Antenatal Corticosteroids and Newborn Screening for Congenital Adrenal Hyperplasia

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Objective: To assess the effect of reported corticosteroid exposure on neonatal levels of 17-hydroxyprogesterone (17-OHP), the cortisol precursor used in newborn screening for congenital adrenal hyperplasia, in newborns weighing less than 2500 g at birth.

Design: A retrospective study of newborns weighing less than 2500 g at birth and exposed to corticosteroids as reported on their newborn screening card compared with newborns weighing less than 2500 g at birth and reported as not exposed to corticosteroids.

Methods: Birth weight, gestational age, age at screening, special care information, and name of screening hospital were obtained from newborn screening cards for 16115 newborns screened in Michigan during the first 3 months of 2000. Levels of 17-OHP, measured by fluoroimmunoassay, were obtained from Michigan’s Newborn Screening Program database.

Results: The mean 17-OHP level for the 69 low-birth-weight newborns in the corticosteroid-exposed group was 52 ng/mL, which was higher than that for the 771 low-birth-weight newborns in the unexposed group (35 ng/mL) (P <.001). Reported corticosteroid use did not decrease the number of expected borderline positive screening results for congenital adrenal hyperplasia (P >.05). Levels of 17-OHP varied by birth weight in corticosteroid-exposed and unexposed newborns.

Conclusions: Corticosteroid exposure may not suppress screening 17-OHP levels. Therefore, newborn screening should not be delayed in premature newborns because of antenatal exposure to corticosteroids.


Congenital adrenal hyperplasia (CAH) refers to a clinical spectrum of autosomal recessive disorders defined by a deficiency in an enzyme necessary for cortisol biosynthesis from cholesterol in the adrenal gland.1,2 The most common of the disorders, 21-hydroxylase deficiency, occurs in approximately 1 of 15000 live births in the United States and accounts for more than 90% of CAH cases.3-5 Boys, in particular, with classic CAH are at risk for delayed diagnosis because there are not any specific physical signs evident immediately after birth.3-5 Newborn screening for CAH plays a major role in identifying these affected infants before development of symptoms or life-threatening biochemical abnormalities.7,8

Newborn screening for CAH is accomplished by assaying for elevated levels of 17-hydroxyprogesterone (17-OHP) on filter paper saturated with a heel stick capillary blood sample. The feasibility of this method was first demonstrated by Pang et al.9 Twenty states, including Michigan, currently screen for CAH, and several states are considering adding the test to their screening panel.10 A major difficulty with CAH screening is that low-birth-weight newborns typically have higher 17-OHP levels than normal-birth-weight newborns. To avoid a high false-positive rate when screening for CAH, many states have adopted weight-adjusted cutoffs. For those states, the less a newborn weighs, the higher his or her 17-OHP level needs to be to have a positive screening result.11

A further complicating factor in newborn CAH screening is that all fetuses at risk of preterm delivery between 24 and 34 weeks’ gestation are considered candidates for antenatal corticosteroid treatment for fetal lung maturation according to a consensus statement from the National Institutes of Health.12 Long-term maternal corticosteroid treatment suppresses fetal 17-OHP levels, as maternal treatment with dexamethasone is successful in preventing or minimizing virilization in most prenatally treated female fe-
MATERIALS AND METHODS

Data were obtained from Michigan’s Newborn Screening Program in Lansing. A total of 16,115 newborn screening cards, collected during January, February, and March of 2000, were examined. Michigan mandates screening for all newborns and recommends specimen collection at an age older than 24 hours. The Michigan newborn screening card requests information on the newborn’s name, sex, hospital of birth, medical record number, order of birth, birth date, specimen date, newborn’s age in hours, gestational weeks, birth weight, ethnic background, and demographic information for the mother and for the physician responsible for newborn follow-up. The mother’s hepatitis B surface antigen test result and responses to 5 special care questions labeled “antibiotics,” “transfused,” “type of feeding,” “neonatal intensive care unit/special care nursery,” and “steroid treatment” are also requested.

Initial newborn screening cards marked with a birth weight of less than 2500 g were excluded for analysis. Age at screening, birth weight, and gestational age were recorded. The state laboratory’s unique identifying sample number was also recorded, as were the yes/no responses to the 5 special care questions. Special note was made if the newborn’s age in hours did not agree with the data fields for birth date and specimen date. Newborn screening cards with unsatisfactory blood samples because of specimen age, layering of blood, insufficient or multiple applications of blood, presence of serum rings, or contamination were excluded from the study.

Initial screening laboratory 17-OHP values were obtained from the Michigan Newborn Screening Program database and matched with appropriate card information using the laboratory’s unique identifying sample number. Michigan uses the Auto DELFIA system (Perkin Elmer Life Sciences, Wellesley, Mass), a fluoroimmunoassay, to determine the 17-OHP values of dried blood spots from heel pricks on filter paper. This method of screening is complicated by cross-reactivity that elevates the apparent 17-OHP level. In this study, “17-OHP level” refers to the apparent 17-OHP level.

