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Dietary fish oil n-3 polyunsaturated fatty acids and alpha-linolenic acid differently affect brain accretion of docosahexaenoic acid and expression of desaturases and sterol regulatory element-binding protein 1 in mice☆

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Abstract

Whether preformed dietary docosahexaenoic acid (DHA) is required for brain accretion has not been clearly determined. In this study, we investigated in mice the different effects of dietary longer-chain n-3 polyunsaturated fatty acids (PUFAs) and α-linolenic acid (LNA) on brain accretion of DHA and the expression of associated desaturases and transcription factors. C57 BL/6J mice were fed for 3 months with four fish oil n-3 PUFA diets — lower, low, high and higher (0.46%, 0.91%, 1.73% and 4.29% total energy, respectively); a flaxseed oil n-3 PUFA (5.01% total energy) diet; and an n-3 PUFA-deficient diet, respectively. Either fish oil or flaxseed oil n-3 PUFA diets increased DHA concentrations in the brain. However, the flaxseed oil n-3 PUFA diet was less effective than the fish oil diet with higher amount of n-3 PUFA in increasing brain DHA content. Furthermore, the expressions of delta-6 desaturase (D6D) and sterol regulatory element binding protein 1 (SREBP-1) in the liver were down-regulated by all fish oil diets with different amounts of n-3 PUFAs, as well as by the flaxseed oil n-3 PUFA diet, whereas in the brain, D6D, delta-5 desaturase (D5D) and SREBP-1 expressions were down-regulated by the higher fish oil n-3 PUFA diet rather than by other fish oil n-3 PUFA and the flaxseed oil n-3 PUFA diets. These results suggest that preformed dietary DHA, different from those converted by LNA inside the body, is better for brain accretion. Dietary longer-chain n-3 PUFAs affect expressions of D6D, D5D and SREBP-1 in the brain differently from their precursor LNA.
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Key words: n-3 polyunsaturated fatty acids; Desaturase; Sterol regulatory element-binding protein; Brain; Mice

1. Introduction

n-3 polyunsaturated fatty acids (n-3 PUFAs), especially docosahexaenoic acid (DHA) (22:6n-3), are important for normal brain development and function. DHA accumulates in high concentrations in the brain and the retina. Changes in brain DHA are positively associated with changes in cognitive and behavioral performance [1,2]. Animal and human studies have shown that n-3 PUFA deficiency reduces DHA concentration in brain, alters neurogenesis [3] and is associated with poorer development of visual acuity and lower indices of neural development [1,2], as well as with a variety of central nervous system disorders, including Alzheimer's disease and psychiatric illnesses, such as schizophrenia and depression and mood disorders [4].

In mammals, n-3 PUFAs must be obtained from diet because they cannot be synthesized *de novo*. However, longer-chain n-3 PUFAs such as DHA can be converted from the precursor α-linolenic acid (LNA, 18:3n-3), via delta-6 desaturase (D6D), delta-5 desaturase (D5D) and elongases, although the conversion is limited [5]. Therefore, mammals obtain DHA either as DHA itself or the precursor LNA, and intermediates between LNA and DHA, including eicosapentaenoic acid (EPA, 20:5n-3) [5–8]. In the brain, the conversion coefficients of LNA to DHA are very low with only 1% of plasma LNA entering the brain converted to DHA. Therefore, circulating plasma DHA derived from diet or biosynthesized from LNA in the liver is the source for brain accretion of DHA [1,5,8–10]. An important question has been whether preformed dietary DHA is required or needs to be met with its precursor LNA. Data from some studies show that LNA appears to be a limited source of longer-chain n-3 PUFAs, and adequate intakes of preformed DHA may be important for maintaining optimal function of organs and tissues including the brain [5,9]. Still, other studies report that adequate intake of only LNA is sufficient to maintain normal brain DHA concentration [11,12].

Abbreviations: D5D, delta-5 desaturase; D6D, delta-6 desaturase; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; LNA, α-linolenic acid; PPAR, peroxisome proliferator-activated receptor; PUFAs, polyunsaturated fatty acids; SREBP-1, sterol regulatory element-binding protein 1.

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An appropriate quantity and ratio of n-3 to n-6 PUFAs in diet are an important determinant of human health. In modern Western diets, the ratio of n-6/n-3 PUFAs increases to 15:1 to 25:1, which may have contributed to the prevalence of many chronic diseases including depression and Alzheimer's disease [4,13]. However, evidence from studies on the evolutionary aspects of diet, modern day hunter–gatherers and traditional diets indicates that human beings evolved on a PUFA-rich diet in which the ratio of n-6/n-3 PUFAs was about 1–2:1 [13], and this n-3 PUFA-enriched diet may probably promote human brain evolution [14]. Therefore, the brain may need more n-3 PUFAs than what we recommend in diets.

D6D and D5D, expressed in many tissues including the liver, brain, heart and placenta, are important enzymes in longer-chain n-3 and n-6 PUFA synthesis [15,16]. Results from studies on peripheral tissues show that D6D and D5D are under strong control by dietary fats and other factors such as insulin and growth hormone [17]. Furthermore, at the transcription level, D6D and D5D are regulated by two transcription factors, sterol regulatory element-binding protein 1 (SREBP-1) and peroxisome proliferator-activated receptor-α (PPARα) [17,18]. Liver SREBP-1 expression is reduced in rodents by dietary intake of PUFAs including EPA and DHA in fish oil [19,20], whereas PUFAs and their metabolites are ligands of PPARs [21]. Most of the studies on SREBP-1 and PPARs have been primarily conducted in peripheral organs and tissues, and less is known about the roles of these transcription factors for brain lipid metabolism. Therefore, we hypothesized that the different effects of DHA and LNA on DHA accretion may be associated with changes in the desaturases and transcription factors in the brain.

In the present study, we utilized two types of diets with n-3 PUFAs from fish oil and flaxseed oil, respectively, as well as fish oil diets with different quantities of n-3 PUFAs, to investigate the differences in brain accretion of DHA and the expression of D6D, D5D, as well as SREBP-1 and PPARβ, the prevalent PPAR isoform in rodent brain.

2. Materials and methods

2.1. Diets

Two dietary regimes — fish oil n-3 PUFAs (rich in DHA) and flaxseed oil n-3 PUFAs (rich in LNA) — and an n-3 PUFA-deficient diet were used. Fish oil n-3 PUFA diets were divided into four subtypes — lower, low, high and higher, according to n-3 PUFA quantity contained. Each of the diets was designed with 6% of fat (wt/wt) (14% total energy) and with the same baseline containing 200 g of casein, 388 g of corn starch, 100 g of maltodextrin, 150 g of sucrose, 35 g of mineral mix, 10 g of vitamin mix, 47 g of cellulose, 40 g of calcium phosphate and 2.5 g of choline per kilogram. The fat content and the fatty acid compositions of the diets are shown in Table 1. The four fish oil n-3 PUFA diets — lower, low, high and higher — provided trace LNA and 3.68%, 6.33%, 10.41% and 28.37% (wt/wt), respectively, of longer-chain n-3 PUFAs (0.46%, 0.91%, 1.73% and 4.29% total energy, respectively), whereas the flaxseed oil diet provided a higher amount of LNA (35.78%, wt/wt) (5.01% total energy) with no longer-chain n-3 PUFAs. The higher fish oil and the flaxseed oil n-3 PUFA diets had an n-6/n-3 PUFA ratio of 1.09:1 and 0.90:1, respectively, close to 1:1. In other fish oil n-3 PUFA diets (lower, low and high), the n-6/n-3 PUFAs ratio was 16.77:1, 9.69:1 and 4.51:1, respectively. The diets were prepared by the Laboratory Animal Center of the Academy of Military Medical Sciences of China.

