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Effects of 6-Keto-prostaglandin E_1 on Perinatal Pulmonary Vascular Resistance (41037)

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Abstract. The effects of a biologically active metabolite of PGI_2, 6-keto-PGE_1, were evaluated in fetal goats and newborn lambs using an *in situ*, constant-flow, isolated lower left lobe preparation. Intrapulmonary injections of 6-keto-PGE_1 (0.074–4.41 μg/kg) produced dose-dependent decreases in pulmonary vascular resistance, mean systemic arterial pressure, and heart rate in fetal goats. Fetal systemic responses to 6-keto-PGE_1 were significantly less following left atrial injections than after intrapulmonary injections. Newborn lambs also responded to intrapulmonary infusions of 6-keto-PGE_1 (0.078–5.15 μg/kg·min) with dose-dependent reductions of pulmonary vascular resistance and systemic arterial pressure. A possible role for 6-keto-PGE_1 in the modulation of perinatal pulmonary vascular resistance is discussed.

Wong *et al.* (1) have shown recently that hepatic metabolism of prostacyclin (PGI_2) in the rabbit may lead to the enzymatic formation of 6-keto-prostaglandin E_1 (6-keto-PGE_1). They suggested that the enzyme, 9-hydroxy-prostaglandin dehydrogenase, can act on either PGI_2 or its relatively inactive metabolite, 6-keto-$PGF_{1\alpha}$, to produce 6-keto-PGE_1. Additional evidence suggests the presence of this enzyme in human plasma (2) and human platelets (3).

The potency of 6-keto-PGE_1 on inhibiting platelet aggregation is similar to that of prostacyclin (4). Quilley *et al.* (5) have shown that 6-keto-PGE_1 is similar to PGI_2 in its ability to produce systemic hypotension and cause renal vasodilation; like PGI_2, 6-keto-PGE_1 escapes inactivation in the lung. Prostacyclin, which may be released from lungs of fetal lambs and goats upon initiation of ventilation (6), is a very potent dilator of the perinatal pulmonary circulation (7–9). If the enzyme systems for formation of 6-keto-PGE_1 are active in the perinatal period, this metabolite could be present in considerable quantities because of its apparent lack of pulmonary inactivation (5) and stability in aqueous solutions (4). Thus, if the actions of 6-keto-PGE_1 on perinatal pulmonary vasculature are similar to those of PGI_2, this compound could be important in maintenance of low pulmonary vascular resistance following the first breath (10).

The effects of 6-keto-PGE_1 on pulmonary and systemic circulations were evaluated in anesthetized fetal goats and newborn lambs using an *in situ*, pump-perfused, isolated lower left lobe preparation. In this model changes in pulmonary arterial pressure reflect changes in pulmonary vascular resistance since perfusion rate is held constant. We have reported our results in newborn lambs in a preliminary communication (11).

Materials and methods. Five newborn lambs (2–14 days of age, 4.0 ± 0.7 kg, mean ± SE) and mothers of five fetal goats (135–145 days gestation, 2.3 ± 0.3 kg) were anesthetized with chloralose (50 mg/kg, iv) and a tracheotomy was performed. Chloralose (10 mg/kg) was administered hourly to maintain anesthesia. Fetuses were delivered by cesarean section with umbilical circulation undisturbed and placed on a warmed table adjacent to the mother. A saline-filled rubber bag was placed over the fetal head to prevent breathing, and the fetal abdomen and chest were sutured to the uterus to minimize exposure of the umbilical cord. A cannula filled with warm saline was introduced into the fetal trachea and the head cover removed. Newborns were ventilated (Harvard respiration pump) with oxygen-enriched room air (O_2 = 30%). In both fetuses and newborn animals, a femoral artery was cannulated in order to monitor systemic arterial pressure

