

Selective endothelial dysfunction in conscious dogs after cardiopulmonary bypass

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Zanaboni, Paul, Paul A. Murray, Brett A. Simon, Kenton Zehr, Kirk Fleischer, Elaine Tseng, and Daniel P. Nyhan. Selective endothelial dysfunction in conscious dogs after cardiopulmonary bypass. *J. Appl. Physiol.* 82(6): 1776–1784, 1997.—It has previously been demonstrated that cardiopulmonary bypass (CPB) causes prolonged pulmonary vascular hyperreactivity (D. P. Nyhan, J. M. Redmond, A. M. Gillinov, K. Nishiwaki, and P. A. Murray. *J. Appl. Physiol.* 77: 1584–1590, 1994). This study investigated the effects of CPB on endothelium-dependent (acetylcholine and bradykinin) and endothelium-independent (sodium nitroprusside) pulmonary vasodilation in conscious dogs. Continuous left pulmonary vascular pressure-flow (LP-Q) plots were generated in conscious dogs before CPB and again in the same animals 3–4 days post-CPB. The dose of U-46619 used to acutely pre-constrict the pulmonary circulation to similar levels pre- and post-CPB was decreased (0.13 ± 0.01 vs. 0.10 ± 0.01 $\text{mg} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$, $P < 0.01$) after CPB. Acetylcholine, bradykinin, and sodium nitroprusside all caused dose-dependent pulmonary vasodilation pre-CPB. The pulmonary vasodilator response to acetylcholine was completely abolished post-CPB. For example, at left pulmonary blood flow of $80 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ acetylcholine ($10 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) resulted in $72 \pm 15\%$ reversal ($P < 0.01$) of U-46619 precontraction pre-CPB but caused no change post-CPB. However, the responses to bradykinin and sodium nitroprusside were unchanged post-CPB. The impaired pulmonary vasodilator response to acetylcholine, but not to bradykinin, suggests a selective endothelial defect post-CPB. The normal response to sodium nitroprusside indicates that cGMP-mediated vasodilation is unchanged post-CPB.

pulmonary circulation; endothelium-dependent vasodilation; endothelium-independent vasodilation; chronic instrumentation; pressure-flow plots

CARDIOPULMONARY BYPASS (CPB) is an essential component of cardiac surgery. However, CPB may result in serious pulmonary complications, including increased pulmonary vascular permeability and pulmonary edema, ventilation-perfusion mismatch, pulmonary hypertension, elevated pulmonary vascular resistance, and resulting in right-heart failure (4, 7, 28). There is increasing evidence that alterations in pulmonary vasoregulation post-CPB may be important in mediating these effects. CPB results in activation of several critical cascade mechanisms, including the complement system, and causes ischemia-reperfusion injury to the lung, both of which may affect pulmonary vasoregulation after CPB. Acute, transient pulmonary vasoconstriction, followed by prolonged pulmonary vas-

cular hyperreactivity post-CPB, has recently been described (26). However, the potential mechanisms responsible for these changes in pulmonary vasoregulation after CPB have not been determined. The purposes of this study were to investigate the effects of CPB on pulmonary vascular endothelium-dependent and -independent vasodilators to determine whether alterations in these mechanisms contribute to pulmonary vascular hyperreactivity post-CPB. CPB causes leukocyte and complement activation, causes ischemia-reperfusion injury to the pulmonary circulation, and results in the formation of free radicals, tumor necrosis factor, and interleukin-1. Each of these mechanisms can result in endothelial dysfunction. Thus we first tested the hypothesis that CPB attenuates the pulmonary vasodilator response to the endothelium-dependent vasodilator acetylcholine, which mediates its effects via nitric oxide (NO). Because CPB is likely to cause generalized (as opposed to selective) endothelial dysfunction, our second hypothesis was that CPB would also attenuate the pulmonary vasodilator response to bradykinin, an endothelium-dependent vasodilator that mediates its effects by a different pattern of endothelial cell mechanisms. Our third hypothesis was that the pulmonary vasodilator response to the endothelium-independent vasodilator sodium nitroprusside would not be altered by CPB.

We utilized chronically instrumented, conscious dogs to investigate the specific effects of CPB on the left pulmonary vascular pressure-flow (LP-Q) relationship. The LP-Q relationship was measured to determine the pulmonary vascular responses to endothelium-dependent and -independent vasodilators before and again 3–4 days after closed-chest CPB. This approach avoids the known effects of general anesthesia on neurohumoral (5, 12, 23, 25) and local (12, 19, 22) mechanisms of pulmonary vasoregulation. Furthermore, measurement of continuous LP-Q plots avoids the inherent limitations of single-point calculations of pulmonary vascular resistance. Finally, and importantly, this model avoids the confounding effects of thoracotomy, direct lung manipulation, cardioplegia, manipulation of the great vessels and activation of reflexes, the effects of local hypothermia, and the direct and indirect effects of aortic cross clamping on pulmonary vasoregulation.

METHODS

All surgical procedures and experimental protocols were approved by the Institutional Animal Care and Use Committee, and the animal facilities are fully accredited by the

American Association for Accreditation in Laboratory Animal Care.

Experimental Preparation

Chronic instrumentation. The chronic instrumentation for this preparation has been described previously (26). Briefly, 14 male mongrel dogs (22–26 kg) were anesthetized with pentobarbital sodium (20 mg/kg iv) and fentanyl citrate (15 µg/kg iv). Anesthesia for the remainder of the surgery was maintained with halothane. Heparin-filled Tygon catheters (1.02-mm ID; Norton) were inserted into the descending thoracic aorta, left and right atria, and the main pulmonary artery after a left thoracotomy. A hydraulic occluder (18 mm; Jones) was placed around the right pulmonary artery, and an electromagnetic flow probe (10 mm; Zepeda) was placed around the left pulmonary artery. The catheters, occluder, and flow probe wires were externalized between the scapulae. A chest tube was placed in the left pleural space and was removed on postoperative *day 1*. Morphine sulfate (2–8 mg im, every 4–6 h) was used for postoperative pain. Cephalozin (1 g) was administered intraoperatively and for 7 days postoperatively via the right atrial catheter. All dogs were allowed to recover for 7–14 days before undergoing the experimental protocols in the conscious state before CPB. After completion of the pre-CPB protocols, the dogs underwent CPB on a separate day, as described below.