Table 1
Fat content and fatty acid composition of the experimental diets

	n-3 PUFA-deficient diet	Fish oil n-3 PUFA diets				Flaxseed oil n-3 PUFA diet
		Lower	Low	High	Higher	
Fat (g/kg diet)						
Sunflower oil	60	54	49	42	18	22
Fish oil	–	6	11	18	42	–
Flaxseed oil	–	–	–	–	–	38
Fatty acid (% of total fatty acids)						
Saturated	11.30	11.01	9.86	9.64	7.42	10.12
Monounsaturated	24.07	23.71	25.24	25.99	28.55	22.10
C18:2n-6	64.63	60.04	55.61	49.24	28.39	32.22
C18:3n-3	–	0.90	1.64	2.68	2.25	35.78
C20:4n-6	–	0.23	0.42	0.68	1.59	–
C20:5n-3	–	2.13	3.52	6.39	15.89	–
C22:4n-6	–	0.43	0.90	1.36	3.42	–
C22:5n-3	–	0.18	0.50	0.61	1.89	–
C22:6n-3	–	1.37	2.31	3.41	10.59	–

2.2. Animals

C57 BL/6J mice (3–4 weeks old, female) were purchased from the Laboratory Animal Center of the Academy of Military Medical Sciences of China and then housed at the animal facilities in the center in a 12-h light/12-h dark cycle and cycles of air exhaustion. After 1-week recovery from transportation, the mice were fed one of the six types of diets for 3 months. At the end of experiments, food was withdrawn during the daylight hours before onset of the dark cycle. Anesthesia was administered by intraperitoneal injection of Avertin (2,2,2-tribromoethanol) (T-4840-2, Aldrich Chemical) (125 mg/kg) to each mouse to obtain blood samples for analysis. Mice were sacrificed immediately by injection of an overdose of Avertin (500 mg/kg), and brains and livers were collected. Tissues were weighed and frozen in liquid N_2 and then wrapped in aluminum foil. Once an entire group of animals was harvested, the tissues were transferred to a −80°C freezer until analysis. All animal experiments were conducted from 0800 to 1200 in compliance with the guidelines of the Animal Care and Use Committee of Academy of Military Medical Sciences of China.

2.3. Fatty acid analysis

Fatty acids in tissues and plasma were measured by gas chromatography (GC). Tissues were first hand homogenized in sodium chloride solution (145 mM) (1:5, wt/vol). Fatty acid methyl esters (FAMEs) from plasma and tissue homogenate were prepared according to a modified method of Lepage and Roy [22,23]. Briefly, 2 ml of methanol-*n*-hexane (4:1, vol/vol) solution containing C15:0 (an internal standard) and 2 μl of BHT (20 mM) were added to 100 μl of plasma or tissue homogenate to prevent lipid oxidation, then 0.2 ml of acetyl chloride was slowly added. After being heated at 100°C for 1 h, 5 ml of 6% K_2CO_3 solution was added to the tube, mixed on a vortex and centrifuged, and the clear *n*-hexane top layer containing FAMEs was transferred to a GC autosampler vial for analysis.

FAMEs were analyzed using an Agilent 6890N GC equipped with a flame ionization detector and injector. Separation of FAMEs was conducted on an HP capillary column (30 m×0.32 mm id×0.25 μm DB, P/N 19091J-433) with cross-linked 5% phenyl methyl siloxane as stationary phase. The split–splitless injector was used in split mode with a split ratio of 1:5. The injection volume of the sample was 1 μl. The injector and detector temperatures were kept at 240°C and 270°C, respectively. The oven temperature was programmed as follows: initial temperature was 80°C for 2 min, then increased by 15°C/min to 220°C for 10 min, again increased by 3°C/min to 240°C and then left to stand for 45 min at 240°C. Nitrogen was used as the carrier gas, with a linear velocity of 15 cm/s. Peaks were identified by comparison with standard mixtures of fatty acid methyl esters (18919-1AMP) (Supelco, Bellefonte, PA, USA) and by comparison with reference group chromatograms. Plasma and tissue fatty acid data were expressed as percent of total fatty acids.

2.4. Quantitative RT-PCR-based gene expression

Total RNA was extracted from mouse livers and brains with TRIzol Reagent (15596-026) (Invitrogen, USA), and cDNA was prepared from total RNA using the SuperScript III First-Strand Synthesis System for RT-PCR (18080-051) kit (Invitrogen, USA) as described [24]. mRNA levels of D6D and D5D were measured with real-time quantitative RT-PCR using an ABI PRISM 7300 sequence detection system (Applied Biosystems, Foster City, CA, USA). Previously published oligonucleotide primers in GenBank were used: D6D (NM_019699) (forward: TGCCTGGTTCATCCTCTCGTA; reverse: GGCTGTGAC-GAGGGTAGGAA), D5D (NM_146094) (forward: CCGCGGCACAACTACCA; reverse: TTGGCGCACAGGGATTG), β-actin (NM_007393) (forward: ACCAGTTCGCCATGGATGAC; reverse: TGCCGGAGCCGTTGTC). Coamplification of the mRNA for mouse β-actin, an invariant internal control, was performed in all samples. Assays were performed in triplicate, and results were normalized to β-actin mRNA levels.

