Influence of Increased Pulmonary Vascular Pressures on the Closure of the Ductus Arteriosus in Newborn Lambs

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ABSTRACT. Neonatal conditions associated with increased pulmonary artery pressure have an increased incidence of patent ductus arteriosus. We operated on 15 near term fetal lambs and placed mechanical occluders into or around both branch pulmonary arteries so that main pulmonary artery blood pressure could be controlled. The lambs were delivered and ventilated for 4 h. In seven lambs, the branch vessels were obstructed so that pulmonary artery pressure increased to equal aortic pressure; in eight lambs (control), the branch vessels were not obstructed. There were no significant differences between the two groups in circulating prostaglandin E_2 or 6 keto $F_{1\alpha}$ concentrations, PaO_2 , pH, or $Paco_2$. Despite these similarities, ductus resistance in the lambs with elevated pulmonary pressure was significantly less than that in the control lambs. After the 4 h measurements, we studied the ductus *in vitro*. We have previously found that ductus arteriosus constriction produces ischemia of its muscle wall that limits its ability to dilate or constrict any further. Ductus from lambs with elevated pulmonary pressure had a significantly increased ability to respond to oxygen, prostaglandin E_2 , and indomethacin compared with ductus from control lambs; these findings are consistent with less ductus constriction *in vivo*. Thus, the high incidence of patent ductus arteriosus in neonates with elevated pulmonary vascular resistance may be due in part to the increased pulmonary vascular pressure, which opposes ductus constriction and preserves ductus responsiveness. Conversely, the normal drop in pulmonary pressure that occurs in full term infants may facilitate the closure of the ductus after delivery. (*Pediatr Res* 25: 136–142, 1989)

Abbreviations

PDA, patent ductus arteriosus
PG, prostaglandin
 F_iO_2 , fraction of inspired oxygen
 Q_{LV} , left ventricular output
 $Q_{ductus\ L \rightarrow R}$, left to right blood flow through the ductus arteriosus
 $Q_{ductus\ R \rightarrow L}$, right to left blood flow through the ductus arteriosus
 $Q_{ductus-total}$, total combined blood flow through the ductus arteriosus
 Q_{FO} , right to left blood flow through the foramen ovale

R_{ductus} , ductus arteriosus resistance
 P_{pa} , pulmonary arterial blood pressure
 Q_{pulm} , pulmonary blood flow
PIP, peak inspired ventilator pressure
IMV, ventilator rate
HR, heart rate
 P_{AO_2} , aortic blood pressure
 P_{SVC} , superior vena caval blood pressure
 P_{LVED} , left ventricular end diastolic pressure
6 keto $PGF_{1\alpha}$, prostaglandin 6 keto $F_{1\alpha}$

Alterations in the balance between the constricting effect of oxygen and the vasodilating effect of prostaglandins play a significant role in the incidence of PDA (1). However, little attention has been paid to the effect that changes in ductus luminal blood pressure might have on the rate of ductal closure. One might expect that the greater the intravascular pressure, which opposes ductus constriction, the slower the rate of ductus closure, *i.e.* the greater the pressure load against which the ductus smooth muscle constricts, the greater the amount of work it must perform to achieve the same degree of constriction.

Several observations lead one to expect that changes in luminal pressure will affect ductus resistance by altering the caliber of the vessel: 1) *In utero*, the circulating PaO_2 is low and prostaglandin E_2 concentration is high so that the ductus is dilated; pulmonary vascular resistance and pressure is greater than systemic vascular resistance and pressure (2, 3). When ductus constriction is induced *in utero*, there is a tendency for the ductus to constrict first at the end with the lower resistance vascular bed: the aortic end (3). 2) After delivery, pulmonary vascular resistance and pressure decreases compared with systemic vascular resistance and pressure. Ductal constriction after delivery also tends to occur first at the end with the lower resistance vascular bed; however, this time it is the pulmonic end (4). 3) Full term infants rarely have evidence of a PDA 48 h after delivery; however, those infants who have idiopathic pulmonary hypertension after delivery frequently have delayed closure of their ductus arteriosus (5–7). We hypothesized that the normal decrease in pulmonary vascular resistance and pressure that occurs in healthy, full term infants after delivery would decrease the force that opposes ductus constriction, and facilitate ductus closure. Conversely, clinical situations, associated with increased pulmonary vascular resistance and pressure, would offer a greater intravascular distending force to oppose ductus constriction and would be associated with delayed closure of the ductus. We therefore studied the effect of increased pulmonary vascular pressure on the ability of the ductus arteriosus to constrict after delivery.

