Biventricular cardiac dysfunction after acute massive pulmonary embolism in the rat

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Sullivan, D. Matthew, John A. Watts, and Jeffrey A. Kline. Biventricular cardiac dysfunction after acute massive pulmonary embolism in the rat. J Appl Physiol 90: 1648–1656, 2001.—Cardiac dysfunction has been documented in vivo after acute massive pulmonary embolism (AMEP). The present study tests whether intrinsic ventricular dysfunction occurs in rat hearts isolated after AMPE. AMPE was induced in spontaneously breathing ketamine-xylazine-anesthetized rats by thrombus infusion until mean arterial blood pressure (MAP) was ∼40% of basal measurement. A hypotensive control group underwent controlled blood withdrawal to produce MAP ∼40% of basal levels. Shams underwent identical surgical and anesthesia preparation but without pulmonary embolization. Hearts were perfused in isovolumetric mode, and simultaneous right ventricular (RV) and left ventricular (LV) pressures were measured. AMPE caused arterial hypotension with hypoxemia (Po2 = 50 ± 14 Torr), acidemia (pH = 7.26 ± 0.11), and high lactate concentration (6.9 ± 1.7 mM). Starling curves from both ventricles demonstrated that AMPE significantly reduced ex vivo systolic contractile function in the RV (P = 0.031) and LV (P = 0.008) compared with both the hypotensive control and sham hearts. AMPE did not alter coronary flow or compliance in either ventricle. Soluble tumor necrosis factor-α decreased in the RV (P = 0.043) and LV (P = 0.005) tissue. These data support the hypothesis that AMPE produces intrinsic biventricular dysfunction and suggest that arterial hypotension is not the principal mechanism of this dysfunction.

The mortality rate of acute massive pulmonary embolism (AMEP) sharply increases in the subset of patients who experience circulatory shock compared with patients without generalized circulatory failure (22). Mortality from AMPE remains high even if fibrinolytic therapy is rapidly administered (5, 24). The prevailing explanation as to how AMPE causes circulatory shock centers on the concept that AMPE blocks the transit of blood to the left ventricle (LV) and thereby reduces cardiac output, causes arterial hypotension, and impairs corporal perfusion. It also has been firmly established from echocardiographic images that AMPE simultaneously causes acute right ventricle (RV) distension and hypokinesis, which may persist for months after pulmonary vascular occlusion is alleviated (16). However, it remains unclear whether selective RV dysfunction is the major limitation to acute recovery of global cardiac function or whether LV dysfunction also contributes to cardiac failure during emergent recanalization after AMPE.

AMPE has been recognized to cause injury to the LV. In 1949, Dack et al. (8) evaluated electrocardiographic and histopathological evidence from a series of patients with pulmonary embolism (PE). They concluded “that when a patient has been lost the fact that the left ventricle is affected deleteriously and often to a greater extent than the right ventricle.” Moreover, in humans with PE, the cardiac index does not correlate to the degree of pulmonary vascular occlusion (19, 28), suggesting that a mechanism other than pulmonary vascular occlusion may limit heart function in AMPE. The first aim of this study was to simultaneously measure intrinsic contractile function in the RV and LV after AMPE. The second aim was to determine whether arterial hypotension alone could reproduce the systolic dysfunction observed after AMPE. We hypothesized that AMPE with hypotension and shock would impair intrinsic function in both ventricles and that arterial hypotension alone would not cause biventricular dysfunction.

METHODS

Experiments were performed according to the National Institutes of Health guidelines on the use of experimental animals. The Institutional Animal Care and Use Committee of Carolinas Medical Center approved all methods. Studies were conducted in male Sprague-Dawley rats weighing between 370 and 522 g.

