Systemic and Myocardial Effects of ATP and Adenosine during Hypoxic Pulmonary Hypertension in Lambs

G. GANESH KONDURI

Departments of Pediatrics, Children's Hospital of Michigan, Hutzel Hospital, and Wayne State University School of Medicine, Detroit, Michigan 48201

ABSTRACT

The systemic and myocardial effects of adenosine and ATP were investigated in 12 newborn lambs, instrumented at 5–7 d of age with catheters in the aorta, pulmonary artery, coronary sinus, and right and left atria and flow transducers around the main pulmonary artery and left circumflex coronary artery. Studies were done 3–7 d after recovery from surgery. Pulmonary hypertension was induced by exposure to alveolar hypoxia (10% O₂, 5% CO₂, and 85% N₂), which was maintained throughout the experiment. Adenosine, ATP, or an equal volume of saline (control) was infused into the right atrial line in doses of 0.04 to 2.5 μmol/kg/min during hypoxia. Alveolar hypoxia caused significant increases in pulmonary artery pressure, pulmonary vascular resistance, left circumflex flow, left ventricular O₂ consumption, and systemic and myocardial O₂ extraction and a decrease in systemic O₂ transport. ATP and adenosine caused selective decreases in pulmonary artery pressure and pulmonary vascular resistance at doses of 0.04 to 0.30 μmol/kg/min and decreases in both pulmonary and systemic pressures and resistances at 0.60 to 2.5 μmol/kg/min. ATP and adenosine caused increases in systemic O₂ transport, left circumflex flow, left ventricular O₂ transport, and left ventricular O₂ consumption and decreases in systemic O₂ extraction and left ventricular O₂ extraction at 0.3 to 2.5 μmol/kg/min. Systemic O₂ con-

sumption did not change during the study. Arterial and coronary sinus blood lactate levels increased during hypoxia and decreased from hypoxia at 2.5-μmol/kg/min infusion rates of adenosine and ATP. Adenosine and ATP cause selective pulmonary vasodilation in lambs with pulmonary hypertension at doses of 0.04 to 0.30 μmol/kg/min and improvement in systemic and myocardial O₂ delivery at larger doses. Adenosine and ATP may have a beneficial role in the management of pulmonary hypertension in children. (*Pediatr Res* 36: 41–48, 1994)

Abbreviations

PVR, pulmonary vascular resistance
SVR, systemic vascular resistance
PAP, pulmonary artery pressure
SOT, systemic O₂ transport
VO₂, systemic O₂ consumption
V_E, systemic O₂ extraction
MOT, left ventricular O₂ transport
MVO₂, left ventricular O₂ consumption
MV_E, left ventricular O₂ extraction
Pao₂, arterial Po₂
Paco₂, arterial Pco₂
FIO₂, fraction of inspired oxygen

Persistent pulmonary hypertension of the newborn is a condition associated with high morbidity and mortality. The affected infants have an increased PVR and hypoxemia secondary to right-to-left shunting of blood across fetal channels (1). Vasodilator drugs that are used in the management of this condition, such as tolazoline, seem to dilate both pulmonary and systemic vessels and cause

hypotension and tachycardia as side effects (2). The increase in heart rate and systemic vasodilation results in increases in VO₂ and MVO₂ during hypoxia (3). Newborn infants have a high resting cardiac output and O₂ consumption (4) and are limited in their ability to augment SOT during hypoxia (5). Therefore, an increase in O₂ consumption during vasodilator therapy may compromise tissue oxygenation in hypoxic infants.

Adenosine and ATP are purine nucleotides that dilate systemic and pulmonary vessels by stimulation of P₁ and P₂ purinergic receptors (6). They are important mediators of O₂-induced pulmonary vasodilation in fetal lambs (7). The nucleotides have an extremely short half-life and are cleared during a single pass through the lung (8) when

Received August 12, 1993; accepted February 1, 1994.

Correspondence and reprint requests: G. Ganesh Konduri, M.D., Department of Pediatrics, Hutzel Hospital, 4707 St. Antoine Blvd., Detroit, MI 48201.

Supported by grants from the American Heart Association of Michigan and the Children's Hospital of Michigan Research Endowment Fund.

¹Presented in part at the Society for Pediatric Research meeting, New Orleans, LA, May 1991.

they are infused in low doses into the right atrium. ATP infusion causes selective pulmonary vasodilation in doses $\leq 0.15 \mu\text{mol/kg/min}$ in newborn lambs with pulmonary hypertension induced by alveolar hypoxia (9) or infusion of thromboxane mimetic U46619 (10) and in newborn piglets (11) during hypoxia. ATP has been shown to cause selective pulmonary vasodilation at doses $\leq 0.15 \mu\text{mol/kg/min}$ in children with congenital heart disease and preoperative and postoperative pulmonary hypertension (12). Adenosine causes selective pulmonary vasodilation in newborn lambs during hypoxia at doses $\leq 0.15 \mu\text{mol/kg/min}$ (13). Preliminary data (14) suggest that adenosine infusion ($0.1\text{--}0.2 \mu\text{mol/kg/min}$) improves oxygenation without causing hypotension and tachycardia in newborn infants with persistent pulmonary hypertension. We proposed the hypothesis that ATP and adenosine do not alter the VO_2 and myocardial O_2 consumption in doses that cause selective pulmonary vasodilation. The objectives of the present experiments were to determine the effects of adenosine and ATP on 1) SOT and VO_2 , 2) MOT and MVO_2 , and 3) arterial and coronary sinus lactate concentrations as an index of systemic and myocardial oxygenation. We chose the newborn lamb as a model because its pulmonary circulation has been studied extensively in the past, and its response to hypoxia has been well documented (15). We used chronically instrumented animals for our studies because this allows us to study pulmonary and systemic circulations without the confounding effects of surgery and anesthesia.

METHODS

Twelve lambs were studied between the ages of 8 and 12 d. Each lamb was studied three times during hypoxia: once each with adenosine, ATP, and normal saline (control). Each of the three experiments was done on a separate day, and the order of experiments was randomized for each animal. The study was approved by the Animal Investigation Committee of Wayne State University (Detroit, MI).

Lambs were brought into our laboratory at 1 d of age and housed in separate Plexiglas cages. They were trained to feed *ad libitum* from an artificial feeding system that contained goat's milk (Irish Hills Farm natural goat's milk, Mason, MI). Lambs adjusted well to the feeding system and gained weight at the rate of $50\text{--}75 \text{ g/kg/d}$ before surgery.

