Inhibition of 20-HETE abolishes the myogenic response during NOS antagonism in the ovine fetal pulmonary circulation

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Mechanisms that maintain high pulmonary vascular resistance (PVR) and oppose vasodilatation in the fetal lung are poorly understood. In fetal lambs, increased pulmonary artery pressure evokes a potent vasoconstrictor, suggesting that a myogenic response contributes to high PVR in the fetus. In adult systemic circulations, the arachidonic acid metabolite 20-hydroxyeicosatetraenoic acid (20-HETE) has been shown to modulate the myogenic response, but its role in the fetal lung is unknown. We hypothesized that acute increases in pulmonary artery pressure release 20-HETE, which causes vasoconstriction, or a myogenic response, in the fetal lung. To address this hypothesis, we studied the hemodynamic effects of N-methylsulfonyl-12,12-dibromododec-11-enamide (DDMS), a specific inhibitor of 20-HETE production, on the pulmonary vasoconstriction caused by acute compression of the ductus arteriosus (DA) in chronically prepared fetal sheep. An inflatable vascular occluder around the DA was used to increase pulmonary artery pressure under three study conditions: control, after pretreatment with nitro-L-arginine (L-NA; to inhibit shear-stress vasodilation), and after combined treatment with both L-NA and a specific 20-HETE inhibitor, DDMS. We found that DA compression after L-NA treatment increased PVR by 44 ± 12%. Although intrapulmonary DDMS infusion did not affect basal PVR, DDMS completely abolished the vasoconstrictor response to DA compression in the presence of L-NA (44 ± 12% vs. 2 ± 4% change in PVR, L-NA vs. L-NA + DDMS, P < 0.05). We conclude that 20-HETE mediates the myogenic response in the fetal pulmonary circulation and speculate that pharmacological inhibition of 20-HETE might have a therapeutic role in neonatal conditions characterized by pulmonary hypertension.

HIGH PULMONARY VASCULAR RESISTANCE (PVR) characterizes the fetal lung, resulting in very low pulmonary blood flow compared with the newborn or adult. At birth, in response to the release of several vasoactive agents, the lung undergoes a marked vasodilatation, and pulmonary blood flow increases dramatically (2, 11, 14, 31). The sustained nature of that vasodilator response stands in marked contrast to the fetal pulmonary circulation. Although the fetal lung is capable of vasodilating to a number of stimuli, including increased oxygen tension, alkalosis, and shear stress (1, 3, 12–14, 23), past studies have demonstrated that the vasodilator response is time limited. Despite ongoing exposure to the dilator stimulus, pulmonary blood flow gradually returns to normal, preserving the fetal phenotype marked by high PVR (1, 4).

Our group (1, 28) recently reported that compression of the ductus arteriosus (DA) in the fetus, which diverts blood flow into the lung and simultaneously increases shear stress and pulmonary artery pressure (PAP), causes a biphasic response. Initially, PVR falls in response to increased shear stress. However, over 30–60 min, a vasoconstrictor response to the increased intraluminal pressure predominates, and PVR rises above baseline levels. This vasoconstriction, or myogenic response, illustrates another example whereby the fetal lung limits pulmonary blood flow. Myogenic reactivity has been described in multiple adult circulations and is believed to play a critical role in autoregulation of organ-specific blood flow (15, 26). In studies that employed a variety of in vitro techniques, vessels from adult renal and cerebral circulations demonstrate active constriction as intraluminal pressure is increased within a physiological range (10, 25). Recent studies have implicated the generation of 20-hydroxyeicosatetraenoic acid (20-HETE), a cytochrome P-450 metabolite, in the myogenic response of these as well as other circulations (18, 20, 22, 37). 20-HETE is formed in vascular smooth muscle cells and increases vascular tone by inhibiting large-conductance, calcium-activated potassium channels and inducing depolarization (27, 36).

Unlike its effects in adult renal and cerebral circulations, several recent studies have demonstrated that 20-HETE is a vasodilator of the adult pulmonary circulation. 20-HETE causes concentration-dependent relaxation of isolated human adult pulmonary arteries and of adult rabbit pulmonary artery rings (9, 21, 34). However, in contrast to the fetal lung, myogenic vasoconstriction is not believed to be present in the low-resistance adult pulmonary circulation. Several studies support the possibility that 20-HETE might modulate the myogenic vasoconstriction that we have previously reported in the fetal lung. Fuloria et al. (16) recently reported that 20-HETE is a modest constrictor of isolated pulmonary resistance arteries from newborn piglets. In addition, our laboratory (27) has shown that tetraethylammonium, which inhibits calcium-activated potassium channels in a fashion similar to 20-HETE, increases PVR in intact fetal lambs. However, the potential role of 20-HETE in mediating the myogenic response of the fetal lung has not been studied.

