesian (HF) bulls were used for the feeding experiment. The details of the experiment and the diet of groups were presented in a previous publication (HOLLÓ et al., 2004, 2005). The measurements for the amino acid composition and mineral analyses had been done at the *longissimus* muscle (11-13 ribs) removed 24 hours after slaughter.

After the homogenisation of this slice the determination of the protein content was carried out with a Kjel-Foss 16200 rapid nitrogen analyser according to the relevant Hungarian standards. The amino acids were analysed by an INGOS AAA amino acid analyser. Hydrolysis of the samples is performed in reusable Pyrex hydrolysis tubes. Seven mL of 6 M HCL is added and nitrogen is bubbled through the tubes through a glass capillary for five minutes. Immediately after bubbling the tubes are sealed and hydrolysis begins at 110 ^{o}C. After hydrolysis (24h), the tubes are allowed to cool down to room temperature. In case of the amino acids containing sulphur performic acid oxidation was made before hydrolysis according to CSAPÓ et al. (1986). After the tubes are opened, the pH of the hydrolysate is adjusted to a value of 2.2, using 4M NaOH. During pH adjustment, a salt/ice cooling mixture is used to maintain the temperature below 30 ^{o}C. The resulting samples are diluted with a citrate buffer to pH value 2.2. Samples are filtered and stored at -25 ^{o}C until the analysis by ion exchange column chromatography. Three sodium buffer systems were used. The first sodium buffer (0.3 0 M Na, pH=3.50) elutes the following amino acids: aspartic acid, threonine, serine, glutamic acid, proline, cystine, glycine, alanine and valine. The second sodium buffer (0.40 M Na, pH=4.25) elutes methionine, isoleucine, leucine. The third sodium buffer (1.12 M Na, pH=7.9) elutes tyrosine, phenylalanine, histidine, lysine and arginine and ammonia. The flow rates of different buffers are 50 cm^3/h and the flow rate of ninhydrine is 25 cm^3/h. The determination of amino acids is performed with post column derivatisation by ninhydrine with photometric detection at 570 nm for all amino acids and 440 nm for proline.

The contents of calcium, copper, iron, magnesium, potassium, sodium and zinc was determined by using atomic absorption spectrometry (SOLAAR M 6) according to European standard (EN ISO 6869), whilst phosphorous content was analysed by spectrometric method according to International Standard (ISO- 6491).

The statistical analysis was made by SPSS (SPSS 10.0) program package. In Tables 1-2 the mean and the standard error of mean were presented. The GLM Type III. procedure and least significant difference (LSD) test was used to investigate the effects of individual factors (breed, diet) as well as interactions (breed x diet) between factors. Pearson correlation of coefficients and their significance level was calculated to determine the relation among variables.

Results

The amino acid composition of *longissimus* muscle is presented in Table 1. Seventeen amino acids were identified in all samples. The main essential amino acids in beef are leucine, lysine and arginine. The extensive diet caused some changes in the proportion of several essential amino acids. There were significant higher concentrations of isoleucine, leucine, and threonine in intensive groups and phenylalanine and valine were lower compared to extensive feeding.

Table 1
Amino acid composition (g/100g protein) of *longissimus* muscle ($Mean_{SEM}$) [Aminosäurezusammensetzung (g/100 g Protein) im *Musculus longissimus dorsi* ($Mean_{SEM}$)]

