Thromboxane Mediation of Cardiopulmonary Effects of Embolism

TAKAYOSHI UTSUNOMIYA, MICHAEL M. KRAUSZ, LAWRENCE LEVINE, DAVID SHEPRO, and HERBERT B. HECHTMAN, *Department of Surgery, Brigham and Women's Hospital, Harvard University School of Medicine, Department of Biochemistry, Brandeis University, Biological Science Center, Boston University, Boston, Massachusetts 02115*

ABSTRACT Humoral factors released from platelets during pulmonary embolism may be the cause of several attendant cardiopulmonary abnormalities. This study examines the role of thromboxanes (Tx) after experimental embolism induced with 0.5 g/kg autologous clot in four groups of five dogs: (*a*) untreated embolized controls; (*b*) pretreatment with the Tx synthetase inhibitor, imidazole 25 mg/kg·h i.v., starting 30 min before embolization; (*c*) pretreatment with the cyclooxygenase inhibitor indomethacin, 5 mg/kg, 12 h per os and 1 mg/kg, 1 h i.v. before the experiment; (*d*) treatment with prostacyclin (PGI_2) 100 ηg/kg·min i.v. for 1 h, 1 h after embolization. Within 30 min, embolization led to increases of 6-keto-$PGF_{1\alpha}$, the stable hydrolysis product of PGI_2, from 0.11±0.08 ηg/ml (mean±SD) to 0.33±0.10 ηg/ml ($P < 0.005$) and TxB_2, the stable product of TxA_2, from 0.10±0.04 ηg/ml to 0.38±0.06 ηg/ml ($P < 0.001$). Increases were observed in total dead space (V_D/V_T) from 0.46±0.03 to 0.61±0.08 ($P < 0.025$, physiologic shunting ($\dot{Q}_S/\dot{Q}_T$) from 16±4% to 38±9% ($P < 0.01$), pulmonary vascular resistance (PVR) from 2.27±0.59 mm Hg·min/liter to 9.21±1.90 mm Hg·min/liter ($P < 0.005$) and mean pulmonary arterial pressure from 14±6 mm Hg to 34±1 mm Hg ($P < 0.001$). Cardiac index (CI) fell from 139±11 ml/kg·min to 95±17 ml/kg·min in 4 h ($P < 0.025$). Imidazole pretreatment prevented a rise of TxB_2, but not 6-keto-$PGF_{1\alpha}$; indomethacin blocked both. Both agents maintained V_D/V_T at base line and limited increases in $\dot{Q}_S/\dot{Q}_T$ and PVR. CI was higher after imidazole pretreatment compared with controls ($P < 0.025$). Indomethacin led to intermediate levels of CI. PGI_2 lowered TxB_2 ($P < 0.025$), V_D/V_T ($P < 0.025$), $\dot{Q}_S/\dot{Q}_T$ ($P < 0.025$) and PVR ($P < 0.05$) within 30 min. During PGI_2 infusion, CI was higher than controls. Concentrations of TxB_2 correlated with V_D/V_T, $r = 0.79$ and $\dot{Q}_S/\dot{Q}_T$, $r = 0.69$ ($P < 0.001$). Treatment of three dogs with the imidazole derivative ketoconazole, 10 mg/kg IV, 30 min after 0.75 g/kg autologous clot resulted in a lowering of physiologic dead space, but no other improvement of cardiopulmonary function. These results show that a number of cardiopulmonary abnormalities induced by pulmonary embolism are related directly or indirectly to platelet secretions and that V_D/V_T is closely allied to TxA_2 levels.

INTRODUCTION

Pulmonary embolism produces cardiopulmonary dysfunction that may result directly from mechanical obstruction of the pulmonary vasculature as well as indirectly by humoral factors (1). It is believed that platelet interaction with a clot leads to platelet release reaction, which may be responsible for several of the attendant cardiopulmonary abnormalities such as pulmonary hypertension and increase of venous admixture (2, 3). Platelet aggregation is inhibited by prostacyclin (PGI_2),[1] which is largely produced by the vascular wall (4), whereas thromboxane (Tx) A_2, a potent proaggregator, is produced by platelets (5). The ratio of circulating PGI_2:TxA_2 is likely to be significant in controlling platelet-endothelial interaction (5), and therefore, aggregation and release of vaso- and bronchoactive agents. The present study was designed to

Address all reprint requests to Dr. Hechtman, Brigham and Women's Hospital.

Received for publication 2 October 1981 and in revised form 2 April 1982.

[1] *Abbreviations used in this paper*: CI, cardiac index; CO, cardiac output; 5-HT, 5-hydroxytryptamine; MAP, mean arterial pressure; MPAP, mean pulmonary arterial pressure; PAWP, pulmonary arterial wedge pressure; PGI_2, prostacyclin; PVR, pulmonary vascular resistance; Tx, thromboxane.

evaluate the cardiopulmonary functional abnormalities after embolism, moderated by PGI_2 and TxA_2.

METHODS

20 mongrel dogs of either sex weighing 17–34 kg were anesthetized with pentobarbital sodium 15 mg/kg i.v. and paralyzed with pancuronium bromide, 2 mg i.v. The dogs were intubated and ventilated with room air at a tidal volume of 15 ml/kg and rate of 12 cycles/min. The animals were placed supine and catheters positioned in the right atrium and femoral artery via a groin cutdown. A 7 Fr thermistor-tipped, pulmonary arterial catheter (Instrumentation Laboratory, Lexington, MA) was introduced through the external jugular vein. Strain gauge transducers were used to measure the following pressures: mean arterial (MAP), mean pulmonary arterial (MPAP), and pulmonary arterial wedge (PAWP). Cardiac output (CO) determined by thermodilution was performed in triplicate and divided by body weight to obtain cardiac index (CI). Pulmonary vascular resistance (PVR) measured in millimeters of mercury per minute per liter was obtained from the ratio MPAP-PAWP/CO.

