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# Effect of high or low protein ration combined or not with rumen protected conjugated linoleic acid (CLA) on meat CLA content and quality traits of double-muscled Piemontese bulls

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#### ABSTRACT

A trial was carried out on double-muscled Piemontese bulls to evaluate the effects of two rations differing in crude protein density (HP = 14.5% DM and LP = 10.8% DM) and top dressed or not with 80 g/d of rumen protected CLA (rpCLA) for a long period (336 d) on meat quality traits and CLA content. Forty-eight bulls were fed one of the four experimental diets based on corn silage and cereals and were slaughtered at an average age and body weight (BW) of  $562 \pm 18$  d and  $668 \pm 56$  kg, respectively. After slaughter the 5th rib cut was dissected into *Longissimus thoracis* (LT), other muscles (OM), inter-muscular fat (IF), cover fat (CF), and bones. Muscles and fatty tissues were analyzed for proximate composition and fatty acid (FA) profiles. Rib was composed by 81.1, 3.7, 1.6 and 13.6% of muscles, IF, FC and bone, respectively; LT and OM contained only 0.8 and 1.4% of lipid, respectively. The treatments did not influence these values, but rpCLA increased, compared to control, both c9,t11-CLA and t10,t10

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#### 1. Introduction

Constraints introduced by the Nitrate Directive of the European Union and the cost of soybeans are forcing farmers to evaluate the use of low protein diets (Xiccato, Schiavon, Gallo, Bailoni, & Bittante, 2005: Yan, Frost, Keaty, Agnew, & Mayne, 2007), Some farmers are using rumen protected CLA (rpCLA) in combination with low protein (LP) diets. The use of CLA is thought to be beneficial because it decreases fat deposition and slightly increases lean tissue growth and feed efficiency in non-ruminant animals (Park, Albright, Liu, Storkson, Cook, & Pariza, 1997) and, to a lesser extent, in beef cattle (Gillis, Duckett, & Sackmann, 2004). Park et al. (1997) and Pariza, Park, and Cook (2001) suggested that CLA could exert some metabolic proteinsparing effects, but unlikely these effects would be observed on conventional cattle kept on conventional diets. These effects could be more likely be evidenced with animals having high protein requirements fed with low protein diets. Compared with other cattle, double-muscled (DBM) young bulls have less bone, less fat, more muscle, a greater dressing percentage and a reduced intake (Arthur, 1995). The reduced intake and the greater potential for lean growth suggested that DBM bulls require diets with increased energy and especially protein density (Fiems, De Campeneere, Bogaerts, Cottyn, & Boucqué, 1998). The protein-sparing effects due to rpCLA might be better exploited using DBM bulls under condition of protein restriction, not only because of their great potential for lean gain, but also because these animals have very poor aptitude for fat gain, so that confounding effects of rpCLA on body fatness and meat fat content are presumably low. In a recent work conducted on DBM Piemontese young bulls, we found that nearly 1 year of administration of a low protein diet (CP = 10.8% DM) had no detrimental effect on average daily gain (ADG) and dry matter intake (DMI) compared to a conventional protein ration (CP = 14.5% DM), but rpCLA addition increased feed efficiency in low protein rations, but not in conventional ones (Dal Maso, Schiavon, Tagliapietra, Simonetto, & Bittante, 2009; Schiavon, Tagliapietra, Dal Maso, Bailoni, & Bittante, 2010). Very few information about the meat quality traits of DBM Piemontese bulls have been published and it is not clear if the level of CP, the addition of rpCLA or their combination will influence the quality of their meat, especially in terms of fatness, fatty acid profile and CLA content. Thus this work was done in order: i) to provide information about meat quality traits of DBM Piemontese bulls; ii) to evaluate if a strong reduction of the dietary CP content, with or without rpCLA addition,

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has consequences on the meat quality of these animals; and iii) to test if the addition of rpCLA alters the CLA content and the fatty acid profile of the fat and lean tissues dissected from the 5th rib.