Surgical procedure. Each lamb underwent one sterile surgical procedure at 5 to 7 d of age. Lambs were given preanesthetic medication consisting of atropine sulfate (0.01 mg/kg s.c.) and ketamine HCl (10 mg/kg intramuscularly). The animal's trachea was intubated with a cuffed endotracheal tube, and anesthesia was maintained by ventilating the lungs with $1\text{--}2\%$ isoflurane and O_2 . Rectal temperature and ECG were monitored continuously, and arterial blood gases were measured periodically during surgery. Core temperature was maintained at

or close to 39°C with a heating pad. An incision was made in the left groin, and catheters were inserted into the descending aorta and inferior vena cava via the femoral artery and femoral vein. A left lateral thoracotomy was done, and catheters were inserted by direct puncture into the main pulmonary artery and right and left atria. A catheter was also inserted into the coronary sinus via the hemiazygos vein. The coronary sinus in the lamb receives the venous return from left ventricular myocardium exclusively (16). Ultrasonic flow transducers were implanted around the main pulmonary artery (size 12 S, Transonic Systems Inc., Ithaca, NY) and around the left circumflex coronary artery (size 2 R, Transonic Systems Inc.). The ductus arteriosus was identified and ligated, and the catheters and flow transducer cables were tunneled to the lamb's back. The animal was allowed to recover from surgery for at least 3 d before experiments were done. Cardiovascular function and O_2 consumption were shown to return to normal by 3 d after thoracotomy in lambs (17). Antibiotics (procaine penicillin, $50\,000 \text{ U/kg}$, and gentamicin, 5 mg/kg , intramuscularly) were administered on the day of surgery and on each postoperative day until the studies were completed.

Experimental protocol. Each lamb was secured in a sling during the study. The vascular catheters were connected to strain gauge manometers (P23XL, Spectramed Inc., Critical Care Division, Oxnard, CA), and the flow transducer cables were connected to a dual-channel ultrasonic transit time blood flow meter (Transonic Systems Inc.). The mean aortic, pulmonary arterial, left and right atrial pressures, and mean pulmonary and left circumflex coronary blood flow were recorded on a Grass model 7D polygraph (Grass Instruments, Quincy, MA). From these data, we calculated cardiac index as cardiac output $\div$ body weight, PVR index as mean (PAP – left atrial pressure) $\div$ cardiac index in L/kg, and SVR index as mean (systemic arterial pressure – right atrial pressure) $\div$ cardiac index in L/kg. Arterial, mixed venous, and coronary sinus blood samples were obtained to measure blood gas tensions and blood pH on a blood gas analyzer (ABL 30, Radiometer, Copenhagen, Denmark). Blood Hb concentration (g/L) and the percentage of saturation of Hb with O_2 were measured with a hemoximeter (OSM 2, Radiometer). Blood O_2 content ($\text{mL O}_2/\text{L}$) was calculated (4) as $(1.36 \times \text{Hb concentration} \times \% \text{Hb saturation}) + (0.003 \times \text{Pao}_2)$. SOT was calculated as arterial O_2 content $\times$ cardiac index, VO_2 as cardiac index $\times$ (arterial-mixed venous) O_2 contents, V_E as $(\text{VO}_2 \div \text{SOT}) \times 100$, MOT as left circumflex coronary flow $\times$ arterial O_2 content, MVO_2 as left circumflex flow $\times$ (arterial-coronary sinus) O_2 contents, and MV_E as $(\text{MVO}_2 \div \text{MOT}) \times 100$.

Ambient temperature of the study room was maintained at $26\text{--}27^\circ\text{C}$ during the experiment. Baseline hemodynamic variables were measured, and blood samples were drawn during normoxia when the animals were awake and resting quietly in the sling. The animals were allowed to rest in the sling for at least 15 min before

baseline variables were measured. Alveolar hypoxia was then induced by placing a loosely fitting plastic bag over the animal's head and allowing it to breathe a gas mixture of 10% O₂, 5% CO₂, and 85% N₂ at a flow rate of 5–8 L/min. Lambs frequently hyperventilate during hypoxia and lower their P_aCO₂, which tends to attenuate the pulmonary vascular response to hypoxia (5). We added 5% CO₂ to the gas mixture to maintain a stable P_aCO₂ at or close to 5.3 kPa (40 mm Hg). The flow rate of the gas mixture was high enough to prevent rebreathing of expired gas. Variables were measured after the animals had stable hypoxemia for 10 min. Adenosine, ATP, or normal saline was infused into the right atrial line starting at 0.04 μ mol/kg/min. Variables were measured after 10 min. The infusion rate was then doubled every 10 min to give the animal 0.08, 0.15, 0.30, 0.60, 1.20, and 2.50 μ mol/kg/min of adenosine or ATP. Each infusion rate was maintained for 10 min, and hemodynamic variables were measured at the end of the 10-min period. Alveolar hypoxia was continued during the infusion of adenosine, ATP, or saline until the end of experiment. The animal was returned to room air at the end of each study and returned to its cage. Subsequent experiments were done after a 24-h recovery. Arterial, mixed venous, and coronary sinus blood samples were drawn at baseline, during hypoxia alone, and at each infusion rate of adenosine, ATP, or saline to measure blood gas tensions, Hb concentration, and Hb O₂ saturation. Arterial and coronary sinus blood samples were also obtained for blood lactate levels at baseline, during hypoxia alone, and at infusion rates of 0.04, 0.15, 0.60, and 2.50 μ mol/kg/min. The infusion rates of ATP and adenosine were chosen to obtain minimal and maximal changes in PVR and cardiac index in newborn lambs as determined from previous studies (9, 13). The half-lives of ATP and adenosine in blood are <10 s (9, 13), and 10 min is sufficient to achieve steady state levels of nucleotide in plasma. The volume of fluid infused during the experiment ranged from 0.04 to 2.5 mL/min, and the total volume was 50 mL or approximately 9 mL/kg over 1.5 h. The infusion of saline at this volume in control studies did not cause significant hemodynamic changes. Animals were awake and did not show agitation during the study. Most of the animals had a shivering response during the last 30 min of hypoxia, even though ambient temperature was kept constant.

Drug preparation. Adenosine and disodium salt of ATP (Sigma Chemical Co., St. Louis, MO) were obtained in powder form, and the solutions for i.v. administration were prepared each day by mixing them with sterile normal saline to give a final concentration of adenosine or ATP of 4 μ mol/mL.

Assay for lactate. Blood samples were collected into tubes with 0.5 mL of 12% trichloroacetic acid kept at 0–4°C. Samples were immediately centrifuged, and the supernatant was separated and stored at –20°C. Samples were assayed for lactate by an enzymatic method based on conversion of lactate to pyruvate by lactate dehydro-

genase. The reduction of NAD from this reaction causes an increase in absorbance at 340 nm, which was measured by a spectrophotometer (Beckman Instruments, Inc., Fullerton, CA). Assays for all samples were run in triplicate. The concentration of lactate in each sample was measured using a four-point standard curve and expressed as mmol/L. The intraassay and interassay coefficients of variation for this method were 3–5% and 5–7%, respectively.

