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OBSERVATIONS ON THE EFFECTS OF HYPOXIA ON THE PULMONARY VASCULAR BED

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Inhalation of mixtures of O_2 and N_2 in which the O_2 concentration is less than 15% causes an increase in pulmonary vascular resistance in the cat. This response occurs in the anaesthetized animal (von Euler & Liljestrand, 1946), in anaesthetized animals with left lung perfusion (Duke, 1954) and in isolated lung preparations (Nisell, 1948, 1951; Duke, 1951, 1954; Duke & Killick, 1952). Von Euler & Liljestrand (1946) found that hypoxia caused a rise of pulmonary arterial pressure in anaesthetized cats after removal of both stellate ganglia. This response was not abolished by ergotoxine (Logaras, 1947), nor did it appear to originate from changes in bronchial calibre or systemic and left auricular blood pressure (Duke, 1954).

A pulmonary pressor response to hypoxia also occurs in anaesthetized dogs. It has been suggested that this is dependent on the integrity of the sympathetic nerves and adrenal glands (Stroud & Rahn, 1953; Nahas, Mather, Wargo & Adams, 1954; Nahas, Visscher, Mather, Haddy & Warner, 1954), or that it is caused by the increase of cardiac output which accompanies hypoxia in the whole animal (Aviado, Cerletti, Alanis, Bulle & Schmidt, 1952; Hürlimann & Wiggers, 1953; Leusen & Demeester, 1955), although Hall (1953) found a constrictor response to hypoxia in a denervated perfused lung lobe.

In view of the conflicting evidence between the cat and the dog it appeared profitable to investigate whether pulmonary constrictor responses to hypoxia occurred in the dog when changes in cardiac output were excluded and also to find whether the pressor response in cats was mediated by neurohumoral influences or by a vasoconstrictor substance.

METHODS

Isolated cat lungs were set up and perfused through the pulmonary artery with the animals' own heparinized blood as previously described (Duke, 1951). Perfusion was at constant volume inflow with a Dale-Schuster pump. The venous outflow from the lungs was collected from the left auricular cannula into a reservoir which fed the pump. Ventilation was by positive pressure using a

Starling 'Ideal' pump; measurements of the resistance of the lungs to inflation were usually made (Konzett & Rössler, 1940). Dextran ('Intradex', Glaxo) was sometimes used to augment the blood obtained from the animal. Dog lungs were perfused in a similar manner (Duke, 1949).

The perfused left lung preparation was made in cats and dogs under chloralose anaesthesia (0·1 g/kg intraperitoneally), using the technique previously described (Duke, 1954). In both species the trachea was cannulated and the blood pressure recorded from the right femoral artery with a mercury manometer. The chest was opened in the mid line and held widely retracted with the animal under positive pressure respiration. Heparin (Liquemin, Roche, 1000 u./kg) was injected intravenously. The pump and its connexions were filled with warm Dextran. A cannula was placed in the right auricle and the left pulmonary artery was tied intrapericardially and cannulated. Perfusion of the left lung was performed at constant volume inflow with blood from the right auricle using a Dale-Schuster pump. The left auricle was cannulated and connected to a manometer filled with 0·9% NaCl solution. The right lung received its blood in the normal way.

In all experiments pulmonary arterial pressure was measured by a manometer filled with 0·9% NaCl solution and recorded by a tambour connected to the pulmonary arterial tubing by a T-piece. Left auricular pressure was measured by a manometer filled with 0·9% NaCl solution connected to the left auricular tubing in isolated lung experiments and to the left auricular cannula in left lung perfusion experiments. Left auricular pressure was recorded by a small volume recorder connected to the top of the manometer. The gas mixtures used were from gas cylinders supplied by the British Oxygen Company. Changes in the ventilating gas mixture were made by attaching Douglas bags to the input of the Starling pump.

In some experiments the possibility that a vasoconstrictor substance was liberated from the lungs during ventilation with N_2 was investigated. For this purpose the cat's hind limb was perfused by cannulating the femoral artery and femoral vein and severing the limb from the body of the animal. The limb was placed so that the venous outflow drained on to a tray and thence into a venous reservoir. The perfusion of the hind limb was at constant volume inflow through the femoral artery with blood which had previously been perfused through a pair of isolated lungs. The femoral blood pressure was recorded with a mercury manometer.

