ORIGINAL ARTICLE

Maternal Folate Intake during Pregnancy and Childhood Asthma in a Population-based Cohort

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Abstract

Rationale: A potential adverse effect of high folate intake during pregnancy on children's asthma development remains controversial.

Objectives: To prospectively investigate folate intake from both food and supplements during pregnancy and asthma at age 7 years when the diagnosis is more reliable than at preschool age.

Methods: This study included eligible children born 2002–2006 from the Norwegian Mother and Child Cohort Study, a population-based pregnancy cohort, linked to the Norwegian Prescription Database. Current asthma at age 7 was defined by asthma medications dispensed at least twice in the year (1,901 cases; n = 39,846) or by maternal questionnaire report (1,624 cases; n = 28,872). Maternal folate intake was assessed with a food frequency questionnaire validated against plasma folate. We used log-binomial and multinomial regression to calculate adjusted relative risks with 95% confidence intervals.

Measurements and Main Results: Risk of asthma was increased in the highest versus lowest quintile of total folate intake with an adjusted relative risk of 1.23 (95% confidence interval, 1.06–1.44) that was similar for maternally reported asthma. Mothers in the highest quintile had a relatively high intake of food folate (median, 308; interquartile range, 241–366 μg/d) and nearly all took at least 400 μg/d of supplemental folic acid (median, 500; interquartile range, 400–600 μg/d).

Conclusions: In this large prospective population-based cohort with essentially complete follow-up, pregnant women taking supplemental folic acid at or above the recommended dose, combined with a diet rich in folate, reach a total folate intake level associated with a slightly increased risk of asthma in children.

Keywords: folic acid; children's asthma development; birth cohort; epidemiology; maternal diet

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At a Glance Commentary

Scientific Knowledge on the Subject: Periconceptional folic acid supplement use at 400 μg per day is internationally recommended to prevent neural tube defects, and mandatory folic acid food fortification has been implemented in some countries to better target women of reproductive age at the population level. In animal models and human studies, supplementation with folic acid and other methyl donors during pregnancy alters the offspring epigenome, which can influence health outcomes in offspring. Various observational studies have examined the association between maternal folate exposure and children's respiratory outcomes. This literature has been summarized in systematic reviews as inconclusive with limitations from methodologic issues, including possible differential loss to follow-up. Few studies have assessed total folate intake from both food and supplements in relation to asthma at school age when the diagnosis is more certain than at preschool age.

What This Study Adds to the Field: Virtually complete follow-up of children was enabled by linkage to the Norwegian Prescription Database. Folate intake from both food and supplements was assessed with a questionnaire validated against maternal plasma folate measurements. Our findings suggest that pregnant women who take supplements containing folic acid, at or above the recommended dose, combined with a diet rich in folate, reach a total folate intake level associated with a slightly increased risk of children's asthma.

Nutrition in pregnancy plays a key role in fetal growth and development and may impact the long-term health of children (1). The importance of adequate folate status for preventing neural tube defects (2) has led to the current World Health Organization (WHO) recommendation that women should take a folic acid supplement of 400 μg/d from the time of planning a pregnancy until 12 weeks gestation. To better target women of reproductive age in the population as a whole, mandatory folic acid food fortification has been implemented in some countries, including the United States, and is currently under consideration in Europe (3). Although folic acid food fortification and supplementation during pregnancy at these levels are considered safe (2), there are concerns about unintended consequences (4).

Several birth cohort studies have examined the association between maternal folate exposure during pregnancy and subsequent risk of children's respiratory diseases. This literature has been summarized, in several reviews, as conflicting, with limitations caused by heterogeneity in classification of folate intake, the time periods considered for exposure and disease development, and outcome definitions (5–8). Two more recent studies reported associations between asthma in children older than 4 years of age and the use of folic acid supplements containing 1,000 μg or more during pregnancy (9, 10). This dose coincides with, or exceeds, the upper tolerable limit for daily folic acid intake in adults set by the U.S. Institute of Medicine (11).