CPB. After induction of anesthesia with sodium thiamylal (4 mg/kg iv) and fentanyl citrate (15 µg/kg iv), the dogs were intubated with a cuffed endotracheal tube and anesthesia was maintained with halothane. Systemic arterial pressure (SAP), pulmonary arterial pressure (PAP), electrocardiogram, systemic arterial and mixed venous blood gases, hemoglobin (Hb), and rectal and esophageal temperatures were measured during the entire procedure. After sterile preparation, the right femoral artery was cannulated with a 14-Fr Bard cannula for arterial inflow during CPB. Venous drainage for CPB was obtained via 18- to 20-Fr cannulas placed in the right femoral and right external jugular veins. The tips of the venous cannulas were positioned within the thorax to ensure complete drainage of blood from the right atrium and the absence of pulmonary blood flow during CPB. The pulmonary artery was vented during CPB. PAP and left atrial pressures (LAP) were monitored to ensure the absence of a transpulmonary pressure gradient and pulmonary blood flow during CPB. The dogs were heparinized (300 IU/kg iv) to maintain an activated coagulation time >450 s. Additional heparin was given during CPB as needed to maintain anticoagulation. A roller pump (Sarnes 5000) with a bubble oxygenator (Bentley B10+) was used to perfuse the dog during CPB. The perfusion circuit was initially primed with 1,000 ml of lactated Ringer solution and mannitol (0.5 g/kg). After initiation of CPB, nonpulsatile flow rates were maintained between 60 and 80 ml·kg⁻¹·min⁻¹ and adjusted to maintain SAP between 60 and 80 mmHg. The lungs were not ventilated and were open to the atmosphere during CPB. Anesthesia was maintained with halothane delivered via the oxygenator and supplemental intravenous fentanyl. All animals were cooled to 27°C. In all cases, heart rhythm was sinus bradycardia during the hypothermic CPB period. Total CPB time was 150 min. After rewarming to 37°C, the dogs were separated from CPB and heparin's anticoagulant effect was reversed with protamine sulfate (1.3 mg/100 IU heparin iv). The femoral arterial and femoral and external jugular venous cannulas were removed, and the vessels were ligated. This is well tolerated in dogs due to collateral circulation. Three to four days later, the post-CPB experiments were performed.

Experimental Measurements

All vascular pressures (referenced to atmospheric pressure) were measured with strain-gauge pressure transducers (Gould Statham P23 ID) that were positioned at the level of the right atrium. Left pulmonary blood flow (LQ) was measured with an electromagnetic flowmeter (model SWF-4RD, Zepeda). The flow probe was calibrated before and after CPB. This was accomplished by inserting a Swan-Ganz catheter (7-Fr) percutaneously (topical anesthesia with Xylocaine spray) via the left external jugular vein. All flow was diverted through the left pulmonary artery by inflating the vascular occluder around the right pulmonary artery. LQ was then measured by the thermodilution technique (model 9520A, American Edwards). The flows were then indexed to body weight (ml·kg⁻¹·min⁻¹).

Protocols

All experiments were performed on conscious, unsedated dogs trained to lie quietly on their right side. LP-Q plots were generated by slowly inflating the vascular occluder around the right pulmonary artery while the pulmonary vascular pressure gradient (PAP – LAP) and LQ were measured. This technique does not alter systemic hemodynamics or the zonal condition of the lung (20).

In *protocol 1*, we investigated the effects of CPB on the endothelium-dependent pulmonary vasodilator response to acetylcholine. We tested the hypothesis that the pulmonary vasodilator response to acetylcholine would be attenuated in conscious dogs post-CPB. LP-Q plots were generated in the same conscious dogs ($n = 7$) before and again 3–4 days after CPB, in the no-drug condition, after the pulmonary circulation was acutely precontracted with U-46619, and during the cumulative administration of acetylcholine (0.1, 1.0, and 10.0 µg·kg⁻¹·min⁻¹ iv; Sigma Chemical). In each animal the magnitude of the acute precontraction induced by U-46619 was similar pre- and post-CPB. This required titrating the dose of U-46619 post-CPB because CPB causes pulmonary vascular hyperreactivity (26). This allowed us to assess pulmonary vasodilator responses to various agonists at the same level of vasomotor tone. In each protocol, the pulmonary vasodilator responses to acetylcholine, bradykinin (*protocol 2*), and sodium nitroprusside (*protocol 3*) are expressed as a decrease in active U-46619-induced vasoconstriction. Decreases in PAP – LAP are presented at LQ of 80 ml·kg⁻¹·min⁻¹. In *protocol 2*, we investigated the effects of CPB on the endothelium-dependent pulmonary vasodilator response to bradykinin. We tested the hypothesis that the pulmonary vasodilator response to bradykinin would be attenuated post-CPB. LP-Q plots were generated in the same conscious dogs ($n = 7$) before and again 3–4 days after CPB, in the no-drug condition, after acute precontraction of the pulmonary circulation with U-46619, and during the cumulative administration of bradykinin (1, 2, 5, and 10 µg·kg⁻¹·min⁻¹ iv; Bachem).

In *protocol 3*, we investigated the effects of CPB on the endothelium-independent pulmonary vasodilator response to sodium nitroprusside. We tested the hypothesis that the pulmonary vasodilator response to sodium nitroprusside would not be altered post-CPB. LP-Q plots were generated in the same conscious dogs ($n = 7$) before and again 3–4 days after CPB, in the no-drug condition, after acute precontraction of the pulmonary circulation with U-46619, and during the cumulative administration of sodium nitroprusside (1, 2, 5, and 10 µg·kg⁻¹·min⁻¹ iv; Abbott).