Thrombus mixture preparation. After anesthesia with an intraperitoneal injection of 100 mg/kg of ketamine and 4.4 mg/kg of xylazine, a donor rat’s neck was shaved and incised, and a tracheostomy was performed. The trachea was cannulated with PE-240 tubing for airway protection and to facilitate spontaneous respiration. The right femoral artery was dissected and cannulated with PE-50 tubing to allow for collection of 10–15 ml of arterial blood. Whole blood was allowed to clot at 23°C in polystyrene tubes. To produce thrombus fragments of appropriate size for the pulmonary embolization protocol, clotted blood was mechanically dis-
ruptured with a Tissue Tearor 985-370 (Dremel, Racine, WI) for 2 s. The homogenate was washed with 0.9% NaCl at 23°C and centrifuged at 2,000 rpm for 5 min consecutively until the supernatant was clear (~5–7 washes). The last supernatant was reserved for use as a vehicle in sham animals. Finally, a slow centrifugation at 500 rpm for 5 min was performed to remove larger clots, and the supernatant was retained as thrombus mixture. Five rats were used for blood donation; all were then euthanized immediately after blood withdrawal and were not used in other experiments.

**Pulmonary embolization protocol.** Experimental animals were anesthetized and cannulated in the above fashion. Both the right carotid artery and external jugular vein were dissected and cannulated with Millar Mikro-Tip catheter transducers (Millar Instruments, Houston, TX). A 2-Fr Millar catheter monitored arterial blood pressure in the carotid artery. A 2-Fr bent Millar catheter was advanced through the external jugular vein to monitor right atrial pressure (42). The left external jugular vein was dissected and cannulated with PE-90 tubing for administration of thrombus mixture. The right external jugular vein was cannulated for arterial blood sampling in the same fashion as the blood donor rats. After cannulation, a 1.0-ml arterial blood sample was obtained for basal control blood chemistry measurements. Arterial blood gas results were obtained using a Novastat Profile Ultra (Nova Biomedical, Waltham, MA). Basal control hemodynamic measurements were then obtained. Data from the Millar transducer were amplified by a Gould amplifier (Gould Electronics, Millersville, MD) prior to either a DASH-10 paper chart recorder (Astro-Med, West Warwick, RI) or an MP-100 computer interface (Biopac Systems, Santa Barbara, CA). For experimental animals, thrombus mixture was administered at 0.5 ml/min using a Harvard 22 syringe pump (Harvard Apparatus, Holliston, MA) until mean arterial blood pressure (MAP) decreased to 40% of the basal control level. Rats were maintained at the desired level of hypotension (40–45 mmHg) with successive infusion of thrombus as needed. Rats were monitored continuously during the embolization, and, if apnea was witnessed, transient ventilation was performed using an RSP 1002 pressure-controlled rodent respirator (pressure-controlled ambient air, 20 cm water pressure, 30 breaths/min) (Kent Scientific, Chantilly, VA). Absorbance values were determined at 570 nm.

**Langendorff preparation.** Hearts were rapidly excised and immediately placed in ice-cold, modified Krebs-Henseleit-bicarbonate buffer made with distilled, deionized water, and contained (in mM) 118 NaCl, 4.7 KCl, 21 NaHCO3, 1.25 CaCl2, 1.2 MgSO4, 1.2 KH2PO4, and 11 D-glucose. Total Na+ concentration was 140 mM and total K+ concentration was 5.6 mM. Buffer was filtered through Millipore (Millipore, Bedford, MA) paper prior to use. It was gassed with 95% O2 and 5% CO2, which produced PO2 = 600–650 Torr and PCO2 = 35–40 Torr. Within 30 s of removal, hearts were perfused with Krebs-Henseleit-bicarbonate buffer (37°C) using the Langendorff technique and 60 mmHg retrograde aortic perfusion pressure. Immediately after perfusion was initiated, the pulmonary artery was incised to allow free ejection from the RV, and a stab incision was made in the LV apex to allow for LV thebesian venous drainage. Initial coronary flow was determined immediately after the incisions. This measurement was performed ~1 min after perfusion began and before placement of balloons in the ventricles. Latex balloons, attached to PE-60 tubing, were then placed via the mitral valve and pulmonary valve into the LV and RV, respectively. Both balloons were simultaneously filled with water until end-diastolic pressure equaled zero in both ventricles. Each balloon was pretested to determine its threshold distension volume (i.e., volume that would raise the static pressure of a balloon over 0 mmHg). A proper-sized balloon was used to ensure that the balloon was not filled over its distension volume during the construction of Starling curves. Balloon pressures were measured with a Gould P23 pressure transducer (Gould Electronics, Millersville, MD). Approximately 15 min after unpaced measurements, a platinum needle was inserted into the LV apex, and hearts were electrically paced (300 beats/min, using 5-ms duration, and voltage set at two times the pacing capture threshold) using a Grass SD9 stimulator (Astro-Med). Contractile function curves were then constructed by simultaneously increasing both balloon volumes to achieve a relative increase in total balloon volume of 50% and recording the ventricular pressures. After completion of data collection, hearts were frozen in liquid nitrogen. Heart tissue was stored at −70°C.