Chronic sympathectomies were performed in the atropinized animal under pentobarbitone and ether anaesthesia with full aseptic precautions. The sympathetic chain was removed on one side from T5–6 up to and including the stellate ganglion. A similar operation was performed 8–10 days later on the other side. The removal of the stellate ganglion was confirmed after recovery from the anaesthetic by the relaxation of the nictitating membrane on the operated side. A bilateral cervical sympathectomy was performed 8–10 days after the second thoracic operation by removing a length of chain between the thyroid cartilage and the middle cervical ganglion. Vagotomies were performed on the right side below the recurrent laryngeal nerve at the same time as the right thoracic sympathectomy and the left cervical vagus was cut during the cervical sympathectomy. The animals were used for acute experiments 10–15 days after cervical sympathectomy and were without exception in good condition at this time.

RESULTS

Isolated perfused lung experiments

Ventilation of the lungs with N_2 instead of air for 2–6 min produced a rise of pulmonary arterial pressure and a slight bronchoconstriction in the lungs of dogs. The pulmonary pressor response to 4–5 min ventilation with N_2 or 5% O_2 in N_2 is shown in Fig. 1. The effect of N_2 is more pronounced than that of 5% O_2 in N_2.

Table 1 shows the effect of 3–5 min ventilation with N_2 on the pulmonary arterial and left auricular pressures of cat lungs. The arterial pressure and

resistance are invariably increased and in four out of five experiments the left auricular pressure was slightly reduced. The reduction of left auricular pressure coincident with the rise in arterial pressure is well shown in Fig. 2. In this experiment the venous pressure had been artificially raised by partial obstruction of outflow tubing. The pressor effect of N_2 ventilation was also found in all three preparations made from cats which had been subjected to a chronic sympathectomy and vagotomy (Fig. 3*B*, *D*).

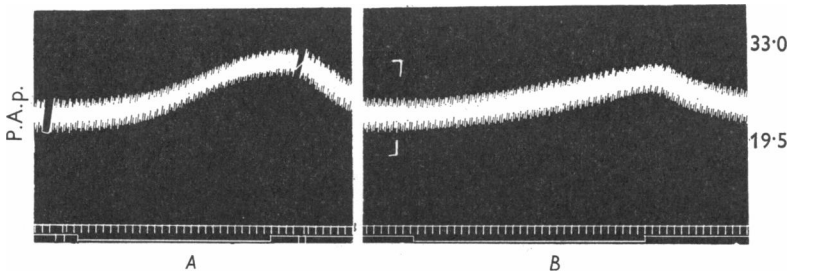

Fig. 1. Dog, 17·0 kg. Isolated perfused lung. Perfusion begun 11.30 a.m., ventilation with air; *A*, 2.32 p.m., change to N_2 during signal; *B*, 2.50 p.m., change to 5% O_2 in N_2 during signal; P.A.p., pulmonary arterial pressure (cm 0·9% NaCl soln.); time marker: 10 sec in all figures.

TABLE 1. Isolated perfused cats' lungs. Before and 3–5 min after changing the ventilating gas mixture from air to N_2

Expt. no.	P.A.p. (cm 0·9% NaCl soln.) Before	P.A.p. (cm 0·9% NaCl soln.) After	L.A.p. (cm 0·9% NaCl soln.) Before	L.A.p. (cm 0·9% NaCl soln.) After	R Before	R After	R Increase (%)
28	17·5	29·75	3·4	3·2	14·1	26·55	88
29	18·4	20·9	3·0	2·7	15·4	18·2	18
31	25·6	34·9	3·5	3·5	22·1	31·4	42
34	16·1	25·9	4·1	4·0	12·0	21·9	82·5
35	16·0	31·6	3·5	3·1	12·5	28·5	133

P.A.p., pulmonary arterial pressure; L.A.p., left auricular pressure; R, P.A.p. – L.A.p. which, at constant volume inflow perfusion, is directly proportional to the resistance of the pulmonary vascular bed.

Three experiments were also performed on cats in which the left hind limb was perfused in series with the lungs. In these experiments ventilation of the lungs with N_2 caused an increase of pulmonary arterial pressure, but the femoral arterial pressure fell (Fig. 4).