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MATERIALS AND METHODS

Neonatal lambs—in vivo. Fifteen mixed breed, Western, time-dated, pregnant ewes at 138–142 days of gestation (140 ± 1 , $\pm$ SD days, term 145 days) were operated on under epidural anesthesia. Ketamine was administered for sedation to the ewe every 15–30 min as needed (usually a total of 250–300 mg). Local anesthesia with 0.25% lidocaine hydrochloride was used for all fetal incisions. Using this combination of ketamine and local anesthesia, fetuses do not respond to surgical incisions. A hysterotomy was performed, and polyvinyl catheters were inserted into the pedal vein and artery and right brachial artery of the fetus and advanced into the inferior vena cava, descending aorta, and ascending aorta, respectively. A left thoracotomy was performed, and the left lung was retracted to expose the great vessels. Taking particular care to stay away from the ductus arteriosus, the following procedures were performed: ligation of the left hemizygous vein, placement of a mechanical snare (made from a polyvinyl-covered cardiac catheterization guide wire and a catheter sheath) around the left main pulmonary artery, direct placement of a 4 F Swan-Ganz balloon catheter (through a catheter sheath in the main pulmonary trunk) into the right pulmonary artery, and placement of a catheter in the main pulmonary trunk. The pulmonary catheter, snare, and balloon catheter were brought through the fetal chest wall, and the thoracotomy was closed. The fetus was given pancuronium bromide (0.4 mg intravenously) for paralysis and diazepam (0.3 mg/kg intravenously) for sedation. The trachea was intubated with a 4.5 F cuffed endotracheal tube. The umbilical cord was ligated, and the lamb was delivered to a warming table and ventilated (in the prone position) with an infant ventilator. Through a peripheral cutdown in the neck (using local anesthesia), additional intravascular catheters were passed into the newborn lamb's left ventricle and superior vena cava, and an umbilical artery also was cannulated from the umbilical cord.

The initial ventilator settings were: PIP, 28 cm water; positive end expiratory pressure, 3 cm water; inspiratory time, 0.6 sec; and respiratory rate, 40 breaths/min. The initial F_iO_2 was 1.0. The PaO_2 was maintained between 50 and 170 torr by adjusting the F_iO_2 ; the $PaCO_2$ was maintained between 25 and 40 torr by adjusting the PIP and respiratory rate. An intravenous infusion of 5% dextrose with 0.2 N NaCl in water at a rate of 5 ml/kg/h was begun after delivery. The lambs initially were towel-dried and covered with a plastic sheet to reduce evaporative losses. Rectal temperature was maintained between 38° and 39°C with aquamatic K-pads and with radiant heat lamps. Heparinized placental blood was transfused to replace blood lost from sampling.

Descending P_{AO} , P_{pa} , and right atrial and left ventricular pressures were measured continuously using Beckman pressure transducers (Beckman Instruments, Inc., Fullerton, CA) and a Sormedics Dynograph R611 multichannel recorder (Anaheim, CA). The signals were averaged electronically to obtain mean pressures.

Although it would be desirable to measure phasic instantaneous blood flow across the ductus using a flow probe, this technique cannot be used accurately with the ductus because the contact with the probe changes as the ductus constricts; in addition, application of probes around the ductus will damage the underlying tissue and make later measurements of ductus contractility impossible. Therefore, we used the radioactive microsphere method (8–10) to determine mean blood flows at particular time points. We injected three different radiolabeled 15 μ m microspheres (approximately 10^6 of each) into the left ventricle, inferior vena cava, and superior vena cava whereas reference blood samples were withdrawn from the ascending and descending aorta (8–10). Nine different microspheres were used: ^{153}Gd , ^{57}Co , ^{114}In , ^{51}Cr , ^{113}Sn , ^{85}Sr , ^{95}Nb , ^{54}Mn , and ^{65}Zn . After the experiment, the lamb was autopsied to check catheter placement and function of the mechanical snare and balloon catheter.

The organs were removed, weighed, and placed in formalin. They were incinerated at 325°C for 72 h, pulverized, and packed into counting vials. The amount of each radionuclide in the organs and the reference blood samples was measured with a well-type γ -scintillation counter and a multiple channel pulse height analyzer.

Q_{LV} and $Q_{ductus\ L \rightarrow R}$ were calculated using the concentration of microspheres (that were injected into the left ventricle) in the reference samples, the total number of microspheres recovered from the whole animal, and the total number of microspheres in the lungs (8–10). In this study (see "Results"), we found that bronchial flow to the left lung (measurements made in the lambs in which the left pulmonary artery had been ligated) was only $0.6 \pm 0.3\%$ (mean $\pm$ SD, $n = 21$ measurements) of Q_{LV} . If we assume that bronchial blood flow to the left lung represents $1/3$ of the bronchial flow to the total lung, then the total bronchial flow was less than 2% of Q_{LV} . In comparison, the $Q_{ductus\ L \rightarrow R}$ -total was $30 \pm 3\%$ of Q_{LV} during the first 4 h after delivery (see "Results," Figs. 1 and 2). Therefore, the bronchial flow makes up less than 10% of the $Q_{ductus\ L \rightarrow R}$ -total. Measurements of Q_{FO} and $Q_{ductus\ R \rightarrow L}$ were made using microspheres that were injected into the inferior and superior vena cava (9). Pulmonary blood flow was calculated as the sum of the venous return to the right atrium plus $Q_{ductus\ L \rightarrow R}$ minus ($Q_{FO} + Q_{ductus\ R \rightarrow L}$) (9).

