Introduction

Certain fatty acids are indispensable for human development and health, but cannot be synthesized de novo by humans. Therefore, they need to be consumed with the diet. These fatty acids are collectively known as ‘essential polyunsaturated fatty acids’ (EPUFAs) and comprise the ‘parent’ essential fatty acids (EFAs) and their longer chain, more unsaturated derivatives, the long-chain (LC) polyunsaturated fatty acids (PUFAs). EFAs and LC-PUFAs are important structural and functional membrane components. In addition, some LC-PUFAs are precursors of prostanoids (prostaglandins and thromboxanes) and leukotrienes, local hormone-like substances with important bioregulatory functions [1].

There are two EPUFA families, the n-6 and the n-3. The parent fatty acids of these families are linoleic acid (LA, 18:2n-6) and α-linolenic acid (ALA, 18:3n-3), respectively. These EFAs, which are mainly present in seed oils (LA + ALA) and green leafs (mainly ALA), can be enzymatically desaturated and elongated in the human body to LC-PUFA. Arachidonic acid (AA, 20:4n-6) and docosahexaenoic acid (DHA, 22:6n-3) are considered the most important LC-PUFAs. AA is involved in the regulation of a large variety of metabolic and physiological processes, whereas DHA is the major LC-PUFA in the central nervous system [1, 2].

[^1]: Fatty acids are indicated by the general formula x:y(n – z), in which x indicates the number of C atoms in the molecule, y the number of double bonds, and z the position of the first double bond, counted from the methyl head group.
In humans, the endogenous formation of LC-PUFA from their respective EFA precursors is rather inefficient. Since the two parent EFAs compete for the same desaturation and elongation enzymes, and the habitual Western diet usually contains much more LA than ALA, endogenous DHA formation is particularly low [3]. Therefore, an adequate LC-PUFA status requires the direct consumption of DHA and possibly AA, which are present in fatty fish (mainly DHA), egg yolk (mainly AA), lean meat, and dietary supplements.

If insufficient EPUFAs are available to meet the physiological requirements, the body starts to synthesize certain fatty acids with a comparable molecular structure but lacking the specific essential functions. These ‘surrogate’ fatty acids are hardly present under normal conditions and can, therefore, be used as EPUFA status markers [4]. The best-known marker is Mead acid (20:3n-9), the increased presence of which indicates a general shortage of all EPUFAs. If there is a functional shortage of DHA, the body increases the synthesis of Osbond acid (ObA, 22:5n-6).

Therefore, under steady-state conditions, the ratio between DHA and ObA is a reliable indicator of the functional DHA status.

**Maternal LC-PUFA Status During and After Pregnancy**

Pregnancy is associated with a generalized lipidemia, and it appears that the plasma amounts (mg/l) of the phospholipid (PL)-associated DHA and AA increase by about 100 and 35%, respectively [5]. These pregnancy-associated LC-PUFA changes have been confirmed under highly different dietary and cultural conditions and, therefore, are a rather general phenomenon. They start already very early in pregnancy and cannot be explained by a changing LC-PUFA intake [6]. Therefore, the pregnancy-associated LC-PUFA increase may be caused by LC-PUFA mobilization from maternal stores or by a metabolic LC-PUFA shift from energy production to structural use.

The proportional amounts of the EPUFA status markers increase considerably stronger than those of the EPUFAs. Thus, Mead acid and ObA concentrations increase by 244 and 348%, respectively (calculated from Al et al. [5] and Otto et al. [6]). This indicates that under the present dietary conditions, pregnancy is associated with a reduction in the functional LC-PUFA status. This is also suggested from the significant reduction in plasma PL of the relative concentrations (percent of total PL-associated fatty acids) of AA, DHA, and most other EPUFAs [5, 7].

After delivery, normalization of the essential PUFA status in maternal plasma PL takes place, but this appears to be a relatively slow process, taking more than 6 months [5]. Since human milk contains LC-PUFA, lactating women continue to transfer their own LC-PUFA to their infants, whereas non-lactating women do not. As a result, normalization of the maternal DHA status takes longer for lactating than for non-lactating mothers [8]. Moreover,
the relative DHA concentrations in plasma (fig. 1) and erythrocyte PLs become significantly lower in lactating as compared to non-lactating women, which could not be explained by differences in EPUFA intakes. After weaning of the infants, the maternal DHA values increase rapidly to values comparable to those of non-lactating women [8].