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(SAP) (Statham P23DC transducer), heart rate (HR), and arterial blood gases and pH (Instrumentation Laboratory pH/Blood Gas Analyzer 213). Left thoracotomy was performed with removal of the fourth rib to permit access to the left lung and left pulmonary artery. Following isolation and dissection from surrounding tissue of the left pulmonary artery, animals were anticoagulated with heparin (2000 U/kg, ia). Blood was withdrawn, via a catheter placed in a femoral vein, from the inferior vena cava above the level of the liver and pumped (Cole Parmer Masterflex pump) into the left pulmonary artery. Between 32 and 42% of the total lung as determined by weight was perfused in this manner. In fetal animals, left pulmonary blood flow ($\dot{Q}$) (Statham flowmeter with In Vivo Metric 3.0-mm cannulating electromagnetic flow probe) was maintained constant at a rate at which mean left pulmonary arterial pressure ($\overline{PAP}$) was equal to or slightly greater than mean SAP ($\overline{SAP}$). In newborns, flow was maintained constant at a rate which produced approximately 20 mm Hg pressure in the left pulmonary artery. A catheter was tied into the left atrial appendage for measurement of mean left atrial pressure ($\overline{LAP}$). Pressures, flow, and heart rate were recorded continuously on a Gould Brush 480 eight-channel polygraph.

6-Keto-prostaglandin E_1 (Upjohn) in dry form and in ethanol (7.5 mg/ml) was stored at −20°. Prior to experiments, aliquots of the ethanol stock solution were diluted with saline to final concentrations of 1, 10, and 150 μg/ml.

Linear regression analyses were performed by the least-squares method (12) for dose–response curves generated from fetuses and newborns. Responses evaluated were percentage change (percentage maximal change from baseline) in $\overline{PAP}$, $\overline{SAP}$, HR, and left pulmonary vascular resistance (PVR), calculated as ($\overline{PAP}$-$\overline{LAP}$) × body wt/$\dot{Q}$. Differences cited were significant at $P < .05$.

Results. Figure 1 shows the effects of an intrapulmonary injection of 6-keto-PGE$_1$ (0.89 μg/kg) on $\overline{PAP}$ in a fetal goat. The response was rapid in onset and was followed by systemic hypotension of lesser duration. Calculated PVR decreased by

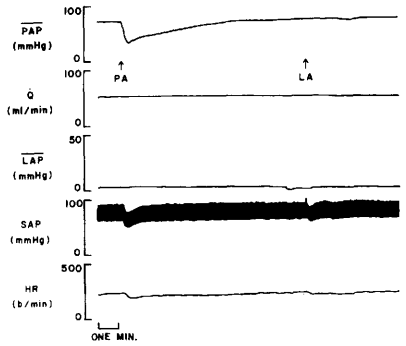

FIG. 1. Effects of intrapulmonary (PA) and left atrial (LA) injections of 6-keto-PGE$_1$ (0.89 μg/kg) on mean pulmonary arterial pressure ($\overline{PAP}$), mean left atrial pressure ($\overline{LAP}$), systemic arterial pressure (SAP), and heart rate (HR) during conditions of controlled pulmonary blood flow ($\dot{Q}$) in fetal goat 24.

57%, with a concomitant fall of 18% in $\overline{SAP}$.

The effects of 17 intrapulmonary injections of 6-keto-PGE$_1$ (0.074–4.41 μg/kg) in fetal goats for percentage change in PVR, $\overline{SAP}$, and HR are shown in Fig. 2. The regressions shown for each response were significantly related to log dose, with extrapolated threshold doses for pulmonary and systemic effects of 0.013 and 0.064 μg/kg, respectively. Similar responses were obtained when 6-keto-PGE$_1$ was infused into the pulmonary artery at equivalent doses for 1 min.

To eliminate the effects of passage through the pulmonary vascular bed on the response of the systemic circulation, 6-keto-PGE$_1$ was given at a postpulmonary site (via the left atrial catheter). Figure 1 shows the response of a left atrial injection (0.89 μg/kg). The systemic response (9% decrease in $\overline{SAP}$) was less than that observed for the equivalent dose given into the pulmonary circuit (18% decrease in $\overline{SAP}$). Left atrial injections produced significant reductions of $\overline{SAP}$ and HR with moderate decreases in $\overline{PAP}$. However, the response of the systemic vasculature to 6-keto-PGE$_1$ was significantly greater following intrapulmonary injections than following left atrial injections (Table I and Fig. 3), although extrapolated threshold doses were similar (0.064 and 0.044 μg/kg, respectively).