Statistical analysis. Data are expressed as mean $\pm$ SD. Data obtained during normoxia and during hypoxia alone from different days on each lamb were compared by single-factor analysis of variance for repeated measures (18) to determine whether baseline variables and response to hypoxia were similar on different days. Data obtained during control, ATP, and adenosine infusions were compared by two-way analysis of variance for repeated measures; the two factors affecting the outcome were hypoxia and dose of ATP, adenosine, or saline. When significant differences ($p < 0.05$) were found, a Duncan's multiple range test (18) was done to determine which means were different.

RESULTS

Baseline variables measured on different days and their response to hypoxia were comparable and showed no significant differences. Alveolar hypoxia resulted in significant increases in heart rate, cardiac index, PAP, PVR, left circumflex flow, and MVO₂ and a significant decrease in SOT (Tables 1 and 2). VO₂ did not change and V_E and MV_E increased during hypoxia. There were no further changes in heart rate, cardiac output, SOT, PAP, and PVR when the animals received normal saline infusion. MOT and MVO₂ decreased to baseline values during infusion of saline with ongoing hypoxia (Table 2). Administration of ATP and adenosine caused selective decreases in PAP and PVR at doses of 0.04 to 0.30 μ mol/kg/min (Table 1). Infusion of ATP and adenosine at doses of 0.60 to 2.5 μ mol/kg/min caused decreases in PAP, PVR, aortic pressure, and SVR and an increase in cardiac index (Table 1). SOT, left circumflex flow, MOT, and MVO₂ also increased at infusion rates of adenosine and ATP of 0.3 to 2.5 μ mol/kg/min (Table 2). VO₂ did not change and V_E and MV_E decreased at infusion rates of ATP and adenosine of 0.30 to 2.50 μ mol/kg/min during hypoxia.

There were significant decreases in pH and P_aO₂ and an increase in P_aCO₂ during alveolar hypoxia in control, ATP, and adenosine experiments (Table 3). The increase in P_aCO₂ is probably caused by addition of 5% CO₂ to the inspired gas mixture. The plastic bag was loosely tied around the neck of the lambs, and the flow rate was kept high enough to prevent rebreathing of expired gas. Blood pH and P_aCO₂ did not change further during the experiments with ATP or adenosine compared with control studies. P_aO₂ increased significantly from hypoxia alone at infusion rates of ATP and adenosine of 2.5 μ mol/kg/

Table 1. Systemic and pulmonary vascular effects of ATP, adenosine (Ade) and saline (Control) during alveolar hypoxia (Hyp)*

Variable	Study	Baseline	Hyp	Infusion rates ($\mu\text{mol/kg/min}$)					
				0.04	0.08	0.15	0.30	0.60	2.50
Heart rate (bpm)	Control	188 $\pm$ 22	251 $\pm$ 38†	242 $\pm$ 34†	252 $\pm$ 23†	248 $\pm$ 43†	252 $\pm$ 41†	255 $\pm$ 35†	258 $\pm$ 24†
	ATP	188 $\pm$ 25	229 $\pm$ 42†	227 $\pm$ 42†	225 $\pm$ 45†	240 $\pm$ 46†	245 $\pm$ 42†	252 $\pm$ 39†	233 $\pm$ 44†
	Ade	184 $\pm$ 30	232 $\pm$ 32†	247 $\pm$ 39†	246 $\pm$ 43†	252 $\pm$ 36†	247 $\pm$ 30†	249 $\pm$ 37†	228 $\pm$ 42†
Cardiac index (mL/kg/min)	Control	170 $\pm$ 32	200 $\pm$ 26†	209 $\pm$ 36†	198 $\pm$ 32†	195 $\pm$ 41†	201 $\pm$ 31†	200 $\pm$ 48†	196 $\pm$ 41†
	ATP	160 $\pm$ 19	190 $\pm$ 21†	190 $\pm$ 22†	190 $\pm$ 29†	200 $\pm$ 25†	210 $\pm$ 31†	235 $\pm$ 32†‡	230 $\pm$ 31†‡
	Ade	166 $\pm$ 28	194 $\pm$ 21†	210 $\pm$ 41†	206 $\pm$ 36†	208 $\pm$ 31†	218 $\pm$ 38†	232 $\pm$ 41†‡	235 $\pm$ 32†‡
SAP (mm Hg)§	Control	63 $\pm$ 9	69 $\pm$ 10	65 $\pm$ 11	66 $\pm$ 10	69 $\pm$ 9	67 $\pm$ 9	68 $\pm$ 3	64 $\pm$ 7
	ATP	66 $\pm$ 6	65 $\pm$ 10	62 $\pm$ 9	61 $\pm$ 10	59 $\pm$ 10	60 $\pm$ 11	56 $\pm$ 9†‡	54 $\pm$ 9†‡
	Ade	60 $\pm$ 5	60 $\pm$ 8	59 $\pm$ 10	61 $\pm$ 9	56 $\pm$ 9	54 $\pm$ 9	50 $\pm$ 9†‡	41 $\pm$ 8†‡
PAP (mm Hg)§	Control	11 $\pm$ 4	20 $\pm$ 3†	19 $\pm$ 3†	19 $\pm$ 4†	20 $\pm$ 4†	22 $\pm$ 4†	21 $\pm$ 5†	24 $\pm$ 6†
	ATP	11 $\pm$ 2	22 $\pm$ 4†	16 $\pm$ 3†‡	17 $\pm$ 3†‡	15 $\pm$ 4†	16 $\pm$ 2†‡	16 $\pm$ 3†‡	16 $\pm$ 3†‡
	Ade	10 $\pm$ 3	20 $\pm$ 3†	15 $\pm$ 4†	16 $\pm$ 2†‡	16 $\pm$ 3†‡	16 $\pm$ 2†‡	16 $\pm$ 4†‡	15 $\pm$ 3†‡
LAP (mm Hg)§	Control	3 $\pm$ 2	1 $\pm$ 1	2 $\pm$ 1	2 $\pm$ 1	2 $\pm$ 2	2 $\pm$ 1	2 $\pm$ 1	1 $\pm$ 1
	ATP	3 $\pm$ 1	1 $\pm$ 1	2 $\pm$ 1	3 $\pm$ 1	2 $\pm$ 1	2 $\pm$ 1	4 $\pm$ 2	3 $\pm$ 1
	Ade	3 $\pm$ 1	2 $\pm$ 1	2 $\pm$ 1	1 $\pm$ 1	2 $\pm$ 1	2 $\pm$ 2	3 $\pm$ 1	3 $\pm$ 1
SVR index (mm Hg·L ⁻¹ ·kg ⁻¹ ·min ⁻¹)	Control	480 $\pm$ 78	420 $\pm$ 70	480 $\pm$ 82	500 $\pm$ 76	497 $\pm$ 82	520 $\pm$ 96	440 $\pm$ 72	420 $\pm$ 90
	ATP	449 $\pm$ 82	400 $\pm$ 72	403 $\pm$ 76	422 $\pm$ 78	370 $\pm$ 82	352 $\pm$ 72	310 $\pm$ 68†	301 $\pm$ 70†
	Ade	433 $\pm$ 76	380 $\pm$ 81	350 $\pm$ 62	348 $\pm$ 71	321 $\pm$ 80	300 $\pm$ 70	260 $\pm$ 62†	214 $\pm$ 76†
PVR index (mm Hg·L ⁻¹ ·kg ⁻¹ ·min ⁻¹)	Control	88 $\pm$ 20	126 $\pm$ 19†	136 $\pm$ 26†	130 $\pm$ 21†	127 $\pm$ 28†	150 $\pm$ 32†	139 $\pm$ 36†	139 $\pm$ 40†
	ATP	78 $\pm$ 23	135 $\pm$ 21†	102 $\pm$ 18†	100 $\pm$ 17†	102 $\pm$ 19†	91 $\pm$ 18†	82 $\pm$ 19†	80 $\pm$ 16†
	Ade	77 $\pm$ 19	122 $\pm$ 20†	100 $\pm$ 18†	102 $\pm$ 19†	91 $\pm$ 19†	90 $\pm$ 18†	82 $\pm$ 20†	76 $\pm$ 18†