The DHA content in plasma PL of primigravidas is significantly higher than that of expecting women who have been pregnant before [9]. Actually, a significant negative relationship was observed between this DHA content at delivery and parity number. This indicates that certain maternal DHA stores may not be fully replenished after pregnancy, as a result of which DHA mobilization during pregnancy is compromised. Alternatively, DHA synthesis from precursor fatty acids may become diminished as a result of repeated pregnancies [10]. Since a highly significant and positive relationship exists between the LC-PUFA status of the neonate and that of its mother (see below), first-born infants have a significantly higher DHA status than their later-born siblings [9].

The Fetal and Neonatal EPUFA Status

Since EFAs and their LC-PUFAs cannot be synthesized de novo by humans, the fetal EPUFA supply will strongly depend on maternal EFA and LC-PUFA consumption, metabolism, and placental transport. This dependence is
convincingly illustrated by the significant, positive maternal-fetal correlations for most EFAs and their LC-PUFAs [5, 7, 11].

In spite of these strong correlations, the plasma and erythrocyte PL fatty acid profiles of neonates are very different from that of their mothers. In general, relative LC-PUFA values (percent of total PL-associated fatty acids) are considerably higher, whereas the concentrations of the parent EFAs are greatly reduced in neonates as compared to their mothers [5, 7, 11, 12]. When expressed in absolute figures, however (mg/l plasma), all fatty acid amounts are much lower in neonatal than in maternal plasma, which is due to considerably smaller neonatal plasma PL pools.

Preterm infants have a significantly lower EPUFA status than term neonates [13]. However, the EPUFA amounts in cord plasma of preterm infants at birth are not lower than that in cord plasma obtained by fetal blood sampling of ongoing pregnancies at a comparable gestational age [14]. Therefore, the low EPUFA status of preterm infants is most probably a physiological situation and not a pathological condition.

The usually observed declines in the maternal EFA and LC-PUFA statuses occurring during pregnancy [5, 7] may imply a suboptimal EPUFA status of their newborn infants. This view is supported by the observation that the EPUFA status of neonatal (cord) blood vessel walls is lower than that of the walls of adult blood vessels [15]. In addition, newborn singletons have a higher EPUFA status than infants born after multiple pregnancies [16, 17].

The EPUFA status of the walls of the umbilical vein (the supplying fetal blood vessel) is considerably higher than that of the umbilical arteries, which carry the blood away from the fetus back to the placenta (fig. 2). Although certain tissues may be preferred sites of EFA/LC-PUFA uptake, the EPUFA statuses of umbilical venous and arterial walls likely reflect the PUFA status of ‘upstream’ and ‘downstream’ fetal tissue, respectively. Consequently, the typical fatty acid profiles of umbilical veins and arteries indicate that the EFA status of the developing fetus is relatively low, and is lower in ‘downstream’ as compared to ‘upstream’ areas.

**Habitual Fatty Acid Intake and LC-PUFA Status**

Humans are unable to synthesize essential fatty acids de novo, and LC-PUFA synthesis from EFA precursors is inefficient in man. Therefore, the EPUFA status of pregnant women is most likely determined by their intake of EFA and LC-PUFA. Several investigators have now confirmed this suggestion. Thus, Al et al. [18] observed a significant, positive correlation between the dietary intake and the plasma PL contents of LA. Interestingly, a significant, negative relationship was observed between the maternal LA intake and the amounts of the n-3 LC-PUFA 20:5n-3 (eicosapentaenoic acid), 22:5n-3, (docosapentaenoic acid) and DHA in maternal as well as
As discussed before [3], this may be due to an inhibitory effect of LA on the incorporation of n-3 PUFAs in plasma and tissue PL. No significant relationship was found between LA intake and AA concentrations (fig. 2).

