Prostaglandins, Leukotrienes and Essential Fatty Acids 86 (2012) 93–98

Contents lists available at SciVerse ScienceDirect

Prostaglandins, Leukotrienes and Essential Fatty Acids

journal homepage: www.elsevier.com/locate/plefa

Low linoleic acid may facilitate Δ6 desaturase activity and docosahexaenoic acid accretion in human fetal development ☆

E.M. Novak, D.J. King, S.M. Innis *

Nutrition and Metabolism Research Program, Child and Family Research Institute, Department of Paediatrics, University of British Columbia, 950 West 28th Ave., Vancouver, B.C., Canada V5Z 4H4

ARTICLE INFO

Article history:
Received 21 December 2011
Received in revised form
9 February 2012
Accepted 10 February 2012

Keywords:
n-3 fatty acids
n-6 fatty acids
Docosahexaenoic acid
Linoleic acid
Pregnancy
Fetal development
Human

ABSTRACT

The *n*-3 and *n*-6 fatty acids are transferred across the placenta with consistently higher 22:6*n*-3 and lower 18:2*n*-6 in fetal than maternal plasma. This study sought to determine whether maternal and fetal cord blood red blood cell (RBC) phospholipid fatty acids show similar saturation with 22:6*n*-3, and also addressed the relationship between 18:2*n*-6 and Δ6 desaturase product/precursor ratios for 97 mothers and newborns. Despite higher fetal than maternal plasma phospholipid 22:6*n*-3, the maternal and fetal RBC phospholipid 22:6*n*-3 showed similar curvilinear relationships to the plasma phospholipid 22:6*n*-3. Risk of failure to achieve high RBC phospholipid 22:6*n*-3 increased sharply below a plasma phospholipid 22:6*n*-3 of 6.5 g/100 g fatty acids. Higher maternal and fetal 18:2*n*-6 was associated with lower RBC phospholipid 22:6*n*-3/22:5*n*-3, 22:5*n*-6/22:4*n*-6 and 18:3*n*-6/18:2*n*-6. These findings suggest low placental transfer of 18:2*n*-6 may be a specific mechanism to prevent inhibition of fetal Δ6 desaturase and facilitate fetal cellular phospholipid 22:6*n*-3 accretion.

1. Introduction

The *n*-3 fatty acids are considered essential dietary fatty acids for humans and other animals due to the absence of enzymatic activities in animal cells leading to creation of a double bond between carbons 12 and 13 from the methyl terminus of a fatty acid carbon chain. The three most well-understood *n*-3 fatty acids are the 18 carbon chain α-linolenic acid (18:3*n*-3), and its desaturation – elongation derivatives, eicosapentaenoic acid (20:5*n*-3) and docosahexaenoic acid (22:6*n*-3). Prior to birth, *n*-3 fatty acids are transferred across the placenta, with levels of 22:6*n*-3 consistently higher in fetal than maternal plasma lipids [1–4]. The higher 22:6*n*-3 in fetal as compared to maternal plasma lipids has been attributed, at least in part, to placental fatty acid binding and transport proteins that facilitate the placental uptake and transfer of 22:6*n*-3 from the mother to the fetus [5]. In contrast to 22:6*n*-3, the *n*-6 linoleic acid (18:2*n*-6) shows an opposite pattern, with lower levels in fetal plasma lipids than in the mother, although arachidonic acid (20:4*n*-6), the desaturation – elongation product of 18:2*n*-6, like 22:6*n*-3 is higher in fetal than maternal plasma lipids [1–4].

Docosahexaenoic acid (22:6*n*-3) is found primarily in membrane phospholipids, with a distinctive tissue, cell, membrane and phospholipid enrichment that includes higher 22:6*n*-3 in ethanolamine than choline containing phospholipids (PE and PC, respectively), and higher 22:6*n*-3 in brain and retina than in other organs. Cells, including red blood cell (RBC) phospholipids are synthesized through several pathways with deacylation–reacylation contributing to the achievement and maintenance of specific membrane phospholipid *n*-3 and *n*-6 fatty acid patterns [6], also constrained by the finite membrane phospholipid acylation sites. The presence of cell and phospholipid specific *n*-3 fatty acid patterns suggest that moving from an *n*-3 fatty acid deficient to adequate diet should result in a curvilinear increase in membrane phospholipid 22:6*n*-3, as has been shown in studies addressing *n*-3 fatty acid requirements [7,8]. Plasma phospholipid fatty acids, on the other hand, are a transport pool reflecting intestinal and hepatic lipoprotein phospholipid synthesis and secretion, and intravascular metabolism.