* Mean $\pm$ SD for $n = 12$. SAP, systemic arterial pressure; LAP, left atrial pressure. Hyp data were obtained at FIO₂ of 0.10 and fraction of inspired CO₂ of 0.05.

† $p < 0.05$ from baseline.

‡ $p < 0.05$ from Hyp alone.

§ Conversion factor for SI units: 1 mm Hg = 0.133 kPa.

Table 2. Effects of ATP, adenosine (Ade), and saline (Control) on oxygen transport during alveolar hypoxia (Hyp)*

Variable	Study	Baseline	Hyp	Infusion rates ($\mu\text{mol/kg/min}$)					
				0.04	0.08	0.15	0.30	0.60	2.50
SOT (mL O ₂ /kg/min)	Control	18 $\pm$ 2	15 $\pm$ 2†	14 $\pm$ 2†	13 $\pm$ 3†	13 $\pm$ 3†	13 $\pm$ 2†	13 $\pm$ 3†	15 $\pm$ 2†
	ATP	17 $\pm$ 2	13 $\pm$ 3†	13 $\pm$ 3†	13 $\pm$ 3†	14 $\pm$ 4†	16 $\pm$ 3†	16 $\pm$ 2†	16 $\pm$ 2†
	Ade	18 $\pm$ 2	15 $\pm$ 2†	16 $\pm$ 2	14 $\pm$ 3†	14 $\pm$ 2†	18 $\pm$ 2†	18 $\pm$ 3†	18 $\pm$ 3†
VO ₂ (mL O ₂ /kg/min)	Control	8 $\pm$ 1	8 $\pm$ 2	6 $\pm$ 2	7 $\pm$ 2	7 $\pm$ 2	6 $\pm$ 2	7 $\pm$ 2	7 $\pm$ 2
	ATP	7 $\pm$ 1	7 $\pm$ 1	7 $\pm$ 1	6 $\pm$ 2	6 $\pm$ 2	7 $\pm$ 1	9 $\pm$ 2	9 $\pm$ 1
	Ade	7 $\pm$ 1	6 $\pm$ 2	6 $\pm$ 2	6 $\pm$ 1	7 $\pm$ 2	6 $\pm$ 1	6 $\pm$ 2	6 $\pm$ 2
V _E (%)	Control	32 $\pm$ 6	40 $\pm$ 6†	40 $\pm$ 9†	41 $\pm$ 7†	40 $\pm$ 7†	38 $\pm$ 9	41 $\pm$ 11†	41 $\pm$ 10†
	ATP	29 $\pm$ 3	43 $\pm$ 6†	44 $\pm$ 4†	40 $\pm$ 6†	37 $\pm$ 7	32 $\pm$ 6†	32 $\pm$ 7†	32 $\pm$ 6†
	Ade	32 $\pm$ 8	41 $\pm$ 8†	42 $\pm$ 8†	35 $\pm$ 8	34 $\pm$ 9	32 $\pm$ 10†	31 $\pm$ 10†	26 $\pm$ 10†
LCF (mL/min)	Control	14 $\pm$ 6	35 $\pm$ 8†	30 $\pm$ 9†	25 $\pm$ 6†	25 $\pm$ 6†	28 $\pm$ 11†	27 $\pm$ 9†	33 $\pm$ 10†
	ATP	16 $\pm$ 5	41 $\pm$ 12†	30 $\pm$ 12†	29 $\pm$ 11†	40 $\pm$ 13†	55 $\pm$ 16†‡	78 $\pm$ 17†‡§	79 $\pm$ 18†‡§
	Ade	17 $\pm$ 8	39 $\pm$ 10†	44 $\pm$ 17†	43 $\pm$ 16†	55 $\pm$ 16†	58 $\pm$ 15†‡	63 $\pm$ 18†‡	63 $\pm$ 16†‡
MOT (mL O ₂ /min)	Control	1.8 $\pm$ 0.5	3.1 $\pm$ 0.6†	2.0 $\pm$ 0.2	2.1 $\pm$ 0.3	1.9 $\pm$ 0.2	1.8 $\pm$ 0.4	1.8 $\pm$ 0.5	2.2 $\pm$ 0.6
	ATP	2.1 $\pm$ 0.6	2.9 $\pm$ 0.6†	2.4 $\pm$ 0.7	2.4 $\pm$ 0.6	3.2 $\pm$ 1†	4.6 $\pm$ 1.5†‡	5.9 $\pm$ 2†‡	6.6 $\pm$ 2†‡
	Ade	2.5 $\pm$ 0.9	4.1 $\pm$ 1.3†	3.9 $\pm$ 1†	3.5 $\pm$ 1.3†	4.9 $\pm$ 1.5†	6.3 $\pm$ 2†‡	7.2 $\pm$ 2†‡	7 $\pm$ 2.1†‡
MVO ₂ (mL O ₂ /min)	Control	0.7 $\pm$ 0.03	1.5 $\pm$ 0.2†	1 $\pm$ 0.1	1.1 $\pm$ 0.2	1 $\pm$ 0.1	1 $\pm$ 0.2	1 $\pm$ 0.1	1.2 $\pm$ 0.4
	ATP	0.8 $\pm$ 0.3	1.6 $\pm$ 0.3†	1.4 $\pm$ 0.2	1.3 $\pm$ 0.1	1.3 $\pm$ 0.3	1.4 $\pm$ 0.4†	2.1 $\pm$ 0.2†	2.8 $\pm$ 0.6†‡
	Ade	1.2 $\pm$ 0.1	1.8 $\pm$ 0.15†	1.3 $\pm$ 0.2	1.3 $\pm$ 0.1	1.6 $\pm$ 0.2	2.5 $\pm$ 0.6†	2.5 $\pm$ 0.8†	2.3 $\pm$ 0.4†
MV _E (%)	Control	33 $\pm$ 9	52 $\pm$ 10†	46 $\pm$ 11†	50 $\pm$ 10†	48 $\pm$ 7†	52 $\pm$ 11†	45 $\pm$ 10	55 $\pm$ 12†
	ATP	40 $\pm$ 6	54 $\pm$ 9†	42 $\pm$ 8	42 $\pm$ 9	38 $\pm$ 9†	28 $\pm$ 8†	26 $\pm$ 9†	32 $\pm$ 8†
	Ade	39 $\pm$ 9	52 $\pm$ 9†	54 $\pm$ 10†	45 $\pm$ 7	43 $\pm$ 9	40 $\pm$ 7†	35 $\pm$ 9†	36 $\pm$ 9†