Based on its vasoconstrictor effects in isolated newborn piglet pulmonary arteries and its importance as a mediator of the myogenic response in other circulations, we hypothesized
that production of 20-HETE modulates the myogenic response in the fetal pulmonary circulation. To address this hypothesis, we studied the effects of N-methylsufonyl-12,12-dibromododec-11-enamide (DDMS), a specific inhibitor of 20-HETE, on the myogenic response provoked by brief compression of the DA in chronically prepared fetal lambs. We report that pretreatment with DDMS completely abolishes the fetal pulmonary myogenic response. Our findings suggest that endogenous 20-HETE modulates myogenic vasoconstriction in the fetal lung and support the concept of fundamental differences in pulmonary vasoregulation in the fetal and adult lung.

METHODS

Pregnant, mixed-breed (Columbia-Rambouillet) ewes were used in this study. All procedures and protocols were reviewed and approved by the Animal Care and Use Committee of the University of Colorado Health Sciences Center and followed the Guide for the Care and Use of Laboratory Animals (National Research Council, 1996).

Fetal surgical preparation. Surgery was performed at 126 ± 2 days of gestation (term = 147 days) after ewes had fasted for 24 h. Animals were given intramuscular penicillin G (600,000 U) and gentamicin (80 mg) immediately before surgery, sedated with intravenous ketamine (8 ml) and diazepam (2 ml), and intubated and ventilated with 1–2% isoflurane for the duration of the surgery. Under sterile conditions, a midline abdominal incision was made and the uterus was externalized. A hysterotomy was made, and the left fetal forelimb was exposed. Polyvinyl catheters (20 gauge) were placed in the left axillary artery and vein and advanced into the ascending aorta and superior vena cava, respectively. A left thoracotomy and pericardial incision were made, and the heart and great vessels were exposed. Using a 16-gauge intravenous placement unit (Angiocath; Travenol, Deerfield, IL), we placed a 22-gauge catheter through purse-string sutures into the left pulmonary artery to allow for selective drug infusions. Using a 14-gauge intravenous placement unit (Angiocath; Travenol), we placed 20-gauge catheters into the main pulmonary artery (MPA) and left atrium. The DA was exposed using gentle blunt dissection. During infusion of prostaglandin E1 (0.1 μg/min) into the MPA to maximally dilate the MPA, the wall of the DA was infiltrated with 0.5 ml of formalin to prevent later constriction. An inflatable vascular occluder was placed around the DA. A catheter was placed in the amniotic cavity to serve as a pressure referent. The flow transducer was connected to an externally calibrated flowmeter (Transonic Systems, Ithaca, NY) to measure left pulmonary artery blood flow (QLPA). Hemodynamic variables, which included PAP, aortic pressure, LAP, and QLPA, were measured continuously for the duration of each study protocol. Left lung PVR was calculated using the formula (PAP – LAP)/QLPA. Heart rate was determined from phasic pressure tracings. Arterial blood-gas measurements included pH, Pco2, and Po2 (ABL 500, Radiometer, Copenhagen, Denmark), as well as oxygen saturation and hemoglobin (OSM3 Hemoximeter, Radiometer).

The general study design for protocols 1–3 is shown in Fig. 1. These protocols were designed to study the hypothesis that release of endogenous 20-HETE in the pulmonary circulation modulates the myogenic response provoked by brief DA compression in the presence of nitric oxide (NO) inhibition. Before each protocol, a 20-min period of stable baseline hemodynamics was established. The vascular occluder around the DA was then inflated with saline to increase mean PAP by 20–30% above baseline and was maintained throughout the study period (40 min). The pulmonary vascular response to this maneuver was measured under each of the three study protocols outlined below. Fetal lambs who developed a pressure gradient of >4 mmHg between the pulmonary and systemic circulation that was sustained (e.g., when vascular occluder was deflated) at any time before completion of all study protocols were excluded to avoid the potential confounding effects of evolving pulmonary hypertension on the myogenic response. Protocols were performed on consecutive days in each animal following at least 48 h of recovery from surgery.

