Effect of ventilation style on cardiovascular and renal adaptation in preterm newborn lambs

NORIHISA WADA, M. GORE ERVIN, AND MACHIKO IKEGAMI
Perinatal Research Laboratories, Departments of Pediatrics and Obstetrics and Gynecology, University of California, Los Angeles School of Medicine, Harbor-University of California-Los Angeles Medical Center, Torrance, California 90502

Effect of ventilation style on cardiovascular and renal adaptation in preterm newborn lambs. Am. J. Physiol. 275 (Regulatory Integrative Comp. Physiol. 44): R836–R843, 1998.—Renal adaptive responses during the 24 h after delivery in term newborn lambs include marked increases in both glomerular filtration rate (GFR) and sodium reabsorption. This study investigated the effects of ventilation style on cardiovascular, renal, and endocrine adaptations in preterm newborn lambs. Lambs (n = 62) were delivered by cesarean section at 131 days gestation (term = 150 days), treated with surfactant, and randomized to one of three ventilation strategies: high-frequency oscillation (12 Hz), high rate (50 breaths/min; tidal volume = 8 ml/kg), or low rate (15 breaths/min; tidal volume = 15 ml/kg). Lambs (5 or 6/group) were ventilated for 2, 5, 10, and 24 h to maintain arterial PCO2 between 45 and 50 mmHg. Plasma vasopressin levels decreased to <25 pg/ml by 10 h, and fractional sodium excretion decreased to <1% by 16 h in all groups. However, cardiac output, renal plasma flow, and GFR values did not change over time for any of the groups. The style of ventilation employed had no measurable effects on overall cardiovascular, renal, or endocrine function. We conclude in ventilated preterm lambs that 1) the ventilation style does not affect the time course for postnatal adaptation, 2) adaptive changes in renal tubular sodium reabsorption are evident by 16 h after birth, and 3) changes in preterm newborn renal sodium reabsorption occur in the absence of postnatal changes in renal plasma flow or GFR.

Methods

Study groups. All animal handling, surgical procedures, and study protocols were reviewed and approved by the Harbor-University of California-Los Angeles Animal Care and Use Review Committee.

Pregnant Western mixed breed ewes were obtained from the Nebecker ranch (Palmdale, CA). Sixty-two 130 ± 1 day gestation preterm lambs (term = 150 days) were randomized to one of three ventilation strategies: ventilation with high-frequency oscillation (HFOV), conventional intermittent mandatory ventilation with high rates of 50 breaths/min and small tidal volumes of ~8 ml/kg (R50), or conventional intermittent mandatory ventilation with low rates of 15 breaths/min and large tidal volumes of 15 ml/kg (R15). Five to seven lambs from each group were ventilated for 2, 5, 10, or 24 h. The numbers of animals were HFOV: n = 21 (2h 5, 5 h = 5, 10 h = 6, 24 h = 5); R50: n = 20 (2 h = 5, 5 h = 5, 10 h = 5, 24 h = 5); and R15: n = 21 (2 h = 5, 5 h = 5, 10 h = 6, 24 h = 5) for a total of 62 animals. Physiological lung function and surfactant metabolism were also investigated and reported elsewhere (11).