To compare relative degrees of ductus constriction *in vivo*, we estimated the average R_{ductus} in the direction of the mean pressure gradient across the vessel (mean P_{AO} minus mean P_{pa} difference). Therefore, R_{ductus} was calculated by dividing the mean P_{AO} minus P_{pa} difference by the appropriate ductus arteriosus flow (either $Q_{ductus\ L \rightarrow R}$ or $Q_{ductus\ R \rightarrow L}$, depending on whether the pressure gradient was $L \rightarrow R$ or $R \rightarrow L$). This estimate assumes that there is laminar flow through the ductus, that mean pressure differences are an adequate reflection of the changes in driving pressure across the ductus during the cardiac cycle, and that net flow through the ductus is unidirectional. Because turbulence may develop at the pulmonic end of the ductus as it constricts, the pulmonary artery catheter was placed as far away from the ductus as possible (close to the pulmonary valve). If there were turbulent flow around the catheter, then just measuring the pressure drop across the ductus arteriosus would overestimate the change in driving energy across the vessel. This would lead to an overestimation of R_{ductus} . This measurement of R_{ductus} has been found to be a useful estimate of ductus constriction when flow through the ductus has been unidirectional from aorta to pulmonary artery (8, 11–13).

Arterial pH, $PaCO_2$ and PaO_2 were measured with a Corning 158 blood gas analyzer (Corning Medical and Scientific, Medfield, MA).

For PGE_2 and 6 keto $PGF_{1\alpha}$ measurements, 10-ml blood samples from pulmonary artery and descending aorta were withdrawn into chilled, heparinized tubes that contained indomethacin (2 μ g/ml blood) and were centrifuged immediately at $2000 \times g$ at 4°C for 30 min. The procedures for extraction, chromatographic separation, and RIA of PGE_2 and 6 keto $PGF_{1\alpha}$ have been detailed previously (11, 12, 14). In brief, 2 ml of plasma were mixed with 2000 dpm of 3H - PGE_2 (130 Ci/mmol, New England Nuclear, Boston, MA) or 3H -6 keto $PGF_{1\alpha}$ (110–130 Ci/mmol, New England Nuclear) for calculation of recovery and acidified to pH 4.0 with citric acid. The prostaglandins in the plasma samples were extracted with a cyclohexane:ethyl acetate (1:1) mixture for PGE_2 or ethyl acetate alone for 6 keto $PGF_{1\alpha}$.

We measured PGE_2 or 6 keto $PGF_{1\alpha}$ by RIA after their separation by silicic acid chromatography. For each concentration point of the standard curve for PGE_2 or 6 keto $PGF_{1\alpha}$, we carried a 2-ml sample of lamb plasma, preincubated with activated charcoal to remove PGE_2 or 6 keto $PGF_{1\alpha}$, through the purification process used for the 2-ml experimental plasma samples. Appropriate PGE_2 or 6 keto $PGF_{1\alpha}$ standards were added to each of these evaporated residues. This procedure yielded reproducible standard curves with linear titration from 2.5–250

pg PGE₂ and 2.5–250 pg 6 keto PGF_{1 α} . The cross reactivity of the antiserum to PGE₂ (Institut Pasteur, Paris) and 6 keto PGF_{1 α} (Advanced Magnetics, Inc., Cambridge, MA) have been described previously (12, 14). The recovery of ³H-PGE₂ (50–60%) or ³H-6 keto PGF_{1 α} (40–60%) was determined for each sample before RIA. Values were corrected for sample recovery.

Fifteen lambs were delivered and ventilated for 4 h after birth. Elevated main P_{pa} was produced in seven lambs by totally occluding the left pulmonary artery (with the snare) and partially occluding the right pulmonary artery by inflating the balloon catheter. Within 15 min after delivery, the snare and balloon were manipulated to keep mean pulmonary blood pressure within 2–5 torr of P_{AO}. This was maintained for the next 4 h. In eight control lambs, the left pulmonary artery snare was not tightened and the balloon was not inflated; there was a predictable drop in P_{pa} after delivery in these animals. Hemodynamic and blood flow measurements were made at 1, 2.5, and 4 h after delivery. PGE₂ and 6 keto PGF_{1 α} were measured at 2 and 4 h after delivery.

