Review

Anti-obesity effects of conjugated linoleic acid, docosahexaenoic acid, and eicosapentaenoic acid

Jing-Jing Li[1], Chang J. Huang[2] and Dong Xie[1]

[1] Institutes for Nutritional Sciences, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai, P. R. China
[2] Hillblom Islet Research Center, Department of Medicine, UCLA School of Medicine, Los Angeles, USA

Obesity has become a prevailing epidemic throughout the globe. Effective therapies for obesity become attracting. Food components with beneficial effects on "weight loss" have caught increasing attentions. Conjugated linoleic acid (CLA), docosahexaenoic acid (DHA), and eicosapentaenoic acid (EPA) belong to different families of polyunsaturated fatty acids (PUFA). However, they have similar effects on alleviating obesity and/or preventing from obesity. They influence the balance between energy intake and expenditure; and reduce body weight and/or fat deposition in animal models, but show little effect in healthy human subjects. They inhibit key enzymes responsible for lipid synthesis, such as fatty acid synthase and stearoyl-CoA desaturase-1, enhance lipid oxidation and thermogenesis, and prevent free fatty acids from entering adipocytes for lipogenesis. PUFA also exert suppressive effects on several key factors involved in adipocyte differentiation and fat storage. Despite their similar effects and shared mechanisms, they display differences in the regulation of lipid metabolism. Moreover, DHA and EPA exhibit "anti-obesity" effect as well as improving insulin sensitivity, while CLA induces insulin resistance and fatty liver in most cases. A deeper and more detailed investigation into the complex network of anti-obesity regulatory pathways by different PUFA will improve our understanding of the mechanisms of body weight control and reduce the prevalence of obesity.

Keywords: Conjugated linoleic acid / Docosahexaenoic acid / Eicosapentaenoic acid / Obesity / Polyunsaturated fatty acid

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1 Introduction

Obesity has become a significant health problem in industrialized and developing countries. In the adult U.S. population, for instance, more than 60% is overweight or obese, which means their body weight exceeds the upper limit of body mass index (BMI) (http://www.cdc.gov/nchs/hus.htm).

Correspondence: Professor Dong Xie, Institute for Nutritional Sciences, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, 294 Tai Yuan Rd. Shanghai 200031, P. R. China
E-mail: dxie@sibs.ac.cn
Fax: +86-21-5492-0291

Abbreviations: BAT, brown adipose tissue; **BMI**, body mass index; **c**, cis; **CLA**, conjugated linoleic acid; **CPT**, carnitine palmitoyltransferase; **DHA**, docosahexaenoic acid; **EPA**, eicosapentaenoic acid; **FAO**, fatty acyl-CoA oxidase; **FAS**, fatty acid synthase; **FFA**, free fatty acid; **HR-LPL**, heparin releasable-lipoprotein lipase; **HSL**, hormone-sensitive lipase; **LPL**, lipoprotein lipase; **PPAR**, peroxisome proliferator activated receptor; **SCD**, stearol-CoA desatuarase; **SREBP**, sterol regulatory element-binding protein; **t**, trans; **WAT**, white adipose tissue

Overweight or obesity always increases the risk of developing many diseases, including type 2 diabetes mellitus (T2DM), hypertension, dyslipidemia, coronary heart disease, congestive heart failure, stroke, gallbladder disease, hepatic steatosis, osteoarthritis, sleep apnea, endometrial, breast, prostate, and colon cancers. Not surprisingly, the World Health Organization has defined obesity as one of the top ten global health problems.

Changes in diet and life style seem to be two major causes of obesity and are associated with increasing industrialization, urbanization, and mechanization. Increasing evidence has shown that high-fat diet, in particular, enriched in saturated fat while lacking appropriate portion of unsaturated fat, may directly facilitate the prevalence of obesity worldwide. However, it is the amount and composition of dietary fat, but not the fat *per se*, that accounts for obesity. Numerous studies have suggested that specific fatty acids can influence body adiposity, while the precise mechanisms by which the PUFA exert their anti-obesity effects are still largely unknown. Illustration of underlying

Table 1. Nomenclature, geometrical characteristic and major source of CLA, DHA and EPA

Trivial name	Systematic name		Classification	Major source
CLA	Biologically active major isomers	c9,t11- octadecadienoic acid t10, c12- octadecadienoic acid	ω-6 family	Natural source: milk and meat from ruminant animals, etc. Commercial synthesis: via alkali isomerization of linoleic acid.
	Relative minor isomers	t11,t13-, t10,t12-, t9,t11-,t8,t10-,c11,t13-,t8,c11- octadecadienoic acid ,etc.		
DHA	4,7,10,13,16,19-docosahexaenoic acid		ω-3 family	Fish oil
EPA	5,8,11,14,17-eicosapentaenoic acid			Fish oil

mechanisms will be beneficial to prevention and therapy of obesity. Thus, this review will summarize the anti-obesity effects of conjugated linoleic acid (CLA), docosahexaenoic acid (DHA), and eicosapentaenoic acid (EPA), and discuss the putative mechanisms at the cellular and molecular levels.

2 Composition and sources of CLA, DHA and EPA

CLA is the acronym for a class of positional and geometric conjugated dienoic isomers of linoleic acid. Ha *et al*. [1] coined the term when they reported the biological activity (*i.e*. anti-carcinogenic activity) associated with CLA isolated from grilled ground beef. CLA is formed when reactions shift the location of one or both of the double bonds of linoleic acid, which generates various cis- and trans-isomeric combinations. The most predominant occurring CLA isomer found in diet is cis-9, trans-11, octadecadienoic acid (c9, t11 CLA), followed by c7, t9 CLA, c11, t13 CLA, c8, t10 CLA, and t10, c12 CLA isomer. CLA is found naturally in many animal products, especially those from ruminant sources, where it is synthesized by rumen bacteria from linoleic acid [2]. CLA can also be synthesized by non-ruminants [3], and was found in non-ruminant meat sources. Nowadays, CLA can be synthesized commercially for laboratory use or dietary consumption. In certain kinds of synthetic products, c9, t11 and t10, c12 isomers constitute 80∼90% portion of the mixture. These two isomers are most likely the major effectors representing various biological functions of CLA.

Docosahexaenoic acid (DHA, 22:6 n-3) and eicosapentaenoic acid (EPA, 20:5 n-3) are two important long-chain unsaturated fatty acids in essential omega-3 family. The richest sources of DHA and EPA are high-fat (10 – 15%), cold-water fish like salmon, sardines, mackerel, herring, trout, and pilchards. DHA and EPA make up 15∼30% of the oil content of these fish. Both of them can be made by the body from the essential fatty acid alpha-linolenic acid (α-LA) of flaxseed and hemp oils, but sometimes this capacity is impaired. Thus, fish oil remains the best source (Table 1).

