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Relationship between adrenal steroid hormones in cord blood and birth weight: The Sapporo Cohort, Hokkaido Study on Environment and Children's Health

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Abstract

Objectives: We investigated the relationship between steroid hormone levels in cord blood and birth weight.

Methods: Among 514 participants in a prospective birth cohort study in Sapporo, the following hormone levels were measured in 294 stored cord blood samples from 135 males and 159 females: androstenedione, dehydroepiandrosterone (DHEA), cortisol, and cortisone. Birth weight information was obtained from medical records.

Results: androstenedione/DHEA was significantly higher in males than in females, while DHEA was significantly higher in females. Birth weight was significantly higher in males than in females. Regarding cortisone, androstenedione/DHEA, and cortisone/cortisol, a correlation was observed with birth weight in males but not in females.

Conclusions: Prenatal adrenal steroids as well as converting enzymes such as 11ß-hydrosteroid dehydrogenase type 2 and 3ß-hydrosteroid dehydrogenase may have an impact on prenatal physical development.

Introduction

The prenatal environment affects infant development. Adaptive changes to an adverse environment *in utero*, which results in low birth weight, have the potential to result in permanent changes in physiology, structure, and metabolism, which are collectively termed fetal ‘programming’ (Moisiadis and Matthews, 2014a; Moisiadis and Matthews, 2014b; O'Connor and Barrett, 2014) and are considered independent factors contributing to adult outcomes.

Glucocorticoids are an important factor in physiological functions, and the hypothalamic-pituitary-adrenal axis is a major system involved in the stress response and its regulation. Maternal cortisol passes through the placenta and affects both fetal cortisol levels and hypothalamic-pituitary-adrenal development, and is known as a glucocorticoid signal between the mother, placenta, and fetus (Moisiadis and Matthews, 2014a; Moisiadis and Matthews, 2014b; O'Connor and Barrett, 2014). Previous studies have reported that glucocorticoid levels in maternal blood and amniotic fluid correlate with birth weight (Baibazarova, et al., 2013; Field, et al., 2006). However, few studies have investigated the relationship between birth weight and glucocorticoid levels in cord blood which directly reflect the infant’s hormonal environment at birth. In this study, we examined whether adrenal steroid hormone levels in cord blood were associated with infant birth weight.

Participants and Methods

Participants

This prospective birth cohort study was based on the Sapporo Cohort, Hokkaido Study on Environment and Children's Health (Kishi, et al., 2013). Details regarding the population, data collection, sampling of biological specimens, and contents of the questionnaire have been described previously (Kishi, et al., 2013). Among 514 pregnant women who were enrolled in this cohort study, we excluded women who had miscarriages, stillbirths, relocated, voluntarily withdrew (n = 11), and those who delivered twins (n = 6). Finally, 294 participants, who had data of hormones in cord blood and birth weight, were enrolled in the present study.

The current study was approved by the Institutional Ethical Board for Epidemiological Studies at Hokkaido University Graduate School of Medicine and Hokkaido University Center for Environmental and Health Sciences. All participants signed informed consent, and informed consent was provided by parents on behalf of the children enrolled.

Hormone measurements in cord blood samples

At the time of delivery, a 10-30 mL blood sample was collected from the umbilical cord and stored at -80°C for subsequent analysis. On 294 stored cord blood samples (135 males and 159 females), the following steroid hormones were measured using highly specific liquid chromatography-tandem mass spectrometry: cortisol, cortisone, androstenedione, and dehydroepiandrosterone (DHEA) (Yamashita, et al., 2007). Ratio of androstenedione/DHEA and cortisol/cortisone were also calculated for parameters of

11ß-hydrosteroid dehydrogenase type 2 (11ß-HSD2) and 3ß-hydrosteroid dehydrogenase (3ß-HSD) activities in the placenta and fetal adrenal gland, respectively.

All hormone measurements were performed by Aska Pharma Medical Co., Ltd. (Kanagawa, Japan). The detection limits for each hormone were as follows: cortisol, 0.250 ng/mL; cortisone, 0.100 ng/mL; androstenedione, 0.010 ng/mL; and DHEA, 0.010 ng/mL.

