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Maternal methyl-donor nutrient intake and serum one-carbon metabolites in relation to small- and large-for-gestational-age births: a prospective cohort study

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ABSTRACT: Evidence regarding the relationship between maternal methyl donor nutrients (MDNs) status and neonatal birth weight remains limited. This study aims to investigate the associations between maternal intakes of MDNs (vitamins B_2, B_6, folate, B_{12}, choline, betaine, methionine), serum one-carbon metabolism biomarkers (including serum MDNs, S-adenosyl methionine [SAM], S-Adenosyl-L-homocysteine [SAH], and homocysteine), and the risk of small- (SGA) and large-for-gestational-age (LGA) births. A total of 2121 mother-infant pairs from the Tongji Birth Cohort were included. During the second trimester, we conducted dietary assessments using a validated food frequency questionnaire and collected fasting blood samples. Serum biomarkers of one-carbon metabolism were quantified via ultra-performance liquid chromatography-tandem mass spectrometry. Robust Poisson and weighted quantile sum (WQS) regressions were employed for analysis. Among participants, 147 SGA and 288 LGA births were identified. Higher food-derived vitamin B_2 and methionine intakes were associated with reduced SGA risk (RR = 0.61, 0.40-0.95 and RR = 0.46, 0.29-0.72, respectively). Intakes of supplement-derived vitamin B_2, B_6, folate, and B_{12} were inversely associated with the risk of LGA, showing reductions of 35%, 32%, 33%, and 29% in the highest tertile. MDN-rich dietary patterns and higher MDN scores were inversely associated with SGA and LGA risks, respectively. Elevated serum vitamin B_{12} (0.60, 0.37-0.99), choline (0.58, 0.34-0.98), betaine (0.64, 0.42-0.98), and SAH (0.37, 0.20-0.68) levels were associated with a reduced risk of SGA. Conversely, elevated homocysteine levels were linked to an increased SGA risk (27.52, 9.61-78.82). Higher serum betaine (0.74, 0.56-0.98) and SAM (0.71, 0.51-0.99) levels were inversely associated with LGA risk. WQS regression showed inverse associations between mixed one-carbon metabolites and both SGA and LGA risks. Adequate maternal MDN intakes and a favorable one-carbon metabolism status are associated with reduced risks of SGA and LGA births, highlighting the importance of maternal nutrition during pregnancy.

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1. Introduction

Birth weight is a key indicator of fetal development and is closely associated with perinatal outcomes and long-term risks of metabolic and cardiovascular diseases [1, 2]. Infants born small (SGA) or large (LGA) for gestational age illustrate deviations from optimal intrauterine growth [2], forming a U-shaped association with later-life health risks [3-5]. Despite improvements in maternal and child health, the global burden of abnormal birth weight remains substantial [6], including China [7, 8]. Identifying modifiable maternal factors that influence fetal growth is therefore essential for early prevention.

Methyl donor nutrients (MDNs), including vitamins B_2, B_6, folate, B_{12}, choline, betaine, and methionine, play a vital role in one-carbon metabolism, a biochemical pathway essential for placental function and epigenetic regulation [9, 10]. Key intermediates of this pathway include S-adenosylmethionine (SAM), S-adenosylhomocysteine (SAH), and homocysteine (Hcy). Disruptions of one-carbon metabolism, particularly elevated maternal Hcy, have been implicated in endothelial dysfunction, placental insufficiency, and impaired fetal growth [11-13]. Emerging evidence also suggests that maternal vitamin B_{12} deficiency may contribute to adverse birth outcomes [14]. MDNs are primarily derived from food intake, and supplementation is often recommended during pregnancy to meet increased nutritional demands. While maternal folate intake has been associated with a reduced risk of SGA [15, 16], evidence regarding other MDNs remains limited and inconsistent [17, 18]. To better capture the cumulative effects of MDNs, recent studies have explored composite MDN scores and MDN-rich dietary patterns [9], but their relationships with fetal growth outcomes remain poorly understood.

Therefore, this prospective cohort study aimed to investigate the associations between maternal MDN intake and SGA and LGA births. We further assessed a broad panel of serum one-carbon metabolism biomarkers to evaluate their individual and combined effects on fetal growth.

2. Methods

2.1 Study design and population

The Tongji Birth Cohort (TJBC) is an ongoing dynamic cohort study based in Wuhan, Hubei province, China [19]. Between March 2018 and October 2023, the TJBC enrolled 2491 participants, of whom 2349 (94.3%) completed a food frequency questionnaire (FFQ) during the second trimester of pregnancy (14-27^{+6} weeks). This study focused on mid-pregnancy dietary data to mitigate potential biases from first-trimester pregnancy symptoms (e.g., nausea and vomiting), which may significantly alter dietary habits and further affect serum indicator levels. After excluding participants with missing delivery data, twin births (n = 203), or extreme daily energy intakes (<500 kcal/day or >3500 kcal/day, n = 25), the final analytical sample comprised 2121 eligible subjects (**Fig. S1**). The research protocol has been registered on the clinical trial website with the registration number ChiCTR1800016908.

2.2 Assessment of dietary and supplement intake

The total intake of MDNs was calculated separately for food and supplement sources. Dietary intake was assessed using a previously validated semi-quantitative FFQ through face-to-face interviews [19]. The reliability and validity correlation coefficients of the FFQ for MDNs ranged from 0.31 to 0.51 and 0.36 to 0.39, respectively. Participants reported their intake of 200-items over the month preceding questionnaire completion. The primary food groups included cereals, grains, tubers, meats, aquatic products, eggs, beans, vegetables, nuts, and dairy products. Nutrient and energy intakes from foods were calculated using the China Food Composition Database.

Supplement-derived MDN intakes (vitamins B_2, B_6, folic acid, and B_{12}) were recorded through an interviewer-administered questionnaire, which included details on the type, dosage, frequency, and gestational weeks when supplementation started and ended. Total MDN intake was the sum of food- and supplement-derived intakes. Since supplement intakes of choline, betaine, and methionine were minimal in this cohort, their total intakes were estimated exclusively from food sources.

2.3 Determination of serum one-carbon metabolism biomarkers

Following a minimum fasting period of 10 hours, venous blood samples were collected by certified laboratory physicians. The samples were centrifuged at 3000 × g for 10 minutes to separate the serum, which was then stored at -80°C. Serum levels of one-carbon metabolism biomarkers were quantified using a modified Ultra Performance Liquid Chromatography-Tandem Mass Spectrometry method (ACQUITY UPLC-TQ-XS, Waters) [20]. Isotope-labeled internal standards were used to ensure the accuracy and precision of the measurements. For values below the limit of detection (LOD), the LOD divided by the square root of two was used as a substitute.

2.4 Outcomes and definitions

The primary outcome variables were SGA and LGA, which were defined using sex-specific birth-weight-for-gestational-age percentiles based on the Chinese population [21]. SGA was classified as a birth weight below the 10^{th} percentile, while LGA was defined as a birth weight above the 90^{th} percentile.

