Direct fetal glucocorticoid treatment alters postnatal adaptation in premature newborn baboons

Ervin, M. Gore, Steven R. Seidner, M. Michelle Leland, Machiko Ikegami, and Alan H. Jobe. Direct fetal glucocorticoid treatment alters postnatal adaptation in premature newborn baboons. Am. J. Physiol. 274 (Regulatory Integrative Comp. Physiol. 43): R1169–R1176, 1998.—Abnormalities of premature newborn adaptation after preterm birth result in significant perinatal mortality and morbidity. We assessed the effects of short-term (24 h) fetal betamethasone exposure on preterm newborn baboon pulmonary and cardiovascular regulation and renal sodium handling during the first 24 h after birth. Male fetal baboons (Papio) (124-day gestation, term 185 days) received ultrasound-guided intramuscular injections of saline (n = 5) or betamethasone (0.5 mg/kg; n = 5). Fetuses were cesarean delivered 24 h later, treated with 100 mg/kg surfactant, and ventilated by adjusting peak inspiratory pressures to maintain P<sub>CO2</sub> values of 35–50 mm Hg for 24 h. Betamethasone- vs. saline-treated mean ± SE newborn body weights (0.45 ± 0.02 vs. 0.41 ± 0.01 kg) were similar. Although prenatal betamethasone did not affect postnatal lung function (P<sub>CO2</sub> arterial/alveolar O<sub>2</sub> gradient, or dynamic compliance), plasma hormones (cortisol or thyroxine), or catecholamine levels, mean arterial pressure (25 ± 1 vs. 32 ± 1 mm Hg), plasma sodium concentration (132 ± 2 vs. 138 ± 1 meq/l), glomerular filtration rate (24.07 ± 0.07 ml·min<sup>-1</sup>·kg<sup>-1</sup>), and renal total sodium reabsorption (1.5 ± 0.5 vs. 16.0 ± 3.0 meq·min<sup>-1</sup>·kg<sup>-1</sup>) values were significantly lower in saline-treated than in betamethasone-treated newborns at 24 h. We conclude that despite the fact that there are no pulmonary and endocrine effects, antenatal glucocorticoid exposure alters premature newborn baboon vascular and renal glomerular function and improves sodium reabsorption after preterm delivery.

THE MAJOR CAUSES OF INFANT morbidity and mortality in the United States are diseases associated with prematurity. Because lung immaturity is the principal contributor to premature newborn mortality, the lungs have been the primary focus of strategies to improve premature newborn survival (7). Although glucocorticoid-induced fetal lung maturation was demonstrated in a large clinical trial over two decades ago (20), widespread application of this therapy has occurred only recently (12, 13). In addition to their pulmonary effects, prenatal glucocorticoids improve postnatal outcomes with their pleiotropic effects, including reduced incidences of patent ductus arteriosus and intraventricular hemorrhage and improved postnatal blood pressure regulation (22). We have demonstrated that antenatal glucocorticoid treatment significantly enhances renal sodium reabsorption in premature newborn sheep (9). Kidney immaturity and the resulting inability to limit excessive fluid and electrolyte losses is a common problem in the clinical management of the preterm newborn and is a significant source of postnatal morbidity (12, 13). Thus, in contrast to term newborns in which postnatal renal adaptations include marked increases in glomerular filtration rate (GFR) and sodium reabsorption (33), preterm newborns (particularly those <30 wk gestation) do not appropriately increase renal sodium reabsorption at birth (3) and retain a fetal pattern of excessive natriuresis. The effects of prenatal glucocorticoids on postnatal adaptation have been examined in detail in preterm sheep, and limited, nonrandomized observations regarding steroid effects on renal function in preterm newborns are available. However, renal function and its hormonal regulation have not been evaluated in premature humans or other primates. Preterm baboons can be delivered as early as 125 days (0.67 of total length of gestation, with term 185 days) and will survive with the development of bronchopulmonary dysplasia if surfactant treated at birth (29). The purpose of the present study was to determine the effect of antenatal steroid administration on postnatal lung, kidney, and endocrine function in the premature newborn primate.