3 The effects of CLA, DHA and EPA on body weight and composition

3.1 Animal studies

Anti-obesity effect of these PUFA has been investigated and evaluated in animal models. In this section, we only focus on the rodent model including various species of mice and rats, which are usually employed in the studies.

As shown in Tables 2 and 3 the effects of CLA in modulating body weight have been supported by most studies [4 – 16]. Decrease in body weight seems largely due to reduced adipose deposition, and sometimes is accompanied by an increase in lean mass or protein content [5, 6, 12, 17, 18]. Although the weight-lowering effect of CLA was widely reported, some studies have demonstrated no effect, or even reverse effect of CLA on body weight [19 – 28] and it seems that rats are less sensitive to the body weight-lowing effect exerted by CLA [25 – 30]. The discrepancy is possibly attributed to the low levels of CLA (<=0.5% in diet) [6, 19, 20] and/or short-term treatment of the latter studies [20]. As compared to c9, t11 CLA, the t10, c12 isomer is considered to be the major component mediating the anti-obesity effect [6, 15, 22].

Dietary DHA and/or EPA showed little effect on body weight [31 – 35] (Table 4), however, supplementation of DHA and/or EPA decreased adipose deposition in both mice and rats [31 – 40]. The evidence supported the beneficial effects of DHA and EPA on improving obesity and indicated the differences in the mechanism underlying the anti-obesity effect between the n-3 and n-6 PUFA.

3.2 Human studies

While plenty of evidence suggests that CLA can effectively alter body composition in animals, human clinical studies conducted so far have not shown consistent results (Table 5). Most of the studies failed to show any decrease in

body weight, independent of the age, gender and physiological condition of the subjects, after supplemented with CLA raging from 0.7 to 6.8 g/day [41 – 53]. Dietary CLA has been reported to reduce body fat mass (BFM) in some studies [46 – 48, 54 – 57]. However, the body fat-lowering effect of CLA in humans was less prominent than that in mice.

Table 2. Effects of CLA on body weight, energy intake, body composition in mouse study.

Model	Dietary Fat (%)	CLA dosage (% in diet)	CLA composition	Control	Duration (d/m)[a]	Body weight	Food/ Energy intake	protein content/lean mass	Adipose deposition	Reference
ICR mice	5.5	0.5	NA[b]	corn oil	28 – 32d	NS[c]	NS	↑	↓	19
ICR mice	5.5	0.5	c9,t11 40.8-41.1%, t10,c12 43.5-44.9%	corn oil	32d	NS	↓	↑	↓	17
ICR mice	6	(1)0.5	(1)c9,t11 41.4%, t10,c12 43.5%	corn oil	28d	NS	↓	↑	↓	6
ICR mice	6	(2)0.3	(2)c9,t11 72.4%, t10,c12 13.0%	corn oil	28d	NS	NS	↑	↓	6
ICR mice	6	(3)0.25	(2)c9,t11 16.2%, t10,c12 79.2%	corn oil	28d	↓	↓	↑	↓	6
ICR mice	5.5	0.5	c9,t11 40.1%, t10,c12 41.6%	corn oil	4d	NS	NA	NA	NS	20
ICR mice	20	1	c9,t11 34.1%, t10,c12 35.9%	safflower oil	56d	↓	NS	NA	↓	7
ICR mice	13	1.41	c9,t11 32.9%, t10,c12 33.7%	linoleic acid	21d	↓	NS	NA	↓	8
C57BL/6J mice	13	1.41	c9,t11 32.9%, t10,c12 33.7%	linoleic acid	21d	↓	↓	NA	↓	8
C57BL/6J mice	11	1	c9,t11/t9,c11 34%, t10,c12 36%	safflower oil	5m	NS	NA	NA	↓	21
C57BL/6J mice	6	1	(1)c9,t11	oleic acid	28d	NS	NA	NA	NS	22
C57BL/6J mice	6	1	(2)t10,c12	oleic acid	28d	↓	NA	NA	↓	22
C57BL/6J mice	6	1	(1)c9,t11	oleic acid	28d	↓	↑	NA	NS	9
C57BL/6J mice	6	1	(2)t10,c12	oleic acid	28d	↓	↑	NA	↓	9
C57BL/6J mice	4	0.1	c9,t11 33.0%, t10,c12 34.8%	safflower oil	5m	NS	NS	NA	NS	10
C57BL/6J mice	4	1	c9,t11 33.0%, t10,c12 34.8%	safflower oil	5 m	NS	NS	NA	↓	10
C57BL/6N mice	5.5	0.5	c9,t11/t9,c11 41.9%, t10,c12 43.5%	corn oil	28d	↓	NA	↑	↓	11
C57BL/6N PPAR-anull mice	5.5	0.5	c9,t11/t9,c11 41.9%, t10c12 43.5%	corn oil	28d	↓	NA	↑	↓	11
AKR/J mice	45	1	c9,t11/t9,c11 39.1%, t10,c12 40.7%	corn oil	35d	NS	NS	NA	↓	23
AKR/J mice	5	0.25	c9,t11/t9,c11 39.1%, t10,c12 40.7%	corn oil	39d	↓	↑	NS	NS	5
AKR/J mice	5	0.5	c9,t11/t9,c11 39.1%, t10,c12 40.7%	corn oil	39d	↓	NS	NS	↓	5
AKR/J mice	5	0.75	c9,t11/t9,c11 39.1%, t10,c12 40.7%	corn oil	39d	↓	NS	NS	↓	5
AKR/J mice	5	1	c9,t11/t9,c11 39.1%, t10,c12 40.7%	corn oil	39d	↓	NS	↑	↓	5
Balb-C mice	20	1.5	c9,t11 29.6%, t10,c12 30.1%	sunflower oil	39d	↓	NS	↑(non-restricted)	↓	12
Balb-C mice	20	0.5	c9,t11 36.9%, t10,c12 38.2%	sunflower oil	21d, 84d	↓	NA	↑	↓	4
MH, ML, C57BL/6J mice	7	1,2	c9,t11/t9,c11 41%, t10,c12 44%	soy oil	12d	NS	↓	NA	↓	24
M16 mice	7	2	c9,t11/t9,c11 41%, t10,c12 44%	soy oil	14d	↑	NA	NA	↓	24
Std ddY mice	4	0.25	c9,t11/t9,c11 34.0%, t10,c12 35.1%	safflower oil	28d, 56d	↓	NS	NA	↓	13
Std ddY mice	4	0.5	c9,t11/t9,c11 34.0%, t10,c12 35.1%	safflower oil	28d, 56d	↓	NS	NA	↓	13
Std ddY mice	4	1	c9,t11/t9,c11 34.0%, t10,c12 35.1%	safflower oil	28d, 56d	↓	NS	NA	↓	13
ICR mice(non-obese control)	7	1	t10,c12 92%	linoleic acid	14d	↓	↓	NA	↓	14
M16-selected(obese)	7	1	t10,c12 92%	linoleic acid	14d	↓	↓	NA	↓	14

a) d: day, m: month
b) Not available
c) No significant difference as compare to control.