Protocol 1: effects of brief DA compression on pulmonary vascular tone. The purpose of this protocol was to establish the pulmonary hemodynamic response to a simultaneous increase in PAP and blood flow caused by DA compression under control conditions. EtOH (50% solution, 4 ml/h for 60 min) was infused into the LPA starting 20 min before DA compression (see Fig. 1) to serve as a vehicle control for DDMS, which was infused in protocol 3.

Study Design

![Study Design](https://via.placeholder.com/150)

**Fig. 1.** Schematic of the study design. Three separate protocols were each completed in chronically catheterized fetal lambs (n = 5) to determine the effects of 20-hydroxyecosatetraenoic acid (20-HETE) inhibition of the myogenic response caused by ductus arteriosus (DA) compression. DDMS, N-methylsufonyl-12,12-dibromododec-11-enamide; EtOH, ethanol; L-NA, nitro-L-arginine.
Protocol 2: effects of t-NA on the pulmonary hemodynamic response to brief DA compression. The purpose of this protocol was to establish the pulmonary hemodynamic response to a brief increase in intraluminal pressure after inhibition of NO-induced vasodilation. Starting 40 min before DA compression, t-NA (30 mg in 1 ml), a specific inhibitor of nitric oxide synthase (NOS), was infused into the LPA (see Fig. 1) over 10 min. In previous studies, we found that this dose of t-NA causes blockade of NO release for ~2 h. As in protocol 1, EtOH (50% solution, 4 ml/h for 60 min) was infused into the LPA to serve as a vehicle control for DDMS.

Protocol 3: effects of combined t-NA and DDMS on the pulmonary hemodynamic response to brief DA compression. The purpose of this protocol was to determine whether a selective antagonist of 20-HETE production would attenuate the pulmonary vasoconstrictor response to increased intraluminal pressure. This protocol is identical to protocol 2, except that DDMS (6 mg/h for 60 min or 2 mg·kg\(^{-1}\)·h\(^{-1}\)) was infused into the LPA starting 20 min before DA compression. This dose of DDMS was selected based on previous in vivo studies, in which DDMS was infused intravenously in rats at 10 mg·kg\(^{-1}\)·h\(^{-1}\) to serve as a vehicle control for DDMS.

Protocol 4: effects of DDMS on basal pulmonary hemodynamics. The purpose of this protocol was to determine whether release of 20-HETE modulates basal pulmonary vascular tone in the fetal lung. After a 20-min period of stable baseline hemodynamics, DDMS (2 mg) or vehicle (50% EtOH) was infused for 30 min at 3 ml/h into the LPA during continuous hemodynamic monitoring.

Statistical analysis. Statistical analysis was performed using the GraphPad Prism version 3.00 for Windows (GraphPad Software, San Diego, CA). Hemodynamic variables over time were compared using repeated-measures ANOVA with Newman-Keuls post hoc testing. The change in PVR at discrete time points between treatments was compared by paired t-test. Significance was set as P < 0.05.

RESULTS

Protocol 1: effects of brief DA compression on pulmonary vascular tone. DA compression increased PAP by 26 ± 1% above baseline. PVR fell progressively for the first 30 min, from 0.49 ± 0.12 mmHg·ml\(^{-1}\)·min at the start of compression to 0.37 ± 0.15 mmHg·ml\(^{-1}\)·min after 30 min (Fig. 2). Arterial blood-gas tensions and LAP did not change during the compression period (Table 1).

Protocol 2: effects of t-NA on the pulmonary hemodynamic response to brief DA compression. Infusion of t-NA increased basal PVR by 80% (0.88 ± 0.19 vs. 0.49 ± 0.12 mmHg·ml\(^{-1}\)·min; P < 0.05). DA compression increased mean PAP by 29 ± 3%, similar to values achieved in the control study. In contrast to the response with protocol 1, pretreatment with t-NA prevented the progressive rise in \(\dot{Q}_{LPA}\), resulting in a 52 ± 10% increase in PVR after 40 min (Fig. 3). PVR peaked after 30 min of compression (1.33 ± 0.24 mmHg·ml\(^{-1}\)·min) and was higher at both 30 and 40 min (P < 0.05) than in the control study (protocol 1). There were no significant changes in arterial blood-gas variables (Table 1) or LAP during the compression period.