We previously reported that use of folic acid supplements or higher maternal plasma folate levels in pregnancy was associated with increased risk of wheeze and respiratory tract infections up to 18 months (12) and asthma at 3 years of age (13) from the Norwegian Mother and Child Cohort Study (MoBa).

The objective of the current study was to investigate the association between maternal total folate intake during pregnancy and asthma in MoBa children who have reached age 7 years, an age when the asthma diagnosis is more reliable than at preschool age. Norway offers advantages for the assessment of folate intake from foods because very few foods are fortified with folic acid.

The primary analysis of asthma at age 7 years was based on prescription registry data enabling near complete follow-up of the cohort. We performed secondary analyses using maternal report of asthma and atopy on the questionnaire mailed at age 7 years for comparison with our main results and to evaluate whether associations with asthma differed by current atopy.

Methods

The online supplement provides details on methods.

Study Population

MoBa (14, 15) is a population-based pregnancy cohort (95,248 mothers and 114,761 children born 1999–2009) linked to the Norwegian birth registry and Norwegian Prescription Database (follow-up to April 1, 2014). Asthma at age 7 years was defined in eligible children (sample selection and eligibility criteria, which included reaching the required age) (Figure 1) by prescription data (n = 39,846; cases 1,901) or maternal report (n = 28,872; cases 1,624). Both samples included births from 2002 to 2006 and partially overlapped (n = 23,199). Maternal plasma folate was available for a random sample of children born 2002–2003 (16): 2,724 with maternal food frequency questionnaire (FFQ) data (validation sample) and 2,681 with prescription follow-up and covariate information for an exploratory asthma analysis.

Measures of Maternal Folate Intake and Folate Status

Total folate intake (expressed as folic acid equivalents or dietary folate equivalents (11) included food folate and folic acid from supplements, estimated from a validated FFQ administered at about 22 weeks gestation (17, 18). Maternal plasma folate concentrations were measured (19) at Bevital AS laboratories (www.bevital.no; Bergen, Norway) in a single nonfasting venous blood sample drawn around 18 weeks gestation.

Outcome Measures of Children's Asthma and Atopy

In separate analyses we examined current asthma in children around age 7 using either at least two pharmacy dispensations of asthma medications (inhaled β_2-agonists, inhaled glucocorticoids, combination inhalers with β_2-agonists and glucocorticoids, and leukotriene receptor antagonists) or maternal report of the child ever having doctor-verified asthma plus either asthma symptoms or asthma medication use in the past year (listing a valid brand). From maternal reports of the child's eczema or allergy to either pollen or animal hair (cat or dog) with symptoms in the past year we created four mutually