Delivery of preterm lambs. Preterm lambs were delivered by cesarean section as previously described (22). Briefly, ewes were sedated with ketamine (20 mg/kg im) and after spinal epidural anesthesia [10 ml 2% lidocaine–0.5% marcarene 1:1 (vol/vol)] the fetal head and neck were exteriorized through abdominal and uterine incisions. The fetus was sedated with a ketamine (10 mg/kg im) and acepromazine (0.1 mg/kg im) mixture administered on the basis of estimated fetal body weight, and sedation was maintained by repeated ketamine–acepromazine administration every 4 h. Although possible adverse effects of sedation cannot be excluded, this ketamine–acepromazine mixture has been used extensively without apparent effects on newborn blood pressure, heart rate, or cardiac output (10–14, 22). After local anesthesia with 2% lidocaine, the trachea was exposed and a 4.5 mm ID tracheal tube was tied into the trachea. Tracheal fluid was aspirated with gentle negative pressure by syringe, and the endotracheal tube was clamped. The umbilical cord was clamped and cut, and blood was collected from the placental umbilical vein into heparinized syringes for pH, blood gas determinations, and newborn transfusion. After delivery, the lambs were weighed, dried, and treated with 100 mg/kg lipid-extracted sheep surfactant (12). Surfactant was given via direct intratracheal instillation before the first breath. Lambs were ventilated by one of three ventilation strategies. HFOV
animals were ventilated using a high-frequency oscillatory ventilator (Sensormedics, Anaheim, CA) with initial settings of fractional inspired O₂ (FＩ₂, O₂) of 1.0, MAP of 15 cmH₂O, and frequency of 12 Hz. FＩ₂, O₂ and amplitude were changed to maintain PaＯ₂, values of 200 mmHg and PaＣO₂, values in the 45- to 50-mmHg range. Both R50 and R15 animals were ventilated using time-cycled pressure-limited neonatal ventilators (Sechrist Industries, Anaheim, CA) with initial settings of positive end-expiratory pressure (PEEP) of 3 cmH₂O, inspiratory time of 1.0 s, rate of 30 breaths/min, and FＩ₂, O₂ of 1.0. Peak inspiratory pressure (PIP) was adjusted to maintain tidal volumes of 10 ml/kg for the first 15 min after birth. Afterward, R50 animal ventilator settings were inspiratory time of 0.4 s and rate of 50–60 breaths/min, with PIP adjusted to maintain tidal volumes of ~8 ml/kg. Ventilation parameters for the R15 animals included inspiratory time of 0.7 s and rate of 15–25 breaths/min, with PIP adjusted to maintain tidal volumes of ~15 ml/kg. R50 and R15 settings for FＩ₂, O₂, PIP, and expiratory time were adjusted to maintain PaＯ₂, values of 200 mmHg and PaＣO₂, values of 45–50 mmHg.

After initiation of ventilation, a 5.0-Fr catheter was placed in the descending aorta via an umbilical artery for blood sampling and blood pressure monitoring. A single 10 µCi dose of [methoxy-3H]inulin (DuPont-NEN, Boston, MA) was injected and followed by continuous infusion of 0.25 µCi·kg⁻¹·h⁻¹ for determinations of plasma inulin clearance. All lambs also received a 10 ml/kg transfusion of filtered umbilical cord blood at this time. A urinary bladder catheter was placed by suprapubic percutaneous cytostomy to permit timed urine collections. Sixty minutes before the end of the study the skin on one side of the neck was infiltrated with 2% lidocaine, either a left or right carotid artery was isolated, and a 5.0-Fr catheter was inserted into the left ventricle for injection of microparticles. Fluid administration included continuous infusion of 5% dextrose in water containing 0.25 µCi/ml [methoxy-3H]inulin and 0.9% saline for a total volume of 120 ml·kg⁻¹·day⁻¹ via the umbilical artery. Fluid administration was increased from our usual levels of 104 ml·kg⁻¹·day⁻¹ (2, 14, 22) to the higher 120 ml·kg⁻¹·day⁻¹ rate because the higher air flows used in high-frequency oscillatory ventilation are associated with increased fluid loss through the airways (6). Aortic blood pressure and heart rate were monitored continuously via pressure transducers connected to a recorder (R-612, Beckman Instruments, Fullerton, CA). Ventilation pressures in R50 and R15, including PIP, PEEP, and MAP, were monitored with an airway pressure monitor (model 400, Sechrist Industries, Anaheim, CA). In the HFOV groups, MAP values indicated by the respirator were recorded. Ventilatory parameter values were recorded whenever the conditions were changed. Tidal volumes in the R50 and R15 groups were monitored continuously by a pulmonary monitor (CP-100, Bicore Monitoring Systems, Irvine, CA). Arterial blood gas (PaO₂, PaCO₂) and pH values determined on a blood gas analyzer (Stat Profile 3, NOVA Biomedical, Waltham, MA) were assessed at least every 30 min and 15 min after any changes in ventilator settings to monitor ventilation and metabolic status.

At the end of the studies, the lambs were deeply anesthetized with pentobarbital sodium (25 mg/kg iv) and killed by exsanguination via cutting of the abdominal aorta. The dry body weight was measured and used for all calculations.

