Concentrations of Antioxidant Vitamins in Maternal and Cord Serum and Their Effect on Birth Outcomes

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Summary Background: Emerging evidence indicates that maternal oxidative stress during pregnancy could impair fetal growth and that antioxidant vitamins (e.g. vitamins A, E and C) have a significant role in maintaining physiological processes of pregnancy and growth. Aims: To determine the concentrations of vitamins A, E, and C in pair-matched maternal and cord serum samples of neonate, and thus to investigate the relationship between maternal serum levels of these vitamins at delivery and birth outcomes. Methods: A total of 143 mother-neonate pairs were recruited into the cross-sectional descriptive study. Demographic information was investigated by questionnaire. After delivery, both cord and maternal blood were collected for quantification of serum levels of vitamins A, E and C by HPLC. Results: Maternal serum levels of vitamins A and E were significantly higher than those in cord serum. In contrast, vitamin C level in cord serum was significantly higher than that in maternal serum. Further, we found that maternal vitamin A status was significantly correlated to both birth weight ($r=0.19$, $p=0.0419$) and birth height ($r=0.21$, $p=0.0311$), and these were manifested by these findings: (i) per 250.2 g reduction in birth weight concomitant with 1 $\mu\text{mol/L}$ increase in maternal serum vitamin A level ($p<0.01$; 95% CI: 56.9–451.5); and (ii) per 1% increase in the ratio of serum vitamin A level of neonate to mother concomitant with 0.8 cm increase in birth height ($p=0.049$; 95% CI: 0.004–1.639). Conclusion: Maternal vitamin A, but not vitamins E and C, during pregnancy had a significant effect on birth outcomes. Further studies are necessary to investigate the role of these antioxidant vitamins in fetal growth at various gestation stages.

Key Words antioxidant vitamins, maternal, cord blood, fetal growth

Previous studies have shown that maternal stress during pregnancy plays an important role in the development of chronic diseases in adulthood (1, 2). Events, especially nutritional alterations, occurring in the uterus obviously affect fetal development and growth, and as well as increase the incidence of disorders in adulthood (3, 4). Scholl and Stein (5) and Kim et al. (6) have reported that maternal oxidative stress during pregnancy contributes to low birth outcomes and antioxidant vitamins, such as vitamins A, E and C, may have an important role in fetal growth.

There are complicated antioxidant defense systems, including both enzymatic and non-enzymatic components, against the damage caused by oxygen free radicals on proteins, nucleic acids, carbohydrates, and lipids (7). Vitamins A, E and C are non-enzymatic antioxidants. Deficiency of vitamin A during pregnancy is associated with a high risk of xerophthalmia and blind-

ness, impairment of cellular function, growth failure, anemia, depressed immunity, and increased morbidity and mortality of infectious diseases (8, 9). Vitamin A is known to be an important natural antioxidant capable of counteracting oxygen free radicals and exerts a protective antioxidant effect (10). Vitamin C, a hydrophilic antioxidant, may protect the fetus from insults resulting from oxygen free radicals by scavenging hydroxyl radicals (11, 12). Vitamin E, a lipophilic antioxidant, appears to be the most important micronutrient involved in the protection of low-density lipoproteins (LDLs) from oxidation (13). Previous reports have shown that serum contents of fat-soluble nutrients in newborns have been shown to be much lower than those in mothers (14); but in contrast, serum contents of hydrophilic nutrients in newborns have been higher than those in mothers (15).

It remains unknown whether the serum contents of vitamins A, E and C are different in newborn and mother. In addition, whether alterations of antioxidant vitamins can affect birth outcomes is still controversial (16–20). Thus, in the present study, serum levels of

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vitamins A, E and C both in the blood of mother and her cord were determined and the relationship between serum levels of antioxidant vitamins in maternal and cord blood and birth outcomes was studied.

The aims of the present study were (i) to determine serum concentrations of vitamins A, E, and C in pair-matched maternal and cord blood, including full-term and preterm infants, and examine the possible relationship in antioxidant vitamin levels between maternal and cord blood; and (ii) to investigate the relationship between maternal serum vitamin levels and birth outcomes.

MATERIALS AND METHODS

Ethical approval. This was a cross-sectional descriptive study. The research protocol was reviewed and approved by the institutional ethical committee of the Third People's Hospital in Changzhou City, Jiangsu Province, China. One of the authors and several assistants explained the objectives and procedures of the study to the selected pregnant women. Informed written consent was obtained from subjects. Participation was voluntary, and volunteers could withdraw from the study at any time.

