# Essential fatty acids in the nutrition of severely neurologically disabled children

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Essential fatty acids (EFA) are important for the normal development and functioning of the brain, retina and immune competent cells. Severely neurologically handicapped children often have feeding difficulties, and the composition of the diet may be critical with respect to an optimal nutrient content. The aim of the present investigation was to evaluate if the dietary intakes and serum phospholipid concentrations of EFA were adequate in a group of severely neurologically impaired children in an institution. To achieve this, a prospective study was done. The investigation showed low dietary intakes of both *n*-6 fatty acids (FA) and *n*-3 FA. The serum concentrations of total *n*-6 FA, linoleic acid and 22:6*n*-3 (docosahexaenoic acid) as proportions of the total serum phospholipid FA concentration were initially low. The serum concentrations of 20:3*n*-9 and 22:5*n*-6 cholesterol, triacylglycerol, total saturated FA, total monounsaturated FA and apolipoproteins A-I and B were high compared with levels in a reference group of healthy children. Following supplementation with fish oil and soyabean oil, the serum lipid profile approached normal. We conclude that the study children had suboptimal intakes of EFA and that elevated serum concentrations of 20:3*n*-9 and 22:5*n*6 were useful serological markers of suboptimal EFA status. Recommended dietary allowances for EFA given as a percentage of energy underestimate EFA requirements in children with a low energy intake. Severely disabled children with feeding difficulties should probably be monitored with serum phospholipid FA measurements or calculation of dietary absolute intakes of EFA.

**Essential fatty acids: Disabled children: Phospholipids**

The nutritional importance of polyunsaturated fatty acids (PUFA) has been increasingly acknowledged during the past decades (Decsi & Koletzko, 1994). Essential fatty acids (EFA) have been shown to be important for the normal development and functioning of the brain and retina. Deficiency may give reduced visual acuity and electroretinographical response as well as disorders of cognitive behaviour and reduced neurodevelopmental quotient (Bjerve *et al.* 1993; Carlson *et al.* 1993; Carlson & Wilson, 1994; Uauy-Dagach *et al.* 1994; Agostini *et al.* 1995; Makrides *et al.* 1995; Willatts *et al.* 1998). PUFA are also important for the normal functioning of different immune competent cells (Robinson, 1987; Bjerve *et al.* 1989; Endres *et al.* 1989). The role of PUFA in attention-deficit–hyperactive disorders has also been emphasized (Aman *et al.* 1987; Mitchell *et al.* 1987; Stevens *et al.* 1995). Severely neurologically impaired children are at high risk for under- and malnutrition and represent a risk population with respect to intakes of certain essential nutrients (Hals *et al.* 1996).

The aim of the present investigation was to evaluate the adequacy of the diet, with respect to the content of EFA, and measure the serum phospholipid fatty acid (FA) concentrations in a group of children with severe neurological impairment living in an institution. Further, having established that the children had suboptimal intakes and serum concentrations of several EFA, we placed them on an improved diet (containing fish oil and soyabean oil) for 9 months before repeating the serum phospholipid FA measurements.

## Materials and methods

### *Patients*

Thirteen severely neurologically disabled children, nursed

**Abbreviations:** Apo, apolipoprotein; DHA, docosahexaenoic acid; E%, percentage of total dietary energy; EFA, essential fatty acids; EPA, eicosapentaenoic acid; FA, fatty acid; PUFA, polyunsaturated fatty acids.

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in an institution, were studied. The first and general part of the study has been published previously by Hals *et al.* (1996). Table 1 shows distribution of sex, age, diagnoses and antiepileptic drugs for the subjects. All children were profoundly mentally retarded, with no verbal communication, no voluntary movements and with severe oral motor problems, requiring additional or total tube feeding. Most of the children had occasional vomiting, two had gastro-oesophageal reflux and had undergone an operation to prevent this (funduplication a.m. Nissen and gastrostomy) before the study period. None of the children had food intolerance, heart, kidney or liver disease or frequent infections. The amount of vomiting, the medications, and the other factors mentioned were constant during the study period. Three of the children were transferred to other institutions some time after the initial assessment and were not subject to the complete intervention. Consequently, thirteen children constitute the original (intention to treat) group initially compared with the reference group. The three children who did not undergo the complete intervention were excluded from the post-interventional comparison, and the ten remaining children were compared, after the intervention, with the reference group and with themselves before intervention.