Renal function assessment. Serial urine samples were collected every 30 min during the first 2 h, every 60 min through 10 h, and every 120 min through 24 h. Urine samples were assessed for urine flow, osmolality, electrolyte concentrations, and inulin-specific activity. Arterial blood samples were obtained at 2, 5, 10, 16, and 24 h for determination of hematocrit, plasma osmolality, and electrolyte (Na, K, Cl) concentrations, and plasma inulin-specific activity. Blood samples were transferred immediately into chilled test tubes containing lithium heparin (10 U/ml blood), and tubes were vortexed and centrifuged at 4°C. Plasma and urine sodium, potassium, and chloride concentrations were determined on an electrolyte analyzer (NOVA Biomedical, Waltham, MA), and osmolalities were determined by freezing-point depression (Advanced Digmatic Osmometer, model MO, Advanced Instruments, Needham Heights, MA). When urine sodium concentrations were below the sensitivity range of the analyzer, the lower limit of detection (1 mmol/l) was used for all calculations. Plasma and urine [methoxy-3H]inulin-specific activities were assessed by counting aliquots (100 µl) in Ecoscint scintillation solution (National Diagnostics, Atlanta, GA) in a 1214 Rackbeta liquid scintillation counter (Wallac, Gaithersburg, MD). GFR was determined from the calculated [3H]inulin clearance.

Cardiovascular function assessment. Cardiac output was assessed 30 min before the end of studies by injection into the left ventricle of a known activity of 57Co-labeled microspheres (5.5 ± 0.1 µCi/ml; DuPont-NEN) (9) mixed in 2 ml of filtered maternal blood. Renal blood flow was assessed by postmortem measurements of 57Co-microspheres in both kidneys. Blood and tissue 57Co-microspheres specific activities were measured with a 1282 Compu gamma Universal gamma counter (Wallac, Gaithersburg, MD). Renal plasma flow values were calculated with renal blood flow values (at 30 min before the end of the study) and hematocrit values at the end of the study. Filtration fraction values were calculated with renal plasma flow and GFR values at the end of the study.

Endocrine function assessment. Blood samples obtained at 2, 5, 10, 16, and 24 h were transferred immediately into chilled test tubes containing either lithium heparin (10 U/ml blood), for determinations of concentrations of arginine vasopressin (AVP), aldosterone, and plasma renin activity (PRA), or aprotinin (500 KiU/ml blood) and K₂EDTA (3 mg/ml blood) for determination of plasma ANG II and plasma atrial natriuretic factor (ANF) concentrations. All tubes were vortexed and centrifuged at 4°C, and plasma samples were frozen at −20°C for assay within 4 wk. Plasma AVP extraction and RIA were performed as previously described (4). The sensitivity of the AVP assay was 0.8 pg of AVP/tube with intra-assay and interassay coefficients of variation of 6 and 9%, respectively. PRA was measured indirectly using the RIANEN ANG I 125I-labeled RIA kit (DuPont-NEN). Plasma aldosterone levels were determined by RIA with kits by ICN Biomedicals (Costa Mesa, CA). Plasma ANG II concentrations were determined with ANG II-specific RIA kits (Peninsula Laboratories, Belmont, CA), with intra-assay and interassay coefficients of variation of 6 and 9%, respectively, and an overall assay sensitivity of 2 pg/tube. Plasma ANF determinations were conducted by RIA as previously described from our laboratory (5).

Statistical analysis. Values are expressed as means ± SE. Differences in gender among the groups were assessed by χ² analysis. Differences over time for the 24-h animals were assessed by repeated-measures ANOVA and post hoc comparisons with the Student-Newman-Keuls procedure. Differences among the groups (including all animals) were assessed by ANOVA and post hoc comparisons by the Student-Newman-Keuls method. Unless indicated otherwise, data presented represent all the animals studied at a given time. Statistical significance for all analyses was accepted at P ≤ 0.05.
RESULTS

There were no differences among the three groups in terms of gender or average final body weight (2.7 kg).

Respiration. $P_{aCO_2}$, $P_{aO_2}$, pH, and MAP values determined at the end of the studies are shown in Table 1 and Fig. 1. There were no differences in $P_{aCO_2}$, $P_{aO_2}$, and pH values among the three groups or over time. However, MAP values were significantly different among the groups at every time point (Fig. 1), with MAP in HFOV > R50 > R15 ($P = 0.05$).