Table 3. Effects of CLA on body weight, energy intake, body composition in rat study

Model	Dietary Fat (%)	CLA dosage (% in diet)	CLA composition	Control	Duration (d/m)[a]	Body weight	Food intake	protein content	Adipose deposition	Reference
ZDF Rats	5	1.5	c9,t11 90.5%, t10,c12 0.8%	Low CLA butter	14d	NS[c]	↑	NA[b]	NS	15
ZDF Rats	5	1.5	c9,t11 47.0%, t10c12 47.9%	Low CLA butter	14d	↓	↓	NA	↓	15
Lean Zucker Rats	7	0.5	c9,t11 42.6%, t10,c12 45.6%	soybean oil	35d,56d	NS	NA	NA	↓	25
Obese Zucker Rats	7	0.5	c9,t11 42.6%, t10,c12 45.7%	soybean oil	35d,56d	NS	NA	NA	↑	25
Obese Zucker Rats	4.25	NA	c9,t11 45%, t10,c12 45%	corn oil	21d	↓	NA	NS	NS	16
Obese Zucker Rats	4.25	NA	c9,t11 53%, t10,c12 17%	corn oil	21d	NS	NA	NS	NS	16
Obese Zucker Rats	4.25	NA	c9,t11 8%, t10,c12 72%	corn oil	21d	↓	NA	NS	NS	16
OLETF Rats	6.5	1	c9,t11/t9,c11 33.2%, t10,c12 34.2%	safflower oil	28d	↓	NS	NA	↓	58
OLETF Rats	10	1	c9,t11 92.7%, t10,c12 5.8%	safflower oil	14d	NS	NS	NA	NS	26
OLETF Rats	10	1	c9,t11 3.5%, t10,c12 89%	safflower oil	14d	NS	NS	NA	↓	26
Sprague-Dawley Rats	8	0.74	c9,t11 34.1%, t10,c12 35.9%	safflower oil	28d	NS	NS	NA	↓	27
Sprague-Dawley Rats	10	3	c9,t11/t9,c11 34.6%, t10,c12 18.4%	sunflower oil	39d	↓	NA	↑	↓	18
Sprague-Dawley Rats	23	1	c9,t11 31.29%, t10,c12 36.65%	beef tallow	56d	↑	NS	NA	NA	28
Sprague-Dawley Rats	23	1	c9,t11 76.50%, t10,c12 17.23%	beef tallow	56d	↓	NS	NA	NA	28
Sprague-Dawley Rats	23	1	c9,t11 10.38%, t10,c12 89.62%	beef tallow	56d	↑	NS	NA	NA	28
Wistar rats	10	1	c9,t11 91.2%, t10,c12 3.5%	sunflower oil	42d	NS	NA	NA	NS	29
Wistar rats	10	1	c9,t11 3.0%, t10,c12 94.1%	sunflower oil	42d	NS	NA	NA	NS	29
Wistar rats	10	2	c9,t11 47.1%, t10,c12 48.85%	sunflower oil	42d	NS	NA	NA	NS	29
Wistar rats	6.5	1.5	c9,t11 39.2%, t10,c12 38.5%	soybean oil	28d	NS	NS	NA	NS	30
Ficher 344 rats	60	1	c9,t11 41.9%, t10,c12 43.5%	corn oil	18m	NS	↓	NA	NS	57

a) d: day, m: month
b) Not available
c) No significant difference as compare to control.

The dose administered, the length of the feeding period, and the differences in metabolic rates may explain the lower efficacy of CLA supplementation in humans [58].

A few reports suggested that dietary CLA could decrease body weight [54, 56, 59]. In the study conducted by Belury *et al.* [59], the subjects with type 2 diabetes mellitus had received CLA-mix supplement for 8 weeks. Plasma level of t10, c12 CLA isomer, but not c9, t11 CLA, was inversely associated with body weight. Gaullier *et al.* [56] reported that 1-year supplementation with CLA-free fatty acid (FFA) and CLA-triacylglycerol (TG) reduced BFM in healthy overweight adults, and CLA-TG could induce significant weight and BMI reductions. The 12-month extension study conducted by this group further confirmed the results of the previous study, and suggested that CLA may help maintain initial reductions in BFM and weight in long term [54].

Results from the above studies suggested that CLA might reduce body weight of diabetic subjects. For non-diabetic individuals, an inverse association of dietary CLA with body fat mass was shown in several studies, and possibly more effective on overweight and/or obese subjects [46, 48, 54]. More studies with better design are required to evaluate the effect of CLA on human body weight and composition. Careful preparation of dietary supplemented CLA with accurate composition and high purity is necessary. Other factors, such as dosage of CLA, duration of the treatment and the physiological conditions of the subjects should be prudently considered and appropriately chosen so that convincible evaluation could be made.

It was found that levels of fish-derived EPA in the plasma of both Eskimos [60] and Japanese [61] were high and the incidence of obesity-induced disease was low in their population. This suggested the beneficial effect of n-3 PUFA enriched fish oil on preventing and/or alleviating obesity. Kunesova *et al.* [62] reported that n-3 PUFA enhanced weight loss in obese females treated by very low calorie diet and DHA seemed to be the active component. In a recent study conducted by Thorsdottir *et al.* [63] in young, over-

Table 4. Effects of DHA and/or EPA on body weight, energy intake, body composition in mouse and rat study.