Data Analysis

Phasic and mean vascular pressures and $\dot{L}\dot{Q}$ measured continuously at end expiration were obtained by using passive electronic filters with a 2-s time constant and displayed on an eight-channel strip-chart recorder (model 2800, Gould-Brush). All vascular pressures were referenced to atmosphere before each LP- \dot{Q} determination. Each individual experiment was analyzed by linear regression to calculate the PAP – LAP (or PAP – 0 if LAP < 0) over the empirically measured range of $\dot{L}\dot{Q}$. The composite data are summarized at intervals of 10 ml · kg⁻¹ · min⁻¹ within the empirically measured range of $\dot{L}\dot{Q}$.

Bivariate analysis of variance (ANOVA) in the form of Hotelling's T² (30) was used to assess changes in the LP- \dot{Q} plots in response to U-46619 and the subsequent pulmonary vasodilators before and after CPB. One-way ANOVA was used to assess changes in the LP- \dot{Q} plots in response to cumulative doses of acetylcholine, bradykinin, and sodium nitroprusside both pre- and post-CPB. Two-way ANOVA and Student's *t*-test were used to assess the effect of CPB on the pulmonary vascular responses to acetylcholine, bradykinin, and sodium nitroprusside. One-way ANOVA and Student's *t*-test were used to determine the effect of CPB on blood gases, Hb concentrations, and steady-state hemodynamics pre- and post-CPB. Values are presented as mean ± SE.

RESULTS

Effect of CPB on Pulmonary Vascular Response to U-46619

The pulmonary circulation was acutely precontracted with U-46619 to a similar degree pre- and post-CPB. Consistent with our previous study (26), the dose of U-46619 required to cause a comparable degree of acute pulmonary vasoconstriction was significantly decreased ($P < 0.01$) from 0.13 ± 0.01 to 0.10 ± 0.01 $\mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ post-CPB. CPB decreased ($P < 0.01$) Hb from 11.4 ± 0.5 to 7.9 ± 0.6 g/dl because crystalloid was used to prime both the oxygenator and tubing.

Effect of CPB on Pulmonary Vasodilator Response to Acetylcholine

The pulmonary vascular responses to acetylcholine (10 $\mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ iv) after precontraction with U-46619 in conscious dogs before and 3–4 days after CPB are summarized in Fig. 1. Acetylcholine decreased PAP – LAP at each common level of $\dot{L}\dot{Q}$ from values measured during U-46619 pre-CPB. However, acetylcholine had no effect on PAP – LAP from U-46619 values post-CPB, i.e., acetylcholine caused pulmonary vasodilation pre-CPB but did not cause pulmonary vasodilation post-CPB. Figure 2 summarizes the effect of CPB on the acetylcholine dose-response relationship. Changes in PAP – LAP at $\dot{L}\dot{Q}$ of 80 ml · kg⁻¹ · min⁻¹ in response to acetylcholine are presented. After precontraction with U-46619, acetylcholine caused pulmonary vasodilation at all but the lowest dose pre-CPB. In contrast, the pulmonary vasodilator response to acetylcholine was entirely absent at all doses post-CPB. Thus acetylcholine-induced vasodilation was entirely abolished post-CPB.

Effect of CPB on Pulmonary Vasodilator Response to Bradykinin

The pulmonary vascular responses to bradykinin (10 $\mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ iv) after precontraction with U-46619 in conscious dogs before and 3–4 days after CPB are illustrated in Fig. 3. Bradykinin decreased PAP – LAP at each common level of $\dot{L}\dot{Q}$ from values measured during U-46619 both pre- and post-CPB, i.e., bradykinin caused pulmonary vasodilation both before and after CPB. Figure 4 summarizes the effect of CPB on the bradykinin dose-response relationship. After precontraction with U-46619, bradykinin decreased PAP – LAP at all but the lowest dose both before and after CPB. Moreover, the magnitude of the pulmonary vasodilator response to each dose of bradykinin was similar pre- and post-CPB.

Effect of CPB on Pulmonary Vasodilator Response to Sodium Nitroprusside

The pulmonary vascular responses to sodium nitroprusside (10 $\mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ iv) after precontraction with U-46619 in conscious dogs before and 3–4 days after CPB are summarized in Fig. 5. Sodium nitroprusside decreased PAP – LAP at each common level of $\dot{L}\dot{Q}$ from values measured during U-46619 pre- and post-CPB, i.e., sodium nitroprusside caused pulmonary vasodilation before and after CPB. Figure 6 summarizes the effect of CPB on the sodium nitroprusside dose-response relationship. After precontraction with U-46619, sodium nitroprusside decreased PAP – LAP at the two highest doses before and after CPB. Thus sodium nitroprusside-induced pulmonary vasodilation was not altered by CPB.

Effects of CPB on Steady-State Systemic Arterial and Mixed Venous Blood Gases and Hemodynamics

These effects are summarized in Tables 1 and 2. There were no systematic hemodynamic or blood-gas changes that could account for the differential influence of CPB on the pulmonary vasodilator effects of acetylcholine, bradykinin, and sodium nitroprusside.

DISCUSSION

In this study, the effects of CPB on endothelium-dependent and -independent pulmonary vasodilation in conscious dogs were systematically investigated. Our experimental approach has several unique features that allow our results to be attributed specifically to the effects of CPB. First, general anesthetics are recognized as modulators of vasoregulation, including pulmonary vasoregulation (5, 12, 22, 23, 25). The use of conscious animals avoids this potential confounding influence. Similarly, utilizing chronically instrumented dogs avoids the potential influence of acute surgical trauma on the pulmonary circulation. Finally, LP- \dot{Q} plots allow us to distinguish between vasoactive effects and flow-dependent effects on the pulmonary vasculature. This study is consistent with a previous study (26) in that CPB caused pulmonary vascular hyperreactivity;