Systemic and renal circulation. Mean arterial blood pressures and heart rates determined at the end of the studies are shown in Fig. 1. There was a statistically significant decrease in mean blood pressure in the R50 group at 10 and 24 h, but no differences among the groups. Heart rate significantly decreased in the R50 and R15 groups. In contrast, changes in heart rate were minimal in the HFOV group, and HFOV heart rate values were significantly higher than in the other two groups at 24 h.

Cardiac output, renal plasma flow, and filtration fraction values at 30 min before the end of studies for each group are summarized in Table 2. Values did not change with time and there were no differences among the groups.

Hematocrit and plasma sodium concentration. Hematocrit and plasma sodium concentration values at the end of the 2-, 5-, 10-, and 24-h studies and 16-h values in lambs studied for 24 h are shown in Fig. 2. There were no changes over time in any group. However, the HFOV group hematocrit value at 24 h was significantly above the R50 group. Although the HFOV group plasma sodium value at 24 h was higher than in the other two groups, the differences were not statistically significant.

Table 1. Arterial blood gas and pH values at the end of ventilation

<table>
<thead>
<tr>
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<th>HFOV</th>
<th>R50</th>
<th>R15</th>
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<tbody>
<tr>
<td>$P_{aCO_2}$, mmHg</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>2 h</td>
<td>48.4 ± 2.7</td>
<td>48.0 ± 5.6</td>
<td>48.8 ± 1.4</td>
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<tr>
<td>5 h</td>
<td>48.2 ± 2.6</td>
<td>49.9 ± 3.3</td>
<td>45.8 ± 1.1</td>
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<tr>
<td>10 h</td>
<td>48.7 ± 4.4</td>
<td>48.4 ± 3.0</td>
<td>47.7 ± 2.9</td>
</tr>
<tr>
<td>24 h</td>
<td>47.6 ± 2.6</td>
<td>52.2 ± 3.3</td>
<td>51.0 ± 2.3</td>
</tr>
<tr>
<td>$P_{aO_2}$, mmHg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 h</td>
<td>213.6 ± 32.2</td>
<td>187.4 ± 18.8</td>
<td>215.6 ± 45.6</td>
</tr>
<tr>
<td>5 h</td>
<td>148.2 ± 39.0</td>
<td>156.1 ± 44.1</td>
<td>190.4 ± 6.4</td>
</tr>
<tr>
<td>10 h</td>
<td>169.8 ± 28.2</td>
<td>232.8 ± 29.5</td>
<td>208.3 ± 11.6</td>
</tr>
<tr>
<td>24 h</td>
<td>158.4 ± 12.4</td>
<td>187.0 ± 16.6</td>
<td>168.6 ± 9.7</td>
</tr>
<tr>
<td>pH</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 h</td>
<td>7.30 ± 0.01</td>
<td>7.30 ± 0.04</td>
<td>7.31 ± 0.01</td>
</tr>
<tr>
<td>5 h</td>
<td>7.29 ± 0.03</td>
<td>7.28 ± 0.02</td>
<td>7.35 ± 0.04</td>
</tr>
<tr>
<td>10 h</td>
<td>7.29 ± 0.03</td>
<td>7.30 ± 0.01</td>
<td>7.31 ± 0.01</td>
</tr>
<tr>
<td>24 h</td>
<td>7.29 ± 0.02</td>
<td>7.26 ± 0.03</td>
<td>7.27 ± 0.01</td>
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</table>

Values are means ± SE; n = 5 animals/group for those ventilated with high-frequency oscillation (HFOV) for 2, 5, and 24 h and 6 for 10 h; 5 for those with conventional intermittent mandatory ventilation with high rates of 50 breaths/min and small tidal volumes of 8 ml/kg (R50) for 2, 5, 10, and 24 h; and 5 for those with conventional intermittent mandatory ventilation with low rates of 15 breaths/min and large tidal volumes of 15 ml/kg (R15) for 2, 5, and 24 h.

Renal function. Urine flow and urine osmolality values are shown in Fig. 3. Urine flow significantly decreased in all groups within 1–2 h after delivery. Urine osmolality values were similarly significantly decreased in all groups within 1–2 h after delivery.