Lamb ductus—in vitro. After completing the 4 h *in vivo* measurements, the lambs were killed rapidly by exsanguination. The ductus was dissected free from the loose adventitial tissue and divided into 1-mm thick rings that were placed in separate 150-ml organ baths kept in the dark within an enclosed box. The rings were suspended between two stainless steel hooks in a modified Krebs-Tris solution at 38°C (11, 15). Isometric responses of circumferential tension were measured by Grass FT03C force transducers (Grass Instrument Co., Quincy, MA) (11, 15).

Each of the rings was stretched to an initial length of 7–8 mm. At this length, rings of ductus arteriosus have their maximal contractile response to increases in Po₂ (15). Initially the Po₂ of the bath solution was maintained at 20–25 torr, and the rings were allowed to equilibrate for 45 min until a steady tension developed. The bath solution was then bubbled with 100% O₂ (to a Po₂ of 680–700 torr) until the tension achieved a new plateau. Indomethacin was added to the bath in a final concentration of 2 μ g/ml (5.6×10^{-6} M), and the rings were allowed to achieve a new increase in tension over the next h. A cumulative dose response to PGE₂ was obtained in the oxygen- and indomethacin-contracted rings; we allowed the tension to achieve a new plateau before higher concentrations of PGE₂ were added. After the experiment, the tissues were removed from the baths and blotted dry so that their wet wt could be determined. The tension developed in the ring was expressed as force/cross-sectional area (g/mm²) (15).

To measure PGE₂ production by the isolated ductus arteriosus, 1 mm ductus rings from the same lambs were suspended (as described above) in 5-ml organ baths designed for the collection of the bath solution. The 5-ml bath solution was changed every 40 min. Initially the bath solution was bubbled with 100% N₂ for 80 min (Po₂: 26–32 torr) and the ring was allowed to equilibrate until a steady tension developed. The bath solution was then bubbled with 100% O₂ (Po₂: 600–700 torr) for 80 min. Indomethacin containing solution (5.6×10^{-6} M) was then added while the rings were bubbled with 100% O₂ for another 80 min. The 5 ml bath solutions from the 40- to 80-min oxygen exposure and the 40- to 80-min oxygen-plus-indomethacin exposure were collected, extracted, purified, and assayed for PGE₂ as described previously (11).

Indomethacin (Sigma Chemical Co., St. Louis, MO) was prepared in ethanol (16 mg/ml), and aliquots were added to the bath solution. PGE₂ and 6 keto PGF_{1 α} , generously supplied by Dr. J. E. Pike of the Upjohn Co. (Kalamazoo, MI), were dissolved in ethanol before use. Concentrations of PGE₂ and 6 keto PGF_{1 α} are expressed as free acid concentrations. The protocol outlined above was approved by the Committee on Animal Research of the University of California, San Francisco.

Statistics. We compared measurements between control lambs

and lambs with elevated P_{pa} by an unpaired *t*-test, or Mann Whitney U test where appropriate. The level of significance was *p* < 0.05 (two tailed). The level of significance was adjusted for multiple comparisons of group means with a Bonferroni correction. The effect of elevated P_{pa} on blood pressures, flows, and blood gases at various times after delivery was tested by two-factor (elevated blood pressure and age) ANOVA with replication. When a significant interaction between elevated pressures and time was found, the effect of elevated pressure was tested independently by an unpaired *t*-test. The data are reported as means $\pm$ SD.

RESULTS

The effects of partial obstruction of branch pulmonary blood flow on blood pressures are depicted in Table 1. In the control animals, the P_{pa} rapidly dropped to 60% of P_{AO} after delivery and remained at this level for the next 4 h. In contrast, in the experimental animals with elevated P_{pa}, we were able to keep the P_{pa} close to P_{AO} during the 4 h after delivery. Lambs with mechanically elevated P_{pa} had significantly greater P_{pa} and significantly lower P_{AO} than the control lambs at each measurement point. In addition, $\dot{Q}_{LV}$ and $\dot{Q}_{pulm}$ were significantly diminished in the lambs with elevated P_{pa} (Fig. 1).

Despite the increase in main P_{pa} in the mechanically obstructed group, there was no change in $\dot{Q}_{FO}$ when compared with controls (Fig. 2). Elevated P_{pa} produced a small but significant increase in $\dot{Q}_{ductus\ R \rightarrow L}$ during the first 4 h after delivery (Fig. 2).

Lambs with mechanically elevated P_{pa} had their left pulmonary arteries totally occluded. Therefore, microspheres from the left ventricular injection that lodged in the left lung represent the proportion of $\dot{Q}_{LV}$ that came through the bronchial circulation. This was only $0.6 \pm 0.3\%$ of $\dot{Q}_{LV}$. In these lambs, $98 \pm 2\%$ (*n* = 21) of the microspheres injected into the left ventricle that were recovered in the lungs were found in the right lung and represent $\dot{Q}_{ductus\ L \rightarrow R}$. The left pulmonary artery was not occluded in the control lambs; in this group, $65 \pm 12\%$ (*n* = 24) of $\dot{Q}_{ductus\ L \rightarrow R}$ was distributed to the right lung. Elevated P_{pa} produced a significant decrease in the net left to right ductus shunt that was present during the first 4 h after delivery (Fig. 2).

