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Dietary Intake of Natural Sources of Docosahexaenoic Acid and Folate in Pregnant Women of Three European Cohorts

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Key Words
Docosahexaenoic acid · Folate · Nutrition · Pregnancy

Abstract

Background: Folic acid plays a fundamental role in cell division and differentiation. Docosahexaenoic acid (DHA) has been associated with infantile neurological and cognitive development. Thus, optimal intrauterine development and growth requires adequate supply of these nutrients during pregnancy. ***Methods:*** Healthy pregnant women, aged 18–41 years, were recruited in Granada (Spain; n = 62), Munich (Germany; n = 97) and Pécs (Hungary; n = 152). We estimated dietary DHA and folate intake in weeks 20 (w20) and 30 of gestation (w30) using a food frequency questionnaire with specific focus on the dietary sources of folate and DHA. ***Results:*** Both w20 and w30 Spanish participants had significantly higher daily DHA intakes (155 ± 13 and 161 ± 9 mg/1,000 kcal) than the German (119 ± 9 and 124 ± 12 mg/1,000 kcal; $p = 0.002$) and Hungarian participants (122 ± 8 and 125 ± 10 mg/1,000 kcal; $p = 0.005$). Hungarian women had higher folate intakes in w20 and w30 (149 ± 5 and 147 ± 6 μg/1,000 kcal) than Spanish (112 ± 2 and 110 ± 2 μg/1,000 kcal; $p < 0.001$) and German participants (126 ± 4 and 120 ± 6 μg/1,000 kcal; $p < 0.001$), respectively. ***Conclusion:*** Dietary DHA and folate intake of pregnant women differs significantly across the three European cohorts. Only 7% of the participants reached the recommended folate intake during pregnancy, whereas nearly 90% reached the DHA recommended intake of 200 mg per day.

Introduction

Nutrient requirements increase markedly during pregnancy to support fetal growth and expansion of maternal tissues. The quality of nutrient supply during pregnancy is associated with maternal health, pregnancy outcome, rate of complications as well as fetal development and growth [1, 2]. Poor maternal nutrition is one of the key factors leading to compromised fetal growth and adverse effects on child health [3]. Pregnant women often do not meet their increased nutrient needs, particularly of folic acid [4–6]. Folate deficiency is one of the most common vitamin deficiencies worldwide [7]. This vitamin is essential for DNA synthesis, amino acid metabolism and cell division [8, 9]. Poor folate status during early pregnancy is associated with increased rates of neural tube defects [10–12]. Moreover, poor folate supply can lead to increased plasma homocysteine, an established risk factor of placental abruption, preterm delivery and increased rates of low birth weight [13, 14]. Considering these facts, an optimal supply with folate during the

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whole pregnancy – not only at the beginning – might improve pregnancy outcome [15].

Food provides two forms of folate, pteroylmonoglutamate and pteroylpolyglutamate. These two forms differ in their bioavailability, because a hydrolysis of the polyglutamate side chain is necessary before absorption. The estimated average bioavailability of folate from omnivorous diets is about 50% [15], whereas synthetic folic acid from supplements is almost completely available for the metabolism [16]. Folate is sensitive to heat and light, and is easily oxidized during food preparation. Thus, it is hard to achieve a well-balanced folate supply from food [17], and pregnant women are recommended to consume synthetic folic acid from fortified foods, supplements or both, in addition to consuming folate from a varied diet. However, there exists no single recommended value for women in childbearing age or pregnant women throughout Europe. On average, a supplementation of 400 μg/day of folic acid is recommended (e.g. Germany: 600 μg/day, Hungary and Spain: 400 μg/day) [18, 19].