2.5 Assessment of covariates

Participants completed an interviewer-administered questionnaire to collect sociodemographic information, lifestyle factors, and reproductive history. Pre-pregnancy body mass index (BMI) was calculated using participants' height and pre-pregnancy weight. Education levels were categorized by years of completed schooling: primary (≤ 9 years), secondary (10-12 years), college (13-15 years), and university (≥ 16 years). Monthly family income per capita (in Chinese Yuan) was categorized into < 5000, 5000-9999, 10000-14999, and ≥ 15000. Lifestyle behaviors, including pregnancy exercise, drinking, and smoking, were assessed as binary variables (yes/no). Pregnancy complications, such as gestational diabetes mellitus (GDM) and pregnancy-induced hypertension (PIH), were diagnosed by qualified physicians. Gestational weight gain

was calculated as the difference between pre-pregnancy weight and weight at delivery. Overall dietary quality was assessed using the Alternate Healthy Eating Index for Pregnancy (AHEI-P) score [22].

2.6 Statistical methods

Continuous variables were described as mean ± standard deviation or median (interquartile range), and categorical variables as frequencies (percentages). Group comparisons were conducted using the Kruskal-Wallis or Chi-square test, as appropriate. Missing data were addressed by multiple imputation. Dietary data were energy-adjusted using the nutrient residual method [23]. Methyl donor nutrient scores (MDNS) were calculated by summing the z-scores of seven MDNs. Reduced-rank regression was used to identify dietary patterns with MDNs as response variables [24]. For the supplement-derived analysis, intakes of vitamins B_2, B_6, and B_{12} were categorized as nonusers, low, or high intake, using cutoff values of 1.8 mg/day, 2.6 mg/day, and 4.0 μg/day, respectively. Folic acid intake was divided into four groups based on thresholds of 400 and 800 μg DFE/day. Each nutrient was assigned a score (0-2 or 0-3), and a total supplement score ranging from 0 to 9 was calculated. Participants were then classed into three groups based on their total score: nonusers (0 points), low-dosage intake (1-5 points), and high-dosage intake (6-9 points).

Exposure variables were analyzed both as continuous variables and by tertiles (T1-T3). Serum one-carbon metabolism biomarkers were log-transformed. Spearman's rank correlation was used to assess associations among MDNs and biomarkers. Modified Poisson regression models were applied to estimate risk ratios (RRs) and 95% confidence intervals (CIs). Linear trends were evaluated by modeling the median value of each tertile as a continuous variable, with multiple comparisons adjusted using the Benjamini-Hochberg false discovery rate (FDR < 0.05). Restricted cubic spline (RCS) models were used to evaluate dose-response relationships. We employed weighted quantile sum (WQS) regression (gWQS R package [25]) to assess combined effects of multiple serum biomarkers. This approach generates a WQS index representing cumulative mixture effects and component weights (0-1) reflecting individual biomarker contributions. To avoid reversal paradox, analyses were constrained to negative-direction effects.

Stratified analyses were performed by maternal age (< 30 vs. ≥ 30 years), GDM status (yes vs. no), and pre-pregnancy BMI (< 24.0 vs. ≥ 24.0 kg/m^2). For MDN intake, three models were applied: Model 1 (unadjusted); Model 2 (adjusted for maternal age, pre-pregnancy BMI, education, family income per capita, primiparity, smoking, drinking, pregnancy exercise, gestational weight gain, GDM, PIH, and gestational week at data collection); and Model 3 (further adjusted for total energy intake and intakes of vitamin C and E). Source-specific models additionally included adjustments for supplement use or the AHEI-P score. For serum biomarker analyses, multivariable models included the same covariates as Model 2.

All statistical analyses were performed using IBM SPSS Statistics 26.0 and R version 4.4.1. A two-sided *P* value < 0.05 was considered statistically significant.

3. Results

3.1 Characteristics of the study populations

A total of 2121 mother-infant pairs were included in this study, comprising 147 SGA and 288 LGA infants (**Table 1)**. The mean maternal age was 29.2 years, and the mean gestational week at data collection was 22.6 weeks. Compared with mothers of appropriate for gestational age (AGA) infants, those in the SGA group had lower pre-pregnancy BMI, and a higher proportion of primiparity (both $P = 0.001$). In contrast, mothers in the LGA group were older ($P = 0.001$), had higher pre-pregnancy BMI, greater gestational weight gain, and a higher prevalence of GDM, but a lower proportion of primiparity.

Table 1 Baseline characteristics of mothers and infants according to birth size category

Characteristics	Total	SGA	AGA	LGA	P^1	P^2	P^3
n	2121	147	1686	288			
Maternal information							
Maternal age (years)	29.4 ± 3.4	29.1 ± 3.6	29.3 ± 3.4	29.9 ± 3.7	0.531	0.010	0.039
Pre-pregnancy BMI (kg/m^2)	21.2 ± 3.0	20.3 ± 3.0	21.1 ± 2.9	22.0 ± 3.2	0.001	< 0.001	< 0.001
Gestational weight gain (kg)	15.0 ± 5.1	14.3 ± 4.7	14.7 ± 5.0	16.6 ± 5.2	0.164	< 0.001	< 0.001
Gestational week at data collection	21.1 ± 4.8	20.7 ± 5.0	21.1 ± 4.8	21.3 ± 4.7	0.809	0.791	0.689
Ethnicity (Han Chinese), *n* (%)	2077 (97.9)	145 (98.6)	1648 (97.7)	284 (98.6)	0.677	0.347	1.000
Education, *n* (%)					0.095	0.051	0.004
Primary (≤ 9 years)	143 (6.7)	7 (4.8)	113 (6.7)	23 (8.0)			
Secondary (10-12 years)	319 (15.0)	13 (8.8)	257 (15.2)	49 (17.0)			
College (13-15 years)	661 (31.2)	45 (30.6)	512 (30.4)	104 (36.1)			
University (≥16 years)	998 (47.1)	82 (55.8)	804 (47.7)	112 (38.9)			
Family income per capita (CNY/month), *n* (%)					0.267	0.520	0.597
< 5000	25 (1.2)	0 (0.0)	23 (1.4)	2 (0.7)			
5000-9999	238 (1.2)	17 (11.6)	189 (11.2)	32 (11.1)			
10000-14999	1212 (57.1)	84 (57.1)	970 (57.5)	158 (54.9)			
≥ 15000	646 (30.5)	46 (31.3)	504 (29.9)	96 (33.3)			
Primiparity, *n* (%)	1635 (7.1)	132 (89.8)	1307 (77.5)	196 (68.1)	0.001	< 0.001	< 0.001
Pregnancy exercise, *n* (%)	1058 (49.9)	70 (47.6)	852 (50.5)	152 (47.2)	0.498	0.299	0.938
Drinking, *n* (%)	126 (5.9)	4 (2.7)	107 (6.3)	14 (4.9)	0.074	0.331	0.289
Smoking, *n* (%)	51 (2.4)	3 (2.0)	37 (2.2)	11 (3.8)	1.000	0.098	0.480
Gestational diabetes, *n* (%)	360 (17.0)	25 (17.0)	259 (15.4)	74 (25.7)	0.597	< 0.001	0.041
Pregnancy-induced hypertension, *n* (%)	85 (4.0)	10 (6.8)	64 (3.8)	12 (4.2)	0.076	0.763	0.235
Multivitamin use, *n* (%)	1236 (58.3)	81 (55.1)	998 (59.2)	156 (54.2)	0.327	0.110	0.853
AHEI-P	54.8 ± 10.8	53.5 ± 10.5	54.9 ± 10.8	54.7 ± 11.1	0.080	0.798	0.188
Delivery information							
Delivery gestational age (weeks)	39.2 ± 1.3	39.4 ± 1.2	39.2 ± 1.3	39.0 ± 1.5	0.293	0.008	0.016
Birth weight (g)	3292.7 ± 423.2	2661.1 ± 273.0	3248.7 ± 322.4	3868.4 ± 353.1	< 0.001	< 0.001	< 0.001
Birth length (cm)	50.0 ± 1.6	48.5 ± 1.5	49.9 ± 1.4	51.2 ± 1.9	< 0.001	< 0.001	< 0.001
Male infant, *n* (%)	1115 (52.6)	81 (55.1)	889 (52.7)	145 (50.3)	0.580	0.455	0.348