Subjects. Subjects were 160 neonates born to healthy Chinese women who delivered at the Department of Gynaecology and Obstetrics in the Third People's Hospital during the period from 1 March, 2007 to 1 June, 2007. Only healthy mothers and their infants were admitted into the study. Exclusion criteria were pregnancy toxemia, hypertension, diabetes mellitus, thyroid disease, bronchial asthma, active hepatitis, chronic renal failure and heart failure. The sample size was estimated based on a confidence interval of 95%, at a desired accuracy of 0.05 with an assumed 30% prevalence of vitamin A deficiency.

Personal interview. A questionnaire of duration of about 45 min was given by a trained interviewer after delivery. The questionnaire elicited demographic information, lifetime residential history (location of birth and duration of residence), history of active and passive smoking (including number of household members who smoke), occupational exposure, medication information, and alcohol use during each trimester of pregnancy. Socioeconomic information related to maternal age, educational level, height, weight before pregnancy, and income level was also collected. Gestational age was estimated based on the maternal report for the last menstrual period and on ultrasound measurement by obstetricians. Preterm birth was defined as gestational period <37 wk.

Anthropometric measurements. Trained nurses in the delivery room routinely measured birth weight, birth length and head circumference immediately after parturition and recorded this information on medical charts. Birth weight was measured to the nearest 50 g using a digital scale, and crown-heel length and head circumference were recorded to the nearest 0.1 cm with a non-stretch plastic tape measure. The data for birth weight, birth length and head circumference in the

present study were obtained from medical charts. Scales were checked daily against standard metal weights and calibrated if necessary.

Information was abstracted by research workers from the medical records of the mother and infant after delivery. These included date of delivery, gestational age at birth, sex of neonate, birth weight, birth length, head circumference, malformations, maternal height, prepregnancy weight, complications of pregnancy and delivery, as well as medications used during pregnancy.

Collection of biological sample and analysis. Maternal blood (10 mL) was collected within 1 d postpartum. Umbilical cord blood (40–60 mL) was collected at delivery. Samples were transported to the field laboratory at the Third People's Hospital immediately after collection and processed there. For blood samples, the buffy coat, packed red blood cells and plasma were separated and stored at -70°C . The serum samples prepared for measurement of vitamin A were protected from light.

Serum concentrations of vitamin A (retinol) and vitamin E (α -tocopherol) were determined using high-performance liquid chromatography (HPLC) according to the previous method (21, 22) with minor modification. Briefly, vitamin A was extracted with hexane after deproteinization with ethanol containing retinyl acetate (the external standard), and then evaporated to dryness with nitrogen gas. The residue was dissolved in 0.1 mL methanol. Twenty microliters of sample was injected into the column (Symmetry Shield RP₁₈ 3.9×150 mm) installed with the HPLC apparatus (Waters 1525 Binary HPLC Pump, Waters Breeze, USA). The mobile phase was a methanol-DH₂O (distilled water) mixture (97:3, for α -tocopherol: 98:2). The concentration of vitamin A was determined by spectrophotometry (Waters 2487 Dual λ Absorbance Detector, USA) at 315 nm (for vitamin E: 280 nm). Serum concentration of vitamin C was measured by HPLC according to the previous method (23, 24) using 100 mmol/L potassium dihydrogen phosphates as the mobile phase at pH 3.5 by phosphoric acid, and the detected absorbance λ was 254 nm. Procedures were carried out in a dark room to protect the serum from the light.

Duplicate analyses for serum vitamin A were performed on one-tenth of the samples and the estimated variability was 0.015 $\mu\text{mol/L}$. Three control samples with low (0.70 $\mu\text{mol/L}$), medium (1.40 $\mu\text{mol/L}$) and high (2.80 $\mu\text{mol/L}$) concentrations of serum vitamin A were provided by vitamin A standard solution (Sigma, USA) with pooled serum. The between-day CVs for low, medium and high concentrations of serum vitamin A were 6.21, 4.55 and 3.19%, respectively. The between-day CVs for serum vitamin E were 3.7, 3.1 and 2.3% at 10, 20 and 40 $\mu\text{mol/L}$, respectively, and for serum vitamin C were 4.2, 3.8 and 1.9% at 30, 60 and 120 $\mu\text{mol/L}$, respectively.