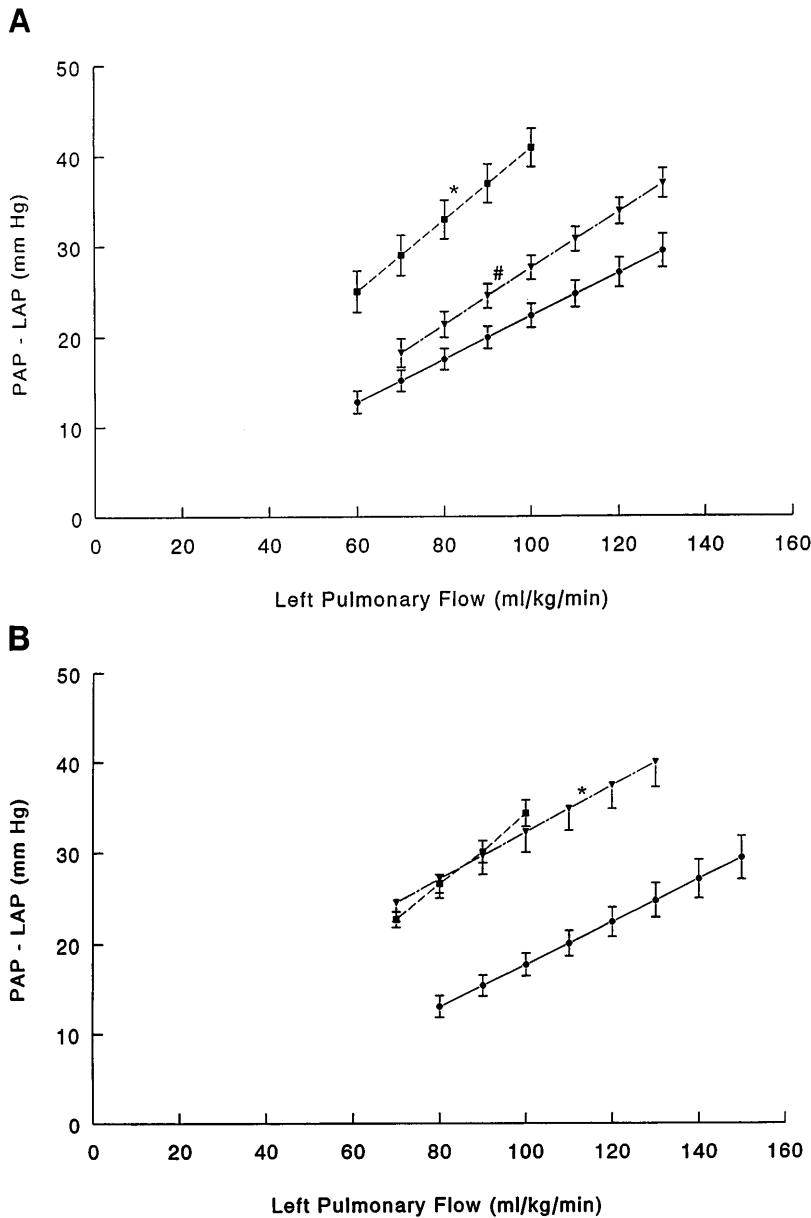


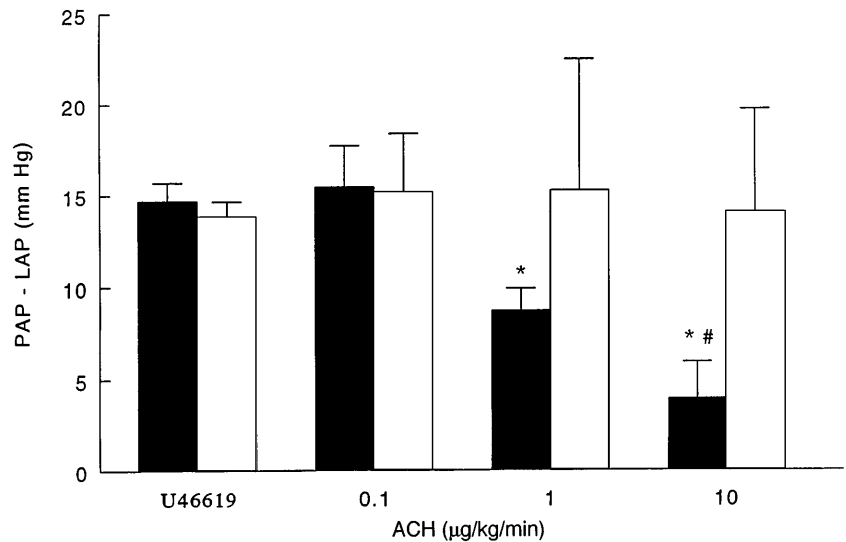
Fig. 1. Composite left pulmonary vascular pressure-flow (LP-Q) plots in 7 conscious dogs after precontraction with U-46619 and during continuous infusion of acetylcholine (ACh; $10 \mu\text{g}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ iv) pre (A)- and 3-4 days post-CPB (B). ●, No drug; ■, U-46619; ▼, ACh. Pulmonary vascular pressure gradient [pulmonary arterial pressure (PAP) - left atrial pressure (LAP)] was increased ($*P < 0.01$) by U-46619 over empirically measured range of flow both pre- and post-CPB. PAP - LAP was decreased ($\#P < 0.01$) during ACh administration pre- but not post-CPB; i.e., ACh-induced pulmonary vasodilation was abolished post-CPB.

i.e., the dose of U-46619 required to cause an equivalent degree of pulmonary vasoconstriction was significantly decreased post-CPB. CPB also caused a marked impairment of the endothelium-dependent response to acetylcholine. Surprisingly, the pulmonary vasodilator response to a second endothelium-dependent agent, bradykinin, was not altered by CPB. Moreover, the response to the endothelium-independent vasodilator sodium nitroprusside was not altered post-CPB. These results indicate that CPB causes selective endothelial dysfunction in response to muscarinic receptor activation. Identifying the mechanism(s) responsible for this defect could have important therapeutic implications for patients undergoing CPB.

