Effects of fat source and dietary sodium bicarbonate plus straw on the conjugated linoleic acid content of milk of dairy cows

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Abstract

The effects of fat source (0.7 kg of fatty acids from extruded soybeans or palmitic acid), of sodium bicarbonate (0.3 kg) plus straw (1 kg) and the interaction of these treatments on the content of conjugated linoleic acid (CLA) in the milk of dairy cows were examined. During nine weeks a group of 10 cows received a ration with palmitic acid and bicarbonate plus straw (ration PAB). During three periods of three weeks a second group of 10 cows received successively a ration with extruded soybeans and bicarbonate plus straw (ration ESB), a ration with palmitic acid without bicarbonate or straw (ration PA), and a ration with extruded soybeans without bicarbonate or straw (ration ES). Rations ES and ESB increased the content of polyunsaturated fatty acids in milk, but decreased milk fat content, compared to rations PAB and PA. Ration ESB led to the greatest milk CLA content, by a synergy between the high amount of dietary fat, and the action of bicarbonate plus straw, favouring *trans*11 isomers of CLA and C18:1, presumably via a ruminal pH near neutrality. Ration ES favoured *trans*10 isomers, not desaturated in the mammary gland, so that the milk CLA content was lower than with ration ESB, and resulted in the lowest milk fat content. In conclusion, a ration supplemented with both extruded soybeans and bicarbonate plus straw, was an efficient way to increase the CLA content in the milk of dairy cows.

Keywords: *Conjugated linoleic acid, dairy cow, milk, extruded soybeans, sodium bicarbonate, straw*

1. Introduction

Conjugated linoleic acids (CLA) are a group of positional and geometric isomers of linoleic acid (C18:2). Among them, C18:2 *cis*-9,*trans*-11 (c9t11-CLA) and C18:2 *trans*-10,*cis*-12 (t10c12-CLA) are presumed to have beneficial dietetic properties for humans, mainly against cancer (Parodi 1999; Whigham et al. 2000; Ip et al. 2002). However, t10c12-CLA can have also adverse effects in humans (Riserus et al. 2002). CLA can be found in many human foods,

among which dairy products have the highest concentrations. In dairy cows, CLA is synthesized in the rumen during biohydrogenation of C18:2 by isomerization of C18:2 (Harfoot et al. 1973), and in the mammary gland by desaturation of *trans*-vaccenic acid (t11-C18:1), another intermediate of biohydrogenation of polyunsaturated fatty acids (PUFA) (Griinari et al. 2000).

The amount of CLA in milk can be enhanced by different ways (Troegeler-Meynadier & Enjalbert 2005). The addition of C18:2 in the rumen systematically increased the concentration of CLA in digesta content and milk (Beam et al. 2000; Dhiman et al. 2000; Kim et al. 2000; Chouinard et al. 2001; Troegeler-Meynadier et al. 2003). A rumen pH near neutrality favours the *in vitro* production of CLA and t11-C18:1 (Martin & Jenkins 2002; Troegeler-Meynadier et al. 2003). *In vivo*, without added fat, a high forage ration increases CLA and t11-C18:1 in milk, compared with a high concentrate ration (Dhiman et al. 1999). On the contrary, when unsaturated fat is added to the ration, the increase of CLA and t11-C18:1 in milk and duodenum content is higher with a high concentrate ration than with a high forage ration (Piperova et al. 2002). Moreover, ruminal environment influences the concentration of CLA and the profile of *trans* octadecenoic acids isomers (t-C18:1) (Piperova et al. 2000; Troegeler-Meynadier et al. 2003): low rumen pH and added dietary concentrates favour *trans*10 (t10) isomers, including t10c12-CLA, known to reduce milk fat content and mammary desaturase activity (Baumgard et al. 2000). Taken as a whole, the published literature demonstrates the importance of rumen pH on CLA formation, which is related to the forage/concentrate ratio, and that this effect depends on fat addition. But until now, a possible interaction between the level of unsaturation of fat and rumen pH was not studied.

The purpose of the present study was to investigate the effects of a combination of straw and sodium bicarbonate, as means to maintain a high rumen pH, and of level of unsaturated fat (palmitic acid vs. extruded soybeans), on CLA content of milk from dairy cows, and to investigate a possible interaction between these two factors.