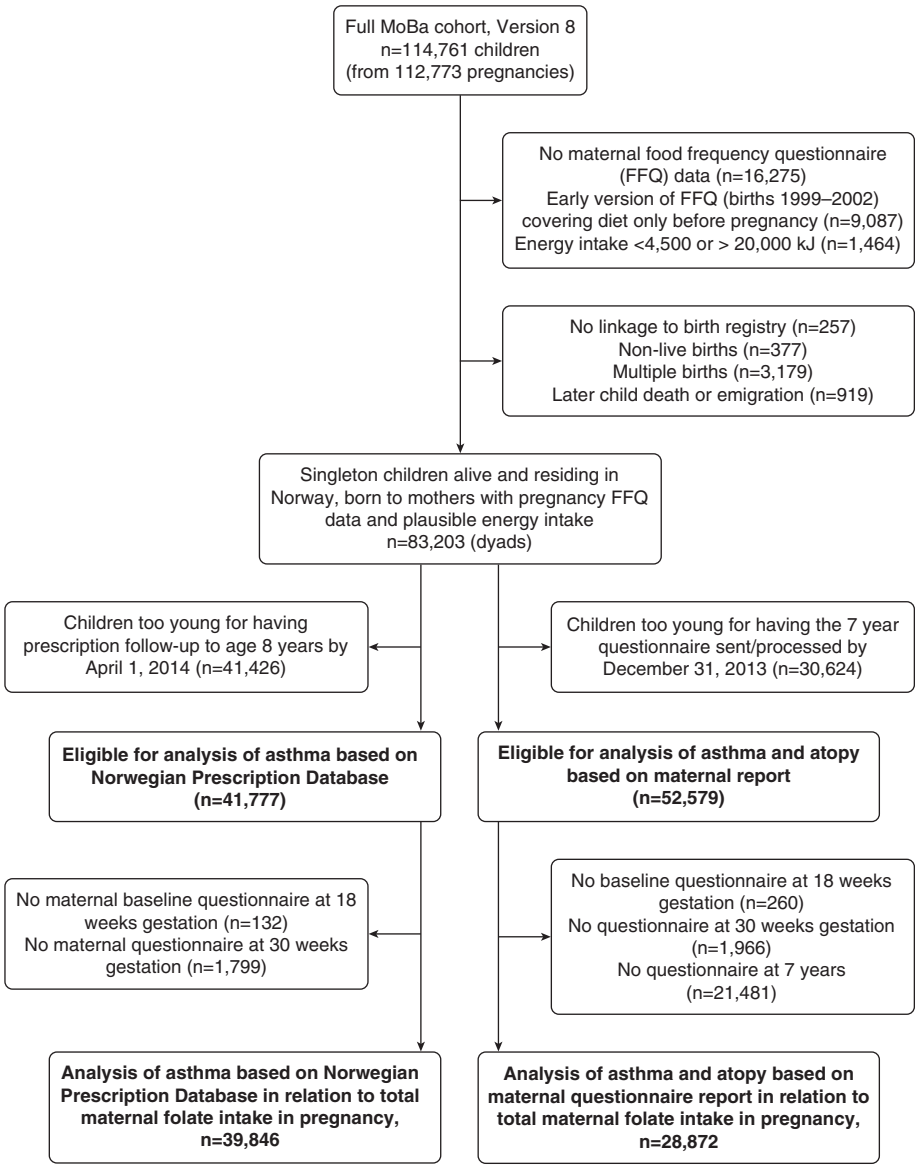

Figure 1. Sample selection. MoBa = Norwegian Mother and Child Cohort Study.

exclusive outcome groups: (*1*) atopy only, (*2*) asthma only, (*3*) asthma with atopy, and (*4*) neither (reference).

Covariates

Potential confounders and other covariates evaluated (Table 1) were based on data from the birth registry (maternal age at delivery, parity, child's sex, and birth weight) or MoBa questionnaires completed around gestational weeks 18 (baseline) and 30, and when the child was aged 6 or 18 months.

Statistical Analyses

We used log binomial regression or multinomial logistic regression with robust cluster variance estimation to calculate relative risks with 95% confidence intervals for children's asthma. The covariates adjusted for are listed in the footnotes of Tables 2 and 3. In sensitivity analyses we additionally adjusted for the child's birth weight (potential mediator), birth year and season, or postnatal child exposures at 6–18 months (duration of breast feeding, dietary supplement intake, and postnatal maternal smoking). Covariates were categorized as shown in Table 1 and entered as dummy variables, except for maternal age and energy intake (both continuous). We tested for multiplicative interaction between total folate intake and prepregnancy body mass index, maternal history of atopy, and smoking. Missing values in covariates were handled by multiple imputation (10 imputations using chained equations). The statistical significance level was 5% for all tests. The analyses were conducted in Stata 13.0 (StataCorp LP, College Station, TX).

Results

Characteristics of the two main study samples (prescription registry sample and maternal report sample) and the validation subsample compared with those eligible for analysis are shown in Table E1 in the online supplement. In the larger prescription registry sample, 57.6% of children (22,957/39,846) had a mother who reported any folic acid supplement use in pregnancy. Among women who took folic acid, the median intake from supplements alone was 400 μg/d (interquartile range, 200–414). Among all women, the median intake of folate from food sources was 257 μg/d (interquartile range, 205–321). In the validation subsample (n = 2,724), the plasma folate concentration (median, 8.7; interquartile range, 5.9–14.7) increased across quintiles of total folate intake (medians, 6.5, 6.9, 8.8, 11.2, and 16.0 nmol/L for quintiles 1–5 in order) (*see* Table E2) and the overall Spearman correlation (continuous) was 0.45.