**TNF-α measurements.** RV was removed from LV while frozen in liquid nitrogen. Tissues were then powdered while frozen. Approximately 100 mg of the frozen, powdered ventricle was homogenized in 1.0 ml K2HPO4 buffer (0.1 M, pH 7.4) for 30 s and centrifuged at 5,000 rpm for 5 min. Tumor necrosis factor-α (TNF-α) measurements were performed on the supernatant from heart tissue and on thawed plasma samples obtained in vivo using a Quantikine M murine TNF-α sandwich enzyme linked immunosassay kit (R&D Systems, Minneapolis, MN) according to the manufacturer’s specifications. Spectrophotometric readings were made using a Dynatec MR 5000 microplate reader (Dynatec Laboratories, Chantilly, VA). Absorbance values were determined at 450 nm, and a correction for background interference was made for each sample well at 570 nm.
Statistical analysis. Before statistical tests were made, data were tested for both homogeneity and equal variance (SigmaStat, v.2.03, Jandel, San Rafael, CA). Data are presented as means ± SE. P < 0.05 was considered statistically significant. The in vivo and ex vivo data were compared between the three groups using a one-way ANOVA with Tukey’s post hoc test. Function curves were compared using a two-way repeated measures ANOVA using SigmaStat (version 2.03, Jandel), which uses a general, linear model to calculate P values. An overall group effect P < 0.05 was used to reject the null hypothesis that sham- and hypotensive-controlled heart function was not different from hearts subjected to PE. Slopes of compliance curves were compared using the t-test with Bonferroni correction of α = 0.01 (2) [confidence interval analysis (13)].

RESULTS

Characteristics of thrombus mixture. The thrombus mixture consisted of microemboli with an average, largest dimension of 415.7 ± 75.3 μm, as measured under light microscopy using a reference caliper under ×20 magnification. The thrombi appeared to consist of erythrocytes bound in a protein matrix (Fig. 1). The vehicle used for sham infusion also contained residual protein, but total free hemoglobin content was <0.1 g/dl.

In vivo data. Mean body weight and basal control heart rate, right atrial pressure, and blood pressure were not statistically different between sham (n = 8), hypotensive controls (n = 8), and AMPE rats (n = 9) before the induction of AMPE. Likewise, basal control measurements of pH, PO2, PCO2, and lactate were not different between sham and embolized rats. These data are shown in Table 1. The target MAP (60% decrease from baseline) was achieved after infusion of 0.66 ml ± 0.05/100 g body wt thrombus mixture in AMPE rats. The target blood pressure was maintained for 40 min in six AMPE rats, whereas three developed a pulse pressure <5 mmHg at ~30 min, at which time hearts were removed. Figure 2 compares MAP for AMPE rats with those for hypotensive controls and demonstrates that the depth and duration of MAP in hypotensive

<table>
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<th>Table 1. In vivo data obtained immediately before removal of hearts for ex vivo perfusion for three groups studied</th>
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<td>Body weight, g</td>
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<td>Hemodynamic data</td>
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<td>MAP, mmHg</td>
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<td>PO2, Torr</td>
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<td>Lactate, mmol/l</td>
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Values are means ± SE. AMPE, acute massive pulmonary embolism; BC, basal control; MAP, mean arterial blood pressure; RAP, right atrial pressure. *Significant change at shock vs. basal control by paired t-test; **Significant data at shock compared with ANOVA and Tukey’s post hoc; data with different superscripted letters are significantly different (P < 0.05).
controls were held equal to or slightly lower than those for AMPE rats \( (P = 0.311, \text{unpaired } t\text{-test}). \)

