Original Article

Polyunsaturated fatty acid consumption and concentration among South Indian women during pregnancy

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In recent years there is growing interest on the role of long chain ω-3 polyunsaturated fatty acids (ω-3 LC-PUFA) in pregnancy and the growth and development of the offspring. We aim to characterize and provide baseline data on the intake of LCPUFA (ω-3 and ω-6) in a prospective cohort of 829 pregnant Indian women and report associations between LCPUFA intake and erythrocyte membrane phospholipid fatty acid concentration in a subgroup at baseline (1st trimester), the 2nd and 3rd trimesters of pregnancy. The dietary intake of all the macronutrients and of α-linolenic acid(ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) significantly increased over the 3 trimesters of pregnancy while that of ω-6 fatty acids (FA) remained unchanged. Median ω-3 FA intakes of ALA, EPA and DHA, however, were on average low at 0.56, 0.003 and 0.011 g/d, respectively while LA was 14.6 g/d during pregnancy. Consequently, the intake ratio of ALA to LA in the women in the present study was very low at 1:26. A significant decline in erythrocyte membrane arachidonic acid (AA) concentration but not of DHA was observed throughout pregnancy. This might be due to increased efficiency in terms of elongation of parent ω-3 FA. Dietary methods for improving the consumption of ω-3 FA need to be considered in the diets of young women as well as during pregnancy. As newborns primarily depend on placental transfer of ω-3 FA there is need to examine the ω-3 LC-PUFA concentration in infants of mother's with low intakes of ω-3 FA.

Key Words: long chain polyunsaturated fatty acid (LCPUFA), linoleic acid (LA), α linolenic acid (ALA), pregnancy, South India

INTRODUCTION

The dietary intake of long chain ω-3 polyunsaturated fatty acids (ω-3 LC-PUFA) during pregnancy is gaining prominence due to their role in the growth and development of the offspring. Docosahexaenoic acid (DHA), a ω-3 LC-PUFA is an important structural constituent, involved in brain growth and function.[1,2] DHA is also known to have a functional role in visual and neural processes.[1] We have previously reported an association between maternal fish and ω-3 LC-PUFA intake during pregnancy and birth weight in Indian infants.[3] During pregnancy, the requirements for DHA are particularly high. In humans, DHA can be synthesized from its precursor, ALA through a series of desaturations and elongations in the liver. As reviewed by Rioux and colleagues, the capacity of the placenta to synthesize LC-PUFA from its precursor is limited. Therefore, the fetal requirements for these fatty acids and their precursors should be met by the mother's diet.[4]

One of the richest sources of the LC ω -3 PUFAs, eicosapentaenoic acid (EPA) and DHA, is fatty fish. During pregnancy, marginal intakes of DHA are provided mainly from sporadic fatty fish intake.[4-6] Therefore, largely cereal consuming populations, with negligible fish intakes, such as those in India, depend on the consumption of α-linolenic acid (ALA) and its endogenous conversion to DHA. However, previous data suggest that the conversion of ALA to ω-3 LCPUFAs is an inefficient process.[7-9] In addition, the sparse dietary intake data available in Indian populations suggest that the intake of ω-6 FA, which is much higher than ω-3 FA, could affect the elongation of the latter to DHA.[10] A diet rich in ω-3 fatty acids, particularly LC-PUFA, may therefore be crucial to ensure optimum ω-3 LCPUFA concentrations in populations with low fish and high cereal intakes. This paper aims to characterize and provide data on daily LCPUFA (both ω-3 and ω -6) intakes in a cohort of 829 pregnant women, as well as on associations with their erythrocyte phospholipid fatty acid concentration.