2.5. Immunoblotting

Brain and liver nuclear extracts from five to six mice of each group were prepared using the Nuclear and Cytoplasmic Extraction Kit (kc-435) (Kangchen Bio-tech, Shanghai, China) according to the method described by the manufacturer. The equal volume of nuclear extraction solution was applied to 10% SDS-polyacrylamide gel electrophoresis and transferred to cellulose membranes by using the Western blotting detection system. Membrane sheets were first incubated with antibody against SREBP-1 (sc-366) or PPARβ (sc-7197) for 1 h at room temperature and with antibody against glyceraldehyde-3-phosphate dehydrogenase (GAPDH) (sc-25778) as control, and then washed several times and incubated with goat anti-rabbit IgG-HRP (sc02004) as previously described [25]. The antibodies were manufactured by Santa Cruz Biotechnology (Santa Cruz, CA, USA). Western signal-intensity data were gathered

Table 2
Brain fatty acid composition of the experimental diet groups

	n-3 PUFA-deficient diet	Fish oil n-3 PUFA diets				Flaxseed oil n-3 PUFA diet
		Lower	Low	High	Higher	
C16:0	22.28±0.03[a]	21.64±0.06[b]	21.91±0.23[b]	21.61±0.14[b]	21.93±0.03[b]	21.97±0.17[b]
C18:0	23.52±0.02[a]	23.43±0.09[a]	23.64±0.19[a]	23.29±0.07[a]	22.77±0.06[b]	23.63±0.13[a]
C16:1	1.02±0.01[a]	1.00±0.01[a]	0.94±0.01[a]	1.05±0.02[a]	1.25±0.09[b]	0.98±0.01[a]
C18:1	17.68±0.01[a]	19.41±0.07[b]	18.67±0.33[ac]	19.79±0.07[b]	20.00±0.10[b]	18.85±0.13[c]
C18:2n-6	0.04±0.00	0.04±0.00	0.05±0.01	0.05±0.02	0.04±0.01	0.04±0.01
C18:3n-6	0.02±0.00[a]	0.02±0.00[a]	0.03±0.01[a]	0.04±0.02[a]	0.03±0.01[a]	ND[b]
C20:3n-6	0.57±0.02[a]	0.71±0.01[b]	0.73±0.02[b]	0.76±0.03[b]	0.67±0.01[b]	0.72±0.02[c]
C20:4n-6	12.32±0.03[a]	11.04±0.09[b]	11.10±0.12[b]	9.91±0.05[c]	9.02±0.05[d]	10.28±0.05[f]
C22:4n-6	2.89±0.02[a]	2.32±0.03[b]	2.33±0.07[b]	2.11±0.06[c]	1.65±0.11[d]	2.16±0.07[c]
C22:5n-6	0.61±0.02[a]	0.12±0.03[b]	0.15±0.07[b]	0.09±0.00[c]	0.09±0.02[c]	0.13±0.05[b]
C18:3n-3	3.84±0.01[a]	3.98±0.03[b]	3.76±0.03[c]	4.08±0.04[d]	4.12±0.04[d]	3.82±0.02[a]
C20:4n-3	0.18±0.02	0.16±0.07	0.18±0.04	0.20±0.01	0.19±0.05	0.20±0.02
C20:5n-3	ND[a]	ND[a]	ND[a]	0.46±0.05[b]	0.76±0.04[c]	0.48±0.06[b]
C22:5n-3	0.25±0.03[a]	0.54±0.12[b]	0.70±0.27[c]	0.68±0.14[c]	0.81±0.05[d]	0.82±0.16[d]
C22:6n-3	14.79±0.04[a]	15.58±0.03[b]	15.82±0.27[bc]	15.89±0.16[c]	16.68±0.20[d]	15.92±0.26[c]
ΣSFAs	45.80±0.05[a]	45.07±0.14[a]	45.55±0.41[a]	44.89±0.20[b]	44.70±0.08[b]	45.60±0.28[a]
ΣMUFAs	18.71±0.02[a]	20.42±0.07[b]	19.60±0.32[c]	20.84±0.06[d]	21.24±0.07[e]	19.83±0.13[c]
Σn-6PUFAs	16.44±0.03[a]	14.25±0.12[b]	14.39±0.16[b]	12.96±0.08[c]	11.50±0.12[d]	13.33±0.11[e]
Σn-3PUFAs	19.06±0.04[a]	20.27±0.15[b]	20.45±0.48[b]	21.31±0.23[c]	22.56±0.14[d]	21.24±0.33[c]
AA/DHA	0.83±0.00[a]	0.71±0.01[b]	0.70±0.01[b]	0.62±0.01[c]	0.54±0.01[d]	0.65±0.01[c]
Σn-6/Σn-3	0.86±0.00[a]	0.70±0.01[b]	0.70±0.01[b]	0.61±0.01[c]	0.51±0.01[d]	0.63±0.01[c]

ND, Not detected. Values are means±S.D., n=5–8. Within a fatty acid, means without a common letter in a row differ, P<.05.

by the AlphaImage molecular imaging system (Alpha Innotech, USA), and the expressions of SREBP-1 and PPARβ were normalized to GAPDH.

2.6. Statistical analysis

One-way analysis of variance was used to compare means in all groups. The Scheffe's test was used for *post hoc* analysis. For the unequal variances, Games–Howell analyses were used to compare the data differences. Statistical analyses were performed using SPSS Release 11.5 for Windows. All values were expressed as means±S.D.

3. Results

3.1. Effects of different dietary n-3 PUFAs on fatty acid profiles in mouse plasma, brain and liver

As shown in Tables 2, 3 and 4, dietary supplementation of fish oil or flaxseed oil n-3 PUFAs affected the fatty acid profiles in mouse brain, liver and plasma similarly. Specifically, in all three tissues, as compared with the n-3 PUFA-deficient diet group, the levels of AA decreased and DHA concentrations increased significantly (P<.05), accompanied by marked decrease of AA/DHA ratio in all fish oil and flaxseed oil n-3 PUFA diet groups (P<.05). Also, with the n-3 PUFA-deficient diet, docosapentaenoic acid (22:5n-6) concentration in plasma, liver and brain increased (P<.05). Meanwhile, concentrations of total n-6 PUFAs (Σn-6 PUFA) were lower, and total n-3 PUFA (Σn-3 PUFA) and monounsaturated fatty acid concentrations were higher in all n-3 PUFA diet groups than in the n-3 PUFA-deficient diet group, which led to the decreased ratios of Σn-6/Σn-3 PUFAs in all n-3 PUFA diet groups (P<.05). Furthermore, in fish oil n-3 PUFA diets, with the content of n-3 PUFAs increasing, the concentrations of DHA and Σn-3 PUFAs increased, and those of AA and Σn-6 PUFAs were reduced, resulting in decreased ratios of AA/DHA and Σn-6/Σn-3 PUFAs.

