

## Fatty acid levels in the muscle tissue of eight beef cattle breeds

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### Abstract

The objective of the study was to compare the chemical composition and fatty acid profile in ten clinically healthy bulls of similar weight from eight meat cattle breeds. The animals were raised by extensive grazing under the same geographical conditions. Significant differences were observed between the monitored genotypes in terms of the dry matter content, nitrogenous matter and fats ( $P \leq 0.05$ ) in the *musculus longissimus thoracis*. The concentration of fats ranged from 68.5 ± 18.81 g (Galloway cattle) to 171.6 ± 43.3 g (Aberdeen Angus) per kg of dry matter. The total content of unsaturated fatty acids ranged from 20.0 ± 2.25 g (Aberdeen Angus) to 42.4 ± 7.87 g (Gascon); the total sum of monounsaturated fatty acids ranged from 17.0 ± 2.26 g (Aberdeen Angus) to 33.8 ± 3.61 g (Gascon); and the total sum of polyunsaturated fatty acids ranged from 3.0 ± 1.42 g (Aberdeen Angus) to 5.1 ± 3.99 g (Limousin) per 100 g of fat. The total content of n-6 fatty acids ranged from 2.4 ± 1.28 g (Aberdeen Angus) to 4.2 ± 3.59 g (Limousin) and the total content of n-3 fatty acids from 0.5 ± 0.16 g (Salers) to 1.1 ± 0.04 g (Gascon) per 100 g of fat. The properties we studied may predict the biological, dietetic, and culinary value of the meat.

*Musculus longissimus thoracis*, MUFA, PUFA, n-6 PUFA, n-3 PUFA

Along with vegetable oils, lipid sources in human nutrition also include fats of animal origin. Findings regarding the presence of fatty acids in vegetable and animal fats have nutritional, dietary and health implications for both people and animals. These effects include the physiological functions of fats in the living organism as well as the functions of individual fatty acids (FA) in the study of diets and in the prevention and treatment of diseases related to a temporary deficiency or increased intake. In this regard, attention is also given to the lipid component in beef (Scollan et al. 2006), where the need to increase the ratio of n-3 polyunsaturated fatty acids (PUFA) has been emphasized (Griel et al. 2006; Kris-Etherton 2007).

It is generally accepted that the concentration of FA in the meat of food animals significantly depends on the content of their diet (Aldai et al. 2008). Several authors (Enser et al. 1998; Duynisveld et al. 2006; Ponnampalam et al. 2006; Garcia et al. 2008; Smith et al. 2009; Daley et al. 2010; Yüksel et al. 2012; Turner et al. 2015) have studied the presence of FA in beef in relation to the diet. These studies compared diets based on grazing and grain-fed beef. Grazing increased the amount of the precursors of vitamins A and E, as well as antioxidants such as glutathione and superoxide dismutase (Daley et al. 2010). In particular, vitamin E maintains the PUFA concentration in beef and contributes to ensuring its quality during processing (Wood et al. 2004). Calf meat on pasture had a higher percentage of saturated fatty acids (SFA), PUFA and n-3 PUFA and a lower percentage of intramuscular fat, monounsaturated fatty acids (MUFA), and n-6 PUFA than animals that were fed with corn while grazing or fed a dose of corn, soybean

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and alfalfa (Garcia et al. 2008). Yüksel et al. (2012) discovered that a diet based on grazing had a favourable impact on the FA profile in meat. However, the resulting meat was darker and tougher.

The relationships between cattle breeds, their nutrition, the presence of fatty acids in intramuscular fat and the quality of meat have been studied by various researchers (Holló et al. 2001; Dannenberger et al. 2003; Raes et al. 2003; Garcia et al. 2008; Xie et al. 2012; Liu et al. 2015). For example, Garcia et al. (2008) report that the diet had a greater influence on the composition of the fatty acids of intramuscular fat than the breed. In three breeds (Holstein, Simmental, and Chinese LongDong Yellow Cattle), Liu et al. (2015) proved that the presence of fatty acids differed in fat of the *musculus longissimus thoracis* and in kidney fat. In contrast, Holló et al. (2001) stated that the proportion of saturated, mono- and polyunsaturated FA were not significantly influenced by the breed (Hungarian Simmental, Holstein-Friesian) or by live weight.