Table 2. Values for cardiac output and renal parameters 30 min before the end of studies for 2, 5, 10, and 24 h

<table>
<thead>
<tr>
<th></th>
<th>HFOV</th>
<th>R50</th>
<th>R15</th>
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<tbody>
<tr>
<td>Cardiac output, ml·kg$^{-1}$·min$^{-1}$</td>
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<tr>
<td>1.5 h</td>
<td>172.2 ± 16.3</td>
<td>210.4 ± 27.1</td>
<td>248.8 ± 47.0</td>
</tr>
<tr>
<td>4.5 h</td>
<td>167.3 ± 11.3</td>
<td>175.2 ± 14.1</td>
<td>194.5 ± 15.6</td>
</tr>
<tr>
<td>9.5 h</td>
<td>175.3 ± 25.4</td>
<td>175.7 ± 14.1</td>
<td>206.9 ± 34.0</td>
</tr>
<tr>
<td>23.5 h</td>
<td>168.5 ± 38.4</td>
<td>165.1 ± 52.4</td>
<td>132.4 ± 25.4</td>
</tr>
<tr>
<td>Renal plasma flow, ml·kg$^{-1}$·min$^{-1}$</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>1.5 h</td>
<td>4.2 ± 0.9</td>
<td>5.8 ± 1.2</td>
<td>4.7 ± 0.8</td>
</tr>
<tr>
<td>4.5 h</td>
<td>3.6 ± 0.4</td>
<td>3.9 ± 0.4</td>
<td>5.1 ± 0.9</td>
</tr>
<tr>
<td>9.5 h</td>
<td>5.5 ± 1.2</td>
<td>5.0 ± 0.8</td>
<td>5.3 ± 0.8</td>
</tr>
<tr>
<td>23.5 h</td>
<td>6.3 ± 1.0</td>
<td>6.3 ± 1.9</td>
<td>4.1 ± 0.9</td>
</tr>
<tr>
<td>Filtration fraction, %</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>1.5 h</td>
<td>12.5 ± 1.1</td>
<td>17.0 ± 2.9</td>
<td>11.8 ± 1.6</td>
</tr>
<tr>
<td>4.5 h</td>
<td>20.8 ± 7.5</td>
<td>9.0 ± 2.8</td>
<td>12.9 ± 3.3</td>
</tr>
<tr>
<td>9.5 h</td>
<td>20.5 ± 4.0</td>
<td>15.7 ± 4.5</td>
<td>20.8 ± 3.2</td>
</tr>
<tr>
<td>23.5 h</td>
<td>11.7 ± 3.7</td>
<td>22.1 ± 11.9</td>
<td>7.4 ± 6.4</td>
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Values are means ± SE; see Table 1 for n values.
increased in all groups at 2 h. Although there was an increasing trend through 6 h, mean urine osmolarity values never exceeded 500 mosmol/kg H2O, and there were no differences among the groups. However, after 12 h, urine osmolarity significantly decreased in the R15 group (differences were not detected by post hoc comparisons) and remained below the HFOV and R50 values through 24 h.

GFR and FENa values are shown in Fig. 4. GFR values were \( \frac{1 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}}{\text{kg}} \) in all groups. Although GFR values tended to be higher at 10 h, there were no significant changes over time and there were no differences among the groups. FENa values decreased to \( <1\% \) by 16 h in all groups. Although there was a significant change over time in the HFOV group, there were no differences among the groups.