Table 3
Liver fatty acid composition of the experimental diet groups

	n-3 PUFA-deficient diet	Fish oil n-3 PUFA diets				Flaxseed oil n-3 PUFA diet
		Lower	Low	High	Higher	
C16:0	29.68±0.15	30.02±0.20	30.32±0.13	29.68±0.20	29.64±0.04	29.06±0.19
C18:0	13.70±0.12[a]	13.48±0.32[a]	12.99±0.14[ab]	11.20±0.33[b]	11.61±0.06[b]	14.13±0.19[a]
C16:1	1.86±0.24[a]	1.16±0.02[b]	1.36±0.02[ab]	2.33±0.12[c]	2.34±0.05[c]	1.38±0.01[ab]
C18:1	13.63±0.27[a]	11.59±0.20[b]	11.85±0.18[b]	15.23±0.63[c]	14.15±0.14[c]	17.25±0.40[d]
C18:2n-6	17.19±0.08[a]	20.25±0.26[b]	20.20±0.08[b]	19.00±0.15[c]	18.26±0.05[cd]	17.14±0.15[a]
C18:3n-6	0.27±0.00	0.29±0.12	0.16±0.12	0.29±0.25	0.29±0.02	0.30±0.03
C20:3n-6	1.05±0.01	1.04±0.02	1.12±0.02	1.10±0.05	1.13±0.01	1.04±0.01
C20:4n-6	15.54±0.14[a]	11.07±0.11[b]	9.64±0.10[c]	7.06±0.39[d]	7.46±0.04[d]	8.47±0.05[e]
C22:4n-6	0.70±0.02[a]	0.76±0.03[a]	0.91±0.06[a]	1.08±0.08[ab]	1.18±0.06[bc]	1.26±0.06[c]
C22:5n-6	0.63±0.01[a]	0.06±0.01[b]	ND[c]	ND[c]	ND[c]	ND[c]
C18:3n-3	2.09±0.03[a]	2.01±0.02[a]	1.73±0.03[a]	2.25±0.11[a]	2.23±0.03[a]	ND[b]
C20:4n-3	0.20±0.01	0.18±0.02	0.18±0.01	0.23±0.03	0.23±0.01	0.27±0.01
C20:5n-3	0.38±0.02[a]	0.92±0.01[b]	1.64±0.02[c]	2.58±0.10[d]	2.91±0.02[e]	3.40±0.02[f]
C22:6n-3	3.07±0.05[a]	7.16±0.10[b]	7.90±0.09[c]	7.99±0.57[c]	8.57±0.11[d]	6.29±0.06[e]
ΣSFAs	43.38±0.24[a]	43.50±0.52[a]	43.31±0.15[a]	40.88±0.18[b]	41.25±0.09[c]	43.19±0.36[a]
ΣMUFAs	15.50±0.35[a]	12.75±0.19[b]	13.21±0.17[b]	17.56±0.53[c]	16.49±0.15[cd]	18.63±0.40[e]
Σn-6 PUFAs	35.38±0.10[a]	33.47±0.26[b]	32.02±0.15[c]	28.52±0.19[d]	28.33±0.10[d]	28.22±0.16[d]
Σn-3 PUFAs	5.74±0.06[a]	10.27±0.11[b]	11.45±0.06[c]	13.04±0.53[d]	13.94±0.13[e]	9.96±0.07[bd]
AA/DHA	5.06±0.04[a]	1.55±0.01[b]	1.22±0.02[c]	0.88±0.02[d]	0.87±0.01[d]	1.35±0.02[e]
Σn-6/Σn-3	6.16±0.05[a]	3.26±0.01[b]	2.80±0.01[c]	2.19±0.08[d]	2.03±0.01[e]	2.83±0.02[c]

Values are means±S.D., n=5–8. Within a fatty acid, means without a common letter in a row differ, P<.05.

Table 4
Plasma fatty acid composition of the experimental diet groups

	n-3 PUFA-deficient diet	Fish oil n-3 PUFA diets				Flaxseed oil n-3 PUFA diet
		Lower	Low	High	Higher	
C16:0	21.88±0.20[a]	21.35±0.31[a]	19.27±0.12[b]	19.03±0.98[b]	16.76±0.95[c]	17.01±0.07[c]
C18:0	9.50±0.09[a]	8.22±0.05[b]	6.85±0.07[c]	7.82±1.11[d]	7.58±0.88[d]	13.42±0.08[e]
C16:1	3.59±0.07[a]	4.32±0.06[b]	7.39±0.06[c]	8.35±1.87[d]	8.59±0.92[d]	2.11±0.04[e]
C18:1	18.44±0.21[a]	22.58±0.18[b]	36.39±0.21[c]	31.36±2.76[d]	35.03±2.85[e]	20.00±0.03[f]
C18:2n-6	12.75±0.05[a]	10.44±0.12[b]	5.31±0.21[c]	7.44±1.08[d]	7.28±1.20[d]	6.66±0.15[d]
C18:3n-6	0.45±0.07[a]	0.54±0.11[a]	0.23±0.32[b]	0.29±0.41[b]	0.55±0.53[a]	0.32±0.08[b]
C20:3n-6	1.68±0.26[a]	1.81±0.17[a]	1.13±0.24[b]	1.28±0.50[a]	0.83±0.15[b]	1.17±0.28[b]
C20:4n-6	25.67±0.24[a]	21.06±0.30[b]	8.51±0.12[c]	7.73±1.35[c]	5.93±1.42[d]	8.23±0.05[c]
C22:5n-6	1.00±0.07[a]	0.54±0.04[b]	0.08±0.18[c]	0.06±0.14[c]	0.17±0.24[c]	ND[d]
C18:3n-3	2.38±0.03[a]	3.03±0.05[b]	4.62±0.08[c]	5.60±1.18[d]	7.21±0.95[e]	18.34±0.09[f]
C20:5n-3	1.01±0.16[a]	2.99±0.20[b]	8.69±0.13[c]	9.05±1.01[c]	8.02±1.10[c]	7.78±0.09[c]
C22:6n-3	1.65±0.05[a]	1.52±0.05[a]	1.53±0.08[a]	1.99±0.28[b]	2.04±0.13[b]	4.95±0.15[c]
ΣSFAs	31.38±0.28[a]	29.57±0.36[b]	26.12±0.17[c]	26.84±2.08[c]	24.34±1.82[d]	30.43±0.13[ab]
ΣMUFAs	22.03±0.25[a]	26.90±0.24[b]	43.79±0.24[c]	39.71±4.19[d]	43.62±3.47[c]	22.11±0.04[a]
Σn-6PUFAs	41.55±0.34[a]	34.39±0.12[b]	15.26±0.43[c]	16.81±2.06[cd]	14.77±2.22[c]	16.38±0.25[cd]
Σn-3PUFAs	5.04±0.21[a]	9.15±0.21[b]	14.83±0.16[c]	16.64±0.65[d]	17.27±0.93[e]	31.07±0.18[f]
AA/DHA	15.57±0.58[a]	6.75±0.12[b]	5.58±0.06[c]	3.88±0.48[d]	2.91±0.33[d]	1.66±0.05[e]
Σn-6/Σn-3	8.25±0.29[a]	3.76±0.09[b]	1.03±0.04[c]	1.01±0.13[c]	0.86±0.16[d]	0.53±0.01[e]

Values are means±S.D., n=5–8. Within a fatty acid, means without a common letter in a row differ, P<.05.

Comparison between the higher fish oil n-3 PUFA diet and the flaxseed oil diet showed that plasma concentrations of LNA and DHA were 2.54- and 2.42-fold higher in the flaxseed oil n-3 PUFA diet group than in the higher fish oil n-3 PUFA diet group (P<.05), with 43% and 38% lower ratios of AA/DHA and total Σn-6/Σn-3 PUFAs, respectively (P<.05) (Table 4). However, the concentrations of brain DHA were lower, and the ratios of both AA/DHA and Σn-6/Σn-3 PUFAs were higher in the flaxseed oil n-3 PUFA diet group than in the higher fish oil n-3 PUFA diet group (P<.05) (Table 2).