* Mean $\pm$ SD for $n = 12$. LCF, left circumflex flow. Hyp data were obtained at FIO₂ of 0.10 and fraction of inspired CO₂ of 0.05.

† $p < 0.05$ from baseline.

‡ $p < 0.05$ from Hyp alone.

§ $p < 0.05$ from 0.30- $\mu\text{mol/kg/min}$ infusion rate of ATP.

Table 3. Arterial blood gas values, blood pH, lactates, and temperature during hypoxia (Hyp) and infusion of saline (Control), ATP, and adenosine (Ade)*

Variable	Study	Baseline	Hyp	Infusion rates ($\mu\text{mol/kg/min}$)					
				0.04	0.08	0.15	0.30	0.60	2.50
pHa	Control	7.45 ± 0.04	$7.39 \pm 0.04^\dagger$	$7.37 \pm 0.04^\dagger$	$7.39 \pm 0.04^\dagger$	$7.36 \pm 0.04^\dagger$	$7.35 \pm 0.03^\dagger$	$7.36 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$
	ATP	7.44 ± 0.04	$7.38 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.37 \pm 0.04^\dagger$	$7.35 \pm 0.04^\dagger$	$7.35 \pm 0.06^\dagger$
	Ade	7.43 ± 0.04	$7.39 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.35 \pm 0.05^\dagger$	$7.36 \pm 0.05^\dagger$	$7.36 \pm 0.04^\dagger$	$7.36 \pm 0.05^\dagger$
Paco_2 (kPa)‡	Control	4.8 ± 0.4	$5.6 \pm 0.5^\dagger$	$5.9 \pm 0.7^\dagger$	$6.0 \pm 0.8^\dagger$	$5.9 \pm 0.4^\dagger$	$5.9 \pm 0.4^\dagger$	$5.6 \pm 0.4^\dagger$	$5.9 \pm 0.7^\dagger$
	ATP	4.7 ± 0.3	$5.9 \pm 0.5^\dagger$	$5.9 \pm 0.7^\dagger$	$5.9 \pm 0.7^\dagger$	$5.6 \pm 0.5^\dagger$	5.3 ± 0.4	$5.6 \pm 0.7^\dagger$	$5.6 \pm 0.4^\dagger$
	Ade	4.8 ± 0.4	$5.6 \pm 0.5^\dagger$	$5.6 \pm 0.4^\dagger$	$5.6 \pm 0.4^\dagger$	$5.6 \pm 0.8^\dagger$	$5.5 \pm 0.4^\dagger$	5.3 ± 0.4	$5.6 \pm 0.4^\dagger$
Pao_2 (kPa)‡	Control	10.9 ± 0.5	$5.1 \pm 0.8^\dagger$	$4.8 \pm 0.8^\dagger$	$4.5 \pm 0.5^\dagger$	$4.4 \pm 0.8^\dagger$	$4.8 \pm 0.8^\dagger$	$4.7 \pm 0.9^\dagger$	$4.8 \pm 0.8^\dagger$
	ATP	10.5 ± 1	$4.7 \pm 0.7^\dagger$	$5.2 \pm 0.7^\dagger$	$4.4 \pm 0.4^\dagger$	$4.7 \pm 0.7^\dagger$	$4.9 \pm 0.7^\dagger$	$5.3 \pm 0.8^\dagger$	$5.6 \pm 0.7^\dagger$ §
	Ade	10.1 ± 0.8	$4.5 \pm 0.5^\dagger$	$4.8 \pm 0.5^\dagger$	$4.5 \pm 0.8^\dagger$	$4.8 \pm 0.9^\dagger$	$5.3 \pm 0.8^\dagger$	$5.3 \pm 0.9^\dagger$	$5.7 \pm 0.7^\dagger$ §
Arterial Hb concentration (g/L)	Control	94 ± 6	98 ± 5	98 ± 6	97 ± 10	97 ± 10	96 ± 5	98 ± 10	90 ± 10
	ATP	90 ± 7	$98 \pm 6^\dagger$	$96 \pm 5^\dagger$	95 ± 6	90 ± 4	91 ± 7	94 ± 7	88 ± 10
	Ade	98 ± 6	$105 \pm 5^\dagger$	101 ± 7	99 ± 6	98 ± 7	95 ± 10	97 ± 7	88 ± 8 §
Arterial lactate (mmol/L)	Control	0.13 ± 0.04	$0.30 \pm 0.06^\dagger$	$0.30 \pm 0.1^\dagger$		$0.52 \pm 0.2^\dagger$		$0.47 \pm 0.2^\dagger$	$0.65 \pm 0.3^\dagger$
	ATP	0.21 ± 0.06	$0.42 \pm 0.1^\dagger$	$0.42 \pm 0.2^\dagger$		$0.71 \pm 0.3^\dagger$		$0.52 \pm 0.3^\dagger$	0.36 ± 0.2
	Ade	0.24 ± 0.08	$0.50 \pm 0.1^\dagger$	$0.56 \pm 0.2^\dagger$		$0.61 \pm 0.2^\dagger$		$0.50 \pm 0.2^\dagger$	0.30 ± 0.1 §
Coronary sinus lactate (mmol/L)	Control	0.04 ± 0.02	0.10 ± 0.04	$0.16 \pm 0.05^\dagger$		$0.25 \pm 0.1^\dagger$		$0.30 \pm 0.15^\dagger$	$0.32 \pm 0.12^\dagger$
	ATP	0.06 ± 0.02	0.09 ± 0.03	$0.12 \pm 0.04^\dagger$		$0.21 \pm 0.1^\dagger$		$0.18 \pm 0.06^\dagger$	0.10 ± 0.05
	Ade	0.08 ± 0.03	0.12 ± 0.04	$0.16 \pm 0.05^\dagger$		$0.24 \pm 0.15^\dagger$		$0.16 \pm 0.1^\dagger$	0.12 ± 0.1
Core temperature ($^\circ\text{C}$)	Control	39.4 ± 0.2	39.2 ± 0.3	39.0 ± 0.2		39.0 ± 0.3		38.6 ± 0.2	$38.5 \pm 0.3^\dagger$
	ATP	39.4 ± 0.3	39.2 ± 0.3	39.0 ± 0.2		38.8 ± 0.2		$38.5 \pm 0.2^\dagger$	$38.4 \pm 0.2^\dagger$
	Ade	39.6 ± 0.2	39.3 ± 0.2	39.0 ± 0.3		38.9 ± 0.2		38.6 ± 0.2	$38.5 \pm 0.3^\dagger$

* Data are mean $\pm$ SD for $n = 12$. pHa, arterial pH.† $p < 0.05$ from baseline.