Endocrine. Plasma AVP and ANF values are summarized in Fig. 5, and PRA, ANG II, and aldosterone values are summarized in Fig. 6. Plasma AVP values significantly decreased by 10 h in all groups, and there were no statistically significant differences among the groups. Plasma ANF and ANG II values were obtained only from animals studied for 24 h. Although there was an increasing trend in plasma ANF values over time, the increase was statistically significant only in the R50 group. There were no differences among the groups. PRA values increased in the R15 group to a peak value at 10 h, but decreased by 24 h. However, PRA values did not change over time in the HFOV and R50 groups, and there were no differences among the groups. ANG II values did not change over time and did not differ among the groups in animals studied for 24 h. Plasma aldosterone values significantly changed over time only in the R15 group (\( P = 0.04 \)). However, there were no differences in plasma aldosterone values among the groups at 24 h. Plasma ANF and ANG II values were obtained only from animals studied for 24 h. Although there was an increasing trend in plasma ANF values over time, the increase was statistically significant only in the R50 group. There were no differences among the groups. PRA values increased in the R15 group to a peak value at 10 h, but decreased by 24 h. However, PRA values did not change over time in the HFOV and R50 groups, and there were no differences among the groups. ANG II values did not change over time and did not differ among the groups in animals studied for 24 h. Plasma aldosterone values significantly changed over time only in the R15 group (\( P = 0.04 \)). However, there were no differences in plasma aldosterone values among the groups.
aldosterone (C0.05); *changes over time (P and 24-h R15 values (blood pressure < 50 mmHg and heart rate < 150 beats/min) obtained from our previous studies in preterm lambs of similar gestation (2). Decreases in mean blood pressure in the R50 group were statistically significant due to relatively high 2- and 5-h values and low SE at 10 and 24 h. However, the patterns for mean blood pressure and heart rate were similar to cesarean-delivered term newborn lambs, with no changes in mean blood pressure and a decrease in heart rate during the first 24 h (19). The observed decreases in heart rate in the R50 and R15 groups are consistent with previous observations that newborn lamb plasma catecholamine levels are elevated at birth and decrease over time (19, 21). In contrast, heart rate in the HFOV group did not change and was significantly higher at 24 h than in the other two groups. Differences in HFOV-related baroreflex effects do not appear accountable given that recent studies in premature baboons (16), human infants (1), and premature lambs (20) demonstrated no differences in cardiovascular function between high-frequency oscillatory ventilation and conven-

DISCUSSION

In the present study, preterm lambs were delivered by cesarean section at 131 days gestation, treated with surfactant, and ventilated by one of three approaches: high-frequency oscillatory ventilation, conventional high-rate intermittent mandatory ventilation, or conventional low-rate intermittent mandatory ventilation. The ventilation strategies resulted in equivalent gas exchange without indications of lung injury (11). Therefore, we had the opportunity to study cardiovascular, renal, and endocrine adaptations in a preterm newborn model without dealing with differences in respiratory function. This study uniquely provides the first data available regarding long-term (24 h) integrative cardiovascular, renal, and endocrine postnatal adaptive changes in preterm newborn lambs. The renal results include marked decreases in FE\textsubscript{NO} by 16 h and a notable absence of GFR changes in all groups, with minimal differences among the groups in overall cardiovascular, renal, and endocrine adaptive responses.

Ventilation pressure-associated increases in intrathoracic pressure may decrease intrathoracic blood volume and cardiac output and activate the renin-angiotensin system, increase catecholamine release, increase AVP release, and increase renal sympathetic tone (8, 31). Thus respiratory therapy with high continuous positive airway pressures in human preterm infants (7, 31) and continuous positive-pressure ventilation in comparison with intermittent positive-pressure ventilation in adult models (8) are effective in decreasing GFR and sodium excretion. Therefore, we hypothesized the wide variations in ventilation pressure currently employed for mechanical ventilation of preterm newborns might affect the overall pattern of preterm cardiovascular, renal, and endocrine adaptations.

The similar Pa\textsubscript{CO\textsubscript{2}} values maintained throughout the study illustrate our success in ventilating the three study groups despite variations in ventilation style and MAP values achieved. However, despite the different ventilatory approaches, the rate and magnitude of adaptive changes among the groups in the current study were similar. These observations are consistent with the work of Kinsella et al. (16), who reported no hemodynamic differences between high-frequency oscillatory ventilation and conventional intermittent mandatory ventilation in preterm newborn baboons. They speculated that lung overdistension might allow for excessive transmission of airway pressure to the pleural space, impeding venous return and diminishing cardiac output. In the present study, ventilation pressure adjusted concomitant with changes in blood gas values and monitored tidal volume, and appropriate Pa\textsubscript{CO\textsubscript{2}} values indicated by measured pH values suggest that overventilation and lung overdistension did not occur. Therefore, the absence of lung overdistension among the groups appears to be consistent with the absence of differences in cardiovascular (except heart rate at 24 h), renal, and endocrine functions among the groups despite differences in MAP values. If assisted ventilation is performed in a manner that prevents lung overdistension, large differences in ventilation style or MAP values may not affect preterm cardiovascular, renal, and endocrine adaptations. The question of whether mechanical ventilation alone might affect overall adaptive responses in the preterm newborn cannot be addressed because preterm lambs <131 days gestation cannot survive in the absence of surfactant treatment and assisted ventilation (10).

