Experimental Physiology

Reflex responses from the main pulmonary artery and bifurcation in anaesthetised dogs

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This study was undertaken to determine the reflex cardiovascular and respiratory responses to discrete stimulation of pulmonary arterial baroreceptors using a preparation in which secondary modulation of responses from other reflexes was prevented. Dogs were anaesthetised with α-chloralose, artificially ventilated, the chests widely opened and a cardiopulmonary bypass established. The main pulmonary arterial trunk, bifurcation and extrapulmonary arteries as far as the first lobar arteries on each side were vascularly isolated and perfused through the left pulmonary artery and drained via the right artery through a Starling resistance which controlled pulmonary arterial pressure. Pressures distending systemic baroreceptors and reflexogenic regions in the heart were controlled. Reflex vascular responses were assessed from changes in perfusion pressures to a vascularly isolated hind limb and to the remainder of the subdiaphragmatic systemic circulation, both of which were perfused at constant flows. Respiratory responses were assessed from recordings of efferent phrenic nerve activity. Increases in pulmonary arterial pressure consistently evoked increases in both perfusion pressures and in phrenic nerve activity. Both vascular and respiratory responses were obtained when pulmonary arterial pressure was increased to above about 30 mmHg. Responses increased at higher levels of pulmonary arterial pressures. In 13 dogs increases in pulmonary arterial pressure to 45 mmHg increased systemic perfusion pressure by 24 ± 7 mmHg (mean ± S.E.M.) from 162 ± 11 mmHg. Setting carotid sinus pressure at different levels did not influence the vascular response to changes in pulmonary arterial pressure. The presence of a negative intrathoracic pressure of −20 mmHg resulted in larger vascular responses being obtained at lower levels of pulmonary arterial pressure. This indicates that the reflex may be more effective in the intact closed-chest animal. These results demonstrate that stimulation of pulmonary arterial baroreceptors evokes a pressor reflex and augments respiratory drive. This reflex is likely to be elicited in circumstances where pulmonary arterial pressure increases and the negative excursions of intrathoracic pressure become greater. They are likely, therefore, to be involved in the cardio-respiratory response to exercise as well as in pathological states such as pulmonary hypertension or restrictive or obstructive lung disease. *Experimental Physiology* (2000) **85.4**, 411–420.

The main pulmonary artery and bifurcation are richly innervated with vagal unmyelinated fibres and to a lesser extent with myelinated fibres (Coleridge *et al*. 1961, 1973). Bianconi & Green (1959) described a 'massive' baroreceptor discharge arising from the pulmonary artery in cats during distal occlusion. However, although histological and electrophysiological evidence for the existence of functional baroreceptors within this region is convincing, the reflex cardiovascular responses to physiological stimulation of these receptors is less clear.

Due to the complexity of applying discrete stimuli to the pulmonary artery and bifurcation, some investigators have distended this region by inflation of a balloon and this has resulted in variable responses of blood pressure and heart rate (Lewin *et al*. 1961; Osorio & Russek, 1962; Laks *et al*. 1975; Crisp *et al*. 1988). It is difficult, however, to relate the responses to balloon distension with those to more normal physiological stimuli.

A more satisfactory method for investigating responses from this region has been to isolate the region vascularly and to distend it with blood at different pressures. This technique has been employed by various investigators but the vascular responses to this have also been inconsistent. Distensions with pressures less than about 80 mmHg have been reported to result in either no change or a fall in arterial pressure (Coleridge & Kidd, 1963) or a small increase (Ledsome & Kan, 1977). Distensions with pressures in excess of 80 mmHg usually cause increases in arterial pressure (Lewin *et al*. 1961; Coleridge & Kidd, 1963; Ledsome & Kan, 1977).

There has been no adequate explanation for the divergent responses of previous investigators and this and the very high pressures previously used implies that we do not yet know what the physiological role of these receptors is likely to be (Daly, 1986). One factor common to the earlier experiments was that no attempts were made to ensure that changes in the stimuli to other reflexogenic areas could not have modified the

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measured response. Regions which could be affected include the arterial baroreceptors and various cardiac receptors. Another consideration is that the earlier work only reported changes in arterial blood pressure, and, without control of blood flow, these may not necessarily reflect vascular resistance changes.

The aim of this study, therefore, was to reinvestigate reflex cardiovascular and respiratory responses during blood distension with various pressures of the main pulmonary trunk, bifurcation and extrapulmonary parts of the left and right arteries, using a preparation in which the stimuli to other major reflexogenic areas, particularly arterial baroreceptors and cardiac receptors, were carefully controlled. We investigated whether setting the level of stimulus to the carotid sinus baroreceptors had any influence on the responses to changes in pulmonary arterial pressure. In a few experiments we also created a closed-chest preparation to determine whether the presence of a negative intrathoracic pressure influenced the pulmonary artery reflexes.

METHODS

Dogs of either sex, weighing between 14.5 and 21.5 kg were premedicated with pentobarbitone sodium (6 mg kg^{-1}; Rhône Mérieux Ltd, Dublin, Ireland) and anaesthetised with an intravenous 1 % solution of α-chloralose (100 mg kg^{-1}; Vickers Laboratories Ltd, Leeds, UK) dissolved in saline. Anaesthesia was maintained by a continuous I.V. infusion of α-chloralose (0.5–1.0 mg kg^{-1} min^{-1}). Before major surgery, morphine sulphate (1 mg kg^{-1}; Evans Medical Ltd, Leatherhead, UK) was slowly infused I.V. The depth of anaesthesia was assessed from the stability of blood pressure and heart rate, the absence of a response to toe pinch and only very small reflex movements to a loud auditory stimulation.

The trachea was cannulated and the animal was artificially ventilated with O_2-enriched air by a Starling 'Ideal' pump, initially set at 17 ml kg^{-1} and 18 strokes min^{-1}. The carotid sinuses were vascularly isolated by ligating all branches arising from the bifurcation of the common carotid artery, except the lingual artery, whilst leaving innervation intact. The sternum was split along the midline and the left side of the chest was divided between the 4th and 5th ribs. When the pleural cavity was opened a positive end-expiratory pressure was maintained at 3 cmH_2O.