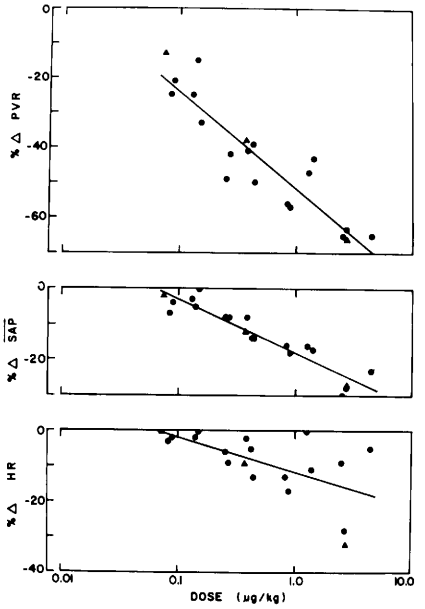

FIG. 2. Effects of intrapulmonary injections (●) and infusions (▲) of 6-keto-PGE$_1$ on pulmonary vascular resistance (PVR), mean systemic arterial pressure ($\overline{SAP}$), and heart rate (HR) in fetal goats.

One-minute infusions of 6-keto-PGE$_1$ (0.078–5.15 μg/kg·min) directly into the pulmonary artery of newborn lambs produced significant reductions in PVR (Fig. 4). The reduction in $\overline{SAP}$ with intrapulmonary infusions was similar to the pulmonary response; however, extrapolated threshold doses were 0.008 μg/kg·min for pulmonary effects and 0.080 μg/kg·min for systemic effects. The effects of 6-keto-PGE$_1$ on HR in lambs were variable. The single injection of 6-keto-PGE$_1$ (1.67 μg/kg) fell within the range of the infusion data.

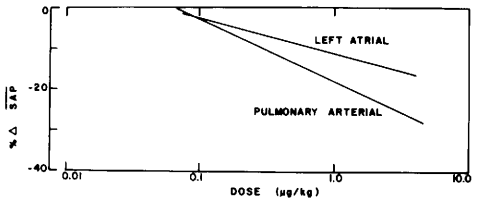

FIG. 3. Effect of pulmonary arterial and left atrial injections of 6-keto-PGE$_1$ on mean systemic arterial pressure ($\overline{SAP}$) in fetal goats. Lines are based on least-squares regression analyses of data given in Table I.

Discussion. Results of the present experiments indicate that 6-keto-PGE$_1$ is a powerful dilator of the perinatal pulmonary and systemic circulations. By maintaining pulmonary blood flow at a constant rate, decreases in $\overline{PAP}$ reflect decreases in PVR, or reductions in vascular smooth muscle tone. These findings in anesthetized,

TABLE I. COMPARISON OF ROUTE OF ADMINISTRATION OF BOLUS INJECTIONS OF 6-KETO-PGE$_1$ ON $\overline{SAP}$ IN FETAL GOATS

Pulmonary artery		Left atrium	
Dose (μg/kg)	%Δ$\overline{SAP}$	Dose (μg/kg)	%Δ$\overline{SAP}$
0.074[a]	−2	0.083	−6
0.083	−7	0.089	−3
0.089	−4	0.13	−8
0.13	−3	0.14	−12
0.14	−5	0.15	0
0.15	0	0.25	−9
0.25	−8	0.27	−3
0.27	−8	0.27	−3
0.37[a]	−12	0.27	−4
0.38	−8	0.38	−3
0.42	−14	0.42	−9
0.44	−14	0.44	+1
0.83	−16	0.83	−20
0.89	−18	0.89	−9
1.25	−16	1.25	−15
1.39	−17	1.39	+2
2.50	−30	1.47	−13
2.68	−28	2.68	−25
2.73[a]	−27	3.75	−17
4.41	−23		

[a] Values from 1-min infusions (μg/kg·min).