Abbreviation: AGA, appropriate for gestational age; AHEI-P, alternate healthy eating index for pregnancy scores; BMI, body mass index; CNY, Chinese yuan; LGA, large for gestational age; SGA, small for gestational age.
Values are mean and SD or number and percentage. *P* values are from the Kruskal-Wallis or chi-square test.
P^1, P value for comparison of SGA vs AGA; P^2, P value for comparison of LGA vs AGA; P^3, P value for comparison of SGA vs LGA.

3.2 MDN intake, serum one-carbon metabolites, and their correlations

Inadequate intake of MDNs from food was common (**Table S1**), with more than 50% of participants failing to meet the estimated average requirement (EAR) during pregnancy, particularly for vitamin B_6. In this study, 58.3% of the pregnant women reported having taken multivitamin supplements during the dietary survey (**Table 1**). When supplement intake was considered, the average daily intake of vitamins B_2, B_6, folate, and B_{12} was 1.9 mg, 1.8 mg, 994.9 μg DFE, and 6.5 μg, respectively (**Table 2**). The mean daily intakes of choline, betaine, and methionine were 278.7 mg, 72.1 mg, and 1072.3 mg, respectively. Compared with the AGA group, mothers in the LGA group had lower dietary intakes of vitamins B_6 and B_{12}, while a lower proportion of mothers in the SGA group met the EAR or recommended nutrient intake (RNI) for vitamin B_{12}.

Table 2 Maternal MDN intake and one-carbon metabolism biomarkers by birth size category

Nutrients and One-Carbon Biomarkers	Total	SGA	AGA	LGA	P^1	P^2	P^3
MDNs intake							
Daily vitamin B_2 intake (mg/d)	1.9 ± 0.1	1.9 ± 0.2	1.9 ± 0.1	1.8 ± 0.1	0.240	0.136	0.959
Daily vitamin B_6 intake (mg/d)	1.8 ± 0.1	1.7 ± 0.2	1.8 ± 0.1	1.6 ± 0.1	0.188	0.052	1.000
Daily folate intake (μg DFE/d)	994.9 ± 13.0	956.6 ± 48.0	1006.1 ± 14.7	948.9 ± 33.7	0.398	0.155	0.888
Daily vitamin B_{12} intake (μg/d)	6.5 ± 0.1	6.3 ± 0.4	6.6 ± 0.1	5.9 ± 0.2	0.091	0.026	0.908
Daily choline intake (mg/d)	278.7 ± 1.8	270.8 ± 7.2	279.2 ± 2.0	279.9 ± 4.8	0.238	0.619	0.190
Daily betaine intake (mg/d)	72.1 ± 0.7	72.2 ± 2.4	72.0 ± 0.8	72.8 ± 1.8	0.737	0.342	0.713
Daily methionine intake (mg/d)	1072.3 ± 6.8	1031.4 ± 26.9	1075.5 ± 7.6	1074.5 ± 18.2	0.081	0.933	0.122
Serum one-carbon biomarkers							
Serum vitamin B_2 (nmol/L)	15.9 (8.6, 27.4)	17.6 (11.2, 25.2)	15.8 (8.3, 27.5)	15.9 (8.8, 27.9)	0.210	0.818	0.360
Serum vitamin B_6 (nmol/L)	132.2 (115.0, 152.9)	128.2 (116.3, 151.3)	132.5 (114.8, 152.6)	131.5 (115.1, 155.4)	0.653	0.978	0.736
Serum folate (ng/mL)	3.4 (3.1, 3.7)	3.4 (3.0, 3.9)	3.4 (3.2, 3.6)	3.4 (3.2, 3.8)	0.828	0.124	0.606
Serum vitamin B_{12} (pmol/L)	569.2 (426.4, 805.3)	497.5 (389.3, 665.6)	576.6 (431.0, 815.3)	553.8 (415.8, 800.8)	< 0.001	0.223	0.016
Serum choline (μmol/L)	43.3 (17.6, 67.9)	35.2 (9.8, 59.0)	43.5 (17.7, 67.9)	47.6 (17.3, 73.8)	0.020	0.159	0.005
Serum betaine (μmol/L)	14.9 (12.6, 17.5)	14.6 (12.5, 17.4)	15.1 (12.7, 17.6)	14.4 (12.1, 16.9)	0.392	0.003	0.242
Serum methionine (μmol/L)	47.8 (42.6, 53.3)	47.6 (41.2, 52.9)	47.7 (42.6, 53.3)	48.1 (42.8, 53.3)	0.360	0.805	0.343
Serum SAM (nmol/L)	116.3 (72.2, 200.2)	107.9 (65.3, 200.0)	118.4 (75.6, 199.0)	108.7 (58.3, 213.3)	0.149	0.030	0.890
Serum SAH (nmol/L)	59.9 (39.1, 82.0)	53.1 (31.7, 64.1)	60.6 (39.5, 83.4)	61.3 (42.4, 82.4)	< 0.001	0.925	0.001
Serum homocysteine (μmol/L)	6.8 (3.8, 17.4)	18.1 (13.6, 26.0)	6.0 (3.6, 15.5)	9.2 (4.5, 18.3)	< 0.001	< 0.001	< 0.001

Abbreviation: SGA, small for gestational age; AGA, appropriate for gestational age; LGA, large for gestational age; MDN, methyl-donor nutrient; SAH, S-adenosyl homocysteine; SAM, S-adenosyl methionine.
Values are mean and SD or median and interquartile range. *P* values are from the Kruskal-Wallis test.
P^1, P value for comparison of SGA vs AGA; P^2, P value for comparison of LGA vs AGA; P^3, P value for comparison of SGA vs LGA.

The median serum concentrations of vitamins B_2, B_6, folate, B_{12}, choline, betaine, methionine, SAM, SAH, and Hcy were 15.9 nmol/L, 132.2 nmol/L, 3.4 ng/mL, 569.2 pmol/L, 43.3 μmol/L, 14.9 μmol/L, 47.8 μmol/L, 116.3 nmol/L, 59.9 nmol/L, and 6.8 μmol/L, respectively. Mothers of SGA infants had higher serum Hcy levels and lower concentrations of vitamin B_{12}, choline, and SAH compared with mothers of AGA infants. In the LGA group, serum Hcy levels were also elevated, while betaine and SAM concentrations were lower.