It is well recognized that CPB can cause clinically significant pulmonary morbidity (4, 7, 28). Pulmonary morbidity may be manifested as pulmonary edema, bronchoconstriction, impaired compliance, or pulmo-

nary hypertension secondary to elevated pulmonary vascular resistance. CPB-induced alterations in pulmonary vascular resistance may occur in any patient but are most likely to occur in patients with congenital heart disease, mitral valve disease, left-heart failure, interstitial lung disease, and preexisting pulmonary vascular disease. Increased pulmonary vascular resistance and pulmonary vascular hyperreactivity can result from an imbalance of vasoconstrictor and vasodilator mechanisms post-CPB. These vasoregulatory mechanisms may be neural, humoral, or local, including endothelium-dependent mechanisms. CPB activates both leukocytes and the complement cascade and results in the formation of oxygen radicals, tumor necrosis factor, and interleukin-1. Each of these factors may cause endothelial dysfunction and thus contributes to a form of ischemia-reperfusion injury to the lung post-CPB. This pulmonary endothelial dysfunction could

Fig. 2. ACh dose-response relationship measured in 7 conscious dogs pre (solid bars)- and 3-4 days post-CPB (open bars). Increase (Δ) in PAP - LAP representing acute pulmonary vasoconstriction induced by U-46619 and its reversal with cumulative doses of acetylcholine at left pulmonary blood flow (LQ) = $80 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ are shown. ACh decreased ($*P < 0.01$ compared with U-46619 values) Δ PAP - LAP at all but lowest dose pre-CPB. However, ACh did not decrease Δ PAP - LAP post-CPB. Thus magnitude of ACh-induced pulmonary vasodilation post-CPB was reduced ($\#P < 0.01$) compared with pre-CPB.



be responsible for pulmonary vascular hyperreactivity post-CPB. The mechanisms responsible for the abnormal pulmonary vascular response to acetylcholine post-CPB are unknown. There are no apparent systematic hemodynamic or blood-gas changes that would account for this effect. The loss of acetylcholine-induced pulmonary vasodilation post-CPB could be due to 1) changes in endothelial muscarinic receptors; 2) changes in endothelial cell signaling between the muscarinic receptor and NO synthase; 3) decrease in endothelial-dependent relaxing factor-NO (EDRF-NO) synthesis in response to muscarinic-receptor stimulation; 4) concurrent release of contracting factors; 5) concurrent release of superoxide anion that may inactivate EDRF-NO; 6) abnormal vascular smooth muscle response to EDRF-NO; or 7) accentuated vasoconstrictor response mediated by vascular smooth muscle muscarinic receptors. Because the pulmonary vasodilator response to sodium nitroprusside was not impaired post-CPB, dysfunction of vascular smooth muscle vasodilatory mechanisms seems unlikely. The differential responses to acetylcholine and sodium nitroprusside observed in this study post-CPB are consistent with those of Wessel et al. (29) in patients and with those of Shafique et al. (27), who examined the vasodilator response of pulmonary microvessels in sheep subjected to CPB. Kirshbom et al. (17) also demonstrated an impaired pulmonary vasodilator responses to acetylcholine in piglets after CPB with deep hypothermic circulatory arrest. These studies demonstrated an impaired vasodilator response to acetylcholine, and the authors suggested that CPB caused generalized endothelial dysfunction. Morphological correlates of generalized endothelial dysfunction have also been demonstrated (2). In contrast, our results demonstrating a normal vasodilator response to the endothelium-dependent agent bradykinin suggest that CPB-induced pulmonary vascular endothelial injury is more complex and specific and does not necessarily result from generalized endothelial dysfunction.

Acetylcholine causes pulmonary vasoconstriction in several species when preexisting vasomotor tone is low

(15). Moreover, acetylcholine-induced pulmonary vasoconstriction has also been observed in certain canine experimental preparations, even when preexisting vasomotor tone is elevated (9). It has been clearly demonstrated that acetylcholine causes dose-dependent pulmonary vasodilation in normal conscious dogs with elevated vasomotor tone (21). However, acetylcholine causes pulmonary vasoconstriction after left lung autotransplantations in this canine model (21). Thus it is possible that an accentuated vasoconstrictor response to acetylcholine post-CPB could contribute to the attenuated vasodilator response. Acetylcholine-induced vasodilation is mediated primarily by endothelial muscarinic-receptor activation of a pertussis toxin-sensitive G_i protein, resulting in NO synthase stimulation and EDRF-NO release. Endothelial-dependent hyperpolarizing factor (EDHF) may also contribute to acetylcholine-induced pulmonary vasodilation (6). Whereas the contributions of EDRF-NO, vasodilator prostaglandins (16), and EDHF in bradykinin-induced pulmonary vasodilation are well established in other models, this response appears to be primarily mediated by EDRF-NO in conscious dogs, (20) with only a minor role for vasodilator prostaglandins (24). Thus a differential pattern of endothelial mediator release in response to acetylcholine (EDRF-NO, EDHF) and bradykinin (EDRF-NO) could be an explanation for the contrasting responses observed post-CPB. Alternatively, CPB may alter muscarinic (but not bradykinin) receptors or the activity of endothelial G_i protein coupled to muscarinic receptors (but not G_q protein coupled to bradykinin receptors). It is also possible that endothelial cell signaling distal to G_i protein but proximal to NO synthase is selectively altered by CPB. Sodium nitroprusside-induced pulmonary vasodilation was not significantly altered post-CPB, although there was a trend toward an enhanced response. This result would be consistent with impaired EDRF-NO release during the post-CPB period (29).