METHODS

Animal Selection and Fetal Treatment

The fetal treatments and delivery studies were performed at the Southwest Foundation for Biomedical Research, San Antonio, Texas. All animal husbandry, animal handling, and procedures were reviewed and approved to conform with the American Association for Accreditation of Laboratory Animal Care guidelines as detailed in the Guide for the Care and Use of Laboratory Animals (National Research Council). Pregnancies were dated using cycle dates and growth parameters from prenatal ultrasounds at 70 and 120 days estimated fetal gestational age. The pregnant baboons were sedated with intramuscular ketamine (10 mg/kg) for each of prenatal treatments. Amniocentesis was performed under ultrasound guidance at 70 days fetal gestational age, and Y-specific DNA amplification of cultured amniocytes was used to determine fetal gender (27). Male fetuses were used in an attempt to minimize variation due to possible differential maturation between males and females. At 124 ± 2 days gestation (term 185 days), the fetuses were again imaged using ultrasound...
after maternal sedation with 10 mg/kg ketamine. Each animal was randomly assigned to receive betamethasone (Celestone Soluspan; Schering Pharmaceuticals, Kenilworth, N. J.) at a fetal dose of 0.5 mg/kg estimated fetal weight in 1 ml or an equivalent volume of vehicle (0.15 M saline). Each treatment was injected under ultrasound guidance into the fetal thigh with a 22-gauge spinal needle. The female baboon was then returned to her cage.

Delivery

The pregnant baboons were sedated with ketamine (10 mg/kg im) 24 h after fetal injection, intubated, and anesthetized with 1.5% halothane. The preterm fetuses were delivered by cesarean section, weighed, and intubated using 2.0- or 2.5-mm endotracheal tubes. The newborns received 100 mg/kg surfactant (Survanta, Ross Products, Columbus, OH) by tracheal instillation, and ventilation was initiated using pressure-limited infant ventilators. After delivery of the fetus and repair of the maternal incisions, recovery of the female baboons was monitored daily for 2 wk, with all animals released to outside gang cages after 4 wk.

Management

The newborn baboons were maintained sedated with intramuscular ketamine (10 mg/kg) and intravenous diazepam (0.1–0.2 mg/kg) if needed. An arterial catheter was placed percutaneously via the saphenous vein into the inferior vena cava for administration of fluids and drugs. All animals received a single intravenous injection of [3H]inulin (6 µCi) for measurements of GFR (9). The animals were cared for on servo-controlled infrared warmers. The animals were not fed and were given parenteral fluids containing amino acids and multivitamins. Intravenous fluids were administered with appropriate electrolytes at 12.5 ml·kg⁻¹·h⁻¹ initially, with infusion rates increased when heart rates were >180 beats/min or for hematocrit increases of >10% or with a small cardiac silhouette on the chest radiograph and an increasing base deficit. Sodium bicarbonate was administered (2 meq/kg) when the base deficit exceeded ~8 meq/kg, Ampicillin (50 mg·kg⁻¹·day⁻¹ in 2 divided doses) and gentamycin (5 mg·kg⁻¹·day⁻¹ in 2 divided doses) were given intravenously. Local anesthesia with 2% lidocaine and additional ketamine were administered for any invasive procedures.

Blood sampling and cardiovascular measurements. In addition to routine arterial blood samples for assessments of pH, PO₂, PCO₂, and hematocrit, blood samples (4–5 ml) were collected from the umbilical cord at delivery and at 2, 6, 12, 18, and 24 h for plasma electrolytes, osmolality, [3H]inulin concentrations, and hormone analysis. Not all measurements were made at all times to minimize blood sampling. An additional plasma sample (1 ml) for determination of plasma catecholamine levels was collected 30 min after delivery. Blood gases and hematocrit values also were monitored at regular intervals. Blood samples were placed volumetrically with heparinized adult baboon blood. Arterial blood pressure, heart rate, oxygen saturation, and electrocardiogram were monitored continuously with a Gould P23 pressure transducer (Gould Instrument Systems, Cleveland, OH) and a cardiorespiratory monitor (model 700; Biomedical Systems, Branford, CT).