	Hungarian Grey		Hungarian Holstein				
	extensive	intensive	extensive	intensive	Breed	Diet	Interaction
Essential							
Histidine	$3.88_{0.19}$	$3.83_{0.11}$	$3.57_{0.06}$	$3.22_{0.23}$	0.007	NS	0.025
Isoleucine	$4.55_{0.14}$	$4.89_{0.10}$	$4.60_{0.11}$	$5.11_{0.17}$	NS	0.003	0.020
Leucine	$7.81_{0.14}$	$8.56_{0.15}$	$8.14_{0.18}$	$8.65_{0.19}$	NS	0.001	0.003
Lysine	$9.58_{0.27}$	$9.01_{0.15}$	$9.33_{0.23}$	$9.17_{0.12}$	NS	NS	NS
Methionine	$2.53_{0.07}$	$2.48_{0.14}$	$2.60_{0.09}$	$2.39_{0.07}$	NS	NS	NS
Phenylalanine	$4.35_{0.11}$	$4.26_{0.11}$	$4.72_{0.15}$	$4.10_{0.15}$	NS	0.012	0.013
Threonine	$4.03_{0.09}$	$4.29_{0.06}$	$4.11_{0.08}$	$4.20_{0.11}$	NS	0.050	NS
Valine	$4.89_{0.17}$	$4.56_{0.12}$	$4.94_{0.17}$	$4.50_{0.09}$	NS	0.009	NS
Arginine	$6.51_{0.23}$	$6.43_{0.12}$	$6.23_{0.13}$	$6.08_{0.22}$	NS	NS	NS
Total	$48.13_{0.44}$	$48.31_{0.36}$	$48.24_{0.27}$	$47.42_{0.29}$	NS	NS	NS
Non essential							
Alanine	$5.61_{0.12}$	$5.43_{0.20}$	$5.59_{0.09}$	$5.87_{0.17}$	NS	NS	NS
Aspartic acid	$9.73_{0.21}$	$9.74_{0.18}$	$9.66_{0.11}$	$9.80_{0.18}$	NS	NS	NS
Cysteine	$0.97_{0.08}$	$1.27_{0.11}$	$0.94_{0.09}$	$1.15_{0.06}$	NS	0.005	0.037
Glutamic acid	$17.53_{0.27}$	$17.24_{0.19}$	$17.44_{0.21}$	$17.73_{0.18}$	NS	NS	NS
Glycine	$4.57_{0.15}$	$4.75_{0.09}$	$4.29_{0.10}$	$4.81_{0.10}$	NS	0.004	0.011
Proline	$4.92_{0.13}$	$4.66_{0.23}$	$4.99_{0.13}$	$4.40_{0.10}$	NS	0.009	0.041
Serine	$3.51_{0.11}$	$3.86_{0.08}$	$3.70_{0.06}$	$3.78_{0.10}$	NS	0.021	0.049
Tyrosine	$3.78_{0.11}$	$3.94_{0.13}$	$3.74_{0.09}$	$3.87_{0.15}$	NS	NS	NS
Total	$50.62_{0.52}$	$50.89_{0.37}$	$50.35_{0.26}$	$51.41_{0.23}$	NS	NS	NS
E/NE	$0.95_{0.02}$	$0.95_{0.01}$	$0.96_{0.01}$	$0.92_{0.01}$	NS	NS	NS
NH3	$1.32_{0.10}$	$0.83_{0.07}$	$1.36_{0.11}$	$1.15_{0.08}$	NS	0.001	0.001