In the same 288 pregnant women, a significant positive relationship was observed between the maternal ALA consumption and the ALA amounts in maternal plasma PLs as well as the neonatal eicosapentaenoic acid concentrations [3, 7]. However, a higher maternal ALA consumption was not associated with a higher maternal or neonatal DHA status. This agrees with recent findings that tripling the ALA consumption by pregnant women hardly increases their DHA status [19]. As is the case for non-pregnant subjects, the habitual intake of n-3 LC-PUFAs is reliably reflected by the n-3 LC-PUFA content of plasma and erythrocyte PLs [3].

Industrial hydrogenation of edible oils is a common procedure to improve the technological and organoleptic quality of these oils. However, this process causes the formation of trans isomers of unsaturated fatty acids, which are known to interfere with the conversion of parent EFAs into their derived LC-PUFAs, especially when the parent EFA levels are low [20]. It has now convincingly been demonstrated that the presence of trans fatty acids in neonatal blood and cord tissue is associated with proportionally lower amounts of EPUFAs [21, 22].

Fig. 2. Maternal linoleic acid intake in mid gestation (g/day) is negatively related to neonatal DHA (□, p for trend < 0.01) but not AA (■) availability (percent of fatty acids in cord plasma phospholipids) [7].
Maternal LC-PUFA Status and Pregnancy Complications

**Pregnancy-Induced Hypertension**

From observational studies it has been suggested that a reduced n-3 LC-PUFA status may contribute to pregnancy-induced hypertension (PIH). However, in a prospective nested case-control study, Al et al. [23] observed a slightly higher n-3 LC-PUFA status in women with PIH. Moreover, in a series of prophylactic and therapeutic trials it was demonstrated that supplementation during pregnancy with up to 6.1 g of n-3 LC-PUFAs/day does not lower PIH risk [24]. Therefore, a causal role of LC-PUFAs in the etiology of PIH seems unlikely.

**Postpartum Depression**

Hibbeln [25] observed that a higher seafood consumption is associated with a lower prevalence of postpartum depression. Otto et al. [26] recently demonstrated that the risk of developing depressive symptoms after delivery is lower in women with a quick recovery of the postnatal DHA status than in women with a relatively slow normalization of the DHA status. Moreover, De Vriese et al. [27] observed that women who developed postpartum depression had significantly lower DHA concentrations in their plasma PLs and cholesterol esters shortly after delivery than women who did not develop this condition. In contrast, the daily administration to lactating women of 200 mg DHA for 4 months did not lower their risk of postpartum depression [28]. It should be noted, however, that supplementation was initiated after delivery, whereas depressed mood after childbirth often starts already during pregnancy. Therefore, further studies are required to elucidate the potential role of DHA in the prevention of depression during and after pregnancy.

**LC-PUFA Status and Birth Outcome**

**Preterm Delivery**

Olsen and Secher [29] extensively studied the relationship between the maternal n-3 LC-PUFA intake and preterm delivery. Until recently, their results were inconsistent, but their most recent prospective cohort study among 8,729 pregnant women clearly demonstrated that the length of gestation is positively related to the intake of n-3 LC-PUFAs, and that low fish consumption is a strong risk factor for preterm delivery [29].

**Birth Weight**

Using dietary history data obtained in a group of 372 pregnant women during their 22nd week of pregnancy and after adjustment for potential confounders, Badart-Smook et al. [30] observed that birth weight and ponderal index (birth weight/cube of birth length) were not significantly related
to maternal EPUFA consumption midway in gestation. In a later study with 627 mother-infant pairs, Rump et al. [31] confirmed that birth weight is not closely associated with maternal EPUFA consumption during pregnancy as represented by the EPUFA amounts in maternal plasma PLs. Relationships between maternal fish intake during pregnancy and infant birth weight have been found to be inconsistent, but tend to be positive. Although fish consumption during pregnancy has been reported to be associated with a reduced risk of intrauterine growth retardation [29], fish oil supplementation did not reduce this risk [24].