Model	Dietary fat (%)	PUFA dosage (% in diet)	control	Duration (d/m)[a)]	Body weight	Energy/ Food intake	Lean mass/ Protein content	Adipose deposition	Reference
C57BL/6J mice	35.2	EPA 0.46%, DHA 3.67%	rapeseed oil, sunflower oil	49d	↓	NS[c)]	NA[b)]	↓	36
C57BL/6J mice	35.2	EPA 1.16%, DHA 10.28%	rapeseed oil, sunflower oil	49d	↓	NS	NA	↓	36
C57BL/6J mice	20	EPA 0.72%, DHA 1.6%	flaxseed oil	49d	NS	NS	NA	NS	36
C57BL/6J mice	20	EPA 2.12%, DHA 4.72%	flaxseed oil	49d	↓	NS	NA	↓	36
C57BL/6J mice	20	EPA 0.6%, DHA 5.4%	corn oil	49d	↓	NS	NA	↓	36
C57BL/6J mice	20	EPA 4.26%, DHA 0.74%	corn oil	49d	↓	NS	NA	NS	36
C57BL/6J mice	35	EPA 0.32%, DHA 2.68%	rapeseed oil, sunflower oil	35d	↓	NS	NA	↓	37
KK-Ay/TaJcl mice	10	EPA 0.297%, DHA 0.54%	Rerilla oil	84d	NS	NS	NA	↓	31
KK-Ay/TaJcl mice	10	EPA 0.297%, DHA 0.54%	Soybean oil	84d	NS	NS	NA	↓	31
KK-Ay/TaJcl mice	10	EPA 0.297%, DHA 0.54%	lard	84d	NS	NS	NA	NS	31
C57BL/6NJcl mice	10	EPA 0.15% DHA3.2%	NA	4m	↓	NS	NA	↓	38
Wistar rats	18	Fish oil 2.5%, 15%	soyabean oil	56d	↑	NS	↑	NS	35
Wistar rats	20	EPA 5.48%	monosaturated FA	28d	NS	NS	NA	↓	32
Wistar rats	20	EPA 0.22%, DHA 5.38%	monosaturated FA	28d	NS	NS	NA	↓	32
Wistar rats	20	EPA 1.98%, DHA 3.04%	monosaturated FA	28d	NS	NS	NA	↓	32
Wistar rats	20	EPA 2.66%, DHA 1.9%	monosaturated FA	28d	NS	NS	NA	↓	32
Wistar rats	20	EPA 1.88%, DHA 2.9%	monosaturated FA	28d	NS	NS	NA	↓	60
Wistar rats	20	EPA 5.38%	monosaturated FA	28d	NS	NS	NA	NS	60
Wistar rats	20	EPA 0.22%, DHA 5.3%	monosaturated FA	28d	NS	NS	NA	↓	60
Wistar rats	NA	EPA 0.1%	saturated FA with the same length	35d	NS	↓	NA	↓	39
Sprague-Dawley rats	20	EPA 2%, DHA 6.52%	safflower oil	21d	NS	↓	NA	↓	40
OLETF rats	5	EPA 0.1%	safflower oil	175d	NS	NS	NA	↓	33
OLETF rats	7	EPA 0.24%, DHA 0.562%	egg-PC	28d	NS	NS	NA	↓	34

a) d: day, m: month
b) Not available
c) No significant difference as compare to control.

weight men, addition of either lean or fatty fish, or fish oil as part of an energy-restricted diet resulted in more weight loss than control diet after 4 weeks. Interestingly, this diet showed no effect on weight loss in women. DHA and EPA exhibited multiple beneficial effects on health other than anti-obesity. Unlike CLA, they are rarely used as purely anti-adiposity supplements. However, the above studies suggested that addition of seafood rich in DHA and EPA to a nutritionally balanced energy-restricted diet might boost weight loss.

4 Underlying mechanisms of anti-obesity effects of CLA, DHA, and EPA

Obesity reflects a status of excessive energy storage. The development of obesity is a complex biological process and is regulated by multiple factors. PUFA may exert their anti-obesity effects through their influence on the following aspects: (i) the balance between energy intake and energy expenditure; (ii) lipid metabolism; (iii) the status of adipocytes; and (iv) neuroendocrine system. Since their influence in the fourth aspect is complex and there have already been excellent reviews in this field, we only focus on the first three in this section.

4.1 PUFA decrease energy intake and/or increase energy expenditure

Several studies have shown that CLA supplementation reduced food/energy intake [6, 8, 14, 15, 17, 24, 64]. Loss in body weight and/or fat mass did not always demonstrate positive association with reduction in food/energy intake [5, 6, 8, 17, 24, 65, 66]. As for EPA and DHA, loss in fat mass was found accompanied by a reduction in energy intake in some studies [39, 40]. However, discrepancy between decrease in body weight/fat mass and energy intake has also been reported in a large part of studies on