2. Materials and methods

2.1. Experimental design and feeding

Twenty lactating Holstein cows received maize silage (296 g DM/kg; per kg DM: 63 g CP, 249 g starch, 495 g NDF and 288 g ADF) *ad libitum*, completed with one of four different mixtures shown in Table I, so that cows were assigned to a ration with palmitic acid and sodium bicarbonate plus straw (PAB), a ration with palmitic acid without sodium bicarbonate or straw (PA), a ration with extruded soybeans and sodium bicarbonate plus straw (ESB), or a ration with extruded soybeans without sodium bicarbonate or straw (ES). The mixtures were top-dressed on the maize silage in order to obtain a total intake of the mixture by the cows. The quantity of maize silage ingested by cows was recorded daily on a pen basis. Palmitic acid (90% purity) was added in order to examine the effect of the level of unsaturation independently on the effect of amount of added FA, which was 0.7 kg with each ration.

Cows were allocated to two groups according to BW, parity, days in milk (average 89 days for Group 1 and 96 days for Group 2), milk production, milk fat and protein contents. Each group contained 6 primiparous and 4 multiparous cows. The experiment comprised 3 successive periods of 3 weeks: Group 1 received ration PAB during the whole experiment of 9 weeks, and Group 2 received successively rations ESB, PA and ES. Each period comprised 20 days adaptation; samples were taken at day 21. Each ration was distributed in 2 equal meals, at 07:00 and at 17:00 h. Cows were group fed, feed offered and feed refused were recorded daily during the last week of each experimental period.

Table I. Ingredients, nutrient composition and fatty acid pattern of the four rations offered to dairy cows.

Fat source	Palmitic acid		Extruded soybeans	
Bicarbonate + straw	+	−	+	−
Diets	PAB	PA	ESB	ES
Ingredients [kg DM per day]				
Wheat	3.5	3.5	3.5	3.5
Soybean meal	3.8	3.8	0.9	0.9
Straw	1.0	0.0	1.0	0.0
Extruded soybeans	0.0	0.0	3.5	3.5
Palmitic acid	0.7	0.7	0.0	0.0
Sodium bicarbonate	0.3	0.0	0.3	0.0
Mineral-vitamin mix[†]	0.2	0.2	0.2	0.2
Nutritive value [g/kg DM]				
Crude proteins	241.1	274.5	228.5	260.5
NDF	240.3	182.3	277.8	224.6
ADF	111.3	68.1	118.6	75.8
Starch	253.1	290.7	256.1	294.7
Total fatty acids	80.8	92.4	82.8	94.9
Fatty acid profile [% of total fatty acids]				
C16:0	82.3	82.5	11.0	10.9
C18:0	1.0	1.0	4.0	4.0
C18:1 *cis*9	2.7	2.6	17.0	17.1
C18:2	10.6	10.6	54.6	54.7
C18:3	1.5	1.5	10.4	10.4

[†]Contained per kg: P, 50 g; Ca, 140 g; Na, 60 g; Zn, 4 g; Mn, 3.2 g; Fe, 3 g; Cu, 0.8 g; vitamin A, 250 800 IU; vitamin D_3, 62 700 IU, vitamin E, 112 IU.

2.2. Samples

Milk of cows was collected at the evening milking, the last day of each period, since Engle et al. (2001) showed that there were no differences in fatty acid composition between the morning and the evening milk samples. Milk samples were weighed and quickly frozen at −18°C. Then they were transferred to the laboratory under cold conditions, to be freeze-dried (Virtis Freezemobile 25; Virtis, Gardiner, NY, USA), weighed and homogenized. They were kept at −18°C until analysis.

Milk production was recorded daily during the last week of each period, and samples of milk were taken at day 17, 19 and 21 for fat and protein determination.

2.3. Analytical procedures

Chemical compositions of maize silage and of the mixtures were determined according to the Association française de normalisation (AFNOR) procedures: NF V18-109 (AFNOR 1981) for DM; NF V18-100 (AFNOR 1977) for crude proteins; NF V18-121 (AFNOR 1997a) for starch; NF V18-122 (AFNOR 1997b) for NDF/ADF (assayed with a heat stable amylase and expressed with exclusive of residual ash).