A further nutrient with particular relevance for perinatal development is docosahexaenoic acid (DHA), an n-3 long-chain polyunsaturated fatty acid (FA) mainly found in fatty sea fish [20]. DHA is an indispensable component of all cell membranes in the brain and other tissues, with major relevance for fetal neurological development [21–24]. In the last 3 months of gestation, the fetus accumulates up to 50 mg DHA per day in the brain and adipose tissue [25]. After birth, breast-fed infants are provided with DHA through breast milk. Several controlled studies found that DHA availability during pregnancy is associated with improved cognitive and visual development as well as reduced risk of early preterm birth [26–28]. Therefore, an average dietary DHA intake of at least 200 mg/day has been recommended for pregnant and breast-feeding women [29].

Given the relevance of DHA and folate supply during pregnancy, we assessed the dietary intake of these nutrients in women participating in the Nutraceuticals for a Healthier Life (NUHEAL) Study in Germany, Hungary and Spain, to obtain data on current dietary intakes during pregnancy in these European cohorts.

Subjects and Methods

Subjects and Recruitment

The study population was comprised of the participants of the NUHEAL Study, a prospective cohort study, which compared the effects of dietary supplementation with DHA and/or methyltetrahydrofolate from week 21 of gestation until child birth in mothers from three different European countries [30, 31].

From November 2001 to March 2003, apparently healthy pregnant women (aged 18–41 years) attending antenatal care clinics were recruited between week 12 and 20 of gestation. Recruitment took place at three study sites: the University Hospital of Granada in Spain, the Ludwig Maximilians University in Munich, Germany, and the University of Pécs, Hungary. Further inclusion criteria were: body weight at study entry between 50 and 92 kg, uncomplicated singleton pregnancy, no participation in another clinical trial, no use of fish oil supplements from the beginning of pregnancy and no use of folate or vitamin B_{12} supplements after the 16th week of gestation [30].

Dietary Assessment

Dietary intake was recorded using a food frequency questionnaire (FFQ) containing standard portion sizes, which was based on previous studies evaluating dietary intakes (the MONICA study [32], the nutrition protocol of Freiburg, Germany, and the GISELA study). To address reproducibility and the possibility of changes in dietary intakes, nutritional assessment was completed in week 20 ± 1 (w20) and week 30 ± 1 (w30) of gestation. Intake of nutrients was calculated from the portion size and frequency of food consumption using the German nutrient database (Bundeslebensmittelschlüssel) [33], version II.3. It was decided to use only one nutrient database because of possible systematic errors, which would increase in case of using three different databases [34]. Nutrient intake is expressed as intake per 1,000 kcal. Thirty-three food items were included in the questionnaire primary focused on dietary sources of DHA and folate. For this reason, certain food categories like milk, dairy products and beverages were disregarded. We did not assess the intake of processed food fortified with folate because the used nutrient database contained only incomplete information about those foods.

In the FFQ, details on the following supplements were also recorded: multivitamin juice/pills, beer yeast, wheat bran, flaxseeds and evening primrose oil. However, intakes from the NUHEAL study supplements were not included.

In addition to the amount and frequency of food consumption, women were asked to provide information on the mode of preparation and special dietary habits (e.g. vegetarian diet). Single food items were combined in several food groups such as meat (beef, pork, poultry, liver and processed meat including sausage); seafood (divided into lean fish, medium fat fish and fatty fish); vegetables (raw vegetables, cooked vegetables and legumes), fats (for warm dishes, cold dishes or spreads), fruits, soy products, cheese, eggs, baked goods and potatoes. The frequency of intake was categorized into never/1 or 2–3 times per day/1 or 2–3 or 4–6 times per week/1–3 times per month. Record sheets were checked by an experienced dietician for the presence of implausible amounts or inadequate description to ensure accuracy. Body height of the participants was determined in w20 of gestation and body weight in both w20 and w30. Estimated basal metabolic rate was calculated from weight and height based on the formula of Schofield [35]. Participants whose energy intake calculated from the FFQ was less than the basal metabolic rate were defined as 'under-reporters' and excluded from nutritional analyses.