Table 5. Effects of CLA on body weight and composition in humans

Subject (F/M)	Physical condition	BMI (kg/m^2)	Intervention period (d/w/m/y)[a)]	Dosage (g/day)	CLA composition	Placedo	Body weight	BFM/LBM	Reference
57(0/57)	with metabolic syndrome	27∼39	12 w	3.4	c9,t11 35.4% t10,c12 35.9%	NA[b)]	NS[c)]	NS/NS	43
57(0/57)	with metabolic syndrome	27∼39	12 w	3.4	(2)c9,t11 2.9% t10,c12 76.5%	NA	NS	NS/NS	43
21(NA)	with type2 Diabetes	NA	8 w	6	c9,t11 ∼37%, t10,c12 ∼39%	safflower oil	↓	NA/NA	59
47(NA)	healthy	25∼35	12 w	1.7	c9,t11:t10,c12=50:50	olive oil	NS	NS/NS	46
47(NA)	healthy	25∼35	12 w	3.4	c9,t11:t10,c12=50:50	olive oil	NS	↓/NS	46
47(NA)	healthy	25∼35	12 w	5.1	c9,t11:t10,c12=50:50	olive oil	NS	NS/NS	46
57(NA)	healthy	25∼35	12 w	6.8	c9,t11:t10,c12=50:50	olive oil	NS	↓/NS	46
17(17/0)	healthy	NA	64 d	3	c9,t11 17.6%, t10,c12 22.6%	safflower oil	NS	NS/NS	53
50(NA)	healthy	19.1∼34.5	12w	4.2	c9,t11:t10,c12=50:50	olive oil	NS	↓/NA	57
22(9/13)	healthy	<30	8 w	0.7 4 w + 1.4 4 w	NA	soybean oil	NS	↓/NA	47
51(33/18)	healthy	<25	8 w	3.0	(1)c9,t11 31.0% t10,c12 31.5%	Linoleic acid	NS	NA/NA	42
51(33/18)	healthy	<25	8 w	3.0	(2)c9,t11 44.9%, t10,c12 11.0%	Linoleic acid	NS	NA/NA	42
54(28/26)	healthy	25∼30	13 w	1.8	Tonalin CLA 75%TG	oleic acid	NS	NS/↑	41
54(28/26)	healthy	25∼30	13 w	3.6	Tonalin CLA 75%TG	oleic acid	NS	NS/↑	41
25(0/25)	healthy	27∼35	12 w	3	c9,t11 83.3%, t10,c12 7.3%	olive oil	NS	NS/NS	49
50(35/15)	healthy	27∼35	12 m	6	c9,t11 37.3%, t10,c12 37.6%	sunflower oil	NS	NS/NA	51
81(NA)	healthy	25∼30	18 w	1.5, 3.0	(1)>80% c9,t11	sunflower oil	NS	NS/NS	44
81(NA)	healthy	25∼30	18 w	1.5, 3.0	(2)>80% t10,c12	sunflower oil	NS	NS/NS	44
180(149/31)	healthy	25∼30	1y	CLA-FFA 3.6	c9,t11 39%, t10,c12 41%	olive oil	NS	↓/↑	56
180(149/31)	healthy	25∼30	1y	CLA-TG 3.4	c9,t11 38%, t10,c12 38%	olive oil	↓	↓/NS	56
83(47/36)	healthy	28∼35	1 y	3.4	c9,t11 39%, t10,c12 41%	olive oil	NS	NS/NS	50
83(NA)	healthy	28∼32	6 m	3.4	c9,t11 37.5%, t10,c12 38%	olive oil	NS	↓/NS	48
44(22/22)	healthy	mean 25.2 (SEM 0.21)	14 w	3.76	c9,t11 35%, t10,c12 35%	cream	NS	NS/NS	45
44(11/33)	healthy	25∼35	12 w	3	NA	CLA(-) skimmed milk.	NA	↓in BMI<=30 subjects/NS	55
48(35/13)	healthy	30∼35	12 w	3.2	c9,t11:t10,c12=50:50	safflower oil	NS	NS/NS	52
48(35/13)	healthy	30∼35	12 w	6.4	c9,t11:t10,c12=50:50	safflower oil	NS	NS/↑	52

a) d: day, w: week, m: month, y: year.
b) Not available
c) No significant difference as compare to control.

EPA and/or DHA in animal models [31, 33, 36, 37, 67]. Data from these experiments suggested that other mechanisms are involved in the PUFA-induced declines in body weight and/or adipose deposition, while reduction in energy intake may account in part for the anti-obesity effects.

On the other hand, West *et al.* [23] reported that CLA treatment reduced adipose depot weights by ~50% without any significant effect on either body weight or energy intake. However, CLA increased energy expenditure by an average of 7.7% in AKR/J mice throughout the 5-week experiment duration. Similarly, low concentration of CLA was shown enough to suppress body fat accumulation and increase energy metabolism [13]. In another study conducted on BALB-C mice, CLA in the form of either triacylglycerols (TAG) or FFA resulted in similar phenotypes: less body fat deposition, lower percentage of energy intake stored, and higher percentage of energy intake expended [68]. In another study, it was indicated that the decrease in body fat in mice fed CLA was due to increase in energy expenditure and energy loss in excreta [12]. The collective evidence suggests that CLA may increase energy expenditure, thus prevent excess energy accumulation as fat. Moreover, its impact on energy intake seems weaker compared to energy expenditure. As for DHA and EPA, they were reported to reduce fat mass at specific fat depots (*i.e.* retroperitoneal and epididymal white fat pads), and this effect

was partially mediated via inducing thermogenic activity of brown adipose tissue (BAT) [67].

The evidence presented above indicates that PUFA may enhance thermogenesis in adipose tissue, thus resulted in loss in body weight and/or fat mass. In addition, this effect might be partially attributed to the alteration in the expression of genes encoding uncoupling proteins (UCP), which play an important role in the regulation of mitochondrial ATP synthesis. The UCP protein family is composed of three members expressed differently in various tissues: UCP1 is expressed exclusively in BAT [69]. UCP2 is ubiquitously expressed in multiple tissues, while UCP3 is predominately expressed in skeletal muscle, and both of them have been identified as new potential molecular targets for the regulation of energy efficiency [70 – 72].

It was reported that fish oil reduced white fat pad mass while up-regulated the expression of mRNA for UCP1 in BAT of the rat [40]. In the later study conducted by the same group, dietary CLA was found to cause a decrease in the mRNA level of UCP1 and UCP3 in BAT of both C57BL/6J and ICR mice. In contrast, CLA greatly up-regulated the expression of UCP2 in BAT [8]. West *et al.* [23] also failed to detect a CLA-dependent increase in UCP-1 mRNA level in AKR/J mice, in spite of reduced adipose depot weights and increased energy expenditure, while UCP3 mRNA level in skeletal muscle was found increased by t10, c12 CLA-supplemented diet in the study conducted by Roche *et al.* [73].

Compared to UCP1 and UCP3, accumulating evidence suggested that UCP2 plays a more important role in the PUFA-induced alterations of energy expenditure. The expression level of UCP2 was reported to be up-regulated by PUFA in white adipose tissue (WAT) [8, 15, 21, 31, 73], BAT [23], liver [21, 73, 74] and skeletal muscle [8, 15] in different species of mice and rats. Hepatic UCP2 expression was up-regulated by EPA potentially through a prostaglandin/peroxisome proliferator activated receptor (PPAR) α-mediated pathway [74]. In addition, PPARa pathway is possibly employed in the regulation of UCP2 expression by CLA and DHA.

4.2 PUFA influence lipid metabolism

One prominent effect of CLA, DHA and EPA is their ability to alter adipose deposition, which indicates their effects on lipid metabolism. PUFA might participate in the regulation in various steps of lipid metabolism, including lipid uptake (here only refer to adipocytes), lipid catabolism (oxidation and peroxidation), lipid anabolism (here refer to biosynthesis of fatty acids) and lipolysis (intracellular and extracellular).