As a reference group for the serum phospholipid FA and serum lipids we used twenty-one healthy children admitted as out-patients for elective surgery. Sex and mean age of the reference group did not differ from those of the patients, and the age distributions were comparable in the two groups ($P = 0{\cdot}977$).

### *Dietary assessment*

The dietary intake of each child was assessed at the start of the study by a 4 d weighed record of food intake (three week days and one weekend day). The diet consisted of fresh raw food materials based on traditional dietary recommendations for Norwegian children, mixed together to be given by spoon or by tube if the child was unable to eat the whole portion. Computerized calculation of the energy and fat contents of each child's diet was based on data from Nordic food tables (Blaker & Rimestad, 1991; Statens livsmedelsverk, 1993). One child was sick during recording, leaving twelve children for the initial dietary assessment. The children were given a daily vitamin supplement, Biovit® (Nycomed, Oslo, Norway) 5 ml, yielding retinyl palmitate 380 μg, thiamin 1 μg, riboflavin 1·4 mg, niacin 10 mg, ascorbic acid 25 mg, cholecalciferol 5 μg, α-tocopherol 5 mg.

### *Measurements*

Serum total phospholipid FA were quantified as described previously (Bjerve *et al.* 1987) at the Department of Clinical Chemistry, Trondheim University Hospital, Norway. Lipids were extracted with *n*-butanol. Phospholipids were then isolated, transmethylated and quantified by GLC after adding diheptadecanoylglycerophosphorylcholine and butylated hydroxytoluene as internal standard and antioxidant respectively. Analytical performance was monitored by analysing a human control serum. The day-to-day precision values expressed as the CV (*n* 55) for determination of the

**Table 1.** Sex, age and weight of the neurologically disabled children used in the present study, together with their disease diagnoses and the antiepileptic drugs prescribed

| Patient | Sex | Age (years)* | Weight (kg)* | Diagnoses and antiepileptic drugs |
|---|---|---|---|---|
| 1 | M | 5·8 | 17·9 | Encephalitis, CP, spastic tetraplegia, epilepsia; phenobarbital, Na-valproate, benzodiazepine |
| 2 | M | 4·3 | 13·8 | Severe anoxic brain damage, intracerebral haemorrhage, CP, spastic tetraplegia, epilepsia; Na-valproate |
| 3 | F | 13·4 | 18·1 | Birth asphyxia, ALTE, Varicella-encephalitis, infantile spasms/epilepsia, CP, spastic tetraplegia; carbamazepine, Na-valproate, benzodiazepine |
| 4 | M | 2·8 | 10·1 | Prematurity, anoxic brain damage, CP, spastic tetraplegia and dystonia, epilepsia; Na-valproate, benzodiazepine, lamotrigine (from September 1993) |
| 5 | M | 6·3 | 16·2 | Birth asphyxia, intracerebral haemorrhage, CP, spastic tetraplegia, epilepsia; Na-valproate, benzodiazepine |
| 6 | M | 7·5 | 16·9 | Mental retardation of unknown origin. Syndrome with microcephali, cryptorchidism, agenesia of the spleen, myelinization disorder, epilepsia; no antiepileptic drugs |
| 7 | M | 5·3 | 14·5 | Birth asphyxia, CP, spastic tetraplegia, epilepsia; phenobarbital |
| 8 | F | 2·3 | 11·2 | Herpes simplex encephalitis, microcephali, CP, spastic tetraplegia, infantile spasms, epilepsia; carbamazepine, Na-valproate |
| 9 | F | 4·3 | 15·0 | Prenatal intracerebral haemorrhage, CP, spastic tetraplegia, epilepsia; benzodiazepine, Na-valproate (from September 1993) |
| 10 | F | 2·8 | 12·5 | Birth asphyxia, brain infarction, hydrocephalus operated, CP, spastic tetraplegia, epilepsia, GER operated (Funduplication a.m. Nissen/gastrostomia); carbamazepine |
| 11 | M | 5·4 | 10·0 | Prematurity, severe GBS-infection with anoxic brain damage, CP, spastic tetraplegia, epilepsia, GER operated; phenobarbital, benzodiazepine |
| 12 | M | 4·5 | 11·4 | Prematurity, extreme SGA, microcephali, brain infarctions, CP, spastic tetraplegia, infantile spasms, epilepsia; Na-valproate, benzodiazepine |
| 13 | M | 8·5 | 15·6 | Congenital hydranencephalia operated, CP, spastic tetraplegia, epilepsia; phenobarbital |

M, male; F, female; CP, cerebral paresis; ALTE, apparent life-threatening event; GER, gastroesophageal reflux; GBS, group B streptococci; SGA, small for gestational age.
* In November 1992.