Pulmonary management and measurements. To standardize management, ventilatory rate was held constant at 40 breaths/min, and the peak end-expiratory pressure was set at 4 cmH₂O, with an inspiratory time of 0.6 s. Arterial Pco₂ values within the target range of 35–45 mmHg were maintained by adjusting peak inspiratory pressures. Oxygenation was regulated by adjusting inspired oxygen content. The arterial/alveolar (A/a) gradient was calculated as PO₂/(FiO₂ × 713) – (PCO₂/0.8), where FiO₂ is the fractional concentration of inspired O₂. 713 is the atmospheric pressure corrected for the partial pressure of water vapor at physiological temperature, and 0.8 is the respiratory quotient. Tidal volumes and dynamic compliances were measured with a VT1000 Vital Station Neonatal Plethysmograph (VitalTrends Technology, Wallingford, CT). The dynamic compliance was calculated as C = dP/dV, where dP is the difference between the airway pressures at the end and beginning of inspiration and dV is the difference between the airway volumes at the end and beginning of inspiration.

At 22 h, each newborn received a single intravenous injection of [125I]-labeled albumin for postnatal assessment of lung protein leak. After 24 h of ventilation, each baboon received 50 mg/kg pentobarbital sodium to achieve deep anesthesia and was ventilated for 2 min with 100% O₂. The endotracheal tube was disconnected from the ventilator to allow passive deflation of the lungs and was clamped. Cardiac activity continued for 2 min to permit absorption atelectasis and was followed by pentobarbital (50 mg/kg) and exsanguination. Static deflation pressure-volume curves were measured in situ in the open chest by inflating the lungs with air to 35 cmH₂O, followed by sequential volume measurements with incremental decreases in pressure as previously outlined in premature newborn lambs (6). The lungs were then removed, weighed, and thoroughly lavaged with cold saline (16). Alveolar wash protein and lung homogenate protein and hemoglobin levels were assessed to determine postnatal lung protein leak (17).

Renal measurements. Urine samples were collected continuously into an inverted syringe barrel placed around the newborn penis, and the total volume of urine produced was measured at 4-h intervals during the 24 h of study. Urine samples were assessed for osmolality, electrolytes (Na, K, and Cl) and [3H]inulin specific activity. The GFR was determined from the calculated plasma [3H]inulin clearance.

Analytic Techniques

Blood pH, PO₂, and PCO₂ values were determined with a model 995 blood gas analyzer (AVL Scientific, Roswell, GA). Plasma and urine osmolalities were measured by freezing point depression (Advanced Digimatic Osmometer, model MO; Advanced Instruments, Needham Heights, MA). Blood and urine electrolyte (Na, K, and Cl) concentrations were determined with a Nova electrolyte analyzer (Nova Biomedicals, Waltham, MA). Plasma and urine [3H]inulin specific activities were assessed by measuring radioactivity in aliquots (0.1 ml) of plasma and urine.

Blood samples were divided immediately after withdrawal into chilled test tubes, vortexed, and centrifuged immediately. Plasma aliquots were frozen (–20°C) for determinations of cortisol, thyroxine (T₄), triiodothyronine (T₃), argi- nindeaminase (ARG), angiotensin II (ANG II), and atrial natriuretic factor (ANF) levels using radioimmunoassay kits (Nichols Diagnostics, San Juan Capistrano, CA) standardized for fetal plasma. Plasma AVP extraction and radioimmunoassay (RIA) were performed as previously de-
scribed (10, 36); assay sensitivity is 0.8 pg of AVP per tube, with intra- and interassay coefficients of variation of 6 and 9%, respectively. Plasma ANF levels were determined by RIA, with an assay sensitivity of 2 pg/tube and intra-assay and interassay coefficients of variation of 11 and 13%, respectively (11). Plasma ANG II levels were determined from the ANF plasma extracts by use of RIA kits obtained from Peninsula Laboratories (Belmont, CA). Intra-assay and interassay coefficients of variation for the ANG II assay averaged 6 and 9%, respectively, with an overall assay sensitivity of 2 pg/tube. PRA was determined with commercially available reagents, and plasma aldosterone levels were determined by use of RIA kits obtained from ICN Radiochemicals (Costa Mesa, CA). Plasma catecholamine levels were determined by radioenzymatic assay (30).