Serum SAM levels were negatively correlated with serum concentrations of vitamin B_6, folate, B_{12}, choline, and methionine, but positively associated with daily folate intake (**Fig. S2)**. Serum Hcy levels were inversely associated with daily methionine intake and most serum biomarkers, except for vitamin B_2, betaine, and SAM, which showed positive associations.

3.3 Association of MDN intake with the SGA and LGA risk

Higher daily methionine intake was associated with a reduced risk of SGA, with an RR of 0.46 (95% CI: 0.29-0.72, P_{trend} = 0.001) for the highest versus the lowest tertile (T3 vs. T1) (**Table 3**). Additionally, higher intakes of vitamins B_2 ($RR_{T3\ vs\ T1}$ = 0.70; 95% CI: 0.51-0.97, P_{trend} = 0.031), B_6 ($RR_{T3\ vs\ T1}$ = 0.72; 95% CI: 0.52-0.99, P_{trend} = 0.040), folate ($RR_{T3\ vs\ T1}$ = 0.68; 95% CI: 0.48-0.97, P_{trend} = 0.036) and B_{12} ($RR_{T3\ vs\ T1}$ = 0.71; 95% CI: 0.51-0.98, P_{trend} = 0.038) were negatively associated with the risk of LGA. Source-specific analyses revealed that higher food-derived vitamin B_2 intake was associated with a 39% reduced risk of SGA ($RR_{T3\ vs\ T1}$ = 0.61, 0.40-0.95) (**Fig. 1a**). An interaction was observed between the source of vitamin B_2 and SGA risk (P interaction = 0.008, **Table S2**). In contrast, the inverse associations between MDN intake and LGA risk were more pronounced for supplemented-derived intakes (**Fig. 1b**).

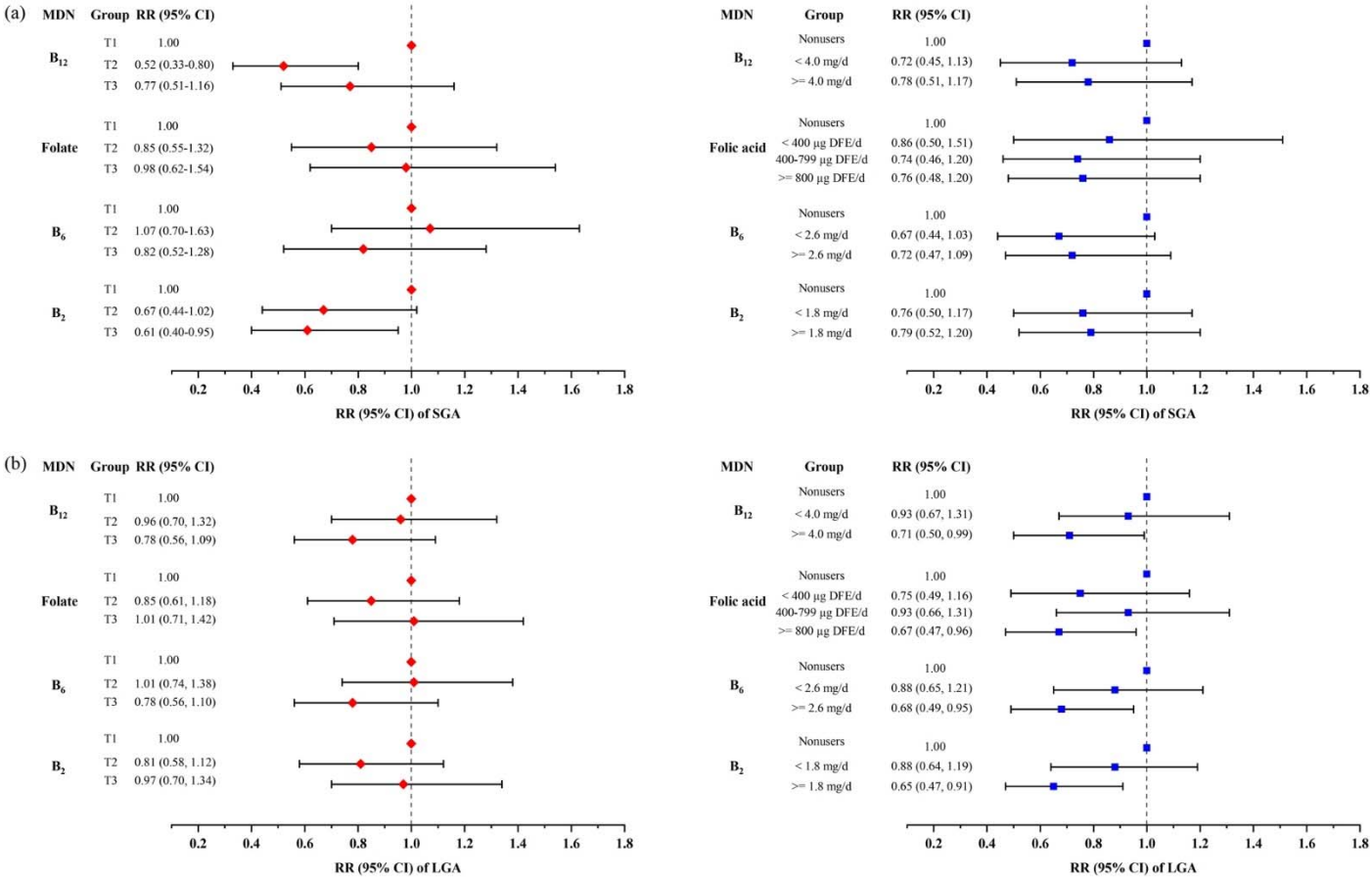

Fig. 1 Associations between maternal MDN intakes from diet (left) and supplements (right) and the risks of SGA (a) and LGA (b). Adjusted for maternal age, pre-pregnancy BMI, education (primary, secondary, college, or university), family income per capita (<5000, 5000~9999, 10000~14999, or ≥ 15000), primiparity (yes/no), smoking (yes/no), drinking (yes/no), pregnancy exercise (yes/no), gestational weight gain, gestational diabetes (yes/no), pregnancy-induced hypertension (yes/no), gestational week at data collection, corresponding MDN intake from supplements or food, energy intake, and dietary intakes of vitamin C and E. BMI, body mass index; CI, confidence interval; LGA, large for gestational age; MDN, methyl donor nutrient; RR, risk ratio; SGA, small for gestational age.