The descending aorta was mobilised by tying and dividing the 2nd to 7th pairs of intercostal arteries. A loose thread was passed around the left pulmonary artery just proximal to the first lobar arteries which also had loose snares passed round them. A loose snare was also passed round the upper right lung lobe, and a portion of the right pulmonary artery proximal to this lobe was freed from connective tissue for later cannulation. Extreme care was taken to avoid damage to nerves running over the lung roots. The pericardium was opened to expose the left and right atria and right ventricle for subsequent cannulation.

Prior to cannulation of the blood vessels and attachment of the perfusion circuit the animal was given heparin I.V. (500 i.u. kg^{-1}). The perfusion circuit (Fig. 1*A*) was partly filled with a heparinised mixture of equal parts of mammalian Ringer solution (g l^{-1}: NaCl, 6.9; KCl, 0.35; $CaCl_2$, 0.28; $MgSO_4$, 0.14; $NaHCO_3$, 2.09; KH_2PO_4, 0.16; glucose, 1.0), dextran in dextrose solution (50 g l^{-1} dextran, molecular weight 181 000) and blood cells obtained from a previous experiment and washed. This extracorpeal circuit was attached to the animal in the following sequence. A cannula was inserted into the central end of the thoracic aorta, initially to convey blood into the pressurised main reservoir and, after establishing the cardiopulmonary bypass, to control pressure to the aortic arch and the coronary and cephalic circulations. Another cannula was inserted into the distal end of the thoracic aorta at the level of the diaphragm through which blood was pumped to perfuse the subdiaphragmatic circulation at constant flow. Cannulae (7 mm i.d.) were inserted into the left and right atria via their appendages and into the right ventricle via a stab wound and secured by means of a purse string suture. Another cannula (10 mm i.d.) drained the inferior vena cava. All four cannulae drained blood into an open reservoir from which it was pumped through a Monolyth Membrane Oxygenator (Sorin Biomedica Cardio, Saluggia, Italy) to the main reservoir for distribution to the various parts of the circuit. Both common carotid arteries were cannulated and perfused with blood at constant pressure and the regions drained via cannulae inserted into the lingual arteries into the open reservoir. In some animals in addition to constant flow perfusion of the subdiaphragmatic circulation, a hind limb was also vascularly isolated and perfused at constant flow by a method previously described (Challenger *et al*. 1987).

A cannula (4 mm i.d.) was inserted centrally into the right pulmonary artery immediately before its first branch. The right upper lung lobe was then firmly tied at its root. A similar cannula was then inserted into the left pulmonary artery, again just before its first division and the upper left lobar arteries were ligated. This created a pouch of the main pulmonary trunk and the extra pulmonary parts of the right and left arteries, bordered by the pulmonary valve (see Fig. 1*B*). This pouch was perfused with blood from the inferior vena cava through the right pulmonary artery, and was drained from the left pulmonary artery cannula via a Starling resistor which controlled pulmonary artery pressure. Perfusion pressure generated by the pump was pulsatile with a pulse amplitude of 10–20 mmHg.

In five animals efferent nerve discharge was recorded from the left phrenic nerve in the thorax. The nerve was cut distal to the recording site and the whole nerve was placed in a small plastic tray filled with warm (37 °C) paraffin oil. A binocular microscope was used to assist removal of the nerve sheath, exposing the nerve trunk from which efferent activity was recorded using bipolar silver electrodes. The electrode output was amplified and filtered (Neurolog, Digitimer Ltd, Welwyn Garden City, Herts, UK). The action potentials were subsequently displayed on a digital storage oscilloscope (Model OS 1420, Gould Ltd, Hainault, Essex, UK). The signal also passed into a spike processor (Model D130, Digitimer Ltd).

In three animals, all tubing connected to cannulae in the chest were externalised through the chest wall and the chest was re-sealed by strapping the ribs and sternum together, and then suturing the overlying muscle and skin tightly together. A tube inserted in the chest cavity was attached to a vacuum source which was used to apply a controlled negative intrathoracic pressure, which was measured by an air-filled catheter positioned inside the chest cavity.

The temperatures of the blood in the circuit and the animal were recorded by thermister probes in the circuit and in the oesophagus and these were maintained at 37–39 °C by the heat exchangers in the circuit and in the oxygenator and by heaters under the animal table. Molar sodium bicarbonate was infused to maintain normal blood pH values (see below).

Blood pressures were recorded using saline-filled nylon catheters attached to strain gauges (Gould-Statham P23 ID), connected to: the left pulmonary cannula (pulmonary pouch pressure), the lumen