Data Analysis

All values are expressed as means ± SE. Differences over time and differences between saline and betamethasone-treated groups were assessed by two-way repeated-measures analysis of variance (ANOVA), with time as the between-subjects factor and treatment as the among-subjects factor. Multiple comparisons to identify differences among groups were conducted with the Student-Neuman-Keuls procedure. Paired or unpaired t-tests were also used as appropriate. Statistical significance was accepted at P < 0.05.

RESULTS

Pulmonary

There were no differences in mean body weights between the control (0.41 ± 0.01 kg) and betamethasone-treated (0.45 ± 0.02 kg) preterm newborn baboons. Indexes of ventilatory status (arterial PCO₂, A/a gradient, and dynamic compliance) did not differ between control and betamethasone-treated preterm newborn baboons over the 24-h study period (Fig. 1). At 24 h, there were no differences, respectively, between control and betamethasone-treated values for arterial pH (7.27 ± 0.03 vs. 7.27 ± 0.03), PO₂ (59 ± 8 vs. 62 ± 4 mmHg), PCO₂ (43 ± 5 vs. 42 ± 2 mmHg), peak inspiratory pressure (25 ± 2 vs. 25 ± 2 cmH₂O), or maximal lung volumes measured at 35 cmH₂O (17 ± 4 vs. 15 ± 4 ml/kg body wt). Radiolabeled albumin accumulated equivalently in alveolar wash (0.6%) and lung tissue (~2%), indicating minimal development of lung injury. Dry-to-wet lung weights were 0.141 ± 0.019 and 0.141 ± 0.005 g/g in control and betamethasone-treated animals, respectively. Prenatal betamethasone exposure did not alter overall postnatal lung function.

Cardiovascular, Electrolytes, and Catecholamines

Mean blood pressure values were not different between control and betamethasone-treated newborns 2 h after delivery (Fig. 2). However, by 12 h, mean blood pressure was significantly higher in the betamethasone-treated group and remained higher at 24 h (Fig. 2). Mean heart rate values significantly increased in both groups between 2 and 12 h of life and remained elevated at 24 h. Over the 24-h observation period, least-squares regression analysis revealed an inverse relationship between heart rate and mean blood pressure in both groups, suggesting the presence of an intact baroreflex response in these very premature newborn baboons. Although heart rate values were significantly higher in the control animals after 12 h of ventilation, heart rate values were not different between the two study groups at 24 h.

At delivery, umbilical cord blood hematocrit and plasma osmolality, sodium, and potassium values were similar between control and betamethasone-treated preterm newborn baboons (Table 1). However, plasma chloride concentrations were significantly higher in the betamethasone-treated lambs. Because plasma sodium and chloride concentrations decreased significantly in
the control newborns during the 24 h of ventilation, plasma sodium and chloride concentrations were significantly higher in the betamethasone-treated baboons relative to controls (Fig. 2 and Table 1). There were no differences in total fluid administered, hematocrit, plasma osmolality, or plasma potassium concentrations between control and betamethasone-treated baboons at any time during the 24 h of ventilation.

Mean umbilical cord plasma epinephrine and norepinephrine levels were low and were not different between control and betamethasone-treated animals (Fig. 3). Plasma epinephrine and norepinephrine levels significantly increased in both groups by 30 min after delivery, remained elevated at 2 h, and were further elevated by 24 h. Plasma epinephrine and norepinephrine levels were similarly elevated in both groups from 2 to 24 h.

Renal

The measurements of renal function are summarized in Table 2 and Fig. 4. During the first 12 h, GFR values were not different between groups. By 16 h, GFR was significantly higher in the betamethasone-treated lambs and remained above the control values at 24 h. Urine flow values also were significantly higher in the betamethasone-treated baboons during the final urine collection period (20–24 h), and urine osmolalities and sodium concentrations were significantly lower than in the control animals (Table 2). In the betamethasone-treated baboons, total sodium reabsorption was significantly higher (Fig. 4), and osmolar clearance per 100 ml GFR values were significantly below (Table 2) the control animal values. Free water clearance also was significantly elevated in the betamethasone-treated baboons.