4.2.1 PUFA reduce lipid uptake by adipocytes via suppressing lipoprotein lipase

Lipoprotein lipase (LPL) hydrolyses triglycerides in triglyceride-rich lipoproteins (such as VLDL and chylomicrons), generating free fatty acids that can serve as a direct energy source for muscle tissue or can be stored in the forms of triglycerides in adipocytes. Park *et al.* [6] reported that t10, c12 CLA suppresses heparin releasable LPL (HR-LPL) activity in 48-h 3T3-L1 cell culture. Since the chronically inhibitory effect observed in *in vitro* study was difficult to be extrapolated to the situation *in vivo*, the acute effects were investigated by another group using major isomers of CLA: c9, t11 and t10, c12 CLA. They found that at low concentrations (<30 μmol/L) both isomers moderately increased intracellular and HR-LPL activity, but at a concentration of 100 μmol/L, they suppressed HR-LPL activity compared with BSA control level and the intracellular LPL activity returned to the initial level. The underlying mechanism is not clear, but it was suggested that the CLA isomers might reduce the LPL activity in a way different from TNF-α [75]. Another study conducted on female ICR mice showed that short-term feeding diet supplemented with t10, c12 CLA significantly reduced both HR-LPL and intracellular LPL activity in the parametrial adipose tissue [76]. In a later study conducted by Brown *et al.* [77], t10, c12 CLA, but not c9, t11 isomer significantly reduced LPL mRNA level of human stromal vascular cells containing newly differentiated adipocytes. The collective data suggest that t10, c12 CLA seems the major isomer responsible for down-regulation of LPL activity.

In a study conducted by Raclot *et al.* [32], the mRNA level of LPL was decreased in the retroperitoneal tissue of rats fed with high-fat diets supplemented with EPA (decreased but not significantly), DHA and EPA-DHA mixture. It is possible that CLA, DHA and EPA exert their fat mass-loss effect by suppressing LPL activity, thus preventing free fatty acid from entering adipocytes.

4.2.2 PUFA increase lipid catabolism via enhancing fatty acid oxidation and/or peroxidation

Catabolism of lipids can be achieved by β-oxidation in mitochondria or peroxidation in peroxisomes. Several studies have confirmed the ability of CLA to increase fatty acid oxidation [78 – 80]. Carnitine palmitoyltransferase-1 (CPT-1) acts as a rate-limiting enzyme in mitochondrial oxidation system, while fatty acyl-CoA oxidase (FAO) functions as the rate-limiting enzyme in peroxisomal oxidation system. One group showed that a 4-week supplementation of both forms of CLA in the form of either FFA or triacylglycerol (TG) resulted in enhanced CPT activity in BAT, perirenal adipose tissue, red astrocnemius muscle and the liver [65]. In agreement with this study, Martin *et al.* [81] reported that the activity of CPT-1 in the liver and adipose tissue was increased in rats after consuming a diet containing 1% of t10, c12 CLA for 6 weeks.

EPA has been shown to promote mitochondrial fatty acid oxidation in both hepatocyte [82] and adipocyte [83], which is closely associated with increased CPT-1 mRNA level

[82] and/or activity [83]. In a study conducted by Willumsen, administration of DHA on rats tended to increase the peroxisomal beta-oxidation in the liver, which was accompanied by a significant increase of FAO activity, but not CPT-1 activity. However, EPA showed hypotriglyceridemic effect and resulted in an increase of mitochondrial and peroxisomal oxidation of fatty acids in this study, and CPT-1 activity was stimulated by EPA treatment [84]. In another study, Totland *et al.* [85] reported that DHA treatment on male Wistar rats significantly increased CPT-1 mRNA expression in the muscle, while feeding with EPA increased CPT-1 mRNA level in the liver, and both of them significantly elevated FAO expression in the liver. Increase of fatty acid oxidation by EPA has also been suggested by another study conducted by Willumsen *et al.* [86]. Conclusively, it is strongly suggested that CLA, DHA and EPA participate in the regulation of fatty acid oxidation and/or peroxidation, which contributes to their influence on lipid metabolism and further lipid-lowering effects.

4.2.3 PUFA decrease lipid synthesis through inhibiting stearol-CoA desatuarase and/or fatty acid synthase

Fatty acid synthase (FAS) and stearol-CoA desatuarase (SCD) are most essential enzymes that are involved in fatty acid synthesis. FAS is a multifunctional enzyme complex. SCD induces a cis-double bond in the delta-9 position of certain saturated fatty acids and behaves as the rate-limiting enzyme in the biosynthesis of monounsaturated fatty acids. Four mouse SCD and two human SCD genes have been cloned and characterized [87 – 91]. Whereas the liver expresses SCD1 exclusively, adipose tissue expresses both SCD1 and SCD2 in mice [87, 88]. A recent study showed that among the total 14 fatty acids examined, c9, t11 CLA exhibited the greatest inhibitory effect on liver FAS activity, followed by t10, c12 CLA. DHA was also shown to inhibit FAS activity but to a lesser extent than CLA [92]. In the study conducted by Tsuboyama-Kasaoka *et al.* [21], the mRNA level of FAS showed marked decrease in female C57BL/6J mice after 5 months of supplementation with 1% CLA. Kim *et al.* [93] reported that high fish oil feeding resulted in a decreased liver FAS and SCD-1 expression. The suppression of hepatic and/or adipose tissue FAS expression by DHA and/or EPA was also observed in other studies [94 – 96].

However, there is some contrary evidence about the regulation of FAS by CLA. Lionel Clément *et al.* [97] reported that dietary t10, c12 CLA increased liver FAS mRNA level and this was responsible for t10, c12 CLA-induced fatty liver. The treatment with t10, c12 CLA demonstrated an increased liver lipid level accompanying body fat loss [21, 22]. The discrepancy between the results of different studies might be partially due to the composition of CLA (mixture or highly-purified isomers), the strains of animals and the duration of feeding (short-term or long-term).

A study conducted by Kang *et al.* [98] declared that anti-obesity effect of CLA is independent of SCD1 expression and enzyme activity. In this study, it has been found that t10, c12 CLA showed similar effects in reducing body fat, blood triglyceride, and free fatty acid level on both wide-type and SCD1-null mice. Since both of SCD1 and SCD2 are expressed in adipose tissue, whereas SCD1 is, also expressed in the liver, there may be some functional compensation that masks the effects of CLA via SCD1. However, a number of studies indicate similar impacts on SCD1 by CLA, DHA, and/or EPA. Lee *et al*. [99] reported that CLA-supplemented diet suppressed SCD1 mRNA expression in the liver, and the same inhibitory effect was observed in H2.35 liver cells. A subsequent study showed that treatment with t10, c12 CLA on 3T3-L1 cells resulted in smaller lipid droplets, with reduced levels of the major monounsaturated fatty acids: palmitoleic acid and oleic acid. This phenomenon could be ascribed to a dose-dependent decrease in expression of SCD1 gene [100]. EPA has also been shown to decrease hepatic SCD1 mRNA level [101]. In the study conducted by Engler *et al.* [102], a DHA-enriched diet for 6 weeks was found to decrease delta-9 desaturase activity in hepatic microsomes of spontaneously hypertensive rats (SHR), accompanied by an increase in 16:0 and a reduction in 16:1 n-7 content in hepatic microsomes. Because monounsaturated fatty acids constitute a large portion of cellular lipids, decreasing their biosynthesis via inhibition of SCD may account for the PUFA fat-lowering effect.