Protocol 3: effects of combined t-NA and DDMS on the pulmonary hemodynamic response to brief DA compression. t-NA infusion increased basal PVR to a level similar to that shown with protocol 2 (0.97 ± 0.16 mmHg·ml\(^{-1}\)·min). DA compression increased PAP by 25 ± 5%, similar to the previous protocols. \(\dot{Q}_{LPA}\) did not change during the compression period, and PVR after 40 min of compression was similar to baseline values (0.97 ± 0.16 vs. 0.98 ± 0.18 mmHg·ml\(^{-1}\)·min, baseline vs. 40 min; P = not significant) (Fig. 4). As in the previous protocols, arterial blood-gas variables and LAP did not change during the compression period (Table 1).

Figure 5 shows the response to DA compression at 10-min intervals during the 40-min study period for each of the three protocols. Response is expressed as the percent change in PVR from baseline (at the start of compression) and is compared among the three study protocols. The change in PVR in response to DA compression is different at the 20-, 30-, and 40-min time points (P < 0.05) after treatment with t-NA (protocol 2) when compared with control (protocol 1). Like-
Fig. 3. **Protocol 2**: hemodynamic responses to 40-min DA compression (gray bar) after L-NA administration to inhibit shear stress vasodilation. PAP was increased by 20–31% above baseline, resulting in a marked and progressive increase in PVR. *P < 0.05 compared with 40-min time point.

Fig. 4. **Protocol 3**: effects of DDMS infusion (to block 20-HETE production) on the pulmonary myogenic response caused by 40-min DA compression (gray bar) after L-NA. Although PAP was increased by 18–23%, the progressive rise in PVR demonstrated in **Protocol 2** was completely blocked. *P < 0.05 compared with 40-min time point.

Table 1. *Comparison of arterial blood-gas values at baseline and after 30 min of DA compression for each protocol*

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Baseline</th>
<th>DA Compression</th>
<th>Baseline</th>
<th>DA Compression</th>
<th>Baseline</th>
<th>DA Compression</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.36±0.01</td>
<td>7.33±0.01</td>
<td>7.36±0.00</td>
<td>7.33±0.02</td>
<td>7.36±0.02</td>
<td>7.30±0.05</td>
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<tr>
<td>PaCO₂, Torr</td>
<td>49±2</td>
<td>52±1</td>
<td>49±1</td>
<td>51±2</td>
<td>50±1</td>
<td>56±5</td>
</tr>
<tr>
<td>PaO₂, Torr</td>
<td>21±1</td>
<td>20±1</td>
<td>20±2</td>
<td>21±1</td>
<td>20±1</td>
<td>19±2</td>
</tr>
<tr>
<td>O₂ saturation, %</td>
<td>55±6</td>
<td>50±4</td>
<td>52±5</td>
<td>53±3</td>
<td>53±5</td>
<td>44±6</td>
</tr>
</tbody>
</table>

Values are means ± SE. DA, ductus arteriosus; PaCO₂, arterial PCO₂; PaO₂, arterial PO₂. See text for explanation of protocols. There are no differences in any parameter within or between protocols.
wise, during infusion of DDMS (protocol 3), the change in 
PVR is different from the L-NA response at 30 and 40 min 
(P < 0.05). Time points showed no differences between 
protocols 1 and 3.

Protocol 4: effects of DDMS on basal pulmonary hemody-
namics. Three fetal lambs were treated in this protocol. Data 
are shown in Table 2. Baseline PVR was similar before 
infusion of vehicle (0.61 ± 0.10 mmHg·ml⁻¹·min) and 
DDMS (0.58 ± 0.11 mmHg·ml⁻¹·min). Hemodynamic vari-
dables, including PAP, Q̇LPA, and PVR, did not change during 
the study period after either vehicle or DDMS. PVR was not 
different between groups at any time point.

DISCUSSION

We found that treatment with DDMS, a specific pharma-
cological inhibitor of 20-HETE generation, does not change 
baseline pulmonary vascular tone in the fetus. However, when 
given during compression of the DA, which acutely raises 
PAP, DDMS completely abolishes the resultant myogenic 
response in the fetal lung. Our findings are the first to ascribe 
a physiological role for endogenous 20-HETE in the fetal 
 pulmonary circulation. These results suggest that the effects of 
20-HETE in the fetal pulmonary circulation are similar to those 
in the adult systemic circulation, where it mediates myogenic 
vasoconstriction, and are in marked contrast to the adult lung, 
where it acts as a vasodilator (9, 21, 26).