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MATERIAL AND METHODS

The present study was a part of a prospective pregnancy cohort conducted at St. John's Medical College Hospital, Bangalore, India, to study the association between maternal nutrition and pregnancy outcomes. In this paper, dietary EFA intakes and concentration during pregnancy are reported. The Institutional Ethical Review Boards at St. John's Medical College Hospital approved all study procedures, and a signed informed consent was obtained from each study subject at the time of enrolment from January 2002 to March 2006. All pregnant women aged 16-40 years, registered for antenatal screening at the Department of Obstetrics and Gynecology and willing to participate were recruited early in their pregnancy (1st trimester at up to 12 weeks of gestation) and were followed until delivery. Women with multiple pregnancies, those with a clinical diagnosis of chronic illness such as diabetes mellitus, hypertension, heart disease and thyroid disease, those who tested positive for hepatitis B surface antigen (HbSAg), HIV or syphilis infection or those who anticipated to move out of the city before delivery were excluded from the study. Socio-demographic details, age, parity and obstetric history were collected at baseline (12.3±3.0 weeks of gestation). Gestational age was confirmed from ultrasonographic measurements within 2 weeks of the baseline visit. A total of 1300 pregnant women were contacted. Of these, 237 refused to participate, 403 dropped out of the study while 166 did not meet the inclusion criteria. During the study, 68 subjects were lost to follow up such that 829 subjects completed the study. Routine antenatal care (folic acid, iron, calcium supplements, and tetanus toxoid) was prescribed to the study participants. None of the pregnant women consumed fish oil or DHA supplements during pregnancy. Information on dietary intake, anthropometry and biochemical concentration was collected at each trimester i.e. at baseline, second trimester (24.1±2.0 weeks) and third trimester (34.0±.5 weeks).

Habitual dietary intake for the preceding 3 months of each trimester of pregnancy was assessed using a pretested, interviewer administered, food frequency questionnaire (FFQ), adapted from that developed for an urban South Indian middle class population.[11] During the interview, standard food measures were placed before the respondent to enable them to quantify the portion size of each food item. In the laboratory, recipes for all the cooked food items had been tested and standardized for raw ingredients, and volume-to-weight conversions for each cooked food item. Nutrient composition of foods were calculated using Indian food conversion tables,[12] wherever available, or from the USDA food composition data.[13] The FFQ was validated against 24 hour food recalls (mean of three records per trimester) obtained at each trimester of pregnancy (n=100, Dwarkanath et al, unpublished data, 2006).

The subjects' height was measured to the nearest 0.1 cm and weight was recorded to the nearest 0.1 kg on a digital weighing scale (Soehnle, Germany) at baseline and monitored throughout pregnancy. Body mass index (BMI) was calculated using body weight and height at baseline.

Measurement of erythrocyte membrane phospholipid fatty acid concentration is discussed in a previous publication.[3]

All statistical analyses were performed with SPSS (version 13.0, SPSS, Chicago, IL). A repeated measures ANOVA was performed to compare the intakes and concentration of major fatty acid components in erythrocytes during pregnancy at the three trimesters. Post hoc Bonferrroni adjusted t-tests were used for pairwise comparison between trimesters. Spearman's rank correlation was used to assess the relationship between ω-3 fatty acid intake (from the validated FFQs) and its content in erythrocyte membrane phospholipids. Levels of significance were set at 0.05.

RESULTS

The study participants were on average 24.7±4.0 years

Table 1a. Macronutrient and fatty acid intakes in the 3 trimesters of pregnancy.