Blood gases, pH, oxygen saturation, and hemoglobin of arterial and mixed venous blood were measured with Clark and Severinghaus electrodes and by spectrophotometry using extinction coefficients specific for dog blood (Instrumentation Laboratory, models 813 and 282). The P_{CO_2} of mixed expired gas was also measured. Physiologic shunting ($\dot{Q}_S/\dot{Q}_T$) was calculated from the Berggren equation (6)

$$\frac{\dot{Q}_S}{\dot{Q}_T} = \frac{C_cO_2 - C_aO_2}{C_cO_2 - C_{\bar{v}}O_2} \times 100, \qquad [1]$$

where C_aO_2 and $C_{\bar{v}}O_2$ are oxygen contents in arterial and mixed venous blood. Capillary oxygen content, C_cO_2, was derived from the alveolar gas equation with the assumption that the respiratory quotient was one. $\dot{Q}_T$ is blood flow exiting the lung with the same oxygen content as mixed venous blood. Dead space, including anatomic and physiologic components, (V_D/V_T) was derived from a modified version of the Bohr equation

$$\frac{V_D}{V_T} = \frac{P_aCO_2 - P_ECO_2}{P_aCO_2}, \qquad [2]$$

where the subscripts "a" and "E" indicate arterial blood and mixed expired gas. Platelets were counted in arterial blood by means of phase microscopy.

Prostaglandin assay. TxB_2 and 6-keto-$PGF_{1\alpha}$, the stable derivatives of TxA_2 and PGI_2, respectively, were measured by radioimmunoassay (7–9). Arterial blood was collected in cooled plastic syringes containing heparin. The blood was immediately centrifuged at 1,500 *g* for 20 min at 4°C. Plasma was separated and stored at −20°C until testing.

Specific rabbit Tx and PG antisera were stored in 0.5-ml vol at −80°C until use when the antiserum was thawed. The Tx antiserum was diluted 1/10,000, and the PG antiserum was diluted 1/4,000 using isogeltris composed of 0.1% gelatin, 0.9% NaCl in 0.1 M tris (hydroxymethyl) amino methane-HCl (Tris-HCl) (Sigma Chemical Co., St. Louis, MO) buffer (pH 7.3). Test plasma (0.2 ml), dilute rabbit Tx or PG antisera (0.2 ml), and [^{3}H]TxB_2 or [^{3}H]6-keto-$PGF_{1\alpha}$ tracer (New England Nuclear, Boston, MA) were incubated for 1 h at 37°C. The added tracer (0.1 ml) had an activity of 6,000–7,000 cpm. A control assay was run where isogeltris (0.2 ml) was substituted for test plasma. A blank assay was also used where isogeltris (0.2 ml), rabbit plasma or serum (0.1 ml, mixed 9 to 1 with 0.1 M ethylene diamine tetraacetic acid) were incubated with Tx or PG tracer (0.1 ml). After 1 h of incubation, the reaction was stopped by the addition of rabbit plasma (0.1 ml) for the TxB_2, or rabbit serum (0.1 ml) for the 6-keto-$PGF_{1\alpha}$ assay. Goat anti-rabbit gamma globulin (0.1 ml) (Arnel Products Co., Brooklyn, NY) was added and precipitation allowed to proceed overnight. Then 0.1 M ethylene diamine tetraacetic acid (0.5 ml) was added. The precipitate was collected by centrifugation at 1,200 *g* for 20 min, dissolved in 0.1 ml Protosol and mixed with 2 ml scintillation fluid (Biofluor, New England Nuclear). The scintillation counts from test plasma, control and blank assays were used to calculate percent inhibition from the formula

$$\% \text{ inhibition} = 1 - \left(\frac{\text{test-blank}}{\text{control-blank}}\right) \times 100. \qquad [3]$$

The acceptable inhibitory range was between 15 and 85%. The lower limit permitted the minimum detectable amount of TxB_2 and 6-keto-$PGF_{1\alpha}$ to be 6 or 10 pg, respectively.

Studies were performed to assess the serologic specificities of the antisera used in the radioimmunoassays. In the TxB_2 system 28 pg TxB_2 inhibited homologous binding by 50%; PGD_2, PGE_2, 13,14-dihydro-15,keto-PGF_2, $PGF_{2\alpha}$, and 6-keto-$PGF_{1\alpha}$ crossreacted <1%. In the 6-keto-$PGF_{1\alpha}$ system, 140 pg 6-keto-$PGF_{1\alpha}$ inhibited homologous binding by 50%; 6-keto-PGE_1 crossreacted 3% while PGD_2, PGE_2, 13,14-dihydro-15-keto-PGE_2, $PGF_{1\alpha}$, $PGF_{2\alpha}$, and TxB_2 crossreacted <1%.

Embolization. Autologous blood was clotted with thrombin 10 U/ml blood. The clot was cut into 3- to 5-mm cubes, washed with saline, and 0.5 g/kg embolized intravenously to the lungs. Previous experience with this preparation using ^{131}I-fibrinogen to label the clot has shown the disappearance rate of ^{131}I activity to range between 3.2 and 5.3%/h during the 4-h observation period (10). Pulmonary angiograms at 4 h demonstrated peripheral emboli in all lobes.