In the present study, mean blood pressure and heart rate values at 2 h were consistent with the average values (blood pressure = 50 mmHg and heart rate = 150 beats/min) obtained from our previous studies in preterm lambs of similar gestation (2). Decreases in mean blood pressure in the R50 group were statistically significant due to relatively high 2- and 5-h values and low SE at 10 and 24 h. However, the patterns for mean blood pressure and heart rate were similar to cesarean-delivered term newborn lambs, with no changes in mean blood pressure and a decrease in heart rate during the first 24 h (19). The observed decreases in heart rate in the R50 and R15 groups are consistent with previous observations that newborn lamb plasma catecholamine levels are elevated at birth and decrease over time (19, 21). In contrast, heart rate in the HFOV group did not change and was significantly higher at 24 h than in the other two groups. Differences in HFOV-related baroreflex effects do not appear accountable given that recent studies in premature baboons (16), human infants (1), and premature lambs (20) demonstrated no differences in cardiovascular function between high-frequency oscillatory ventilation and conven-

![Fig. 6. Mean ± SE plasma renin activity (PRA; A), ANG II (B), and aldosterone (C) values. n = 5 for each group. †Differs from the 2-, 5-, and 24-h R15 values (P ≤ 0.05); ††differs from 2-h R15 value (P ≤ 0.05); *changes over time (P ≤ 0.05).](http://ajpregu.physiology.org/)
ional intermittent mandatory ventilation. Alternatively, although fluid administration and urine outputs were similar in all three groups, larger insensible fluid losses in the HFOV group could have contributed to a mild decrease in intravascular volume. The significantly higher hematocrit values and the more than threefold higher plasma AVP level levels observed in the HFOV group would be consistent with this possibility. Regardless of the exact mechanism(s) accounting for the difference in heart rate between the HFOV and the conventionally ventilated groups at 24 h, the similar cardiac outputs in all three groups indicate overall cardiovascular function was maintained.

The pattern of renal adaptation in term newborn lambs during the first 24 h after birth includes decreases in urine flow and increases in urine concentration, GFR, and sodium reabsorptive capacity (19). In the present ventilated preterm newborn lamb study, initial postnatal adaptive responses also included an increase in urine osmolality and a decrease in urine flow, consistent with the elevated plasma AVP levels measured (Fig. 5). However, there were no changes in GFR over time in any group. Additionally, GFR values in the present and previous (2) studies average only 60–70% of GFR values (1 ml·kg⁻¹·min⁻¹) measured in chronically catheterized fetal lambs of similar gestation. This observation is consistent with a number of studies describing mechanical ventilation-associated impairment of renal function (7, 8, 17).

One possible mechanism explaining the absence of changes in GFR in the preterm newborn may relate to the regulation of intrarenal blood flow distribution. Although nephrogenesis is essentially complete in sheep after 130 days gestation, fetal intrarenal blood flow distribution primarily reflects medullary and inner cortical regions, with minimal outer cortical flow (3). Thus the postnatal increase in GFR observed in term newborn lambs appears to reflect increases in outer cortical blood flow and recruitment of outer cortical nephrons (19) rather than changes in renal perfusion pressure or total renal blood flow (3). The regulation of changes in renal vascular resistance after birth in the term newborn is a complex process potentially influenced by a variety of factors, including renal sympathetic nerve activity; circulating factors, including catecholamines, AVP, and ANG II; and intrarenal regulatory factors, including prostaglandins. For example, ovine renal sympathetic nerve activity markedly increases at birth in the term newborn (26). However, whereas renal denervation is associated with a pronounced diuresis and natriuresis in the immediate postnatal period, renal denervation has no effect on postnatal changes in GFR (28). In contrast, renal sympathetic activity does not increase postnatally in the preterm newborn (25). Thus the absence of appropriate maturational changes in one or more of the systems regulating renal perfusion may serve to limit changes in intrarenal blood flow distribution and glomerular adaptation in the preterm newborn.