Biochemical indices were measured by expert examiners in the clinical laboratory of the Third People's Hospital.

Statistical analysis. Using the Kormogov-Smirnov goodness-of-fit test, the distributions of each set of data

were tested for normality before analysis. If necessary, data were normalized using natural-log transformations. The concentration of each antioxidant vitamin determined in this study was expressed as the mean $\pm$ SD (standard deviation). Tests of significance were two-tailed and $p < 0.05$ was considered statistically significant. Paired Student's *t*-tests were used to compare paired mother-neonatal vitamin A, E and C data with normal distribution and homogeneous variance. Paired Wilcoxon sign-rank test was used for data that were not normally distributed. Correlations between maternal and cord serum nutrient concentrations were measured by linear-regression analysis using Pearson correlation coefficients (normal distribution data) or Spearman correlation coefficients (abnormal distribution data) which describes the simple correlation between dependent variable and independent variable. When discussing the correlation between variables and birth outcomes, the Spearman correlation partial coefficient was used, which describes the behavior of dependent variable and independent variable 1 when independent variable 2, 3, ... n are held fixed and the partial correlation between dependent variable and independent variable 1 holding independent variable 2, 3, ... n fixed is denoted.

The following covariates were examined: sex of neonate (1: male, 2: female), gestational age (d), educational level (1: <High school, 2: $\geq$ High school), parity (1: no parity, 2: 1 parity, 3: 2 parities, 4: 3 parities, 5: 4 parities before), exposure to passive smoking (1: yes, 2: no), maternal intake of vitamins (1: yes, 2: no), maternal age at first hospital visit (years), reported pre-conceptional weight (kg), maternal height (m), maternal head circumference (cm), and body mass index (BMI; as kg/m²) before conception.

Simple linear regression analysis was used to identify the independent variables that were statistically associated with birth outcomes (birth weight, length and head circumference). Multivariate regression models were used to estimate the effects of independent variables on birth outcomes. Gestational age, neonate sex, parity, cord and maternal retinol were found by univariate analysis and simple regression models to significantly influence birth weight. They were included in the multivariate regression models and the R^2 value was

used as an indicator of how well the model fitted the data. Qualitative variables were treated as dummy variables in the multivariate statistical analysis. Variance inflation factor (VIF, a method of detecting the severity of multi-collineation of every independent variable) was calculated, and the variable with >10 VIF was excluded from these models. To avoid instability in the analysis models, only mother-neonate paired cases with valid data for all of the variables (no missing values) were included.

RESULTS

A total of 160 mother-neonate pairs were recruited during the period from 1 March, 2007 to 1 June, 2007. Complete epidemiologic and clinical data were obtained from 143 pairs. The demographic and clinical characteristics of 143 mother-neonate pairs from this cross-sectional study are shown in Table 1.

Status of maternal vitamins A, E and C

Maternal vitamin A concentration was 1.13 ± 0.371

Table 1. Demographic and clinical characteristic of 143 mother-neonate pairs.

Characteristic	Value		
	Mean	SD ¹	Median
Mothers			
Age (y)	24.6	3.76	24.0
Height (cm)	159.2	3.77	157.5
Weight before pregnancy (kg)	50.16	5.25	49.0
BMI before pregnancy (kg/m ²)	19.2	2.11	19.0
Educational level			
<High school [n (%)]		75 (52.4)	
$\geq$ High school [n (%)]		68 (47.6)	
Passive smoking exposure [n (%)]		80 (55.9)	
Vitamin intake [n (%)]		34 (23.8)	
Neonates			
Gestational age (d)	276.5	11.33	279.0
Birth length (cm)	50.8	1.77	50.0
Birth weight (g)	3317.4	400.8	3461.0
Birth head circumference (cm)	33.5	1.15	33.8
Gender of neonate [n (%) female]		74 (51.7)	

¹ SD standard deviation.

Table 2. Concentrations of vitamins A, E and C in maternal and cord plasma.

Nutrients	Maternal plasma (μ mol/L)			Cord plasma (μ mol/L)			p -value ²	Mother (%)		
	Mean	SD ¹	Median	Mean	SD ¹	Median		Mean	SD ¹	Median
A	1.13	0.371	1.00	0.69	0.162	0.72	$<0.0001^3$	78.3	35.2	71.5
E	25.7	10.38	19.8	15.9	11.08	13.9	$<0.0001^4$	90.8	66.1	80.1
C	79.1	32.65	70.9	101.3	38.91	93.1	$<0.0001^3$	161.2	98.1	149.2

¹ SD: standard deviation.