Animals were divided into four groups of five dogs in each: (*a*) untreated embolized controls; (*b*) pretreatment with the thromboxane synthetase inhibitor, imidazole 25 mg/kg·h i.v., starting 30 min before embolization; (*c*) pretreatment with the cyclooxygenase inhibitor, indomethacin, 5 mg/kg per os 12 h before the experiment and 1 mg/kg indomethacin i.v. 1 h before embolization; (*d*) treatment with PGI_2 1 h after embolization; PGI_2 100 ηg/kg·min was infused intravenously for 1 h. The PGI_2 was stabilized in Tris-HCl buffer at a pH of 10. An ice water-jacketed syringe maintained temperature of the infusate at 2 to 3°C. An additional three dogs received a larger mass of clot, 0.75 g/kg. 30 min later an intravenous bolus of the imidazole derivative, ketoconazole, 10 mg/kg was given to observe the effects of reducing TxA_2 concentration in this more extreme setting of cardiopulmonary dysfunction. In unpublished observations of seven dogs this dose of ketoconazole given 1 h after embolization led to a fall in TxB_2 levels to 0.02 ηg/ml within 30 min. All measurements except V_D/V_T were the same. To eliminate the anatomic component and directly estimate physiologic dead space (V_{D_p}/V_T), end tidal (ET) Co_2 concentration was measured by infrared analysis (Instrumentation Laboratory, model 200). After converting Co_2 concentration to end tidal P_{CO_2}, this value was substituted for P_{ECO_2} in Eq. 2 to obtain V_{D_p}/V_T. Our experience in 18 anesthetized dogs has yielded an average base-line value of 7%.

All data in the table and text are presented as the mean±1 SD. The standard error is used in figures. Statistics are based on an analysis of variance, and when differences between groups were found a nonpaired *t* test was used for intergroup

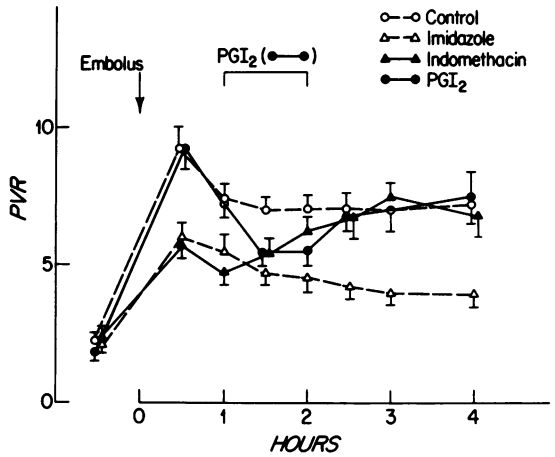

FIGURE 1 The increase in pulmonary vascular resistance 1 h after embolization was significantly less in imidazole and indomethacin-pretreated dogs ($P < 0.005$). PGI_2 lowered PVR, but not significantly.

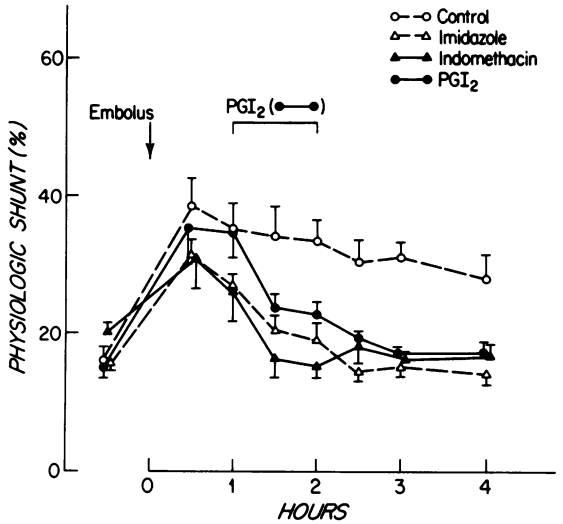

FIGURE 2 Relative to untreated control animals, the rise in $\dot{Q}_S/\dot{Q}_T$ after imidazole or indomethacin was transient. PGI_2 rapidly restored $\dot{Q}_S/\dot{Q}_T$ to base line.

comparisons. Linear regression is used to examine the relationship between variables.

RESULTS

Pulmonary embolization in five control animals caused an increase in MPAP from 14±6 mm Hg to 34±1 mm Hg ($P < 0.001$) within 30 min. By 2.5 h MPAP had stabilized at 29±4 mm Hg. PVR promptly rose from 2.27±0.59 mm Hg·min/liter to 9.21±1.90 mm Hg·min/liter ($P < 0.005$, Fig. 1). After 1 h PVR stabilized.

Arterial oxygen tension (Pa_{O_2}) fell from 85±4 mm Hg to 69±5 mm Hg ($P < 0.01$) by 30 min and then increased to 72±7 mm Hg at 4 h. Initially, at 30 min $\dot{Q}_S/\dot{Q}_T$ rose from 16±4% to 38±9% ($P < 0.01$, Fig. 2) and V_D/V_T rose from 0.46±0.03 to 0.61±0.08 ($P < 0.025$, Fig. 3). Thereafter, $\dot{Q}_S/\dot{Q}_T$ and V_D/V_T decreased, and at 4 h were 28±8% and 0.57±0.04, respectively. There were no significant changes in the base-line MAP of 134 mm Hg and PAWP of 7 mm Hg through the 4-h period of observation (Table I). For the first 1 h, CI was unchanged. Thereafter, it decreased and at 4 h was 60% of base line ($P < 0.025$, Fig. 4).