Table 1 shows hemodynamic and blood chemistry data for all three groups measured prior to heart removal. In sham rats, infusion of vehicle produced a transient increase in MAP (mean increase of 20%) followed by the return of MAP to normal within minutes. The MAP then remained stable throughout the remainder of the sham experiment. In AMPE rats, the right atrial pressure and arterial lactate concentration increased significantly, whereas the pH, PO2, and PCO2 decreased significantly compared with sham animals (Table 1). Thus AMPE produced hyperlactacidemia that was not entirely compensated by hyperventilation, as is seen with AMPE in humans (24). Hypotensive control animals also exhibited a significant increase in arterial lactate concentration, but their decrease in PCO2 was substantial enough to hold the arterial pH at a level not different from shams. In contrast to the rats subjected to AMPE, hypotensive controls showed an increase in arterial PO2 compared with that shown in sham rats.

**Mechanical data.** Sham, hypotensive control, and AMPE hearts demonstrated no significant differences in initial balloon volumes (RV and LV end-diastolic pressures equal to zero), voltage required for pacing, coronary flow, or coronary vascular resistance. Table 2 shows that induction of electrical pacing did not significantly alter RV mechanical function in any group. RV function was decreased significantly both before and after pacing in hearts that were isolated after AMPE compared with sham hearts \( (P < 0.001, \text{AMPE vs. sham}) \) and hypotensive control hearts \( (P = 0.010 \text{ vs. hypotensive control}) \). LV systolic pressure was decreased marginally \( (P = 0.075, \text{AMPE vs. sham}) \) before pacing, when the inherent variability in heart rate was greater but was significantly decreased \( (P = 0.031, \text{AMPE vs. sham}) \) after pacing reduced the variability in heart rate. Compared with that shown in hypotensive control hearts, LV function decreased significantly after AMPE \( (P = 0.004) \). Therefore, when ventricular function is compared between the sham, hypotensive control, and AMPE hearts at identical perfusion conditions, it is evident that PE, but not hypotension alone, compromised intrinsic systolic function in both ventricles (Table 2). In terms of relative reduction, hearts subjected to AMPE demonstrated a relative decrease in RV systolic pressure of 37% (sham) and 42% (hypotensive control) and a relative decrease in LV systolic pressure of 24% (sham) and 39% (hypotensive control).

Figure 3 demonstrates systolic function curves for RV and LV, respectively. ANOVA demonstrated an overall significant depression in the RV function curve obtained from AMPE hearts vs. sham hearts \( (P = 0.031, \text{group effect}) \), as well as a significant depression in LV function after AMPE compared with shams \( (P = 0.008, \text{group effect}) \). Hypotensive control hearts were

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**Table 2. Data from ex vivo beating hearts with latex balloons placed in both ventricles for systolic pressure**