Table 3 Association between daily MDN intake and the risk of SGA and LGA

Variables	Case (*n*, %)	RR (95% CI)		
		Model 1	Model 2	Model 3
SGA				
Daily vitamin B_2 intake (mg/d)				
T1 (< 1.1)	54 (2.9)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (1.1-2.5)	49 (2.7)	0.90 (0.60, 1.35)	0.79 (0.52, 1.19)	0.80 (0.53, 1.22)
T3 (> 2.5)	44 (2.4)	0.80 (0.53, 1.21)	0.69 (0.45, 1.06)	0.70 (0.46, 1.09)
P trend		0.293	0.088	0.112
Daily vitamin B_6 intake (mg/d)				
T1 (< 0.7)	52 (2.8)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (0.7-2.6)	54 (2.9)	1.04 (0.70, 1.55)	0.91 (0.61, 1.37)	0.92 (0.61, 1.40)
T3 (> 2.6)	41 (2.2)	0.77 (0.51, 1.18)	0.68 (0.44, 1.05)	0.68 (0.44, 1.06)
P trend		0.247	0.080	0.091
Daily folate intake (μg DFE/d)				
T1 (< 612.6)	52 (2.8)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (612.6-1485.6)	49 (2.7)	0.94 (0.62, 1.41)	0.82 (0.54, 1.25)	0.82 (0.54, 1.19)
T3 (>1485.6)	46 (2.5)	0.88 (0.58, 1.32)	0.76 (0.49, 1.17)	0.77 (0.50, 1.19)
P trend		0.528	0.220	0.246
P trend (FDR)		0.616	0.257	0.287
Daily vitamin B_{12} intake (μg/d)				
T1 (< 4.3)	56 (3.1)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (4.3-7.3)	48 (2.6)	0.85 (0.57, 1.26)	0.80 (0.53, 1.21)	0.80 (0.53, 1.21)

T3 (> 7.3)	43 (2.3)	0.75 (0.50, 1.14)	0.69 (0.45, 1.06)	0.70 (0.45, 1.07)
P for trend		0.172	0.092	0.100
Daily choline intake (mg/d)				
T1 (< 251.0)	57 (3.1)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (251.0-298.9)	48 (2.6)	0.83 (0.56, 1.24)	0.79 (0.53, 1.20)	0.78 (0.52, 1.18)
T3 (> 298.9)	42 (2.3)	0.72 (0.47, 1.09)	0.66 (0.43, 1.02)	0.66 (0.43, 1.01)
P trend		0.115	0.057	0.055
Daily betaine intake (mg/d)				
T1 (< 57.5)	49 (2.7)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (57.5-80.8)	50 (2.7)	1.02 (0.68, 1.54)	1.04 (0.69, 1.58)	1.05 (0.69, 1.59)
T3 (> 80.8)	48 (2.6)	0.98 (0.65, 1.48)	0.98 (0.64, 1.50)	0.99 (0.65, 1.52)
P trend		0.916	0.940	0.973
Daily methionine intake (mg/d)				
T1 (< 983.0)	63 (3.4)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (983.0-1134.4)	48 (2.6)	0.74 (0.50, 1.10)	0.68 (0.45, 1.02)	0.65 (0.43, 0.98)
T3 (> 1134.4)	36 (2.0)	0.55 (0.36, 0.83)	0.49 (0.31, 0.76)	0.46 (0.29, 0.72)
P trend		0.005	0.001	0.001
LGA				
Daily vitamin B_2 intake (mg/d)				
T1 (< 1.1)	110 (5.6)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (1.1-2.5)	95 (4.8)	0.84 (0.62, 1.13)	0.86 (0.63, 1.17)	0.84 (0.61, 1.15)
T3 (> 2.5)	83 (4.2)	0.72 (0.53, 0.98)	0.71 (0.51, 0.98)	0.70 (0.51, 0.97)
P trend		0.035	0.039	0.031
Daily vitamin B_6 intake (mg/d)				
T1 (< 0.7)	114 (5.8)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (0.7-2.6)	87 (4.4)	0.73 (0.54, 0.98)	0.77 (0.56, 1.05)	0.75 (0.54, 1.03)
T3 (> 2.6)	87 (4.4)	0.73 (0.54, 0.98)	0.73 (0.53, 1.00)	0.72 (0.52, 0.99)
P trend		0.035	0.048	0.040
Daily folate intake (μg DFE/d)				
T1 (< 606.2)	104 (5.3)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (606.2-1450.4)	103 (5.2)	0.99 (0.74, 1.33)	1.04 (0.75, 1.44)	1.03 (0.74, 1.43)
T3 (> 1450.4)	81 (4.1)	0.75 (0.55, 1.02)	0.69 (0.48, 0.98)	0.68 (0.48, 0.97)
P trend		0.073	0.038	0.036
Daily vitamin B_{12} intake (μg/d)				
T1 (< 4.2)	108 (5.5)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (4.2-7.3)	100 (5.1)	0.91 (0.68, 1.22)	0.87 (0.64, 1.19)	0.87 (0.64, 1.19)
T3 (> 7.3)	80 (4.1)	0.70 (0.52, 0.96)	0.71 (0.51, 0.99)	0.71 (0.51, 0.98)
P trend		0.028	0.041	0.038
Daily choline intake (mg/d)				
T1 (< 251.6)	104 (5.3)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (251.6-300.1)	89 (4.5)	0.83 (0.61, 1.13)	0.93 (0.66, 1.30)	0.89 (0.65, 1.23)
T3 (> 300.1)	95 (4.8)	0.90 (0.67, 1.22)	0.69 (0.49, 0.97)	0.99 (0.72, 1.37)
P trend		0.482	0.908	0.948
Daily betaine intake (mg/d)				
T1 (< 58.6)	81 (4.1)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (58.6-81.5)	102 (5.2)	1.31 (0.96, 1.79)	1.31 (0.95, 1.82)	1.32 (0.95, 1.83)
T3 (> 81.5)	105 (5.3)	1.35 (0.99, 1.85)	1.28 (0.99, 1.77)	1.27 (0.92, 1.76)
P trend		0.061	0.150	0.159
Daily methionine intake (mg/d)				
T1 (< 984.8)	111 (5.6)	1 (ref.)	1 (ref.)	1 (ref.)
T2 (984.8-1139.23)	84 (4.3)	0.72 (0.53, 0.98)	0.72 (0.52, 0.99)	0.73 (0.53, 1.02)
T3 (>1139.23)	93 (4.7)	0.81 (0.60, 1.09)	0.84 (0.61, 1.16)	0.86 (0.62, 1.19)
P trend		0.160	0.281	0.366

Modified Poisson regression models were as follows:

Model 1: unadjusted;

Model 2: adjusted for maternal age, pre-pregnancy BMI, education (primary, secondary, college, or university), family income per capita (<5000, 5000~9999, 10000~14999, or ≥ 15000), primiparity (yes/no), smoking (yes/no), drinking (yes/no), pregnancy exercise (yes/no), gestational weight gain, gestational diabetes (yes/no), pregnancy-induced hypertension (yes/no), and gestational week at data collection;

Model 3: further adjusted for energy intake, dietary intakes of vitamin C and E.

Abbreviations: BMI, body mass index; CI, confidence interval; LGA, large for gestational age; MDN, methyl donor nutrient; RR, risk ratio; SGA, small for gestational age.

P for trend were FDR-adjusted.

Stratified analyses (**Fig. S3A-C**) showed that among females younger than 30 years, higher intakes of vitamin B_6, choline, methionine, and food-derived vitamin B_2 were associated with a lower risk of SGA. Similar inverse associations for food-derived vitamin B_2 and methionine were observed among females with a pre-pregnancy BMI < 24.0 kg/m² and those without GDM. Conversely, the negative associations between MDN intake and LGA risk were primarily observed in subgroups of females aged < 30 years, those with a pre-pregnancy BMI ≥ 24.0 kg/m², and those without GDM.