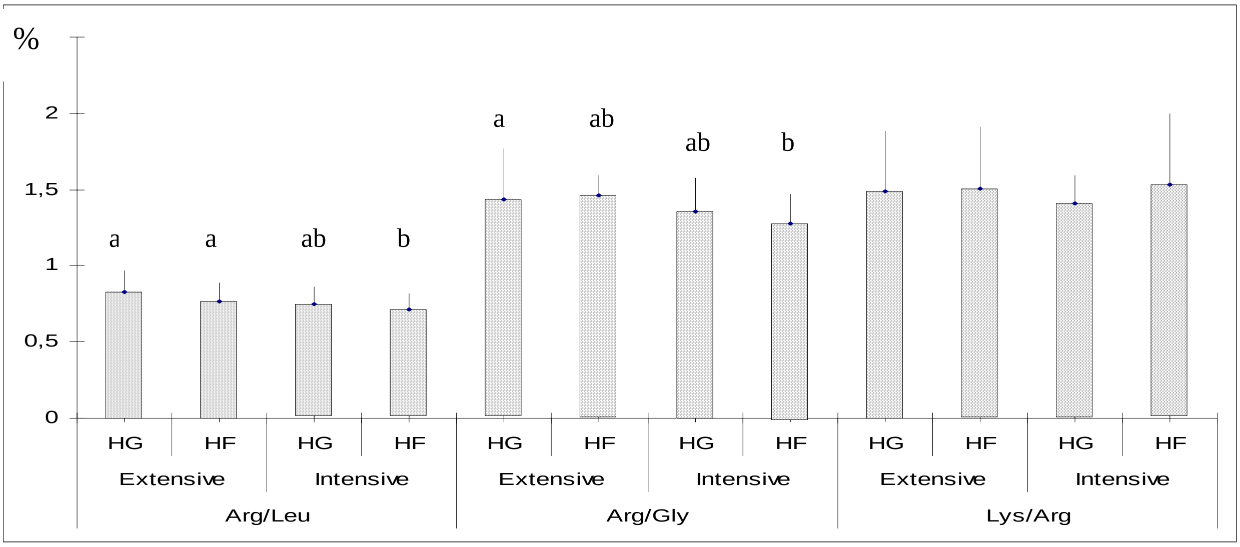
The effect of breed was significant only in the case of histidine. Breed and diet interactions were detected for histidine, isoleucine, leucine and phenylalanine. Glutamic acid and aspartic acid proportions were detected as the highest among non essential amino acids. The non essential amino acid composition did not differ significantly between breeds, however the diet and breed x diet interaction influenced ($P<0.05$) several amino acids including cysteine, glycine, proline, serine and ammonia content values in *longissimus* muscle. In extensive groups the proline percentage is increased, whilst the cysteine, glycine and serine proportions in muscle were decreased.

Table 2
Mineral composition (mg/100g) of *longissimus* muscle ($Mean_{SEM}$) [Mineralstoffzusammensetzung (mg/100g) im Musculus longissimus dorsi ($Mean_{SEM}$)]

	Hungarian Grey		Hungarian Holstein				
	extensive	intensive	extensive	intensive	Breed	Diet	Interaction
Ca	$3.27_{0.55}$	$3.89_{0.15}$	$4.99_{0.28}$	$4.17_{0.25}$	0.007	NS	0.010
P	$200.05_{1.38}$	$176.86_{1.18}$	$190.11_{2.42}$	$173.48_{2.19}$	NS	0.000	0.000
Mg	$19.18_{0.47}$	$20.39_{0.37}$	$21.46_{0.46}$	$18.09_{0.62}$	NS	NS	0.000
K	$350.60_{6.13}$	$310.78_{6.13}$	$317.90_{8.54}$	$338.40_{10.18}$	NS	NS	0.004
Na	$55.37_{1.52}$	$62.42_{2.30}$	$58.11_{1.34}$	$73.92_{3.88}$	0.028	0.000	0.000
Cu	$0.08_{0.01}$	$0.13_{0.03}$	$0.09_{0.01}$	$0.14_{0.02}$	NS	0.015	NS
Zn	$3.47_{0.08}$	$3.38_{0.12}$	$3.37_{0.10}$	$3.64_{0.06}$	NS	NS	NS
Fe	$1.82_{0.09}$	$1.42_{0.09}$	$1.56_{0.06}$	$1.47_{0.09}$	NS	0.008	0.009

The nutritional value of the protein depending on the protein composition of the different tissues is determined by the amount and the proportion of the essential amino

acids (PIVA and GUGLIELMETTI, 1978). The essential and non essential amino acid ratio was ranged between 0.92-0.96 among the treatments. There were no significant differences. The ammonia content was significantly higher in extensive groups.
The mineral composition of *longissimus* muscle is shown in Table 2. The potassium and magnesium content controversial changed in different feeding groups of breeds, the beef in the extensive group of Hungarian Grey contained lower magnesium but higher potassium content than those of the same Holstein group. The P and iron level was significantly increased, whilst the Na and Cu content was decrease in extensive fed animals.