<table>
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<tr>
<th></th>
<th>Sham</th>
<th>HC</th>
<th>AMPE</th>
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<tr>
<td>RV balloon volume, μl</td>
<td>60.8 ± 15.2\textsuperscript{a}</td>
<td>84.9 ± 12.0\textsuperscript{a}</td>
<td>58.8 ± 9.8\textsuperscript{a}</td>
</tr>
<tr>
<td>LV balloon volume, μl</td>
<td>116.6 ± 24.5\textsuperscript{a}</td>
<td>114.4 ± 11.9\textsuperscript{a}</td>
<td>81.4 ± 11.1\textsuperscript{a}</td>
</tr>
<tr>
<td>Pacing voltage, V</td>
<td>4.24 ± 0.79\textsuperscript{a}</td>
<td>3.39 ± 0.41\textsuperscript{a}</td>
<td>5.59 ± 2.76\textsuperscript{a}</td>
</tr>
<tr>
<td>Coronary flow, ml/min</td>
<td>15.11 ± 1.29\textsuperscript{a}</td>
<td>16.15 ± 0.71\textsuperscript{a}</td>
<td>17.15 ± 2.63\textsuperscript{a}</td>
</tr>
<tr>
<td>Unpaced</td>
<td>15.65 ± 2.46\textsuperscript{a}</td>
<td>17.60 ± 0.93\textsuperscript{a}</td>
<td>15.51 ± 1.56\textsuperscript{a}</td>
</tr>
<tr>
<td>Coronary vascular resistance, mmHg·ml\textsuperscript{-1}·min\textsuperscript{-1}</td>
<td>4.13 ± 0.33\textsuperscript{a}</td>
<td>3.77 ± 0.17\textsuperscript{a}</td>
<td>3.95 ± 0.61\textsuperscript{a}</td>
</tr>
<tr>
<td>Unpaced</td>
<td>4.84 ± 1.37\textsuperscript{a}</td>
<td>3.48 ± 0.20\textsuperscript{a}</td>
<td>3.71 ± 0.53\textsuperscript{a}</td>
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<tr>
<td>RV systolic pressure, mmHg</td>
<td>40.7 ± 2.6\textsuperscript{a}</td>
<td>44.4 ± 5.1\textsuperscript{a}</td>
<td>25.4 ± 2.0\textsuperscript{b}</td>
</tr>
<tr>
<td>Unpaced</td>
<td>40.2 ± 1.8\textsuperscript{a}</td>
<td>43.6 ± 5.3\textsuperscript{a}</td>
<td>25.3 ± 3.0\textsuperscript{b}</td>
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<tr>
<td>LV systolic pressure, mmHg</td>
<td>86.3 ± 9.6\textsuperscript{a,b}</td>
<td>114.4 ± 9.0\textsuperscript{a}</td>
<td>62.6 ± 7.1\textsuperscript{b}</td>
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<tr>
<td>Unpaced</td>
<td>84.0 ± 5.3\textsuperscript{a}</td>
<td>104.1 ± 9.8\textsuperscript{b}</td>
<td>63.7 ± 6.6\textsuperscript{b}</td>
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Values are means ± SE. HC, hypotensive control. Pacing voltage is twice threshold. Data for systolic pressures were obtained with end-diastolic pressure equal to zero in both chambers and are shown at the native heart rate (unpaced) and after electrical pacing at 5 Hz (paced). RV, right ventricle; LV, left ventricle. Data with different superscripts are significantly different from each other \( (P < 0.05) \).
not significantly different from sham hearts in either the RV \((P = 0.932, \text{group effect})\) or LV \((P = 0.228, \text{group effect})\). Because both RV and LV balloon volumes were not different between groups at the start of each curve, this significant difference in Starling curves suggests the presence of intrinsic myocardial contractile dysfunction after PE. However, there was a tendency toward a lower balloon volume in the AMPE LV, and the data would only permit an 11% power to detect a significant difference in this measurement. The repeated-measures ANOVA test also revealed that there was no significant group × volume interaction \((P = 0.963 \text{ RV}, P = 0.252 \text{ LV})\). This indicates that, for a given amount of added balloon volume, the change in systolic pressure was not significantly different between AMPE and sham rats for either ventricle. Although volume loading did increase the systolic pressure in AMPE ventricles in parallel to sham and hypotensive control ventricles, neither AMPE ventricle-generated systolic pressure value is equal to sham hearts after a 50% increase in biventricular volume. This suggests that AMPE causes intrinsic cardiac dysfunction that cannot be completely reversed by a clinically relevant degree of volume loading.

Static compliance curves were constructed for RV and LV (Fig. 4). The slope of each line equals the compliance of the ventricle. Pairwise comparison of slopes by \(t\)-test with a Bonferroni correction of \(\alpha = 0.01\) was performed. With this test, the power to demonstrate a 30% difference in slope was greater than 80% for all comparisons. Two significant differences were found. First, when the LV after AMPE was compared with the sham and hypotensive control LV, the LV after AMPE was more compliant than either the sham or hypotensive control LV \((P < 0.01\) for each comparison). Second, RV in hypotensive controls was more compliant than RV from either AMPE or shams \((P < 0.01\) for each comparison).