Platelet counts decreased 30%, 30 min after embolization ($P < 0.025$) and then remained stable (Table I). There was an increase in 6-keto-$PGF_{1\alpha}$ from 0.11±0.08 ηg/ml to 0.33±0.01 ηg/ml ($P < 0.005$, Fig. 5) and TxB_2 from 0.10±0.04 ηg/ml to 0.38±0.06 ηg/ml ($P < 0.001$, Fig. 6). Both cyclooxygenase prostanoids then decreased, and at 4 h were 0.18±0.09 ηg/ml and 0.14±0.06 ηg/ml, respectively.

Imidazole infusion blocked the production of TxA_2. TxB_2 levels did not increase with embolization, while 6-keto-$PGF_{1\alpha}$ rose to values similar to untreated controls. In further contrast to controls, $\dot{Q}_S/\dot{Q}_T$ rose transiently from 16±2% to 31±6% ($P < 0.025$) and then returned to base line (Fig. 2); V_D/V_T remained unchanged through the experiment (Fig. 3); MPAP rose from 20±4 mm Hg to 37±3 mm Hg ($P < 0.001$), and at 3 h had fallen below control values ($P < 0.05$); the rise in PVR was less than controls at all time periods ($P < 0.05$, Fig. 1); and CI was higher than con-

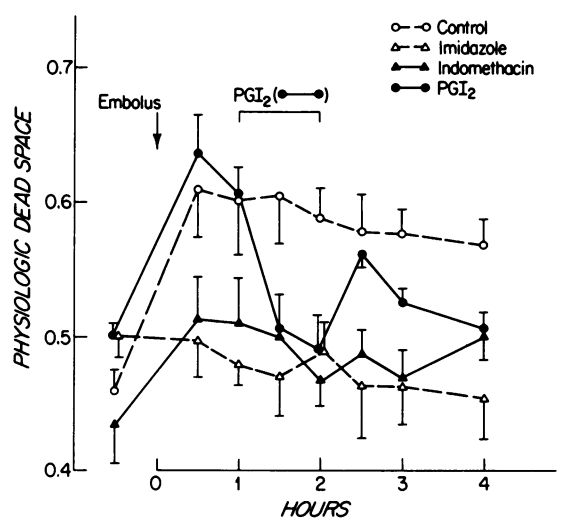

FIGURE 3 Imidazole and indomethacin prevented a rise in V_D/V_T, while PGI_2 rapidly restored this variable to base line. A transient increase in V_D/V_T occurred after cessation of PGI_2.

TABLE I
Cardiovascular and Platelet Response to Emboli

	Base line	Hours 0.5	1.0	1.5	2.0	2.5	3.0	4.0
MAP, *mm Hg*								
Control	134±15	133±9	124±14	127±11	132±14	134±17	134±16	140±16
Imidazole	133±8	125±10	124±12	123±10	126±10	126±11	128±7	127±5
Indomethacin	133±5	132±14	131±13	135±12	137±11	144±12	145±16	137±18
PGI_2	138±25	140±19	129±22	96±19*	97±18*	137±19	141±18	142±18
MPAP, *mm Hg*								
Control	14±6	34±1	31±3	33±5	30±3	29±4	29±3	29±4
Imidazole	20±4	37±3	33±2	29±1	27±2	26±3	25±3*	27±3
Indomethacin	18±4	32±6	26±3*	26±6	26±4	26±6	29±5	29±6
PGI_2	17±2	33±4	30±2	25±2*	24±4*	25±3	24±2*	25±4
PAWP, *mm Hg*								
Control	7±2	9±2	7±2	8±2	7±2	7±2	8±3	8±3
Imidazole	9±2	10±4	10±3	10±3	10±3	10±2	11±1	11±2
Indomethacin	8±2	9±1	9±2	8±2	8±2	8±2	9±2	10±2
PGI_2	8±3	9±3	8±3	8±3	6±4	8±4	8±4	8±5
Platelets, $\times 10^3/mm^3$								
Control	212±65	151 60	151±55	135±42	135±11	131±26	150±45	150±70
Imidazole	199±56	160 40	146±54	142±59	131±49	147±49	154±40	139±36
Indomethacin	186±48	110 27	122±50	115±65	117±66	102±36	103±53	110±64
PGI_2	208±68	145 54	144±40	131±38	132±47	149±51	153±80	149±58

$P < 0.05$ compared with controls.

trols ($P < 0.05$, Fig. 4). Platelets decreased after embolization in a manner similar to controls ($P < 0.05$, Table I).

In indomethacin-pretreated dogs, the base-line value of 6-keto-$PGF_{1\alpha}$ was 0.008±0.001 ηg/ml, which was much lower than that of other groups ($P < 0.001$). After embolization, the concentration did not change (Fig. 4). TxB_2 concentration was also lower than that

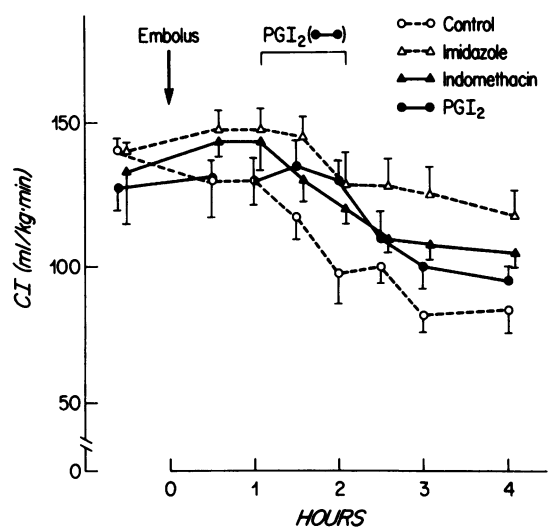

FIGURE 4 Cardiac index gradually decreased after embolization, an event prevented by imidazole and less well by indomethacin. After cessation of PGI_2, the CI approached control values.

