Effects of Dietary Supplementation of Seabuckthorn (*Hippophae rhamnoides*) Oils on Fatty Acids in Patients with Atopic Dermatitis

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Abstract: Forty-nine patients received a daily dose of 5 g seabuckthorn (*Hippophae rhamnoides*) seed oil (from seeds), pulp oil (from pulp/peel of the berries) or paraffin oil for four months. The fatty acid compositions of plasma phospholipids and neutral lipids of the patients were analyzed before, after one month and at the end of the treatment. Skin biopsies were taken from sixteen patients, and the fatty acid compositions of skin glycerophospholipids were analyzed before and after the treatment. Supplementation of the seed oil significantly increased the proportion of α-linolenic acid in plasma neutral lipids ($p < 0.01$) and total n-3 fatty acids in plasma phospholipids and neutral lipids ($p < 0.05$). Increases in the proportion of α-linolenic acid, linoleic acid and eicosapentaenoic acid in plasma phospholipids by the seed oil treatment were close to significant ($0.05 < p < 0.1$). The pulp oil treatment increased the proportion of palmitoleic acid and lowered the proportion of pentadecanoic acid in both plasma phospholipids and neutral lipids ($p < 0.01$). Seed oil treatment slightly increased the level of docosapentaenoic acid (22:5n-3) and decreased the level of palmitic acid (16:0) in skin glycerophospholipids ($0.05 < p < 0.1$). These results indicate a higher efficiency of incorporation and metabolism of α-linolenic acid than linoleic acid and a relatively stable fatty acid composition of skin glycerophospholipids.

Key words: Seabuckthorn oils, *Hippophae rhamnoides* L., atopic dermatitis, fatty acid composition, plasma, skin

1. Introduction

After Hansen (1933) first described the fatty acid abnormality in atopic dermatitis (AD), the issue has been investigated and discussed by many researchers. Abnormal levels of fatty acids have been recognized in the plasma, skin, adipose tissue and breast milk of AD patients compared to healthy controls (Manku et al. 1984, Oliwiecki et al. 1991, Strannegård et al. 1987, Duchen et al. 1998, Wright 1990, Schäfer and Kragballe 1991). This abnormalty is thought to be due to a deficiency in both incorporation and metabolism, especially Δ-6 desaturation, of essential fatty acids in AD patients (Oliwiecki et al. 1991, Wright 1990). However, contradictory results have also been reported by some investigators (Pfeiffer et al. 1996, Zevenbergen and Houtsmuller 1989). The effects of dietary fatty acid supplementations on the fatty acid compositions of tissues, especially of the skin, require further study.

Seabuckthorn seed oil contains a high content of the two essential fatty acids, linoleic acid and α-linolenic acid (Chen et al. 1990), which are precursors of other polyunsaturated fatty acids such as arachidonic and eicosapentaenoic acids. The oil from the pulp/peel of seabuckthorn berries is rich in palmitoleic acid and oleic acid (Chen et al. 1990). In the present study, we tested the effects of dietary supplementation with the two oils on the fatty acid composition of plasma phospholipids, plasma neutral lipids and skin glycerophospholipids of AD patients.

2. Materials and Methods

The Ethical Committee of Turku University Central Hospital approved the experiment. The purpose of the investigation was explained to all participating patients and they signed written consents.

The seeds and soft parts (berry flesh and peel) were separated from the dried press residue of the seabuckthorn juice. Seed oil (SO) was extracted from the seeds and pulp oil (PO) from the soft parts by an aseptic supercritical CO_2 process (Manninen et al. 1997). The fatty acid compositions of the two oils analyzed as methyl esters with gas chromatography are shown in Table 1.

Table 1. Fatty acid composition of seabuckthorn seed and pulp oils (weight percentage).

Oils	Fatty acids (%)						
	16:0	16:1n-7	18:0	18:1n-9	18:1n-7	18:2n-6	18:3n-3
Seed oil	11.3	4.4	2.6	18.9	3.2	34.1	24.9
Pulp oil	33.4	24.9	1.0	26.2	7.3	5.1	1.6

Forty-nine patients were randomly divided into three groups taking orally ten oil capsules (5g oil) of SO, PO or paraffin oil per day, the whole treatment period lasting four months. The patients were asked to follow their normal diet throughout the trial. Plasma samples were taken from the patients at the beginning and after one and four months. Punch biopsies of skin (1mm thick of epidermal layer) were taken from sixteen patients before and after the treatment. Lipids were extracted from plasma samples and skin biopsies with a solvent mixture of chloroform:methanol (2:1) and fractionated into neutral lipids and phospholipids with solid phase extraction (Yang et al. 1999a, Yang et al. 1999b). The fatty acid compositions of plasma neutral lipids and phospholipids as well as skin glycerophospholipids were analyzed with a gas chromatograph and calculated as weight percentages of fatty acid methyl esters (Yang et al. 1999a). The data analyses were carried out with statistical programme packages Statistic/W version 4.5 (Stat Soft Inc., Tulsa, OK) and SPSS 7.5 (SPSS Inc., Chicago, IL).