Nutrients	1st trimester (n=827)	2nd trimester (n=547)	3rd trimester (n=510)
Macronutrient intake per day			
Energy, kcal†	1968 (1657 - 2390)	2147‡ (1838-2526)	2106§ (1775-2662)
Protein, g†	57.1 (46.4-71.5)	63.6‡ (52.2-74.8)	62.0§ (51.7-77.6)
Fat, g†	53.1 (41.6-68.1)	58.3‡ (47.3-73.1)	56.7§ (46.7-72.8)
Carbohydrate, g†	318 (268-379)	342‡ (292-396)	342§ (285-413)
Total saturated fat†	18.4 (13.5-24.4)	20.7‡ (16.0- 26.5)	21.0§ (16.0-26.5)
Total monounsaturated fat†	13.2 (10.3-16.9)	14.8‡ (11.8-18.3)	14.2§ (11.4-18.6)
Total polyunsaturated fat	14.7 (10.2-19.3)	16.0 (11.7-20.0)	15.2 (11.5-20.0)
Fatty acid intake (Nomenclature)			
Linoleic acid, g (18:2 ω -6)	14.1 (0.8-18.5)	15.3 (11.1-19.1)	14.5 (10.9-19.1)
A- linolenic acid, g† (18:3 ω -3)	0.51 (0.40-0.66)	0.59‡ (0.47-0.70)	0.58§ (0.47-0.73)
Arachidonic acid, mg (20:4 ω -6)	32.7 (13.5-48.8)	35.8 (18.0-50.6)	33.2 (14.7-50.7)
Eicosapentaenoic acid, mg† (20:5 ω -3)	2.1 (0.6-5.8)	2.6 (0.8-6.5)	3.0 (0.9-6.1)
Docosapentaenoic acid, mg (22:5 ω -3)	0.7 (0.1-1.9)	0.9 (0.2-2.0)	1.0 (0.3-1.9)
Docosahexaenoic acid, mg† (22:6 ω -3)	10.2 (4.3-18.0)	11.2 (4.8-21.2)	11.2 (4.9-19.5)

Values are median (inter-quartile range)
† Significantly different across 3 trimesters using an ANOVA of repeated measures ($p<0.05$)
‡ Significantly different between the 1st and 2nd trimesters using a post-hoc Bonferroni-corrected pair-wise t-test
§ Significantly different between the 1st and 3rd trimesters using a post-hoc Bonferroni-corrected pair-wise t-test

Table 1b. Erythrocyte membrane concentration in the 3 trimesters of pregnancy

Erythrocyte fatty acid concentration (% by weight)	1st trimester (n=133)	2nd trimester (n=130)	3rd trimester (n=123)
Linoleic acid	11.5 (10.2-12.8)	11.3 (9.84-12.8)	11.5 (10.0-12.8)
α-linolenic acid	0.14 (0.12-0.17)	0.15 (0.12-0.18)	0.15 (0.12-0.18)
Arachidonic acid †	8.80 (7.64-10.07)	8.14‡ (6.95-9.21)	7.80§ (6.64-8.84)
Eicosapentaenoic acid	0.05 (0.04-0.09)	0.05 (0.03-0.09)	0.05 (0.03-0.07)
Docosahexaenoic acid	1.97 (1.57-2.65)	2.10 (1.47-2.65)	2.02 (1.45-2.66)

Values are median (inter-quartile range)
† Significantly different across 3 trimesters using an ANOVA of repeated measures ($p<0.05$)
‡ Significantly different between the 1st and 2nd trimesters using a post-hoc Bonferroni-corrected pair-wise t-test
§ Significantly different between the 1st and 3rd trimesters using a post-hoc Bonferroni-corrected pair-wise t-test

Table 2. Fatty acid composition of some important edible oils used in India and in the present study population.

Type of oil	Location of use in India	% Consumption in our study population
Coconut oil	South	4.0
Mustard	North, East	0.4
Groundnut	North, South	16.6
Soyabean	North	0.1
Palm oil	South	1.6
Sunflower/ Safflower	North	77.3

Table 3. Correlation between fatty acid dietary intakes and concentrations across different trimesters.

Nutrients	I trimester (n=133)	II trimester (n=130)	III trimester (n=123)
Linoleic acid	0.094	0.209*	0.191*
A-linolenic acid	-0.007	0.048	-0.140
Arachidonic acid	-0.091	-0.106	-0.154
Eicosapentaenoic acid	0.431**	0.331**	0.370**
Docosahexaenoic acid	0.402**	0.322**	0.413**

Values represent r values
* Correlation significant at the 0.05 level (2-tailed)
** Correlation significant at the 0.01 level (2-tailed)

old. Almost one-third (37.4%) of the women had at least a university degree. Approximately 60% of the women were primiparous. Mean BMI was 22.2 kg/m^2, with 15.9% of the mothers being undernourished (<18.5 kg/m^2).