3.2. Effects of different dietary n-3 PUFAs on the expressions of D6D and D5D in mouse liver and brain

Changes in the expression of D6D and D5D were determined after 3 months of feeding with n-3 PUFA-rich diets. As shown in Table 5, in the liver, compared to the n-3 PUFA-deficient diet group, the levels of mRNA expressions of D6D in all fish oil n-3 PUFA diet and the flaxseed oil n-3 PUFA diet groups were significantly decreased (P<.05), with no differences in D5D expressions among the different diet groups. In the brain, compared to the n-3 PUFA-deficient diet group, the decreased mRNA expressions of both D6D and D5D were only shown in the higher fish oil n-3 PUFA diet group (P<.05), whereas no changes were found in other fish oil n-3 PUFA diet and the flaxseed oil n-3 PUFA diet groups. Also, the expressions of both D6D and D5D in the higher fish oil n-3 PUFA diet group were lower than in the flaxseed oil n-3 PUFA diet group (P<.05).

Table 5
Effects of different n-3 PUFAs on D5D and D6D desaturase expressions in mice

Groups	Liver		Brain	
	D5D	D6D	D5D	D6D
n-3 PUFA-deficient diet	0.79±0.51	2.61±0.65[a]	4.03±2.19[a]	4.52±2.61[a]
Fish oil n-3 PUFA diet				
Lower	0.54±0.03	1.18±0.41[b]	3.19±1.68[a]	2.82±0.98[a]
Low	0.70±0.26	1.54±0.56[b]	4.29±1.51[a]	3.89±1.31[a]
High	0.91±0.46	–	3.07±1.83[a]	2.92±1.07[ab]
Higher	0.94±0.13	0.93±0.43[b]	1.40±0.56[b]	1.80±0.79[b]
Flaxseed oil n-3 PUFA diet	0.75±0.09	1.17±0.19[b]	3.91±0.63[a]	4.72±2.11[a]

Mice were fed one of the six types of diets for 3 months. Brains and livers were collected.
Tissue mRNA expressions of D6D and D5D were determined by RT-PCR, and data were normalized to β-actin mRNA levels. Values are means±S.D., n=5–8. Within an enzyme, means without a common letter in a column differ, P<.05.

3.3. Effects of different dietary n-3 PUFAs on expressions of SREBP and PPARβ in mouse liver and brain

Furthermore, we examined whether dietary intake of very long chain n-3 PUFAs or their precursor LNA could differently affect the expressions of tissue SREBP-1 and PPARβ. As illustrated in Fig. 1, the expressions of hepatic nuclear SREBP-1 were lower in both the higher fish oil n-3 PUFA diet and the flaxseed oil n-3 PUFA diet groups (0.38±0.05 and 0.40±0.1) than in the n-3 PUFA-deficient diet group (0.6±0.13) (P<.05). Whereas, in the brain, decreased expressions of nuclear SREBP-1 occurred only in the higher fish oil n-3 PUFA diet group (0.51±0.06) compared to the n-3 PUFA-deficient diet group (0.63±0.12) (P<.05).

Fig. 2 shows the effects of dietary n-3 PUFA quantity on the expressions of nuclear SREBP-1. In the liver, the expressions of SREBP-1 were lower in all fish oil n-3 PUFA diet (lower, low, high and higher) groups (0.75±0.1, 0.82±0.2, 0.9±0.24 and 0.92±0.14, respectively) than in the n-3 PUFA-deficient diet group (1.39±0.35) (P<.05). Whereas, in the brain, only the diet with higher fish oil n-3 PUFAs had reduced SREBP-1 expressions (0.6±0.08) as compared with the n-3 PUFA-deficient diet (0.78±0.11) (P<.05).

For the PPARβ expression, no differences were found in both the brain and liver among all fish oil n-3 PUFA diet, the flaxseed oil n-3 PUFA diet and the n-3 PUFA-deficient diet groups (data not shown).

4. Discussion

In this study, using mouse models fed n-3 PUFA-rich and -deficient diets, we found that either fish oil or flaxseed oil n-3 PUFA diets significantly increased DHA and decreased AA concentrations in the brain as well as in the liver and plasma. We also found that the flaxseed oil n-3 PUFA diet, with similar higher amount of n-3 PUFAs, was less effective than the fish oil n-3 PUFA diet in increasing brain DHA content. Furthermore, the expressions of D6D and SREBP-1 in the liver were down-regulated by all fish oil diets with different amounts of n-3 PUFAs (EPA and DHA) ranging from 0.46% to 4.29% total energy, as well as by the flaxseed oil diet with higher amount of LNA (5.01% total energy), whereas, in the brain, D6D, D5D and SREBP-

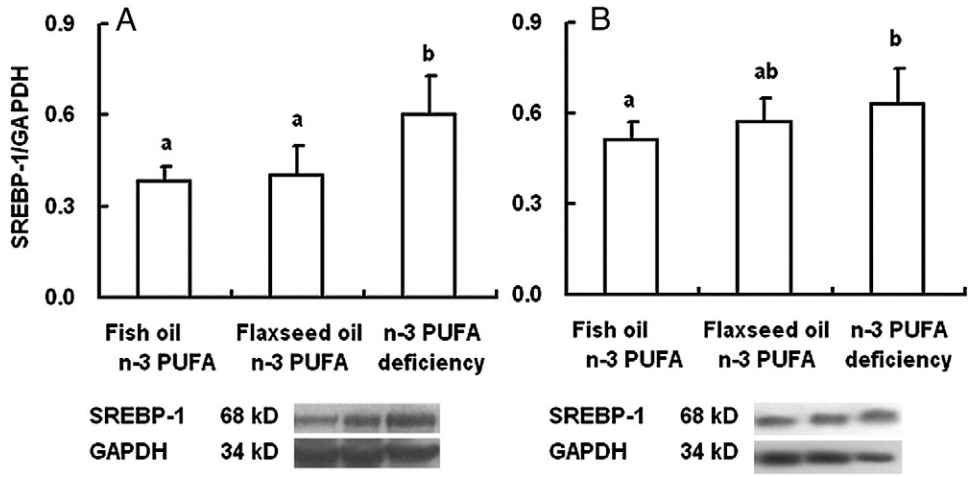

Fig. 1. Western blot analysis of SREBP-1 expressions in mouse brain and liver nuclear extract after 3 months of dietary intake of higher fish oil or flaxseed oil n-3 PUFAs. The protein expressions were normalized to GAPDH. (A) Liver; (B) brain. Values are means±S.D., n=5–6. Means without a common letter differ, P<.05.

1 expressions were down-regulated by the higher fish oil n-3 PUFA diet rather than by other fish oil n-3 PUFA diets and by the flaxseed oil n-3 PUFA diet.

Accretion of DHA in the brain primarily relies on circulating plasma DHA derived from diet or biosynthesized from LNA in the liver, although local biosynthesis in astrocytes occurs [1,8,9,26,27].