3. Results

As shown in Table 2, the proportion of α-linolenic acid in plasma phospholipids in the SO group already increased after one month's administration ($p = 0.08$) and remained stable for the next three months. The proportions of linoleic acid and eicosapentaenoic acid (20:5n-3) in plasma phospholipids were also slightly increased by the SO treatment ($0.05 < p < 0.1$). The level of total n-3 polyunsaturated fatty acids also increased after the SO treatment. Supplementation with PO clearly increased the proportion of palmitoleic acid of phospholipid fatty acids ($p < 0.05$) and decreased the proportion of pentadecanoic acid (15:0) ($p < 0.01$) in plasma phospholipids.

Table 2. Changes in fatty acid composition in plasma phospholipids in the SO and PO groups during follow up. A: at the beginning of the trial; B: after one month treatment; C: after four months treatment. n = 12 in the seed oil group; n = 16 in the pulp oil group.

Fatty acids	Seed oil group			Pulp oil group		
	A	B	C	A	B	C
	mean ± std.	mean ± std.	mean ± std.	mean ± std.	mean ± std.	mean ± std.
15:0	0.17 ± 0.00	0.16 ± 0.03	0.16 ± 0.04	0.18 ± 0.03	0.15 ± 0.00***	0.16 ± 0.03
16:1n-7	1.08 ± 1.75	0.70 ± 0.33	0.60 ± 0.19	0.67 ± 0.04	0.86 ± 0.25**	0.81 ± 0.22*
18:2n-6	22.44 ± 2.55	22.55 ± 2.81	23.89 ± 2.57*	21.99 ± 3.15	21.10 ± 2.99	23.21 ± 3.01
18:3n-3	0.29 ± 0.07	0.37 ± 0.14*	0.37 ± 0.14*	0.31 ± 0.13	0.31 ± 0.14	0.34 ± 0.13
20:5n-3	1.19 ± 0.40	1.41 ± 0.50*	1.30 ± 0.59	1.18 ± 0.62	1.16 ± 0.71	1.21 ± 0.56
n-3	2.44 ± 0.68	2.98 ± 0.65**	2.79 ± 0.88*	2.88 ± 1.06	2.61 ± 0.88	2.56 ± 0.71

* $0.05 < p < 0.1$, ** $p < 0.05$, *** $p < 0.01$.

Table 3 shows the effects of the oil supplements on the fatty acid composition of plasma neutral lipids. At the end of the administration, the percentage of α-linolenic acid increased in the SO group from 1.31 to 1.86 % (Table 3). The increase was thus over 40 %. SO treatment also increased the proportion of total n-3 polyunsaturated fatty acids. The PO administration increased the percentage of palmitoleic acid and lowered the percentage of pentadecanoic acid ($p < 0.01$) (Table 3). All the major fatty acid species such as oleic, linoleic and palmitic acids remained fairly constant in all the participants.

Table 3. Changes in fatty acid composition in plasma neutral lipids in the SO and PO groups during follow up. A: at the beginning of the trial; B: after one month treatment; C: after four months treatment. n = 12 in the seed oil group; n = 16 in the pulp oil group.

Fatty acids	Seed oil group			Pulp oil group		
	A	B	C	A	B	C
	mean ± std.	mean ± std.	Mean ± std.	mean ± std.	mean ± std.	mean ± std.
15:0	0.24 ± 0.08	0.23 ± 0.08	0.23 ± 0.06	0.24 ± 0.05	0.21 ± 0.04***	0.20 ± 0.04***
16:1n-7	3.51 ± 1.18	3.59 ± 1.10	3.13 ± 0.80	3.69 ± 1.11	4.45 ± 1.28**	4.12 ± 1.28*
18:2n-6	27.12 ± 9.12	29.07 ± 9.19	26.39 ± 10.09	29.39 ± 6.78	29.44 ± 7.63	31.22 ± 6.62
18:3n-3	1.31 ± 0.63	1.67 ± 0.63***	1.86 ± 0.57**	1.24 ± 0.50	1.14 ± 0.34	1.24 ± 0.48
20:5n-3	0.58 ± 0.22	0.67 ± 0.32	0.61 ± 0.39	0.64 ± 0.32	0.69 ± 0.55	0.64 ± 0.36
n-3	2.58 ± 0.57	2.90 ± 0.65**	2.75 ± 0.79	2.59 ± 0.89	2.51 ± 0.95	2.61 ± 0.71

* $0.05 < p < 0.1$, ** $p < 0.05$, *** $p < 0.01$.

The paraffin oil supplementation did not lead to any changes in the fatty acid composition of plasma lipids.

The fatty acid composition of skin glycerophospholipids during follow up is shown in Table 4. The SO treatment increased the proportion of docosapentaenoic acid (22:5n-3) and decreased the proportion of palmitic acid (16:0) in skin glycerophospholipids almost significantly ($0.05 < p < 0.1$). The PO treatment slightly increased the proportion of stearic acid ($0.05 < p < 0.1$). A small increase in the proportion of linoleic acid and stearic acid was also observed in the placebo group ($0.05 < p < 0.1$).