Docosahexaenoic acid (22:6*n*-3) can be derived from the diet, or by endogenous synthesis from 18:3*n*-3 involving Δ6 desaturation at two steps, an initial desaturation to 18:4*n*-3 and a second Δ6 desaturation enabling synthesis of 22:6*n*-3 from 22:5*n*-3 Fig. 1, [9–11]. The same Δ6 desaturase is involved in desaturation of 18:2*n*-6, both the initial desaturation to 18:3*n*-6 and later for

☆ Supported by a grant from the Canadian Institute of Health Research (CIHR). SMI supported as a senior scientist at the Child and Family Research Institute (CFRI).

* Corresponding author. Tel.: +604 875 2431; fax: +604 875 3597.
E-mail address: sinnis@mail.ubc.ca (S.M. Innis).

0952-3278/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
doi:10.1016/j.plefa.2012.02.004

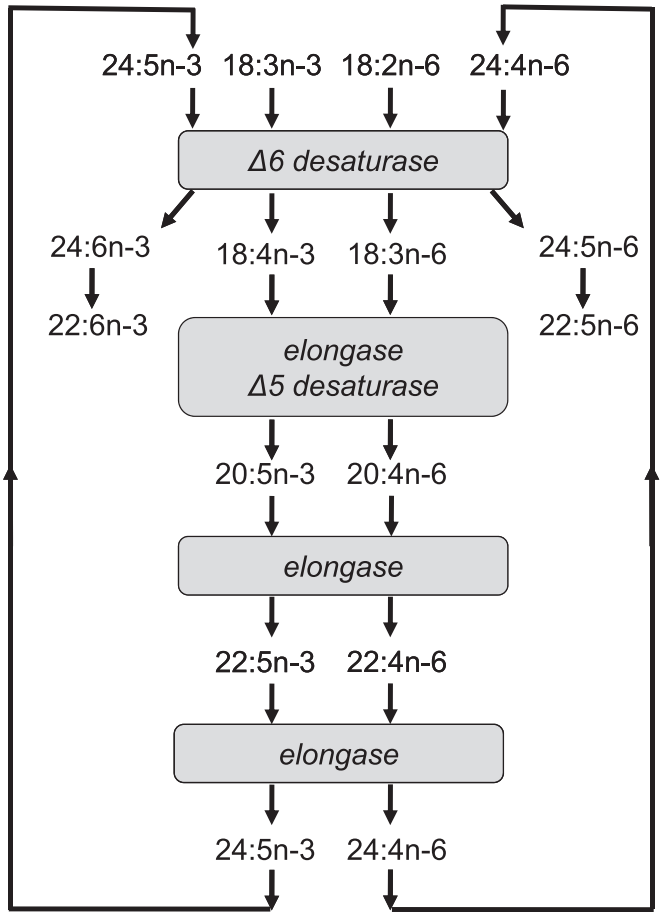

Fig. 1. Schematic illustrating desaturation and elongation of $n-3$ and $n-6$ fatty acids. Adapted from [9,10].

synthesis of 22:5*n*-6 from 22:4*n*-6 [11]. In summary, 22:6*n*-3 accumulated in membrane phospholipids must depend on 22:6*n*-3 availability, either preformed or synthesized, and potential competition by other fatty acids for acyltransferases. Except in severely fat-restricted diets or malabsorption, Δ6 desaturase enzyme activity, not substrate availability, is the most important determinant of fatty acid desaturation. This enzyme appears to require very small amounts of substrate for maximal activity, and perhaps unexpectedly, high amounts of 18:2*n*-6 are associated with decreased Δ6 desaturase gene expression and activity [12], increased 18:2*n*-6 accumulation and decreased tissue 22:6*n*-3 [13–16]. Also important, Δ6 and Δ5 desaturase activity is present in fetal liver, with gene expression about 6-fold higher than in adult liver [17–21]. Thus, we question whether the placenta limits maternal-to-fetal 18:2*n*-6 transfer, facilitating fetal 22:6*n*-3 synthesis and decreasing competition by 18:2*n*-6 for acylation into fetal tissue phospholipids. The present report addresses two questions: first whether maternal and fetal (newborn) RBC phospholipid fatty acids show evidence of similar curvilinear saturation with 22:6*n*-3, and second whether higher maternal and fetal 18:2*n*-6 is associated with inhibition of Δ6 desaturation of *n*-3 and *n*-6 fatty acids as evidenced by product/precursor fatty acid ratios.