Intramuscular fat in beef is very important in terms of taste and tenderness; especially its intercellular ratio, which is divided between muscle fibre in vein-like patterns and forms the so-called marbled meat (Steinhauser et al. 2000). Lipophilic substances, which are released with heat and contribute to the meat's smell and taste (Ingr 2011), are contained in the muscle fat. Jiang et al. (2011) observed the effect of fatty acids, noting a negative influence on the meat's taste for eicosapentaenoic acids (C20:5n3).

Literary data on the presence of FA in the fat of modern beef cattle breeds are hard to find, and a comparison of several breeds with the same diet, same husbandry practices, and in a single location is very rarely done. The presented work should contribute to existing knowledge and attest whether different genotypes of cattle raised by extensive grazing under the same geographical conditions influence the chemical composition of the fat component (especially in relation to the presence of individual groups of FA) in the muscles of contemporary breeds.

### Materials and Methods

The subject of observation was eight beef cattle breeds recognized in the Czech Republic: Aberdeen Angus (AA), Blonde d'Aquitaine (BA), Limousin (Lim), Gascon cattle (Gas), Meat Simmental (MS), Salers (Sal), Galloway cattle (Gall) and Czech multi-colored (Cs). Animals of the selected breeds were raised in the same region of Southern Bohemia (Křišťanov) by extensive grazing. Their diet was based on the intake of grasslands, and their winter diet consisted of hay and straw.

Ten clinically healthy bulls of similar weight between the ages of 16 to 18 months were chosen from each beef cattle breed. After slaughtering, a 500-g muscle sample was taken from the *musculus longissimus, pars thoracis*. The samples were taken from the left half of the body between areas 1 to 10 of the thoracic vertebrae, then the samples were chilled, frozen and gradually analyzed. The muscle samples were provided by the producer ZEFA Volary, Czech Republic.

The dry matter, nitrogenous matter, fat, gross energy (AOAC 2006) and fatty acids were determined in a laboratory. The muscle sample was homogenized, dried at 105 °C under prescribed conditions, and the dry matter was determined. The content of nitrogen was determined using the Kjeldahl method on a Buchi analyzer (Centec automatika, Czech Republic), and the content of nitrogenous matter (crude protein) was calculated by multiplying the value of nitrogen by the coefficient of 6.25. Fat was determined by the ANKOMXT10 Fat Analyzer (O.K. SERVIS BioPro, Czech Republic). Combusted heat was determined by a combusted sample in the calorimetric bomb in an oxygen atmosphere in the Leco (USA) calorimeter. The extraction of fat from the muscle to determine fatty acids was performed according to Hara and Radin (1978). In addition, the standard ČSN EN ISO 12966-1, 12966-2 was used. Fatty acid esters were detected by gas chromatography using a Gas Chromatograph GC-2010 analyzer (Shimadzu, Japan) with a flame ionisation detector and evaluated in the GC PostRun program. Some fatty acids in the muscle sample reached very low values below the detection limit, therefore only certain selected fatty acids were monitored. Samples were converted to the content of the given fatty acid in the sample (g/100 g of fat).

To ensure more accurate reproducibility, all the results are presented in muscle dry matter (g/kg of dry matter). The obtained results were processed by the statistical program Unistat CZ version 5.6 for Excel, in which the mean values and their differences were evaluated by multiple comparison by Tukey-HSD test at the significance level of  $P \leq 0.05$ .

## Results

### Presence of essential nutrients in the muscle

The dry matter of muscle in the eight monitored breeds of bulls (Table 1) was determined at a range from  $245.2 \pm 17.64$  g (Gall) to  $277.6 \pm 11.63$  g (AA) per kg, which proves a positive correlation ( $r = +0.922$ ;  $P < 0.05$ ) with the fat content, whose concentration ranges from  $68.5 \pm 18.81$  g (Gall) to  $171.6 \pm 43.30$  g (AA) per kg.