(a,b- different letters are significant different between all groups at p ≤0.05)

Figure: Changes in leucine, glycine and lysine to arginine ratios in *longissimus* muscle (Änderungen der Quotienten von Arginin/Leucin, Arginin/Glycin und Lysin/Arginin im *Musculus longissimus dorsi*)

Besides this, the breed also affected the sodium level in muscle. The Na and Ca content in Hungarian Grey beef was lower compared to Holstein Friesian muscle. Only in case of zinc there was neither breed nor diet effect.
The Figure shows the ratio of leucine, glycine and lysine to arginine in *longissimus* muscle. The ratio of arginine and leucine as well as ratio of arginine to glycine differed significantly among groups. In muscle of extensive fed Hungarian Grey significantly higher values were found concerning both ratios compared with intensive fed Holstein Friesian.
The relationships between fatty acids and amino acids as well as minerals in *longissimus* muscle are presented in Table 3 and Table 4, respectively. The intramuscular fat content of muscle influenced cysteine, leucine and ammonia content. When intramuscular fat increased, also cysteine and leucine content increased but ammonia proportion decreased with increasing muscle fat. On the other hand cysteine and leucine were negatively correlated with PUFA and *n*-3 and *n*-6 fatty acids and positively with MUFA. A contrary tendency can be seen in case of the ammonia. Among saturated fatty acids palmitic and stearic acid showed a medium relationship with several amino acids such as proline, glycine, cysteine, valine, isoleucine, leucine, phenylalanine and ammonia. Between linolenic acid and proline, valine as well as phenylalanine established a low positive correlation ranged between 0.37-0.43. The

lower *n*-6 and *n*-3 fatty acid ratio in muscle is correlated to a higher proline, valine and phenylalanine and lower glycine, cysteine and leucine proportion.

Table 3
Coefficient of correlation between amino acids and fatty acids in *longissimus* muscle [Korrelationskoeffizienten zwischen Aminosäuren und Fettsäuren im *Musculus longissimus dorsi*]

	Pro	Gly	Cys	Val	Ile	Leu	Phe	NH_3
Intramusc fat content	-	-	0.53	-	-	0.38	-	-0.49
C16:0 palmitic	-0.40	0.40	0.41	-	0.42	0.45	-0.32	-0.59
C18:0 stearic	0.33	-0.43	-0.33	0.39	-0.41	-0.46	0.49	0.49
C18:2 *n*-6 linoleic	-	-	-0.39	-	-0.35	-0.37	-	0.53
C18:3 *n*-3 linolenic	0.43	-0.40	-0.46	0.37	-0.43	-0.52	0.39	0.49
C20:3 *n*-6 eicosatrienoic	-	-0.34	-0.46	-	-	-	-	-
C20:4 *n*-6 arachidonic	0.32	-0.36	0.30	-	-	-	-	0.49
C20:5 *n*-3 eicosapentaenoic	0.37	-	-0.41	-	-0.47	-0.43	-	-
MUFA	-0.32	0.38	0.42	-0.35	0.38	0.40	-0.40	-0.51
PUFA	0.35	-0.36	-0.44	-	-0.37	-0.38	-	0.54
n-3 fatty acids	-	-0.32	-0.40	-	-0.35	-0.35	-	0.53
n-6 fatty acids	0.35	-0.36	-0.44	-	-0.36	-0.35	-	0.53
n-6/*n*-3 ratio	-0.39	0.36	0.35	-0.33	-	0.39	-0.37	-

r ≥ 0.32 are significant at P=0.05 level

Among minerals phosphorous and iron showed a negative relationship to the intramuscular fat level and palmitic acid content in muscle. Moreover, between iron and SFA a negative correlation was found too, whereas the P content was positively correlated to stearic, linoleic, linolenic, eicosatrienoic, arachidonic and eicosapentaenoic acids. The closest correlation was determined between P and linolenic as well as *n*-3 fatty acids (r=0.81). Concerning Fe also a positive, but a weaker relationship was calculated (r=0.43-0.44). The *n*-6/*n*-3 fatty acid ratio is positively correlated to the sodium and negatively to the phosporous content in beef muscle.