Expression of many lipogenic enzymes including FAS and SCD is regulated by the transcription factor, the sterol regulatory element-binding protein (SREBP-1) [103]. Nakatani *et al* [104]. reported that fish oil feeding decreased body weight and fat mass in a dose-dependent manner, in parallel with a decrease in both SREBP-1 mRNA level and mature SREBP-1 protein in the liver, as well as expression of some lipogenic genes including SCD-1. This evidence indicated that DHA and EPA might exert their inhibitory effects on FAS and SCD via their suppression of SREBP-1. This conclusion is also supported by the study of Yahagi *et al.* [105].

The effect of CLA on SREBP-1 seems complicating and conflicting. The c9, t11 CLA diet (c9, t11 CLA: t10, c12 CLA = 4.43:0.84) reduced hepatic SREBP-1c mRNA expression in ob/ob mice, coupled with reduced levels of both membrane-bound precursor and nuclear mature forms of SREBP1-c proteins. In contrast SREBP1-c mRNA expression in WAT was increased by c9, t11 CLA diet. The t10, c12 CLA diet (c9, 11 CLA: t10, 12 CLA = 1.03:4.59) showed a stronger inhibitory effect on body weight gain than c9, t11 CLA diet. However, it did not show any effect on SREBP 1-c expression either in the liver or WAT [73]. Another study using CLA mixture demonstrated that compared to linoleic acid, CLA greatly decreased WAT mass but caused hepatomegaly accompanied by an increased

hepatic mRNA level of SREBP1-c in both C57BL/6J and ICR mice [106]. It seems that the regulation of SREBP-1 by CLA may be isomer dependent (c9, t11 *vs*. t10, c12) and organ/tissue specific (liver *vs*. WAT).

4.2.4 PUFA might increase intracellular lipolysis

Lipolysis includes the release of both extracellular and intracellular fatty acids. Fatty acids from triglycerides coming from fat absorption in the form of chylomicrons are released via extracellular lipolipase, mainly by LPL, as mentioned in the previous section. Hormone-sensitive lipase (HSL) is a cytosolic neutral lipase that hydrolyzes intracellular triglycerides stored within adipocytes, and produces free fatty acids used as energy substrates by a wide range of tissues [107]. HSL is thought to be the rate-limiting enzyme in intracellular lipolysis, thus any changes on HSL expression will modulate lipid mobilization and lipid content in the adipose tissue. A recent study reported that 14-week CLA supplementation (in the form of triacyglycerols 3.76 g with a 50:50 combination of c9, t11 and t10, c12 isomers) added to flavored yoghurt-like products was able to alter body composition in healthy subjects, associated with a significant reduction in mRNA levels of HSL [45]. The study conducted by Raclot *et al*. [32] showed that DHA-diet and DHA, EPA-mixed diet decreased HSL expression in retroperinatal (RP) adipose tissue, which seemed inconsistent with the limitation of local fat cell hypertrophy. However, in this study, the expression of genes encoding for the enzymes involved in lipogenesis, such as FAS, LPL, phosphoenolpyruvate carboxykinase (PEPCK) also decreased. The author commented that the decrease in mRNA level of these genes is closely related to the decrease of fat cell size, and that the impact of DHA and EPA on lipogenesis exceeded that on lipolysis , thus leading to such outcome [32].

4.3 PUFA influence the status of adipocytes

The word "status" refers to two major aspects in the "life of an adipocyte" – preadipocyte differentiation and cell death. In this section, we will summarize studies investigating the effects of CLA, DHA, and EPA on these events.

4.3.1 PUFA alter preadipocyte differentiation

The 3T3-L1 preadipocytes are one of the well-characterized model systems available to study cellular differentiation of adipocytes *in vitro*, thus provide an ideal tool for evaluating the effects exerted by PUFA on adipocyte differentiation. In a study conducted by Kang *et al.*, it was reported that treating differentiating 3T3-L1 preadipocytes with t10, c12 CLA resulted in a decrease of intracellular triglyceride (TG) accumulation as well as a decline of mRNA levels of FAS and aP2. The protein levels of two transcription factors essential for adipogenesis, PPARγ and CCAAT/enhancer binding protein (C/EBPα), were reduced as well. The significantly inhibitory effect on PPARγ and its downstream target-LPL by dietary t10, c12 CLA supplementation was also observed in mouse adipose tissue [108]. An earlier study using CLA mixture showed that continuous treatment with CLA on both pre- and post-confluent 3T3-L1 preadipocytes hindered adipocyte differentiation, as reflected by inhibition of several differentiation-associated markers. This study suggested that fat reduction caused by CLA treatment might be attributed to its inhibition on both proliferation and differentiation of preadipocytes in animals [109]. In another study, the effect of CLA on preadipocyte metabolism was evaluated. It was shown that t10, c12 CLA stimulated oleic acid oxidation, although it also increased glucose and oleic acid incorporation into lipid. Further, it has been concluded that the TG-lowering effects of t10, c12 CLA in cultured 3T3-L1 preadipocytes might be attributed to increase in fatty acid oxidation, which exceeded its stimulatory effects on glucose and oleic acid incorporation into lipid. This study shed some light on the influence of CLA relative to cellular metabolism during preadipocyte differentiation, besides its impact on adipogenic gene expression and cell growth [80]. Such inhibitory effects are probably mediated by the influence of CLA on lipogenic enzymes, adipogenic transcriptional factors and other differentiation-associated factors. The inhibition of adipocyte differentiation by CLA, especially the t10, c12 isomer, has been also reported by other groups [110, 111]. In addition, it was suggested that impaired lipid accumulation by t10, c12 CLA during adipocyte differentiation was dependent on timing and length of the treatment [110]. These results indicate that CLA may decrease body fat partially via inhibiting preadipocyte differentiation.

DHA was reported to decrease the mRNA of adipocyte determination and differentiation-dependent factor-1 (ADD1/SREBP1) in porcine differentiating adipocytes [112]. The study conducted by Kim *et al.* [113] also demonstrated that DHA inhibited 3T3-L1 differentiation to adipocytes. The collective data strongly suggest that the PUFA may exert their anti-obesity effects by inhibiting preadipocyte differentiation.