We estimated the degree of ductus arteriosus constriction that occurred *in vivo* by calculating the R_{ductus}. Lambs in the control group had no net right to left flow and only a unidirectional left to right flow through their ductus. All lambs in the elevated P_{pa} control group during the 4 h after delivery (Table 1). The in the elevated P_{pa} group was significantly less than that in the control group during the 4 h after delivery (Table 1). The decreased amount of ductus constriction in the lambs with elevated main P_{pa} pressure could not be accounted for by differences in arterial blood gases (Fig. 3) or by differences in circulating concentrations of PGE₂ or 6 keto PGF_{1 α} (Table 2).

At the end of 4 h, the animals were killed to see how the effects of elevated main P_{pa} and decreased ductus arteriosus constriction *in vivo* altered the responsiveness of the ductus *in vitro*. When the ductus was removed from the lamb and cut in cross-section, we found that, in the lambs with elevated main P_{pa}, lumen diameter was greater than 2 mm, whereas the diameter in control lambs was less than 2 mm. This finding is consistent with our *in vivo* measurements of R_{ductus}.

As we have previously found, a ductus isolated from a near term control lamb, in which there was a significant drop in P_{pa} and constriction of the ductus after delivery, could generate only a small fraction of the active tension (induced by oxygen and indomethacin) of that generated by a ductus isolated from the near term fetus (Table 3, compare with values reported in Refs. 13 and 16). In addition, PGE₂ was not as effective in relaxing the ductus from our near term control lambs as it was in relaxing the ductus from near term fetal lambs (Table 3, compare with values reported in Refs. 13 and 16).

Table 1. Effect of mechanical obstruction of branch pulmonary arteries on blood pressures, HR, and R_{ductus} *

Time after delivery (h)	Control (n = 8)			Increased P_{pa} (n = 7)		
	1	2.5	4	1	2.5	4
$\bar{P}_{AO}$	71±10†	78±7‡	77±8‡	57±10	65±8	62±8
$\bar{P}_{pa}$	44±14§	43±15‡	46±16‡	56±11	65±9	63±8
$\bar{P}_{SVC}$	2±1	2±1	2±1	3±1	3±1	3±1
$\bar{P}_{LVED}$	6±3	4±2	4±3	5±1	4±1	4±1
HR (beats·min ⁻¹)	204±34	200±39	201±36	174±48	168±23	188±39
R_{ductus} (torr·min ⁻¹ ·min·kg)	0.42±0.24†	0.46±0.24‡	0.35±0.14§	0.10±0.15	0.08±0.06	0.16±0.24

* Values are mean ± SD.

† $p < 0.02$ for differences between control and increased P_{pa} groups.‡ $p < 0.005$ for differences between control and increased P_{pa} groups.§ $p < 0.05$ for differences between control and increased P_{pa} groups. Fig. 1. Effect of mechanical obstruction of branch pulmonary arteries on $\dot{Q}_{LV}$ and $\dot{Q}_{PULM}$. Values are mean ± SD. * $P < 0.05$, ** $p < 0.01$, *** $p < 0.005$ for differences between control and increased (P_{pa}) groups.

In contrast, both the ability to contract actively (to either oxygen or indomethacin) as well as to actively relax (to PGE_2) was significantly greater in the ductus arteriosus of lambs with elevated P_{pa} (Table 3). There was little alteration of the ability of PGE_2 to relax the ductus isolated from lambs with elevated P_{pa} (and delayed ductus constriction *in vivo*) when compared with the ductus from fetal lambs (Table 3). The ductus from lambs with elevated P_{pa} was more than twice as sensitive to PGE_2 than was the ductus from control lambs. Similarly, after the same length of postnatal ventilation, the ductus from lambs with elevated pulmonary blood pressure retained more than twice the contractile activity (to either oxygen or indomethacin) as the ductus from the control lambs with reduced P_{pa} .

This difference in contractile behavior of the isolated ductus could not be accounted for by differences in endogenous PGE_2 production. The ductus from both control lambs and those with

elevated P_{pa} produced similar amounts of PGE_2 when exposed to oxygen; their PGE_2 production rate was inhibited to the same extent when they were exposed to indomethacin (Fig. 4).