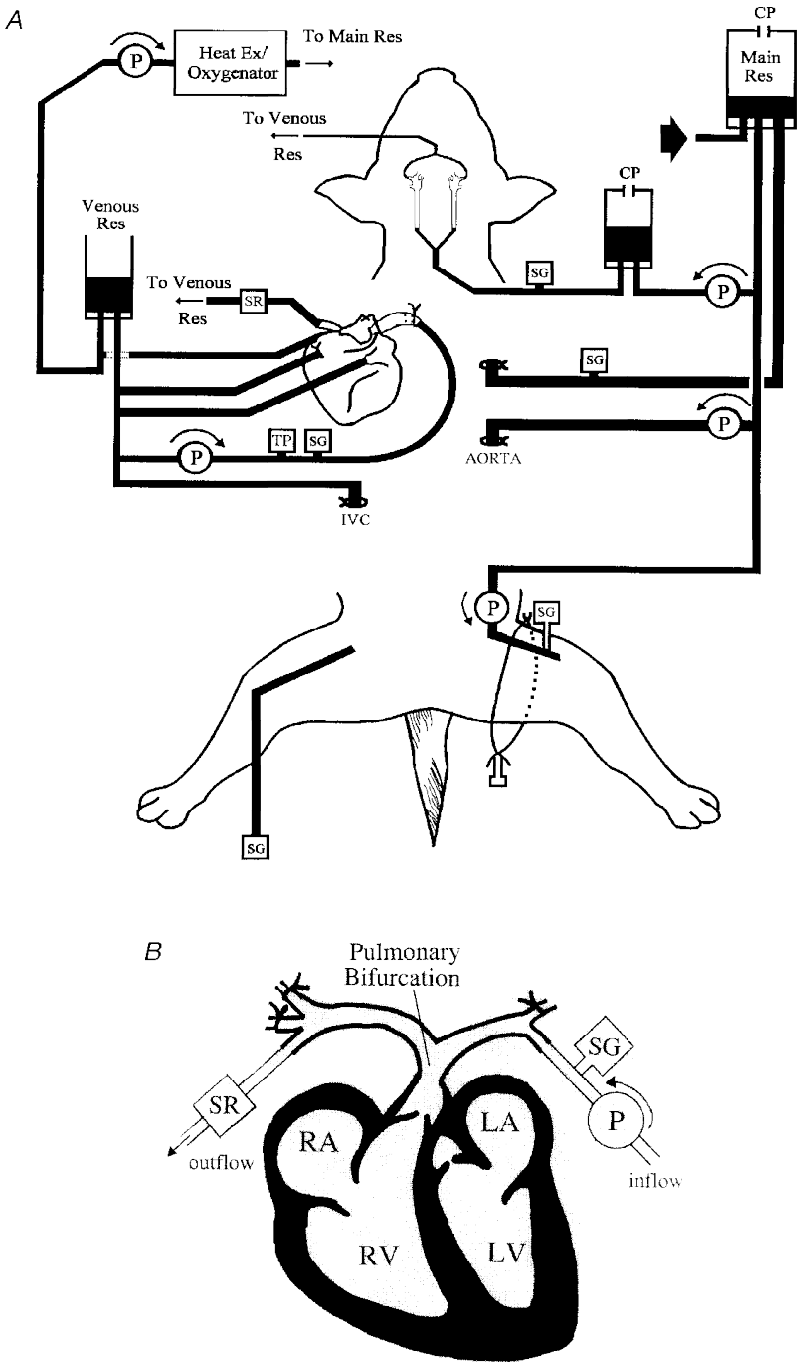

Figure 1

Diagram of experimental preparation (*A*) and the pulmonary bifurcation drainage and perfusion configuration (*B*). *A*, pressure was applied to the main reservoir to maintain perfusion pressure to the coronary and aortic arch baroreceptors via a cannula in the ascending aorta. Carotid sinus regions were perfused at constant pressure from a blood filled reservoir. Cannulae in the inferior vena cava (IVC), left atria, right atria and right ventricle drained blood into the venous reservoir. Venous blood was pumped into the left pulmonary artery and this region was drained through a cannula in the right pulmonary artery into the venous reservoir. Blood from this reservoir was pumped through a membrane oxygenator/heat exchanger to the main reservoir. The subdiaphragmatic and hind limb circulations were perfused at constant flows. CP, constant pressure; P, pump; SG, strain gauge transducer; SR, Starling resistor; TP, thermister probe. *B* shows details of the pulmonary arterial pouch. The pulmonary bifurcation was perfused with venous blood pumped at a constant flow through a cannula introduced into the left pulmonary artery proximal to the first lobar arteries which were ligated. Blood drained from the pulmonary bifurcation via a cannula inserted centrally in the right pulmonary artery. The upper right lung lobe was ligated at its root. LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

of the aortic cannula (thoracic aortic and cerebral perfusion pressure), the right carotid cannula (carotid sinus pressure), the limb perfusion cannula (limb perfusion pressure) and the abdominal aorta (systemic perfusion pressure). Signals of pressures and nerve activity were amplified (EMMA system, SE Laboratories, Feltham, UK) and recorded on VHS tape (Racal V-Store; Racal Recorders Ltd, Southampton, UK) and a direct-writing electrostatic recorder (ES1000, Gould, Ballainvilliers, France) and digitised (100 Hz) for online computer analysis (Fastdaq, Lectromed, Letchworth, UK). Before each experiment the pressure transducers were calibrated over 0–225 mmHg against a mercury column, except the pulmonary artery transducer where the range was 0–150 mmHg.

These experiments were carried out in accordance with the current UK legislation, the Animals (Scientific Procedures) Act, 1986. Experiments were terminated by exsanguination of the animal.

Experimental protocol

After connecting the animal to the perfusion circuit, pressures were allowed to stabilize and blood gases were measured and corrected as necessary to achieve values (means ± S.E.M., $n = 16$) for P_{O_2} of 194 ± 15 mmHg, P_{CO_2} of 40 ± 0.2 mmHg, and pH 7.4 ± 0.02. Haematocrit was 22 ± 1 %.

The protocol involved increasing the pressure in the pulmonary pouch from its control value in steps to 15, 30, 45, 60 and 75 mmHg (approximate mean pressures) for 1–2 min and then lowering it back to the control level for a further 1–2 min before the next increase in pulmonary pouch pressure. Measurements were taken during steady states in the 30 s period before changing the pulmonary artery pressure. The increase in pouch pressure was achieved by applying a pressure to a Starling resistor on the outflow cannula thus impeding outflow of blood from the pouch. Pressure perfusing the aortic arch, coronary and cerebral circulations and carotid sinuses were maintained constant throughout the tests. In five experiments the procedure was repeated with carotid pressure held at two different levels. In three experiments the procedure was repeated in the presence of a negative intrathoracic pressure. Ventilatory rate and stroke volume were unchanged once the cardiopulmonary bypass was established. When recordings were made of phrenic nerve activity the ventilator was switched off and the lungs were allowed to deflate against a pressure of 3 cmH_2O.