Table 2. Effects of 20-HETE inhibitor on basal pulmonary hemodynamics

<table>
<thead>
<tr>
<th>Vehicle</th>
<th>DDMS</th>
<th>Infusion</th>
<th>Baseline</th>
<th>Infusion</th>
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</thead>
<tbody>
<tr>
<td>PAP, mmHg</td>
<td>43±2</td>
<td>49±3</td>
<td>44±2</td>
<td>45±3</td>
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<tr>
<td>LAP, mmHg</td>
<td>3±0</td>
<td>2±1</td>
<td>2±1</td>
<td>1±1</td>
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<tr>
<td>Q̇LPA, ml/min</td>
<td>72±16</td>
<td>97±18</td>
<td>82±19</td>
<td>93±25</td>
</tr>
<tr>
<td>PVR, mmHg·ml⁻¹·min</td>
<td>0.61±0.10</td>
<td>0.51±0.06</td>
<td>0.58±0.11</td>
<td>0.51±0.09</td>
</tr>
<tr>
<td>AoP, mmHg</td>
<td>43±3</td>
<td>48±4</td>
<td>42±3</td>
<td>43±2</td>
</tr>
<tr>
<td>HR, beats/min</td>
<td>154±7</td>
<td>178±14</td>
<td>164±5</td>
<td>171±10</td>
</tr>
</tbody>
</table>

Values are means ± SE. N-methylsafoneryl-12, 12-dibromododec-11-enam-
ide (DDMS; 2 mg) or vehicle (50% ethanol) was infused into the left 
 pulmonary artery for 30 min. AoP, aortic pressure; HR, heart rate; LAP, left 
atrial pressure; PAP, pulmonary artery pressure; PVR, pulmonary vascular 
resistance; Q̇LPA, left pulmonary artery blood flow. There were no differences 
between groups.

To study a potential role for 20-HETE in modulating the 
myogenic vasoconstriction in the fetal lung in this study, we 
used the model of DA compression in the chronically instru-
mented fetal lambs. This model takes advantage of the unique 
fetal physiology, whereby high PVR causes a right-to-left 
shunt at the DA and limits pulmonary blood flow. Compression 
of the DA diverts some of this shunt into the lung and causes 
simultaneous activation of two competing physiological re-
 sponses within the fetal pulmonary circulation: shear-induced, 
NO-dependent relaxation and pressure-mediated constriction 
(1). Although the predominant initial response is vasodilation, 
this response is time limited. Over time, with persistent DA 
compression, a constrictor response develops that serves to 
maintain the high pulmonary vascular tone that characterizes 
the fetal pulmonary circulation (1). As we have shown previ-
ously (28), this vasoconstriction, or myogenic response, can be 
unmasked at the outset of DA compression by pharmacological 
blockade of the early shear-mediated vasodilator response with 
L-NA, a specific NOS inhibitor.

These findings contrast with previous reports on the effects 
of 20-HETE in the adult pulmonary circulation. Birks et al. (9) 
have shown that isolated human pulmonary arteries dilate in a 
dose-dependent fashion to 20-HETE and that micromers from 
adult lung have the enzymatic capacity to synthesize 20-HETE 
from arachidonic acid. 20-HETE relaxes preconstricted iso-
lated rabbit and bovine pulmonary artery rings (34, 35). More-
ever, inhibition of endogenous 20-HETE release results in 
increased basal PAP and enhanced vasoconstrictor responses to 
hypoxia in isolated perfused rabbit lungs, suggesting that 
release of 20-HETE serves to maintain the low baseline PVR 
that characterizes the adult lung (35). However, in contrast to 
the fetal lung, we are unaware of data establishing the presence 
of myogenic responsiveness in the adult lung. Our study is the 
first to evaluate the hemodynamic effects of endogenous 20-
HETE on the intact pulmonary circulation of the fetus, and our 
findings are consistent with those of Fuloria et al. (16), who 
have recently demonstrated that exogenous 20-HETE causes a 
vasoconstrictor response in isolated resistance vessels from the 
lungs of neonatal pigs. Our study design, whereby L-NA is 
used to block endogenous NO release, precludes our determin-
ing whether 20-HETE might also have a role in the initial 
vasodilator response provoked by DA compression under nor-
mal physiological conditions. This consideration is particularly
role relevant given the findings in adult bovine pulmonary artery rings, in which relaxation to 20-HETE is dependent on NO release from the vascular endothelium (34). Nonetheless, together with the study by Fuloria et al., our results suggest the possibility that endogenous 20-HETE release serves to maintain the high PVR that characterizes the fetal lung and further clarifies fundamental differences in vasoregulation between the perinatal and adult pulmonary circulation.