DISCUSSION

These studies were performed in acutely prepared, sedated, and paralyzed neonatal lambs; extrapolating the results to the normal birth situation should be done with caution. With this caveat in mind, we found that both control lambs and those with obstructed branch pulmonary arteries were hemodynamically stable during the 4 h after delivery. Lambs with obstructed pulmonary arteries had significantly lower $\dot{Q}_{pulm}$, left ventricular output, and P_{AO} than control lambs. The decrease in P_{AO} and $\dot{Q}_{LV}$ probably is a reflection of the drop in right ventricular output caused by the marked increase in right ventricular afterload (17). Lambs with obstructed pulmonary arteries also required greater ambient oxygen and greater ventilator pressures and rates to maintain their blood gases in the same range as the control lambs. The blood gas alterations in the lambs with obstructed pulmonary arteries are not surprising when one considers: 1) the increased ratio of $\dot{Q}_{FO}$ and $\dot{Q}_{ductus}$ $R \rightarrow L$ compared to $\dot{Q}_{LV}$ (17% of $\dot{Q}_{LV}$ versus 12% in control lambs), 2) the decreased total P_{pa} , and 3) the decreased recirculation through the lungs of oxygenated blood from the left to right ductus shunt (18, 19).

Although the mechanisms that regulate patency and closure of the ductus arteriosus are only partially understood, most attention has been paid to the factors that directly alter ductal tone (1). There appears to be a balance between the constricting effect of oxygen and the vasodilating effect of the prostaglandins E_2 and I_2 . The marked sensitivity of the lamb ductus to PGE_2 suggests that this is the most important prostanoid for regulation of vessel patency in this species (1). Despite similar circulating concentrations of P_{ao_2} , PGE_2 , and 6 keto $PGF_{1\alpha}$ (a stable metabolite of PGI_2) (Fig. 3; Table 2) and similar rates of endogenous PGE_2 production (Fig. 4), we found decreased ductus constriction (using our estimate of R_{ductus}) in the lambs with elevated main P_{pa} (Table 1). Elevated intravascular pressure could affect ductus arteriosus caliber in several ways. First, for a given stimulus for equal-sized vessels, vasoconstriction would be inhibited in the vessel with the greater luminal pressure. Second, as resting wall tension is passively increased by elevated luminal pressure, the potential for additional active increases in tension may be significantly reduced (15). Our findings have parallels in the pulmonary vascular bed where changes in vascular pressure have a passive effect on vascular caliber (20). An increase in vascular pressure has been found to diminish the response of the pulmonary bed to both hypoxic (21) and nonhypoxic (22) stimuli.

Previously we have found that when the ductus arteriosus is removed from the neonatal lamb several h after delivery and studied *in vitro*, it has lost much of its ability to dilate or contract to vasoactive stimuli (13). This generalized loss of ductus respon-

Fig. 2. Effect of mechanical obstruction of branch pulmonary arteries on $\dot{Q}_{FO}$ and the $\dot{Q}_{\text{ductus } R \rightarrow L}$ and $\dot{Q}_{\text{ductus } L \rightarrow R}$. Values are mean $\pm$ SD. * $p < 0.005$, ** $p < 0.025$ for differences between control and increased Ppa groups.

Fig. 3. Effect of mechanical obstruction of branch pulmonary arteries on the F_iO_2 , PIP, and IMV needed to keep pH, P_{aO_2} , and P_{aCO_2} in a defined range. Values are mean $\pm$ SD. * $p < 0.05$, ** $p < 0.01$ for differences between control and increased Ppa groups.

siveness was directly related to the degree of prior ductus constriction and to the subsequent reduction in lumen diameter *in vivo*. The loss of responsiveness was independent of arterial P_{aO_2} , pH, and PGE_2 concentrations. In addition, this loss of responsiveness prevented the ductus arteriosus from reopening once it had constricted (13). This functional change probably reflects early ischemic damage to the inner muscle wall. In the normal newborn lamb, the increased P_{aO_2} and drop in circulating PGE_2 after delivery stimulate ductus arteriosus constriction (1). As the ductus constricts, intraluminal pressure drops on the pulmonary side due to the drop in pulmonary vascular resistance after birth (23). As pressure decreases on the pulmonary end of the ductus, less muscle tension is needed to overcome the transmural pressure at that end and to continue the progressive constriction. In addition, as the ductus constricts, there may be increased velocity

Table 2. Effect of mechanical obstruction of branch pulmonary arteries on the circulating concentrations of PGE_2 and 6 keto $PGF_{1\alpha}$ in aortic and pulmonary blood*

Time after delivery (h)	Aorta		Pulmonary artery	
	2	4	2	4
PGE_2 (pg/ml)				
Control	91 (1-286)	57 (1-271)	54 (1-68)	38 (1-41)
Increased P_{pa}	62 (1-86)	25 (1-49)	66 (1-163)	27 (1-72)
6 keto $PGF_{1\alpha}$ (pg/ml)				
Control	16 (2-1010)	15 (2-499)	9 (3-364)	9 (4-328)
Increased P_{pa}	3 (2-448)	3 (2-20)	5 (2-206)	5 (2-223)