² Comparison of nutrient concentration between maternal and cord plasma.

³ Paired Student's *t*-test.

⁴ Paired Wilcoxon sign-rank test.

$\mu\text{mol/L}$ (Table 2), and 24 of 143 mothers (16.80%) had vitamin A deficient status (serum vitamin A $<0.7 \mu\text{mol/L}$), while another 52 mothers (36.4%) had marginal vitamin A deficient status (serum vitamin A: $0.7\text{--}1.05 \mu\text{mol/L}$). Associations between maternal vitamin A concentration and maternal/environmental factors were detected. We found that maternal vitamin A concentration had significantly positive correlation with gestational days (Spearman correlation coefficient: 0.21 , $p=0.033$). Maternal vitamin A concentration in women without passive smoking was markedly higher than that in women with passive smoking ($1.09 \pm 0.313 \mu\text{mol/L}$ vs. $0.91 \pm 0.364 \mu\text{mol/L}$, $p=0.0117$). There was no significant correlation between maternal vitamin A status and maternal educational level, BMI before pregnancy or parity.

Maternal concentration of vitamin E and vitamin C were 25.7 ± 10.38 and $79.1 \pm 32.65 \mu\text{mol/L}$, respectively. Our data showed that maternal vitamin C concentration had significantly positive correlation with gestational days (Spearman correlation coefficient: 0.29 , $p<0.01$). Not maternal educational level, nor parity, nor passive smoking exposure, nor BMI had any significant effect on maternal vitamin E or vitamin C status.

Status of neonatal vitamins A, E and C

Cord serum vitamin A concentration was $0.69 \pm 0.162 \mu\text{mol/L}$ (Table 2), and 59 of 143 neonates (41.3%) had serum vitamin A $<0.7 \mu\text{mol/L}$ status, while 81 of 143 neonates (56.6%) had $0.7\text{--}1.05 \mu\text{mol/L}$ serum vitamin A status. Our data showed that cord vitamin A concentration had significant positive correlation with gestational days (Spearman correlation coefficient: 0.18 , $p=0.0437$). Vitamin A concentration of female neonates was markedly higher than that of male neonates ($0.77 \pm 0.138 \mu\text{mol/L}$ vs. $0.62 \pm 0.167 \mu\text{mol/L}$, $p=0.0013$). There was no significant correlation between neonate vitamin A status and maternal educational level, BMI before pregnancy, parity or maternal exposure status to passive smoking.

Cord serum concentration of vitamin E and vitamin C were 15.9 ± 11.08 and $101.3 \pm 38.91 \mu\text{mol/L}$, respectively. Maternal educational level, parity, passive smoking exposure, BMI, gestational age, and neonatal sex all had no significant effect on cord serum vitamin E or vitamin C status.

Relationship between vitamins A, E and C in maternal and cord blood

Newborns had significantly lower concentrations of vitamin A and vitamin E than their mothers (both $p<0.0001$) (Table 2), but no significant correlation between cord and maternal vitamin A or vitamin E was observed. Cord serum levels of vitamin C were higher than that in maternal serum ($p<0.0001$), and no significant correlation was found between the cord and maternal vitamin C levels.

Placental transfer of the different antioxidant vitamins was calculated as the ratio of the concentrations in cord to maternal serum. The average ratios were 78.3, 90.8 and 161.2% for vitamin A, vitamin E and vitamin C, respectively (Table 2).

Simple correlation analysis showed that only gestational age had a significantly negative association with the ratio of neonate to maternal vitamin E concentration (Spearman correlation coefficient: -0.21 , $p=0.0334$). Other maternal factors had no significant correlation with the difference between neonate and mother or with the ratios of neonate to maternal vitamin concentration.

Variables associated with birth outcomes

Spearman partial correlation analysis showed that after adjustment for gestational age, neonate sex, cord vitamins A, E, and C status and some maternal factors, there was positive correlation between maternal vitamin A status and birth weight (Spearman partial correlation coefficient $r=0.19$, $p=0.0419$) and birth height ($r=0.21$, $p=0.0311$). Positive correlation was found between maternal vitamin A status and birth head circumference ($r=0.1$), but it was not significant ($p=0.3274$). No significant association was found between either maternal vitamin E or vitamin C status and birth outcomes.