RESULTS

Of 16,115 newborn screening cards examined for possible inclusion in this study, 1217 (7.6%) reported that the newborn weighed less than 2500 g at birth. Twenty-seven cards (2.2%) were excluded from the study because of an unsatisfactory blood sample. Of the remaining 1190 cards, 840 (70.6%) had the steroid treatment question completed. According to data recorded on the newborn screening cards, 69 of those newborns (8.2%) were exposed to corticosteroids and 771 (91.8%) were not. The mean 17-OHP level of the corticosteroid-exposed group was 52 ng/mL, and the mean 17-OHP level of the unexposed group was 35 ng/mL ($P < .001$). Mean birth weight was 1365 g for corticosteroid-exposed newborns and 2018 g for unexposed newborns ($P < .001$). Based on screening card information, there was no significant difference in mean age at screening between the 2 groups ($P > .05$) (Table 1).

The Figure illustrates the range of the data. Although gestational age and birth weight correlate with 17-OHP levels, birth weight is a more accurate and measurable variable, so it is frequently used in formulating stratified cutoffs. A 2-factor analysis of variance was used to determine whether mean levels of 17-OHP for newborns exposed to corticosteroids and those not exposed were statistically significantly different from each other. One factor in this analysis was corticosteroid exposure, and the second factor was birth-weight class (Table 2). Because the number of newborns in each of the categories was uneven, the analysis of variance was run using a regression approach. Mean levels of 17-OHP were not statistically different.

Fifty-seven (83%) of the 69 corticosteroid-exposed newborns were born in hospitals that reported routinely checking the mother’s prenatal history for antenatal corticosteroid exposure, as opposed to checking only

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<th>Table 1. Comparison of Corticosteroid-Exposed and Unexposed Groups</th>
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<tr>
<td>Exposed (n = 69)</td>
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<tr>
<td>17-Hydroxyprogesterone, mean (SD), ng/mL*</td>
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<td>Birth weight, mean (SD), g*</td>
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*$P < .001$. 

...tuses affected with CAH. A question that remains to be answered is whether short-term maternal corticosteroid treatment also suppresses 17-OHP levels, possibly interfering with validating the results of newborn screening. In many states low-birth-weight infants need to have higher 17-OHP levels to have a positive screening result, if corticosteroid treatment suppresses 17-OHP levels, a true-positive case of CAH may be missed. The purpose of this study was to determine whether corticosteroid treatment as reported on the newborn screening card significantly suppresses newborn 17-OHP levels.
the newborn’s record for antenatal or postnatal corticosteroid exposure. The mean 17-OHP level and birth weight of these 57 corticosteroid-exposed newborns were 50 ng/mL and 1363 g, respectively. The values for unexposed newborns (n = 244) born at these hospitals were 34 ng/mL and 2004 g, respectively. These results were not statistically significantly different from the results described earlier for all corticosteroid-exposed and unexposed newborns.

Eight of 35 newborns whose newborn screening cards indicated that they had been exposed to cortico-steroids but weighed less than 1300 g at birth had borderline positive or strongly positive screening results for CAH, with none confirmed as affected. Twelve of 82 comparable unexposed newborns had positive screening results. This result is not significantly different based on the $\chi^2$ test ($P > .05$).

There were 2 confirmed cases of CAH reported to the Michigan Newborn Screening Program during this study. One newborn was not included in the study because the birth weight was greater than 2500 g. The other newborn was included in the unexposed group. There were no corticosteroid-exposed newborns with CAH in our study population.

The primary goal of newborn CAH screening is to prevent salt-wasting deaths before diagnosis. Additional benefits include early detection of inappropriate sex assignment of girls and prevention of premature epiphysial closure in children with the simple virilizing variant, as well as prevention of other consequences associated with delayed diagnosis.1,13 Newborn screening for classic CAH is effective in lowering morbidity and mortality rates associated with late diagnosis, particularly in boys.

One principle of screening is that there should be minimal, preferably zero, false-negative test results. To achieve this, a small false-positive rate is accepted. To minimize the high false-positive rate in newborn CAH screening, most states that screen for CAH have implemented weight-adjusted cutoffs. This practice has been successful in decreasing the number of false-positive findings without any reports of false-negative findings.10 However, given that premature infants may receive corticosteroids in utero to promote fetal lung maturation and that low-birth-weight newborns require a higher 17-OHP level to have a positive screening result, this study investigated whether corticosteroid exposure might suppress adrenal function enough to cause affected newborns to be missed.

The benefits of antenatal corticosteroid treatment for fetal lung maturation include a large reduction in early neonatal death, respiratory distress syndrome, intraventricular hemorrhage, and necrotizing enterocolitis.12 Despite these benefits, corticosteroid therapy among eligible infants remained low throughout the 1990s. Data from multiple sources17,18 indicate that approximately 20% to 30% of eligible preterm infants received antenatal corticosteroid therapy, although there was great variation in use by institution. The present study found that of 315 newborns who weighed less than 2000 g at birth and had the corticosteroid question answered on the newborn screening card, 54 (17%) were exposed to corticosteroids. A study19 compiling 8 multicenter data sets found that corticosteroid therapy was used most often in infants weighing less than 1250 g and for infants of less than 31 weeks’ gestation. In the present study, 30% of newborns weighing less than 1250 g at birth were exposed to corticosteroids. This is consistent with the literature.