**DISCUSSION**

The present study demonstrates that PE can depress the intrinsic mechanical function of both RV and LV. To our knowledge, this is the first study to examine both RV and LV function after AMPE in the ex vivo perfused heart. We used a spontaneously breathing animal that was subjected to whole blood PE in an effort to reproduce in vivo cardiac stress caused by both respiratory distress and vascular occlusion, while permitting the release of soluble mediators caused by massive PE. We used a sham model in which the rat was exposed to the potential systemic chemical stress induced by the presence of residual protein in the vehicle that was used to deliver the thrombus mixture to AMPE rats. A third group was performed to mimic the effect of arterial hypotension without PE. We then directly measured ventricular systolic function in isolated perfused hearts, which permitted comparison of AMPE and sham hearts without the confounding effect...
of variable ventricular oxygenation, heart rate, loading conditions, or adrenergic reflexes on ventricular function. With this model, we observed that AMPE with shock produced a symmetrical depression in the mechanical systolic contractile function of both ventricles in the presence of adequate coronary flow. However, an equal degree of arterial hypotension from blood withdrawal did not produce systolic dysfunction. These findings indicate that AMPE causes global cardiac depression rather than selective RV depression secondary to RV injury (8). Comparison of AMPE data to data from hypotensive controls indicates that AMPE produces cardiac dysfunction by one or more other mechanisms in addition to arterial hypotension.

Previous experimental and clinical evidence has shown that massive PE causes a sequence of pulmonary vascular occlusion with resultant RV strain, coronary hypotension, and hypoxemia (4, 32). The combination of these insults is commonly thought to reduce RV oxygenation out of proportion to RV oxygen demand (15) and presumed to cause selective RV ischemia and dysfunction (39). As such, clinicians typically report RV kinetic function as a key measurement of heart function during treatment of PE in humans, with relatively little attention given to the LV (30, 31, 33). Our data support the hypothesis that the LV may also play an important role in hemodynamic compromise with severe PE. Biventricular depression suggests that PE produces a diffuse insult, such as myocardial ischemia in vivo.

Previous work in large animals has shown that acute pulmonary arterial hypertension, induced by pulmonary arterial constriction, causes symmetrical reduction in subendocardial blood flow to both ventricles (12, 15). Gold and Bache (15) studied awake, chronically instrumented dogs and found an equal decrease in RV and LV subendocardial blood flow during severe occlusion of the pulmonary artery. They found that in vivo
subendocardial blood flow to both ventricles could be increased above control levels simply by normalizing aortic blood pressure with aortic occlusion (15). This observation suggests that arterial hypotension with massive PE could cause coronary hypoperfusion, which might impair systolic contraction. However, these previous studies did not report indexes of LV or RV contractility. In the present report, the data show that PE significantly impaired systolic function in both ventricles despite the presence of adequate coronary flow ex vivo, but arterial hypotension of equal magnitude did not significantly alter ex vivo systolic function in either ventricle. Hearts subjected to AMPE also showed no significant reduction in compliance (based on data in Fig. 4) and no increase in TNF-α in either ventricle. Although these findings do not suggest the presence of severe myocardial ischemia with necrosis (6, 40), the data do not allow a conclusion regarding whether AMPE caused reversible myocardial ischemia in vivo. Indeed, clinical studies have suggested that PE can cause significant myocardial ischemia, based on the observation of electrocardiographic changes indicative of ischemia (11) and increased serum creatine kinase and troponin levels in patients with large PE (1, 14, 41).

Mechanical damage to myocytes in the RV has been demonstrated in an experimental rat model of PE induced by latex microspheres, suggesting that shear forces play an important role in causing cardiac dysfunction after massive pulmonary vascular occlusion (7). However, RV volume and compliance measurements were normal in AMPE hearts, indicating that shear forces did not grossly distort RV.

