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Influence of Linoleic Acid Isomers on Body Fat

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Summary and Implications

In two studies, mice were fed diets containing either individual conjugated linoleic acid (CLA) isomers or a mixture of isomers in the presence or absence of dietary essential fatty acids. Mice fed the C18:2 Δ10,12 CLA isomer lost as much body fat as mice fed a mixture of isomers. This effect was not observed when the mice were fed the C18:2 Δ9,11 isomer or when feed intake was restricted. The loss of body fat was much greater in mice consuming an essential fatty acid deficient diet versus a control diet. This supports our hypothesis that for CLA to deplete body fat, it must first be metabolized in a manner similar to linoleic acid. Furthermore, we suggest that the loss of body fat may be mediated by metabolism of CLA to an isomer of arachidonic acid. Understanding the mechanism by which CLA causes body fat loss, in pigs as well as mice, will allow for greater regulation of body fat content.

Introduction

Conjugated linoleic acid (CLA) is a group of isomers of linoleic acid (C18:2Δ9,12), which, when consumed, produce health benefits such as reducing the incidence of cancer and cardiovascular disease and reducing body fat content. Furthermore, in swine, dietary CLA has resulted in firmer bellies, reduced backfat, and improved feed efficiency. Our group previously reported (Nebraska Swine Report 2001, pg 27 – 28) that not only did dietary CLA supplementation cause a loss of body fat in mice, but that it also resulted in programmed cell death, or apoptosis, of fat cells. The basis for the following two studies was to further determine the mechanism by which CLA is causing both the body fat loss as well as the apoptosis. The predominant naturally occurring isomer is C18:2Δ9,11 (CLA 9/11), whereas commercially synthesized CLA products usually contain approximately equal amounts of C18:2Δ10,12 (CLA 10/12) and CLA 9/11 as well as smaller quantities of other isomers. The diverse benefits of CLA may depend on different isomers. Therefore our first objective was to determine which isomer(s) are responsible for the loss of body fat in mice.

Arachidonic acid (C20:4Δ5,8,11,14) is synthesized in animals from dietary linoleic acid. Similarly, CLA 10/12 can be metabolized to C20:4Δ5,8,12,14. This product of CLA metabolism could antagonize the normal production of prostaglandins from arachidonic acid. Therefore, mice fed a diet deficient in linoleic acid, and thus arachidonic acid, may be especially sensitive to the anti-

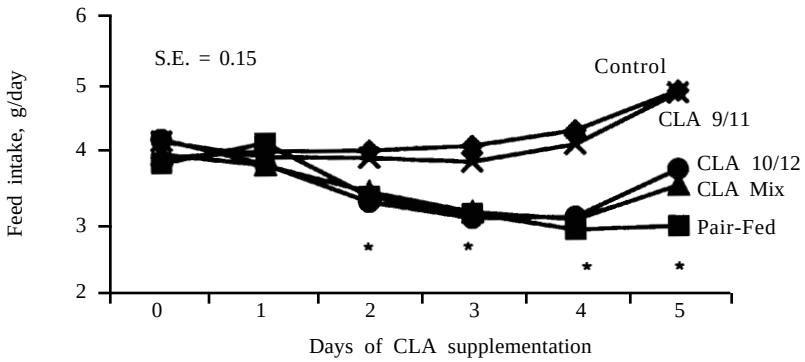

Figure 1. Effect of CLA Mix or individual isomers on feed intake (Experiment 1). *CLA 10/12, CLA Mix, and Pair-Fed differ from Control (P < 0.001).

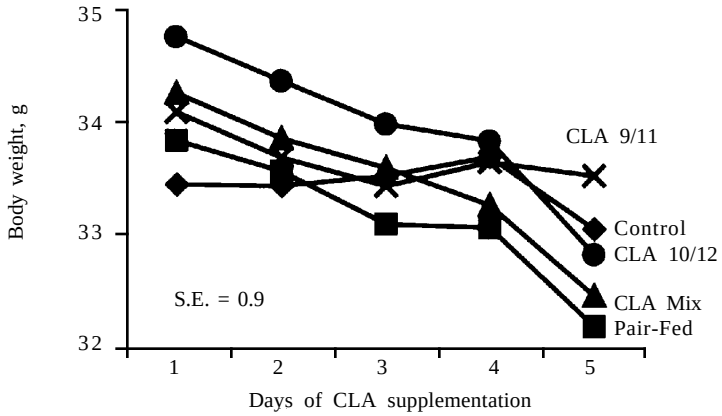

Figure 2. Effect of CLA Mix or individual isomers on body weight (Experiment 1). No effect of dietary treatment on body weight was detected.