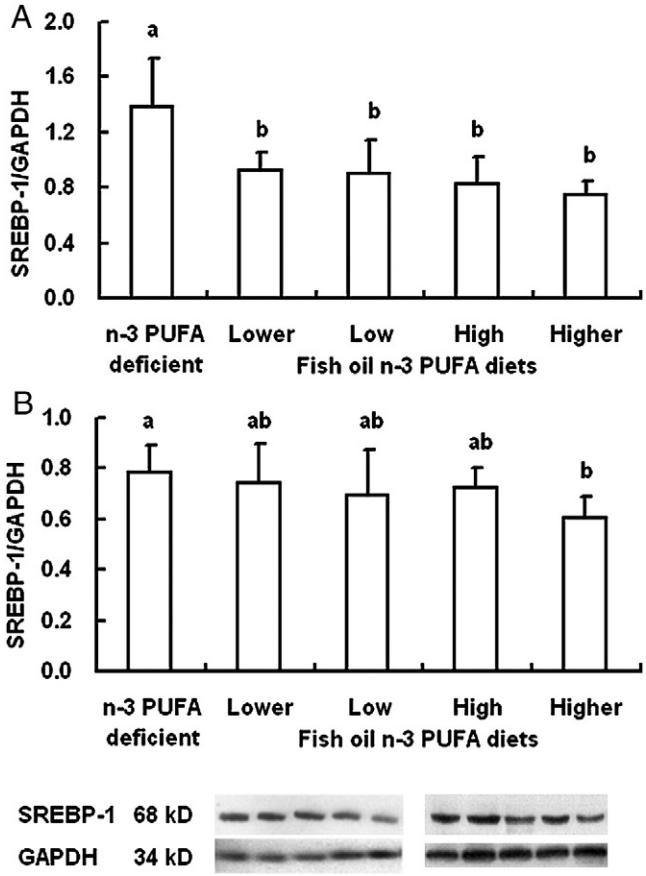

Fig. 2. Western blot analysis of SREBP-1 expressions in mouse brain and liver nuclear extract after 3 months of feeding of fish oil diets with different amounts of n-3 PUFAs — lower, low, high and higher (0.46%, 0.91%, 1.73% and 4.29% total energy, respectively). The protein expressions were normalized to GAPDH. (A) Liver; (B) brain. Values are means±S.D., n=5–6. Means without a common letter differ, P<.05.

DHA concentrations in brain lipids are altered by the type and amount of fatty acids in the diet [28,29]. Our results again confirm previous studies that have demonstrated that dietary restriction of n-3 PUFAs in developing animals results in reduced brain DHA level and that dietary n-3 PUFA supplementation can increase DHA concentration [10,30]. Meanwhile, we also found that dietary intake of n-3 PUFAs decreased AA concentration in the brain, liver and plasma, which may result from the competition between LNA and LA for D6D as well as from the competition between the two 24 carbon fatty acids (24:5n-3 and 24:4n-6) at the second use of the enzyme in the process of longer-chain PUFA synthesis [5].

Some studies demonstrated that the conversion rate of LNA to DHA in the liver is several-fold higher than brain DHA consumption. Therefore, in the absence of dietary DHA, a normal brain DHA content can be maintained by liver conversion of LNA to circulating DHA, provided that sufficient LNA is in the diet [11,31]. However, the general conversion of LNA to EPA in the body is limited and further transformation to DHA is low [5]. Thus, LNA appears to be a limited source of longer-chain n-3 PUFAs, and adequate intakes of preformed longer-chain n-3 PUFAs, in particular DHA, may be important for maintaining optimal tissue (including brain) function [5,9,32]. In keeping with this, we found in this study that the flaxseed oil diet doubled plasma DHA concentration but did not increase brain DHA level to the content of the fish oil diet with equal quantity of n-3 PUFAs, indicating that preformed DHA in diets is better than dietary LNA for brain DHA accretion, although the mechanism by which this occurs is not yet clear. Anderson et al. [33] reported that [^{14}C]DHA-labeled chylomicrons, injected to hepatectomized rats, had an average 65% greater incorporation into the brain than chylomicrons prepared from [^{14}C]16:0, [^{14}C]LA and [^{14}C]LNA.

D6D and D5D, key enzymes in longer-chain PUFA production, are expressed in many tissues, but their expression is highest in the liver and brain [12,13]. Both enzymes are coordinately regulated by various dietary conditions, hormonal levels as well as PUFAs [14]. Although in one report, feeding a diet deficient in LNA to rats for 2 and 6 months did not affect liver D6D activity [34], other studies demonstrate that feeding with fish oil rich in n-3 PUFAs and corn oil rich in LA greatly decreased liver desaturase activities [12,35,36] and that a diet deficient in LA, LNA or both increased desaturase activities in rat liver [37,38]. Therefore, changes in the expression of desaturases indicate that these enzymes are involved in feedback regulation in the production of longer-chain PUFAs.

Very recently, in rats fed an n-3 PUFA-adequate diet (4.6% LNA of total fatty acids, no DHA) compared with an n-3 PUFA-deficient diet

(0.2% LNA, no DHA), mRNA and activity levels of D5D and D6D were down-regulated in the liver but not in the brain [39]. This is consistent with our results showing that D6D mRNA expression in mouse liver, but not in the brain, was down-regulated by dietary supplementation of flaxseed oil LNA. Our findings showing little changes in brain D6D and D5D expression between flaxseed oil and n-3 PUFA-deficient diets indicate that the brain's ability to synthesize DHA from LNA is low and is not altered by LNA deprivation as in other reports [30]. However, in our study, in mice fed the fish oil diet with higher amount of EPA and DHA similar to LNA in the flaxseed oil diet, not only hepatic D6D expression but also brain D6D and D5D expressions were down-regulated compared to the n-3 PUFA-deficient diet. This may account for the higher rates of β-oxidation and esterification of LNA into stable lipids in the liver and brain and the lower conversion of LNA to DHA in rats on a high-DHA-containing diet [30,40].

The mechanisms by which n-3 PUFAs affect the desaturase expression may have a complex explanation. Studies of liver and adipose tissue demonstrated that D6D, D5D and other lipogenic enzymes such as elongases and lipoprotein lipase are regulated at the transcriptional level during different metabolic states by SREBPs [14,41–43]. PUFAs were reported to inhibit SREBP-1 activity by multiple mechanisms with effects of n-3 PUFAs>n-6 PUFAs [44–46]. Feeding rats a diet high in DHA or LNA down-regulates the transcription of liver D5 and D6 desaturase and elongase genes as well as the liver expression of SREBP-1, which positively affects their transcription [12,15,45]. Therefore, it is likely that down-regulation of D6D and D5D mRNA expression by n-3 PUFAs in our findings and other reports [39] could be mediated through suppression of SREBP-1. Of interest, in this study, we found that dietary flaxseed oil n-3 PUFAs inhibited protein expression of SREBP-1 in the liver but not in the brain, whereas dietary fish oil n-3 PUFAs inhibited SREBP-1 expression in both the liver and the brain, consistent with the down-regulation of the expressions of D6D and D5D.