Women in the highest quintile of total folate intake tended to be older, lighter, and less likely to smoke. They were also more likely to be primiparous, have atopy, take other dietary supplements, and were more highly educated (Table 1). Children born to mothers in the highest quintile of total folate intake were more likely to have been breastfed for at least 6 months and to have received dietary supplements at 6 and 18 months (Table 1). Patterns were similar in the maternal report sample (data not shown). All patterns held for quartiles of maternal plasma folate (results not shown), with the exception of a lack of association with maternal atopy. Plasma folate was also associated with gestational week of sample collection.

The prevalence of current asthma at age 7 years based on prescription registry data was 4.8% (1,901/39,846). Children born to women in the highest versus lowest quintile of total folate intake during pregnancy had more frequent asthma. In the

Table 1. Distribution of Maternal and Child Characteristics According to Total Maternal Folate Intake in Pregnancy, by Quintiles (n = 39,846)

	Total Folate Intake (*Folic Acid Equivalents, μg/d*)				
	Q1: ⩽146	Q2: 147–216	Q3: 217–391	Q4: 392–577	Q5: ⩾578
N	7,869	8,046	7,981	7,918	8,032
Mean maternal age at delivery, yr	29.8	30.1	30.0	30.0	30.2
No. of previous children, %					
0	36.4	37.9	43.5	47.1	52.0
1	39.5	36.9	34.3	35.2	32.5
⩾2	24.1	25.2	22.2	17.7	15.5
Maternal education, %*					
Less than high school	11.8	9.9	9.4	6.3	5.5
High school	38.0	33.7	32.9	27.7	28.1
Up to 4 yr of college	36.7	38.9	40.0	44.8	44.7
>4 yr of college	13.5	17.5	17.7	21.2	21.6
Maternal prepregnancy BMI, %*					
<18.5 kg/m^2	2.2	2.6	3.1	2.7	3.0
18.5–24.9 kg/m^2	59.0	63.5	65.1	65.4	67.0
25.0–29.9 kg/m^2	25.4	24.3	21.7	22.8	21.2
⩾30 kg/m^2	13.3	9.6	10.1	9.2	8.7
Mean maternal energy intake, kJ/d	8,150	10,174	10,680	9,387	10,439
Maternal history of atopy, % yes	31.1	30.5	33.2	34.4	36.8
Cod liver oil taken in pregnancy, % yes	29.5	36.6	39.0	44.0	46.3
Other supplements taken in pregnancy, % yes†	34.7	40.6	65.1	61.6	79.2
Maternal smoking in pregnancy, % yes*	12.8	10.6	10.1	7.1	7.0
Postnatal maternal smoking at 18 mo, % sometimes/daily*	20.3	17.6	17.7	13.7	14.3
Child's sex, % boys	52.3	51.0	50.7	50.3	50.7
Child's birth weight, %*					
<2,500 g	2.6	2.6	2.7	2.6	2.6
2,500–4,500 g	92.6	92.6	92.7	93.0	92.7
>4,500 g	4.8	4.8	4.6	4.4	4.7
Any breastfeeding ⩾6 mo duration, % yes	80.5	84.3	84.0	86.0	86.2
Child given supplements at 6 mo, % yes*†	56.2	60.8	62.3	61.8	64.5
Child given supplements at 18 mo, % yes*†	68.9	72.5	76.9	76.8	80.6

Definition of abbreviations: BMI = body mass index; Q = quintile.
Pearson chi-square *P* value less than or equal to 0.001 for all associations except with child's sex ($P = 0.1$) and birth weight ($P = 0.98$).
*Missing values: from less than 0.05% (birth weight) to 20% (child supplements at 18 mo) as indicated in Table E1 in the online supplement.
†Other supplements taken in pregnancy contain at least one of the vitamins B_2, B_6, B_{12}, C, A, D, and E. Children's supplements include cod liver oil and/or liquid multivitamins.

highest quintile of pregnancy intake 5.6% (449/8,032) of children had been dispensed asthma medications at age 7 compared with 4.6% (363/7,869) in the lowest (Table 2). The relative risk after multivariable adjustment was 23% higher in the highest versus lowest quintile (Table 2). The secondary analysis of asthma by maternal report gave similar results (*see* Table E3).