The reported pulmonary pressures are mean values and all other values reported are means ± S.E.M. Statistical significance was assessed by one-way ANOVA, Student's paired t test and two-way ANOVA as appropriate. Statistical analysis was carried out on the mean data from all tests (1–3) carried out in each animal.

RESULTS

Vascular responses at low carotid sinus pressures

Results are presented from 13 dogs. With carotid pressure held at 64 ± 2 mmHg and thoracic aortic pressure at 98 ± 3 mmHg, systemic and hind limb perfusion pressures were 157 ± 9 mmHg ($n = 13$) and 180 ± 13 mmHg ($n = 8$), respectively.

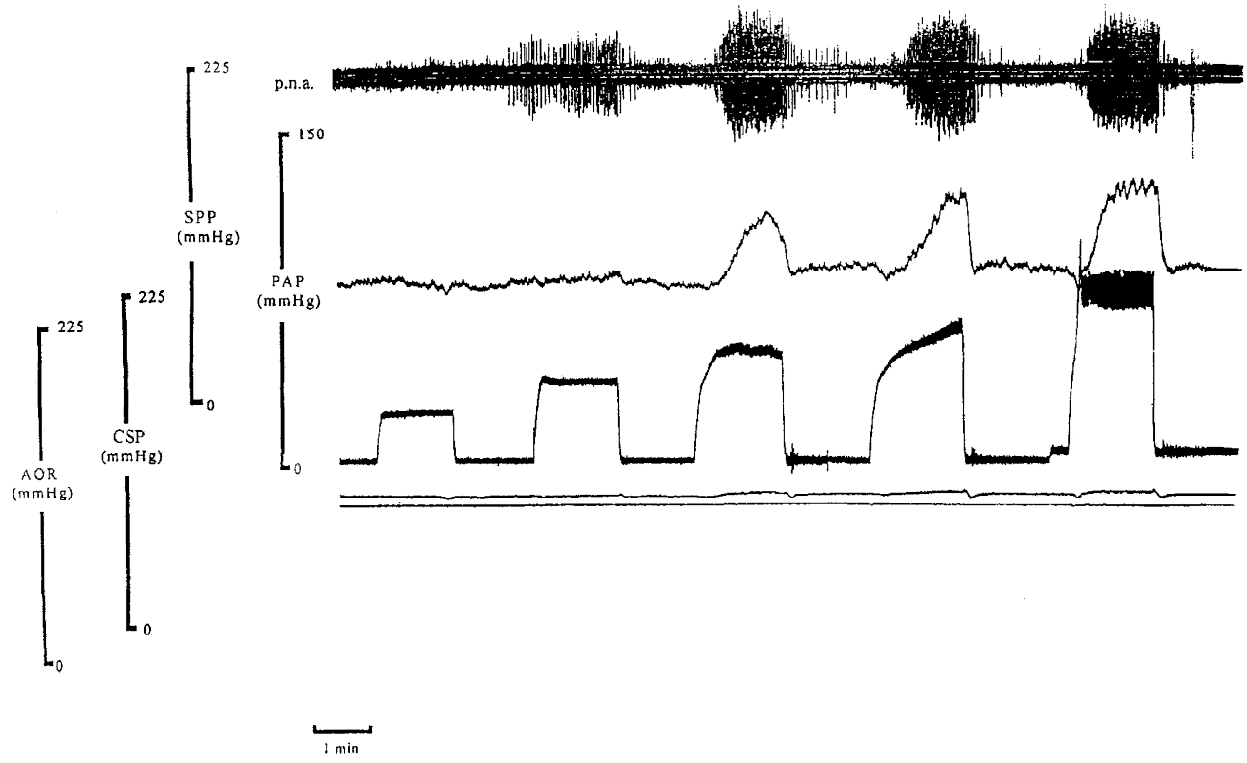

Figure 2

Systemic perfusion pressure and phrenic nerve activity responses to increases and decreases in pulmonary arterial pressure. The traces are of phrenic efferent nerve activity (p.n.a.), systemic perfusion pressure (SPP), pulmonary arterial pressure (PAP), carotid sinus pressure (CSP) and aortic root perfusion pressure (AOR). Note the absence of responses of SPP and phrenic activity at the lowest pressure tested and the increases in SPP and phrenic activity observed at higher pulmonary arterial pressures. Note also the low frequency burst of phrenic activity just prior to the second pressure step. This represents baseline discharge.

Table 1. Maximal transient decreases of systemic perfusion pressure (mmHg) after pulmonary artery pressure had been reduced from various levels to the control value of 6 ± 2 mmHg

	Initial pulmonary artery pressure (mmHg)				
Dog	17 ± 1	29 ± 1	43 ± 2	57 ± 2	70 ± 4
1	0	0	−14	−16	−35
2	0	0	0	−36	−66
3	0	0	0	−51	−61
4	0	0	0	0	−9
5	0	−23	−30	−36	−35
6	0	0	−23	−20	−47
7	0	−23	−56	−31	−35
8	0	0	−33	−35	−53
9	−22	−61	−25	−54	*
No. of dogs showing undershoot	1	3	6	8	9
Mean of those responding	−22	−36	−30	−35	−42

Changes in perfusion pressures in the 9 (out of 13) dogs in which an undershoot was seen. *Not tested in this dog, but vasodilatation was assumed to occur after this pressure.

An example of a test of increasing pulmonary arterial pressure to various levels is shown in Fig. 2. This trace was taken from one dog where mean pulmonary pressure was increased to different values from the control level of 5 mmHg up to a maximum of 81 mmHg. This figure shows that the smallest step increase in pulmonary pressure, to 18 mmHg, induced a very small (2 mmHg) fall in perfusion pressure. The next step, to 40 mmHg caused a small increase (+3 mmHg) perfusion pressure and an increase in phrenic activity. Subsequent steps, to 55 mmHg and above caused definite increases in both perfusion pressure and phrenic activity. Note that there was a spontaneous burst of phrenic activity just before the second step.