An MDN-rich dietary pattern, characterized by lower consumption of cereals and higher intake of aquatic products, eggs, and dairy (**Table S3**), was associated with a lower SGA risk ($RR_{T3\ vs\ T1}$ = 0.65, 95% CI: 0.42-0.99, *P* trend = 0.045) (**Table S4**). Conversely, LGA risk decreased progressively with increasing MDNS, with a 33% reduction in the highest tertile ($RR_{T3\ vs\ T1}$ = 0.67, 95% CI: 0.48-0.93, *P* trend = 0.019). Moreover, higher-dose use of MDN-containing supplements was associated with a 28% lower risk of LGA (**Table S5**).

3.4 Association of serum one-carbon metabolism biomarkers with SGA and LGA

Higher maternal serum levels of vitamin B_{12} ($RR_{T3\ vs\ T1}$ = 0.60, 95% CI: 0.37-0.99), choline ($RR_{T3\ vs\ T1}$ = 0.58, 95% CI: 0.34-0.98), betaine ($RR_{T3\ vs\ T1}$ = 0.64, 95% CI: 0.42-0.98) and SAH ($RR_{T3\ vs\ T1}$ = 0.37, 95% CI: 0.20-0.68) was associated with reduced risk of SGA (**Table 4**). Conversely, elevated maternal Hcy levels were strongly associated with an increased SGA risk ($RR_{T3\ vs\ T1}$ = 27.52, 95% CI: 9.61-78.82). Additionally, serum levels of betaine ($RR_{T3\ vs\ T1}$ = 0.74, 95% CI: 0.56-0.98) and SAM ($RR_{T3\ vs\ T1}$ = 0.71, 95% CI: 0.51-0.99) were inversely associated with LGA risk. RCS analyses revealed an L-shaped association between serum vitamin B_{12} and SGA risk ($P_{non\text{-}linear}$ = 0.004), an inverted U-shaped association for choline ($P_{non\text{-}linear}$ = 0.007), and a J-shaped relationship for Hcy ($P_{non\text{-}linear}$ < 0.001) (**Fig. S4A**). For LGA, an inverted U-shaped association was observed with serum choline ($P_{non\text{-}linear}$ = 0.043) (**Fig. S4B**).

Table 4 Associations between maternal serum one-carbon metabolism biomarker concentration and SGA and LGA risk during the second trimester

One-carbon metabolism biomarkers	SGA (*n*, %)	RR (95% CI)	One-carbon metabolism biomarkers	LGA (*n*, %)	RR (95% CI)
Vitamin B_2 (nmol/L)			**Vitamin B_2 (nmol/L)**		
T1 (< 10.9)	31 (5.4)	1 (ref.)	T1 (< 10.7)	77 (12.5)	1 (ref.)
T2 (10.9-22.3)	54 (9.4)	1.31 (0.85, 2.01)	T2 (10.9-22.3)	89 (14.4)	1.08 (0.82, 1.43)
T3 (> 22.3)	43 (7.5)	1.09 (0.70, 1.69)	T3 (> 22.3)	86 (13.9)	1.13 (0.85, 1.50)
Continuous		1.17 (0.95, 1.43)	Continuous		1.02 (0.90, 1.15)
P trend		0.802	*P* for trend		0.404
Vitamin B_6 (nmol/L)			**Vitamin B_6 (nmol/L)**		
T1 (< 120.4)	44 (7.6)	1 (ref.)	T1 (< 120.4)	86 (13.9)	1 (ref.)
T2 (120.4-144.4)	47 (8.2)	1.12 (0.75, 1.66)	T2 (120.4-144.6)	81 (13.1)	0.89 (0.67, 1.17)
T3 (> 144.4)	37 (6.4)	0.85 (0.55, 1.32)	T3 (> 144.6)	85 (13.8)	0.87 (0.66, 1.16)
Continuous		1.01 (0.43, 2.39)	Continuous		0.53 (0.19, 1.46)

P for trend		0.460	*P* for trend		0.357
Folate (ng/mL)			**Folate (ng/mL)**		
T1 (< 3.3)	55 (9.5)	1 (ref.)	T1 (< 3.3)	82 (13.2)	1 (ref.)
T2 (3.3-3.5)	17 (3.0)	0.43 (0.24, 0.75)	T2 (3.3-3.5)	71 (11.6)	1.17 (0.83, 1.64)
T3 (> 3.5)	56 (9.7)	0.88 (0.60, 1.30)	T3 (> 3.5)	99 (16.0)	1.23 (0.93, 1.63)
Continuous		0.43 (0.11, 1.67)	Continuous		1.14 (0.41, 3.16)
P trend		0.655	*P* for trend		0.155
Vitamin B_{12} (pmol/L)			**Vitamin B_{12} (pmol/L)**		
T1 (< 465.7)	55 (9.5)	1 (ref.)	T1 (< 466.5)	95 (15.4)	1 (ref.)
T2 (465.7-706.7)	47 (8.2)	0.87 (0.59, 1.27)	T2 (466.5-712.5)	77 (12.5)	0.87 (0.65, 1.16)
T3 (> 706.7)	26 (4.5)	0.60 (0.37, 0.99)	T3 (> 712.5)	80 (12.9)	0.98 (0.72, 1.33)
Continuous		0.71 (0.61, 0.83)	Continuous		0.99 (0.85, 1.16)
P trend		0.042	*P* for trend		0.938
Choline (μmol/L)			**Choline (μmol/L)**		
T1 (< 28.8)	51 (8.9)	1 (ref.)	T1 (< 29.6)	82 (13.3)	1 (ref.)
T2 (28.8-58.2)	45 (7.8)	0.81 (0.51, 1.28)	T2 (29.6-59.7)	80 (12.9)	1.20 (0.83, 1.74)
T3 (> 58.2)	32 (5.5)	0.58 (0.34, 0.98)	T3 (> 59.7)	90 (14.6)	1.22 (0.85, 1.76)
Continuous		0.53 (0.27, 1.04)	Continuous		1.24 (0.86, 1.78)
P trend		0.036	*P* for trend		0.342
Betaine (μmol/L)			**Betaine (μmol/L)**		
T1 (< 13.5)	47 (8.2)	1 (ref.)	T1 (< 13.5)	99 (16.0)	1 (ref.)
T2 (13.5-16.7)	40 (6.9)	0.73 (0.50, 1.07)	T2 (13.5-16.6)	85 (13.8)	0.86 (0.66, 1.12)
T3 (> 16.7)	41 (7.1)	0.64 (0.42, 0.98)	T3 (> 16.6)	68 (11.0)	0.74 (0.56, 0.98)
Continuous		0.17 (0.04, 0.83)	Continuous		0.27 (0.10, 0.70)
P trend		0.040	*P* for trend		0.037
Methionine (μmol/L)			**Methionine (μmol/L)**		
T1 (< 44.4)	46 (8.0)	1 (ref.)	T1 (< 44.5)	80 (12.9)	1 (ref.)
T2 (44.4-51.3)	45 (7.8)	1.09 (0.73, 1.62)	T2 (44.5-51.3)	91 (14.7)	1.12 (0.84, 1.48)
T3 (> 51.3)	37 (6.4)	0.95 (0.63, 1.44)	T3 (> 51.3)	81 (13.1)	1.00 (0.75, 1.33)
Continuous		0.60 (0.17, 2.15)	Continuous		1.23 (0.48, 3.14)
P trend		0.818	*P* for trend		0.996
SAM (nmol/L)			**SAM (nmol/L)**		
T1 (< 87.3)	54 (9.4)	1 (ref.)	T1 (< 86.2)	103 (16.7)	1 (ref.)
T2 (87.3-169.1)	31 (5.4)	0.74 (0.48, 1.14)	T2 (86.2-167.1)	73 (11.8)	0.84 (0.63, 1.10)
T3 (> 169.1)	43 (7.5)	0.86 (0.53, 1.40)	T3 (> 167.1)	76 (12.3)	0.71 (0.51, 0.99)
Continuous		0.95 (0.82, 1.11)	Continuous		0.90 (0.82, 0.99)
P trend		0.433	*P* for trend		0.035
SAH (nmol/L)			**SAH (nmol/L)**		
T1 (< 47.9)	51 (8.9)	1 (ref.)	T1 (< 48.7)	80 (12.9)	1 (ref.)
T2 (47.9-72.6)	58 (10.1)	0.93 (0.58, 1.50)	T2 (48.7-73.8)	90 (14.6)	1.13 (0.80, 1.61)
T3 (> 72.6)	19 (3.3)	0.37 (0.20, 0.68)	T3 (> 73.8)	82 (13.3)	1.11 (0.77, 1.61)
Continuous		0.64 (0.51, 0.80)	Continuous		0.94 (0.65, 1.35)
P trend		0.001	*P* for trend		0.647
Hcy (μmol/L)			**Hcy (μmol/L)**		
T1 (< 4.5)	4 (0.7)	1 (ref.)	T1 (< 4.3)	62 (10.0)	1 (ref.)
T2 (4.5-12.5)	21 (3.6)	5.78 (1.99, 16.83)	T2 (4.5-11.1)	75 (12.2)	1.07 (0.76, 1.49)
T3 (> 12.5)	103 (17.9)	27.52 (9.61, 78.82)	T3 (> 11.1)	115 (18.6)	1.26 (0.85, 1.89)
Continuous		11.03 (5.94, 20.47)	Continuous		1.20 (0.80, 1.79)
P trend		< 0.001	*P* for trend		0.246