Table 1. Control and betamethasone-treated premature newborn baboon plasma osmolality, electrolyte, and hormone values at delivery and after 24 h of ventilation

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control Cord</th>
<th>Betamethasone Cord</th>
<th>Control 24 h</th>
<th>Betamethasone 24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hematocrit, %</td>
<td>43.4 ± 0.7</td>
<td>46.0 ± 0.9</td>
<td>40.8 ± 1.5</td>
<td>41.2 ± 2.5</td>
</tr>
<tr>
<td>Osmolality, mosmol/kgH2O</td>
<td>292 ± 5</td>
<td>294 ± 1</td>
<td>289 ± 5</td>
<td>294 ± 3</td>
</tr>
<tr>
<td>Sodium, meq/l</td>
<td>137 ± 2</td>
<td>141 ± 1</td>
<td>132 ± 2*</td>
<td>138 ± 1t</td>
</tr>
<tr>
<td>Potassium, meq/l</td>
<td>4.4 ± 0.2</td>
<td>4.9 ± 0.6</td>
<td>4.5 ± 0.3</td>
<td>4.3 ± 0.2</td>
</tr>
<tr>
<td>Chloride, meq/l</td>
<td>108.1 ± 1</td>
<td>112 ± 1</td>
<td>104 ± 1*</td>
<td>111 ± 1t</td>
</tr>
<tr>
<td>Triiodothyronine, µg/dl</td>
<td>55.1 ± 8.2</td>
<td>66.9 ± 11.1</td>
<td>41.2 ± 3.9</td>
<td>48.6 ± 3.6</td>
</tr>
<tr>
<td>Atrial natriuretic factor, pg/ml</td>
<td>37 ± 8</td>
<td>17 ± 10</td>
<td>63 ± 14</td>
<td>83 ± 13*</td>
</tr>
<tr>
<td>Plasma renin activity, ng/ANG l·ml⁻¹·h⁻¹</td>
<td>6.4 ± 1.8</td>
<td>19 ± 10</td>
<td>131 ± 27*</td>
<td>90 ± 31*</td>
</tr>
<tr>
<td>Aldosterone, pg/ml</td>
<td>65 ± 17</td>
<td>215 ± 104</td>
<td>90 ± 29</td>
<td>398 ± 90†</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 5 animals for both control and betamethasone. *Different from umbilical cord (Cord) value, P < 0.05; † different from control value, P < 0.05.

Table 2. Control and betamethasone-treated premature newborn baboon renal parameters after 24 h of ventilation

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>Betamethasone</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urine Flow, ml·min⁻¹·kg⁻¹</td>
<td>2.84 ± 1.4</td>
<td>5.59 ± 1.25*</td>
</tr>
<tr>
<td>Osmolality, mosmol/kgH₂O</td>
<td>308 ± 6</td>
<td>213 ± 33*</td>
</tr>
<tr>
<td>Sodium, meq/l</td>
<td>137 ± 6</td>
<td>88 ± 14*</td>
</tr>
<tr>
<td>Potassium, meq/l</td>
<td>7.13 ± 0.46</td>
<td>5.04 ± 1.09</td>
</tr>
<tr>
<td>Potassium excretion, µeq·min⁻¹·kg⁻¹</td>
<td>3.04 ± 1.06</td>
<td>4.48 ± 1.01</td>
</tr>
<tr>
<td>Osmolar clearance, ml/100 ml GFR</td>
<td>44 ± 10</td>
<td>20 ± 5*</td>
</tr>
<tr>
<td>Free water clearance, ml/min</td>
<td>0.015 ± 0.017</td>
<td>0.063 ± 0.016*</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 5 animals for both control and betamethasone. GFR, glomerular filtration rate. *Different from control, P < 0.05.

Fig. 3. Plasma epinephrine and norepinephrine levels in control and betamethasone-treated premature newborn baboons ventilated for 24 h.