The responses of systemic and limb perfusion pressures in all dogs to step increases in pulmonary arterial pressure are shown in Fig. 3. The first step increase in mean pulmonary pressures (to 18 ± 1 mmHg) resulted in variable changes. There were decreases in systemic perfusion pressure of between −1 and −11 mmHg in four of the animals and either no change or a small increase in the remainder. Overall there was no significant change. Increases in pulmonary pressures to 32 mmHg and above caused significant increases in systemic perfusion pressure. Responses in the perfused limb were similar although, due to the smaller number of animals in which they were studied, these responses did not become statistically significant until higher distending pressures were applied.

In 9 of 13 animals a large undershoot in systemic perfusion pressure (−43 ± 7 mmHg) was observed after mean pulmonary pressure had been abruptly lowered from a high level (70 ± 4 mmHg) back to the low control level (6 ± 2 mmHg). An example of this is given in Fig. 4 which shows that when pulmonary pressure was decreased from a mean pressure of 70 mmHg to 12 mmHg there was an undershoot in systemic pressure of 46 mmHg below the mean control level of 174 mmHg. This then gradually returned to the control level in 24 s. These transient vasodilatations were sometimes also seen after pulmonary arterial pressure was restored after increasing to lower levels (Table 1). The magnitude of the undershoot was not obviously related to the magnitude of the pulmonary distension, but the probability of inducing the response was increased from only one out of the 13 dogs at the lowest pressure to 9 at the highest levels.

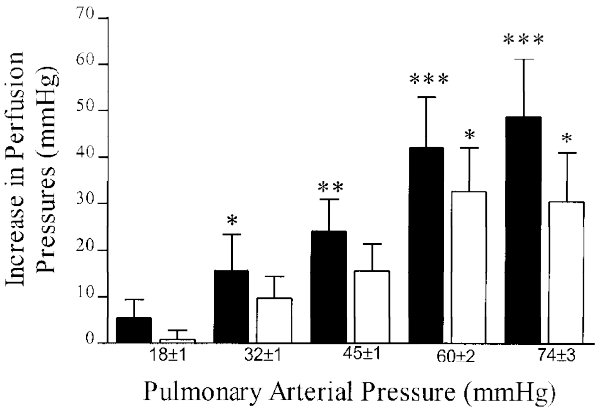

Figure 3

The increases in systemic and limb perfusion pressures at the various pulmonary arterial pressures. ■ shows systemic perfusion pressure responses and □ shows limb perfusion pressure responses. Results of systemic pressure responses were obtained from 13 animals, except the steps at 74 mmHg for which 10 animals were used. For limb responses 8 animals (6 at 74 mmHg) were used. The control pulmonary arterial pressure was 5 ± 1 mmHg and the corresponding values of systemic and limb perfusion pressures were 162 ± 11 mmHg and 180 ± 13 mmHg. Columns and bars represent means ± S.E.M.; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$ (one-way ANOVA) compared to control values.

Phrenic nerve discharge

Recordings of phrenic efferent discharge were made in five of the animals. The mean discharge during the control period was 40 ± 15 impulses s^{-1} with a burst frequency between 0 and 23 bursts min^{-1}. In all animals, increasing pulmonary arterial pressure resulted in bursts of activity in previously silent nerves or increases in mean discharge and burst frequency (e.g. Fig. 2). These responses are summarised in Fig. 5 where the changes in phrenic nerve discharge during the bursts and the change in mean discharge frequency during the pulmonary artery distension from control are plotted against the applied pulmonary pressure. Although all animals showed an increase in activity during pulmonary artery distension, due to the small numbers of animals tested and the variability of the reponses, the changes became significant only at high pulmonary pressures.

Denervation studies

In nine of the dogs increases in pulmonary pressure to 68 ± 2 mmHg, before and after bilateral vagotomy, resulted in changes in systemic perfusion pressure of $+30 \pm 8$ mmHg ($P < 0.006$; Student's paired t test) and -0.3 ± 0.9 mmHg ($P > 0.3$), respectively. In three dogs in which phrenic nerve activity was assessed, it increased by 12, 38 and 231 impulses s^{-1} before and 2, 1 and 19 impulses s^{-1} after vagotomy, respectively. The undershoot of vascular resistance after lowering pulmonary pressure was -33 ± 7 mmHg ($P < 0.005$; Student's paired t test, $n = 6$) before vagotomy and was abolished by vagotomy.

In two animals, cutting the femoral and sciatic nerves to the isolated hind limb abolished the vascular response in this region to increases in pulmonary pressure.

Effect of carotid sinus pressure on vascular responses to pulmonary pressure changes

In five of the animals we examined the effect of setting carotid sinus pressure at 65 ± 1 and 175 ± 16 mmHg on the reflex effects of pulmonary arterial distension. When carotid pressure was ~60 mmHg, systemic perfusion pressure was 201 ± 10 mmHg and when it was 175 ± 16 mmHg the perfusion pressure was significantly different ($P = 0.02$, Student's paired t test) at 149 ± 15 mmHg. However, the magnitude of change in systemic pressure in response to each step increase in pulmonary pressure was unaffected by the prevailing carotid sinus pressure as assessed by two-way ANOVA (see Fig. 6). Recalculation of responses as percentage changes also failed to show significant differences.