‡ Conversion factor: 1 kPa = 7.5 mm Hg.

§ $p < 0.05$ from Hyp.|| $p < 0.05$ from 0.15- $\mu\text{mol/kg/min}$ infusion rate.

min. The increase in Pao_2 was caused by an increase in cardiac output and mixed venous Po_2 , inasmuch as FIO_2 was not changed. The mixed venous Po_2 increased from 2.8 ± 0.5 kPa (21 ± 4 mm Hg) at baseline to 3.6 ± 0.4 kPa (27 ± 3 mm Hg) at an infusion rate of ATP or adenosine of 2.5 $\mu\text{mol/kg/min}$. Blood Hb concentration increased during alveolar hypoxia before the infusion of ATP and adenosine in study animals. The increase averaged 7 g/L and was similar to the increase in Hb concentration during hypoxia reported by Moss *et al.* (5). Arterial blood lactate levels (Table 3) increased during hypoxia and remained elevated during infusion of saline. Arterial lactate levels at 2.5- $\mu\text{mol/kg/min}$ infusion rates of ATP and adenosine were lower than at 0.15 $\mu\text{mol/kg/min}$. Lactate levels in coronary sinus were lower than in aorta during both normoxia and hypoxia, indicating a net lactate uptake by the myocardium. Coronary sinus lactate levels increased during hypoxia and remained high during infusion of saline. However, lactate levels in coronary sinus during infusion of adenosine or ATP at 2.50 $\mu\text{mol/kg/min}$ were lower than at the 0.15- $\mu\text{mol/kg/min}$ infusion rate, consistent with a decrease in MV_E at these doses of ATP and adenosine. The core temperature decreased in all animals with prolonged hypoxia (Table 3) and occurred at the 2.50- $\mu\text{mol/kg/min}$ infusion rate of saline, ATP, and adenosine. Animals have shown a shivering response at the time of decrease in core temperature. The increase in

O_2 delivery during infusion of ATP and adenosine at 0.3 to 2.5 $\mu\text{mol/kg/min}$ did not restore core temperature to baseline.

DISCUSSION

Our study provides the evidence that ATP and adenosine, the purine nucleotides, cause selective pulmonary vasodilation in newborn lambs with hypoxic pulmonary hypertension at doses ≤ 0.30 $\mu\text{mol/kg/min}$. The results confirm the hypothesis that ATP and adenosine do not alter the VO_2 and the myocardial O_2 consumption at doses that cause selective pulmonary vasodilation. Adenosine and ATP had a favorable effect on tissue perfusion as evidenced by decreases in V_E and MV_E . These data indicate a potential role for continuous infusions of adenosine and ATP in the management of newborn infants and children with pulmonary hypertension.

The decrease in PVR caused by adenosine and ATP in hypoxic lambs was associated with a significant decrease in PAP and was not accompanied by a change in left atrial pressure or pulmonary blood flow at doses ≤ 0.30 $\mu\text{mol/kg/min}$. The PAP remained significantly lower at doses ≥ 0.60 $\mu\text{mol/kg/min}$, even though the pulmonary blood flow increased. Therefore, the decrease in PVR caused by ATP and adenosine is probably caused by true pulmonary vasodilation (19) rather than by recruitment of un-

used vascular channels or dilation of pulmonary veins. These results are in contrast to those of other vasodilators such as hydralazine, tolazoline, and diazoxide that cause a decrease in calculated PVR by increasing the flow (19). Heart rate did not change in our study animals during infusion of adenosine and ATP even at doses that caused an increase in cardiac output and a decrease in SVR. Other vasodilators that decrease SVR, such as tolazoline, cause tachycardia (3, 19). ATP and adenosine have been reported to have negative chronotropic effects (20) and are clinically used to decrease heart rate in the presence of supraventricular tachycardia (20). Adenosine is thought to exert its negative chronotropic effects by stimulation of A_1 receptors in heart (21). We did not observe bradycardia during infusion of ATP or adenosine in any of the animals. The favorable effect of adenosine and ATP at doses of 0.60 to 2.50 $\mu\text{mol/kg/min}$ on MV_E may be caused by the increase in coronary flow, without an increase in heart rate above control values.

Adenosine has been implicated as a mediator of coronary vasodilation that occurs during hypoxia and exercise (22). Increased plasma and interstitial fluid levels of adenosine have been observed in association with coronary vasodilation caused by hypoxia (23). We have shown that hypoxia causes a significant increase in coronary flow in newborn lambs and that the increase in flow resulted in a significant increase in O_2 delivery to the left ventricular myocardium. However, the increase in flow was not sufficient to compensate for the increase in O_2 consumption, because O_2 extraction and lactate levels in coronary sinus increased during hypoxia. Infusion of ATP or adenosine during hypoxia causes significant additional vasodilation at doses of 0.3 to 2.50 $\mu\text{mol/kg/min}$. The increase in coronary flow had a favorable effect on MOT and MVO_2 . This was indicated by decreases in O_2 extraction and coronary sinus lactate levels compared with control studies.

The animals in our study did not have a significant decrease in VO_2 during hypoxia. The decrease in SOT was compensated for by an increase in V_E . These results are in contrast to the previous observation of a decrease in VO_2 during hypoxia in newborn lambs (5). Moss *et al.* (5) have reported a decrease in VO_2 and an increase in lactate levels above 1 mmol/L during hypoxia caused by exposure to FIO_2 of 0.08 in newborn lambs. VO_2 and lactate levels did not change significantly at FIO_2 of 0.16 or 0.12. Our studies were done at an FIO_2 of 0.10 and resulted in a Pao_2 of 4.7 ± 0.5 kPa (35 ± 4 mm Hg), which is comparable to the Pao_2 reported by Moss *et al.* at an FIO_2 of 0.12. Our results are therefore consistent with exposure to moderate hypoxia, as reported by Moss *et al.* Lactate levels in our study did not increase above 1 mmol/L during hypoxia. The increase in lactate levels during hypoxia may be caused by increased work of breathing or a decrease in O_2 supply to the tissues. Addition of 5% CO_2 to the gas mixture to prevent a decrease in Paco_2 may have increased the respiratory drive and work of breathing in these animals. However,