#### 2. Material and methods

### 2.1. Experimental procedures

This study was part of a trial aimed to investigate the effects of corn-silage and cereals based rations with different protein levels and top-dressed or not with rpCLA on performance, N excretions and meat quality traits of Piemontese double-muscled bulls. The project was approved by the "Ethical Committee for the care and the use of experimental animals" of the University of Padua. The trial was described by Dal Maso et al. (2009) and by Schiavon et al. (2010) who also reported the main results obtained in vivo and on the carcass. Briefly, 48 intact Piemontese young bulls were weighed and divided in four experimental groups of 12 animals each, balanced by body weight (BW) and age. The bulls were housed in 12 fully slatted floor pens with four animals each. After 28 days of adaptation, the animals of each group were fed one of 4 experimental diets resulting from the combination of two crude protein (CP) densities (High protein, HP: CP = 14.5% DM and Low protein, LP: CP = 10.8% DM) and two top dressed additives (A), rpCLA or hydrogenated soybean oil (HSO). Bulls were weighed monthly and slaughtered after 332 days of trial at  $562 \pm 18$  d of age and  $668 \pm 56$  kg BW.

High protein ration was composed, on DM basis, of corn meal (36.0%), corn silage (25.0%), soybean meal (12.6%), dried sugar beet pulp (10.2%), wheat bran (6.3%), wheat straw (6.0%), vitamin and mineral mix (2.4%), calcium soap (0.8%) and hydrogenated soybean oil (0.7%). LP ration did not differ for ingredient composition except for the levels of inclusion of soybean meal that was 3.3 instead of 12.6% DM, while the other ingredients were proportionally increased. Calcium soap of hydrogenated palm oil was included at the same level in all the rations as source of energy. The two rpCLA groups received 80 g/d of rpCLA. The commercial rpCLA supplement (SILA, Noale, Italy), was manufactured by binding methyl esters of CLA to a silica matrix, and then coating this complex with hydrogenated soybean oil, which contained fatty acids in the triglyceride form. The two HSO groups received 65 g/d of HSO, in addition to the HSO included in the base diet, with the aim of supplementing the same amount of HSO used to coat CLA in the rpCLA.

The composition of Ca-soap, of HSO and RPCLA, resulting from the chemical analysis, is given in Table 1. Ca-soap is rich in saturated (SFA) and monounsaturated (MUFA) fatty acids (42.1 and 30.6% total FA of C16:0 and C18:1, respectively), while HSO is almost completely represented by SFA (C18:0 and C16:0). These fat supplements, because of their low content of C18:2 and C18:3, precursors of *cis*-9

**Table 1**Composition of calcium soap, hydrogenated soybean oil (HSO) and rumen protected CLA (rpCLA).

Item	Ca-soap	HSO	rpCLA
Dry matter, %	96.0	99.9	98.4
Lipids, % as fed	81.1	99.0	80.0
Ash	14.9	0.2	17.8
Fatty acids, % of dietary lipids			
C16:0	42.1	10.3	8.5
C18:0	4.9	77.2	48.5
C18:1 cis	30.6	0.1	8.5
C18:1 trans	-	-	0.2
C18:2	7.4	0.1	0.8
c9, t11-CLA	_	_	9.9
t10, c12-CLA	_	_	9.6
C20:0	2.7	-	0.4
Other fatty acids	1.1	1.0	1.5
Total fatty acids	88.7	88.7	87.9

*trans*-11 CLA, were used to minimize possible interferences due to formation of CLA in the rumen.

Tabled values for the various feed ingredients (Sauvant, Perez, & Tran, 2004) and chemical analysis for fat supplements were used to compute the nutritional value and the fatty acids (FA) profile of whole rations (Table 2). The HSO rations contained about 43.9, 19.4 and 36.7% of total FA as SFA, MUFA and polyunsaturated fatty acids (PUFA), respectively. The replacement of HSO with rpCLA in the rations increased the proportion of PUFA from 36.7 to 40.8% total FA, at the expense of SFA. The daily consumptions of top dressed *c*9, *t*11-CLA and of *t*10, *c*12-CLA were 5.57 and 5.41 g/d, respectively.