Limited data are available on the effect of maternal corticosteroid use on fetal and neonatal endocrine sys-
tems. Plasma cortisol levels have been reported to be lower in preterm infants exposed to antenatal glucocorticoids compared with preterm infants not exposed or exposed at least 7 days before testing. These differences often have not been significant, particularly after being corrected for multiple variables.20,21 Teramo et al22 compared the serum cortisol levels of 24-hour-old newborns who had been treated prenatally with betamethasone with those of newborns treated with placebo. There was no difference between cortisol levels in the groups, indicating that use of betamethasone did not have a sustained effect on fetal cortisol concentration.

Even after repeated antenatal corticosteroid exposure, the neonatal adrenal gland remains capable of secreting cortisol in response to corticotropin. Terrone et al23 administered corticotropin to neonates whose mothers had received 3 or more courses of betamethasone to enhance fetal lung maturity. Although the sample size was small (n=9), the mean cortisol concentration after corticotropin stimulation increased significantly in these infants.

In a small but well-controlled study, Dorr et al24 found no difference in plasma cortisol and 17-OHP levels 2 hours after birth in low-birth-weight newborns who received 2 doses of antenatal betamethasone compared with preterm newborns with uneventful histories who were not exposed to corticosteroids. Based on the works of Teramo22 and Dorr24 and their colleagues, it is not surprising that this study found that corticosteroid treatment, as reported on the newborn screening card, did not suppress 17-OHP levels. In fact, corticosteroid-exposed newborns were more likely than unexposed newborns to have higher 17-OHP levels. One explanation for this finding may be that smaller, more at-risk newborns in utero are more likely to receive the treatment, and those newborns are also more likely to have higher 17-OHP levels. This hypothesis is consistent with the study’s finding that corticosteroid-exposed newborns had a lower mean birth weight than did unexposed newborns. Nevertheless, even when stratified by birth weight, there is no evidence in this study that corticosteroid exposure suppressed 17-OHP levels.

Premature infants are at particular risk of being missed in newborn screening programs.25 Reasons for this may include neonatal transfer, concern about specimen validity, and concern that severe neonatal illness precludes screening. Despite these concerns, screening remains potentially valuable in the low-birth-weight population, especially because medical interventions or prematurity-related illness may mask clinical symptoms.26 Delay of screening until recovery of the adrenal axis does not seem to be justified, and it increases the likelihood of a missed opportunity to screen.

A limitation of this study is that the newborn screening card does not specify whether newborns or mothers received corticosteroid treatment, although it would be unusual for this group of newborns to receive postnatal corticosteroids in the first 3 days of life. Of hospitals that marked the steroid treatment “yes” box on 1 or more newborn screening cards, 60% reported routinely checking the mother’s medical record to get the information. Currently, no Michigan Department of Community Health instructions specifically defining “steroid treatment” have been disseminated to the nurses or technicians filling out newborn screening cards. Therefore, although the intent was to gather data on antenatal exposure, and most birth hospitals reported use of antenatal corticosteroids only, a small proportion of newborns may have been exposed postnatally.

More problematic was that the newborn screening card steroid treatment question was more likely to be left unanswered than were other special care questions. This suggests that individuals completing the cards might not have understood the question, chose not to answer the question, or did not have the proper information readily available. The response to the neonatal intensive care unit/special care nursery question was also omitted on 23.7% of the cards. That question does not require any special information gathering. The high rate of omitting these answers suggests that many individuals did not understand the importance of the information requested. Newborn screening cards provide a means to gather health information on a population basis, but clarity and completeness are important for many applications.

What This Study Adds

Small studies of the effect of antenatal corticosteroid use on the neonatal adrenal axis have demonstrated small and often transient effects on cortisol and 17-OHP levels. However, few data are available on the effect of antenatal corticosteroid use on 17-OHP levels measured in newborn screening for CAH. If antenatal corticosteroid use significantly alters screening test results for CAH, changes in timing or cutoffs would improve validity.

This study showed that reported corticosteroid exposure did not change newborn 17-OHP concentration when corrected for birth weight. In exposed and unexposed newborns, low birth weight is associated with increased 17-OHP levels compared with birth weights greater than 2500 g. Results of this study suggest that newborn screening should not be delayed in newborns exposed to antenatal corticosteroids.

CONCLUSIONS

In this study, newborns reportedly exposed to corticosteroids had screening 17-OHP levels similar to those of unexposed newborns when corrected for birth weight. This suggests that corticosteroid exposure may not result in significant adrenal suppression in neonates. We recommend that newborn screening specimens be collected from all newborns regardless of corticosteroid exposure. Because no data are available on the effect of limited maternal corticosteroid use on affected newborns, false-negative results remain a potential concern. A larger prospective study including maternal dose, timing of dose, and age of newborns at screening should be done to confirm the results of this study.

This study also documented that special care information on the newborn screening card is frequently omitted. Information requested on the cards is important in
interpreting the results of several screening tests. Education emphasizing the importance of the newborn screening information and clarifying what information should be recorded may be effective in increasing proper card completion.

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