Statistical Analyses

Data were analyzed with SPSS for Windows 12.0 (SPSS, Chicago, Ill., USA). Normal distribution was examined using the Kolmogorov-Smirnov analysis. One-way analysis of variance and the post hoc Bonferroni test were used to evaluate differences between study groups in normally distributed data. For non-normally distributed data, the Mann-Whitney U test was employed. A linear model was used to obtain more information regarding variables affecting dietary intake. Non-normally distributed data were logarithmized before the test. Statistical significance was considered at $p < 0.05$. Correlations between parameters were estimated by computing Pearson's correlation coefficient in the case of normally distributed values and the Spearman correlation coefficient (ρ) in the case of other distributions, respectively.

Results

Three hundred and eleven pregnant women agreed to participate in the clinical trial. A total of 271 women with an average age of 30.8 years (mean) completed the study protocol (table 1). More than 98% of the women were Caucasians and >96% were living in a partnership. Most of the women consumed omnivorous diets, <1% of the participants followed a vegetarian diet (n = 2). Forty-eight percent of the participants were living in urban areas. At the time of study entry, 42% of the participating women were not working, 32% had a full time job and 13% worked part time. Women from Munich were significantly older than those from the two other centers ($p < 0.001$). In w20 and w30, 12 and 14%, respectively, of the participants reported to smoke. An overview about socioeconomic characteristics of the three study samples is shown in table 2.

In w20, 15 women were excluded from analysis (Germany: n = 12; Spain: n = 2, and Hungary: n = 1), because of apparent under-reporting of dietary intake. Thus, 256 subjects were included in the calculations. In w30, 35 women were excluded (Germany: n = 24; Spain: n = 5, and Hungary: n = 6), because of apparent under-reporting, thus a total of 236 FFQs were included. Women who dropped out of the study did not differ in age, ethnic group, residency, family status and education from the remaining participants.

Body Weight Progress and Birth Outcomes

Body weight and weight gain (on average 5.7 kg from w20 to w30) did not significantly differ between the three cohorts, but in w20 the body mass index was lower in Munich compared to Granada (p = 0.047, table 1). Body mass index increased from w20 to w30 by 2.1 ± 1.3 kg/m^2 (mean ± SD). The birth outcomes, birth weight, head

Table 1. Characteristics of the study cohorts at study entry

	Germany (n = 68)	Spain (n = 147)	Hungary (n = 55)	p value
Age, years	$33.5 \pm 3.5^{a,b}$	30.1 ± 4.9^{a}	29.4 ± 4.8^{b}	<0.001
Weight, kg	67.8 ± 9.9	67.2 ± 8.9	69.6 ± 11.8	NS
Height, cm	166 ± 6.0	162 ± 6.0	165 ± 7.0	NS
BMI	24.4 ± 3.3^{a}	25.8 ± 3.5^{a}	25.5 ± 4.6	0.003
Vegetarians, n	2	0	0	NS
Smokers, n	3	27	1	NS
Cigarettes n/week	22	55	28	NS

BMI = Body mass index; NS = no significant difference. Statistical differences were tested with ANOVA and the post hoc Bonferroni test (means ± SD).

[a, b] Common superscripts indicate a significant difference.

Cigarettes n/week are related to the subgroup of smokers.

Table 2. Socioeconomic characteristics of the study participants (frequency in %)

	Spain	Germany	Hungary
Education			
None	–	1.4	–
Primary school	39.7	62.6	5.5
General qualification for university	54.4	30.6	60.0
University	5.9	4.1	30.9
Other	–	–	3.6
Graduation			
None	5.9	40.8	74.5
With graduation	44.1	25.2	–
Degree	8.8	4.8	–
University graduation	38.2	23.8	23.6
Other	2.9	2.7	1.8
Career			
Appointee	2.9	18.4	–
Employee	52.9	29.3	96.4
Manager	19.1	1.4	–
Worker	1.5	22.4	1.8
Freelancer	7.4	8.2	1.8
Current job			
None	23.5	46.3	52.7
<15 h per week	2.9	2.7	3.6
Half time	16.2	15.0	3.6
Full time	48.5	27.2	25.5
Maternity leave	8.8	–	14.5
University/education	–	0.7	–
Family status			
Single	1.5	6.1	–
Partnership	98.5	93.9	100.0
Habitat			
Urban area	57.4	31.3	80.0
Rural area	42.6	68.0	20.0