Sensitivity analyses are presented in Table E4. Results were robust to adjustment for the child's birth weight, or birth year and season, or postnatal childhood exposures. As expected, results were virtually identical for total folate expressed in folic acid equivalents and in dietary folate equivalents. Because some other studies have examined use of higher dose supplements, we divided the top quintile into three categories: 579–799, 800–999, and greater than or equal to 1,000 μg (folic acid equivalents). Within the top quintile, risk did not increase with higher intakes, but data were sparse in the upper category (*see* Table E4). In the much smaller plasma folate sample there was no significant association with asthma or elevated risk in the highest category (*see* Table E5).

Among children with current asthma at age 7 defined by maternal report, 45% of cases (729/1,624) also had current atopy (Table 3). Compared with a reference category of neither asthma nor atopy, children born to mothers in the highest versus lowest quintile of total folate intake during pregnancy had a higher relative risk of asthma only (33% increase) and asthma with atopy (51% increase). Although the *P* value for trend across quintiles of folate intake was strongest for asthma with atopy, contrast analysis on the relative risks (highest vs. lowest quintiles) did not show statistically significant differences ($P = 0.49$ for asthma with atopy vs. asthma only, and $P = 0.09$ for asthma with atopy vs. atopy only; complete case analysis, n = 27,369).

In stratified analyses (*see* Table E6) results did not differ significantly according to prepregnancy body mass index ($P_{\text{interaction}} = 0.22$), maternal atopy ($P_{\text{interaction}} = 0.37$), or maternal smoking ($P_{\text{interaction}} = 0.35$). We examined the effect of food folate separately in users and nonusers of folic acid supplements and did not find a significant difference between groups ($P_{\text{interaction}} = 0.20$).

Discussion

In this population-based pregnancy cohort study, we observed a positive association

Table 2. Crude and Adjusted Relative Risk Estimates (95% Confidence Interval) for Current Asthma at Age 7 Years Based on the Norwegian Prescription Database, by Quintiles of Total Maternal Folate Intake in Pregnancy (n = 39,846)

Total Folate*	Cases/Total (*n*)	Crude RR	Adjusted RR†
Q1	363/7,869	1.00 (ref)	1.00 (ref)
Q2	351/8,046	0.95 (0.82–1.09)	0.97 (0.83–1.13)
Q3	393/7,981	1.07 (0.93–1.23)	1.07 (0.92–1.24)
Q4	345/7,918	0.94 (0.82–1.09)	0.98 (0.85–1.14)
Q5	449/8,032	1.21 (1.06–1.39)	1.23 (1.06–1.44)
p_{trend}		0.01	0.01

Definition of abbreviations: Q = quintile; ref = reference; RR = relative risk.
*Quintile limits for total folate intake from food and supplements (folic acid equivalents, μg/d): Q1, ⩽146; Q2, 147–216; Q3, 217–391; Q4, 392–577; Q5, ⩾578.
†Adjusted for maternal age at delivery (continuous), parity (0, 1, ⩾2), maternal education (less than high school, high school, up to 4 yr of college, >4 yr of college), prepregnancy body mass index (<18.5, 18.5–24.9, 25.0–29.9, ⩾30 kg/m^2), maternal history of atopy (no, yes), maternal smoking in pregnancy (no, yes), and use of cod liver oil (no, yes), other dietary supplements (no, yes), and maternal energy intake (continuous) in pregnancy. Missing values in covariates (Table E1) handled by multiple imputation (m = 10) using chained equations.

between total folate intake during pregnancy and asthma at age 7 in the offspring. Children born to women in the highest versus lowest quintile of total folate intake during pregnancy had about 20% higher relative risk of asthma. Essentially all women in the highest quintile of total intake reported folic acid supplement use and took at least 400 μg/d of folic acid (*see* Table E2) with many taking more; the median supplement intake was 500 μg/d, 25% above the 400 μg/d recommended both by the Norwegian government and the WHO. Women in the highest quintile of total intake, in addition to being supplement users, tended to have higher intakes from food sources; their median intake was 308 μg/d, nearly the cutpoint for the highest quintile of dietary folate intake.