Initially, we investigated possible factors related to birth outcomes (indicated by birth weight, birth height and neonate head circumference) by simple linear regression model analysis. We also used a multivariate model, in which the dependent variables were birth weight, birth height and neonate head circumference, to investigate an independent relationship between these variables and birth-outcomes indicators (Tables 3, 4 and 5).

Independent variables in the multiple linear regres-

Table 3. Independent variables in relation to birth weight.

Independent variables	Regression coefficient (β)		95% CI, β	p-value
	Parameter estimate	Standardized estimate		
Gestational age (d)	10.15648	0.28047	(-1202.36884, 2239.77132)	0.0012
Maternal vitamin A status ($\mu\text{mol/L}$)	250.21128	0.22305	(56.90091, 451.52164)	0.0230
Neonate vitamin A status ($\mu\text{mol/L}$)	381.75402	-0.16057	(-56.04255, 819.55059)	0.0868
Neonate vitamin E status ($\mu\text{mol/L}$)	-7.73267	-0.23014	(-13.87741, -1.58820)	0.0197
Parity	59.19052	0.13329	(-17.23203, 135.61307)	0.1277

$R^2=0.21$ (Adjusted $R^2=0.17$).

Table 4. Independent variables in relation to head circumference.

Independent variables	Regression coefficient (β)		95% CI, β	p-value
	Parameter estimate	Standardized estimate		
Gestational age (d)	0.02299	0.22867	(0.00655, 0.03942)	0.0066
Neonate's sex	-0.41048	-0.18294	(-0.77130, -0.04966)	0.0262
Vitamin E ratio of neonate to mother (%)	-0.24137	-0.14383	(-0.52129, 0.03856)	0.0903
Neonate vitamin C status ($\mu\text{mol/L}$)	-0.00691	-0.23654	(-0.01179, -0.00202)	0.0060
Head circumference of mother (cm)	0.26325	0.29887	(0.11812, 0.40838)	0.0005
BMI before pregnancy	0.10397	0.20054	(0.02037, 0.18758)	0.0153

$R^2=0.33$ (Adjusted $R^2=0.27$).

Table 5. Independent variables in relation to birth height.

Independent variables	Regression coefficient (β)		95% CI, β	p-value
	Parameter estimate	Standardized estimate		
Gestational age (d)	0.02294	0.14563	(-0.00487, 0.05076)	0.1050
Neonate sex	-0.89457	-0.25437	(-1.52164, -0.28332)	0.0045
Retinol ratio of neonate to mother (%)	0.82152	0.17721	(0.00383, 1.63920)	0.0490
Maternal head circumference (cm)	0.19651	0.14235	(-0.04688, 0.43991)	0.1124
Maternal vitamin intake	-0.78556	-0.1918	(-1.50788, -0.06323)	0.0333

$R^2=0.18$ (Adjusted $R^2=0.14$).

sion equation of birth weight were shown in Table 3. All independent variables together (R^2) explained 21% of the variability in birth weight (adjusted $R^2=17\%$). Gestational age, maternal vitamin A status and neonate vitamin E status had a significant effect on birth weight ($p=0.0012$, 0.023 and 0.0197, respectively). The model predicted that a 1 $\mu\text{mol/L}$ increase in maternal serum vitamin A level was concomitant with a 250.2 g increase in birth weight adjusted for gestational age, neonate vitamins E and A, and parity.

All independent variables together (R^2) explained 33% of the variability in neonate head circumference (adjusted $R^2=27\%$) (Table 4). But the neonate head circumference was significantly influenced by gestational age, neonate sex, neonate vitamin C status, maternal head circumference and BMI before pregnancy. Independent variables in regression equation of birth length explained 18% (R^2) of the variability (adjusted $R^2=14\%$) (Table 5). Only neonate sex, vitamin A ratio of neonate to mother and vitamin intake of the mother had marked effects on birth height ($p<0.05$).