## 2.2. Sampling and analysis

When the bulls reached approximately 660 kg of BW ( $668\pm56$  kg) the animals were slaughtered (age at slaughter:  $562\pm18$  d). No significant differences of final BW and average daily gain due to the feeding treatment were observed, even if the growth rate was significantly lower with LP diets during the first 4 months of trial, followed by a compensatory growth afterwards (Schiavon et al., 2010).

Twenty-four hours after slaughter the whole cut of the 5th rib was collected. The entire rib was vacuum packed, moved to the laboratory, and aged at 4 °C in a chilling room for 10 days.

After aging, drip losses were assessed as the ratio of the difference between the wet and the dried empty bag and the weight of the rib. Muscles pH was measured using a Delta Ohm HI-8314 pH-meter (Delta Ohm, Padova, Italy) 10 days *post-mortem*.

Color parameters were measured, after 1 h of air exposure, on *Longissimus thoracis* muscle (LT) using a Minolta CM-508c (illuminate: D65, Observer:  $10^{\circ}$ ) on 5 anatomical positions and the mean was taken as final value. Meat color was expressed according to the CIE-Lab color space by reporting  $L^*$ ,  $a^*$  and  $b^*$  values (CIE, 1978).

The rib was dissected into muscles (LT and remaining muscles, OM), inter-muscular fat (IF), cover fat (CF) and bones. Each fraction was weighted and sampled for the analysis.

**Table 2**Chemical composition, nutritional values and fatty acids composition of rations.

Item	Feeding treatment <sup>a</sup>			
	HP <sub>HSO</sub>	$HP_{rpCLA}$	$LP_{HSO}$	$LP_{rpCLA}$
Chemical composition, % DM:				
CP	14.5	14.5	10.8	10.8
Lipids	4.1	4.1	4.2	4.2
NDF	28.7	28.7	30.3	30.3
Net energy for maintenance and gain, MJ/kg DMb	7.8	7.8	7.7	7.8
Fatty acids content, % of total fatty acids <sup>c</sup> :				
SFA <sup>d</sup>	43.9	38.1	43.4	37.8
C14:0	0.2	0.2	0.2	0.2
C16:0	16.7	16.4	17.1	16.8
C18:0	27.1	21.5	26.2	20.8
MUFA <sup>d</sup>	19.4	21.2	19.8	21.5
C16:1	0.1	0.1	0.1	0.1
C18:1	19.3	21.0	19.6	21.3
PUFA <sup>d</sup>	36.7	40.8	36.8	40.8
C18:2	34.5	34.8	34.8	35.1
c9, t11-CLA	0.0	1.9	0.0	1.9
t10, c12-CLA	0.0	1.9	0.0	1.8
C18:3	2.1	2.1	2.0	2.0
Other FA	< 0.1	< 0.1	< 0.1	< 0.1

 $<sup>^{\</sup>rm a}$  HP and LP diets contained 145 and 108 g of CP/kg, respectively; HSO =65 g/d of top dressed hydrogenated soybean oil; rpCLA =80 g/d of top dressed commercial CLA (product coated with hydrogenated soybean oil; SILA, Noale, Italy).

<sup>&</sup>lt;sup>b</sup> Values were computed, according to the French energy system (Sauvant et al., 2004), from actual chemical composition of feed.

<sup>&</sup>lt;sup>c</sup> Values were computed considering: tabled values for feed ingredients (Sauvant et al., 2004), average DM intake recorded during the trial = 8.79 kg DM/d, amount of top dressed HSO or rpCLA and their composition resulting from chemical analysis.

 $<sup>^{\</sup>rm d}$  SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.

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