In previous analyses of younger MoBa children we observed associations between maternal use of folic acid supplements and wheezing and lower respiratory illness up to 18 months of age (12). In a sample of 1,962 children with maternal plasma folate measurements during pregnancy we observed a positive association with asthma at age 3 years (13). The current report extends these findings on maternal folate exposure to school age when asthma is more reliably diagnosed and also includes evaluation of asthma with and without atopic illness.

Limitations and Strengths of this Study

As in other large nationwide population-based studies, we were not able to classify asthma based on clinical examination. Likewise we classified atopy based on questionnaire report of allergic conditions, limited to eczema and two aeroallergens, and some misclassification in our outcome measures cannot be ruled out. Another limitation of our study, as in any observational study, is that we cannot exclude the possibility of residual or unmeasured confounding. Women who take folic acid supplements and have diets higher in folate might differ from other women in ways related to unknown causes of asthma, or health care utilization. Although we cannot exclude this possibility, Norway offers advantages because of the universal access to health care, both during pregnancy and childhood, and prescription medication coverage, coupled with a relatively narrow range of income variability. The objection could be raised that the association we are ascribing to folate could be caused by correlated micronutrients from dietary supplements (20). However, adjustment for the common use of cod liver oil in Norway and other supplements, including multivitamins, had little impact on the results.

Our study has several strengths. We used a validated FFQ to estimate total folate intake from both food and supplements. Many previous studies have examined only supplement use or separately considered diet and supplements. Total folate intake from our FFQ correlated well with plasma

Table 3. Crude and Adjusted Multinomial Logistic Regression Relative Risk Estimates for Current Asthma and/or Current Atopy at Age 7 Years Based on Maternal Questionnaire Report, by Quintiles of Total Maternal Folate Intake in Pregnancy (n = 28,872)

		Current Atopy Only (*n* = *4,460*)			Current Asthma Only (*n* = *895*)			Current Asthma with Atopy (*n* = *729*)		
Total Folate*	Neither Asthma nor Atopy (*n* = *22,788*) Reference (*n*)	Cases (*n*)	Crude RR (95% CI)	Adjusted RR (95% CI)†	Cases (*n*)	Crude RR (95% CI)	Adjusted RR (95% CI)†	Cases (*n*)	Crude RR (95% CI)	Adjusted RR (95% CI)†
Q1	4,190	758	1.00 (ref)	1.00 (ref)	160	1.00 (ref)	1.00 (ref)	116	1.00 (ref)	1.00 (ref)
Q2	4,425	872	1.09 (0.98–1.21)	1.11 (1.00–1.25)	167	0.99 (0.79–1.23)	1.07 (0.85–1.34)	109	0.89 (0.68–1.16)	0.96 (0.73–1.26)
Q3	4,461	819	1.01 (0.91–1,13)	0.99 (0.88–1.11)	165	0.97 (0.78–1.21)	1.02 (0.80–1.29)	133	1.08 (0.84–1.39)	1.11 (0.84–1.47)
Q4	4,878	972	1.10 (0.99–1.22)	1.06 (0.95–1.19)	173	0.93 (0.75–1.16)	0.99 (0.79–1.25)	176	1.30 (1.03–1.66)	1.36 (1.06–1.74)
Q5	4,834	1,039	1.19 (1.07–1.32)	1.12 (1.00–1.26)	230	1.25 (1.01–1.53)	1.33 (1.05–1.68)	195	1.46 (1.15–1.84)	1.51 (1.16–1.96)
p_{trend}			0.001	0.11		0.03	0.02		<0.0001	<0.0001