Birth weight has not been shown to be positively related to fetal n-3 LC-PUFA levels. On the contrary, negative relationships have been reported between birth weight and the concentrations of various n-3 LC-PUFAs in cord plasma and cord serum PLs [31, 32]. In one of these studies (fig. 3), negative associations with birth weight were also observed for AA. The additional significant and positive correlations between birth weight and the umbilical amounts of the EPUFA shortage markers Mead acid and ObA suggest that the maternal-to-fetal LC-PUFA transfer is too limited to secure an adequate, birth weight-independent neonatal LC-PUFA status. Interestingly,

**Fig. 3.** In term neonates, the LC-PUFA status is negatively related to birth weight [31]. **a, b** Significant negative relationship between birth weight and DHA and AA. **c, d** Significant positive relationships between birth weight and LC-PUFA shortage markers Osbond acid (ObA) and Mead acid (MA). All p values ≤0.0001. Birth weight classification: I (n = 81), ≤10th percentile; II (n = 95), >10th to ≤25th; III (n = 339), >25th to <75th; IV (n = 71), ≥75th to 90th; V (n = 41), ≥90th percentile.
birth weight appeared to be positively related to dihomo-γ-linolenic acid (DGLA, 20:3n-6) [31]. This has been reported by others before and warrants further study.

**Head Circumference**

In a group of 110 normal neonates, Al et al. [5] observed that head circumference was significantly and negatively correlated with the LA percentage in umbilical plasma PLs. This finding could imply that neonatal head circumference is negatively influenced by maternal LA intake. Indeed, maternal LA consumption midway through gestation was negatively related with neonatal head circumference [30]. Head circumference is a powerful predictor of brain weight, and AA and DHA are major ‘building bricks’ of the brain. In addition, maternal LA intake during pregnancy is negatively related to the amounts of most neonatal n-3 LC-PUFAs [7] (fig. 2). Therefore, the negative association between LA intake and head circumference could possibly be explained by an overabundant LA availability, resulting in substrate inhibition of the Δ⁶-desaturation reaction required for a proper EFA-to-LC-PUFA conversion [3]. In addition, LA has also been shown to inhibit LC-PUFA incorporation in plasma and tissue PLs [3]. This suggests that the ratio between the amounts of n-3 and n-6 PUFAs in the present diet is too low and needs readjustment, preferably by substituting ALA for LA.

**Early LC-PUFA Availability and Later Neurodevelopment**

Since the brain has its growth spurt during the third trimester of pregnancy and in the neonatal period, it seems feasible to suggest that the fetal and/or neonatal LC-PUFA status could affect early brain growth, maturation, and function. However, no significant associations have been observed between either DHA or AA concentrations in cord blood PL (a proxy for fetal LC-PUFA availability) and cognitive performance at 7 years of age [33]. Likewise, no significant relationship was observed between cognitive performance at 3.5 years of age and LC-PUFA status of neonatal erythrocytes [34]. However, DHA status at birth was significantly and positively related to movement quality and to visual acuity at 7–8 years of age. Speed of visual information processing, measured by visual evoked potentials and electroretinograms at follow-up, were also positively related to DHA levels at birth (manuscripts in preparation).

The perinatal DHA availability was also significantly related to infant behavior at age 7 years: the higher the DHA status at birth, the lower the problem score for internalizing behavior (manuscript in preparation). None of these functional outcome measures were significantly associated with AA concentrations in umbilical plasma or with LC-PUFA levels at follow-up. These results indicate that a higher perinatal DHA availability may promote
certain aspects of later neurodevelopment, brain function and infant behavior. They also suggest that an ample prenatal DHA supply, and consequently and adequate maternal DHA intake during pregnancy, may be of at least equal importance for cognitive, motor, visual, and behavioral development as dietary LC-PUFAs during childhood.