2. Subjects and methods

The present study involved 97 mother and infant pairs for whom maternal blood was collected close to term, at 36 week gestation, with collection of cord blood from the infants at delivery. The women and their newborn infants were subjects in a prospective study that involved enrollment at 16 wk gestation with subsequent follow-up of the mothers and their infants [22]. At enrollment, the women were aged 21–40 years with a mean height of 164 cm (range 148–182 cm) and pre-pregnancy weight of 63.6 kg (range 45.5–100 kg), and 52% were expecting their first child. Women expecting to deliver more than one infant, at risk for preterm delivery or any other pregnancy complication, consuming a vegan diet or taking supplemental sources of 22:6*n*-3 were not enrolled. For the 97 women in this study, 49 took no supplemental sources of 22:6*n*-3 during their pregnancy and 48 were given a supplement of 400 mg/day 22:6*n*-3 from 16 week gestation until delivery. The protocol and procedures were approved by the Committee for Ethical Review of Research Involving Human Subjects at the University of British Columbia and the British Columbia Children's and Women's Hospital. All subjects provided written informed consent before participation.

2.1. Biochemical measures of RBC fatty acids

Blood was collected in tubes with EDTA as the anticoagulant, then the RBC were separated from plasma by centrifugation and stored at −70 °C until analyzed [1]. The RBC total lipids were extracted based on Rose and Oklander [23], then PC and PE were separated and the fatty acid components analyzed by gas–liquid chromatography (GLC) as described in detail [1]. PC and PE were specifically chosen as PC represents the major phospholipid on the outer layer of the membrane bilayer and PE represents the major phospholipid on the inner bilayer [24]. Plasma lipids for a subset of 17 mother- infant pairs were separated using HPLC, then the fatty acid composition of the total phospholipids similarly analyzed by GLC.

2.2. Statistical analysis

Data are represented as means ± SEM unless otherwise indicated. Differences between maternal and infant *n*-3 and *n*-6 fatty acids were analyzed using paired t-tests, with Pearson correlation coefficients used to detect significant associations in the levels of *n*-6 and *n*-3 fatty acids between maternal and infant RBC, and between fatty acids in RBC and plasma phospholipids. The analysis were done using SPSS software version 19.0 (SPSS Inc, Chicago, IL) with differences considered significant at $P < 0.05$.

Table 1
Fatty acid composition of maternal and infant red blood cell (RBC) phosphotidylcholine (PC) and phosphatidylethanolamine (PE).

Fatty acid (g/100 g)	RBC PC		RBC PE	
	Maternal	Infant	Maternal	Infant
22:6*n*-3	2.39 ± 0.10	3.72 ± 0.11*	8.28 ± 0.24	7.85 ± 0.19
22:5*n*-3	0.32 ± 0.01	0.30 ± 0.01	2.87 ± 0.07	0.96 ± 0.03*
20:5*n*-3	0.33 ± 0.02	0.31 ± 0.02	0.67 ± 0.03	0.28 ± 0.01*
18:3*n*-3	0.36 ± 0.01	0.05 ± < 0.01*	0.26 ± 0.01	0.07 ± < 0.01*
22:5*n*-6	0.15 ± 0.01	0.41 ± 0.02*	0.68 ± 0.02	1.37 ± 0.04*
22:4*n*-6	0.27 ± 0.01	0.65 ± 0.01*	4.86 ± 0.12	5.80 ± 0.11*
20:4*n*-6	5.14 ± 0.11	12.0 ± 0.19*	15.7 ± 0.23	19.1 ± 0.24*
18:2*n*-6	20.8 ± 0.25	8.72 ± 0.18*	4.68 ± 0 11	1.97 ± 0.04*
16:0	37.5 ± 0.16	37.4 ± 0.18	21.4 ± 0.32	25.0 ± 0.30*
18:0	8.76 ± 0.80	10.2 ± 0.08*	16.8 ± 0.25	17.1 ± 0.30
18:1	18.8 ± 0.16	18.7 ± 0.15	20.5 ± 0.20	15.6 ± 0.18*

$n = 94$ mother and infant pairs for PC, 97 for PE.

* Different from the same fatty acid in maternal RBC, $P < 0.05$ by paired t-test.