Table 4
Coefficient of correlation between minerals and fatty acids in *longissimus* muscle [Korrelationskoeffizienten zwischen Mineralstoffen und Fettsäuren im *Musculus longissimus dorsi*]

	P	Mg	Na	Cu	Fe
Intramusc fat content	-0.52				-0.34
C16:0 palmitic	-0.63		0.41		-0.50
C18:0 stearic	0.61	0.37	-0.46	-0.41	
C18:2 *n*-6 linoleic	0.46				0.38
C18:3 *n*-3 linolenic	0.81		-0.52	-0.33	0.44
C20:3 *n*-6 eicosatrienoic	0.39				
C20:4 *n*-6 arachidonic	0.45				
C20:5 *n*-3 eicosapentaenoic	0.75		-0.42		0.38
SFA					-0.39
MUFA	-0.61		0.39		
PUFA	0.57		-0.32		0.37
n-3 fatty acids	0.81		-0.50		0.43
n-6 fatty acids	0.46				0.37
n-6/*n*-3 ratio	-0.76		0.58		

r ≥ 0.32 are significant at P=0.05 level

Discussion

In this study the effect of the extensive feeding *versus* intensive diet on the amino acid composition and mineral content of beef was studied. Additionally the relation between fatty acid profile and amino acid composition as well as mineral content of beef was investigated. In agreement with our results FRANCO et al. (2006) found that in beef the highest essential amino acid fractions were lysine and leucine whereas the non-essentials were glutamic acid and aspartic acid. At the same time MOLNÁR and MOLNÁR (1981) established that the amino acid composition of the different muscles in Hungarian Simmental breed is changed according to age, and even the amino acid content is influenced by the stress of the muscles as well. CUTINELLY-AMBESI (1975) proved that the amino acid pattern is very similar of domestic ox, buffalo and African buffalo, only the lysine content was higher as compared with domestic ox. According to previous result (HOLLÓ et al., 2001) the breed also influenced the proportion of some amino acids in beef. The content of histidine, threonine, valine, glycine, glutamic acid and serine differed significant between Holstein and Hungarian Simmental cows. In this investigation the amino acid profile of Holstein and Hungarian Grey bulls showed a similar composition except for histidine. Limiting amino acids in dietary protein are typically the sulphur-containing amino acids, like threonine and lysine. The amount of threonine in *longissimus* muscle was higher than in potato protein. The lysine proportion was also higher than both hen egg and potato protein (MORUP and OLESEN (1976). Beef from extensive groups contained lower threonine and higher lysine level. The diet significantly affected the threonine content of muscle. In human investigations were observed that the amino acid ratios of meals influenced the amino acids in plasma as well as pancreatic hormone secretion (SOUCY and LeBLANC, 1999). The amino acid arginine is a precursor of nitric oxide, a potent vasodilator which can inhibit platelet adhesion and aggregation (SABATÉ et al. 1993, cit. SERRANO et al. 2005). Higher arginine/leucine or arginine/glycine ratios detected in plasma are associated with lower insulin/glucagon ratio. Low insulin/glucagon ratio is associated with hypocholesterolemia (SANCHES and HUBBARD, 1991). In present study the leucine and glycine concentrations in beef are significantly decreased in extensive groups, whilst arginine was increased. These higher arginine and leucine or arginine and glycine ratios detected in extensive groups is favourable of human nutritional point of view (Figure). According to KRITCHEVSKY et al. (1982) besides arginine, a low lysine and arginine ratio reduces atherosclerosis in rabbits. The present results the lysine and arginine ratio did not change significantly in all cases.