FA 18:0, 18:1*n*-9, 18:2*n*-6, 20:3*n*-6, 20:4*n*-6, 20:5*n*-3 and 22:6*n*-3 were 3·3, 4·9, 3·1, 4·0, 3·8, 4·3 and 6·6 % respectively. The following FA were measured: 14:0, 16:0, 18:0, 20:0, 22:0, 24:0, 16:1, 18:1, 20:1, 22:1, 24:1, 20:3*n*-9, 18:2*n*-6, 20:2*n*-6, 20:3*n*-6, 20:4*n*-6, 22:4*n*-6, 22:5*n*-6, 18:3*n*-3, 20:5*n*-3, 22:5*n*-3 and 22:6*n*-3. Results are given as a relative concentration (g/100 g total phospholipid fatty acids). The samples were collected in November 1992 and January 1994. The venous blood was separated within 1 h and the serum was stored for 4 weeks in a freezer at −70° before being analysed.

Serum total cholesterol, triacylglycerol, and apolipoproteins (Apo) A-I and B were measured by routine methods previously described (Bønaa *et al*. 1992*a*). The blood samples from the reference group were treated in the same way as those from the patients except that they were collected during a time period of 4 weeks. The sera were immediately stored in a freezer at −70° and then analysed together with the patient samples in the same laboratory at the same time.

Measurements of haemoglobin, packed cell volume, cell indices, serum concentrations of Fe, ferritin, albumin, cobalamin, folic acid, α-tocopherol, retinol, 25-hydroxycholecalciferol, Zn, Se, Cu and erythrocyte folic acid were made together with anthropometric measurements, and have been presented in a separate publication (Hals *et al*. 1996).

### *Intervention*

Based on the preliminary results from the initial dietary assessment and the biochemical measurements in November 1992, we changed the diet in April 1993, making the new assessment in January 1994 (Hals *et al*. 1996). The amount of whole milk was reduced, and more fish, meat and wholemeal flour and its products were used in the general composition of the diet for the ten children who were still living in the institution. Time and number of meals were not changed. A supplement containing 7 g carbohydrate energy formula (Semper Energy®, Semper Nutrition, Stockholm, Sweden), 5 g fish oil (Peter Møller, Oslo, Norway; corresponding to 0·44 g eicosapentaenoic acid (EPA) and 0·52 g docosahexaenoic acid (DHA)) and 40 g soyabean oil, yielding a total of 1890 kJ (450 kcal) energy, was given to each child daily. The intake of FA is expressed as a percentage of total energy intake (E%) and compared with the Nordic recommendations (Sandstrøm *et al*. 1996).

### *Statistical analyses*

The unpaired two-tailed *t* test was used to compare the original group of patients (*n* 13) before intervention with the reference group, and the intervention group (*n* 10) after the intervention with the reference group. The paired two-tailed *t* test was used to compare the intervention group (*n* 10) before and after intervention. Differences were considered statistically significant for probability levels of $P < 0{\cdot}05$. Statistical calculations were made by the authors using the computer package Statview TM SE+ Graphics for Macintosh (1988; Abacus Concepts Inc., Berkeley, CA, USA) and their validity was confirmed by Parexcel Medstat AS, Lillestrøm, Norway.

## Results

The initial intakes (g) of linoleic acid (18:2*n*-6), arachidonic acid (20:4*n*-6), α-linolenic acid (18:3*n*-3), EPA (20:5*n*-3) and DHA (22:6*n*-3) are presented in Table 2, together with the intakes of total fat, *n*-3 FA, *n*-6 FA and EFA (total *n*-6 + *n*-3 FA) expressed as E%. Mean total energy intake before intervention was 60 % of the average energy allowance (National Research Council, 1989) for the group (Hals *et al*. 1996).

The concentrations of serum phospholipid FA (g/100 g total serum phospholipid FA) and the serum lipid and lipoprotein concentrations before and after intervention are shown in Table 3. Compared with the reference group, patients showed initially low concentrations of DHA, linoleic acid and total *n*-6 FA. Serum concentrations of arachidonic acid, EPA, α-linolenic acid and total *n*-3 FA were not different from those of the reference group. The concentrations of 20:3*n*-9 and 22:5*n*-6 were initially higher than in the reference group.