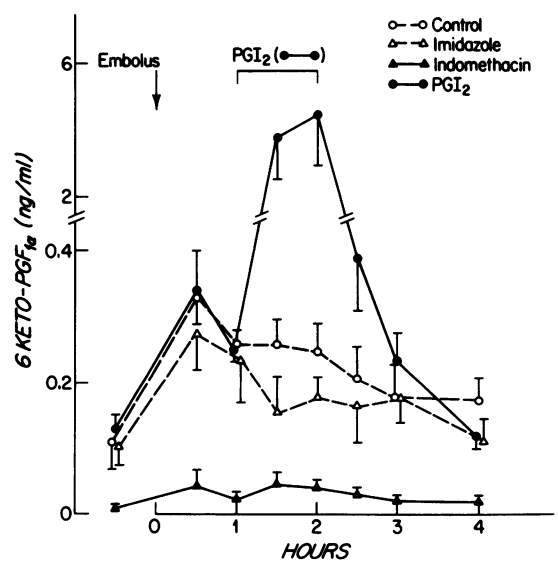

FIGURE 5 Embolization caused a rise in concentrations of 6-keto-$PGF_{1\alpha}$, which was prevented with the cyclooxygenase inhibitor indomethacin, but not the thromboxane synthetase inhibitor imidazole. Infusion of PGI_2 led to a 10-fold increase in concentrations of 6-keto-$PGF_{1\alpha}$.

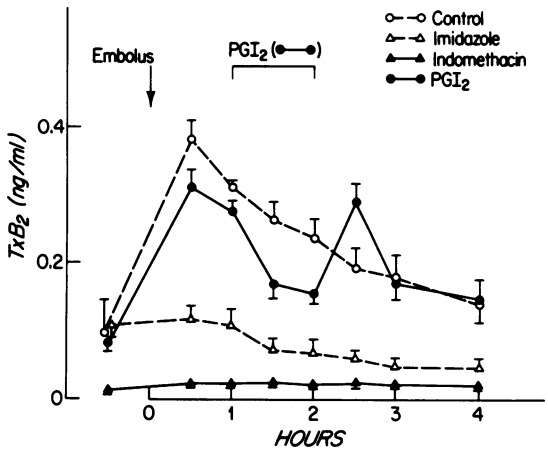

FIGURE 6 Imidazole given just after the first sample was drawn (30 min prior to embolization) and indomethacin given 12 and 1 h before the experiment inhibited the rise in TxB_2, while an infusion of PGI_2 led to its rapid lowering. 30 min after cessation of PGI_2, there was a significant increase in TxB_2 ($P < 0.05$).

of other groups ($P < 0.005$, Fig. 5). After embolization, levels of TxB_2 rose from 0.014±0.003 ηg/ml to 0.023±0.005 ηg/ml ($P < 0.05$), which was still significantly below controls. Embolization led to an insignificant decrease of PaO_2; after 3 h, PaO_2 had fallen to 75±5 mm Hg ($P < 0.025$). $\dot{Q}_S/\dot{Q}_T$ and V_D/V_T were not significantly changed by embolization (Figs. 2, 3). The increase in MPAP was similar to controls. PVR rose; at 1 h it was significantly lower than controls and reached equivalency at 3 h. CI was well maintained within the first 2 h, but then decreased in the next 2 h to a value 79% of base line (Fig. 4). Platelets decreased in the same fashion as controls.

PGI_2 infusion was started 1 h after embolization. The plasma concentration of 6-keto-$PGF_{1\alpha}$ rose to 4.51±3.25 ηg/ml at the end of the infusion period. After cessation of PGI_2, the concentration returned to base line in 30 min. After 30 min, PGI_2 lowered TxB_2 concentration ($P < 0.005$), reducing the value below control animals ($P < 0.025$). However, 30 min after cessation of PGI_2, TxB_2 levels rose above control ($P < 0.05$). PGI_2 infusion reversed the fall in PaO_2 and rise in $\dot{Q}_S/\dot{Q}_T$ ($P < 0.025$, Fig. 2). V_D/V_T also decreased 30 min after the start of PGI_2 infusion ($P < 0.025$) to levels lower than control ($P < 0.05$). However, 30 min after cessation of PGI_2 infusion, V_D/V_T rose ($P < 0.025$) concomitant with the increase of TxB_2. Arterial and MPAP were also lowered during PGI_2 ($P < 0.05$), as was PVR ($P < 0.05$). CI was maintained during the infusion ($P < 0.05$), but 30 min later dropped to values similar to controls. Platelet numbers were not restored by PGI_2 infusion.

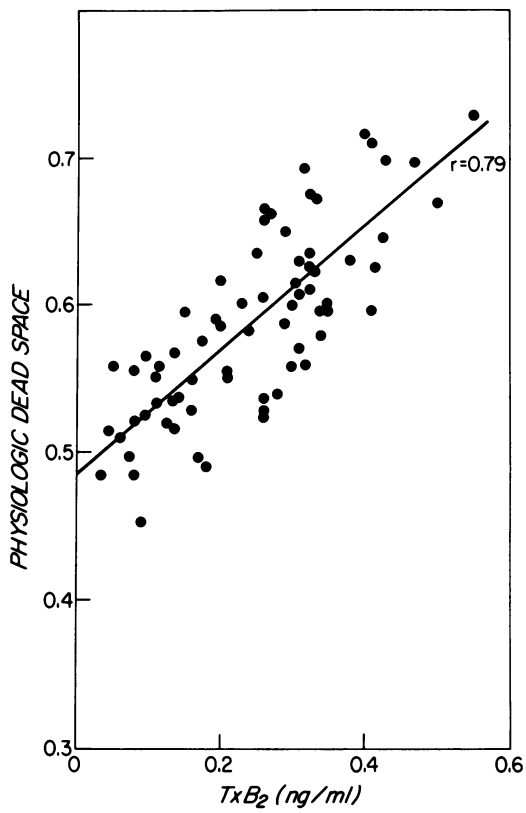

FIGURE 7 A significant relation exists between TxB_2 and V_D/V_T ($P < 0.001$). Data from control and PGI_2 groups are plotted before the PGI_2 infusion was started.

