Conjugated Linoleic Acid and Fatty Acids Profile in Buffalo Meat

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ABSTRACT

Aim of the present study was to evaluate the fatty acids profile of buffalo meat. The trial was carried out on 16 buffalo calves fed a total mixed ratio (2.7% body weight) composed by a commercial concentrate, mixed hay and corn silage (CP: 14.9 % DM; 0.91 VFU/kg DM). All the animals were slaughtered when the BW of 350 kg was reached (about 420 d of age). Samples of *Longissimus thoracis* (LT), *Semitendinosus* (ST), *Iliopsoas* plus *Psoas minor* (IP) were analysed for fatty acids profile by gas chromatography (GC). To this purpose total fat was extracted and subsequently turned into methyl esters (FAMEs) by direct methylation. The Atherogenic Index (AI) and the Thrombogenic Index (TI) were also calculated. The muscle type highly affected the results: ST showed the most favourable fatty acids profile and consequently, lower values for both index (AI: 0.60, 0.42, 0.57 and TI: 1.50, 1.02, 1.51, for IP, ST and LT, respectively) as well as CLAs contents (0.20, 0.25, 0.21 g/100g; for IP, ST and LT, respectively). In each case, the results of present trial should confirm the favourable assessment of the nutritional characteristics of the meat from buffalo young bulls.

Keywords: Meat, Italian Mediterranean Buffalo, fatty acid profile, CLA.

INTRODUCTION

Ruminant meat has been longer criticized for its high levels of mutagens and carcinogens (Belury, 1995) and considered one of the factors that may lead to the development of human cardiovascular diseases, obesity, hypertension, and cancer, especially due to the high levels of saturated fatty acids (SFA) and cholesterol. However, mainly in the case of grass-fed ruminants, meat and milk are the best dietary sources of CLAs, which has been shown to have health promoting effects, as well as immunomodulating and anticarcinogenic activity (Pastuschenko et al., 2000; Whigham et al., 2000). Additionally, CLAs has been reported to reduce atherosclerosis and total serum cholesterol (Lee et al., 1994). The CLAs are a group of positional and geometric fatty acid isomers derived from linoleic acid (Parodi, 1999) of which the main representative isoform is the cis-9, trans-11 CLA, known as rumenic acid (Kramer et al., 1998). Comparing bovine and buffalo meat, Infascelli et al. (2004) reported several favourable nutritional characteristics for buffalo meat: lower SFA and higher monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) percentages. Recently, Giordano et al. (2010) found that consumption of buffalo meat is associated with several beneficial effects on cardiovascular risk profile, including lower carotid atherosclerotic burden and susceptibility to oxidative stress. Aim of the present study was to evaluate the fatty acids profile and, in particular, CLA content of the meat in the Italian Mediterranean Buffalo bred in an intensive system.

MATERIAL AND METHODS

The trial was carried out on 16 Italian Mediterranean buffalo calves (age 30 d, BW 55 kg) that received 6 l/head/d of acidified milk replacer (180 g/l of water) until 56 d. Subsequently, the replacer amount was gradually decreased, whilst administering the same volume. Roughly chopped

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alfalfa hay and weaning concentrate were available from the fifth week of life and corn silage was administered from 70 d of age. After weaning until the trial beginning, the animals were fed ad libitum hay and corn silage; the concentrate was administered in the amount of 2 kg/d. At the age of 84 d (average 30 kg BW), each animal was placed in individual box up to the slaughtering weigh and was fed (2.7% body weight) a total mixed ratio (CP: 14.9 % DM; 0.91 VFU/kg DM) composed by a commercial concentrate, mixed hay and corn silage. All the animals were slaughtered in an authorized slaughterhouse according to EU legislation (EU Regulation EC No 882/2004) when the BW of 350 kg was reached (about 420 ± 59.4 d of age). Samples of *Longissimus thoracis* (LT), Semitendinosus (ST), Iliopsoas plus Psoas minor (IP) muscles were collected seven days (at 3-5°C) after slaughter, to simulate what usually is done in Italy to improve meat tenderness and water holding capacity (Cutrignelli et al., 2008a). Fatty acids profile was analysed by gas chromatography (Chiofalo et al., 2011). To this purpose total fat was previously extracted (Folch et al., 1957) and subsequently turned into methyl esters (FAMEs) by direct transesterification (Christie, 1993). The FAMEs were analyzed by GC-FID (Agilent Technologies 6890N, Palo Alto, CA, U.S.A.) with a split/splitless injector, a flame ionization detector and fused silica capillary column Omegawax 250 (Supelco, Bellefonte, PA, U.S.A.), 30m x 0.25mm I.D., 0.25 µm film thickness. Column temperature was programmed: initial isotherm of 160°C (6 min.), increment of 3°C/min and final isotherm of 250°C (30 min.). Temperature of the injector and detector: 250°C. Injection volume: 1.0 µL. Carrier gas: helium (1 mL/min). Split ratio: 1:50. Identification of fatty acids was made by comparing the relative retention times of FAME peaks from samples with standards from Supelco (Bellefonte, PA, U.S.A.). Chromatogram peak areas were acquired and calculated by Chemstation software (Agilent, Palo Alto, CA, U.S.A.). The concentration of each fatty acid was expressed as g/100 g, considering 100 g the summation of the areas of all fatty acid methyl esters identified. For each sample the chromatographic analysis was replicated three times. Identification of fatty acids was made by comparing the relative retention times of FAME peaks from samples with an external standards (Supelco). The Atherogenic Index (AI) and the Thrombogenic Index (TI) were also calculated as follows:

$$AI = \frac{C12:0 + (4 \times C14:0) + C16:0}{n - 6 PUFA + n - 3 PUFA + MUFA}$$
$$TI = \frac{C14:0 + C16:0 + C18:0}{(0.5 \times MUFA) + (0.5 \times n - 6 PUFA) + (3 \times n - 3 PUFA) + (n - 3 PUFA/n - 6 PUFA)}$$

were MUFA and PUFA were expressed as g/100 g. Data set were statistically processed using the SAS (2000) GLM procedure.

RESULTS AND DISCUSSION

The muscle type highly affected the results (Table 1). The semitendinosus (ST) showed the most favourable fatty acids profile: lower SFA, higher PUFA, MUFA, ω -3, ω -6, and consequently, lower values for both index (AI: 0.60, 0.42, 0.57 and TI: 1.50, 1.02, 1.51, for IP, ST and LT, respectively). In each case, these last values are particularly interesting; indeed the AI was lower than those reported by Ulbricht and Southgate (1991) for raw minced beef, Cutrignelli et al (2008b) for Marchigiana meat, as well as TI was lower than that reported by Cutrignelli et al (2008b). As concerns the CLAs, also in this case the ST had the more favourable results. Indeed, the contents of either the single isomers or of the total CLA were significantly higher in this muscle compared to the LT and IP. In each case, the average value found in this trials were very interesting (0.23 g/100g), taking into account that meat was from animals bred in intensive system, without use of pasture or linseed in the diet, and close to the data reported by other authors for Italian Mediterranean buffaloes (Infascelli et al., 2004). It is well known that the presence of CLA in the intramuscular lipids of large ruminants depends, in part, on the ruminal biohydrogenation of the diets LA (Bauman et al., 1999). As reported by de Mendoza et al. (2005), many authors recorded differences in rumen microbial metabolism, rumen ciliate protozoa population, forage-eating behaviour, consumption and rumination, development and size of the rumen-reticulum between

domestic buffalo as compared to cattle. These data suggest that the rumen supply of CLA readily available for intestine absorption and further deposition in the intramuscular lipids of these herbivorous would also differ. In conclusion, the results of present trial should confirm the favourable assessment of the nutritional characteristics of the meat from buffalo young bulls.