The median intakes of macronutrients and selected ω-3 and ω-6 fatty acids during pregnancy are shown in Table 1. The dietary intake of all macronutrients and ω-3 fatty acids significantly increased over the 3 trimesters of pregnancy while that of ω-6 fatty acids remained unchanged. With respect to fat intake, significant increase occurred only for saturated and monounsaturated components of fat intake, while PUFA intake remained unchanged. In consequence, there were no significant changes in LA or AA intakes. When analyzed for trimester-based changes, the macronutrient intakes showed a significant increase in both the 2nd and 3rd trimesters. There was also a significant but small increase (15-20%) in the ALA intakes in the 2nd and 3rd trimesters. While DHA intakes in 2nd and 3rd trimester were similar, a trend towards an increase across trimesters was noted only with EPA intakes.

An overview of the oils commonly consumed in the different parts of India is presented in Table 2.[14] Edible oils used for cooking in this population were primarily sunflower oil (77 % of women) and groundnut oil (17 % of women), both of which contain high amounts of LA. (Table 2). Intake of ω-3 fatty acid rich foods such as flaxseed, soya and mustard oil and flesh foods namely, meat and eggs, was low. About half of the women in the study occasionally consumed fish, with the median intakes being extremely low (1.19 g/d). Median ω-3 fatty acid intakes of ALA, EPA and DHA, were 0.56, 0.003 and 0.011 g/d, respectively while that of LA (ω-6 fatty acid) was 14.6 g/d.

The ω-3:ω-6 parent fatty acid ratio in erythrocyte membranes had a similar trend as that of the intake ratio. However, the trends of ω-3 dietary fatty acid intakes during pregnancy were not reflected in the erythrocyte membrane content. Comparison of individual fatty acid intakes and their red cell membrane concentration of ω-3 and ω-6 fatty acids (Table 3) showed that EPA and DHA intake and concentration were positively correlated throughout pregnancy ($p<0.01$). There was no association between ALA intakes and the concentrations of its long chain derivatives (data not shown). Linoleic acid intakes were significantly related to its erythrocyte content only in the 2nd and 3rd trimester.

DISCUSSION

In the pregnancy cohort study, we observed low dietary intakes of the parent ω-3 fatty acid ALA and its long chain derivatives, EPA and DHA. The median dietary ALA intake in our population, mainly from the consumption of lentils and green leafy vegetables, was 50% of the recommended intake of 1.0 g/d during pregnancy.[15] Not surprisingly, the median DHA intake of about 0.011 g/d throughout pregnancy was found in women in the present study, who traditionally consume low amounts of eggs, meat and fish. This was 10 times lower than intakes reported in the diets of pregnant women in Denmark (0.182 g/d) and the United Kingdom (0.147 g/d). This intake was 3% of the daily intake of 300 mg of preformed DHA that is recommended during pregnancy.[16]

The increase in the consumption of ω-6 PUFA-rich sunflower and safflower oils over recent years in Indian diets has resulted in a current ω-3:ω-6 intake ratio of

1:30-70, which is much higher than the recommended ratio of 1:5-10 for optimal health benefits.[17] The study population showed an intake ratio of 1:26 of ω-3:ω-6. While a high LA intake inhibits conversion of ALA to EPA and DHA,[18] and conversely, LA conversion is inhibited by ALA as confirmed by labeled isotope studies. There is recent evidence to suggest that it is the absolute amounts of both ALA and LA in the diet that determine ALA elongation in the body,[19] and not their ratio as previously believed.[10] These studies suggest that there could be poor conversion of ALA into DHA in women with very low total ω -3 fatty acids, and ω-3:ω-6 ratios. Therefore, in the context of a diet containing relatively low amounts of ALA, it would appear that a diet rich in preformed ω-3 LC-PUFA may be crucial to ensure optimum ω-3 LC-PUFA concentration in the pregnant mother. This possibility is strengthened by the finding in the both the present and other studies,[20] that the dietary intakes of LCPUFA correlated well with erythrocyte phospholipid concentrations.