V_D/V_T and TxB_2 concentration in control and PGI_2 groups correlated well ($r = 0.79$, $P < 0.001$, Fig. 7). The $\dot{Q}_S/\dot{Q}_T$ was also related to TxB_2 ($r = 0.69$, $P < 0.001$). 30 min after embolization, the increase of TxB_2 related to a decrease of platelets in control and PGI_2 groups ($r = -0.64$, $P < 0.001$).

Ketoconazole given 30 min after embolization to three dogs, appeared to influence only V_{D_p}/V_T. 2 h after embolus, V_{D_p}/V_T was 22% (Table II), while

TABLE II
Cardiopulmonary Effects of Ketoconazole Given 0.5 h after Embolization (n = 3)

	Hours					
	Base line	0.5	1.0	2.0	3.0	4.0
MPAP, mm Hg	11	49	37	30	31	30
CI, ml/kg·min	127	117	116	107	110	99
PVR, mm Hg·min/liter	1.8	11.7	8.8	7.0	6.8	8.2
$\dot{Q}_S/\dot{Q}_T$, %	11	52	44	30	29	24
V_{D_p}/V_T, %	7	37	27	22	23	30

V_{D_p}/V_T was 34±3% in a series of seven untreated dogs given 0.75 g/kg autologous clot. After 2 h MPAP was 30 mm Hg and PVR was 7.0 mm Hg · min/liter. These values were within one standard deviation of those in untreated dogs where MPAP was 29±5 and PVR was 10.3±4.5 mm Hg · min/liter. Finally, the decline of $\dot{Q}_S/\dot{Q}_T$ from 52 to 30% at 2 h was similar to untreated animals whose level at 2 h was 30±15%.

DISCUSSION

Pulmonary embolism obstructs the pulmonary vasculature and causes a rise in dead space and hypoxemia. These effects are not entirely explicable in terms of mechanical obstruction. A humoral agent(s) released by the clot has been thought to contribute to the cardiopulmonary distress (1, 11, 12). Present investigations show that emboli cause production of the antiaggregator and vasodilator PGI_2, as well as the proaggregator and constrictor, TxA_2. PGI_2 is produced by vessel walls, while TxA_2 is synthesized in platelets and perhaps lung parenchyma (4, 13, 14). Our observation that the concentration of TxB_2, the stable degradation product of TxA_2 is temporally related to the fall in platelet count ($r = -0.64$) suggests that in this setting TxA_2 may originate in platelets that have aggregated. Inhibition of TxA_2 synthesis with imidazole, but not the thrombocytopenia, indicates that aggregation after embolization is independent of TxA_2.

Induced thrombocytopenia or inhibition of platelet function by indomethacin has in the past been shown to modify the effects of microembolism (15, 16). Present data suggest that TxA_2, directly by smooth muscle constriction or indirectly by promoting release of smooth muscle constrictors such as platelet serotonin (5-hydroxytryptamine, 5-HT), is responsible for at least some of the cardiopulmonary abnormalities after experimental pulmonary embolism. Inhibition of TxA_2 synthetase by pretreatment with imidazole and inhibition of cyclooxygenase by pretreatment with indomethacin (Figs. 5, 6) modified the rise in $\dot{Q}_S/\dot{Q}_T$ and prevented any increase in V_D/V_T (Figs. 2, 3). The CI was better maintained along with lower PVR (Figs. 3, 4).

Infusion of PGI_2 led to decreased concentration of TxA_2 and prompt reductions of $\dot{Q}_S/\dot{Q}_T$, V_D/V_T, MPAP, and PVR with maintenance of CI. These salutary cardiopulmonary effects of PGI_2 have been reported and have been related to a direct action on pulmonary arterial smooth muscle causing vasodilation as well as enhanced fibrinolytic activity (10, 17). The very prompt reduction in $\dot{Q}_S/\dot{Q}_T$ and V_D/V_T excludes PGI_2-induced clot lysis as the mechanism of action. A selective PGI_2 effect of vaso- and bronchodilation has been suggested by our finding that a PGE_1 infusion will reduce MPAP to levels lower than PGI_2, but surprisingly, will worsen $\dot{Q}_S/\dot{Q}_T$ and V_D/V_T (18). Other actions of PGI_2 are to antagonize the vaso- and broncho-constriction induced by 5-HT infusion (19) and to lower plasma 5-HT levels after experimental pulmonary embolization (20). Since PGI_2 modifies plasma and platelet 5-HT transport, it is possible that PGI_2 not only acts directly to relax vascular and bronchial smooth muscle, but also acts indirectly in inhibiting the synthesis and/or release of the platelet agents, TxA_2 and 5-HT, which mediate smooth muscle constriction. The present data support the postulate that release of platelet secretions can be modified by PGI_2, and that this event is related to improvement in cardiopulmonary function.