The level of nitrogenous substances were the lowest in AA ( $791.1 \pm 45.83$  g/kg of dry matter). The highest values were discovered in Lim ( $897.6 \pm 60.36$  g/kg of dry matter).

The highest content of fat was found in the AA muscle ( $171.6 \pm 43.30$  g/kg dry matter), which also had the highest content of gross energy (GE) among the monitored breeds ( $25.1 \pm 0.67$  MJ/kg). In contrast, the lowest GE value was found in Gall ( $23.2 \pm 0.33$  MJ/kg).

Total content of saturated and unsaturated FA in the muscle fat

The total content of saturated fatty acids ( $\Sigma$  SFA) ranged from  $31.8 \pm 3.05$  g ( $\check{C}s$ ) to  $38.8 \pm 1.57$  g (Gas) per 100 g of fat. The total content of unsaturated fatty acids ( $\Sigma$  UFA) ranged from  $20.0 \pm 2.25$  g (AA) to  $42.4 \pm 7.87$  g (Gas) per 100 g of fat. While no significant differences in the values of  $\Sigma$  SFA were proven in the monitored breeds, several significant ( $P \leq 0.05$ ) to highly significant ( $P \leq 0.01$ ) differences were confirmed for  $\Sigma$  UFA (Table 2).

Total content of MUFA and PUFA in the muscle fat

Of the unsaturated FA, MUFA predominates, ranging from  $5.5 \times$  (Lim) to  $8.9 \times$  (Sal) more in comparison with PUFA (Table 3) in the muscle fat. The total content of MUFA ranges from  $17.0 \pm 2.26$  g in AA to  $33.8 \pm 3.61$  g in Gas per 100 g of fat. PUFA had a total presence ranging from  $3.0 \pm 1.42$  g (AA) to  $5.1 \pm 3.99$  g (Lim) per 100 g of fat.

Table 1. Content of dry matter, nitrogenous substances (N-substances) and fat in g/kg of dry matter and gross energy (GE) in MJ/kg of dry matter of muscle of eight cattle breeds.

Breed	AA	BA	Lim	Gas	MS	Sal	Gall	$\check{C}s$
Dry matter ( $\times$ ) $\pm$ SD	$277.56 \pm 11.63$	$277.27 \pm 22.27$	$251.10 \pm 13.26$	$269.74 \pm 8.37$	$271.53 \pm 7.75$	$267.51 \pm 23.96$	$245.21 \pm 17.64$	$261.77 \pm 12.05$
N-Matter ( $\times$ ) $\pm$ SD	$791.06 \pm 45.83$	$821.99 \pm 72.06$	$897.57 \pm 60.36$	$868.29 \pm 25.09$	$849.44 \pm 25.79$	$835.68 \pm 60.59$	$858.94 \pm 42.62$	$868.76 \pm 47.01$
Fat ( $\times$ ) $\pm$ SD	$171.61 \pm 43.30$	$145.00 \pm 81.06$	$80.44 \pm 59.11$	$108.81 \pm 23.57$	$129.70 \pm 30.31$	$118.86 \pm 58.28$	$68.50 \pm 18.81$	$102.99 \pm 41.16$
GE MJ/kg ( $\times$ ) $\pm$ SD	$25.16 \pm 0.67$	$24.57 \pm 1.36$	$23.52 \pm 0.95$	$24.00 \pm 0.51$	$24.37 \pm 0.62$	$24.03 \pm 0.96$	$23.20 \pm 0.33$	$24.00 \pm 0.80$

Aberdeen Angus (AA), Blonde d'Aquitaine (BA), Limousin (Lim), Gascon cattle (Gas), Meat Simmental (MS), Salers (Sal), Galloway cattle (Gall), Czech multi-coloured ( $\check{C}s$ ),  $\times$  - mean; SD - standard deviation; <sup>ab, cd</sup>  $P \leq 0.05$

Table 2. Total content of SFA and UFA in the muscle fat of eight breeds of cattle in g/100 g of fat.