**Fetal Availability of \(\gamma\)-Linolenic Acid and Later Risk of Type-2 Diabetes mellitus (fig. 4)**

According to the ‘fetal origins of adult disease’ hypothesis, unbalanced nutrition during intrauterine development may contribute to a later risk of cardiovascular disease, including disturbed lipoprotein metabolism, type-2 diabetes, insulin resistance and obesity. Studies on the nutritional factors involved, however, are limited. Therefore, we investigated whether the fetal availability of EPUFAs (as represented by their levels in cord plasma PLs)
relates to childhood plasma lipoprotein levels, glycemic control, and body composition. Total cholesterol, HDL cholesterol, LDL cholesterol, apolipoprotein A1, apolipoprotein B, and lipoprotein[a] concentrations, measured at 7 years of age, did not relate to the PL fatty acid composition of umbilical cord plasma at birth. Plasma triacylglycerol concentrations, however, were negatively associated with concentrations of γ-linolenic acid (GLA, 18:3n-6) and DGLA (20:3n-6) in umbilical cord plasma PLs (manuscript in preparation). In addition, cord plasma GLA and DGLA concentrations were negatively related to fasting insulin and pro-insulin concentrations and to insulin resistance (homeostasis model assessment index) at 7 years of age [35]. The GLA concentrations were also negatively related to body fatness as calculated from skin-fold measurements and reflected by plasma leptin concentrations at age 7. No associations were observed for other EPUFA concentrations at birth. These results suggest that a low intrauterine availability of GLA and possibly DGLA could be one of the factors predisposing individuals to obesity and insulin resistance later in life. If the observed relationships turn out to be causal, maternal GLA supplementation during pregnancy to improve the fetal (D)GLA status may present a simple and safe way to lower the risk of newborns for later insulin resistance and obesity.

Maternal EPUFA Supplementation during Pregnancy and Lactation: Biochemical and Functional Effects on Neonates and Breast-Fed Infants

Intervention studies demonstrated that it is feasible to increase the EPUFA status of neonates or breast-fed infants by dietary supplementation of their mothers. Thus, maternal supplementation with LA increased the neonatal n-6 PUFA status, but this was associated with a reduction of the n-3 LC-PUFA status [36]. Supplementation of pregnant women with fish oil [37–39] results in an increase in the neonatal n-3 PUFA status, but this is often associated with a lower n-6 LC-PUFA status [3]. Therefore, it seems that an overall increase in the maternal and, consequently, neonatal LC-PUFA status would require an increased maternal consumption of both n-6 and n-3 fatty acids. For the maternal LC-PUFA status this has been confirmed by a series of studies with single-cell oils rich in DHA or AA [40, 41].

It has also been shown that the DHA status of breast-fed infants was significantly and positively related to the DHA dose their mothers were supplemented with [42]. However, no neurodevelopment differences were observed between the various groups [43], but it should be realized that group sizes of 8–12 might have been too small for a reliable assessment, considering the many potential sources of variability.

As compared to a placebo, maternal fish oil supplementation during pregnancy and lactation resulted in higher mental processing scores (Kaufmann
ABC) after 4 years of follow-up [44], but not before [38]. This strongly indicates that maternal intake of n-3 LC-PUFAs during pregnancy and lactation may be favorable for later mental development of children.

The use of cod liver oil supplements during pregnancy has also been reported to be associated with a lower risk of type 1 (insulin-dependent) diabetes mellitus in the offspring, both unadjusted and after adjustment for age, sex, breastfeeding, maternal education and maternal use of ‘other supplements’ [45].

Implications for Nutrition during Pregnancy

From the results summarized above, it may be felt necessary to increase the dietary EPUFA intake of pregnant women in order to prevent the decrease in their EPUFA status during pregnancy and to optimize that of their newborns. This may be of particular importance for preterm infants, because they have a significantly lower EPUFA status than term neonates [13]. In addition, positive relationships were observed between the amount of DHA in umbilical artery PLs and birth weight, head circumference and birth length of preterm infants and, moreover, the EPUFA status at birth appeared the strongest determinant of the EPUFA status at the expected date of delivery [46]. Therefore, a higher DHA status may be of benefit to preterm neonates, not only for their intrauterine development, but for their postnatal development as well.

As discussed above, the DHA content of maternal plasma PLs is significantly lower in multiparous as compared to primiparous women and infants born to multiparous women have significantly less DHA in umbilical vessel wall PLs than infants born to primiparous women. Whether or not this has functional consequences for these infants is not known as yet. However, there is now good evidence that the pre- and early postnatal DHA status has important consequences for growth and function of the central nervous system and, consequently, for neurologic and cognitive development. Therefore, a lower pre- and perinatal DHA availability may, at least in part, present an explanation for observations that first-born children, in general, do better than their younger siblings on several developmental, behavioral and intelligence tests [47, 48].