Statistical analyses

Participant characteristics and hormone values were analyzed using one-way ANOVA or Chi-square. Hormone values were converted to a log10 scale because the data were not normally distributed. Half the detection limit listed above was used when levels were below the detection limit for individual hormones. The relationships between birth weight and steroid hormone levels in cord blood samples were calculated using multiple linear regression analyses. Inclusion of covariates was based on biological considerations, and adjustments were made for gestational age (continuous), maternal smoking and alcohol consumption during pregnancy (yes or no), annual household income (<5 or ≥5 million yen/year), educational level (≤12 or ≥13 years), and parents' body weights (continuous). Statistical analyses were performed using JMP pro 10 (SAS institute Inc., NC, USA). Significance levels were set to 0.05 for all comparisons.

Results

1) Characteristics of participants and their children

The characteristics of parents and their children were shown in Table 1. Birth weights of children ranged from 1784 g to 4162 g (median: 3096 g). Only 4 infants were born with low birth weight (<2500 g). Birth weights were significantly higher in males (mean ± standard deviation: 3139.3 ± 28.1 g) than in females (3072.0 ± 25.9 g) (p=0.002). Maternal pre-pregnancy weight was significantly associated with female birth weight (ρ=0.2685, p<0.001) but not that of males (ρ= 0.0977, p= 0.128). Paternal weight was not associated with the birth weights of males (ρ= 0.0310, p= 0.631) or females (ρ= 0.0400, p= 0.525).

2) Sex hormones in cord blood samples

Median concentration of androstenedione/DHEA was significantly higher in males than in females, whereas that of DHEA was significantly higher in females. Other hormones were not different between males and females.

3) Relationship between birth weight and sex hormones

A multivariate regression model showed that birth weight was positively correlated with cortisone, cortisone/cortisol, and androstenedione/DHEA in males only. No significant correlations were observed between birth weight and any other hormones, in males or females (Table 2).

Discussion

The results of our investigation suggest that prenatal adrenal steroids are associated with birth weight in males but not in females. Furthermore, androstenedione/DHEA and cortisol/cortisone, which are indicative of converting enzymes in the placenta and fetal adrenal gland, also correlated with birth weight in males only.

Supraphysiologic levels of glucocorticoids may induce fetal growth retardation. High glucocorticoid concentrations in both maternal and fetal human studies have been associated with intrauterine growth retardation (Field, et al., 2006; McTernan, et al., 2001). Elevated concentrations of glucocorticoids have been found to be induced by stress, maternal malnutrition, and placental insufficiency due to restriction of placental blood flow. On the other hand, the fetus is generally protected from the effects of maternal glucocorticoids by 11ß-HSD2 in the placenta. This enzyme rapidly catalyzes cortisol (in the form of active glucocorticoids) to cortisone, a physiologically inactive glucocorticoid. In an animal study using rats, maternal stress during gestation induced decreased fetal weight and placental 11ß-HSD2 activity (Mairesse, et al., 2007). Thus, 11ß-HSD2 functions to prevent the transport of maternal glucocorticoids to the fetus (Benediktsson and Seckl, 1998). Regarding 3ß-HSD, there is no evidence for the relationship of 3ß-HSD and fetal growth during the prenatal period, from either human epidemiological or animal studies. Although we speculate that some pathways in the steroidogenesis of progesterone and 3ß-HSD in the placenta and fetus may be involved in fetal growth, this has not yet been demonstrated.

The sexually dimorphic effects of glucocorticoids on human fetal growth have been reported in previous studies. Ellman et al. (2008) reported that an increase in cortisol

levels in mothers during early pregnancy was correlated with physical maturity in newborn males only (Ellman, et al., 2008). Further, difficulty in adapting placental function to an adverse maternal environment may also induce sexual dimorphism in fetal growth (Clifton, 2010). Therefore, the effects of glucocorticoids during gestation may induce sexual dimorphism in fetal development, and glucocorticoid metabolic activity is less adaptive to high maternal glucocorticoid concentrations in the male placenta. However, the mechanisms underlying the sexually dimorphic effects of glucocorticoids on fetal growth remain unclear, and thus further studies are necessary.