PPARs play an important role in the pathogenesis of various disorders of the CNS [47]. All three isotypes (PPARα, PPARβ and PPARγ) are co-expressed in the nervous system during late embryogenesis. Whereas PPARβ remains highly expressed in this tissue, the expression of PPARα and γ decreases postnatally in the brain [48,49]. Tang et al. [50] have shown that the promoter region of D6D contains the response element for the ligand-activated transcription factor PPARα and that binding of DHA to PPARα suppresses the transcription of D6D, but we did not examine this. Considering that PPARβ is the prevalent isotype both prenatally and postnatally [48,49] and may be related to lipid metabolism in the brain [47], we determined its expression in the present study. Our results showed that the expression of PPARβ was not affected by dietary n-3 PUFA status.

Still, when the effects of fish oil diets with different amounts of n-3 PUFAs on brain accretion of DHA and on expressions of desaturases and SREBP-1 were compared, all four fish oil n-3 PUFA-containing diets reduced hepatic D5D and SREBP-1 expressions. However, in the brain, only the diet with higher amount of n-3 PUFAs inhibited expressions of D6D, D5D and SREBP-1, whereas the other three diets with less n-3 PUFAs had the same effects as the n-3 PUFA-deficient diet. Note that, in humans, in the United States, the average intake of n-3 fatty acid is about 0.7% of total energy [51], while in Eskimos, the average intake of n-3 fatty acid, mostly from fish oils, is about 5% of total energy [52]. Thus, the higher fish oil n-3 PUFA diet containing 4.29% of total energy from DHA plus EPA (Table 1) in our study corresponds to the average fish oil intake in Eskimos. These data suggest that more dietary longer-chain n-3 PUFAs may be needed for DHA accretion in the brain than what we have recommended, and this idea needs further study.

Taken together, dietary intake of fish oil longer-chain n-3 PUFAs had higher ability to enhance brain DHA concentration than flaxseed oil LNA, indicating that preformed dietary DHA, different from those converted by LNA inside the body, is better for brain accretion. Meanwhile, high intake of n-3 PUFAs may be needed for DHA accretion in the brain. Dietary longer-chain n-3 PUFAs affect the expressions of D6D, D5D and SREBP-1 in the brain differently from their precursor LNA.

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References

[1] Innis SM. Dietary (n-3) fatty acids and brain development. J Nutr 2007;137:855–9.
[2] Mccann JC, Ames BN. Is docosahexaenoic acid, an n-3 long-chain polyunsaturated fatty acid, required for development of normal brain function? An overview of evidence from cognitive and behavioral tests in humans and animals. Am J Clin Nutr 2005;82:281–95.
[3] Kawakita E, Hashimoto M, Shido O. Docosahexaenoic acid promotes neurogenesis in vitro and in vivo. Neuroscience 2006;139:991–7.
[4] Assisi A, Banzi R, Buonocore C, Capasso F, Di Muzio V, Michelacci F, et al. Fish oil and mental health: the role of n-3 long-chain polyunsaturated fatty acids in cognitive development and neurological disorders. Int Clin Psychopharmacol 2006;21:319–36.
[5] Burdge GC, Calder PC. Conversion of alpha-linolenic acid to longer-chain polyunsaturated fatty acids in human adults. Reprod Nutr Dev 2005;45:581–97.
[6] Seo T, Blaner WS, Deckelbaum RJ. Omega-3 fatty acids: molecular approaches to optimal biological outcomes. Curr Opin Lipidol 2005;16:11–8.
[7] Yehuda S, Rabinovitz S, Mostofsky DI. Essential fatty acids are mediators of brain biochemistry and cognitive functions. J Neurosci Res 1999;56:565–70.
[8] Qi K, Hall M, Deckelbaum RJ. Long-chain polyunsaturated fatty acid accretion in brain. Curr Opin Clin Nutr Metab Care 2002;5(2):133–8.
[9] Lefkowitz W, Lim SY, Lin Y, Salem Jr N. Where does the developing brain obtain its docosahexaenoic acid? Relative contributions of dietary alpha-linolenic acid, docosahexaenoic acid, and body stores in the developing rat. Pediatr Res 2005;57:157–65.
[10] Igarashi M, DeMar Jr JC, Ma K, Chang L, Bell JM, Rapoport SI. Docosahexaenoic acid synthesis from alpha-linolenic acid by rat brain is unaffected by dietary n-3 PUFA deprivation. J Lipid Res 2007;48:1150–8.
[11] Igarashi M, DeMar Jr JC, Ma K, Chang L, Bell JM, Rapoport SI. Upregulated liver conversion of alpha-linolenic acid to docosahexaenoic acid in rats on a 15 week n-3 PUFA-deficient diet. J Lipid Res 2007;48:152–64.
[12] Connor WE, Neuringer M, Reisbick S. Essential fatty acids: the importance of n-3 fatty acids in the retina and brain. Nutr Rev 1992;50:21–9.
[13] Simopoulos AP. Evolutionary aspects of diet, the omega-6/omega-3 ratio and genetic variation: nutritional implications for chronic diseases. Biomed Pharmacother 2006;60:502–7.
[14] Gibbons A. Humans' head start: new views of brain evolution. Science 2002;296:836–7.
[15] Cho HP, Nakamura MT, Clarke SD. Cloning, expression, and nutritional regulation of the mammalian Delta-6 desaturase. J Biol Chem 1999;274:471–7.
[16] Cho HP, Nakamura M, Clarke SD. Cloning, expression, and fatty acid regulation of the human delta-5 desaturase. J Biol Chem 1999;274:37335–9.
[17] Nakamura MT, Nara TY. Essential fatty acid synthesis and its regulation in mammals. Prostaglandins Leukot Essent Fatty Acids 2003;68:145–50.
[18] Matsuzaka T, Shimano H, Yahagi N, Amemiya-Kudo M, Yoshikawa T, Hasty AH, et al. Dual regulation of mouse Delta (5)- and Delta (6)-desaturase gene expression by SREBP-1 and PPARalpha. J Lipid Res 2002;43:107–14.
[19] Xu J, Nakamura MT, Cho HP, Clarke SD. Sterol regulatory element binding protein-1 expression is suppressed by dietary polyunsaturated fatty acids. A mechanism for the coordinate suppression of lipogenic genes by polyunsaturated fats. J Biol Chem 1999;274:23577–83.
[20] Nakatani T, Kim HJ, Kaburagi Y, Yasuda K, Ezaki O. A low fish oil inhibits SREBP-1 proteolytic cascade, while a high-fish-oil feeding decreases SREBP-1 mRNA in mice liver: relationship to anti-obesity. J Lipid Res 2003;44:369–79.
[21] Houseknecht KL, Cole BM, Steele PJ. Peroxisome proliferator-activated receptor gamma (PPARgamma) and its ligands: a review. Domest Anim Endocrinol 2002;22:1–23.
[22] Lepage G, Roy CC. Direct transesterification of all classes of lipids in a one-step reaction. J Lipid Res 1986;27:114–20.
[23] Rodríguez-Palmero M, Lopez-Sabater MC, Castellote-Bargallo AI, De la Torre-Boronat MC, Rivero-Urgell M. Comparison of two methods for the determination of fatty acid profiles in plasma and erythrocytes. J Chromatogr A 1997;778:435–9.
[24] Whelan JA, Russell NB, Whelan MA. A method for the absolute quantification of cDNA using real-time PCR. J Immunol Methods 2003;278:261–9.
[25] Lawler Jr JF, Yin M, Diehl AM, Roberts E, Chatterjee S. Tumor necrosis factor-alpha stimulates the maturation of sterol regulatory element binding protein-1 in human hepatocytes through the action of neutral sphingomyelinase. J Biol Chem 1998;273:5053–9 Related articles, links.