A significant, negative relationship has been observed between the amount of trans fatty acids in cord arterial tissue, the neonatal LC-PUFA status, and birth weight [22]. Dietary trans unsaturated fatty acids have also been shown to increase cardiovascular risk as reflected by the plasma lipoprotein profile and cardiovascular risk may already be programmed during early development. Therefore, maternal intake of trans fatty acids should be reduced as much as possible, even if the negative effects of trans fatty acids on fetal development have not been ascertained [49].

Finally, if supplementation with EPUFA during pregnancy is considered, it should be recalled that the two PUFA families compete for the same metabolic
enzymes [3]. Therefore, the supplement of choice should contain a mixture of n-6 and n-3 (LC)PUFAs.

So far, no official recommendations have been made for the LC-PUFA intake of pregnant and lactating women. It is felt that this would require more functional studies [50]. However, since pregnant and lactating mothers are the major source of LC-PUFAs for their infants, and pregnancy and lactation are associated with a reduced (biochemical) LC-PUFA status, it seems prudent for pregnant and lactating women to increase their LC-PUFA intake.

References

EFAs in Mother and Child


27 De Vriese SR, Christophe AB, Maes M: Lowered serum n-3 polyunsaturated fatty acid (PUFA) levels predict the occurrence of postpartum depression: Further evidence that lowered n-PUFAs are related to major depression. Life Sci 2003;73:3181–3187.


EFAs in Mother and Child


Discussion

Dr. Di Renzo: I am a little puzzled about your showing a maternal-fetal, or let’s say neonatal, correlation of docosahexaenoic acid (DHA). How do you explain that in preterms? However this doesn’t seem to be so true for term-babies, considering the fact that it is decreasing. In your preterm babies are there any pathologies that decrease DHA content? Or is it that you can speculate about the preterm correlation to the term correlation?

Dr. Hornstra: This is a very interesting question. What we noted in our studies is that during fetal development there is an increase in the DHA status of the fetus. It starts to be very low but at the end of pregnancy, and especially in the last trimester, the DHA status of a normal fetus increases quite dramatically. To me this indicates that in one way or another the placenta adapts itself to transfer more DHA to the fetus in late pregnancy as compared to early pregnancy. The preterm baby does not benefit from this later improvement in placental function because it is born before this latter period of pregnancy, and that is, we think, the explanation for the difference in DHA status between preterm and term babies. We know this because we did studies in which fetal blood samples were taken during ongoing pregnancies (between 18 and 39 weeks of gestation), and we found that the DHA concentration in the blood increases with the increasing duration of gestation [1]. So there seems to be a learning process by the placenta. The placenta enables a larger transfer of DHA at the end of pregnancy as compared to the beginning of pregnancy. Does that answer your question?
Dr. Di Renzo: Yes partially, because you can probably speculate that the concentration of DHA in the mother is also reflected in the newborn, unless it is at term. This is probably your conclusion because you say that the concentration levels are very similar between neonatal cord blood and maternal blood in term pregnancy, but apparently it is not the case in the preterm babies.

Dr. Hornstra: In the study mentioned above, we also demonstrated that throughout the gestational period studied, a significant maternal-fetal correlation exists for most essential fatty acids and their long-chain polyunsaturated fatty acids (LC-PUFAs), DHA included.

Dr. Di Renzo: I would like to have your opinion about supplementation. There is some trend according to which supplementation should be given with a certain fixed ratio between the n-6 family and n-3 family. Is it fact or fiction for you?

Dr. Hornstra: Theoretically speaking, if you decide that supplementation would do something good, then you have to supplement both n-3 and n-6 because these two families compete with each other, and if you increase the n-3 intake you see a reduction in n-6, and vice versa [2]. So if you decide that DHA supplementation is beneficial, then you should consider to use a combination of DHA and arachidonic acid to prevent a simultaneous reduction in the latter. I have to say immediately that this has still not been studied sufficiently in pregnancy [3].