DISCUSSION

Relationship between maternal and cord vitamins A, E and C

Extra vitamin A is required during pregnancy for the fetus growth, and for maternal tissue and fetal stores of vitamin A (25). Requirements are highest during the third trimester when fetal growth is most rapid. In the present study, the prevalence of vitamin A deficiency and marginal vitamin A deficiency in pregnant women were unexpectedly high. According to reports from the

WHO (26), vitamin A deficiency is classified as a moderate public health problem for pregnant women in communities, and programs combating such deficiencies should be implemented. Though normal values of cord serum vitamin A concentrations are not clearly established, its concentration below 0.7 $\mu\text{mol/L}$ suggests deficiency (27), indicating a severe public health problem for neonates in the locality. Consistent with previous reports (28, 29), cord serum vitamin A levels in our study were significantly lower than those of maternal serum. In contrast to other studies (18, 28), significant correlation between cord and maternal serum vitamin A was not observed in our study. Plasma vitamin A is transported by vitamin A binding protein or in a form of retinyl ester in lipoprotein (14), which probably contributes to our observation that no significant relationship exists between maternal and cord serum vitamin A concentration in these healthy mother-neonate pairs. Placental transfer was calculated as the ratio of newborn to maternal serum vitamin A concentration. In our study, the ratio was about 78% and was higher than those from previous reports (50–60%) (27, 29).

Environmental tobacco smoke is a major source of oxygen free radicals (30). Our data showed that the extent of maternal exposure to environmental tobacco had a significant effect on maternal, but not neonatal, serum vitamin A concentration, which is consistent with previous reports (31, 32). The different effect of environmental tobacco smoke on maternal and neonate vitamin A status was probably related to the efficient buffering function of the placenta. The placenta is able to control vitamin A exchange between mother

and fetus by releasing vitamin A to the fetus when maternal intake is deficient (33). Results in our study revealed that cord vitamin A level most likely was increased with gestational age, and this was consistent with a previous report that vitamin A accumulates in the fetus throughout the last third of pregnancy (34).

Our findings clearly demonstrated that vitamin E levels in cord blood are lower than those in maternal blood as previously reported (28, 29). In contrast with other studies, no correlation was found between maternal and cord plasma vitamin E concentrations, which is probably due to the different subject population. Several studies have shown that whatever the vitamin E status of mother and neonate were, the ratio of maternal to cord serum vitamin E levels persisted at 4:1 in rats (35). The ratio was far less than that in our study. This low placental transfer may be due to the inefficient transfer of plasma lipids (14), but the mechanism of transportation has not been clarified.

Although levels of vitamins A and E were lower in cord blood than in maternal blood, the level of vitamin C was significantly higher in cord blood than in maternal blood, which was consistent with a previous report (28). In addition, we found that cord serum vitamin C level was increased with gestational age, which was not consistent with Das and Powers (36). The high placental transfer (about 160% in the present study) most likely relates to placenta transferable function. It has been reported that the placenta is permeable to dehydroascorbic acid but not to ascorbic acid, and the fetus can convert dehydroascorbic acid to ascorbic acid, so accumulation of ascorbic acid in the fetus ensued (37). *Effect of mother-neonate paired vitamin status on birth outcomes*

VIF calculation values indicated that the multi-collinearity between mother-neonate paired vitamin A, vitamin E and vitamin C, and as well as the corresponding differences between them, cannot be included in the same multiple linear regression equation. The purpose of this study was to investigate the effect of maternal vitamin status on birth outcomes, so models including the maternal vitamin status but not the difference in mother-neonate paired vitamins, were recruited into the present study. The coefficient of determination (R^2) and adjusted R^2 of these two regression models were similar for each dependent variable of birth outcomes.

Effect of maternal vitamin A status on birth outcomes

As expected, neonate's sex, gestational age and maternal BMI indices before pregnancy exert significant effects on birth weight, birth height and head circumference. Most indices of maternal vitamin status were not associated with birth outcomes measured in our study. The associations between levels of maternal serum vitamin A during pregnancy and birth outcomes have been examined by other investigators, who also found the positive relationship (18, 38, 39), but others did not (20). Several reports also have found a negative correlation between maternal vitamin A status and birth weight (40).

There are few studies about the effect of maternal

vitamin A status on neonatal birth length. Villamor et al. (41) reported that supplementation of vitamin A and β -carotene to women during pregnancy not only had no beneficial effect on neonatal birth weight, length or head circumference but also could impair the beneficial roles of other multivitamins. Muslimatun et al. (42) also demonstrated that serum vitamin A status during pregnancy did not significantly influence neonatal weight or length. Our data indicated a positive correlation between maternal serum vitamin A status at delivery and neonatal birth length. Multiple variables regression analysis in the present study showed that the vitamin A ratio of neonates to pregnant women had a beneficial effect on birth length, regardless of the minimum changes of birth length (each 1% increase in the vitamin A ratio was concomitant with a 0.8 cm increase in birth length). Compared with the index of maternal serum vitamin A, the vitamin A ratio of neonate to pregnant women could preferably reflect the transfer efficiency of vitamin A from maternal body to neonate across the placenta. We concluded that high efficiency of vitamin A transfer in our study, regardless of the low maternal serum vitamin A status could also manifest the effect of vitamin A on birth outcomes.