Definition of abbreviations: CI = confidence interval; Q = quintile; ref = reference; RR = relative risk.
*Quintile limits for total folate intake from food and supplements (folic acid equivalents, μg/d): Q1, ⩽146; Q2, 147–216; Q3, 217–391; Q4, 392–577; Q5, ⩾578.
†Adjusted for maternal age at delivery (continuous), parity (0, 1, ⩾2), maternal education (less than high school, high school, up to 4 yr of college, >4 yr of college), prepregnancy body mass index (<18.5, 18.5–24.9, 25.0–29.9, ⩾30 kg/m^2), maternal history of atopy (no, yes), maternal smoking in pregnancy (no, yes), and use of cod liver oil (no, yes), other dietary supplements (no, yes), and maternal energy intake (continuous) in pregnancy. Missing values in covariates (Table E1) handled by multiple imputation (m = 10) using chained equations.

folate. Because there was virtually no fortification with folic acid in Norway in the study period our estimation of intake of folate from foods was simplified and therefore possibly more accurate than folate estimates from populations consuming fortified foods. In addition, we had a large sample size (1,901 cases) providing good power to study asthma at school age. We also had data on a large number of potential confounders including postnatal child exposures.

Our primary outcome of asthma was defined by the filling of two prescriptions for asthma medications within a 12-month period. This is an objective outcome, requiring physician diagnosis. In a validation study of the MoBa 7-year questionnaire items regarding asthma, we have found that even a single dispensing of asthma medication was very rare in the absence of the mother's report of doctor diagnosis of asthma (21). The requirement for a second prescription within 12 months in the current study decreases the possibility that the medication is being prescribed for self-limited wheezing illness after a viral infection or as part of an evaluation to determine the child's symptoms response to asthma medication. By requiring two prescriptions we expect a higher positive predictive value, which has been reported to be the most important property of an asthma definition when the goal is to estimate relative risks as in our study (22). In addition to the validation in our own MoBa study population (21), an earlier validation study in Sweden, with a similar health care system to Norway, concluded that asthma medication is a suitable proxy for asthma in older children and adults based on comparison of data from their prescription registry and national patient registries (23).

The use of this objective outcome also addresses potential selection bias caused by loss to follow-up at age 7 years based on failure to return the questionnaire at this age because the prescription registry covers the entire population. Similar associations for asthma by maternal report and prescription data indicate that loss to follow-up had little influence. As expected, the prevalence of asthma at age 7 years based on two dispensations of asthma medication (4.8%) is slightly lower than the reported national prevalence of around 6% for only one dispensation among children aged 6–12 years (24), but quite similar, suggesting little selection with regard to asthma among MoBa participants. Furthermore, the prevalence among those eligible for the current study was the same as in study participants (*see* Table E1).

Comparison with Other Studies and Potential Mechanism

The current study is the largest to date that examines total folate intake during pregnancy in relation to children's asthma at school age. The total folate intake in these Norwegian women is very similar to that of women of reproductive age in the United States, which practices mandatory folic acid food fortification. Specifically, the median total folate intake in dietary folate equivalents (*see* Table E1) was 482 versus 490 reported for nonpregnant women aged 19–30 years in the U.S. National Health and Nutrition Examination Survey study from 2003–2006 (25). Two recent studies reported 20–40% increased risk of asthma at school age (4.5–8 yr) associated with dispensing of high-dose maternal folic acid supplements: 1,000 μg in the study of Veeranki and colleagues (9) and 5,000 μg in the study of Zetstra-van der Woude and colleagues (10). Our study suggests that a lower intake of total folate is associated with increased risk of asthma. We only observed an increased risk in the top quintile, which could suggest a threshold effect. This level of intake (≥578 μg/d of folic acid equivalents) can be reached by taking the WHO recommended dose of 400 μg folic acid combined with either a multivitamin containing folic acid or at least 300 μg/d of food folate.

Four prospective studies of childhood asthma at school age (5–8 yr) have examined folic acid supplement use during pregnancy without finding an association (26–29); sizes ranged from 130 to 605 cases. Thus, differences may be caused in part by the greater power in the current study with 1,901 cases. However, because these studies did not include a measure of total folate intake, from both foods and supplements, their null findings may not be in conflict with ours. The U.S. study (29) recruited pregnant women from 1997 to 2000, mostly after folic acid food fortification was implemented. Thus, the folate intake among the reference group of women, who did not take folic acid supplements, could already be above a threshold where risk of asthma does not increase much further. Of note, a U.S. study found no evidence for protection against neural tube defects from folic acid supplement use in pregnancy and interpreted this result as evidence that fortification had already increased intake sufficiently to prevent these birth defects (30).