Endothelial dysfunction post-CPB could result from profound changes in pulmonary blood flow during CPB,

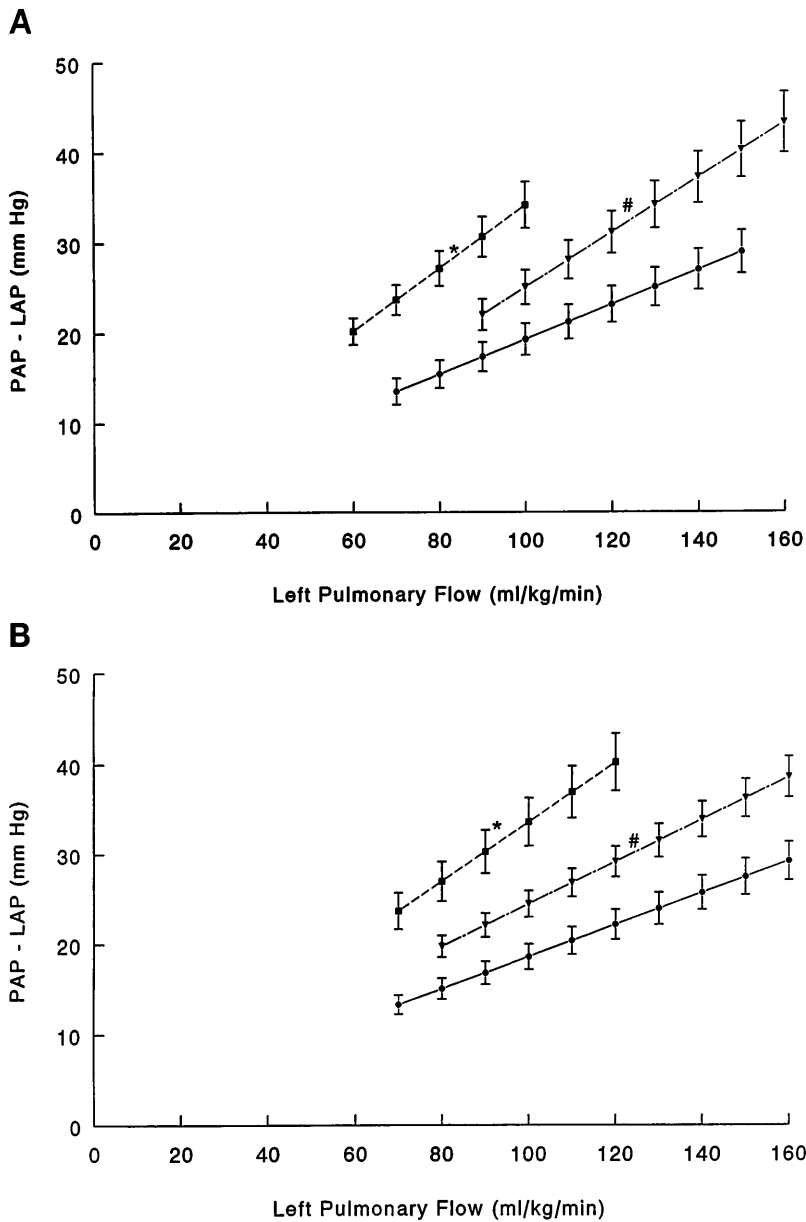


Fig. 3. Composite LP- \dot{Q} plots in 7 conscious dogs after precontraction with U-46619 and during continuous infusion of bradykinin (BK; $10 \mu\text{g}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ iv), pre (A)- and 3-4 days post-CPB (B). \bullet , No drug; \blacksquare , U-46619; \blacktriangledown , BK. PAP - LAP was increased ($*P < 0.01$) by U-46619. PAP - LAP was decreased ($\#P < 0.01$) during BK administration both pre- and post-CPB; i.e., BK caused pulmonary vasodilation both pre- and post-CPB.

Fig. 4. BK dose-response relationship measured in 7 conscious dogs pre (solid bars)- and 3-4 days post-CPB (open bars). Δ PAP - LAP in response to U-46619 and its reversal with BK at $L\dot{Q} = 80 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ are shown. BK decreased ($*P < 0.01$ compared with U-46619 value) Δ PAP - LAP at all but lowest dose both pre- and post-CPB. Magnitude of BK-induced pulmonary vasodilation was not altered by CPB.

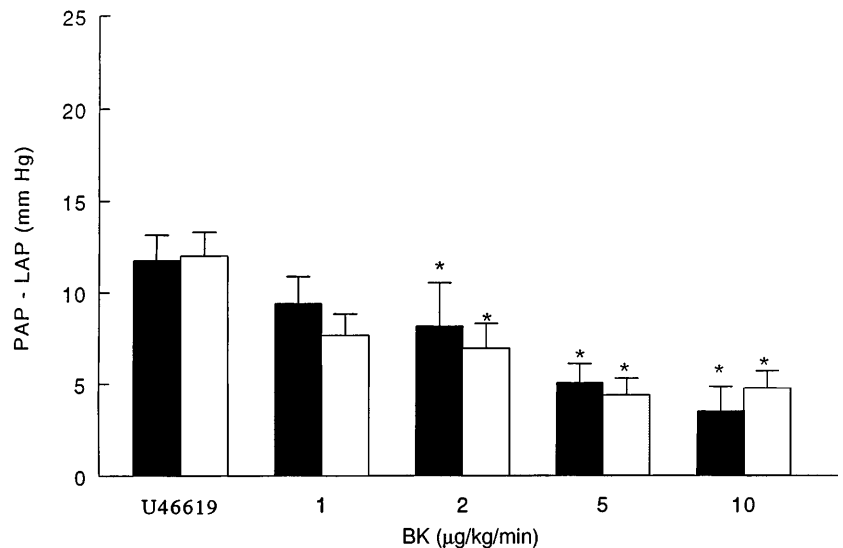


Fig. 5. Composite LP-Q plots in 7 conscious dogs after precontraction with U-46619 and during continuous infusion of sodium nitroprusside (SNP; 10 $\mu\text{g}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ iv), pre (A)- and 3-4 days post-CPB (B). ●, No drug; ■, U-46619; ▼, SNP. PAP - LAP was increased ($*P < 0.01$) by U-46619. PAP - LAP was decreased ($\#P < 0.01$) during SNP administration both pre- and post-CPB, i.e., SNP caused pulmonary vasodilation both pre- and post-CPB.

