

Dietary conjugated linoleic acid isomers and selenium affect the fatty acid profile in rat liver*

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ABSTRACT

The influence of dietary conjugated linoleic acid (CLA) isomers and/or Se (as Na₂SeO₄) on the content of CLA isomers and other fatty acids (FAs) in rat liver was investigated. Feeding CLA isomers resulted in an elevated hepatic CLA isomer content. The diet with Se and *cis9trans11* CLA most effectively elevated the liver content of *cis9 trans11*, *cis,tras/trans,cis*, the sum of CLA isomers, C20:4 and C20:6. Adding Se, *cis9trans11*CLA or a CLA isomer mixture decreased the capacity of $\Delta 6$ -, $\Delta 5$ -desaturases and elongase, while *trans10cis12* had the opposite effect. Feeding Se or CLA isomers reduced $\Delta 9$ -desaturase capacity.

KEY WORDS: CLA isomers, fatty acids, rats, liver, GLC, HPLC

INTRODUCTION

Numerous investigations have demonstrated that CLA isomers alter lipid metabolism in the liver and adipose tissue. Indeed, PUFA have been shown to decrease SREBP-1 mRNA² in liver, therefore leading to reduced lipogenic gene expression. Moreover, in some studies it has been reported that dietary CLA isomers can modify the oxidation of fatty acids (FAs).

Therefore, the aim of this study was to investigate the CLA isomer profile and composition of other FAs in the liver of rats fed a mixture of CLA isomers: *cis9,trans11* (*c9,t11*), *trans10,cis12* (*t10,c12*) and/or 2 ppm Se (as Na₂SeO₄).

* Supported in part by the State Committee for Scientific Research, Grant No. 3 PO6Z 034 22

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² sterol regulatory element binding proteins (SREBP) mRNA (type of SREBP: SREBP-1a, -1b and -1c)

MATERIAL AND METHODS

Ten groups of 7-8 female rats each (Wistar, Ifz: BOA), 8 weeks of age and an initial body mass ~200 g were housed individually as described previously (Czauderna et al., 2004). Rats were fed the Labofeed diet or one enriched in 1-2% CLA isomers, 2 ppm Se (as Na₂SeO₄) (Table 1), *ad libitum*. After 28 days the rats were killed by CO₂ and their livers were removed and freeze-dried.

All reagents used and the saponification methylation methods were as previously described (Czauderna et al., 2005). CLA isomers and other FAs containing conjugated double bonds (CFA) were determined using ion (Ag⁺) liquid chromatography (Ag⁺-HPLC), while analysis of all FAs in rat liver was carried out using GLC (Czauderna et al., 2005).

The effects of CLA isomers or Se treatments were subjected to statistical analysis using the nonparametric Mann-Whitney U test, while the simultaneous Se and CLA treatment was analysed by two-factorial analysis. Statistica (ver. 6) and Excel 2000 were used.

RESULTS AND DISCUSSION

No lesions or symptoms of harmful effects of Se were found in rats fed diets with Se. The FA composition in rat liver was altered by the treatment with CLA isomers and/or Se (Table 1). Incorporation of isomers *c9t11* and *t10c12* and *t,t* was found to be selective; in particular, the percentage contribution of *t,t* isomers in liver was relatively higher compared with their share (~15%) in the dietary CLA isomer mixture. The *t,t* isomers are catabolized more slowly and are poor species for β-oxidation; they are more efficiently accumulated into liver membrane phospholipids due to their geometrical configuration. On the other hand, the percentage contribution of *c9t11* and *t10c12* was lower than in the administered CLA isomer mixture and the content ratio of *t10c12* to *c9t11* in the liver was smaller than in the CLA isomer mixture. The *c9t11* and *c11,t13* isomers were preferentially metabolized (like other *c,t/t,c* isomers) to form CFA (i.e. C18:3, C20:3, C20:4), while *t10c12*, *t10t12* and *t8c10* isomers were more efficiently driven through β-oxidation than their 9,11 homologues. Se most efficiently elevated the content of *c9t11* (285%), *c,t/t,c* (286%), the sum of CLA isomers (281%), C20:4 and C20:6 in the liver of rats simultaneously fed Se and *c9t11* CLA (positive interaction). Our recent study has also indicated that dietary Se increased the *c6* C18:1 content in the liver of rats fed Se and *c9t11* or other CLA isomers, probably due to stimulation of Δ6-desaturase capacity (a positive interaction). These results suggest that dietary Se and isomer *c9t11* most efficiently stimulated the capacity of Δ6-, Δ5-desaturases and elongase; so feeding diet 8_{+Se} increased the content of CFA, C20:4 and C22:6 in the liver. This effect may be also attributed to the ability of dietary Se