Several reports have shown that cord vitamin A concentration is significantly correlated with neonatal head circumference (38, 39, 43). The relationship between maternal vitamin status and neonatal head circumference remains to be confirmed further.

Effect of maternal vitamin C and vitamin E status on birth outcomes

We found no significant relationship between maternal serum vitamin E or vitamin C at delivery and neonate birth weight, birth length or head circumference. Other studies reported that the concentration of vitamin E and vitamin C in women during pregnancy was positively and/or negatively associated with birth outcomes.

Lee et al. (17) and Rao et al. (44) had demonstrated that serum vitamin C levels during the second trimester had positive correlation with neonatal weight and length in full-term babies. Other investigators showed that supplementation with vitamins C and E during pregnancy did not reduce the risk of delivering an infant with a birth weight below the 10th percentile for gestational age (45), however increased the risk of a preterm infant (46). Scholl et al. (16) reported that at 28 wk, higher circulating concentrations of vitamin E were positively associated with several indicators of fetal growth. Masters et al. (38) also reported that infants whose mothers had low plasma α -tocopherol concentrations (below the median) weighed 92.9 g less and had 0.41 cm smaller head circumference than did infants whose mothers had high α -tocopherol concentrations. Other studies reported that maternal vitamin E levels at gestation of 18 and 30 wk were not related to birth outcomes (47).

Even though serum vitamin levels had only a limited effect on birth weight or length in this study, they may have a greater effect on the magnitude of health prob-

lems at the population level. Several reports have shown that even normal variations in size and weight at birth are associated with chronic diseases, such as coronary heart disease, hypertension and type-II diabetes in adulthood (2, 48).

Our data suggested that there may be a role for maternal vitamin A status during pregnancy in determining birth weight and length of infants and that vitamin A plays an important role in fetal growth. The low maternal and neonatal vitamin A serum concentrations in normal subjects in Changzhou City, where food is plentiful and vitamin A-rich foods are easily accessible throughout the year for all socioeconomic groups, maybe attribute, at least partially, to their lifestyle peculiarities and dietary preferences. The diets of local residents mainly consist of rice, with low availability of iron and without preformed vitamin A or pre-vitamin A.

Antioxidant defense systems are important to protect tissues and cells from damage caused by oxidative stress, so an imbalance between increased oxidative stress and decreased antioxidant defenses can impair fetal growth. There has been little research on the effect of the levels of vitamins A, E and C on fetal growth. It is notable that maternal serum concentrations of antioxidant vitamins during pregnancy are associated with birth outcomes. Our results do not suggest that healthy circulating concentrations of vitamins E and C in the mother improve fetal growth among relatively well-nourished women. This association is still not confirmed, and cautions should be highlighted in the use of supplements containing antioxidant vitamins.

As with observational designs, the main weakness of the study was that unknown confounding factors capable of affecting birth outcomes were not adjusted for, such as zinc (49) and *n*-3 fatty acids (50). In addition, subjects were recruited only from one exclusive clinical hospital, so the characteristics of women visiting the clinic for prenatal care may have influenced the results. Regardless of the investigation of vitamin intake during pregnancy, we did not measure dietary intakes of nutrients by 24-h recall questionnaires. This led to insufficient evaluation of the effect of maternal dietary habits on maternal nutrient status and birth outcomes.

CONCLUSION

Our investigation suggested that serum levels of vitamin A and vitamin E in maternal blood at delivery were significantly higher than those in cord blood; in contrast, serum levels of vitamin C in maternal blood at delivery were lower than those in cord blood. We found that there is a positive correlation between maternal vitamin A status and birth weight and height. Multiple linear regression analysis showed those maternal vitamin A status did not exert a beneficial effect on birth weight, whereas the transfer rate of vitamin A had a beneficial influence on birth length. Further studies are necessary to investigate the role of these antioxidant vitamins in fetal growth at various gestation stages.

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