Effect of a negative intrathoracic pressure on vascular responses to pulmonary pressure changes

In three additional dogs in which the perfusion lines were exteriorised and the chests were closed, we investigated the

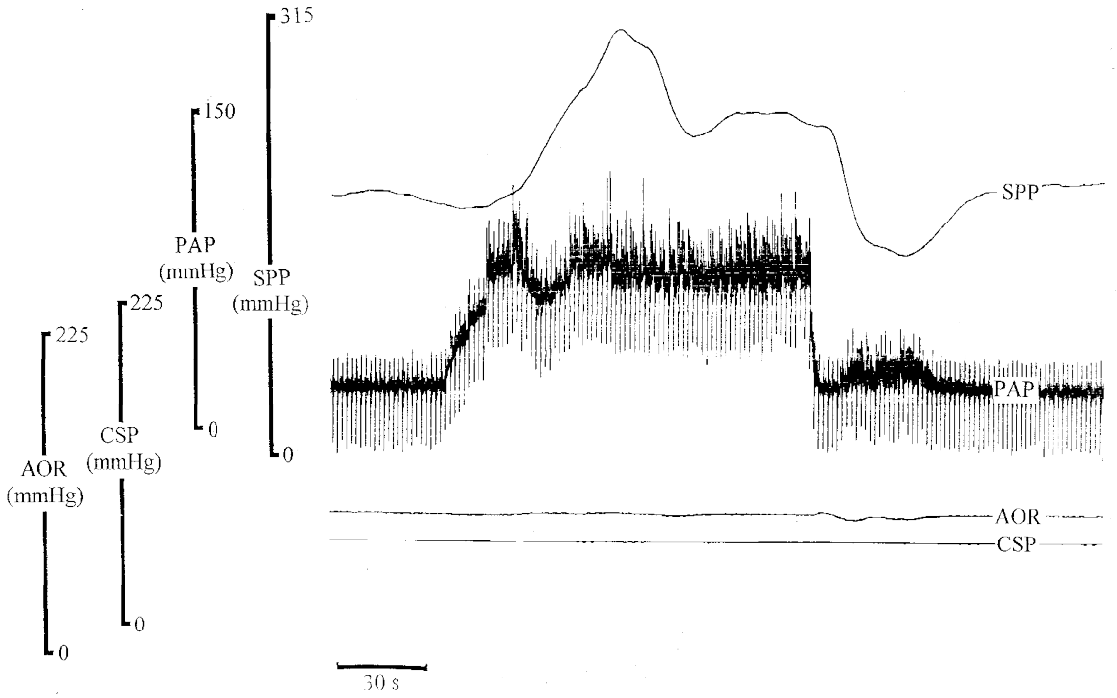

Figure 4

Systemic perfusion pressure response to an increase and decrease in pulmonary arterial pressure. The traces are of systemic perfusion pressure (SPP), pulmonary arterial pressure (PAP), carotid sinus pressure (CSP) and aortic root perfusion pressure (AOR). Note the large undershoot of SPP following the decrease in pulmonary arterial pressure. In this test there was also, unusually, an overshoot in SPP following the increase in pulmonary arterial pressure.

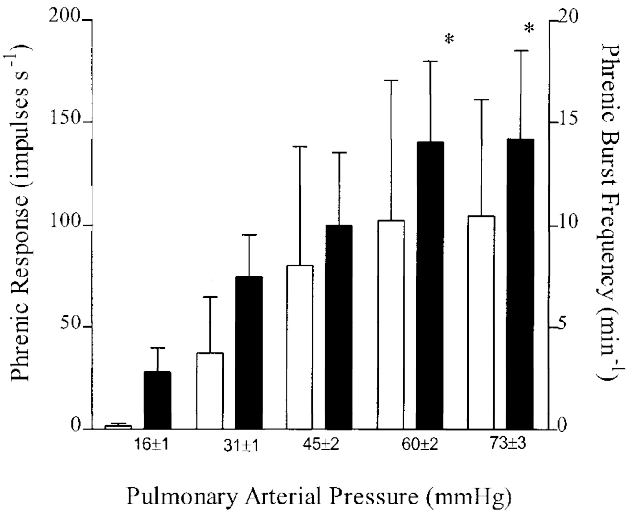

Figure 5

Responses of phrenic nerve activity to step increases in pulmonary arterial pressure expressed as changes from control values. Control pulmonary arterial pressure was 1 ± 1 mmHg. ■, phrenic burst frequency (min^{-1}); □, phrenic nerve response (impulses s^{-1}). Columns and bars represent means ± S.E.M.; *$P < 0.05$ (one-way ANOVA) compared to control values, ($n = 5$).

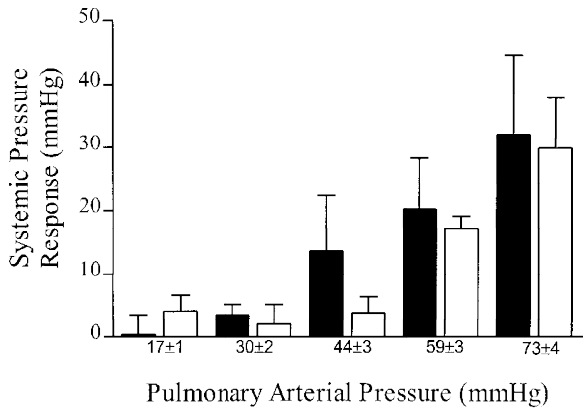

Figure 6

Responses of systemic perfusion pressure to step increases in pulmonary arterial pressure at low and high carotid sinus pressures. ■, carotid sinus pressure = 60 mmHg; □, carotid sinus pressure = 175 ± 16 mmHg. Control values of pulmonary arterial pressure were 6 ± 2 mmHg; values of systemic perfusion pressure at the low and high carotid pressures were 201 ± 10 mmHg and 149 ± 15 mmHg. Columns and bars represent means ± S.E.M. There were no significant differences between the systemic response to increases in pulmonary arterial pressure at low and high carotid pressure ($P > 0.05$, two-way ANOVA, $n = 5$).

systemic vascular responses to graded steps in pulmonary arterial pressure, with either atmospheric or a subatmospheric (−20 mmHg) pressure in the thorax.

An example of the responses from one animal is shown in Fig. 7. An increase in mean pulmonary arterial pressure from 6 to 17 mmHg with intrathoracic pressure at atmospheric level had almost no effect on systemic perfusion pressure (Fig. 7*A*). However, with a −20 mmHg intrathoracic pressure the same stimulus resulted in a increase in systemic perfusion pressure, of 12 mmHg (12 %) from the control level of 100 mmHg (Fig. 7*B*).