Modified Poisson regression model was adjusted for maternal age, pre-pregnancy BMI, education (primary, secondary, college, or university), family income per capita (<5000, 5000~9999, 10000~14999, or ≥ 15000), primiparity (yes/no), smoking (yes/no), drinking (yes/no), pregnancy exercise (yes/no), gestational weight gain, gestational diabetes (yes/no), pregnancy-induced hypertension (yes/no), and gestational week.

Continuous, log-transformed concentrations of serum one-carbon metabolism biomarkers were used.

Abbreviations: BMI, body mass index; CI, confidence interval; Hcy, homocysteine; LGA, large for gestational age; RR, risk ratio; SAH, S-adenosyl homocysteine; SAM, S-adenosyl methionine; SGA, small for gestational age.

P for trend were FDR-adjusted.

Stratified analyses showed that the inverse associations of serum vitamin B_{12}, choline, betaine, and SAH with SGA remained among females aged < 30 years, those with a pre-pregnancy BMI < 24.0 kg/m^2, and those

without GDM (**Fig. S5A-C[a]**). The adverse effect of elevated Hcy on SGA was more pronounced in females with a BMI < 24.0 kg/m^2 or without GDM, but was not modified by maternal age. In contrast, the protective associations of higher serum betaine and SAM levels against LGA were more evident in females with pre-pregnancy BMI ≥ 24.0 kg/m^2 or with GDM (**Fig. S5A-C[b]**).

The WQS index was inversely associated with both SGA ($RR_{T3\ vs\ T1}$ = 0.29, 95% CI: 0.16-0.52, P_{trend} < 0.001) and LGA risk ($RR_{T3\ vs\ T1}$ = 0.73, 95% CI: 0.55-0.98, P_{trend} = 0.034) (**Fig. 2**). In the WQS model for SGA, the top three contributors were SAM (30.8%), SAH (28.1%), and choline (20.6%), while Hcy contributed minimally (< 0.01%). For LGA, the most influential biomarkers were betaine (33.0%), SAM (22.0%), and SAH (12.9%), with Hcy again showing the least contribution (0.07%).

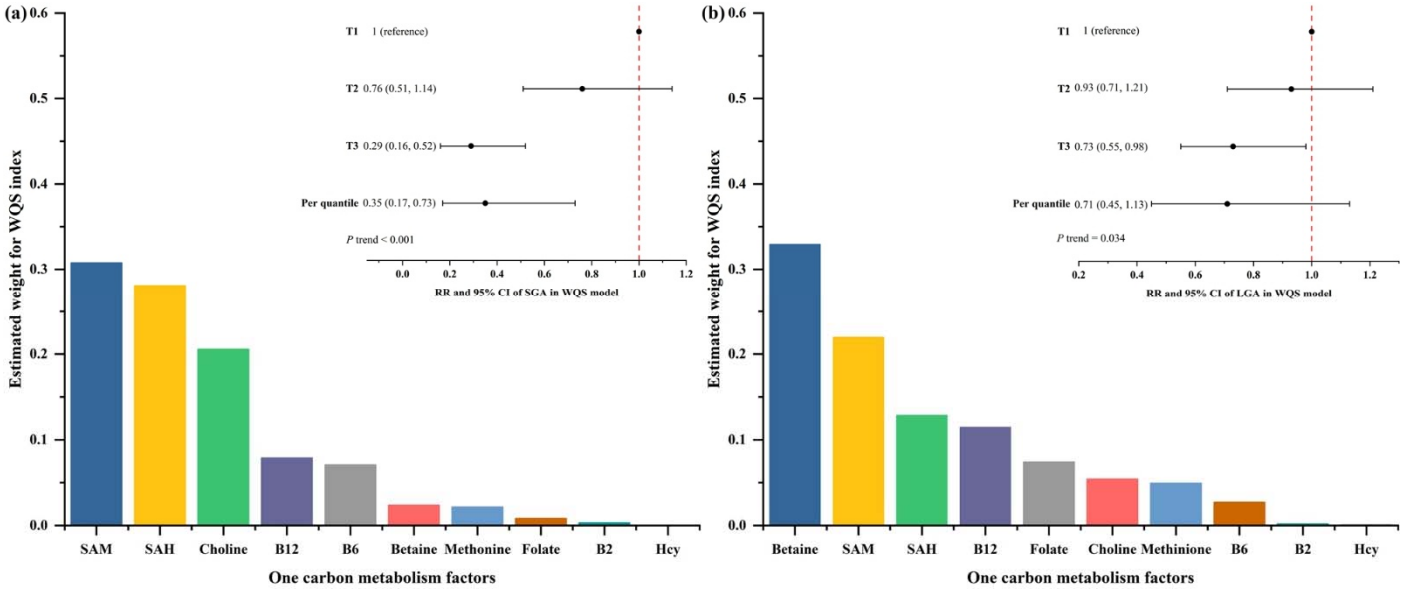

Fig. 2 Weighted values of maternal serum one-carbon metabolism biomarkers for SGA (a) and LGA (b) risks in the WQS models. Adjusted for maternal age, pre-pregnancy BMI, education (primary, secondary, college, or university), family income per capita (<5000, 5000~9999, 10000~14999, or ≥ 15000), primiparity (yes/no), smoking (yes/no), drinking (yes/no), pregnancy exercise (yes/no), gestational weight gain, gestational diabetes (yes/no), pregnancy-induced hypertension (yes/no), and gestational week. BMI, body mass index; CI, confidence interval; LGA, large for gestational age; RR, risk ratio; Hcy, homocysteine; SAH, S-adenosyl homocysteine; SAM, S-adenosyl methionine; SGA, small for gestational age; WQS, weighted quantile sum.