Breed	AA	BA	Lim	Gas	MS	Sal	Gall	$\check{C}s$
$\Sigma$ SFA ( $\times$ ) $\pm$ SD	$34.02 \pm 2.93$	$32.56 \pm 4.46$	$35.77 \pm 7.11$	$38.80 \pm 1.57$	$33.71 \pm 4.24$	$33.58 \pm 3.61$	$33.25 \pm 2.92$	$31.82 \pm 3.05$
$\Sigma$ UFA ( $\times$ ) $\pm$ SD	$20.04 \pm 2.25$	$26.31 \pm 6.32$	$33.12 \pm 5.04$	$42.47 \pm 7.87$	$30.50 \pm 6.60$	$31.77 \pm 8.36$	$26.75 \pm 1.05$	$25.87 \pm 3.56$
$\Sigma$ SFA: $\Sigma$ UFA	1 : 0.59	1 : 0.81	1 : 0.93	1 : 0.91	1 : 0.90	1 : 0.95	1 : 0.80	1 : 0.81

Aberdeen Angus (AA), Blonde d'Aquitaine (BA), Limousin (Lim), Gascon cattle (Gas), Meat Simmental (MS), Salers (Sal), Galloway cattle (Gall), Czech multi-coloured ( $\check{C}s$ ), saturated fatty acids (SFA); unsaturated fatty acids (UFA),  $\times$  - mean; SD - standard deviation; <sup>ab</sup>  $P \leq 0.05$

Table 3. Total content of MUFA and PUFA in the muscle fat of eight breeds of cattle in g/100 g of fat.

Breed	AA	BA	Lim	Gas	MS	Sal	Gall	Čs
ΣMUFA (×) ± SD	17.00 <sup>a</sup> ± 2.26	22.56 <sup>b</sup> ± 6.43	28.02 ± 6.83	33.81 <sup>a</sup> ± 3.61	27.05 ± 6.56	28.56 ± 8.05	22.67 <sup>b</sup> ± 3.00	22.38 <sup>b</sup> ± 3.12
ΣPUFA (×) ± SD	3.04 ± 1.42	3.75 ± 2.59	5.11 ± 3.99	4.54 ± 2.85	3.45 ± 0.88	3.21 ± 0.68	4.08 ± 2.45	3.49 ± 0.84
ΣMUFA:ΣPUFA	1 : 0.18	1 : 0.17	1 : 0.18	1 : 0.13	1 : 0.13	1 : 0.11	1 : 0.18	1 : 0.16

Aberdeen Angus (AA), Blonde d'Aquitaine (BA), Limousin (Lim), Gascon cattle (Gas), Meat Simmental (MS), Salers (Sal), Galloway cattle (Gall), Czech multi-coloured (Čs), monounsaturated fatty acids (MUFA); polyunsaturated fatty acids (PUFA), × - mean; SD - standard deviation; <sup>a,b</sup> P ≤ 0.05

Table 4. Total content of n-6 and n-3 FAs in the muscle fat of eight breeds of cattle in g/100 g of fat.

Breed	AA	BA	Lim	Gas	MS	Sal	Gall	Čs
Σn-6 FAs (×) ± SD	2.47 ± 1.28	3.03 ± 2.11	4.21 ± 3.59	3.39 ± 0.39	2.92 ± 0.84	2.72 ± 0.56	3.50 ± 2.52	2.88 ± 0.86
Σn-3 FAs (×) ± SD	0.57 ± 0.20	0.72 ± 0.51	0.90 ± 0.55	1.15 ± 0.04	0.53 ± 0.15	0.49 ± 0.16	0.57 ± 0.09	0.61 ± 0.16
Σn-3:Σn-6 FAs	1 : 4.35	1 : 4.19	1 : 4.70	1 : 2.94	1 : 5.50	1 : 5.58	1 : 6.11	1 : 4.69

Aberdeen Angus (AA), Blonde d'Aquitaine (BA), Limousin (Lim), Gascon cattle (Gas), Meat Simmental (MS), Salers (Sal), Galloway cattle (Gall), Czech multi-coloured (Čs), fatty acids (FAs), × - mean; SD - standard deviation

Total content of n-6 and n-3 fatty acids in the muscle fat

The total content of n-6 FA ranged from  $2.4 \pm 1.28$  g (AA) to  $4.2 \pm 3.59$  g (Lim) per 100 g of fat.