Obstruction of the pulmonary vasculature by emboli leads to ventilation without perfusion. That this rise in V_D/V_T could be prevented or reversed with inhibitors of TxA_2 synthesis was unexpected since this functional abnormality has usually been considered to be a simple mechanical event. It is likely that inhibition of TxA_2 secretion by platelets layering onto the embolus prevents intense constriction distal to the clot. As long as there is some perfusion through these embolized segments, the measured V_D/V_T will appear unchanged (21). This is related to the method of measurement, using the very soluble gas CO_2, which is sensitive only to very high ventilation/perfusion ratios.

The PGI_2 group illustrates the close association of TxA_2 and V_D/V_T. When PGI_2 with $t_{1/2}$ of 2–3 min was stopped, TxA_2 concentrations rapidly rose along with V_D/V_T (Figs. 3, 6). As TxA_2 decreased, so did V_D/V_T. The excellent correlation of TxB_2 with V_D/V_T in control and PGI_2 studies strongly suggests a causal relationship (Fig. 7). The ability of ketoconazole given after embolization to reduce TxB_2 and V_{D_p}/V_T adds further support to this thesis.

The fact that MPAP was little altered and PVR only moderately reduced in the treatment groups after embolization shows that significant reductions in the cross-sectional area of the pulmonary vascular bed still existed despite normal V_D/V_T. It is likely that other vasoactive platelet secretions are involved in the control of PVR. Serotonin is important. Administration of the antagonist cyproheptadine before embolization, using the same preparation as reported in this study prevented any rise in PVR (20).

There are a number of possible causes of the decline in CI after pulmonary embolism. Most noteworthy is right ventricular failure secondary to pulmonary hypertension. In addition, TxA_2 has been reported by our laboratory to be associated with a circulating agent that causes reduction in cardiac output and cardiac contractility (22, 23). The present results do not permit identification of the mechanism whereby PG and TX manipulations maintain CI.

One of the most perplexing events after emboliza-

tion is hypoxemia. Mechanical effects of the clot cannot explain the increase in $\dot{Q}_S/\dot{Q}_T$ that is caused by perfusion of poorly or nonventilated lung segments. Our observation that the concentration of TxB_2 correlates with $\dot{Q}_S/\dot{Q}_T$ ($r = 0.69$) indicates that TxA_2 may directly or indirectly induce bronchoconstriction. An indirect role is suggested by the observation that neither imidazole nor indomethacin completely prevented the increase in $\dot{Q}_S/\dot{Q}_T$ (Fig. 2), and also that ketoconazole was without beneficial effect. Further, PGI_2-infused animals maintained a low $\dot{Q}_S/\dot{Q}_T$ even after cessation of PGI_2, a time when TxA_2 concentrations rose transiently. Other bronchoconstrictors, particularly serotonin, may also participate in producing the hypoxemia of embolization (24). An infusion of 5-HT will lower arterial oxygen tension that can be restored by a simultaneous infusion of PGI_2 (19). In this setting, PGI_2 may be acting both as a bronchodilator as well as a promotor of rapid 5-HT clearance by platelets (20). Pretreatment with imidazole and indomethacin inhibits TxA_2 synthesis (Fig. 6), and although these drugs did not inhibit platelet sequestration and loss, it is possible that they modified platelet 5-HT release accounting for the attenuated rise in $\dot{Q}_S/\dot{Q}_T$. Posttreatment with ketoconazole did not alter $\dot{Q}_S/\dot{Q}_T$. At this time, after platelet sequestration, it is unlikely that the drug influenced 5-HT release.

These results suggest that many of the cardiopulmonary effects of embolism are related to platelet secretions. However, extrapolation of these data to the clinical setting must be done with caution. Our experimental use of fresh clot divided into small fragments permitted embolization of peripheral vessels. This varies from the usual findings in patients where older and larger clot fragments with less surface area for platelet activation are the rule. It is possible that patients suffer quantitatively fewer humorally mediated effects of embolism than those noted in this study.

Cardiopulmonary function has also been shown to be altered in other states where platelets are entrapped in the lungs such as microembolism, sepsis, and abdominal aortic aneurysm repair (1, 11, 14, 15, 25, 26). In addition to platelet synthesis of TxA_2, the observation that lung parenchyma can be stimulated by antigen antibody reaction to release thromboxanes (27) indicates that pulmonary metabolic events may also influence cardiopulmonary function.

ACKNOWLEDGMENTS

This work was supported in part by National Institutes of Health grants GM24891-04 and HL16714-06, the U. S. Army Medical Research and Development Command contract DAMD17-78-C-8026; the Brigham Surgical Group, Inc., and the Trauma Research Foundation.