After the intervention the serum concentrations of DHA, EPA and total *n*-3 FA were higher than in the reference group and had increased during the intervention for the ten patients. α-Linolenic acid levels were now lower than in the reference group, but with no significant decrease for the ten patients. Linoleic acid and total *n*-6 FA concentrations were still lower than in the reference group. Mean arachidonic acid concentration did not change and was still not different from that of the reference group. The desaturation products 20:3*n*-9 and 22:5*n*-6 decreased to normal concentrations. The serum phospholipid total *n*-6 FA : total *n*-3 FA ratio was 4 : 1 before and 2·5 : 1 after the intervention.

Compared with the reference group, values for total saturated FA and total monounsaturated FA in the patient group were higher before intervention and decreased to concentrations comparable with those of the reference group after intervention. Compared with the reference group, the values for serum cholesterol, triacylglycerols, ApoA-I and ApoB were higher in the patient group before intervention. During intervention, ApoA-I did not change, while triacylglycerol levels decreased to values comparable with those of the reference group. ApoB and cholesterol concentrations in the patient group after intervention did not differ from those of the reference group or from those for the ten children before intervention.

## Discussion

### *Materials and methods*

Initially in the present study we had thirteen children to evaluate. This was the ‘intention to treat’ group, which is compared with the reference group before intervention. Since three children were subjected only to parts of the intervention, they were excluded from the final comparison after the intervention, leaving ten children who were compared with the reference group after the intervention. They were also compared with themselves before and after the intervention as shown in Table 3. Although the study population was small, the observed changes in EFA were highly statistically significant. The food recordings were made only at the start of the study and not after the dietary

**Table 2.** Initial dietary intakes of energy, fat and selected fatty acids in thirteen children with severe neurological disability, together with values for the supplement given after the initial assessment*

(Individual values together with means, standard deviations and ranges for the whole group)

| Patients | Energy (kJ) | Fat (% energy) | 18:2*n*-6 (g) | 18:3*n*-3 (g) | 20:4*n*-6 (g) | 20:5*n*-3 (g) | 22:6*n*-3 (g) | *n*-6 + *n*-3 (% energy) | *n*-6 (% energy) | *n*-3 (% energy) |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 4131 | 31·6 | 2·22 | 0·39 | 0·02 | 0·01 | 0·01 | 2·42 | 2·05 | 0·38 |
| 2 | 4757 | 35·8 | 3·21 | 0·51 | 0·02 | 0·01 | 0·02 | 3·00 | 2·57 | 0·43 |
| 3 | 4109 | 23·5 | 1·89 | 0·32 | 0·01 | 0·01 | 0·01 | 2·06 | 1·75 | 0·31 |
| 4 | 3869 | 36·4 | 3·26 | 0·52 | 0·02 | 0·01 | 0·01 | 3·73 | 3·20 | 0·53 |
| 5 | 4067 | 28·1 | 1·73 | 0·30 | 0·02 | 0·01 | 0·01 | 1·92 | 1·63 | 0·30 |
| 6 | 5399 | 32·7 | 3·61 | 0·56 | 0·01 | 0·01 | 0·02 | 2·95 | 2·53 | 0·41 |
| 7 | 4557 | 34·7 | 3·21 | 0·51 | 0·02 | 0·01 | 0·02 | 3·13 | 2·68 | 0·45 |
| 8 | 3181 | 29·9 | 1·34 | 0·21 | 0·01 | 0·01 | 0·01 | 1·88 | 1·60 | 0·27 |
| 9 | 4019 | 29·2 | 1·79 | 0·30 | 0·01 | 0·01 | 0·01 | 1·99 | 1·69 | 0·30 |
| 10 | 2574 | 30·6 | 1·32 | 0·21 | 0·01 | 0·01 | 0·01 | 2·29 | 1·95 | 0·34 |
| 11 | 4596 | 45·0 | 2·18 | 0·37 | 0·01 | 0·01 | 0·01 | 2·12 | 1·80 | 0·32 |
| 13 | 3919 | 43·7 | 2·22 | 0·39 | 0·01 | 0·01 | 0·01 | 2·55 | 2·15 | 0·40 |
| Mean | 4098 | 33·4 | 2·33 | 0·38 | 0·014 | 0·01 | 0·013 | 2·50 | 2·13 | 0·37 |
| SD | 729 | 6·2 | 0·79 | 0·12 | 0·005 | | 0·005 | 0·58 | 0·51 | 0·08 |
| Range | 2474–5399 | 23·5–45·0 | 1·32–3·61 | 0·21–0·56 | 0·01–0·02 | 0·01–0·01 | 0·01–0·02 | 1·88–3·73 | 1·6–3·2 | 0·27–0·53 |
| Recommended intake† | | | | | | | | | 3·00 | 0·50 |
| Supplement | 1890 | | 20·46 | 2·97 | 0 | 0·44 | 0·52 | | | |