Table 1. Levels of CLA isomers (µg/g DM) and other FAs¹ (mg/g DM) in livers of rats fed diets enriched in CLA isomers^{3,4,5,6} and/or 2 ppm Se (C_{Se})

Group	Sum of CLA isomers ¹	CFA mg/g ²	<i>t,t</i> CLA ⁸ GLC, HPLC ⁷	<i>c,t/t,c</i> CLA ^{1,9} GLC, HPLC	<i>t10,c12</i> CLA GLC, HPLC	<i>c9,t11</i> CLA GLC, HPLC	9desaturase index ²	MUFA ¹⁰ mg/g	PUFA ¹⁰ mg/g	SFA ¹⁰ mg/g	C20:4 ¹⁰ mg/g	C22:6 ¹⁰ mg/g
1 ^{Control}	-	-	-	-	-	-	0.127 ^{ABCD} BEFG	6.3 ^{ABCD} abc	31 ^{Aabc}	30 ^{ABbb}	10.4 ^{Aabcd}	4.3 ^{ABCbed}
2 ^{+Se}	-	-	-	-	-	-	0.0948 ^{Aba}	3.4 ^{AE}	19 ^{aBCd}	20 ^{ACDc}	6.9 ^{Ba}	2.9 ^a
3 ³	1091	0.48 ^A	494	523	211(R=.84) ¹¹	249	0.078 ^B	3.3 ^B	23 ^b	22 ^a	7.2 ^b	4.1
4 ⁴	594 ^A	0.28 ^C	130 ^A	58 ^A	25 ^A	387 ^A	0.102	3.7 ^{CF}	16 ^{AD}	16 ^{BE}	5.6 ^{AC}	2.7 ^A
5 ⁵	1726	0.24 ^D	538	1092	804	199	0.066 ^C	5.9 ^a	42	38	12.6	8.8 ^{bb}
6 ⁶	2100	2.21 ^{AB}	829	1160	452(R=.73) ¹¹	615	0.071 ^D	3.2 ^D	30	27	9.5	5.8 ^c
3 ^{+Se}	934	0.51	373	530	230(R=.94) ¹¹	217	0.083 ^E	3.7 ^b	24	22 ^b	7.3 ^c	4.3
4 ⁴	2264 ^A	1.38 ^C	444 ^A	767 ^A	72 ^A	1491 ^{AB}	0.101	7.0 ^{EF}	48 ^{BD}	39 ^{CE}	16.7 ^{BCd}	7.5 ^{BD}
5 ^{+Se}	1360	1.53 ^D	319	1002	847	138	0.075 ^{HI}	4.8 ^C	34 ^C	33 ^D	9.1	7.5 ^C
6 ^{+Se}	2367	4.34 ^B	850	1442	599(R=.94) ¹¹	488 ^B	0.075 ^{Gh}	4.2 ^c	30 ^d	28 ^c	8.9	5.7 ^d