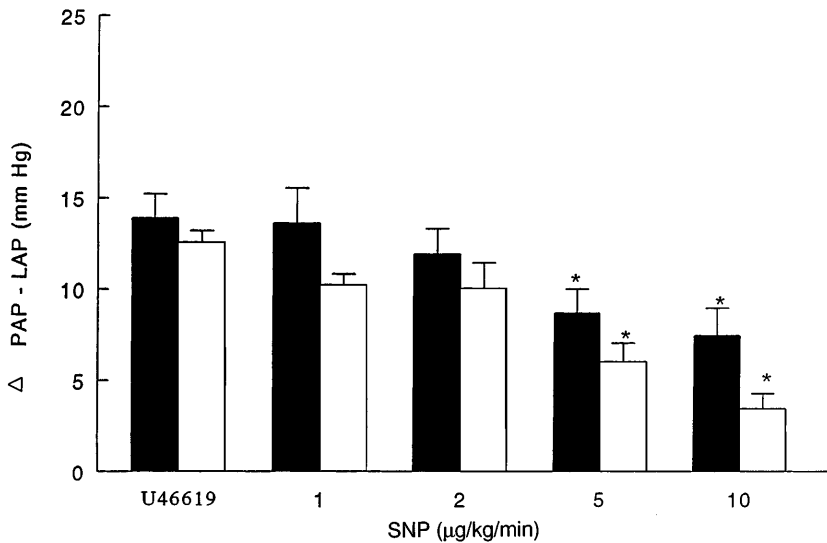
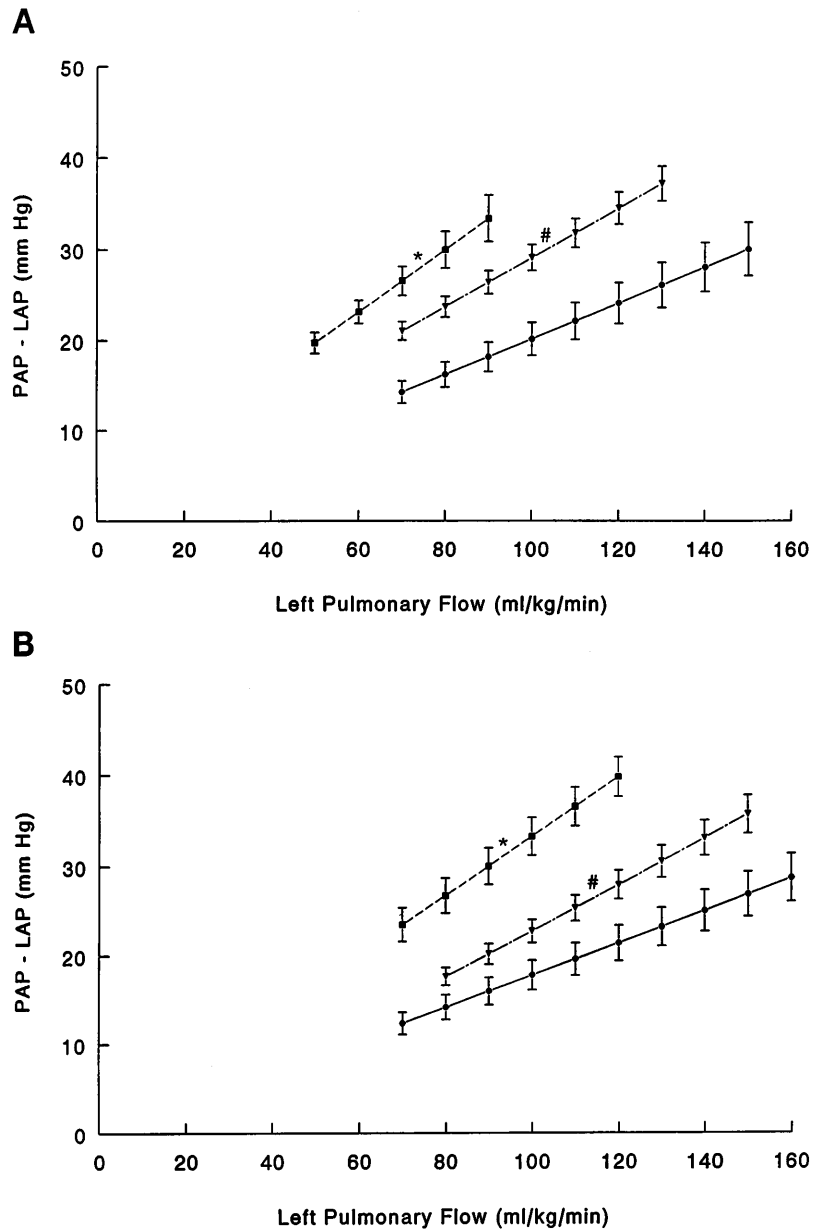


Fig. 6. SNP dose-response relationship measured in 7 conscious dogs pre (closed bars)- and 3-4 days post-CPB (open bars). $\Delta\text{PAP} - \text{LAP}$ representing acute pulmonary vasoconstriction induced with U-46619 and its reversal with SNP at $\text{LQ} = 80 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ are shown. SNP decreased ($*P < 0.01$ compared with U-46619 value) $\text{PAP} - \text{LAP}$ both pre- and post-CPB. Magnitude of SNP-induced pulmonary vasodilation was not altered by CPB.