* For details of subjects and procedures, see Table 1 and pp. 219–221.

† The recommended energy intakes for children vary according to age as follows: 1–3 years, 5460 kJ/d; 4–6 years 7560 kJ/d; 7–10 years, 8400 kJ/d; 11–14 years 10 500 kJ/d.

**Table 3.** Concentrations of serum phospholipid fatty acids (FA; g/100 g total FA), cholesterol, triacylglycerols and apolipoproteins in children with severe neurological disability before and after dietary supplementation with soyabean oil and fish oil, and in a reference group of healthy children‡
(Mean values, standard deviations and ranges)

| | Patients before intervention (*n* 13) | | | Patients after intervention (*n* 10) | | | Reference group (*n* 21) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Mean | SD | Range | Mean | SD | Range | Mean | SD | Range |
| Phospholipid FA (g/100 g) | | | | | | | | | |
| Total *n*-3 FA | 7·5 | 1·0 | 5·6–9·1 | 12·6*††† | 3·7 | 6·5–19·8 | 9·0 | 3·1 | 5·0–16·8 |
| $\alpha$-Linolenic (18:3*n*-3) | 0·1 | 0·1 | 0–0·4 | 0·1*** | 0·1 | 0–0·2 | 0·2 | 0·1 | 0·1–0·4 |
| DHA (22:6*n*-3) | 4·4** | 0·8 | 2·9–5·9 | 7·8**††† | 2·6 | 3·5–11·6 | 6·2 | 2·1 | 3·2–10·5 |
| EPA (20:5*n*-3) | 1·3 | 0·4 | 0·4–1·7 | 2·7***†† | 1·3 | 1·0–6·0 | 1·3 | 1·0 | 0·5–4·3 |
| Total *n*-6 FA | 31·5*** | 2·6 | 28·6–38·1 | 30·6*** | 3·7 | 25·1–36·3 | 36·0 | 3·3 | 29·6–40·8 |
| Linoleic (18:2*n*-6) | 15·4*** | 2·2 | 12·4–19·0 | 15·6*** | 2·8 | 11·8–19·9 | 23·1 | 3·6 | 17·3–29·6 |
| Arachidonic (20:4*n*-6) | 9·8 | 1·6 | 7·1–13·5 | 9·2 | 1·3 | 7·6–11·1 | 8·7 | 1·4 | 6·5–11·8 |
| 20:3*n*-9 | 0·8*** | 0·3 | 0·2–1·3 | 0·2††† | 0·2 | 0–0·4 | 0·2 | 0·2 | 0–0·7 |
| 22:5*n*-6 | 0·6*** | 0·2 | 0·3–0·8 | 0·2††† | 0·2 | 0–0·4 | 0·2 | 0·1 | 0–0·4 |
| Total saturated FA | 44·0*** | 0·8 | 42·4–45·2 | 43·0*††† | 0·92 | 41·4–43·8 | 42·1 | 0·57 | 41·3–43·0 |
| Total monounsaturated FA | 16·5*** | 2·0 | 12·5–20·3 | 13·7††† | 1·2 | 11·7–15·4 | 12·6 | 1·6 | 10·5–16·1 |
| Cholesterol (mmol/l) | 5·7** | 1·2 | 3·5–8·6 | 5·1 | 0·9 | 3·9–6·9 | 4·7 | 0·7 | 3·4–5·9 |
| Triacylglycerol (mmol/l) | 1·3** | 0·4 | 0·8–1·9 | 1·0† | 0·6 | 0·5–2·2 | 0·9 | 0·3 | 0·4–1·5 |
| Apolipoprotein A-I (g/l) | 1·6*** | 0·3 | 1·0–2·1 | 1·43* | 0·27 | 1·0–2·07 | 1·21 | 0·18 | 0·73–1·5 |
| Apolipoprotein B (g/l) | 1·3* | 0·3 | 0·8–1·9 | 1·00 | 0·22 | 0·6–1·44 | 1·04 | 0·21 | 0·63–1·4 |

DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid.
Mean values were significantly different from those for the reference group: * $P<0·05$, ** $P<0·01$, *** $P<0·001$.
Mean values were significantly different from those for the same ten patients before intervention: † $P<0·05$, †† $P<0·01$, ††† $P<0·001$.
‡ For details of subjects and procedures, see Table 1 and pp. 219–221.

changes, and no registration of the reference children's dietary intakes was made. Still we argue that this would have little impact on our results because we initially related the dietary intake to serum analyses of phospholipid FA, establishing a relationship between low intakes and low serum concentrations of certain EFA. Serum phospholipid FA measurements have been shown to reflect the dietary intake of FA in several publications (Bjerve *et al*. 1993; Svensson *et al*. 1993; Andersen *et al*. 1996), and were measured before and after intervention to reflect the changes made during the intervention.

The patients ranged in age from 2 to 13 years, but the reference group had a statistically similar mean age ($P=0·98$), range and distribution. The patient group was heterogeneous with respect to the cause of the neurological impairment and medications, but there was no change during the study, and the patients were comparable with respect to the degree of neurological impairment, feeding difficulties, diet and environment.

## *Results*

We found that the patients had intakes of *n*-6 EFA (2·13 E%), *n*-3 EFA (0·37 E%) and total EFA (2·50 E%) that were lower than recommended (Sandstrøm *et al*. 1996). The fact that the study children initially had an intake of energy below the recommended dietary allowance (Hals *et al*. 1996), suggests that the absolute intake of EFA was even lower than reflected in the intake expressed as E%. Crawford *et al*. (1989) emphasized the importance of this in their study of pregnant women of low socio-economic status who appeared to have a low energy intakes. According to the Nordic recommendations (Sandstrøm *et al*. 1996), a child's intake of *n*-6 FA should be at least 3 E%, and the intake of *n*-3 FA at least 0·5 E%, but the ideal and safe intakes are debated and are probably higher for selected groups (Bjerve *et al*. 1988; Sandstrøm *et al*. 1996), and even higher when the energy intake is low.

We observed that the mean concentration of DHA (g/100 g total phospholipid FA) in the reference group of healthy children was on the lower limit of the Norwegian adult reference scale. This might indicate that the DHA intake of healthy children is lower than that of adults. The serum phospholipid concentrations of DHA, total *n*-6 FA and linoleic acid were initially low in the patients compared with the reference group. For the intervention group (*n* 10) the serum concentrations of DHA, EPA and total *n*-3 FA increased, while levels of $\alpha$-linolenic acid, linoleic acid, arachidonic acid and total *n*-6 FA were statistically unchanged. This was surprising, since the intervention included *n*-6 FA in amounts that would usually be sufficient. To be sure that there had been no technical error in the analysis of *n*-6 FA, we reanalysed all the samples in February 1999 and found values identical with the original analyses. We also double-checked the compliance of the staff in the institution, and found no reason to believe that this was poor. We cannot offer any good explanation for this result, only speculate that there might be several factors that have an influence on the serum concentrations that we measured. The children gained weight at a high rate in the initial phase of the intervention, but by the time the second measurements were done they had reached a stable state in weight gain rate (Hals *et al*. 1996). We have no indication that the preparation of the food during the intervention differed in any way that could have produced *trans*-FA which might have influenced the results (Koletzko, 1997). Severely neurologically impaired children with markedly low physical activity levels might have a different energy metabolism from that of normal children (Berg, 1973), but we do not know if this would influence the metabolism of

EFA, and thus contribute to the explanation of the lack of change in the *n*-6 FA after the intervention.