* Values are median (range). Control group ($n = 8$); increased P_{pa} group ($n = 7$). There were no differences between control and increased P_{pa} groups that reached a level of significance <0.05 .

of red blood cell flow through the closing lumen. As a result there would be less potential energy stretching the vessel walls and therefore less intraluminal pressure to oppose constriction. However, as the ductus constricts it develops ischemic damage and becomes less capable of responding to vasoactive stimuli. How does the ductus remain closed once it has suffered ischemic damage? One may infer from the law of Laplace that as a vessel constricts, greater intravascular pressures will be required to cause any subsequent increase in vessel diameter. Mathematical considerations suggest that there is a critical closing pressure below which the lumen of a vessel will be obliterated (24-26). It would therefore follow that once the ductus is constricted, it may be kept constricted by very little muscular effort (25).

In contrast, in newborn animals with persistently elevated pulmonary vascular resistance after delivery, initial constriction of the ductus and separation of the systemic and pulmonary circulations would not produce a drop in blood pressure on the pulmonary side of the ductus. The elevated P_{pa} would continue to oppose vessel constriction. Because the normal loss of ductus responsiveness to vasoactive stimuli appears to be due to the degree of postnatal ductus constriction, one would expect that the ductus from lambs with postnatal pulmonary hypertension would continue to have a fetal pattern of vasoactive responsiveness. In the present study, there was a reduced rate of ductus closure (*in vivo*) during the first 4 h after delivery in the lambs with elevated pulmonary blood pressure. Consistent with our

Table 3. *In vitro* behavior of ductus arteriosus taken from fetal lambs, newborn control lambs, and newborn lambs with elevated P_{pa} *

	Active tension			PGE ₂ inhibition of tension		
	O ₂ (g/mm ²)	Indomethacin (g/mm ²)	O ₂ + indomethacin (g/mm ²)	Threshold (pg/ml)	ED ₅₀ (pg/ml)	Maximum (%)
Fetal†			4.24±1.39		448±389	73±25
Control	1.17±0.59	0.28±0.09	1.45±0.63	535±508	1491±772	28±18
Increased P_{pa}	2.98±1.72‡	0.66±0.25§	3.63±0.65§	165±224	666±475‡	63±32‡

* Values are mean ± SD. First, each ring (22.7 ± 5.4 mg wet wt) was allowed to equilibrate in a low PO₂ environment; steady state tensions, which were produced by the ring when incubated in a low PO₂ environment were: control group = 2.17 ± 1.15 g/mm² (n = 8); increased P_{pa} group = 2.68 ± 1.07 g/mm² (n = 7). The active tension for oxygen is the difference between the steady state tension at low PO₂ and the tension after the rings had contracted with high PO₂; active tension for indomethacin is the difference between the steady state tension in high O₂ and high O₂ plus indomethacin; active tension for O₂ + indomethacin is the difference between the steady state tension in high O₂ plus indomethacin and the tension at low PO₂. PGE₂ inhibition of tension is expressed for the active O₂ + indomethacin tension. Threshold concentration is the lowest concentration of PGE₂ needed to produce a significant decrease in tension. ED₅₀ is the effective concentration of PGE₂ that will produce 50% of the maximum PGE₂ relaxation. Maximum relaxation is expressed as the percentage of inhibition of the active O₂ + indomethacin tension.

† Values for the ductus from fetal lambs were obtained from 10 near term fetuses (137 ± 1 days gestation) that were delivered by cesarean section and killed before their first breath (values were obtained from Ref. 13).

‡ $p < 0.01$ for differences between control and increased P_{pa} groups.

§ $p < 0.005$ for differences between control and increased P_{pa} groups.

|| $p < 0.05$ for differences between control and increased P_{pa} groups.

Fig. 4. Effect of mechanical obstruction of branch pulmonary arteries on the ability of isolated rings of ductus arteriosus to produce PGE₂. Values are mean ± SD. First, all rings (35.6 ± 14.7 mg) were allowed to equilibrate in a low PO₂ environment. Steady state low PO₂ tensions were: control group = 2.42 ± 0.73 g/mm² (n = 8), increased P_{pa} group = 2.78 ± 0.95 g/mm² (n = 7). The bath solution was then bubbled with 100% O₂. The PGE₂ released by the rings between 40–80 min of exposure to O₂ was measured. The rings were then exposed to indomethacin 2 µg/ml (5.6 × 10⁻⁶ M). The PGE₂ released by the rings between 40–80 min of exposure to indomethacin was measured. Indomethacin inhibited PGE₂ released from the rings by 90%. There were no differences between the two groups.

expectations, the ductus from lambs with elevated pulmonary pressures were still responsive to oxygen and PGE₂ and behaved more like the ductus from fetal lambs than the ductus from newborn lambs with reduced pulmonary blood pressures. These findings in newborn lambs may help to explain both the increased incidence of PDA in full term infants with respiratory failure and pulmonary hypertension (5, 6) and the persistent responsiveness of the ductus in these infants once the increased pulmonary vascular resistance has been removed (7). In addition, this study supports the hypothesis that the normal drop in pulmonary vascular resistance and pressure that occurs in full term infants after delivery may facilitate the closure of the ductus arteriosus.