Dr. Di Renzo: You showed that a good ratio, at least in newborns who actually have good performance at the end, is between 2.5-fold n-6 compared to 1-fold n-3. Is that true, can you maintain supplementation, or am I speculating?

Dr. Hornstra: I am not sure whether I understand what you mean.

Dr. Di Renzo: In showing the PUFA status in the newborn, which you subsequently studied for cognitive performance and so on, you showed that the ratio between the two is around 2.5 against 1. Is it a good ratio or do you think it is just an artifact?

Dr. Hornstra: At birth, the ratio between arachidonic acid and DHA in the plasma phospholipids of our study population was 2.7, whereas the ratio between the total amount of n-6 LC-PUFA and the total amount of n-3 LC-PUFA was 3.4. I don't know whether this is good. It is the average ratio we observed in our 300 infants, but whether this is optimum or not, I simply do not know because we do not have adequate data. But what is interesting is the fact that this ratio is of the same order of magnitude as that in human milk and maybe Mother Nature is teaching us a lesson here that this ratio is alright. On the other hand, I don't think that such ratios are of any functional relevance since we observed positive associations with neuromotor and visual functions for DHA only, and not for arachidonic acid [4].

Dr. Bleker: If you consider supplementation in women, what is the time dependency? How long does it take to reach optimal levels in the plasma?

Dr. Hornstra: I don't know what optimal levels mean because we don't know that, but how long does it take to reach the new steady state? This is how I translate your question. That depends of course on the dose given and the domain you look at. For plasma we know that for supplementation with, let's say, 400 or 500 mg LC-PUFAs/day it requires approximately 2–4 weeks of supplementation before a new steady state is reached. But if you look into the erythrocytes, it takes much longer [5]. For the brain or other tissues it can be expected to last even longer, but I am afraid I cannot answer your question.

Dr. Bleker: What would be the preferable source in food when you think about it worldwide?

Dr. Hornstra: I am a nutritionist, so if it comes to sources of these LC-PUFAs, I think for the n-3 LC-PUFAs we have to refer to fish, whereas meat and eggs are the major sources of arachidonic acid.

Dr. Bleker: Would it be ideal to start months before pregnancy?

Dr. Hornstra: There are no good data to answer that question at this particular moment. But with regard to postpartum depression it was observed that supplementation
with 200 mg DHA/day did not reduce its incidence [6]. Supplementation was started after delivery, which may have been too late since the negative association between maternal DHA status and postpartum depression were seen in observational studies [7–9]. Such studies are related to the habitual situation, so not only during but also before pregnancy. So from that you could infer that it would be better to start as soon as possible and preferably before conception, but this is hypothesis, since the supplementation studies with functional outcomes reported so far were restricted to the 2nd and 3rd trimesters of pregnancy [10–12].

Dr. Cai: In one of your slides you showed that essential fatty acid is lower with breast feeding than with bottle feeding. Have you any long-term follow-up for the prevalence of the breast cancer in later life?

Dr. Hornstra: I am not sure whether I understood your question correctly. Was your question whether we had a long-term follow-up of mothers that have been supplemented?

Dr. Cai: Yes.

Dr. Hornstra: We don’t have those data, I am sorry for that.

Dr. Pencharz: A very provocative talk, but I want to provoke in the levels or rate of appearance and rate of disappearance. As you know I work primarily with amino acids. But because of stable isotopes I have been drawn into fatty acids too. What really struck me is essential fatty acids: there are no mechanisms to protect them against oxidation, whereas for essential amino acids you can downregulate the degradative pathway. So we actually found that the essential fatty acids are highly oxidized, whether it is linoleic acid, α-linolenic acid, DHA, and so on. So what concerns me in your studies is what are the levels? You are primarily dealing with plasma rather than with stores like red cell membranes or anything else like that. What is actually happening? In studies on cystic fibrosis patients by Parsons and Grey, until the energy balance was corrected they were unable to really correct their erythrocyte essential fatty acid status. So I am just trying to see where your women are in terms of their energy balance and rates of formation and rates of removal.