4. Discussion

This study found that MDN intake during pregnancy was generally insufficient among the Chinese population. We observed that higher intakes of food-derived MDNs were associated with a reduced risk of SGA. In contrast, supplement-derived MDNs showed stronger inverse associations with LGA. Biomarker analyses indicated that elevated maternal serum levels of one-carbon metabolites were protective against SGA, except Hcy. Additionally, maternal serum concentrations of betaine and SAM were inversely associated with the risk of LGA. These findings were further supported by WQS models, highlighting a collective protective effect of one-carbon metabolism biomarkers on the risks of both SGA and LGA.

Our research also found multiple factors that contribute to fetal growth abnormalities. In line with previous research, pre-pregnancy BMI, gestational weight gain, and parity are correlated with higher newborn weights [26]. Elderly mothers, particularly those with GDM, have a greater likelihood of giving LGA newborns

[27]. This study investigated the association between the maternal MDNs status and the risk of SGA and LGA, independent of the aforementioned factors.

Our previous research found that higher vitamin B_2 intakes during pregnancy were associated with a reduced risk of GDM, suggesting potential implications for fetal development [28]. In the current study, both dietary and circulating levels of vitamin B_2 were inversely associated with SGA risk. Aligning with the negative association between maternal vitamin B_6 status and preterm birth, we associated elevated serum vitamin B_6 concentrations and a lower risk of SGA [29]. Folate, a key MDN, has been widely recognized for its role in fetal development [16, 30]. We found that higher maternal serum folate was associated with lower SGA risk, particularly among women with lower pre-pregnancy BMI, who often have reduced folate stores [31]. However, the relatively high average folate intake in our cohort (967.3 μg DFE/ day) may limit our ability to detect stronger associations. The lack of a significant correlation between serum folate levels and folate intake observed in this study may be attributed to several factors. First, while dietary folate assessment reflects long-term habitual intake, serum folate concentrations primarily indicate recent intake and are particularly sensitive to short-term dietary fluctuations, supplement consumption, or fasting status. Second, serum folate levels are modulated by genetic determinants (e.g., MTHFR genotype polymorphisms) [32], which may obscure the true relationship with dietary intake. Third, although this study evaluated total folate intake encompassing both food-derived and supplemental sources, the serum analysis was limited to the detection of unmetabolized folate, potentially overlooking other biologically relevant folate forms. Consistent with meta-analyses linking vitamin B_{12} deficiency to low birth weight and preterm birth [14], we observed an inverse association between serum vitamin B_{12} and SGA risk. Our findings also support evidence from clinical trials showing that choline supplementation enhances placental function and fetal growth [33], as higher serum choline levels were associated with reduced SGA risk. Elevated Hcy emerged as a risk factor for SGA, which is biologically plausible given its known role in endothelial dysfunction and placental insufficiency [13]. Notably, serum betaine was inversely associated with both SGA and LGA, reflecting its role as an alternative methyl donor that supports impaired folate-dependent remethylation [34]. Additionally, the associations of SAM and SAH with fetal growth outcomes underscore the importance of maintaining methylation balance during pregnancy [35, 36].

The observed associations between maternal MDN status and LGA risk represent an important and understudied aspect of fetal growth regulation. While one previous study suggested potential adverse effects of continued folic acid supplementation on LGA risk [37], our findings indicate protective associations for multiple MDNs. This discrepancy may reflect differences in study populations or assessment methods, highlighting the need for further research in this area.

Several biological mechanisms may explain our findings. MDNs contribute to fetal growth regulation through: (1) serving as essential cofactors and substrates for DNA methylation and epigenetic regulation [10]; (2) maintaining redox balance and reducing oxidative stress in the placenta [13]; and (3) supporting maternal

metabolic homeostasis during pregnancy [38, 39]. The differential effects we observed for specific nutrients and metabolites underscore the complexity of one-carbon metabolism in fetal development.

Our findings indicate that fmaternal intake of MDNs from foods and supplements may be differentially associated with SGA and LGA. Although it remains uncertain whether these sources act through distinct biological pathways, several possibilities can be considered. First, nutrients from foods are embedded in complex dietary matrices, which may facilitate long-term improvements in overall maternal nutritional status, thereby reducing the risk of growth restriction [40]. In contrast, supplements provide isolated and readily absorbable forms of vitamins at higher doses, which may exert a more immediate effect on one-carbon metabolism, potentially preventing excessive fetal growth. Second, SGA is often related to maternal undernutrition or impaired placental function, conditions more strongly reflected by habitual dietary intake [40]. LGA, however, is frequently associated with maternal metabolic status, such as energy excess and altered glucose-insulin homeostasis, which may be mitigated by adequate micronutrient supplementation [38]. Finally, our multivariable models adjusted for AHEI-P [22], suggesting that the observed associations were not merely confounded by general dietary quality. Together, these findings suggest that dietary and supplemental MDNs may influence fetal growth through overlapping but not identical mechanisms.

This study has several notable strengths. Firstly, we conducted a comprehensive assessment of the associations between maternal MDN intake and serum one-carbon metabolism biomarkers with neonatal birth weight, using a multi-layered analytical approach encompassing nutrient-specific effects, dietary patterns, biomarker levels, and their combined impact. Secondly, the prospective observational design allowed us to examine the temporal relationship between prenatal exposures and birth outcomes in a real-world population setting.

Several limitations of this study warrant consideration. First, the single-time-point measurement may not adequately reflect chronic exposure status, as serum levels fluctuate with recent intake and metabolic turnover. While we adjusted for key covariates, residual confounding from unmeasured factors (e.g., paternal genetic contributions to fetal development, environmental exposures) could persist. Furthermore, the research results we have obtained on the Chinese population have certain limitations of generalization, and more studies are needed to verify these findings in other populations with different genetic backgrounds or dietary habits.

5. Conclusion

In conclusion, our study demonstrates significant associations between maternal MDN status and both SGA and LGA risk, consistent with the established role of one-carbon metabolism in fetal development. These findings advance our understanding of nutritional influences on fetal growth and may inform more comprehensive approaches to prenatal nutrition assessment, while mechanistic interpretations await further investigation.

Ethics approval

All participants provided written informed consent before participation. The study was conducted under the Declaration of Helsinki and was approved by the Ethics Review Committee of Tongji Medical College, Huazhong University of Science and Technology (Ethical approval code: 2021IEC[S044]).

Data availability

All the data that support the findings of this study are available from the corresponding author upon reasonable request.

Use of Artificial Intelligence (AI) tools

AI tools (Grammarly) were used solely for improving readability and grammar. The authors reviewed and edited the content as needed and take full responsibility for the final manuscript.

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Conflict of interest

None declared.

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