Table 3. Dietary intake in w20 and w30 of gestation (medians and interquartile ranges: P25–P75)

	Germany		Spain		Hungary		Total study population	
	1,000 kcal	day	1,000 kcal	day	1,000 kcal	day	1,000 kcal	day
w20								
Energy, kcal		2,204$^{a*, b*}$		3,078^{a*}		3,267^{b*}		2,968
		1,765–3,520		2,492–3,549		2,462–3,994		2,271–3,520
SFA, g	16.0	35.4$^{d, e*}$	15.9	48.1^{d}	16.6	48.0^{e*}	16	45.8
	14.2–20.3	27.9–46.7	14.4–17.9	39.5–59.3	14.4–18.7	38.1–66.7	14.3–18.3	35.2–59.3
MUFA, g	19.7^{a*}	41.4$^{d*, e}$	25.4$^{a*, b*}$	74.2$^{d*, f*}$	19.3^{b*}	60.9$^{e, f*}$	22.90	65.6
	17.5–23.4	32.8–57.8	22.6–28.4	61.0–93.2	17.2–21.3	44.7–72.1	19.3–26.7	47.8–85.2
PUFA, g	9.9^{b*}	20.1$^{d*, e*}$	10.2^{c*}	29.5$^{d*, f*}$	12.4$^{b*, c*}$	39.0$^{e*, f*}$	10.7	29.7
	8.0–12.8	16.5–31.1	9.2–11.8	24.2–40.6	10.9–16.2	29.6–51.9	9.1–12.6	21.9–42.0
DHA, mg	111^{a}	235$^{d*, e*}$	134$^{a, b}$	413$^{d*, f}$	107^{b}	315$^{e*, f}$	126	355
	66.3–140	154–244	103–176	297–327	86.3–151	231–444	91.5–165	240–477
n-6/n-3 ratio	9.0^{a*}	9.0^{d*}	9.1^{b*}	9.1^{e*}	12.0$^{a, b*}$	12.0$^{d*, e*}$	9.3	9.3
	7.9–13.1	7.9–13.1	8.0–10.3	8.0–10.3	9.1–14.5	9.1–14.5	8.1–11.5	8.1–11.5
Folate, μg	123$^{a, b}$	271$^{d, e*}$	110$^{a, c*}$	324$^{d, f*}$	152$^{b, c*}$	429$^{e*, f*}$	116	327
	104–147	206–354	97–124	270–403	123–166	319–610	103–139	262–424
w30								
Energy, kcal		2,203$^{a*, b*}$		2,934^{a*}		2,926^{b*}		2,713
		1,774–2,612		2,301–3,402		2,354–3,662		2,205–3,365
SFA, g	17.8^{a}	37.0$^{d, e}$	16.3^{a}	45.4^{e}	16.1	48.0^{d}	16.4	44.0
	13.9–21.2	30.4–48.2	14.5–17.6	35.4–57.9	14.5–17.8	37.8–59.5	14.5–17.9	35.4–56.7
MUFA, g	19.2^{a*}	40.0$^{d*, e}$	25.5$^{a*, c*}$	71.9$^{d*, f*}$	18.5^{c*}	54.3$^{f*, e}$	23.4	61.8
	17.9–22.1	32.8–61.5	23.5–28.0	55.8–89.3	17.0–21.3	42.7–73.7	19.2–26.6	48.1–85.1
PUFA, g	9.7^{a}	21$^{d*, e*}$	10.8^{b*}	29.5$^{d*, f}$	12.4$^{a, b*}$	37.4$^{e*, f}$	11.0	29.4
	7.7–12.8	15.8–27.2	9.5–12.3	24.0–38.7	10.4–15.8	27.9–56.1	9.5–12.9	22.6–39.2
DHA, mg	107^{a}	259$^{d*, e}$	136$^{a, b*}$	403$^{d*, f}$	110^{b*}	317$^{e, f}$	123	372
	68.3–180	136–363	109–185	325–494	93.3–138	239–458	98.8–175	271–465
n-6/n-3 ratio	9.5^{a}	9.5	9.3^{c*}	9.3^{f*}	12.0$^{b, c*}$	12.0$^{e, f*}$	9.7	9.7
	7.0–13.0	7.0–13.0	8.1–10.8	8.1–10.8	9.4–15.2	9.4–15.2	8.2–11.9	8.2–11.9
Folate, μg	113^{b*}	254$^{d, e*}$	109^{c*}	304$^{d*, f*}$	143$^{b*, c*}$	396$^{e*, f*}$	116	311
	92.1–147	193–320	94.3–124	254–360	119–165	321–526	97.2–138	254–393