The serum phospholipid concentrations of the desaturation products 22:5*n*-6 and 20:3*n*-9 were initially very high in the patient group compared with the reference group and with the adult reference range. It has been shown in animals that the serum phospholipid concentrations of these FA may increase when there is depletion of essential *n*-3 FA (Hagve & Christophersen, 1984; Neuringer *et al*. 1986; Bjerve *et al*. 1988), and Crawford *et al*. (1989, 1990) point to the Mead acid (Mead & Slaton, 1955) as a 'sign of thirst for EFA'. They suggest that the presence of this FA has potential as a diagnostic tool in EFA deficiency. In the study children, levels of these desaturation products normalized when the children were given a supplement of EFA, strongly suggesting that the children initially had an inadequate intake of EFA, even if their intake, expressed as E% was just below the recommended amount. This indicates that the recommended dietary allowance for EFA in children based on E% underestimates the actual need for EFA when the energy intake is low. Since the children in the reference group had normal concentrations of 22:5*n*-6 and 20:3*n*-9 compared with the adult reference values, there was no indication that their serum concentration of DHA, which was low compared with that of Norwegian adults, represented any stress on their EFA metabolism. Whether this means that children's relative intake of DHA or other *n*-3 FA is lower than that of adults, however, remains to be determined.

Several indices have been proposed as markers for EFA deficiency. Holman (1978) suggested the ratio 20:3*n*-9/20:4*n*-6 using a value of greater than 0·2 for the diagnosis of overt *n*-6 FA deficiency. Rivers & Frankel (1981) stated that the ratio 20:4*n*-6/18:2*n*-6 is increased in EFA deficiency. However, irrespective of these ratios, which reflect massive *n*-6 FA deficiency, they concluded that the mere presence of 20:3*n*-9 must be regarded as evidence of impaired nutritional status. This was the case in our study children before intervention, who had markedly elevated serum concentrations of 20:3*n*-9 and 22:5*n*-6, while the 20:3*n*-9/20:4*n*-6 value was 0·08. The value decreased to 0·02 after the intervention, the same value as for the reference group. The value for 20:4*n*-6/18:2*n*-6 was 0·66 both before and after the intervention, reflecting the lack of change in *n*-6 FA as discussed earlier. Arachidonic acid levels were not low either before or after intervention, but this would not be the case unless there was an extreme *n*-6 deficiency. In summary, the children probably did not have massive EFA deficiency, but they had suboptimal intakes and serum concentrations of certain EFA, and the elevated levels of 20:3*n*-9 and 22:5*n*-6 were useful markers to confirm this.

The change in the *n*-6 FA : *n*-3 FA ratio in g/100 g total phospholipid FA from 4 : 1 before to 2·5 : 1 after intervention might indicate a better balance between *n*-6 and *n*-3 FA in the new diet. The ideal ratio in children remains to be determined, but the ratio after intervention corresponds very well to the ratio of 2–3 : 1 found in fetal neuronal tissue and human milk (Svennerholm, 1968; Martinez, 1991).

The serum concentrations of cholesterol, triacylglycerol, ApoB, ApoA-I and total saturated and monounsaturated FA were initially higher in the patients than in the reference group, probably reflecting the high amounts of whole milk in the original diet. After intervention serum concentrations of all except ApoA-I decreased. This could possibly be explained by the reduction in animal fat, but the increased amount of PUFA may also have played a role in reducing triacylglycerol, ApoB and cholesterol concentrations (Harris, 1989; Bønaa *et al*. 1992*b*; Shrapnel *et al*. 1992; Connor *et al*. 1993). In summary, the dietary changes seem to have given the children a serum lipid profile that approached that of the healthy reference group of children.

We observed that the initial $\alpha$-tocopherol level was low in several children (31 %), and was unchanged after the intervention (Hals *et al*. 1996). This is in accordance with other investigators (Decsi & Koletzko, 1995; Koletzko *et al*. 1995) who found no alterations in vitamin E status after introducing PUFA into the diet. The potential effect on the antioxidant status when increasing the amounts of PUFA in the diet remains to be determined.

## Conclusion

The present study shows that our study group of severely disabled children had a low dietary intake of EFA, and that the serum concentrations of several EFA were low compared with those of a reference group of children. The observed initially elevated concentrations of 20:3*n*-9 and 22:5*n*-6 and their return to normal after supplementation, support the observation that the study children had an inadequate intake of EFA before intervention. Our observations further indicate that the recommended dietary allowance for *n*-3 FA should not be given as E% in children with a low energy intake. We believe that severely disabled children with eating difficulties who are at risk of having a low energy intake should be monitored either by calculating their absolute dietary intake of EFA, or by measuring serum phospholipid FA concentrations. Elevated serum concentrations of 20:3*n*-9 and 22:5*n*-6 may be useful indicators of a suboptimal intake of EFA.

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