Table 1. Effect of dietary treatment on feed intake, body weight change, and fat pad weights (Experiment 2).

	Dietary Treatments[d]				
	Control	CLA	EFAD	EFAD +CLA	SEM
Feed intake, g/d					
Before CLA[e]	3.79	3.79	3.82	3.82	0.12
After CLA[e]	4.45[a]	3.95[b]	4.50[a]	3.85[b]	0.08
Body wt. change, g[f]					
Before CLA[e]	16.66	16.66	16.94	16.94	0.47
After CLA[e]	2.75[a]	1.55[b]	2.55[a]	-0.46[c]	0.30
RP wt., g[g]	0.37[a]	0.19[b]	0.34[a]	0.09[c]	0.03
Epi wt., g[h]	0.61[a]	0.45[b]	0.55[a]	0.21[c]	0.04

[abc]Different letters in a row indicate differences, P < 0.05.
[d]Dietary treatments are as follows: Control – 7% soy oil diet for 8 weeks; CLA – 7% soy oil diet for 6 weeks, 0.5% CLA mix + 6.5% soy oil diet for 2 weeks; EFAD – 7% coconut oil diet for 8 weeks; and EFAD + CLA – 7% coconut oil diet for 6 weeks, 0.5% CLA + 6.5% coconut oil diet for 2 weeks.
[e]Feed intake and body weight change before CLA is the first six weeks of the study; after CLA is the final 2 weeks of the study.
[f]Body weight change is calculated as the weight at the final week of the feeding period minus the initial weight of that feeding period.
[g]RP = retroperitoneal fat pads.
[h]Epi = epididymal fat pads.

obesity effect of dietary CLA. Our second objective was to compare the effect of CLA in dietary essential fatty acid-adequate and -deficient diets.

Procedures

Experiment 1

Seventy-two mixed sex mice were allotted to one of five diets (each 7% fat) and allowed to consume ad libitum, except the Pair-Fed group, for 5 days:

Control	purified diet with 7% soy oil
Pair-Fed	control diet at intake of CLA Mix
CLA Mix	2% CLA mixture and 5% soy oil
CLA 9/11	0.82% CLA 9/11 and 6.18% soy oil
CLA 10/12	0.88% CLA 10/12 and 6.12% soy oil

Individual isomers were included the concentrations they were found at in the CLA Mix diet. Feed intake and body weight were measured daily. After 5 days, the mice were killed and retroperitoneal (RP) fat pads were removed and weighed. Body fat was determined on carcasses by ether extraction.

Experiment 2

Eighty, newly weaned male mice were fed either a control diet (7% soy oil) or essential fatty acid deficient (EFAD) diet (7% coconut oil) for 6 weeks. Next, half of the mice in each group were supplemented with 0.5% CLA mixture, replacing either soy or coconut oil, for 2 weeks. Then the mice were killed and RP fat pads, epididymal (Epi) fat pads, and livers were removed and weighed. Body fat was determined by ether extraction.