Despite its capacity to abolish the myogenic response induced by the hemodynamic stresses of DA compression, we found that DDMS treatment did not lower resting PVR in the fetal lung. These findings suggest that 20-HETE does not regulate basal pulmonary vascular tone in the fetus but is produced and released in response to a hemodynamic stress such as that caused by DA compression. We speculate that the lack of effect on basal tone might be related to interaction between 20-HETE and NO, similar to those established in several adult circulations (6–8, 29, 30). In isolated renal and cerebral arteries from rats, NO binds to the heme moiety in CYP4A enzymes and inhibits formation of 20-HETE (29). The resultant loss of 20-HETE-mediated vasoconstriction complements the cGMP-dependent vasodilation of NO and accounts for the cGMP-independent vasodilator effect of NO (6–8, 30).

NO modulates basal pulmonary vascular tone in the late gestation fetus and mediates a substantial portion of the dramatic and sustained vasodilator response to physiological stimuli at the time of delivery (2, 14). Tonic release of NO in the resting fetal pulmonary circulation might inhibit basal release of 20-HETE and account for the lack of effect of DDMS on baseline hemodynamics. Additional studies to determine whether DDMS reduces the vasoconstrictor effects of NOS antagonists on pulmonary vascular tone and to further delineate the interactions between NO and 20-HETE in the transitional pulmonary circulation would be of considerable interest.

Aside from defining a physiological role for 20-HETE in modulating myogenic responsiveness in the fetal lung, our findings could have important implications for understanding the transitional circulation of the neonate. Under normal circumstances, PVR rapidly falls at birth, allowing the lungs to assume their normal role in gas exchange. However, unlike the fetal condition in which the pulmonary vascular response to dilator stimuli is transient and high PVR is preserved (1, 4), the fall in PVR at birth is profound and sustained. Although changes in the physiological condition of the fetal lung, such as rhythmic distension and increased arterial Po2, undoubtedly contribute to the differences between the fetus and neonate, fundamental changes in the production and response to mediators such as 20-HETE may also underlie these differences. It is conceivable that preservation of a fetal vasoconstrictor response to 20-HETE precludes the normal fall in PVR in pathological disease states characterized by persistent pulmonary hypertension of the newborn. Moreover, impairment of endothelial function, as we mimic with L-NA infusion in this study, may result in loss of NO-mediated inhibition of 20-HETE vasoconstriction in the transitional pulmonary circulation and contribute to the pathogenesis of persistent pulmonary hypertension of the newborn, particularly given the prevailing hypothesis that NO release is reduced in the lungs of infants suffering from this condition.

The in vivo design of our study limits our ability to provide additional data supportive to our primary findings. Our results would be strengthened by the demonstration that 20-HETE induces pulmonary vasconstriction in the intact fetus. Unfortunately, because of the avidity with which 20-HETE binds to proteins such as albumin in vivo, studies in which 20-HETE is infused directly into the bloodstream to provoke a vasoconstrictr response are not practical (26). Similarly, we have no method to measure endogenous 20-HETE production locally within the lung vasculature with this in vivo model, and quantitation of serum 20-HETE levels would serve as a poor surrogate for local levels given intracellular binding to fatty acid binding proteins and intravascular albumin binding (33). It is for these reasons that the preponderance of investigation into the vascular effects of 20-HETE has been based on in vitro models. We also cannot exclude the possibility that DDMS, in the dose that we used, did not cause some alteration of other cytochrome P-450 products, which could have contributed to our findings. However, previous studies suggest that DDMS is a highly selective inhibitor of 20-HETE (32) and the dosing that we chose was lower than published intravenous dosing used by Alonso-Galicia et al. (5, 7). Moreover, any inhibition of eicosatetraenoic acid (EET) generation would be expected to cause vasconstriction, as EETs cause vasodilation of newborn pulmonary resistance vessels (17).

In summary, we found that pharmacological inhibition of endogenous 20-HETE production in the fetal lung does not affect baseline PVR but attenuates the myogenic response provoked by compression of the DA. Our findings raise the possibility that perinatal alterations in 20-HETE signaling may underlie the sustained changes in pulmonary vascular behavior at delivery. Furthermore, persistence of fetal responses to 20-HETE may contribute to abnormalities in pulmonary vascular tone that characterize neonatal disease states complicated by pulmonary hypertension.

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