Null Results in Brief

Estrogen and Androgen Concentrations Are Not Lower in the Umbilical Cord Serum of Pre-eclamptic Pregnancies

Rebecca Troisi,[1] Nancy Potischman,[2] Christine Neuser Johnson, James M. Roberts,[3,4] David Lykins,[3] Gail Harger,[3,5] Nina Markovic,[3,5] Pentti Siiteri,[6] and Robert N. Hoover[1]

[1]Division of Cancer Epidemiology and Genetics, [2]Division of Cancer Control and Population Sciences, National Cancer Institute, Bethesda, Maryland; [3]Magee-Womens Research Institute, [4]Department of Obstetrics Gynecology and Reproductive Sciences, [5]Graduate School of Public Health, University of Pittsburgh, Pittsburgh, Pennsylvania; and [6]University of California San Francisco School of Medicine, San Francisco, California

Abstract

Reductions in breast cancer risk observed in daughters of pre-eclamptic pregnancies are hypothesized to be mediated by lower *in utero* estrogen concentrations. Whereas maternal urinary estriol excretion is generally lower in pre-eclamptic women, results for maternal blood concentrations are equivocal, and little is known about estrogen concentrations in the cord of pre-eclamptic pregnancies. Unconjugated estrogen and androgen concentrations were measured in mixed umbilical cord sera from 86 pre-eclamptic and 86 uncomplicated, singleton pregnancies, matched on length of gestation, maternal age, parity, and type of delivery. Pre-eclamptic and uncomplicated pregnancies were similar in maternal age, prepregnancy weight, maternal height, type of delivery, use and type of anesthesia, and sex of offspring. Estriol, estradiol, estrone, dehydroepiandrosterone, dehydroepiandrosterone-sulfate, androstenedione, and testosterone concentrations measured in cord sera were not significantly different in pre-eclamptics compared with uncomplicated pregnancies. Estriol was 9% lower ($P = 0.43$), and all of the other hormones were actually higher in pre-eclamptics with testosterone and estradiol approaching statistical significance ($P = 0.06$ and $P = 0.12$, respectively). These data do not support the hypothesis that the lower breast cancer risk in daughters of pre-eclamptic pregnancies is explained by lower *in utero* estrogen exposure.

Introduction

Reduced *in utero* estrogen exposure is hypothesized to explain the lower risk of breast cancer found in daughters of pre-eclamptic pregnancies (1). Studies investigating the hormonal correlates of conditions such as pre-eclampsia to understand the biological mechanisms underlying effects on cancer risk have relied largely on maternal samples. Whereas maternal urinary estriol excretion is lower in pre-eclamptic women in most studies, results for circulating levels are equivocal, and few studies have assessed concentrations of other, more potent estrogens, such as estradiol (2–4) and estrone (3). Previously, we reported that circulating estrogens, including estriol, estradiol, and estrone, were not reduced in maternal serum from pre-eclamptic compared with uncomplicated pregnancies, with adjustment for gestational age and several other potentially confounding factors (5). In contrast, androgen concentrations were elevated, as noted in two previous studies (4, 6).

The more proximate exposure to the fetus, estrogens in the fetal circulation, has not been studied in pre-eclamptic pregnancies. Because the fetus and placenta are highly integrated, it has been assumed that maternal hormone concentrations reflect those in the fetal circulation. The degree of correlation between hormone concentrations in the maternal and fetal circulation, however, is modest (7). We tested the hypothesis that concentrations of estrogens and androgens in mixed cord blood samples from pre-eclamptic differ from those of uncomplicated pregnancies of similar gestational age.