The coefficients of correlation showed that the higher intramuscular fat content is related to enhanced level of cysteine and leucine proportion in muscle. GALLO et al. (2002) established that the amino acid percentage of meat is negatively correlated with the PUFA content in the diet (r=-0.78). In our findings muscle PUFA concentration is negatively correlated to glycine, cystine, isoleucine and leucine and positively to proline and ammonia proportion. The beef with favourable n-6/*n*-3 fatty acid ratio contained lower proportion of glycine, cysteine and leucine and higher content of proline, valine and phenylalanine. GINGRAS et al. (2006) also found that the *n*-3 fatty acids could also affect the protein metabolism of muscle. They added different supplements containing long chain *n*-3 polyunsaturated fatty acids to the diet of six growing steers. Animals with the marine *n*-3 fatty acids supplementation showed increased sensitivity to insulin and improved protein metabolism. Based on the results of HOLLO et al. (2005) it can be established that the effect of diet besides the fatty acid composition also influenced the isoleucine, leucine, phenylalanine, threonine, valine, cysteine, glycine, proline, serine and ammonia proportion of beef.

As nutrition surveys described in the USA (KENNEDY and GOLDBERG, 1995), Great Britain (SAMUELSON et al., 1996) and Sweden (GREGORY et al., 1995) the intakes of calcium, iron and zinc are often marginal, while the intakes of sodium and magnesium reach or exceed the recommendations.

Currently, one of the major health concerns related to the intakes of calcium in the diet is osteoporosis; the daily recommended value is 800 mg (FERREIRA et al., 2000). The mean Ca content of *longissimus* muscle was 4.08 mg/100 g very small value in comparison with those reported by USDA (1990) (10.59 mg/100 g cooked whole beef rib) and by SERRANO et al. (2005) in case of the beef steak but similar to values measured at 17 months of age zebu crossbred cattle and buffalo (GIUFFRIDA-MENDOZA et al., 2007). The calcium content of *longissimus* muscle in our study was affected by breed but not by diet. ONO et al. (1986) found by milk fed calves higher Ca in muscle than in special fed veal. BARGE et al. (2005) observed that Ca content of *longissimus* muscle negatively correlated with age. On the other hand GIUFFRIDA-MENDOZA et al. (2007) reported for Buffalo and Zebu influenced cattle higher muscle Ca concentrations in 17 to 19 months old cattle compared to 24 months old animals.

Similarly to the studies of LAWRIE (1990) and BARGE et al. (2005) the beef is quantitatively the most enriched meat source of potassium followed by phosphorous content. The P concentration of muscle was affected by diet and there was a breed and diet interaction, too. The value increased in the extensively fed groups, and was also higher in Hungarian Grey groups. GIUFFRIDA-MENDOZA et al. (2007) established significant species and age effects on the P content of *longissimus* muscle. The average level of P (185.13 mg/100g) was higher than those (150-171 mg/100 g) reported by HERMIDA et al. (2006).

The mean Mg content of *longissimus* muscle was detected at 19.78 mg/100 g, agreed with the range for beef in the literature. The Mg content did not relate to the age (BARGE et al., 2005). In our results Mg concentration was affected neither by breed nor by diet. No clear tendency can be seen among treatments for Mg, similar to potassium level. The average K content (329.42 mg/100 g) was slightly lower compared to other values measured in beef in the USA (340 mg/100g USDA, 1990), and in Spain in case of the beef steak (350 mg/100g (SERRANO et al., 2005). Analysing the Na content it was similar to USDA values. K and Na content of beef was influenced by muscle type (SIM and WELLINGTON, 1976; GILLET et al., 1967) and by age (DOORNENBAL and MURRAY, 1981; KOTULA and LUSBY, 1982). Na concentration was significantly lower in extensive and Hungarian Grey groups. High potassium and low sodium level in Hungarian grey extensively fed group indicates nutritional advantage.