Soluble negative inotropic agents such as TNF-α (36), thromboxane A2 (38), and endothelins (9, 35) have been shown to increase after PE. The latter two mediators have been implicated in the development of cardiac failure during circulatory shock after PE. Although we did not measure their concentrations, the absence of increased coronary vascular resistance suggests the absence of a biologically significant effect of either thromboxane A2 or endothelins in the present model. One might expect to find an elevation in myocardial TNF-α following the stress of PE; however, we found lower TNF-α levels in hearts subjected to PE compared with sham. The significance of this is uncertain but probably relates to a suppression of TNF-α production in AMPE hearts. We have previously shown that the procedure of cardiac isolation and perfusion will increase myocardial TNF-α content two- to threefold in unstressed rat hearts compared with TNF-α measured in unstressed hearts freeze-clamped in situ (21). It has been shown that, if the heart is exposed to 5–10 min of hypoxia, then the exposure protects against myocardial injury from subsequent ischemia and reperfusion (10, 17). We therefore speculate that in vivo hypoxia and hypotension during PE caused a preconditioning phenomenon that prevented the increase in TNF-α that is usually caused by the process of isolating the heart and reperfusing it ex vivo. Thus the sequence of events in the experiment may have caused an apparent reduction in myocardial TNF-α levels in AMPE hearts.

The present data support the hypothesis that massive PE causes global myocardial stunning through a combination of insults including acidosis, coronary hypotension, and hypoxemia. Taken as individual insults, it is unlikely that these entities have any significant detrimental effect on ventricular function. In our model, the arterial pH was significantly lower in AMPE rats compared with sham and hypotensive control rats, but the observed arterial pH (7.21) was well above the level that is required for acidosis to impair cardiac contractility in vivo (37) or ex vivo (34). It is also highly unlikely that the level of arterial hypotension produced by AMPE in this model could have caused the degree of contractile dysfunction that we observed in the ex vivo perfused heart, since the hypotensive controls show no significant change in function. Furthermore, previous data from our laboratory (21) also show smaller changes in heart function with a much more severe hemorrhagic shock (MAP of 25 mmHg for 1 h) compared with AMPE hearts. Likewise, although AMPE did produce hypoxemia (Po2 = 49.6 ± 14.0 Torr), the arterial Po2 was probably not depressed to a level that, individually, would have impaired heart function (23, 29). However, the in vivo data in Table 1 show that acidosis, hypotension, and hypoxemia existed simultaneously in our model of AMPE. Prior studies have demonstrated that simultaneous hypoxemia and hypotension will cause greater (18) and more rapid (26) LV dysfunction than either insult individually. In the setting of massive PE, circulatory shock and hypoxemia could cooperate to produce an insult that is equivalent to myocardial stunning, which is usually observed after reversible mild ischemiareperfusion (25).

This study raises the possibility of at least two potential mechanisms of global cardiac dysfunction caused by AMPE. First, the combination of hypoxemia and hypotension may be sufficient to cause the observed biventricular dysfunction. In a separate study, we found in rats that neither a MAP of 30–35 mmHg for 1 h nor 150 s of apnea caused decreased LV function, but 1 h of hypotension followed by 150 s of apnea produced reversible LV dysfunction in the isolated working rat heart (20). In addition, ex vivo perfusion might have led to the generation of reactive oxygen species or peroxynitrite in the AMPE hearts. This potential mechanism will require further study; if free radicals are significant in the present model, then a similar mechanism could occur in vivo with fibrinolytic therapy for AMPE.

A limitation in the present study arises from the finding that LV in AMPE hearts tended to be smaller and have greater compliance than sham LV, but this observation was not found when hypotensive controls were compared with shams. This observation may indicate that AMPE caused the LV chamber to adjust in vivo to the reduced LV filling with simultaneous compression from RV dilation (3), leading to a lower sar-
comere length in the LV at end-diastolic pressure equal to zero. This raises the question of whether the differences in LV contractility that were observed with the Starling curves starting at end-diastolic pressure equal to zero in all groups would have disappeared if curves were performed with LV chamber volume starting at a constant point (normalized for rat body weight) rather than at an equal pressure. In addition, we did not observe contractile dysfunction after controlled hemorrhage, which might appear to contradict the recent work by McDonough et al. (27), which demonstrated that hemorrhage causes intrinsic cardiac contractile dysfunction. However, in that model (using a guinea pig), hypotension was induced to a lower level (35–40 mmHg) and for a longer duration (3 h) than that for the hypertensive controls in the present work.

In conclusion, circulatory shock from AMPE decreases systolic contractile function in both RV and LV. Arterial hypotension is not the sole mechanism responsible for this dysfunction.

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