Table 1. Steady-state systemic arterial and mixed venous blood gases

	Systemic Arterial Blood Gases					
	Pre-CPB			Post-CPB		
	pH	PCO ₂ , Torr	PO ₂ , Torr	pH	PCO ₂ , Torr	PO ₂ , Torr
Control	7.40 ± 0.01	36 ± 2	97 ± 4	7.41 ± 0.01	34 ± 2	99 ± 4
U-46619	7.38 ± 0.01*	36 ± 1	87 ± 4	7.39 ± 0.01	34 ± 2	93 ± 8
U-46619+ACh	7.38 ± 0.02	35 ± 3	77 ± 3	7.38 ± 0.01	34 ± 2	84 ± 5
Control	7.39 ± 0.01	39 ± 1	102 ± 2	7.40 ± 0.01	36 ± 1	104 ± 2
U-46619	7.35 ± 0.01*	41 ± 2	90 ± 4	7.40 ± 0.01	34 ± 1	104 ± 5
U-46619+BK	7.36 ± 0.01	40 ± 2	88 ± 2	7.38 ± 0.01	35 ± 2	97 ± 4
Control	7.40 ± 0.01	37 ± 3	103 ± 3	7.41 ± 0.01	36 ± 1	107 ± 3
U-46619	7.39 ± 0.01	37 ± 2	89 ± 5*	7.38 ± 0.01*	35 ± 1	100 ± 6
U-46619+SNP	7.40 ± 0.01	34 ± 1	87 ± 6	7.40 ± 0.01†	32 ± 2	103 ± 8

	Mixed Venous Blood Gases					
	Pre-CPB			Post-CPB		
	pH	PCO ₂ , Torr	PO ₂ , Torr	pH	PCO ₂ , Torr	PO ₂ , Torr
Control	7.37 ± 0.01	40 ± 2	46 ± 2	7.38 ± 0.01	38 ± 2	45 ± 2
U-46619	7.36 ± 0.02	41 ± 3	44 ± 1	7.36 ± 0.01	40 ± 2	40 ± 2
U-46619+ACh	7.36 ± 0.02	38 ± 3	51 ± 6	7.35 ± 0.02	39 ± 3	42 ± 3
Control	7.37 ± 0.01	42 ± 2	53 ± 1	7.37 ± 0.01	40 ± 1	43 ± 1
U-46619	7.34 ± 0.01*	45 ± 2	47 ± 2	7.37 ± 0.01	38 ± 1	40 ± 2*
U-46619+BK	7.34 ± 0.01	43 ± 2	56 ± 1	7.36 ± 0.01	38 ± 2	48 ± 2†
Control	7.38 ± 0.01	41 ± 1	49 ± 2	7.37 ± 0.01	40 ± 1	45 ± 1
U-46619	7.36 ± 0.01	42 ± 2	43 ± 1*	7.36 ± 0.01*	39 ± 1*	41 ± 1*
U-46619+SNP	7.37 ± 0.01	40 ± 1	45 ± 2	7.36 ± 0.01	37 ± 1†	43 ± 1

Values are means ± SE measured during intravenous administration of acetylcholine (ACh), bradykinin (BK), and sodium nitroprusside (SNP) at 10 µg·kg⁻¹·min⁻¹; n = 7 dogs. CPB, cardiopulmonary bypass. * P < 0.05 vs. control. † P < 0.05 vs. U-46619.

subsequent pulmonary reperfusion and CPB-induced changes in the complement, coagulation, fibrinolytic and kallikrein systems, and leukocytes (11, 28). Pulmonary ischemia-reperfusion, leukocyte activation, complement activation, interleukin-1, and tumor necrosis factor have individually been shown to cause endothelial dysfunction and collectively, as occurs during CPB, could seriously impair normal pulmonary vascular endothelial function. Thus the endothelium may play a pivotal role in mediating the disturbances in pulmonary vasoregulation post-CPB. In general, endothelium-dependent vasodilators tend to inhibit leukocyte adherence to endothelial cells (18), whereas endothelium-dependent vasoconstrictors promote leukocyte adherence (10).

Moreover, there is a balance between endothelial dilator and constrictor systems, with complex interactions and feedback loops (3, 13). Under physiological conditions, endothelial NO inhibits leukocyte adherence to endothelial cells (18). Pulmonary endothelial dysfunction and leukocyte adherence in non-CPB models of pulmonary ischemia-reperfusion suggest a similar pivotal role for the endothelium (1, 8, 14).

In summary, CPB caused pulmonary vascular hyperreactivity 3–4 days post-CPB. The pulmonary vascular response to the endothelium-dependent vasodilator acetylcholine is completely abolished in conscious dogs post-CPB, whereas the response to the endothelium-independent vasodilator sodium nitroprusside is

Table 2. Steady-state hemodynamics

	Pre-CPB			Post-CPB		
	Heart rate, beats/min	SAP, mmHg	LQ, ml·kg ⁻¹ ·min ⁻¹	Heart rate, beats/min	SAP, mmHg	LQ, ml·kg ⁻¹ ·min ⁻¹
Control	96 ± 9	120 ± 13	67 ± 5	115 ± 12	111 ± 13	80 ± 6
U-46619	91 ± 15	121 ± 5	51 ± 3	101 ± 11	124 ± 7	68 ± 5
U-46619+ACh	100 ± 12	117 ± 6	83 ± 4†	137 ± 14†	113 ± 6	72 ± 11
Control	103 ± 9	107 ± 5	70 ± 9	96 ± 4	101 ± 4	71 ± 5
U-46619	82 ± 9*	121 ± 4*	58 ± 5	88 ± 3*	112 ± 4*	67 ± 4
U-46619+BK	92 ± 8	109 ± 4†	92 ± 7†	102 ± 5†	100 ± 3†	87 ± 4†
Control	95 ± 6	107 ± 4	67 ± 5	95 ± 7	106 ± 5	80 ± 6
U-46619	83 ± 7	122 ± 3*	51 ± 4*	83 ± 6	114 ± 6	67 ± 6
U-46619+SNP	123 ± 8†	93 ± 4†	70 ± 7†	132 ± 6†	85 ± 6†	82 ± 7†

Values are means ± SE measured during intravenous infusion of ACh, BK, and SNP at 10 µg·kg⁻¹·min⁻¹; n = 7 dogs. SAP, systemic arterial pressure; LQ, left pulmonary blood flow. * P < 0.05 vs. control. † P < 0.05 vs. U-46619.

normal. In addition, the pulmonary vasodilator response to a second endothelium-dependent vasodilator, bradykinin, is normal post-CPB. These results indicate that endothelial cell regulation of pulmonary vascular tone is selectively altered 3–4 days post-CPB.

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