Despite the absence of changes in GFR, there was a marked decreasing trend in $\text{FE}_\text{Na}$ after delivery such that, by 16 h, $\text{FE}_\text{Na}$ values in all groups were similar to values in term newborn lambs at 24 h (19). Thus tubular sodium reabsorption increased. An absence of changes in outer cortical blood flow may have limited both changes in GFR and maintained juxtamedullary nephron perfusion where sodium reabsorption is more efficient (8). However, additional changes in tubular function, including increases in $\text{Na}^+$/H⁺ exchanger expression and function (24) and/or $\text{Na}^+$/K⁺-ATPase activity (19, 29), may have contributed to the increase in sodium reabsorption and would be consistent with the postnatal change in tubular function noted after delivery of term newborns (19, 27).

PRA levels were similar to previously reported levels in term newborn lambs (19). In contrast, plasma ANG II concentrations were four- to fivefold higher and plasma aldosterone concentrations were two- to threefold above reported term newborn levels (19). These results are consistent with an attenuated response to aldosterone in premature kidneys (15) and a reduced aldosterone response to ANG II (3, 23). In addition to decreases in renal perfusion pressure, renal sympathetic nerve activity also is an important determinant of renin production (18). Available data indicate that mechanical ventilation is associated with an increase in sympathetic nervous system activity, and renal sympathetic nervous system activity increases postnatally in term newborn lambs (26). However, because renal sympathetic nervous system activity has recently been shown to decrease in preterm newborn lambs (25), the basis for the overall increasing trend in PRA during the study and the significant elevations in PRA in the R15 group at 10 and 16 h are not clear. Whatever the mechanism(s) involved, premature delivery and ventilation resulted in sustained activation of the overall renin-angiotensin-aldosterone axis (Fig. 6), and this prolonged activation was independent of the ventilation styles employed.

In summary, we used surfactant-treated preterm lambs and mechanical ventilation to assess long-term (24 h) preterm newborn cardiovascular, renal, and endocrine postnatal adaptations. This study demonstrates that in ventilated preterm lambs 1) differing ventilation styles over the pressure ranges employed in the current study do not affect the time course for cardiovascular, renal, and endocrine adaptations, 2) glomerular adaptation does not occur during the first 24 h, and 3) marked increases in renal tubular sodium reabsorption can be demonstrated by 16 h after birth and occur in the absence of changes in renal plasma flow or GFR. Because the premature lambs studied had essentially normal lungs without underlying disease, a situation very different from the clinical setting, where much higher ventilatory pressures might be employed, we speculate the effects of different ventilation styles on postnatal adaptation may not occur without lung overdistension. Overall, the discordant adaptive changes in glomerular and tubular functions observed indicate that even in “healthy” preterm newborns renal vascular/glomerular adaptive potential is severely limited.
Kidney function during fetal life is characterized by highly exaggerated rates of diuresis and natriuresis relative to term newborns or adults in a variety of species. Although a high urine flow rate is essential to the maintenance of amniotic fluid volume in utero, water and sodium conservation are critical to postnatal adaptation in the extraterine environment. In the term newborn, GFR increases markedly, in keeping with the newborn’s need to manage waste disposal in the absence of the placenta, with concurrent increases in sodium reabsorptive capacity. In contrast, renal adaptive potential of the preterm newborn kidney is limited, resulting in often severe disruption of preterm newborn fluid and electrolyte regulation. Because ventilatory management styles vary markedly, the objective of this study was to determine if differing ventilation approaches affect overall renal adaptive responses. The results suggest that variations in ventilation approach over the range of conditions studied are not a determining factor in renal adaptation. However, this study clearly outlines the delay in renal adaptation/maturation characteristic of the premature kidney. More importantly, although delayed, the demonstrated increase in sodium reabsorptive capacity in the absence of changes in GFR indicates that improvements in preterm newborn sodium management can occur in the absence of changes in renal perfusion and GFR. Thus future studies can begin to distinguish the factors and/or mechanisms that specifically regulate postnatal adaptation of glomerular function versus tubular absorptive capacity.

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Address for reprint requests: M. G. Ervin, PO Box 60, Middle Tennessee State Univ., Murfreesboro, TN 37132.

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