Fig. 4. Glomerular filtration rate (GFR) and total sodium reabsorption in premature newborn baboons delivered 24 h after fetal saline or betamethasone (0.5 mg/kg) treatment and ventilated for 24 h. *Different from control, P < 0.05.
animals (Table 2). Although control animal urine potassium concentration and urinary potassium excretion values were 40% higher and 32% lower, respectively, than the betamethasone-treated animal values, the values were not statistically different between groups (Table 2).

Endocrine

Plasma cortisol levels were similar between groups at delivery (Fig. 5). By 24 h, there were similar significant decreases in plasma cortisol levels in both groups. T4 levels also were similar between control and betamethasone-treated newborns at delivery, and in both groups plasma T4 levels significantly decreased. Umbilical cord plasma T3 levels were similar in both groups after delivery and did not change in either group at 24 h.

Umbilical cord plasma endocrine values including AVP, ANF, and ANG II and aldosterone and PRA values were not different between control and betamethasone-treated newborns (Fig. 5 and Table 1). Relative to the umbilical cord values, plasma AVP, ANG II, and PRA levels increased significantly in both groups at 24 h, but there were no differences between control and betamethasone-treated groups. Plasma ANF levels were not different between groups at delivery. Although there was a statistically significant increase in plasma ANF levels in the betamethasone-treated group by 24 h, plasma ANF levels did not differ between groups. Mean umbilical cord and 24 h plasma aldosterone levels did not differ between groups. However, plasma aldosterone levels were significantly elevated in the betamethasone-treated newborns compared with controls at 24 h (Table 1).

DISCUSSION

In these very premature newborn baboons given prenatal glucocorticoids as a single fetal dose 24 h before delivery, an increase in postnatal blood pressure and improved kidney function were evident after 12 h postnatal age and were sustained to 24 h of postnatal life. However, the anticipated effect of improved lung function consistently observed after prenatal glucocorticoid exposure in preterm sheep was absent. Several possible explanations for this apparent absence of pulmonary effects in the premature baboon model warrant consideration. First, the present preterm newborn baboon studies are unique because the animals were studied very early in gestation (0.67), earlier than previously reported for most other species. In preterm sheep, prenatal glucocorticoids improve renal, cardiovascular, and lung function after maternal or fetal treatments after ~0.8 of gestation (6, 32). Postnatal effects of prenatal glucocorticoid treatment before 120 days (0.8) gestation have not been evaluated in sheep because survival is limited. Thus pulmonary effects may not have been detected in the present studies because the preterm baboons at 0.67 of gestation were too immature to respond. In apparent contradiction to this hypothesis, striking alterations in lung structure follow perterm or term delivery of monkeys exposed to high-dose glucocorticoids over 3 days at midgestation (4). Although species differences in timing of responsiveness might be one explanation, a second possibility is that a treatment-to-delivery interval of 24 h may be insufficient in the baboon to manifest lung maturational effects. In sheep delivered at 0.85 of gestation, glucocorticoid-induced effects on lung function occur within 15 h of fetal or maternal treatment with 0.5 mg/kg betamethasone. In both baboons and monkeys, glucocorticoids also enhance lung function, but only paradigms of 3 days of prenatal maternal glucocorticoid exposure have been studied (18, 19). Thus glucocorticoid treatment-to-delivery intervals of >24 h may be necessary to initiate improvements in lung function in primates. In favor of this explanation is the lack of...
effect of prenatal glucocorticoids on the incidence or severity of respiratory distress syndrome in the human (7) and the observation that the blood pressure and renal effects detected in the current studies were not apparent until 12 h after delivery. It is possible that the postnatal cardiovascular and renal responses observed were delayed relative to the time of delivery because of the short treatment-to-delivery interval.