4. Discussion

The efficiency of incorporation and metabolism of dietary linoleic and α-linolenic acids strongly affects the essential fatty acid status of the human body.

In the present study, clear increases in the levels of α-linolenic acid and total n-3 fatty acids in plasma phospholipids and neutral lipids were already recognized after a one-month administration of SO. The level of eicosapentaenoic acid in plasma phospholipids was also increased. The high content of linoleic acid in SO resulted in only a slight increase in its level in plasma phospholipids at the end of the treatment without affecting the levels of its long chain, desaturated metabolites. These results show that α-linolenic acid was more efficiently incorporated and metabolized compared with linoleic acid. α-linolenic acid may have competitively inhibited the incorporation and desaturation of linoleic acid (Popp-Snijders et al. 1984). The results of the present study do not show strong evidence of Δ-6 desaturase deficiency in AD.

The increase in the level of α-linolenic acid in plasma lipids showed a clear improving effect on AD symptoms (Yang et al. 1999a). These effects of α-linolenic acid may have been due to both changes in the eicosanoid composition and other mechanisms independent of eicosanoid synthesis (The British Nutrition Foundation 1992, Kelley 1992).

The high level of palmitoleic acid in PO significantly increased the proportion of fatty acid in plasma lipids without a clear effect on the levels of polyunsaturated fatty acids or improvement of AD symptoms (Yang et al. 1999a).

The oil supplementation's did not lead to any significant changes in the levels of the major fatty acids in skin glycerophospholipids, indicating a more stable fatty acid composition of glycerophospholipids in skin compared to plasma.

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Table 4. Fatty acid compositions of skin glycerophospholipids in the patients before and after treatment in the three groups (as weight percentage).
A, before treatment; C, after treatment.

Fatty Acids	seed oil group						pulp oil group						placebo group					
	n	A		C		p	n	A		C		p	n	A		C		p
		mean	stdv.	mean	stdv.			mean	stdv.	mean	stdv.			mean	stdv.	mean	stdv.	
14:0	5	0.48	0.18	0,50	0.25	0.67	7	0.59	0.22	0.46	0.16	0.16	4	0.45	0.29	0.77	0.77	0.29
16:0	5	16.24	2.05	14.31	2.32	0,09*	7	14.15	1.87	13,70	1.18	0.11	4	14.74	4.18	15.77	1.37	0.54
16:1 (n-9)	5	2.84	2.27	2.15	1.28	0.43	7	1.85	0.99	1.16	0.51	0.15	4	1.63	1.06	1.52	1.09	0.57
16:1 (n-7)	5	0.59	0.27	0.58	0.23	0.95	7	0.71	0.29	0.66	0.37	0.46	4	0.68	0.14	0.75	0.08	0.15
18:0	5	16.55	2.28	17.58	1.34	0.31	7	16.78	1.21	18.09	2.07	0,07*	4	17.8	2.77	18,50	3.65	0,09*
18:1 (n-9)	5	19.18	1.91	18.67	1.38	0.67	7	18.85	2.25	17.62	1.87	0,10	4	17.18	1.03	16.37	1.24	0.39
18:1 (n-7)	5	1.95	0.17	1.92	0.2	0.78	7	2.15	0.38	2.16	0.42	0.93	4	2.02	0.14	2.15	0.09	0.27
18:2 (n-6)	5	19.87	2.64	20.29	4.87	0.85	7	21.3	3,00	21.01	2.13	0.95	4	19,80	1.24	21.43	1.66	0,05*
20:0	5	0.62	0.22	0,80	0.16	0.18	7	0.62	0.12	0.66	0.13	0.18	4	0.65	0.14	0.58	0.12	0,50
20:1 (n-9)	4	0.35	0.09	0.31	0.09	0.22	5	0.31	0.16	0.28	0.05	0.61	3	0.35	0.07	0.38	0,10	0.77
20:3 (n-6)	5	0.39	0.17	0.29	0.13	0.54	7	2.12	0.35	2.29	0.34	0.38	4	2,30	0.78	2.14	0.51	0.37
20:4 (n-6)	5	12,50	1.33	12.37	2.24	0.83	7	14,10	1,90	14.73	1,50	0.41	3	14.28	1.83	12.78	0.94	0.19
20:5 (n-3)	4	0.35	0.13	0.38	0.14	0.74	5	0.34	0.16	0.33	0.15	0.86	3	0.24	0.08	0.19	0.05	0.36
22:1 (n-9)	5	0.29	0.11	0.35	0.09	0.41	7	0.27	0.06	0.36	0.18	0.27	3	0.41	0.09	0.32	0.06	0.41
22:5 (n-3)	5	1.11	0.16	1.36	0.15	0,07*	7	1.39	0.43	1.56	0.39	0.34	4	1.62	0.32	1.12	0.23	0.16
22:6 (n-3)	5	3.21	0.68	1.09	0.49	0.69	7	3.09	0.54	3.33	1.48	0.68	4	3.78	0,80	2.83	0,70	0,10

* $0.05 < p < 0.1$