Among species veal and beef cuts showed the highest amount of zinc (LOMBARDI-BOCCIA et al., 2005). Many studies have shown that zinc deficient diet affects the immune system in humans. The Zn was affected by age (GIUFFRIDA-MENDOZA et al., 2007), by beef cut (LOMBARDI-BOCCIA et al., 2005), and by sex (LÓPEZ-

ALONSO et al., 2000). In the present study zinc varied between 3.37-3.64 mg/100g, and it was either by diet or breed influenced.

GIUFFRIDA-MENDOZA et al. (2007) suggested the fluctuation in the mineral content observed among ages could be due to confounding effect of feeding (grass availability). LOMBARDI-BOCCIA et al. (2005) concluded that there is a great variability in micronutrients content in meats among studies, furthermore the introduction of new rearing systems contribute to the variability of the micronutrient composition of meat.

LOMBARDI-BOCCIA et al. (2005) measured in raw beef sirloin higher value for Fe and Zn, whilst lower value for Cu compared to our results. Iron deficiency is the most common and widespread nutritional disorder in the world (DeMAEYER and ADIELS-TEGMAN, 1985). At the same time meat is the food richest in heme iron representing the primary source of heme iron in the Italian total diet (LOMBARDI-BOCCIA et al., 2005). The extensive diet caused significantly higher Fe values in this study. In opposite the copper level changed, in intensive groups more Cu was detected. In line with this result LÓPEZ-ALONSO et al. (2000) observed previously that the copper concentrations in muscle are inversely related to muscle lipid concentration. Similarly to the studies of LAWRIE (1990) and BARGE et al (2005) the beef is quantitatively the most enriched meat source in potassium followed by phosphorous content.

The relationship of mineral content and tenderness of beef from different breed, sex and age were investigated by MURRAY et al. (1982). They found no significant correlation between shear values and concentrations of the minerals in the *longissimus*, whilst in the *semimembranosus* muscle Ca and Fe were significant related to tenderness. JEREZ-TIMAURE et al. (2006) used canonical correlation to determine the relationship between mineral concentrations and meat quality traits in buffalo and zebu-type cattle. The Ca content is most highly correlated among the minerals. Results indicated a positive correlation between Ca content and shear force values. They suggested other nutritional components such as fatty acid profile and total lipids might be included in further studies. Previous, BARGE et al. (2005) established a negative correlation between K and Mg as well as intramuscular fat in *longissimus* muscle. In the present investigation a negative relationship was found between Fe and P as well as fat. Fe and P content correlated significantly positive to polyunsaturated fatty acids. At the same time the low *n*-6/*n*-3 ratio is related to lower sodium and higher phosphorous level in muscle.

In conclusion extensive diet with grass/grass silage and concentrate with linseed supplements caused changes in the proportion of some amino acids compared to that of intensive groups. The enhancement of the phenylalanine, valine, proline and ammonia concentration and the decrease of isoleucine, leucine, threonine, cysteine, glycine and serine were observed. Breed differences were detected only for the histidine content.

The *longissimus* muscle of extensive fed animals contained a lower level of sodium and copper and a higher level of iron and phosphorous than intensive ones. Hungarian Grey bulls had a lower Ca and Na content in *longissimus* muscle opposite to Holstein counterparts.

Cysteine and leucine are positively correlated to intramuscular fat content and negatively to *n*-3 and *n*-6 fatty acids. Among minerals the P and Fe content correlated negatively to intramuscular fat content. The closest correlation was determined between P and linolenic as well as *n*-3 fatty acids (r=0.81). Feeding grass and concentrate with linseed supplements (extensive groups) resulted in lower *n*-6 and *n*-3 fatty acid ratio and in higher proline, valine, phenylalanine and phosphorous and a lower glycine, cysteine, leucine and sodium proportions in *longissimus* muscle.

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