the Paco_2 values did not differ between control, ATP, and adenosine studies. The lactate levels in control animals increased gradually during the 90 min of hypoxia. The lactate levels during infusion of adenosine and ATP at 2.5 $\mu\text{mol/kg/min}$ were lower than in control studies, coinciding with an increase in SOT. Therefore, the elevation in lactate levels was most likely caused by a decrease in O_2 supply to the tissues during alveolar hypoxia. Although arterial lactate levels decreased at an infusion rate of ATP and adenosine of 2.5 $\mu\text{mol/kg/min}$, a decrease in V_E was noted at lower doses of ATP and adenosine. These data indicate a lag between optimization of O_2 delivery and clearance of lactate from the blood. The coronary sinus lactate levels were lower than arterial values in newborn lambs, indicating a net lactate uptake by the myocardium. This is consistent with previous studies (24) showing lactate uptake by myocardium in both fetus and newborn. The coronary sinus lactate levels increased with hypoxia and remained elevated in the control experiments. Coronary sinus lactate levels have decreased during infusion of adenosine and ATP at doses of 0.30 to 2.50 $\mu\text{mol/kg/min}$ compared with control values. The change in lactate levels correlate with an increase in O_2 transport and decrease in O_2 extraction at these doses.

The increase in Pao_2 at 2.5 $\mu\text{mol/kg/min}$ infusion rates of adenosine and ATP was caused by an increase in mixed venous Po_2 (2.8 ± 0.5 to 3.6 ± 0.4 kPa) at this dose. The large increase in cardiac output and decrease in V_E at this dose of ATP and adenosine probably caused an increase in mixed venous Po_2 . Arterial Hb concentration increased during hypoxia, probably by hemoconcentration. This may reflect an attempt to optimize SOT during a period of decreased O_2 availability. A similar increase in Hb concentration during hypoxia was previously reported in lambs (5). The Hb concentration gradually returned to baseline as hypoxia was continued. The decrease in Hb may be caused by a dilutional effect of fluid administered during the study. A greater decrease in Hb concentration was noted in animals during infusion of ATP and adenosine at 2.5 $\mu\text{mol/kg/min}$ and may be related to generalized vasodilation observed at this dose.

The core temperature in our animals decreased only after prolonged hypoxia. Each experiment lasted a total of 90 min, and the animals were kept at an FIO_2 of 0.10 during this time. There were no significant differences between the three experiments in the decrease in core temperature. The increase in O_2 delivery and decrease in V_E in ATP and adenosine groups were not accompanied by an increase in core temperature. It is possible that temperature may have increased if the infusion of ATP or adenosine was continued for a longer period of time. The core temperature decreased by an average of 1°C in our lambs. Sidi *et al.* (25) have previously reported that the decrease in core temperature during hypoxia depends on ambient temperature. They observed a decrease of 2.1°C at an ambient temperature of 17°C and a decrease of 0.4°C at an ambient temperature of 25°C in newborn lambs at an FIO_2 of 0.09 for 1 h. Our studies were done

at an ambient temperature of 26°C, and the hypoxia ($\text{FIO}_2 = 0.10$) was maintained for longer than 1 h. Therefore, our results on the decline in core temperature are consistent with the results of Sidi *et al.* at higher temperatures. The lack of a decrease in VO_2 during hypoxia in our animals is also consistent with the observation of Sidi *et al.* in lambs at a higher (25°C) ambient temperature. The animals in our study have shown a shivering response during the decrease in core temperature, presumably to increase heat production. Inasmuch as VO_2 did not decrease, the decrease in temperature is unlikely to be an attempt to sacrifice thermogenesis during decreased O_2 supply. Moss *et al.* (5) speculated that hypothermia in newborn lambs during hypoxia may represent greater heat loss at this age because of increased body surface area. The increase in work of breathing during hypoxia may also cause greater evaporative heat loss from the respiratory tract.

The purine nucleotides seem to exert their vasodilator effects by stimulation of specific receptors present on endothelial and smooth muscle cells of blood vessels. Burnstock (6) proposed that the effects of purine nucleotides are mediated by two types of purinergic receptors. P_1 receptors are more sensitive to adenosine, act via the adenylyl cyclase system, and are inhibited by methylxanthines. P_2 receptors are more sensitive to ATP and are not antagonized by methylxanthines. Based on receptor affinity and ligand binding studies, P_1 receptors have been further classified into A_1 and A_2 subtypes; A_1 receptors inhibit adenylyl cyclase and appear predominantly in the heart, where they mediate the negative chronotropic and inotropic effects of adenosine (21). A_2 receptors are present on the vascular endothelial cells, stimulate adenylyl cyclase, and mediate the vasodilator effects of adenosine (21). P_2 receptors are also classified into P_{2x} , P_{2y} , P_{2z} , and P_{2t} receptors (21). The relative predominance of these receptors on the cell varies between different organs, with P_{2y} receptors being more predominant on vascular endothelial cells. Stimulation of purine receptors on endothelial cells is accompanied by release of endothelium-derived nitric oxide and prostacyclin (26). Both adenosine and ATP have been shown to cause endothelium-dependent vasodilation in newborn (27) and fetal lambs (28). The mechanism of the selective pulmonary vasodilation seen at lower infusion rates of adenosine and ATP is probably a rapid clearance of the nucleotide by the pulmonary vascular endothelium during the first pass. Fineman *et al.* (10) have shown that infusions of ATP in identical doses into the right atrium and the left atrium cause either pulmonary or systemic vasodilation. We have previously reported (13) that adenosine infusion in hypoxic lambs at doses of 0.01 to 0.15 $\mu\text{mol/kg/min}$ causes no change in left atrial adenosine levels, whereas larger doses cause a dose-dependent increase in adenosine levels in the left atrium.

The selective pulmonary vasodilation caused by adenosine and ATP in lower doses may be useful in the management of infants with pulmonary hypertension.

The wide range of doses that seem to be selective for the pulmonary circulation may facilitate better titration of the dose needed for individual patients. The short half-life and rapid clearance of adenosine and ATP by endothelium may allow us to use these drugs in infants with compromised renal function, a situation in which tolazoline therapy seems to be less desirable (29). However, the presence of right-to-left shunts at the level of the foramen ovale or ductus arteriosus in these infants may allow adenosine and ATP to enter the systemic circulation even in low doses and result in systemic vasodilation. We have ligated the ductus arteriosus in the lambs during surgery. The foramen ovale was closed at autopsy in our animals, and this observation is consistent with closure of the foramen by 1 wk of age reported previously by Dawes *et al.* (30). In addition, the PAP in our lambs was increased by alveolar hypoxia, whereas infants with persistent pulmonary hypertension usually have near-systemic PAP in spite of receiving high FIO_2 . The response of pulmonary vessels in infants with pulmonary hypertension may also be different from that of newborn lambs with normal pulmonary circulation because of their structural (31, 32) and possibly functional differences. These differences should be considered when extrapolating our results to infants with persistent pulmonary hypertension. Additional studies are required to evaluate fully the role of purinergic receptors in cardiopulmonary changes seen during the perinatal period and the usefulness of adenosine and ATP as therapeutic agents in the management of newborn infants with pulmonary hypertension.