REFERENCES

- Bauman, D.E., L.H. Baumgard, B.A. Corl and J.M. Griinari. 1999. Biosynthesis of conjugated linoleic acid in ruminants. Proc. American Society of Animal Science (pp. 1–15), Ithaca, NY.
- Belury, M.A. 1995. Conjugated dienoic linoleate: A polyunsaturated fatty acid with unique chemoprotective properties. Nutrition Reviews, 53, 83–89.
- Chiofalo B., V. Lo Presti, G. Savoini, E. D'Alessandro, V. Chiofalo and L. Liotta. 2011. Nucleotides administration in broiler chichen diet: Effect on breast muscles quality. Czech. J. Food Sci., 29 (4): 308-317.
- Christie W.W. 1993. Preparation of ester derivatives of fatty acids for chromatographic analysis. 69–111. In W.W. Christie (ed.) Advances in lipid methodology, 2nd ed. Oily Press, Dundee, UK.
- Cutrignelli M.I., G. Piccolo, F. Bovera, S. Calabrò, S. D'Urso, R. Tudisco and F. Infascelli. 2008a. Effects of protein source and energy value of diet on the performance of young Marchigiana bulls: 1 *Infra vitam* performance and carcass quality. Ita. J. of Anim. Sci., 7: 259-270.
- Cutrignelli M.I., S. Calabrò, F. Bovera, R. Tudisco, S. D'Urso, M. Marchiello, V. Piccolo and F. Infascelli. 2008b. Effects of protein source and energy value of diet on the performance of young Marchigiana bulls: 2. meat quality. Ita. J. of Anim. Sci., 7: 271-285.
- de Mendoza M.G., L.A. de Moreno, N. Huerta-Leidenz, S. Uzcàtegui-Bracho, M.J. Beriain and G.C. Smith. 2005. Occurrence of conjugated linoleic acid in longissimus dorsi muscle of water buffalo (Bubalus bubalis) and zebu-type cattle raised under savannah conditions. Meat Science 69: 93–100.
- Folch J., M. Lees, and Sloane Stanley. 1957. A simple method for the isolation and purification of total lipides from animal tissues. J Biol Chem., 226(1): 497-509.
- Giordano G., P. Guarini, P. Ferrari, G. Biondi-Zoccai, B. Schiavone and Giordano A. 2010. Beneficial impact on cardiovascular risk profile of water buffalo meat consumption. Europ. J Clin. Nutr., 64: 1000–1006.
- Infascelli, F., S. Gigli and G. Campanile. 2004. Buffalo meat production: Performance infra vitam and quality of meat. Veterinary Research Communications. 28 (suppl. 1): 143-148.
- Kramer, J.K.G., P.W. Parodi, R.G. Jensen, M.M. Mossoba, M.P. Yurawecz and R.O. Adlof. 1998. Rumenic acid: a proposed common name for the major conjugated linoleic acid isomer found in natural products. Lipids, 33: 835.
- Lee, K.N., D. Kritchevsky and M.W. Pariza. 1994. Conjugated linoleic acid and atherosclerosis in rabbits. Atherosclerosis, 108: 19–25.
- Parodi P.W. 1999. Conjugated linoleic acid and other anticarcinogenic agents of bovine milk fat. J. Dairy Sci., 82: 1339–1349.
- Pastuschenko V., H.D. Matthes, T. Hein and Z. Holzer. 2000. Impact of cattle grazing on meat fatty acid composition in relation to human nutrition. In: Alföldi, T., Lockeretz, W., Niggli, U. (Eds.), Proc. 13th Int. IFOAM Scientific Conf., Basel, Switzerland, 293–296.
- SAS. 2000 SAS/STAT User's Guide, Version 8.2, Vol. 2, 4th Edition. SAS Institute Inc., Cary, NC.
- Whigham L.D., M.E. Cook and R.L. Atkinson. 2000. Conjugated linoleic acid: implications for human health. Pharmacol. Res., 42: 503–510.

	IP	ST	LT	MSE
C14:0	1.42 ^A	0.92 ^B	1.31 ^A	0.026
C16:0	21.6 ^A	20.3 ^B	21.0^{AB}	0.955
C18:0	27.5 ^A	20.0^{B}	27.2^{A}	2.50
C18:1n-9	27.8^{B}	33.1 ^{Aa}	31.2 ^{Ab}	5.11
C18:1-cis11	1.17^{B}	1.48^{A}	1.16 ^B	0.012
C18:2n-6	8.12 ^{Ab}	9.12 ^{Aa}	6.47 ^B	1.04
C18:3n-6	0.09^{B}	0.14^{A}	0.09^{B}	0.0069
C18:3n-3	0.56^{b}	0.66^{a}	0.51^{b}	0.0073
cis9-trans11 CLA	0.05^{b}	0.06^{a}	0.04^{b}	0.00017
trans10-trans12 CLA	0.15^{b}	0.19^{a}	0.17^{ab}	0.0017
SFA	50.0^{A}	42.2^{B}	49.9 ^A	1.12
MUFA	28.4 ^B	34.2 ^A	32.0 ^A	4.9
PUFA	9.52 ^A	10.50 ^A	8.56 ^B	0.87
Σ CLA	0.20^{b}	0.25^{a}	0.21^{b}	0.0001
AI	0.60 ^A	0.42 ^B	0.57^{A}	
TI	1.50 ^A	1.02 ^B	1.51 ^A	

Table 1. Fatty acid profile (g/100 g) in the three muscles.

IP: *Iliopsoas* plus *Psoas minor*; ST: *Semitendinosus*; LT: *Longissimus thoracis*. MSE: Mean square error. Along the row: A,B,C: P<0.01 and a,b,c: P<0.05.