Statistical differences were tested using the Mann-Whitney U test. SAF = Saturated FAs; MUFA = monounsaturated FAs; PUFA = polyunsaturated FAs. $^{a–f}$ Common superscripts indicate a significant difference; * $p < 0.001$.

circumference and placental weight were not significantly different between the three cohorts (data not shown). Birth length was significantly higher in the German sample ($p < 0.001$) compared with the two other centers.

Nutrient Intake

The highest calculated energy intake was observed in Hungary (table 3). German women had the lowest energy intake from the FFQs ($p < 0.001$ vs. both other cohorts), while there was no significant difference between Spain and Hungary. Dietary fat intake contributed about 45% to energy intake. Spanish women had a higher total fat intake than both other groups at both time points (table 3). The carbohydrate intake was significantly higher in Germany than in Spain in w20 and w30, and there was also a significant difference between Spain and Hungary in w30. Analysis of the total protein intake and protein intake in grams per kilogram body weight was significantly lower in German and Spanish than in Hungarian cohorts.

Our estimate of folate intake includes natural sources of folate in food, folic acid and folic acid equivalents. The daily intake of folate differed significantly between the three centers. The highest intake was observed in Hungarian women (table 3). Significant differences were found between Hungarian and German as well as between Hungarian and Spanish women. Other variables like age, and the mother's education level and family sta-

Table 4. Important food sources of folate, and their percentage to total folate intake

w20	Germany (n = 56)	Spain (n = 146)	Hungary (n = 54)
Vegetables	31.2	23.4	30.0
Fruits	11.9	18.5	13.4
Bread	11.9	10.6	6.4
Meat products	3.5	5.8	10.0
Nuts, oil seeds	5.9	7.9	12.8
Potato products	7.4	7.6	6.7
w30	(n = 43)	(n = 143)	(n = 50)
Vegetables	25.6	23.4	28.9
Fruits	13.8	17.6	15.0
Bread	10.8	10.9	7.0
Meat products	3.8	5.2	8.1
Nuts, oil seeds	4.9	8.9	13.1
Potato products	7.0	8.3	6.6

Table 5. Important food sources of DHA, and their percentage to total DHA intake

w20	Germany (n = 56)	Spain (n = 146)	Hungary (n = 54)
Fish	42.2	48.1	22.0
Poultry	20.3	23.7	35.5
Eggs	7.1	6.9	9.4
Bread	14.9	4.3	2.5
Sweets and pastries	2.1	2.7	2.1
w30	(n = 43)	(n = 143)	(n = 50)
Fish	47.2	51.0	25.6
Poultry	18.9	25.2	37.0
Eggs	8.3	5.5	7.2
Bread	16.8	13.1	11.7
Sweets and pastries	2.4	2.7	2.5

tus had no effect on the folate intake. The correlation between the two time points was statistically significant for the whole study population ($r = 0.5$; $p < 0.001$). Only 7 and 5% of the women attained the recommended folate intake of 600 µg/day during pregnancy in w20 and w30, respectively. The folate intake of 400 µg/day, which is recommended for pregnant women in Hungary (Hungarian National Center of Epidemiology), was met by 29 and 23% of the total study population in w20 and w30, respectively.