¹ analysis was performed using GLC as described previously (Czauderna et al., 2005); results are presented as mean values
² Δ9-desaturase index: (*cis*9C16:1 + *cis*9C18:1)/ (*cis*9C16:1 + *cis*9C18:1 + C16:0 + C18:0)
³ a diet with 1% CLA isomer mixture (the profile of CLA isomer mixture was presented previously, Czauderna et al., 2003)
⁴ a diet with 1% *cis*9*trans*11CLA (*c9,t11*)
⁵ a diet with 1% *trans*10*cis*12CLA (*t10,c12*)
⁶ a diet with 2% CLA isomer mixture
⁷ analysis performed using GLC and Ag⁺-HPLC as described previously (Czauderna et al., 2005); results are mean values obtained from GLC and Ag⁺-HPLC
⁸ *t,t* CLA: *trans* - 11/13, 10/12, 9/11 or 8/10 CLA isomers (~15% *trans,trans* CLA isomers in dietary CLA isomer mixture; Czauderna et al., 2004)
⁹ *c,t/t,c* CLA: *c,t/tc* - 11/13, 10/12, 9/11 or 8/10 CLA isomers; main CLA isomers: main CLA isomers (~80%) in the dietary CLA isomer mixture; Czauderna et al., 2003, 2004
¹⁰ MUFA - *c9*C16:1; *t11*C18:1; *c9*C18:1; *c11*C18:1; PUFA - *c9c12*C18:2; *c9c12c15*C18:3; *c5c8c11c14c17*C20:5; *c7c10c13c16c19*C22:5; *c4c7c10c13c16c19*C22:6; SFA - C16:0; C17:0; C18:0
¹¹ the ratio (R) of the concentration of *t10c12*CLA to *c9t11*CLA in liver of rats fed a diet with CLA isomer mixture; the ratio of the concentration of *t10c12*CLA to *c9t11*CLA in dietary CLA isomer mixture: 1.0242 (Czauderna et al., 2003)
¹² the content of fatty acids containing conjugated double bonds determined using reversed-phase HPLC; data previously published in Czauderna et al. (2004)

to diminish the efficiency of β -oxidation of administered CLA isomers, particularly *c9t11*CLA, as well as other non-CLA FAs in liver. These results suggest that Se, *c9t11* or the CLA isomer mixture decreased the capacity of $\Delta 6$ -, $\Delta 5$ -desaturases and elongase³ (negative interaction), while *t10 c12*CLA increased the capacity of these enzymes. Feeding Se or CLA isomers probably caused reduction of $\Delta 9$ -desaturase capacity (most effectively *t10c12*CLA regardless of Se presence), while Se in the diet with CLA isomers slightly stimulated the capacity of this enzyme (positive interaction). Consequently, the hepatic MUFA and PUFA contents of rats fed Se or CLA isomers numerically or statistically decreased, while extra Se in the diet with CLA isomers usually slightly elevated the content of MUFA in liver (positive interaction).

CONCLUSIONS

Our results indicate that CLA isomers (especially *c9t11*) decreased $\Delta 9$ -, $\Delta 6$ - and $\Delta 5$ -desaturase capacity. Therefore, finding that CLA isomers, especially *c9t11*, modify arachidonate metabolism with concurrent reduction of arachidonate-derived eicosanoid biosynthesis (cancer stimulating metabolites) and increase the level of *c9t11*⁴ and other conjugated fatty acids in the animal body confirm the particular importance of our program of developing functional food by feeding domestic animals a diet enriched in CLA isomers.

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STRESZCZENIE

Wpływ izomerów CLA i Se w diecie na zawartość kwasów tłuszczowych w wątrobie szczurów

Stwierdzono wzrost zawartości izomerów sprzężonego kwasu linolowego (CLA) w wątrobie szczurów żywionych dietą z dodatkiem izomerów CLA i/lub Se (w postaci Na_2SeO_4). Se, *cis9 trans11*CLA i mieszanina izomerów CLA zmniejsza stężenie nasyconych i nienasyconych kwasów tłuszczowych oraz efektywność działania procesu $\Delta 9$ -, $\Delta 6$ - i $\Delta 5$ -desaturacji.

³ probably *c9t11* metabolites compete with C18:3n-3, C18:2n-6 metabolites for these enzymes

⁴ having anti-carcinogenic properties