The total content of n-3 FA varied from  $0.5 \pm 0.16$  g (Sal) to  $1.1 \pm 0.04$  g (Gas) per 100 g of fat in the monitored breeds (Table 4). The ratio of n-6/n-3 ranged from 2.94 (GAS) to 6.11 (Gall).

## Discussion

Presence of essential nutrients in the muscle

Findings on the presence of essential nutrients (dry matter, N-substances, fat), gross energy and especially individual FA in the muscles of bulls of several beef cattle breeds kept under the same conditions are uncommon. However, the values of these indicators may pre-determine the biological, dietary and culinary value of the meat.

Wood et al. (2008) give a value of 3.13 g/100 g for total lipids of the original dry matter in the m. longissimus thoracis of AA breed bulls, aged 19 months and fed a concentrate-based diet. In our case, the value of fat for some breeds was higher (AA, BA, MS), for others it was lower (Lim, Gas, Gall, Čs), and for Salers it was identical.

From the results of the presented analysis (Table 1), we can infer that nutritionally speaking, the muscle of a bull is primarily an important source of protein. However, significant differences were discovered among the breeds. The highest values of nitrogenous substances were discovered in Lim ( $897.6 \pm 60.36$  g/kg of dry matter). From this viewpoint, this breed can be evaluated as having the highest muscle quality.

Bull muscle also has a high energy value (GE). This finding is primarily

determined by the content of fat, as a highly energetic nutrient. Therefore, the energy level corresponds with the content of muscle fat. The highest fat content was found in the AA muscle ( $171.6 \pm 43.30$  g/kg dry matter), which also had the highest content of energy among the monitored breeds ( $25.1 \pm 0.67$  MJ/kg). In contrast, the lowest value of GE was discovered in Gall ( $23.2 \pm 0.33$  MJ/kg).

### Total content of saturated and unsaturated FA in the muscle fat

For consumers, the effect of diet on health and disease prevention is related to their interest in food containing bioactive substances, and in terms of fats, the content of fatty acids. Based on the presence of individual groups of FA in the muscle fat of the eight monitored breeds of cattle, its quality and dietary features may be assessed.

While no significant differences in the values of  $\Sigma$  SFA were proven in the monitored breeds, several significant ( $P \leq 0.05$ ) differences were confirmed for  $\Sigma$  UFA (Table 2). For all the monitored breeds, with the exception of Gas, there was a higher presence of SFA compared to UFA. As Kameník (2014) claims, hydrogenation of UFA and SFA occurs in ruminants as a result of first stomach digestion, which also influences the composition of somatic lipids and their consistency. A high content of SFA in the meat of bulls raised on pastures is also documented in the results published by Garcia et al. (2008).

A different ratio between UFA and SFA (Table 2) was shown among the breeds. In Gas, this ratio was 1:1.09, meaning that the fat had an almost equal presence of SFA and UFA. In AA, the content of SFA was almost double the content of UFA (ratio 1:0.59).

### Total content of MUFA and PUFA in the muscle fat

Of the unsaturated FA, MUFA predominated, ranging from  $5.4 \times$  (Lim) to  $8.9 \times$  (Sal) more in comparison with PUFA (Table 3) in the muscle fat. The work of Garcia et al. (2008) and Xie et al. (2012) warns of the high content of MUFA in beef meat.