Besides preadipocytes, there is a large amount of mature adipocytes in adipose tissue. It is an interesting possibility that the three PUFAs may induce adipocyte dedifferentiation, reduce the proportion of fat-storing adipocytes, thus lead to fat-lowering effects.

4.3.2 PUFA induce apoptosis of preadipocytes and/or adipocytes

CLA treatment was shown to induce apoptosis of 3T3-L1 preadipocytes [114]. The apoptotic effect of CLA was also observed in adipose tissue [115] and fat cells [111], and the apoptosis was induced mainly by t10, c12 isomer. In the study by Perez-Matute *et al.* [39], EPA administration reduced retroperitoneal adipose tissue, which could be secondary to an increase in apoptosis. In addition, DHA was reported to induce apoptosis in postconfluence preadipo-

cytes [113]. Thus, induction of adipocyte apoptosis is likely involved in their anti-obesity effect as well.

4.4 Effects of PUFA on the neuroendocrine system

Obesity is a multifactorial and complex affectation that is characterized by a long-term excess energy intake (EI) above energy expenditure (EE). Since both, energy intake and expenditure, are under control of the neuroendocrine system, influence from dietary fatty acids on the nervous system and the endocrine system has been extensive investigated. Increasing evidence suggests that PUFA can exert anti-obesity effect via the regulation of these systems, including their impact on sympathetic nervous activity [116], hypothalamic gene expression [117] and level of hormones that are essential for body fat control by the nervous system, such as leptin [32, 65, 118 – 121] and adiponectin [37, 118, 120, 121].

5 Side effects of CLA, EPA and DHA supplementation

Although CLA showed beneficial effects in decreasing body fat mass, several adverse effects have been reported and attracted a great deal of attention. The harmful side effects of CLA include induction of hyperinsulinemia, insulin resistance (IR) and massive liver steatosis. In fact, CLA was first reported to normalize impaired glucose tolerance and attenuate fasting hyperinsulinemia and free fatty acid concentration in male Zucker diabetic fatty fa/fa (ZDF/GMI) rats [122]. In addition, some subsequent studies supported its anti-diabetic effects through improving insulin sensitivity [15, 123, 124]. However, many studies on other animals besides ZDF rats exhibited controversial results. CLA treatment was demonstrated to induce insulin resistance on either AKR/J male mice or C57BL/6J female mice [5, 21]. Roche *et al.* [73] reported divergent metabolic effects of CLA isomers in ob/ob mice. This study implicated the t10, c12 CLA isomer in the development of insulin resistance in a mouse model of obesity and hypoleptinemia. Thrush *et al.* [125] reported that mixed-isomer CLA supplementation in overweight non-diabetic humans decreased insulin sensitivity. The t10, c12 CLA was shown to promote insulin resistance through NF-κB-dependent cytokine production in mature human adipocyte [126]. Collective studies supported that t10, c12 CLA is the major isomer inducing insulin resistance in human [43, 127, 128]. ZDF rats are leptin receptor-defective, thus leptin resistant, and this might be responsible for the opposing effects exerted by CLA on this animal model compared to that on other murine models and humans.

In contrast to t10, c12 CLA, both DHA and EPA are beneficial for improving insulin resistance. Oral administration of DHA ethyl ester exerted its hypoglycemic effect on KK-Ay mice with genetic non-insulin-dependent diabetes mellitus (NIDDM), and it was suggested that DHA exhibited this effect by increasing insulin sensitivity [129]. The study of Mori *et al.* [130] indicated that EPA prevented development of insulin resistance in Dahl-S rats, which were fed a high-sucrose diet (HSD). Moreover, EPA was reported to improve or prevent insulin resistance in WBN/Kob rats [131] and Otsuka Long-Evans Tokushima Fatty (OLETF) rats [33, 132].

Dietary CLA supplementation was reported to cause considerable liver steatosis, though a dramatic decrease in body fat mass was observed [97]. The adverse effects to induce fat liver and/hepatic steatosis have been also observed by other groups, and increasing studies indicate that t10, c12 isomer is the major player [9, 22, 97, 133]. However, dietary CLA was reported to alleviate nonalcoholic fatty liver disease or hepatic steatosis in ZDF rats [134, 135]. The discrepancy illustrated a relationship between the state of insulin resistance and liver-lipid metabolism, and suggested that CLA might act to favorably modify lipid metabolism in ZDF rats. In contrast to the adverse effect of CLA on the liver, fish oil-feeding was shown to prevent perfluorooctanoic acid – induced fatty liver in mice [136]. Moreover, DHA was reported to attenuate CLA-induced fatty liver when added to CLA diet in C57BL/6N mice [137].

6 Conclusions

CLA, DHA and EPA show beneficial effects against obesity through various molecular mechanisms. Despite a large amount of investigations, the molecular mechanisms responsible for their anti-obesity effects remain largely unknown. In recent years, increasing concerns have been shifted from anti-adiposity phenomena to digging out the underlying mechanisms. CLA, DHA and EPA have been shown to regulate some nuclear receptors involved in control of body weight and adiposity, either by affecting the expression level of the receptors, or by serving as a ligand, thus modulating their transcriptional activity. Nuclear receptors PPAR [138 – 140], liver X receptor (LXR) [110, 141], retinoid X receptor (RXR) [142, 143] are all reported as potential candidates.

A study conducted by Brown *et al.* [77] indicated that MEK/ERK signaling pathway might be activated in fat-loss action of CLA. It has been reported that DHA participated in the modulation of ERK1/ERK2 phosphorylation stimulated by PMA or transforming growth factor (TGF)-α in NIH/3T3 cells [144]. The three PUFA have been shown to regulate MAPK signaling in several other types of cells besides adipocytes [145 – 148]. It is possible that MAPK pathway gets involved in the anti-obesity effect of the PUFA.

Although CLA, DHA and EPA reveal similar effects on prevention and/or improvement of obesity, and share sev-

eral mechanisms, there are still distinct in several aspects, such as the degree of the fat-lowering effect, side effects of administration, etc. The difference in double-bond position, geometrical structure, and metabolic pathway may partially account for these distinctions. However, a full-scale interpretation remains unavailable. Well-designed experiments are required to interrogate into the roles of these PUFA in specific event. Moreover, such studies are important in ensuring the development of safe and efficacious foods fortified by the PUFA, or PUFA-based nutraceuticals, for the prevention or treatment of obesity.

The authors have declared no conflict of interest.

7 References

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