A third explanation for the absence of pulmonary effects is that lung function in both glucocorticoid-treated and control animals was favorably influenced to a similar degree by the stress associated with repetitive maternal anesthesia and ultrasound examination. In the ovine model, a single maternal exposure to anesthesia and laparotomy can increase circulating fetal cortisol levels sufficiently to induce fetal lung maturation (34). Thus, in our attempt to optimize the quality control of the pregnant baboons through ultrasound examinations performed at 70 and 120 days for measurements of fetal growth, and again at 124 days for fetal therapy, the procedures alone may have been sufficient to evoke a stress response in both groups. Consistent with this hypothesis, the umbilical cord cortisol levels (Fig. 5) in the control animals were high relative to reported newborn sheep or human values and also higher than anticipated values for early gestation controls in this species, given the limited fetal potential for cortisol production at this early gestation (25). Another indicator consistent with chronic fetal stress was the high umbilical cord plasma T3 value. Thus handling and anesthesia of the pregnant baboons may have been sufficient to evoke similar elevations in cortisol and T3 values in both control and betamethasone-treated animals, thereby enhancing lung maturation in both groups. The predominant source of the cortisol in the umbilical cord blood may have been from the mother via placental transfer (25). Assuming the latter hypothesis is correct (induced lung maturation in both groups), it is interesting to note that a single fetal dose of betamethasone was associated with marked effects on preterm newborn blood pressure and kidney function.

Despite a lack of differences in lung function, the betamethasone-treated preterm baboons were characterized by significantly higher mean arterial blood pressure values by 12 h after delivery, and this trend continued through 24 h. The higher plasma sodium concentration in these animals may have been sufficient to sustain intravascular volume better than that of the control animals (22). Because fluid administration was similar in both groups, the higher plasma sodium concentration and larger urinary losses in the betamethasone-treated animals suggest that dehydration might account for the higher plasma sodium concentration. However, the similar hematocrit values and higher rate of sodium reabsorption suggest that overall sodium management was altered in favor of a higher plasma sodium concentration. Thus one speculation is that glucocorticoid-induced changes in vascular permeability may have contributed to a decrease in extravascular fluid volume, consistent with the postnatal adaptation pattern in term newborns.

Glucocorticoids also affect vascular reactivity and vasomotor tone. For example, glucocorticoid administration increases overall cardiac contractility, vascular tone, and cardiovascular receptor expression for catecholamines and ANG II (28, 35). In premature newborn sheep, single-dose fetal or maternal prenatal betamethasone treatment increases mean arterial blood pressure despite causing marked suppression of circulating catecholamines and ANG II levels. Thus, in the lamb, betamethasone-induced increases in catecholamine and ANG II receptor expression and/or postreceptor response mechanisms may augment overall vascular responsiveness to circulating vasoactive agents even when circulating levels are reduced. However, the response pattern was different in these premature newborn baboons. The betamethasone-treated newborns had higher mean blood pressure values with no differences in circulating catecholamine and ANG II levels between betamethasone and control groups. Rather than being suppressed as in the lamb, catecholamine and ANG II levels increased over 24 h.

At birth, the kidneys must rapidly shift from a fetal pattern of high fractional excretion of water and sodium to highly efficient water and sodium conservation. In term human newborns, adaptations including marked increases in GFR and sodium reabsorption are well underway within 24–48 h after delivery. Although preterm newborns also will eventually adapt appropriately if maintained viable for a sufficient period of time (3), delays in shifting from the fetal pattern of excessive water and sodium losses relative to the low GFR often predispose preterm infants to hyponatremia and hypovolemia. Glucocorticoid-induced kidney maturation (increases in GFR and sodium reabsorption) has been reported previously in fetal (15) and near-term newborn sheep (31) in response to prolonged maternal administration. We have also reported increases in GFR and sodium reabsorption in preterm newborn lambs delivered 24 or 48 h after single fetal or maternal betamethasone treatment (2, 9). An acute effect of glucocorticoids to increase GFR is widely accepted and principally reflects selective renal near-parallel afferent and efferent vessel vasodilation to increase glomerular plasma flow and filtration fraction (1). Although postnatal increases in GFR also reflect increases in filtration fraction, the exact mechanism(s) have not been defined. The two- to threefold increase in GFR noted in the betamethasone-treated baboons (Fig. 4) is not attributable to the higher blood pressure in these animals because blood pressure did not increase concurrently with the increase in GFR.