In all study centers, the two most common sources of folate were vegetables and fruits (table 4). Bread was important for the supply in Spain and Germany, but not in Hungary, where meat products and sausages, especially liver, were the major folate sources. Additionally, nuts and oil seeds contributed to a larger extent to the folate supply in Spain and Hungary than in Germany. The correlation between the estimated daily folate intake and the vegetable and fruit intake was significant in the whole study population ($r = 0.6$; $p < 0.001$). Daily folate and liver intake showed the highest correlation coefficient in Hungary ($r = 0.7$; $p < 0.001$), while there was no correlation in Germany.

Fatty Acids

The FA composition and consumption for the three study centers is shown in table 3. Values for monounsaturated FAs were significantly higher in Spain compared to the other centers at both time points. Saturated FA intake was similar in the three centers in w20, whereas in w30 German women showed a significantly higher intake than Spanish women. The n-6 FA intake as well as the ratio of n-6 (linoleic and arachidonic acid) to n-3 (α-linolenic acid, eicosapentaenoic acid and DHA) polyunsaturated FAs was high in all centers. Women from Hungary had the highest n-6 FA intake as well as the highest n-6/n-3 ratio, whereas women from Spain showed the lowest n-6/n-3 ratio and the highest n-3 FA intake. Significant differences in the n-6 intake were found between Hungary and the two other centers at both time points.

The highest DHA intake was found in the Spanish cohort (significantly different from both Hungarian and German cohorts). DHA intake in w20 and w30 correlated with each other ($r = 0.519$; $p < 0.001$) and with daily fish intake ($r = 0.55$; $p < 0.001$). The three most common sources for DHA intake from food in all three study samples were fish, poultry and eggs (table 5). The linear model showed no effect for family status, age, education level and urban/rural residency on DHA intake.

Supplement Intake

Forty-three percent of the women took dietary supplements in w20 and 39% in w30. Hungary provided the biggest group of participants taking one or more supplements in w20 and w30 (78 and 62%, respectively). In the remainder, the number of women taking supplements was about half of those in Hungary (Germany: 36 and 28%, and Spain: 32 and 35%). Multivitamin juices and tablets were the most common supplements, followed by beer yeast, wheat bran and flaxseeds.

FFQ w20 versus FFQ w30

A comparison between the two evaluation time points showed significant differences in energy ($p = 0.01$) and retinol ($p = 0.03$) intake in the total study population. Correlations between FFQ w20 and FFQ w30 were statistically significant for all nutrients ($p < 0.001$). Spearman's rank correlation coefficient ranges from $r_{sp} = 0.49$ to $r_{sp} = 0.62$.

Discussion

This study on the nutrient intake of pregnant women from three different European cohorts indicates a poor folate supply, as well as considerable differences in nutrient supply between the three study centers. To decrease any potential effects of incomplete reporting, we expressed nutrient intakes per energy intake, i.e. compared the nutrient density between the three study centers. About one third of the folate intake was provided by vegetables. Leafy greens such as spinach, legumes, and some fruits as well as vegetables are rich food sources of folate. Staple foods such as bread or potatoes contribute only moderately, but consumed in large amounts they can provide a significant portion of the total folate intake [36]. The relative contribution of other folate sources varied from sample to sample. Nuts and meat products played a particularly important role in the Hungarian cohort, where women showed the highest liver consumption (median; w20: 88 g/week; w30: 59 g/week). German participants obtained their folate supply primarily from vegetables, pastries, fruits and cheese, whereas meat products played only a minor role and liver consumption was negligible. In all centers, about 7% of total dietary folate was supplied by potatoes and potato products, which confirms that staple food can significantly contribute to the total folate intake [36]. While our results indicate that vegetables and fruits were the primary sources of dietary folate intake, Siega-Riz et al. [37] reported that folate-fortified grains and ready-to-eat cereals followed by different kinds of juices were the most important sources for folate intake in pregnant women in the US, where cereals and grains are generally fortified with folic acid.