Dr. Hornstra: That is a very important point you are raising. First of all let me say that so far I did not discuss any supplementation studies during pregnancy. What I discussed here were observational studies, normal life so to speak. But we also measured the antioxidant status of mothers and infants as a function of their LC-PUFA status. The interesting thing is that if you correct for the changes in plasma phospholipid unsaturation, then the ‘relative’ tocopherol levels increase by about 15% during pregnancy, whereas the ‘relative’ carotenoid status decreases by about the same percentage [13]. Davidge et al. [14] observed that the functional antioxidant capacity of serum increased during pregnancy. Therefore, I don’t think that the LC-PUFA status during pregnancy is compromised by increased peroxidation.

Dr. Butte: Your results on γ-linolenic acid (GLA) were really fascinating. Was there any relationship between the child at 7 years of age and the GLA, and what was the possible mechanism with insulin resistance and body fatness?

Dr. Hornstra: This issue is very complicated to explain. What we did find were negative associations of plasma insulin, pro-insulin, leptin and triglyceride concentrations at 7 years of age with plasma phospholipid GLA levels at birth. These correlations were not observed with GLA levels at follow-up. There was one exception though and that was the amount of triglycerides positively associated with GLA at follow-up. This makes things very complicated, but it reminds me of the Barker hypothesis [15, 16]. Let’s say that if you have adaptation to a low availability of an essential component and then the component becomes more abundant, then you are in trouble, and this is exactly what is suggested from our studies. But again this is observational and we should try and confirm it by intervention studies. Now what about the mechanism, I really don’t know. I do know though, and maybe Dr. Uauy can expand a little bit on
this, that GLA is a powerful activator of the so-called peroxisomal proliferator-activated receptors (PPARs) which are ligand-activated transcription factors involved in the regulation of these particular metabolic routes [17–19]. If I was going to investigate the potential mechanisms, I would go into the PPARs, I would put my money on that. But I am sorry, I cannot say anything more than that.

Dr. Kramer: I have two related questions. First, how much of the decrease in DHA concentrations with advancing gestational age is due to plasma volume expansion (i.e. dilution) rather than either increased excretion or metabolism of endogenous DHA? Second, are the decreases that you observe during lactation related to the concentration of DHA in breast milk?

Dr. Hornstra: To start with your last question first, we did not do the calculations so I cannot answer that question, but this seems a very likely explanation. With respect to your first question, again I am afraid that things are a little bit more complicated than I presented in my talk, since we had only very limited time. If you read my chapter though, you will see that yes, there is a reduction in DHA status but that does not necessarily mean that there is a reduction in DHA content. Because one of the first things that happens after conception is an increase in the amount of DHA in the blood. An increase, and why do we say that there is a reduction in DHA status? That is because the DHA shortage marker Osbond acid rises even more quickly than the amount of DHA. Please note that ‘more’ does not necessarily mean ‘functionally sufficient’. In order to know whether there is enough or too little of an essential component, you have to have indicators telling you whether there is a functional sufficiency or shortage. The functional shortage marker for DHA is the docosapentaenoic acid of the n-6 series, Osbond acid (22:5n-6), and we see that during pregnancy Osbond shoots up very quickly, indicative of the fact that the body is telling me, ‘yes, OK, DHA is increasing but it should increase more because I need more’. This is an interpretation of the biochemical data, but our long-term follow-up studies indicate that there is some functional consequence of the DHA changes during pregnancy. So the reduction in DHA status during pregnancy is not primarily due to a reduction in the DHA concentration or amount, it is calculated from the ratio between the amounts of DHA in the blood and the shortage indicator for DHA, which is Osbond acid. In fact, it is a functional interpretation of biochemical changes which from our long-term follow-up appears to have functional consequences.

References

EFAs in Mother and Child

9 De Vriese SR, Christophe AB, Maes M: Lowered serum n-3 polyunsaturated fatty acid (PUFA) levels predict the occurrence of postpartum depression: Further evidence that lowered n-PUFAs are related to major depression. Life Sci 2003;73:3181–3187.