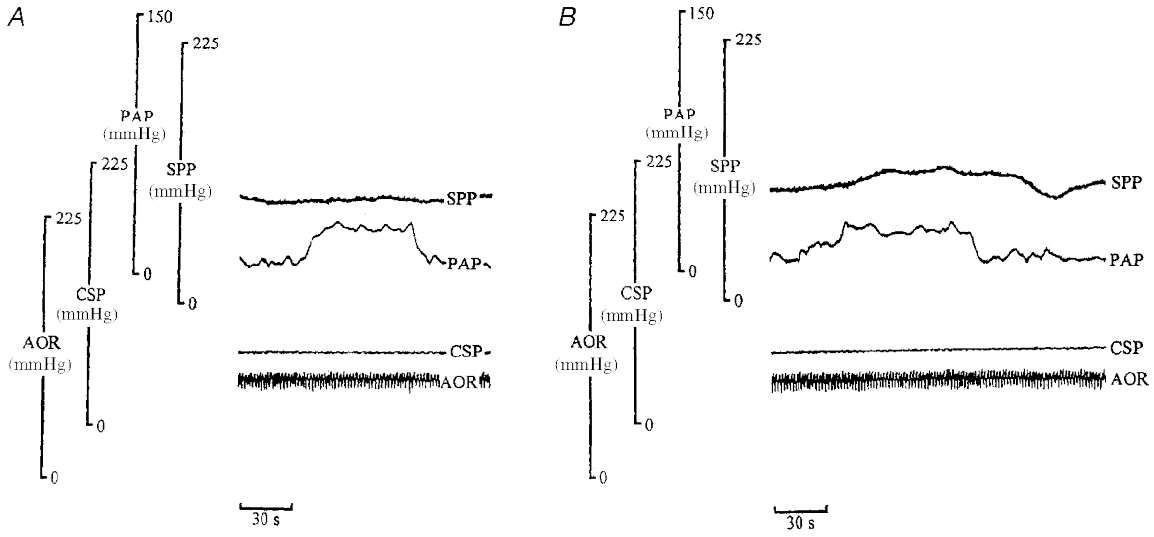

Figure 7

Systemic perfusion pressure responses in closed-chest dog to changes in pulmonary arterial pressure with and without a negative intrathoracic pressure. The traces are of systemic perfusion pressure (SPP), pulmonary arterial pressure (PAP), carotid sinus pressure (CSP) and aortic root perfusion pressure (AOR). *A*, pulmonary pressure test without the negative intrathoracic pressure; note the absence of an SPP response. *B*, pulmonary pressure test in the presence of an intrathoracic pressure of −20 mmHg; note the increase in SPP.

In the three animals tested increasing pulmonary arterial pressure from 8, 13 and 12 mmHg to 48, 44 and 44 mmHg, respectively, with intrathoracic pressure at atmospheric level caused systemic perfusion pressure to change by −2, +12 and +9 mmHg from the control levels of 158, 171 and 95 mmHg. In the presence of a −20 mmHg intrathoracic pressure, similar changes in pulmonary arterial pressures evoked much larger increases in systemic perfusion pressure of +8, +23 and +27 mmHg from the control levels of 104, 139 and 98 mmHg, respectively. Responses were also obtained at lower pulmonary arterial pressures in the presence of the negative intrathoracic pressure. The lowest pressure required to induce responses in these animals was reduced from 30–45 mmHg to 15–22 mmHg.

DISCUSSION

The principal findings from this study were that, using a preparation in which the stimulus was carefully localised to the pulmonary artery and its bifurcation and in which secondary responses from other reflexogenic regions were largely prevented, increases in pulmonary arterial pressure to 40 mmHg or more caused significant increases in peripheral vascular resistance and phrenic nerve activity.

Although there have been previous reports of the effects of changing pulmonary arterial pressure, this is the first study which has applied graded changes in pressure to the pulmonary bifurcation, pulmonary trunk and extrapulmonary arteries whilst controlling the stimuli to other known reflexogenic areas. In particular, we maintained constant pressures to baroreceptors in the carotid sinuses, aortic arch and coronary arteries (McMahon *et al*. 1996), and the cardiopulmonary bypass should have prevented changes in activity from cardiac receptors (Hainsworth, 1991). Previous attempts to assess the role of pulmonary arterial baroreceptors have not satisfied the criteria set out for this study. For example, distension of balloons in and around the pulmonary bifurcation (Lewin *et al*. 1961; Osorio & Russek, 1962; Laks *et al*. 1975; Crisp *et al*. 1988) do not apply a stimulus that is possible to relate to physiological events.

There have been other studies of responses to changes in extrapulmonary pulmonary arterial pressures using blood perfusion of the isolated regions (Lewin *et al*. 1961; Coleridge & Kidd, 1963; Ledsome & Kan, 1977). There are, however, important differences both in techniques used and in the responses obtained in the previous studies and our present report. Coleridge & Kidd (1963) reported that increases in pulmonary artery pressure of up to 60 mmHg induced reflex vasodilatation in 8 out of 18 animals tested and no change in the remainder; above that level the response changed to constriction in 8 out of 10 animals tested. Ledsome & Kan (1977) reported that vasodilatation occurred only when the temperature of the perfusate was abnormally low (< 30 °C) and this was thought to explain some of the responses reported by Coleridge & Kidd (1963): at normal temperatures only vasoconstriction occurred. Both Lewin *et al*. (1961) and Ledsome & Kan (1977) reported results qualitatively similar to ours. However, the physiological significance of their results is questionable due to the high pressures required to induce a response. In Ledsome & Kan's study a 10 % increase in systemic vascular resistance was only obtained when pulmonary arterial pressure was increased to 90 mmHg, whereas in our report pressures of only about 40 mmHg were necessary to cause the same response. The reason for the large difference in the required pressures is not known. However, one likely possibility is that buffering by various baroreceptors and other vascular mechanoreceptors could well have masked the responses in the previous studies. Another consideration is that in our study we controlled systemic arterial blood flow and so we could be confident that changes in blood pressure accurately reflected changes in vascular resistance.