It is known from longitudinal studies that while relative AA concentrations decrease as the pregnancy progresses, the relative DHA concentrations increase during early pregnancy and decrease later, albeit remaining higher than before conception. Normalization occurs only in the postpartum period.[21] Similarly, in the present study, there was a decreasing trend observed in the erythrocyte membrane AA concentrations while the relative DHA concentration remained constant during the course of pregnancy. During the last trimester of pregnancy, there is a significant increase in DHA and the ratio of ω-3/ω-6, starting at approximately 32 weeks of gestational age[22] which could be due to rapid synthesis of brain tissue. It is unclear how DHA concentration was maintained in the women in our study during pregnancy. One possible explanation could be a steady conversion of ALA to DHA, thereby maintaining its concentration in the mother's blood for supply to the developing fetus, although the relatively low ALA and high LA intakes would mitigate this process. On the other hand, it is also possible that the extremely low intakes of DHA lead to adaptive changes that maintain fatty acid desaturation and chain elongation, but this is not known.

In conclusion, South Indian diets during normal pregnancies are low in ω-3 fatty acids, and the red cell membrane concentration of DHA in particular did not change during the course of pregnancy. While this might be due to an increased efficiency of elongation of parent ω-3 fatty acids, dietary methods improving the consumption of ω-3 fatty acids in general need to be considered in the diets of young women as well as during pregnancy. Moreover, it is known that ω-3 fatty acids are important for growth and neural development of the newborns. The fetus primarily depends on placental transfer for its supply and thus, the concentration and supply of the mother is critical. Therefore there is also a need to examine ω-3 fatty acid concentration in infants of mothers with low intake of ω-3 fatty acids.

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AUTHOR DISCLOSURES

The authors have no conflicts of interest with this research.

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南印度的懷孕婦女多元不飽和脂肪酸之攝取與含量

近年來對長鏈 ω-3 多元不飽和脂酸（ω-3 LCPUFA）在懷孕、後代的生長和發育的角色越來越感興趣。本研究目的是探討 829 位懷孕印度婦女的前瞻性世代，描述和提供 LCPUFA（包含 ω-3 和 ω-6）在研究起點（第一孕期）的攝取量資料，並以次群體探討研究起點（第一孕期）、第二孕期、第三孕期 LCPUFA 攝取與紅血球細胞膜磷脂質中脂酸含量之關係。所有巨量營養素、**α-次亞麻油酸(**ALA)、EPA、DHA 的攝取量在 3 孕期中顯著增加，而 ω-6 脂酸攝取量保持不變。ω-3 脂酸中的 ALA、EPA、DHA 攝取量中位數，都低於平均參考值，分別為 0.56、0.003、0.011 克/天，而**亞麻油酸(**LA)在懷孕期則為 14.6 克/天。因此，本研究中的婦女 ALA 與 LA 攝取比值是非常低的 1:26。在紅血球胞膜中花生四烯酸（AA）濃度在整個孕期顯著下降，但 DHA 濃度無改變。這可能由於孕期中體內 ω-3 脂酸延長作用的效率增加。年輕女性以及懷孕婦女需考慮改善飲食以增加 ω-3 脂酸的攝取。因為新生兒的 ω-3 脂肪酸主要取決於胎盤轉移，有必要檢驗母親攝取量低的嬰兒 ω-3 LCPUFA 濃度。

關鍵字：長鏈多元不飽和脂酸、亞麻油酸、α 次亞麻油酸、懷孕、南印度