Studies with null results from European countries without mandatory folic acid fortification of the food supply (26–28) only analyzed food folate or folic acid supplements, but not the combined intake, and thus may not have identified the pregnancies with the highest total intake of folate. Two previous studies of children's asthma at school age used FFQs to assess folate intake amounts from food and supplements, as in our study, during pregnancy (31, 32). Nwaru and colleagues (31) studied children at age 5 years in Finland and found no association of total folate intake with asthma. Although the dietary folate intake was relatively high (mean, 364 μg/d), the folic acid intake from supplements was very low (mean, 48 μg/d). In contrast, the Australian study population of Withrow and colleagues (32) took higher dose folic acid supplements during early pregnancy (median, 658 μg/d) and increased risks of asthma at age 3.5 years, and persistent asthma at ages 3.5 and 5.5 years were observed.

Few studies to date have investigated maternal circulating folate levels in relation to asthma in school age children. Magdelijns and colleagues (27) reported a tendency of an inverse association between red blood cell folate in late pregnancy (wk 35) and asthma risk at age 6–7 years (n = 837; 43 cases in total). The lack of association with plasma folate in the current study likely reflects the much lower statistical power in the plasma subsample with only 127 cases compared with 1,901 in the primary analysis of folate intake and 507 cases in our previous case-control study of asthma at age 3 (13). Very large studies with measurements of red blood cell folate, which reflects longer term status than plasma folate, may be necessary to more definitely answer the question of whether maternal folate status in the first trimester is associated with children's asthma development, because other lifestyle factors, metabolism, and genetics all can affect folate levels in addition to intake.

Folate (natural or as synthetic folic acid) provides, with vitamin B_{12} as cofactor,

methyl groups for the synthesis of methionine and *S*-adenosylmethionine, which acts as a methyl donor (4). Epigenetic modification, more specifically DNA methylation, provides one potential mechanism for an effect of *in utero* exposures to folate and other methyl donors on the offspring (33). Although it did not address relevance to asthma, a recent study identified differential methylation at various loci in newborns in relation to maternal folate levels in pregnancy (34). In an *Agouti* mouse model, supplementation of mothers with folate and other methyl donors just before and during gestation and during lactation, abrogated effects of an environmental contaminant (bisphenol-A) on methylation in offspring (35). Interest in this mechanism with respect to asthma was stimulated by the report of Hollingsworth and colleagues (33) that *in utero* supplementation with methyl donors in pregnant mice altered locus-specific DNA methylation and predisposed to allergic airway disease by directing the differentiation of T lymphocytes toward a TH2 phenotype. One of the top differentially methylated loci, *RUNX3*, in a previous knockout mouse model (36), displayed an allergic asthma phenotype. However, this article was recently retracted because of problems with the airway hyperresponsiveness data; the other data in the article were not affected (37). We acknowledge that the mechanistic data on periconceptional folate, or other methyl donors, and asthma pathogenesis are very limited.

Folic acid supplementation during pregnancy has an established role in the prevention of neural tube defects but the mechanism of this protection remains largely unknown (2). Two MoBa studies have found the use of folic acid supplements during pregnancy to protect against other neurologic outcomes including language delay (38) and autism spectrum disorders (39). Taken together with our results, these findings point toward multiple folate-dependent pathways in the fetus of importance to development of neurologic and immune systems.

Conclusions

In this large prospective pregnancy cohort, pregnant women taking folic-acid-containing supplements at or above the recommended dose, combined with a diet rich in folate, reach a total folate intake level that was associated with a slightly increased asthma risk in children. We note that increased risk of asthma was only seen in the highest quintile of intake in which women also had a generally folate-rich diet. In populations with very low folate intake, women taking the WHO recommended dose of folic acid may not reach this level of intake. On the other end of the spectrum, the specific associations might not be observable in populations where mandatory folic acid food fortification has already been implemented. ■

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