Prenatal glucocorticoid treatment in sheep also increases postnatal sodium reabsorption (31, 32). Proximal rather than distal tubular function appears to be the principal location limiting fetal and perhaps preterm newborn sodium reabsorption (21). Glucocorticoid-induced increases in preterm newborn sodium reabsorption may reflect a direct effect to increase proximal tubular sodium transporter function and expression
(14) and an indirect effect to increase basolateral Na-K-ATPase activity (8). Thus glucocorticoid-induced changes in proximal tubular function may also be important in the premature newborn baboon. However, the significantly higher plasma aldosterone levels (Table 1) measured in the betamethasone-treated fetuses suggest that increases in distal tubular function may have contributed to the overall increase in renal sodium reabsorption (Fig. 4). Given the higher mean blood pressure and GFR observed in the betamethasone-treated premature newborn baboons and the marked suppression of the renin-ANG II-aldosterone axis observed in newborn lambs after prenatal glucocorticoid treatment, the basis for the heightened renin-ANG II-aldosterone axis activity measured in the betamethasone-treated preterm newborns is not clear. Nonetheless, the present results indicate that glucocorticoid exposure for as little as 24 h before delivery can significantly improve preterm newborn baboon renal sodium handling and plasma electrolyte regulation.

The umbilical cord plasma cortisol levels were not suppressed in the betamethasone-exposed fetuses, a surprising result suggesting lack of feedback inhibition, a long plasma cortisol half-life at this gestation, or perhaps simply placental cortisol transfer. The very low umbilical cord plasma catecholamine and AVP levels (Figs. 3 and 5) indicate that the preterm baboons were not "stressed" at the time of delivery. The absence of an effect of antenatal betamethasone exposure on umbilical cord plasma ANG II levels is consistent with fetal sheep data demonstrating that cortisol-induced suppression of fetal angiotensinogen production is gestation dependent (23). The pronounced increases in catecholamines, AVP, and ANG II levels and PRA at 24 h indicate the significant stress associated with preterm delivery and postnatal ventilation. In contrast, cortisol and T3 levels had decreased similarly in both groups by 24 h, with no changes in plasma T3 levels. In fetal sheep, the thyroid axis response to glucocorticoids differs in that T3 levels increase with little change in T4 levels (5, 26). However, a thyroid response to exogenous corticosteroid exposure is variably seen in humans (24). Although changes in cortisol and thyroid hormone metabolism were likely important to the changes in circulating levels noted between delivery and 24 h of postnatal life, these patterns did not appear to be influenced by prenatal betamethasone exposure (Fig. 5).

In summary, these are unique data in very preterm baboons for which there is no comparison information available. A limitation to studies in the baboon is the maternal response to handling and the need for sedation and/or anesthesia associated with procedures. Nonetheless, these studies provide the first integrative information regarding premature newborn pulmonary, cardiovascular, renal, and endocrine responses during postnatal adaptation and the effects of fetal therapy in the severely premature (0.67 gestation) newborn primate. A single fetal dose of betamethasone as little as 24 h before delivery can have pronounced effects on preterm newborn GFR, sodium reabsorption, plasma sodium regulation, and blood pressure stability. The interpretative difficulty is that we do not know how the fetal environment (high cortisol and T3) may have modulated the betamethasone effects. This difficulty is also common to clinical situations in which many preterm deliveries are associated with chronic and/or acute fetal stress. The clinically important conclusion is that prenatal glucocorticoids may augment postnatal adaptation even if the fetus has been stressed in utero and is at a decreased risk of respiratory distress syndrome.

Perspectives

The now widely accepted practice of prenatal glucocorticoid administration in cases of threatened premature labor has profoundly improved postnatal outcomes in premature infants. Nonetheless, prematurity-associated diseases remain the leading cause of infant morbidity and mortality in the United States. Studies assessing prenatal glucocorticoid exposure in the premature newborn model may provide insight into why some infants apparently fail to respond to prenatal glucocorticoid exposure. In addition, glucocorticoids influence a diverse array of systems and functions. However, the actual effect(s) and the sequence of events that improve or augment postnatal adaptation are not known. Thus studies of preterm newborn adaptation and the diverse effects of prenatal glucocorticoid exposure on this process represent an important step toward optimizing therapeutic approaches to improve postnatal outcomes in premature infants.

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