The FA of dried milk and feed samples were extracted and methylated *in situ* according to the procedure of Park and Goins (1994), except that the solution of 14% borontrifluoride in methanol was replaced by a solution of methanol-acetylchloride (10:1). C19:0 was used as an internal standard. The FA methyl esters were then quantified by gas chromatography (Agilent 6890N, equipped with a model 7683 auto injector, Network GC System, Palo Alto,

California, USA). The column was a fused silica capillary (CPSil88, 100 m × 0.25 mm ID, 0.20 μm film thickness; Chrompack-Varian, Middleburg, The Netherlands). A first analysis was made using the method described by Enjalbert et al. (2003), and a second analysis was realized to separate t10-C18:1 and t11-C18:1, as described by Troegeler-Meynadier et al. (2003). Peaks were identified and quantified by comparison with commercial standards (Sigma, St Louis, MO, USA), except t10-C18:1, which was identified by order of elution.

Fatty acids were expressed as percentage of total FA. The CLA isomers assayed were t10c12-CLA and c9t11-CLA, and CLA will refer to the sum of these two isomers. The t-C18:1 assayed were t10-C18:1 and t11-C18:1 and the term t-C18:1 will refer to the sum of these two isomers. The other t-C18:1 isomers did not co-elute with t10-C18:1 and t11-C18:1, but were not determined. The t10/t11 ratio was calculated by dividing the percentage of t10 isomers (t10-C18:1 plus t10c12-CLA) by the percentage of *trans*11 (t11) isomers (t11-C18:1 plus c9t11-CLA). True protein contents and milk fat content were determined by infrared analysis (Milkoscan 605, Foss Electric, F-75001 Paris, France).

2.4. Adjustment of data and statistical analysis

The data obtained from periods 2 and 3 of Group 1, receiving the ration PAB, were used for adjustment. A corrective factor for period effect was calculated for each variable and each period:

$$\text{Corrective factor for period 2 (C2)} = \ln \text{ (mean of value from period 1)}$$
$$- \ln \text{ (mean of value from period 2)}$$

$$\text{Corrective factor for period 3 (C3)} = \ln \text{ (mean of value from period 1)}$$
$$- \ln \text{ (mean of value from period 3)}$$

Each value obtained from Group 2 during periods 2 and 3 was corrected as follow:

$$\text{Period 2 : } \ln \text{ (corrected value from period 2)} = \ln \text{ (value from period 2)} + \text{C2}$$

$$\text{Period 3 : } \ln \text{(corrected value from period 3)} = \ln \text{ (value from period 3)} + \text{C3}$$

The data from period 1 of Groups 1 (ration PAB) and 2 (ration ESB), and the corrected data from periods 2 (ration PA) and 3 (ration ES) of Group 2, were analysed with SYSTAT (version 9, SPPS Inc., 1998 Chicago) according to the model:

$$\text{Variable} = \text{Mean} + \text{Fat effect} + \text{Bicarbonate effect} + \text{Fat} \cdot \text{Bicarbonate interaction} + \varepsilon$$

A linear regression was computed between the milk fat content and the percentage of t10 isomers. Multiple linear regressions were computed between milk fat content and all FA with eighteen carbons (18C FA) assayed. Differences were considered significant at $p < 0.05$.

3. Results

Cows receiving rations PAB, ESB, PA and ES, ingested 5.4, 5.9, 5.5 and 5.7 kg of maize silage (on a DM basis), respectively.

Table II presents milk production and composition. There was no significant effect of rations on milk production and protein content. Extruded soybeans significantly decreased

Table II. Milk production, milk protein and fat content of dairy cows and significance of the effects of fat source (Fat), of bicarbonate and straw addition (B), and of their interaction (Fat × B).

Fat source	Palmitic acid		Extruded soybeans			p		
Bicarbonate + straw	+	−	+	−				
Diets	PAB ($n=30$)	PA ($n=10$)	ESB ($n=10$)	ES ($n=10$)	SEM*	Fat	B	Fat × B
Milk production [kg/d]	33.33	34.01	36.01	35.33	1.30	0.13	1.00	0.61
Milk protein content [g/kg of raw milk]	29.54	29.38	29.19	29.26	0.45	0.60	0.92	0.81
Milk fat content [g/kg of raw milk]	47.14	42.97	39.48	36.47	1.46	<0.01	0.02	0.70

*SEM = Standard error of the mean.

milk fat content, with a significant linear relationship ($r^2 = 0.549$, $p < 0.01$) between milk fat content and percentages of the different 18C FA in milk fat and a negative slope for the unsaturated 18C FA:

$$\text{Milk fat content} = 50.25 + 0.96\ \text{C18:0} - 0.27\ \text{C18:1}\ cis9 - 1.52\ \text{t} - \text{C18:1} - 2.59\ \text{CLA} - 1.20\ \text{C18:2} - 0.72\ \text{C18:3}$$

Rations without bicarbonate or straw led to a lower milk fat content. Moreover, there was a significant linear relationship between milk fat content and t10 isomers percentage (milk fat content = 44.97 − 2.91 t10 isomers, $r^2 = 0.402$, $p < 0.01$). On the contrary, there was no significant interaction between fat source and bicarbonate on milk fat content, but an additive effect could be observed.