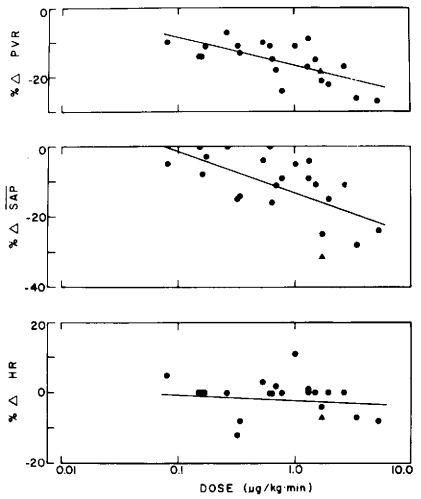

FIG. 4. Effects of intrapulmonary infusions (●) and injection (▲) of 6-keto-PGE$_1$ on pulmonary vascular resistance (PVR), mean systemic arterial pressure ($\overline{SAP}$), and heart rate (HR) in newborn lambs.

open-chest newborn lambs are similar to an earlier study in conscious lambs (13) in which pulmonary blood flow increased with intrapulmonary injections of 6-keto-PGE$_1$. In contrast to Lock *et al.* (13), we show similar responses of the pulmonary and systemic circulations to 6-keto-PGE$_1$ under normoxic conditions (Fig. 4); however, we did not measure cardiac output. Reductions in $\overline{SAP}$ may reflect indirect effects of 6-keto-PGE$_1$ on the heart.

The fetal pulmonary circulation was very sensitive to 6-keto-PGE$_1$. Pulmonary responses of fetal goats were significantly greater than those for PGI$_2$, exhibiting a lower extrapolated threshold dose (0.013 μg/kg and 0.035 μg/kg·min, respectively) (unpublished data). This comparison may not be entirely valid; 6-keto-PGE$_1$ was administered as bolus injections while PGI$_2$ was infused for 1 min. However, three 1-min infusions of 6-keto-PGE$_1$ in fetal goats fit in well with bolus responses (Fig. 2), indicating that method of administration does not appear to be a consideration for the difference in response to PGI$_2$ and 6-keto-PGE$_1$. This difference in response was also true for the systemic circulation, with 6-keto-PGE$_1$ again causing significantly greater reductions in $\overline{SAP}$ (present study, unpublished data). The possibility of converting PGI$_2$ to a more potent metabolite is thus of importance in consideration of using PGI$_2$ in treating human neonates with persistent pulmonary hypertension (14, 15).

We noted significant decreases in HR with increasing doses of 6-keto-PGE$_1$ in fetal goats. Chapple *et al.* (16) have investigated the mechanism for the hypotension and bradycardia induced by PGI$_2$ in chloralose-anesthetized dogs. Atropine attenuated the bradycardia due to infusion of PGI$_2$, while vagotomy reversed the response to PGI$_2$. These findings suggest a reflex vagal pathway mediating prostacyclin-induced bradycardia. Since there are similarities of actions of PGI$_2$ and 6-keto-PGE$_1$ (4, 5), it is possible that the dose-related decreases in HR observed in fetal goats are mediated by the same mechanism. However, this possibility was not tested in the present study.

An interesting observation was the difference in systemic response to 6-keto-PGE$_1$ depending on site of administration (Table I and Fig. 3). The potentiated systemic effect following intrapulmonary injection could be due to: (1) pulmonary conversion of 6-keto-PGE$_1$ to an as yet undiscovered metabolite with even greater biological activity, or (2) 6-keto-PGE$_1$-stimulated release from the lungs of additional vasoactive compounds. The first possibility seems unlikely due to previous reports in adult rats which indicate an absence of pulmonary inactivation (or conversion) of 6-keto-PGE$_1$ (5); however, species or age differences could account for the responses we observed. Additional experiments are needed to determine the mechanism of this observation.

Since prostacyclin is the major product of arachidonic acid metabolism in fetal vessels, including pulmonary artery (17), the cardiovascular actions of PGI$_2$ and its metabolites may play a prominent role in the normal rearrangement of circulation which occurs at birth (10). The report of PGI-like material released by fetal sheep and goat lungs following initiation of respiration (6) lends support to the postulated role of a prostaglandin-mediated action in reduction of pulmonary vascular resistance (10). Results of the present study indicating powerful dilator actions of 6-keto-PGE$_1$ on perinatal pulmonary and systemic circulations could thus implicate this prostacyclin metabolite in the long-term maintenance of low pulmonary vascular resistance.

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