Materials and Methods

The study methods are described in detail elsewhere (5). Briefly, subjects were a sample from an ongoing study of pre-eclamptic pregnancies at the Magee Womens Hospital, University of Pittsburgh, between 1994 and 1998. Cases, defined by explicit criteria ($n = 86$), and controls ($n = 86$) were 14 years of age and older. Controls were uncomplicated deliveries, matched to cases on parity, length of pregnancy at delivery (± 2 weeks), type of delivery (vaginal *versus* C-section, and whether they labored or not), and maternal age (± 5 years). Mixed cord sera were collected at delivery from pre-eclamptic and normal pregnancies, and samples were analyzed for unconjugated estrogens and androgens at Quest Diagnostics (San Juan Capistrano, CA). Laboratory personnel were blinded to case status, and case and control samples were run matched within batches to control for laboratory drift. Blinded aliquots of pooled sera from normal pregnant women from the study population constituted 10% of each batch and were handled exactly the same as the study samples to monitor quality during the laboratories assays. The coefficients of variation (representing total inter- and intra-assay error) were 8.1% for DHEA,[7] 6.6% for DHEAS, 8.5% for androstenedione, 15.2% for testosterone, 10.9% for estradiol, 16.7% for estrone, and 9.2% for estriol. Information on demographics and reproductive and medical history, as well as some details of the pregnancy, was

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Requests for reprints: Rebecca Troisi, 7927 Rubin Building, Dartmouth-Hitchcock Medical Center, One Medical Center Drive, Lebanon, NH 03756. E-mail: troisir@mail.nih.gov.

[7] The abbreviations used are: DHEA, dehydroepiandrosterone; DHEAS, dehydroepiandrosterone-sulfate.

Table 1 Mean cord serum hormone concentrations in pre-eclamptic and uncomplicated pregnancies

Hormone	Pre-eclampsia ($N = 86$)	Uncomplicated pregnancy ($N = 86$)	P
DHEA (ng/dL)			
Median (range)	454 (124–1,598)	531 (103–1,470)	
Adjusted mean[a] (95% CI[b])	462 (332–642)	447 (328–609)	0.70
DHEAS (μg/dl)			
Median (range)	151 (18–709)	187 (20–885)	
Adjusted mean[a] (95% CI)	132 (80–220)	139 (87–224)	0.68
Androstenedione (ng/dl)			
Median (range)	341 (158–1,644)	344 (91–996)	
Adjusted mean[a] (95% CI)	338 (251–454)	323 (245–426)	0.54
Testosterone (ng/dl)			
Median (range)	22.0 (6–187)	20.5 (2–166)	
Adjusted mean[a] (95% CI)	19.3 (11.4–32.9)	15.7 (9.6–25.9)	0.12
Estradiol (pg/ml)			
Median (range)	11,805 (3,332–44,431)	9,510 (402–81,083)	
Adjusted mean[a] (95% CI)	9,901 (5,468–17,929)	7,526 (4,315–13,124)	0.06
Estrone (pg/ml)			
Median (range)	26,356 (4,848–151,824)	30,815 (2,753–147,865)	
Adjusted mean[a] (95% CI)	27,574 (15,549–48,899)	28,135 (16,451–48,117)	0.89
Estriol (ng/ml)			
Median (range)	188 (54–755)	223 (44.7–768)	
Adjusted mean[a] (95% CI)	276 (174–438)	303 (199–461)	0.43

[a] Adjusted means are geometric and from models that include a variable representing the individual match, race (white/other), parity (nulliparous, 1, 2+), gravidity (1, 2, 3, 4+), smoking (yes/no), weeks of gestation (continuous) and hours of labor (continuous).
[b] CI, confidence interval.

obtained by interview and supplemented by medical records of the subjects. Linear regression, accounting for the matched analysis, was used to assess differences in hormones between pre-eclamptic and uncomplicated pregnancies using SAS software (Statistical Analysis System, Inc., Cary, NC).