The fatty acids profile (not including intermediates of biohydrogenation) of milk of dairy cows is presented in Table III. Extruded soybeans increased the percentage of all 18C FA. In the milk fat of cows receiving extruded soybeans, C6:0, C8:0, C10:0, C12:0 and C14:0 proportions were not significantly affected, contrary to the C14:1, C15:0, C16:0 and C16:1 percentages, which were decreased compared with milk of cows receiving palmitic acid. Rations PA and ES, without sodium bicarbonate or straw, led to significantly higher C11:0 and C16:1 percentages, and tended to increase C18:2 ($p = 0.12$) and t-C18:1 ($p = 0.11$) percentages compared with rations PAB and ESB. There was a tendency toward an interaction between fat source and bicarbonate plus straw for percentages of C16:1 ($p = 0.081$) and CLA ($p = 0.066$), where ration PA caused the highest C16:1 percentage and ration ESB the highest CLA percentage.

The percentages of intermediates of C18:2 biohydrogenation are presented in Table IV. The addition of extruded soybeans led to an increase of all intermediates of C18:2 biohydrogenation, but only rations ESB and ES led to a measurable secretion of t10c12-CLA in milk. Rations PA and ES, without sodium bicarbonate or straw, caused a significantly higher percentage of t10 isomers and t10/t11 ratio, and tended to decrease the percentage of t11-C18:1 ($p = 0.067$) compared with rations PAB and ESB. There was a tendency toward an interaction for the proportion of t10-C18:1 ($p = 0.076$) and t11-C18:1 ($p = 0.091$), where after feeding ration ES the highest values of t10 isomers were measured and after feeding ration ESB the highest values of t11 isomers.

Table III. Fatty acids profile (without intermediates of biohydrogenation) of milk of dairy cows, and significance of the effects of fat source (Fat), of bicarbonate and straw addition (B), and of their interaction (Fat × B).

| Fat source | Palmitic acid | | Extruded soybeans | | | | p | |
| Bicarbonate + straw | + | − | + | − | | | | |
Diets	PAB ($n=30$)	PA ($n=10$)	ESB ($n=10$)	ES ($n=10$)	SEM*	Fat	B	Fat × B
Fatty acids [g/100 g total FA]								
C4:0	2.37	2.22	2.59	2.50	0.13	0.06	0.35	0.80
C6:0	1.83	1.61	1.78	1.91	0.12	0.30	0.68	0.14
C8:0	1.18	1.03	1.16	1.33	0.12	0.25	0.95	0.20
C10:0	2.49	2.17	2.36	2.74	0.24	0.36	0.91	0.15
C11:0	0.08	0.15	0.04	0.41	0.11	0.32	0.05	0.17
C12:0	2.87	2.62	2.66	2.79	0.16	0.87	0.72	0.24
C13:0	0.14	0.14	0.12	0.13	0.01	0.23	0.43	0.55
C14:0	9.69	8.90	9.80	9.67	0.32	0.19	0.17	0.31
C14:1	1.04	1.17	0.83	0.95	0.09	0.02	0.15	0.96
C15:0	1.23	1.15	0.97	0.97	0.06	<0.01	0.55	0.52
C16:0	45.94	46.54	26.50	27.96	1.02	<0.01	0.32	0.68
C16:1	2.97	3.69	1.64	1.72	0.18	<0.01	0.03	0.08
C17:0	0.70	0.67	0.74	1.10	0.19	0.22	0.38	0.31
C18:0	4.63	3.80	10.28	9.74	0.55	<0.01	0.22	0.80
C18:1 *cis*9	14.43	14.75	20.51	19.70	0.64	<0.01	0.70	0.39
C18:2	1.60	1.79	4.23	4.89	0.27	<0.01	0.12	0.38
C18:3	0.17	0.19	0.63	0.71	0.06	<0.01	0.42	0.63
C20:4	0.14	0.16	0.17	0.17	0.02	0.15	0.72	0.48

*SEM = Standard error of the mean.