There are some limitations to this study. First, the correlation between birth weight and prenatal hormone levels was relatively small, indicating the possibility of marked individual differences in human biological variation at birth. Second, the cord blood samples were collected throughout the day and night due to unpredictable deliveries. Therefore, the diurnal rhythms of circulating glucocorticoids may have affected these data. Third, stress from the delivery itself could have affected levels of steroid hormones in the cord blood (Keelan, et al., 2012). However, we did not investigate this factor, since there are no data establishing the impact of stress on steroid hormone levels during delivery. Fourth, the effects of hormone exposure during critical gestational periods on fetal growth are still unknown because we analyzed hormonal exposure using cord blood at delivery. Therefore, further studies are required to reveal the effects of steroid hormone levels *in utero* on physical changes in children.

Conclusions

Prenatal adrenal steroid exposure during the prenatal period has the potential to affect fetal growth. Converting enzymes, such as 11ß-HSD2 and 3ß-HSD in the placenta and fetal adrenal gland, respectively, may also impact birth weight. Since these relationships were observed in males only, sensitivity to steroid hormones and the activities of enzymes in steroidogenesis may induce sexually dimorphic effects of glucocorticoids on fetal growth.

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Abbreviations

3ß-HSD: 3ß-hydrosteroid dehydrogenase

11ß-HSD2: 11ß-hydrosteroid dehydrogenase type 2

DHEA: dehydroepiandrosterone

HPA: hypothalamic-pituitary-adrenal

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Table 1 Characteristics of participants

The values in brackets represent percentages.

			Participants with data of hormones and birth weight
Characteristics of parents			
		n	Mean ± SD
Age at delivery (years old)		294	30.4 ± 5.0
Body weights of mothers at pre-pregnancy (kg)		294	52.6 ± 7.8
Body weights of fathers (kg)		289	68.5 ± 11.0
			n (%)
Parity	Primiparous		149 (50.7)
	Multiparous		145 (49.3)
Annual household income (million yen per year)	<5		206 (70.6)
	≥5		86 (29.5)
Educational level (years)	≤12		133 (45.2)
	≥13		161 (54.8)
Smoking during pregnancy	Non-smoker		238 (81.0)
	Smoker		56 (19.0)
Alcohol consumption during pregnancy	Non-drinker		193 (65.7)
	Drinker		101 (34.3)
Infant characteristics			
			n (%)
Gender	Males		135 (45.9)
	Females		159 (54.1)
		n	Mean ± SD
Birth weight (g)		294	3126.8 ± 331.6
Gestational age (weeks)		294	39.3 ± 1.0

SD: standard deviation

Table 2 Relationship between birth weight and sex hormones in cord blood

*: p<0.05

Hormone levels	Males		Females	
	B	R^2	B	R^2
	95% CI		95% CI	
Cortisol	0.1203	0.1014	0.0060	0.2372
	-57.534 / 163.72		-78.199 / 84.852	
Cortisone	0.1741*	0.1432	0.0211	0.2375
	4.240 / 183.06		-51.437 / 68.884	
Androstenedione	0.1266	0.1089	-0.1115	0.2415
	-76.522 / 403.87		-357.66 / 43.907	
DHEA	-0.099	0.1232	-0.0148	0.2373
	-345.17 / 91.470		-162.87 / 132.84	
Cortisone / Cortisol	0.2125*	0.1553	0.0407	0.2386
	43.255 / 383.83		-103.24 / 179.88	
Androstenedione / DHEA	0.1746*	0.1436	--0.0593	0.2406
	10.269 / 385.74		-176.53 / 74.033	

CI: confidence interval, DHEA: dehydroepiandrosterone.

Co-variates: gestational age, maternal smoking and alcohol consumption during pregnancy, annual household income, educational level, and body weights of parents.