Results

Experiment 1

Feed intake was reduced (P < 0.001) in mice fed CLA Mix and CLA 10/12 as

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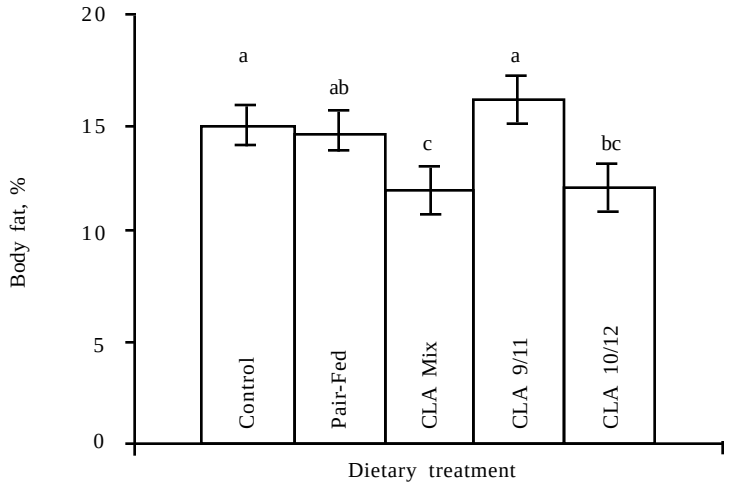

Figure 3. Effect of CLA Mix or individual isomers on body fat (Experiment 1). [abc]Bars with different superscripts differ ($P < 0.10$).

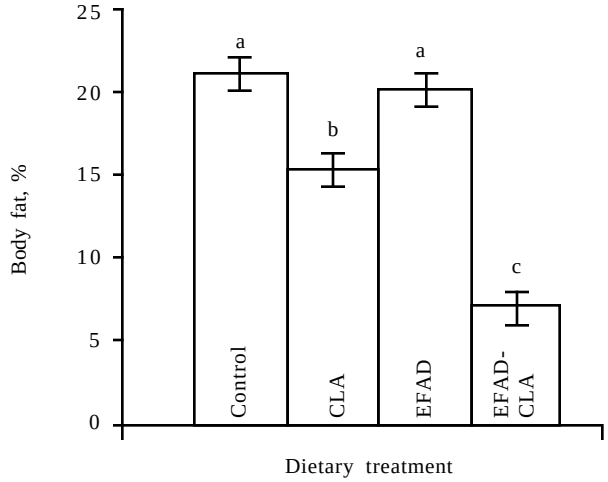

Figure 4. Effect of essential fatty acid deficiency (EFAD) and CLA supplementation on body fat (Experiment 2). [abc]Bars with different superscripts differ ($P < 0.001$).

well as the Pair-Fed mice, starting on day 2 (Figure 1). However, there were no significant differences in body weight among dietary treatment groups in this short time period (Figure 2). After 5 days of CLA supplementation, body fat content of mice fed the CLA Mix and the CLA 10/12 isomer was 20% less ($P < 0.10$) than that of mice fed the Control diet (Figure 3).

Experiment 2

Supplementation of CLA reduced ($P < 0.05$) both feed intake and body weight change in the final 2 weeks of the study (Table 1). CLA reduced ($P < 0.001$) RP (49%) and Epi (19%) fat pad weights when added to the control diet (Table 1). Furthermore, CLA reduced total body fat by 27% (Figure 4). The EFAD diet alone had no effect on feed intake, body weight, or body fat. However, when CLA was fed to mice deficient in essential fatty acids its effects were greatly amplified ($P < 0.001$); a reduction of 73% in RP, 57% in Epi, and 66% in total body fat (Figure 4).

Discussion

Our results indicate that CLA 10/12 is responsible for the loss of body fat observed when mice are fed a mixture of CLA isomers. This loss of body fat may be mediated through metabolism of CLA to an isomer of arachidonic acid. This was the basis for the design of Study 2. Arachidonic acid is a precursor to the series 2 prostaglandins, some of which appear to protect against cell death. Therefore, CLA-mediated inhibition of the conversion of arachidonic acid to prostaglandin could explain the fat cell death caused by feeding CLA. Essential fatty acids (linoleic and linolenic) protect against the full effect of CLA, which may indicate that CLA and these fatty acids are metabolized via a common metabolic path.

The knowledge that CLA 10/12 is responsible for the full fat-reducing-effect of CLA will allow both researchers, as well as swine producers to more accurately formulate diets on the active CLA isomer (CLA 10/12), instead of on the total CLA content. This becomes especially important when different sources of CLA are used as different manufacturing procedures produce different ratios of isomers. In addition, recognizing the metabolic pathway through which CLA acts should facilitate development of better methods to manipulate body fat in the future.

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