Table IV. Profile of intermediates of C18:2 biohydrogenation, t10/t11 ratio in milk of dairy cows, and significance of the effects of fat source (Fat), of bicarbonate and straw addition (B), and of their interaction (Fat × B).

| Fat source | Palmitic acid | | Extruded soybeans | | | | p | |
| Bicarbonate + straw | + | − | + | − | | | | |
Diets	PAB ($n=30$)	PA ($n=10$)	ESB ($n=10$)	ES ($n=10$)	SEM*	Fat	B	Fat × B
Fatty acids [g/100 g total FA]								
T10-C18:1	0.32	0.54	1.26	2.61	0.31	<0.01	0.02	0.08
t11-C18:1	0.66	0.64	2.61	2.12	0.14	<0.01	0.07	0.09
t-C18:1	0.98	1.18	3.87	4.73	0.32	<0.01	0.11	0.32
t10c12-CLA	0.00	0.00	0.02	0.04	0.01	<0.01	0.17	0.17
c9t11-CLA	0.61	0.69	1.53	1.25	0.09	<0.01	0.28	0.06
CLA	0.61	0.69	1.55	1.30	0.09	<0.01	0.23	0.07
t10 isomers	0.32	0.54	1.28	2.61	0.31	<0.01	0.02	0.08
t11 isomers	1.27	1.33	4.15	3.37	0.21	<0.01	0.10	0.06
t10/t11 ratio	0.24	0.39	0.36	0.82	0.11	0.02	0.01	0.16

*SEM = Standard error of the mean.

4. Discussion

4.1. Effect of fat source

A decreased milk fat content after feeding a ration rich in unsaturated fat was previously observed by Kalsheur et al. (1997). This could be attributed to an inhibition of short and medium chain FA synthesis by PUFA (Kalsheur et al. 1997), or their intermediates after

biohydrogenation (Loor et al. 2005) in the mammary gland. In goats, Schmidely et al. (2005) noticed that an increase in dietary long chain FA from extruded soybeans mainly reduced medium-chain FA in goats' milk. Furthermore, Bernard et al. (2005) showed that PUFA or their intermediates inhibited *de novo* medium-chain FA synthesis in the mammary gland of goats. In our study, extruded soybeans increased the percentages of unsaturated FA (see Table III) along with a decreased milk fat content. We did not observe a significant effect of PUFA on proportions of C6:0, C8:0, C10:0, C12:0 and C14:0 (see Table III). The lower proportions of C16:0 and C16:1 after feeding rations ES and ESB compared to rations PA and PAB are probably caused by the high alimentary level of these FA (Enjalbert et al. 1998).

When extruded soybeans were fed, the high proportion of 18C FA was in part due to the great C18:2 content of soya. The slowness of biohydrogenation induced by a great dietary concentration of C18:2, led to an accumulation of CLA and t-C18:1 in the rumen (Harfoot et al. 1973; Kim et al. 2000; Troegeler-Meynadier et al. 2003). The lower percentage of C14:1 obtained with extruded soybeans could be in part explained by a lower desaturation of C14:0. Inhibition of $\Delta 9$ desaturase by various 18C FA was noted previously in cows (Bickerstaffe & Annison 1970; Kay et al. 2004), and goats (Bernard et al. 2005; Schmidely et al. 2005). The strongest inhibitors would be, in a decreased order of inhibition capacity: C18:3, C18:2, C18:1 *cis*9 and CLA (Bickerstaffe & Annison 1970; Baumgard et al. 2000).

The t10/t11 ratio depended on fat source and was greater with extruded soybeans, mainly with ration ES, than with palmitic acid (see Table IV). While percentage of t10 isomers in milk of cows receiving extruded soybeans was 4.5 times greater than that of milk from cows receiving palmitic acid, the percentage of t11 isomers was only 2.8 times greater. The production of t10 isomers needs both, an addition of C18:2 and a low ruminal pH (Kucuk et al. 2001; Piperova et al. 2002), but probably also starch (Troegeler-Meynadier et al. 2003). Indeed the t10 isomerization is realized by the enzymes of *Megasphaera elsdenii* (Kim et al. 2002), a bacteria that develops with high-grain rations. All rations offered contained large proportions of maize silage and wheat, whose starch could enhance the population of *Megasphaera elsdenii*. So the ration ES provided favourable conditions for the production of t10 isomers. On the other hand, the production of t11 isomers could be limited: *in vitro* studies (Troegeler-Meynadier et al. 2003) have already shown that the isomerization can be saturated by high amounts of substrate, and this saturation could affect the t11 rather than the t10 pathway.