REFERENCES

1. Fox WW, Gewitz MH, Dinwiddie R, Drummond WH, Peckham GJ 1977 Pulmonary hypertension in the perinatal asphyxia syndromes. *Pediatrics* 59:205-211
2. Drummond WH, Gregory GA, Heymann MA, Phibbs RA 1981 The independent effects of hyperventilation, tolazoline and dopamine on infants with persistent pulmonary hypertension. *J Pediatr* 98:603-611
3. Konduri GG, Dawe EJ 1989 Tolazoline improves systemic and myocardial oxygen transport and oxygen consumption in hypoxemic newborn lambs. *Pediatr Res* 25:241A(abstr)
4. Lister G, Walter TK, Versmold HT, Dallman PR, Rudolph AM 1979 Oxygen delivery in lambs: cardiovascular and hematologic development. *Am J Physiol* 237:H668-H675
5. Moss M, Moreau G, Lister G 1987 Oxygen transport and metabolism in the conscious lamb: the effects of hypoxemia. *Pediatr Res* 22:177-183
6. Burnstock G 1990 Overview: purinergic mechanisms. In: Dubyak GR, Fedan JS (eds) *Biological Actions of Extracellular ATP*. New York: Academy of Sciences, New York, pp 1-18
7. Konduri GG, Gervasio CT, Theodorou AA 1993 Role of adenosine triphosphate and adenosine in oxygen-induced pulmonary vasodilation in fetal lambs. *Pediatr Res* 33:533-539
8. Konduri GG, Theodorou AA, Mukhopadhyay A, Deshmukh DR 1992 Adenosine triphosphate and adenosine increase the pulmonary blood flow to postnatal levels in fetal lambs. *Pediatr Res* 31:451-457
9. Konduri GG, Woodard LL 1991 Selective pulmonary vasodilation by low dose infusion of adenosine triphosphate (ATP) in newborn lambs. *J Pediatr* 119:94-102
10. Fineman JR, Crowley MR, Soifer SJ 1990 Selective pulmonary vasodilation with ATP-MgCl₂ during pulmonary hypertension in lambs. *J Appl Physiol* 69:1836-1842
11. Paidas CN, Dudgeon DL, Haller JA, Clemens MG 1988 Adenosine triphosphate: a potential new therapy for hypoxic pulmonary hypertension. *J Pediatr Surg* 23:1154-1160
12. Soifer SJ, Fineman JR, Brook M, Heymann MA 1992 ATP-MgCl₂ produces selective pulmonary vasodilation in children with postoperative pulmonary hypertension. *Pediatr Res* 31:36A(abstr)
13. Konduri GG, Woodard LL, Mukhopadhyay A, Deshmukh DR 1992 Adenosine is a pulmonary vasodilator in newborn lambs. *Am Rev Respir Dis* 146:670-676

14. Konduri GG, Garcia DC, Kazzi NJ, Shankaran S 1993 Adenosine infusion improves oxygenation in infants with persistent pulmonary hypertension of newborn. *Pediatr Res* 33:219A(abstr)
15. Custer JR, Hales CA 1985 Influence of alveolar oxygen on pulmonary vasoconstriction in newborn lambs *versus* sheep. *Am Rev Respir Dis* 132:326-331
16. Downing SE, Lee JC, Taylor JFN, Halloran K 1973 Influence of norepinephrine and digitalis on myocardial oxygen consumption in the newborn lamb. *Circ Res* 32:471-479
17. Sidi D, Kuipers JR, Heymann MA, Rudolph AM 1982 Recovery of cardiovascular function in newborn lambs after thoracotomy. *Pediatr Res* 16:705-710
18. Winer BJ 1971 *Statistical Principles in Experimental Design*, 2nd Ed. McGraw-Hill, New York
19. Rich S, Martinez J, Lam W, Levy PS, Rosen KM 1983 Reassessment of the effects of vasodilator drugs in primary pulmonary hypertension: guidelines for determining a pulmonary vasodilator response. *Am Heart J* 105:119-127
20. Belhassen B, Pelleg A 1984 Electrophysiologic effects of adenosine triphosphate and adenosine on the mammalian heart: clinical and experimental aspects. *J Am Coll Cardiol* 4:414-424
21. Paton DM, Taerum T 1990 A comparison of P_1 - and P_2 -purinoceptors. In: Dubyak GR, Fedan JS (eds) *Biological Actions of Extracellular ATP*. New York Academy of Sciences, New York, pp 165-171
22. Berne RM 1980 The role of adenosine in the regulation of coronary blood flow. *Circ Res* 47:807-813
23. Berne RM, Knabb RM, Ely SW, Rubio R 1983 Adenosine in the local regulation of blood flow: a brief review. *Fed Proc* 42:3136-3142
24. Fisher DJ, Heymann MA, Rudolph AM 1980 Myocardial oxygen and carbohydrate consumption in fetal lambs *in utero* and in adult sheep. *Am J Physiol* 238:H399-H405
25. Sidi D, Kuipers JRG, Heymann MA, Rudolph AM 1983 Effects of ambient temperature on oxygen consumption and the circulation in newborn lambs at rest and during hypoxemia. *Pediatr Res* 17:254-258
26. Pearson JD, Slakey LL, Gordon JL 1983 Stimulation of prostaglandin production through purinoceptors on cultured porcine endothelial cells. *Biochem J* 214:273-276
27. Fineman JR, Heymann MA, Soifer SJ 1991 N-nitro-L-arginine attenuates endothelium-dependent pulmonary vasodilation in lambs. *Am J Physiol* 260:H1299-H1306
28. Konduri GG, Theodorou AA 1992 Nitro-L-arginine attenuates the pulmonary vasodilation caused by adenosine in fetal lambs. *Pediatr Res* 31:62A(abstr)
29. Ward RM, Kendig JW, Daniel CH 1982 Potentially lethal accumulation of tolazoline in neonates with oliguria. *Pediatr Res* 16:313A(abstr)
30. Dawes GS, Mott JC, Widdicombe JG 1955 Closure of the foramen ovale in newborn lambs. *J Physiol (Lond)* 128:384-395
31. Haworth SG, Reid L 1976 Persistent fetal circulation: newly recognized structural features. *J Pediatr* 88:614-620
32. Murphy JD, Rabinovitch M, Goldstein JD, Reid L 1981 The structural basis of persistent pulmonary hypertension of the newborn infant. *J Pediatr* 98:962-967