Muscle fat contained the most acids C18:1n9c/C18:1n9t (oleic/elaidic acid) from the MUFA group. In the significant  $\Sigma$  MUFA: $\Sigma$  PUFA ratio, MUFA predominates over PUFA by a ratio of 1:0.11 (Sal) to 1:0.18 (Lim and Gall). Xie et al. (2012) observed the presence of fatty acids in five cattle breeds and reported a significantly higher presence for MUFA (from 26.78 to 36.02 mg/g) compared to PUFA (from 5.05 to 6.96 mg/g), which corresponds with our findings.

Of the PUFA group, n-6 FA were most present in the muscle fat, compared to the n-3 PUFA group of fatty acids. From the n-6 FA group, the muscle fat contained the most linoleic /linolelaidic acid (C18:2n6c/C18:2n6t).

### Total content of n-6 and n-3 fatty acids in the muscle fat

The total content of n-3 PUFA varied in the monitored breeds from  $0.5 \pm 0.16$  g (Sal) to  $1.1 \pm 0.04$  g (Gas) per 100 g of fat (Table 4). Somewhat higher n-3 PUFA values ranging from 1.35 to 2.57 g were observed by Garcia et al. (2008), primarily in bulls on pasture, compared to those grazed with a supplement of corn grain in their diet or those fed a concentrate without access to pasture.

The ratio of n-6/n-3 ranges from 2.94 (Gas) to 6.11 (Gall). In the Holstein and Simmental breeds, Dannenberger et al. (2003) recorded an n-6/n-3 ratio of 1.68 to 2.03. Xie et al. (2012) observed the presence of n-6 and n-3 PUFA in five cattle breeds and recorded significant differences in the presence both of n-6 (4.35–4.98: 6.19 mg/g) and n-3 fat (0.31–0.39: 0.8 mg/g), which complies with our results. It is obvious that the presence of the n-3 PUFA group is significantly lower in the muscle fat of the monitored breeds. Wood et al. (2004) state that the meat of ruminants is a relatively good source of n-3 PUFA with regard to the presence of 18:3 in the grass. A further increase is possible in animals when incorporating linseed or linseed oil, provided it is “protected” against biodegradation. Efforts to increase the presence of PUFA in the meat of ruminants by providing a diet rich in n-3 FA are also described by Raes et al. (2004).

In terms of dietary health, the relationship between  $\Sigma$  n-6: $\Sigma$  n-3 fatty acids is often monitored. A 1:5 ratio is considered optimal. The narrower the ratio is, the higher the quality of the evaluated fat. In the scope of the monitored meat breeds of cattle, the  $\Sigma$  n-6: $\Sigma$  n-3 ratio ranged from 1:2.94 (Gas) to 1:6.11 (Gall). That is, for a specific breed,

there were 3 to 6 times more  $\Sigma$  n-6 PUFA than  $\Sigma$  n-3 PUFA in the fat. The requirement for a higher intake of polyunsaturated fatty acids in human food (Griel et al. 2006; Kris-Etherton 2007) is fulfilled by the Gascon breed (1:2.94). The values were significantly higher (1:4.19 to 1:6.11) in other breeds.

The composition of FA in fatty muscle tissue has a large impact on the meat quality. It determines the strength/oiliness of fat tissue and the meat's oxidation stability, which has an influence on its taste and colour (Wood et al. 2008). Several studies comparing several breeds were done in order to determine the presence of FA in fats in modern cattle breeds (Wood et al. 2004; Garcia et al. 2008; Xie et al. 2012). However, our monitored set of eight breeds is completely exceptional in that multiple modern breeds of beef cattle coming from one location and the same husbandry practices were compared. The different genotypes of the cattle, raised extensively by grazing under the same geographic conditions, influenced the chemical composition of the fat component (especially the presence of individual groups of FA) in the muscle of the slaughtered bulls.

The indicators we studied help predict the biological, dietetic and culinary value of the meat. The results confirmed our hypothesis that within the selected beef cattle breeds there are intermediate differences in the nutritional value of the muscles. Considering the results, it can be generally concluded that dietetically, meat from beef cattle is a high quality food for its substantial content of nutritionally high quality fats, based on the content of different fatty acid groups and their relative proportions.

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