Results

As we reported previously (6), prepregnancy weight, maternal height, delivery method, use and type of anesthesia, and sex of the babies were similar in cases and controls, although the cases were more likely to be white, to be in their first pregnancy, and less likely to be highly parous or to smoke, but these latter three differences were not statistically significant. Despite the attempt to match, pre-eclamptic babies had a slightly shorter gestation (37.0 weeks, SD = 2.2 *versus* 37.8 weeks, SD = 2.4; $P = 0.04$) and, with adjustment for gestational age, appeared smaller with a lower mean birth weight than babies from uncomplicated pregnancies (2704 g *versus* 2932 g; $P = 0.15$), length (48.2 cm *versus* 49.1 cm; $P = 0.05$), and head circumference (32.9 cm *versus* 33.7 cm; $P = 0.0007$).

Estrogen and androgen concentrations measured in the cord sera of pre-eclamptic and uncomplicated pregnancies showed no statistically significant differences, with adjustment for several potentially confounding factors (Table 1). Whereas a difference in cord estradiol approached statistical significance, it was higher in pre-eclamptic pregnancies than in uncomplicated pregnancies. Testosterone appeared higher in the cord serum of pre-eclamptic compared with uncomplicated pregnancies, but the difference was not statistically significant.

Women with pre-eclampsia were more likely to have been treated with magnesium sulfate (81.4% *versus* 1.2% ($n = 1$); $P < 0.001$) and prostaglandins (29.1% *versus* 8.1%; $P < 0.001$) and were slightly more likely to be given oxytocin (84.9% *versus* 72.1%; $P = 0.06$) than controls. Adjustment for prostaglandin or oxytocin use did not materially change the results.

Assuming an α of 0.05, 86 pairs and a SD = 146 (observed in the control group), the power was 88% to find a 20% difference in mean cord estriol between the cases and controls in a paired analysis.

Discussion

These data do not provide support for the hypothesis that reduced breast cancer risk in daughters of pre-eclamptic pregnancies are mediated through lower fetal estrogen exposure. We observed an ~9% lower estriol concentration in pre-eclamptic pregnancies. Because our study was not powered to detect such a small difference, we cannot exclude the possibility of an effect at this level. The etiological significance of this finding is cast in doubt, however, by findings for cord estradiol, a much more potent estrogen, which was higher in pre-eclamptic pregnancies and of borderline statistical significance. These findings are consistent with the lack of difference in maternal estrogen concentrations between pre-eclamptic and uncomplicated pregnancies that we observed in a previous study (6). Likewise, testosterone, a potent androgen, was elevated in both the maternal and cord circulations, although it lacked statistical significance in the latter.

Limitations of the main study from which these data derive have been detailed in a previous report (6). Briefly, cases arose from the entire obstetric service, which included patients from both private and hospital practices, whereas the comparison group of uncomplicated pregnancies was drawn only from the practice of the hospital. Whereas our results could be biased if private- and hospital-practice patients differed with respect to factors associated with hormone concentrations, the findings remained with adjustment for race and smoking, although we cannot discount confounding by other factors.

Random measurement error in the hormones may have resulted in the lack of an association, although the combined inter- and intra-assay laboratory error, calculated using blinded replicates, ranged from 6.6% for DHEAS to 16.7% for estrone. The coefficient of variation for estriol was only 9.2%, suggesting laboratory error was unlikely to have obscured the results. Cord sera were collected by necessity after delivery, and we

attempted to reduce bias from extraneous factors associated with labor and delivery, such as type of delivery and duration of labor, by matching for them in the design and including them in the regression analyses, respectively.

In conclusion, we found no difference in concentrations of unconjugated estrogens and androgens in the cord sera of pre-eclamptic and uncomplicated pregnancies. These data are not consistent with the hypothesis that reduced cancer risk in offspring of pre-eclamptic pregnancies is attributable to lower estrogen concentrations in the fetal circulation. In fact, similar to our findings in maternal blood, the cord concentrations of both estradiol and testosterone were actually higher in pre-eclampsia. Future studies focusing on uncovering the mechanism responsible for the protective effect on breast cancer risk should attempt to address more comprehensively the changes in all potentially relevant pregnancy hormones and growth factors. Indeed, such explorations should probably also include assessments of changes in immunological and other nonhormonal exposures in pre-eclampsia.

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