The average dietary folate intake of the study population was 327 μg/day in w20 and 311 μg/day in w30. Thus, only 6% of the participants reached the intake of 600 μg/day recommended in Germany, Austria and Switzerland, and only 26% of the participants reached 400 μg/day. However, we may have underestimated total folate intake, because we could not account for some fortified foods. The consumption of a folate-rich diet and folic acid supplements is recommended for women of childbearing age [38–40]. Folic acid fortification of foods is an alternative option to cover the needs of women who get pregnant [41–43]. Nationwide fortification programs of staple foods such as flour are well established in many countries around the world [7, 41, 44, 45], primarily because folic acid supplementation during the first weeks of pregnancy decreases the incidence of neural tube defects. For example, cereal fortification with folic acid in Canada has reduced the prevalence of neural tube defects >50% [46]. The rather low folate intake in pregnant women found in this study was also reported in other studies [5, 37]. The folate intake of the Hungarian subjects reported in the present study exceeded the results of a previous Hungarian nutritional survey carried out between 1990 and 1994 [47]. This trial revealed a mean daily folate intake of 166 μg in w20 and 149 μg in w30 of gestation, and hence folate intake may have increased in pregnant Hungarian women during the last decade. However, the 3rd Hungarian dietary survey showed that folic acid intake of adult women did not meet the criteria of the Hungarian recommendations [48]. German non-pregnant women aged 25–51 years were reported to have a mean daily folate intake of about 225 μg/day [49] and, thus, to have a lower intake than the NUHEAL participants, potentially due to an increased health consciousness of a population participating in a dietary intervention trial during pregnancy and different data collection methods.

Also Ortega et al. [50] showed in their survey that in 319 Spanish women aged 18–35 years none of them reached the recommended 400 μg/day of folate.

DHA is primarily contained in fatty sea fish such as salmon, mackerel and herring, as well as in certain micro algae [51]. In the used FFQ, women were asked to separate their intake into lean fish, medium fat fish and fatty fish as well as into the kind of preparation (cooked, fried, conserved, crumbed or soup). Eighty-five percent of the Spanish women ate fatty fish at least once a month. In comparison, only 4% of the Hungarian women consumed fatty fish with high DHA contents, they rather consumed red and sea bass, swordfish and trout, with moderate fat and lower DHA contents. German participants tended to eat more lean fish (e.g. halibut, cod or sole pike) with low DHA contents, which possibly explains their lower DHA intake.

Our results agree with previous data reporting a higher availability of fish in Spain (75 g/day) than Hungary (4 g/day) and Germany (12 g/day) [52]. Authors reported also that cooking and frying are the main types of preparing fish, followed by the use of canned fish, while fish soups play only a minor role. Other possible dietary DHA sources are meats and eggs. A European Commission-funded evidence-based consensus recommendation recently advised that pregnant and lactating women should reach an average intake of at least 200 mg DHA per day [29]. Nearly 90% of the participating women achieved this intake. The WHO has recommended an n-6/n-3 FA ratio between 3:1 and 4:1 [53]. The subjects in this study reached an n-6/n-3 ratio of 10:1, i.e. much higher than the WHO goal. The ratio in Spanish women (9.4:1) is very similar to the 9.8 ± 7.3 previously reported in 162 Spanish women [54]. A somewhat higher dietary DHA intake would contribute to lowering the n-6/n-3 ratio.

While mean macronutrient intake of our study population seems quite adequate, our findings clearly show shortcomings particularly in folate intake, and to a lesser extent also in the amount of DHA intake, in pregnant women in the study population. While there were differences between the three cohorts studied, intake in w20 and w30 within study populations was very similar. Other variables like age, family status or education level did not have significant effects either on folate or on DHA intake from food. An increased dietary micronutrient density and micronutrient supplementation could enhance micronutrient intake [37].

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