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Announcements

1989 Annual Meetings

The American Pediatric Society, the Society for Pediatric Research, and the Ambulatory Pediatric Association will have meetings on May 1-5, 1989 at the Washington Sheraton Hotel, Washington, D.C.

Contact: SPR—Ms. Debbie Wogenrich, 2650 Yale Blvd., S.E., Suite 104, Albuquerque, NM 87106 (505)764-9099. APS—Dr. Audrey K. Brown, Secretary-Treasurer, Department of Pediatrics, SUNY, Health Science Center at Brooklyn, 450 Clarkson Avenue, Box 49, Brooklyn, NY 11203 (718)270-1692. APA—Ambulatory Pediatric Association, 6728 Old McLean Village, McLean, VA 22101 (703)556-9222.

Symposium on Developmental Endocrinology

A two-day symposium on Developmental Endocrinology sponsored by Ares-Serono Symposia will be held October 23-24, 1989, at the Hotel PENTA, Geneva, Switzerland. Three different sessions will cover new trends on *Fetal and Neonatal Endocrinology*, *Puberty*, and new aspects of the *Biology and the Molecular Biology of Growth and Development*.

This symposium is dedicated to Drs. Selma L. Kaplan and Melvin M. Grumbach.

For further information, please contact: Dr. P.C. Sizonenko/Dr. M.L. Aubert, Clinique de Pédiatrie, Hôpital Cantonal Universitaire, 30 Bd de la Cluse, CH-1211 Geneva 4, Switzerland, or Dr. S. Rossetti, Ares-Serono Symposia, Via Ravenna 8, I-00161 Rome Italy.

Symposium on Pediatric Neuro-Oncology

The premiere session of the International Symposium on Pediatric Neuro-Oncology is June 1-3, 1989, in Seattle, Washington.

The Symposium is sponsored by Children's Hospital & Medical Center, Seattle, Children's Hospital of Philadelphia, and University of Washington School of Medicine.

For program information, contact CME, SC-50, University of Washington, Seattle, Washington 98195.

Poster and oral presentations are invited; for abstract form, due February 15, contact Dr. Bleyer at (206) 526-2107 or Fax 206-527-3838.

Fetal and Neonatal Physiology

International Union of Physiological Sciences satellite symposium "Fetal and Neonatal Physiology," Karolinska Institute, Stockholm, Sweden, July 5-8, 1989.

The meeting will include symposia and poster sessions on fetal growth, neuromodulators during development, perinatal neuroendocrinology, and water and electrolyte transport. Keynote lectures will focus on molecular biology in perinatal medicine, neonatal thermogenesis, lung maturation, asphyxia, and prostaglandins.

For information write: Dr. Hugo Lagercrantz, Neonatal Research Unit, Karolinska Institute, Box 60400, S-104 01 Stockholm, Sweden.

Annual Meeting of the European Society for Pediatric Research

The Annual Meeting of the European Society for Pediatric Research will be held in Kraków, Poland, June 11-14, 1989. In addition to free paper and poster sessions, plenary sessions and workshops will be held on topics including acute renal failure, advances in genetics and immunology, applied genetics, perinatal epidemiology, neonatology, diabetes, and atrial natriuretic peptides. European Working Groups on Allergy and Clinical Immunology, Mineral Metabolism, Neonatology, and Perinatal and Pediatric Microcirculation will participate. *Deadline for receipt of abstracts will be January 31, 1989.*

For abstract forms and information, contact the President of the Meeting: Jacek J. Pietrzyk, 1st Department of Pediatrics, Institute of Pediatrics, 30-663 Kraków, Wielicka 265, Poland, phone 55-02-56, telex 032-5795 IP AM PL.

European Chief Editor

Since the term of European Chief Editor Dr. Kari O. Raivio will expire at the end of 1989, the position of European Chief Editor of **Pediatric Research** will be open.

Duties include evaluation and editorial decisions regarding European manuscripts and collaboration with the Editor-in-Chief and the Board of Trustees of the International Pediatric Research foundation toward developing the journal scientifically. Financial support for the office is provided.

Interested applicants should submit a curriculum vitae and the names of at least four local editors from different fields of pediatrics who would help evaluate manuscripts. Inquiries and applications should be sent by **March 1, 1989** to: Dr. Kari O. Raivio, University of Helsinki, Children's Hospital, 00290 Helsinki, Finland.