4.2. Effect of dietary bicarbonate plus straw

Bicarbonate is known to increase the rate of rumen passage (Hart & Polan 1984), which would result in an increase of C18:2 proportion in milk. The contrary was observed in our study. The lower C18:2 percentages obtained with rations ESB and PAB than with rations PA and ES could be explained by a higher ruminal pH with sodium bicarbonate plus straw, leading to a greater extent of biohydrogenation (Troegeler-Meynadier et al. 2003). The rations without bicarbonate or straw presumably favoured a low pH and consequently the formation of t10 isomers (Kucuk et al. 2001; Piperova et al. 2002), by allowing the development of the *Megasphaera elsdenii* population, to the detriment of t11 isomers. The decrease of t11 isomers could be due to an inhibition of the $\Delta 12$ isomerase by a low pH (Troegeler-Meynadier et al. 2003) induced by the lack of bicarbonate and straw in rations. The increase of t10 isomers and the decrease of t11 isomers without bicarbonate or straw led to an increased t10/t11 ratio (see Table IV). The tendency ($p = 0.11$) toward an increase of t-C18:1 percentage was mainly due to the great increase of t10-C18:1.

This greater percentage of t10 isomers could explain the lower milk fat content when cows did not receive bicarbonate or straw (see Table IV), because t10 isomers are known to

decrease milk fat content. The t10c12-CLA decreased milk fat content (Baumgard et al. 2000; De Veith et al. 2004), but t10-C18:1 may also be involved (Piperova et al. 2004; Loor et al. 2005). These two t10 isomers are not independent of each other because t10-C18:1 comes from ruminal biohydrogenation of t10c12-CLA, so that the determination of their respective effects would be difficult when FA are not post-ruminally infused.

4.3. Interaction between fat source and bicarbonate plus straw

There was a tendency toward a synergy between fat source and the addition of bicarbonate plus straw for the CLA and t-C18:1 isomers, leading to the highest percentages of CLA and t11 isomers with ration ESB, and to the highest percentage of t10 isomers with ration ES (see Table IV). The results obtained for ration ES were in part due to the addition of unsaturated fat and in part to the t10 isomers favoured by the high dietary starch content and by the low pH obtained with the ration without bicarbonate or straw, as described above.

Among fatty acids, C18:2 is most efficiently transformed to CLA and t-C18:1 (Chouinard et al. 2001), and the trends toward interactions observed in the current study support that the effect is linked to changes in the rumen environment. In our experiment, when extruded soybeans were added, sodium bicarbonate plus straw presumably led to a higher ruminal pH, favouring t11 isomers: c9t11-CLA represented 99% of milk CLA and t11-C18:1 67% of measured milk t-C18:1, t11-C18:1 being the major source of milk CLA via mammary desaturation (Griinari et al. 2000; Kay et al. 2004). Consequently, after feeding ration ESB CLA concentration reached 550 mg/l milk, which was twice more compared to rations PA and PAB and 1.3 times more compared to ration ES.

Without bicarbonate and straw addition, the ruminal pH was presumably lowered and the t10 isomers were favoured to the detriment of t11 isomers: t10-C18:1 represented 55% of milk t-C18:1 and a low amount of t10c12-CLA (3.4% of milk CLA) was obtained. But t10-C18:1 is not desaturated because there is no Δ12 desaturase in the mammary gland of dairy cows (Lawson et al. 2001), and only 21% of the duodenal t10c12-CLA (compared with 33.5% for c9t11-CLA) is secreted into milk (Chouinard et al. 1999). Consequently, the CLA concentration in milk increased only slightly compared with rations without extruded soybeans (425 mg/l for ration ES vs. 260 mg/l for ration PAB and 267 mg/l for ration PA).

5. Conclusions

A combined supplementation with extruded soybeans and sodium bicarbonate plus straw would be an interesting way to enhance the biologically interesting *trans* FA amounts (c9t11-CLA and t11-C18:1) in the milk of dairy cows, without strong depression of milk fat content: soybeans provided C18:2, and bicarbonate plus straw turned the biohydrogenation of C18:2 towards the *trans*11 isomers pathway. Extruded soybeans, bicarbonate and straw are useful for farmers since they are common feed for cattle. But further studies are necessary to optimize the quantities that must be added to rations of dairy cows in order to obtain a maximum content of c9t11-CLA and t11-C18:1 in their milk.

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