REFERENCES

1. Halmagy, D. F., B. Starzecki, and G. J. Horner. 1965. Humoral transmission of cardiorespiratory changes in experimental lung embolism. *Circ. Res.* **14:** 546–554.
2. Elkins, R. C., M. Lane, and L. J. Greenfield. 1978. Pulmonary vascular response to experimental embolism and reversal by embolectomy. *J. Surg. Res.* **25:** 135–142.
3. Woolverton, W. C., and A. L. Hyman. 1973. The pulmonary hemodynamic effects of lung thromboemboli in dogs. *Surgery (St. Louis).* **73:** 572–578.
4. Moncada, S., and J. R. Vane. 1977. Arachidonic acid metabolites and the interaction between platelets and blood vessel wall. *N. Engl. J. Med.* **300:** 1142–1147.
5. Moncada, S., and J. L. Amezcua. 1979. Prostacyclin, thromboxane A_2, interactions in haemostatis and thrombosis. *Haemostasis.* **8:** 252.
6. Berggren, S. M. 1942. The oxygen deficit of arterial blood caused by nonventilating parts of the lungs. *Acta Physiol. Scand. (Suppl.).* **11:** 1–92.
7. Levine, L., I. Alam, and J. Langone. 1979. The use of immobilized ligands and ^{125}I protein A for immunoassays of thromboxane B_2, prostaglandin D_2, 13-14-dihydro-prostaglandin E_2, 5-6 dihydro-prostaglandin I_2, 6-keto-prostaglandin $F_{1\alpha}$, 15-hydroxy-9_α,11_α (epoxymethano) prosta-5,13-dienoic acid and 15-hydroxy-11_α,19_α (epoxymethano) prosta-5,13-dienoic acid. *Prostaglandins Med.* **2:** 177–189.
8. Gaudet, R. J., A. Iftekhar, and L. Levine. 1980. Accumulation of cyclooxygenase products of arachidonic acid metabolism in gerbil brain during reperfusion after bilateral common carotid artery occlusion. *J. Neurochem.* **35:** 653–658.
9. Alam, I., K. Ohuchi, and L. Levine. 1979. Determination of cyclo-oxygenase products and prostaglandin metabolites using high pressure liquid chromatography and radioimmunoassay. *Anal. Biochem.* **93:** 339–345.
10. Utsunomiya, T., M. M. Krausz, C. R. Valeri, D. Shepro, and H. B. Hechtman. 1980. Treatment of pulmonary embolism with prostacyclin. *Surgery (St. Louis).* **88:** 25–30.
11. Clay, T. P., and J. M. B. Hughes. 1978. Humoral mediation of bronchoconstriction following pulmonary microembolism in the guinea pig. *J. Physiol. (Lond.).* **275:** 84P.
12. Vargaftig, B. B., J. Jefort, D. Joseph, and F. Fouque. 1979. Mechanism of broncho-constriction and thrombocytopenia induced by collagen in the guinea pig. *Eur. J. Pharmacol.* **58:** 273–284.
13. Greenwald, J. E., L. K. Wong, M. Rao, T. R. Bianchine, and R. V. Panganamal. 1978. A study of three vasodilating agents as selective inhibitors of thromboxane A_2 biosynthesis. *Biochem. Biophys. Res. Commun.* **84:** 1112–1118.
14. Dawson, W., and J. R. Boot. 1976. Release of novel prostaglandins and thromboxanes after immunological challenge of guinea pig lung. *Nature (Lond.).* **262:** 699–702.
15. Gjesdal, V. K. 1976. Lung responses and platelet release reaction following induced intravascular platelet aggregation in the cat. *Scand. J. Clin. Lab. Invest.* **36:** 641–648.
16. Vaage, J., and A. Hauge. 1977. Small airway constriction and closure after induced intravascular platelet aggregation. *Acta Physiol. Scand.* **100:** 221–230.
17. Hechtman, H. B., T. Utsunomiya, A. M. Vegas, G. A. Grindlinger, G. A. McLoughlin, M. M. Krausz, and D. Shepro. Prostaglandin medication of pulmonary fibri-

nolytic activity. *In* Tissue Hormones In The Acutely Ill. Raven Press. In press.
18. Utsunomiya, T., M. M. Krausz, C. R. Valeri, L. Levine, D. Shepro, and H. B. Hechtman. 1981. Treatment of pulmonary embolism with positive end-expiratory pressure and prostaglandin E_1. *Surg. Gynecol. Obstet.* **153:** 161–168.
19. Spannhake, E. W., J. L. Levin, B. T. Mellion, C. A. Gruetter, A. L. Hyman, and P. J. Kadowitz. 1980. Reversal of 5-HT induced bronchoconstriction by PGI_2: distribution of central and peripheral actions. *J. Appl. Physiol.* **49:** 521–527.
20. Utsunomiya, T., M. M. Krausz, D. Shepro, and H. B. Hechtman. 1981. Prostacyclin control of plasma and platelet 5-hydroxytryptamine in normal and embolized animals. *Am. J. Physiol.* **241:** H766–H771.
21. Hechtman, H. B., M. H. Reid, B. C. Dorn, and R. D. Weisel. 1973. Indicator diluation measurements of lung volumes and alveolar air exchange during breathing. *J. Clin. Invest.* **52:** 1215–1229.
22. Utsunomiya, T., M. M. Krausz, B. Dunham, J. A. Mannick, P. A. Allen, D. Shepro, and H. B. Hechtman. 1981. Maintenance of cardiodynamics with aspirin during abdominal aortic aneurysmectomy. *Ann. Surg.* **194:** 602–608.
23. Dunham, B. M., G. A. Grindlinger, T. Utsunomiya, M. M. Krausz, H. B. Hechtman, and D. Shepro. 1981. Role of prostaglandins in positive end-expiratory pressure induced negative inotropism. *Am. J. Physiol.* **241:** H783–H788.
24. Rosoff, C. B., E. W. Salzman, and V. Gurewich. 1971. Reduction of platelet serotonin and the response to pulmonary emboli. *Surgery (St. Louis).* **70:** 12–19.
25. Hechtman, H. B., E. A. Lonergan, and D. Shepro. 1978. Platelet and leukocyte lung interactions in patients with respiratory failure. *Surgery (St. Louis).* **83:** 155–163.
26. Hechtman, H. B., E. A. Lonergan, H. P. B. Staunton, R. C. Dennis, and D. Shepro. 1978. Pulmonary entrapment of platelets during acute respiratory failure. *Surgery (St. Louis).* **83:** 277–283.
27. Schulman, E. S., H. H. Newball, L. M. Demers, F. A. Fitzpatrick, and N. F. Adkinson. 1981. Anaphylactic release of thromboxane A_2, prostaglandin D_2, and prostacyclin from human lung parenchyma. *Am. Rev. Respir. Dis.* **124:** 402–406.