Another interesting observation in the present study which has not previously been reported was the large undershoot in perfusion pressure which frequently occurred when pulmonary arterial pressure was abruptly decreased, particularly from high levels. Responses also sometimes occurred when pressure was decreased from lower levels, and the likelihood of this response occurring increased with increasing distension pressures. The significance of this response is uncertain, but it may be relevant in conditions where pulmonary artery distension changes abruptly.

The other effect of an increase in pulmonary arterial pressure was an increase in phrenic nerve activity, seen as increases in both the frequency of the bursts and in the mean discharge. These effects on respiration have been seen by others, albeit in response to much higher pressures (Kan *et al*. 1979; Ledsome *et al*. 1980; Crisp *et al*. 1988).

The reflex nature of the responses was demonstrated by their dependence on the vagus nerves and, in the case of the limb responses, on the sciatic and femoral nerves. Although afferent sympathetic nerves with endings in the pulmonary artery are also known to be present (Nishi *et al*. 1974; Uchida, 1975), since all the responses obtained in the present study were abolished by bilateral vagotomy, it is unlikely that sympathetic afferents made any major contribution.

Before we can speculate on the possible physiological significance of pulmonary arterial baroreceptors, we need to consider the range of pressures over which they induce responses. In the present experiments the lowest pressures needed to induce reflex responses were still well above those usually encountered within the extrapulmonary arteries and bifurcation. For both the vascular and the respiratory responses it was necessary to increase the pulmonary arterial pressure to more than 25 mmHg and often to as much as 40 mmHg. In the conscious resting dog mean pulmonary arterial pressure has been reported to be 17 mmHg (Katz & Steinitz, 1939) and in healthy resting humans values of around 15 mmHg have been reported (Harris & Heath, 1977). This pressure may increase to 25 mmHg or a little more during strenuous exercise in healthy humans (Harris & Heath, 1977) and pulmonary hypertension is characterised by sustained pressures of 25 mmHg or more (Wagenvoort & Wagenvoort,

1977). Our results, therefore, would seem to indicate that, in both normal dogs and also in normal humans, if their pulmonary baroreceptors behave similarly, pulmonary arterial baroreceptors are unlikely to induce major reflex responses under physiological conditions. There are, however, some considerations which may allow for a more physiological role for the pulmonary arterial reflex.

Firstly, it is possible that, in preparations in which the pulmonary artery is vascularly isolated, the surgical procedures involved may have resulted in some damage to the receptors or to their nerve supply. We took extreme care to avoid damage to nerves but cannot entirely rule out the possibility that some might have been damaged. A further possibility is that the extensive surgery and the anaesthetic could have depressed the reflex. This, therefore, raises the possibility that in the intact situation pulmonary baroreceptors may elicit responses at pressures lower than those reported here. This possibility is supported by the observations of Bianconi & Green (1959) and Coleridge & Kidd (1961) that in intact animals (with closed chests) there was an afferent discharge from the pulmonary arterial baroreceptors at normal levels of pulmonary arterial pressure.

Another factor which is very likely to influence the activity of pulmonary arterial baroreceptors is the effect of respiration on pressure gradients across the pulmonary vessels. Pleural and intrathoracic pressure at rest changes from about −8 mmHg during normal inspiration to −3 mmHg during expiration. In exercise, respiratory effort is enhanced and this may shift intrathoracic pressure to −23 mmHg during inspiration, and during expiration the pressure ranges from +4 to +23 mmHg (Janicki *et al*. 1996). In the major part of our study the chest was open so the pulmonary arterial pressure could not have been influenced by changes in intrathoracic pressure. It is likely, however, in the intact animal that the reflex responses from pulmonary arterial baroreceptors would be observed at lower pulmonary arterial pressures due to the effect of the negative intrathoracic pressure on transmural pulmonary arterial pressure. We tested this hypothesis in three animals in which a negative intrathoracic pressure of −20 mmHg was applied. The systemic vascular response in the presence of the −20 mmHg intrathoracic pressure was over three times greater than the response observed without the negative pressure and the value of the pulmonary pressure required to induce the vascular response was greatly reduced by the addition of the negative intrathoracic pressure. This effect would be particularly important during the large changes occurring in intrathoracic pressure during exercise. It may also be influenced by some lung diseases, including both obstructive and restrictive lung diseases, which may affect intrathoracic pressures.

If we assume that the pulmonary arterial baroreflex exists in humans and that it is elicited by the transmural pressures encountered in healthy subjects during exercise and, to a greater extent in patients with pulmonary hypertension or lung disease, we need to speculate on its possible physiological or pathophysiological significance. Other baroreceptor reflexes – carotid, aortic and coronary – serve as negative feedback systems, so that as pressure increases the resulting vasodilatation would tend to return it towards its previous levels. It is less obvious what might be controlled by pulmonary baroreceptors. One possibility is that the reflex could be concerned with mediating the responses to exercise. An increased inflow to the heart together with an increase in pulmonary ventilation would increase the stimulus to pulmonary arterial baroreceptors, leading to generalised systemic vasoconstriction which would divert blood to metabolically active muscle. It may also contribute to the increased ventilatory response occurring during exercise and this could function as a feedforward mechanism (Wasserman *et al*. 1986) in that stimulation causes an increased ventilation which may then cause an even greater stimulation of the pulmonary arterial baroreceptors.

In conclusion, this study has demonstrated the existence of a pulmonary artery baroreflex which induces peripheral vasoconstriction and an increased respiratory drive. This reflex is evoked at high pulmonary artery transmural pressures which may be encountered during exercise and in diseases of the lung including pulmonary hypertension.

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