[26] Bernoud N, Fenart E, Bénistant C, Pageaux JF, Dehouck MP, Molière P, et al. Astrocytes are mainly responsible for the polyunsaturated fatty acid enrichment in blood–brain barrier endothelial cells in vitro. J Lipid Res 1998;39:1816–24.
[27] Kim HY. Novel metabolism of docosahexaenoic acid in neural cells. J Biol Chem 2007;282:18661–5.
[28] Ozias MK, Carlson SE, Levant B. Maternal parity and diet (n-3) polyunsaturated fatty acid concentration influence accretion of brain phospholipid docosahexaenoic acid in developing rats. J Nutr 2007;137:125–9.
[29] Diau GY, Hsieh AT, Sarkadi-Nagy EA, Wijendran V, Nathanielsz PW, Brenna JT. The influence of long-chain polyunsaturate supplementation on docosahexaenoic acid and arachidonic acid in baboon neonate central nervous system. BMC Med 2005; 3:11.
[30] Anderson GJ, Neuringer M, Lin DS, Connor WE. Can prenatal n-3 fatty acid deficiency be completely reversed after birth? Effects on retinal and brain biochemistry and visual function in rhesus monkeys. Pediatr Res 2005;58:865–72.
[31] Rapoport SI, Rao JS, Igarashi M. Brain metabolism of nutritionally essential polyunsaturated fatty acids depends on both the diet and the liver. Prostaglandins Leukot Essent Fatty Acids 2007;77:251–61.
[32] Bowen RA, Clandinin MT. Maternal dietary 22:6n-3 is more effective than 18:3n-3 in increasing the 22:6n-3 content in phospholipids of glial cells from neonatal rat brain. Br J Nutr 2005;93:601–11.
[33] Anderson GJ, Tso PS, Connor WE. Incorporation of chylomicron fatty acids into the developing rat brain. J Clin Invest 1994;93:2764–7.
[34] Dinh TK, Bourre JM, Durand G. Effect of age and alpha-linolenic acid deficiency on delta 6 desaturase activity and liver lipids in rats. Lipids 1993;28:517–23.
[35] Garg ML, Thomson AB, Clandinin MT. Effect of dietary cholesterol and/or omega 3 fatty acids on lipid composition and delta 5-desaturase activity of rat liver microsomes. J Nutr 1988;118:661–8.
[36] Garg ML, Sebokova E, Thomson AB, Clandinin MT. Delta 6-desaturase activity in liver microsomes of rats fed diets enriched with cholesterol and/or omega 3 fatty acids. Biochem J 1988;249:351–6.
[37] Peluffo RO, Nervi AM, Brenner RR. Linoleic acid desaturation activity of liver microsomes of essential fatty acid deficient and sufficient rats. Biochim Biophys Acta 1976;441:25–31.
[38] Bernert Jr JT, Sprecher H. Studies to determine the role rates of chain elongation and desaturation play in regulating the unsaturated fatty acid composition of rat liver lipids. Biochim Biophys Acta 1975;398:354–63.
[39] Igarashi M, Ma K, Chang L, Bell JM, Rapoport SI. Dietary n-3 PUFA deprivation for 15 weeks upregulates elongase and desaturase expression in rat liver but not brain. J Lipid Res 2007;48:2463–70.
[40] Demar Jr JC, Ma K, Chang L, Bell JM, Rapoport SI. alpha-Linolenic acid does not contribute appreciably to docosahexaenoic acid within brain phospholipids of adult rats fed a diet enriched in docosahexaenoic acid. J Neurochem 2005;94(4): 1063–76.
[41] Wang Y, Botolin D, Christian B, Busik J, Xu J, Jump DB. Tissue-specific, nutritional, and developmental regulation of rat fatty acid elongases. J Lipid Res 2005;46: 706–15.
[42] Wang Y, Torres-Gonzalez M, Tripathy S, Botolin D, Christian B, Jump DB. Elevated hepatic fatty acid elongase-5 activity affects multiple pathways controlling hepatic lipid and carbohydrate composition. J Lipid Res 2008;49: 1538–52.
[43] Okamoto K, Kakuma T, Fukuchi S, Masaki T, Sakata T, Yoshimatsu H. Sterol regulatory element binding protein (SREBP)-1 expression in brain is affected by age but not by hormones or metabolic changes. Brain Res 2006;1081: 19–27.
[44] Jump DB. The biochemistry of n-3 polyunsaturated fatty acids. J Biol Chem 2002; 277(11):8755–8.
[45] Botolin D, Wang Y, Christian B, Jump DB. Docosahexaenoic acid (22:6n-3) regulates rat hepatocyte SREBP-1 nuclear abundance by Erk- and 26S proteasome-dependent pathways. J Lipid Res 2006;47:181–92.
[46] Xu J, Teran-Garcia M, Park JH, Nakamura MT, Clarke SD. Polyunsaturated fatty acids suppress hepatic sterol regulatory element-binding protein-1 expression by accelerating transcript decay. J Biol Chem 2001;276:9800–7.
[47] Heneka MT, Landreth GE. PPARs in the brain. Biochim Biophys Acta 2007;1771: 1031–45.
[48] Braissant O, Foufelle F, Scotto C, Dauça M, Wahli W. Differential expression of peroxisome proliferator-activated receptors (PPARs): tissue distribution of PPAR-alpha, -beta, and -gamma in the adult rat. Endocrinology 1996;137: 354–66.
[49] Braissant O, Wahli W. Differential expression of peroxisome proliferator-activated receptor-alpha, -beta, and -gamma during rat embryonic development. Endocrinology 1998;139:2748–54.
[50] Tang C, Cho HP, Nakamura MT, Clarke SD. Regulation of human delta-6 desaturase gene transcription: identification of a functional direct repeat-1 element. J Lipid Res 2003;44:686–95.
[51] Kris-Etherton PM, Taylor DS, Yu-Poth S, Huth P, Moriarty K, Fishell V, et al. Polyunsaturated fatty acids in the food chain in the United States. Am J Clin Nutr 2000;71:179S–88S.
[52] Feskens EJ, Kromhout D. Epidemiologic studies on Eskimos and fish intake. Ann N Y Acad Sci 1993;683:9–15.