Injection of 5-hydroxytryptamine (HT) into the pulmonary artery produced a pulmonary arterial pressor effect, and the dose of HT was adjusted so that the response was of similar size to that produced by ventilation of the lungs with N_2 for 2–3 min. Lysergic acid diethylamide (LSD) did not reduce the pressor response to N_2 (Fig. 3) but abolished that to much larger doses of HT than had previously been given.

Left lung perfusion experiments

In the only experiment on a dog (Fig. 5) ventilation with 5 % O_2 in N_2 caused a rise of left pulmonary arterial pressure and slight bronchoconstriction with no significant change in left auricular pressure. In the cat the left pulmonary arterial pressor response to anoxia was not prevented by removal of the adrenal glands, although it was sometimes reduced.

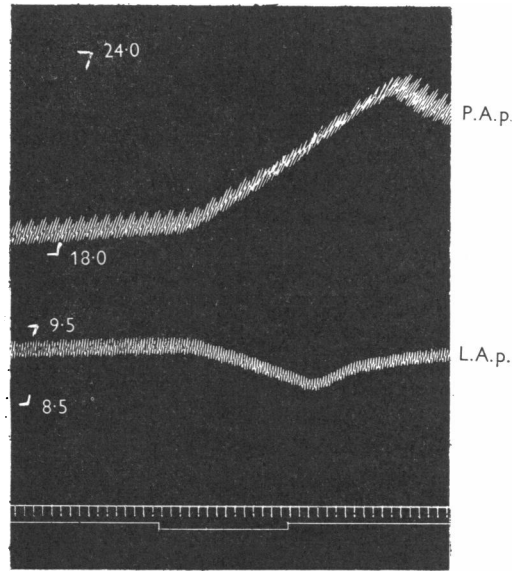

Fig. 2. Cat, 2·2 kg. Isolated perfused lung. Positive pressure respiration with air; during signa ventilation changed to N_2. L.A.p., left auricular pressure (cm 0·9 % NaCl soln.).

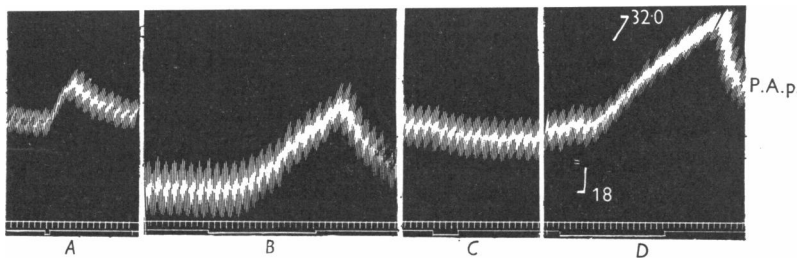

Fig. 3. Cat, 2·4 kg. Previous history: 16. ii. 55, right thoracic sympathectomy and right thoracic vagotomy; 23. ii. 55, left thoracic sympathectomy; 2. iii. 55, bilateral cervical sympathectomy and left cervical vagotomy.

15. iii. 55, isolated perfused lung; *A*, 1.07 p.m., 10 μg 5-HT into pulmonary artery at signal; *B*, 1.27 p.m., ventilation changed from air to N_2 during signal; 1.33 p.m., 0·24 mg LSD injected into pulmonary artery; *C*, 1.55 p.m., 100 μg HT at signal; *D*, 2.07 p.m., ventilation changed from air to N_2 during signal.

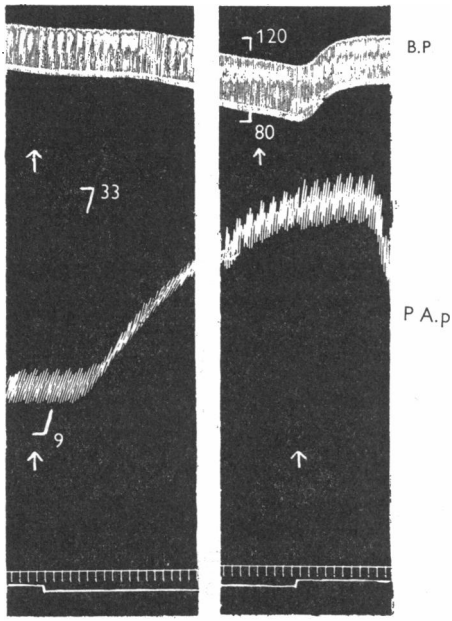

Fig. 4

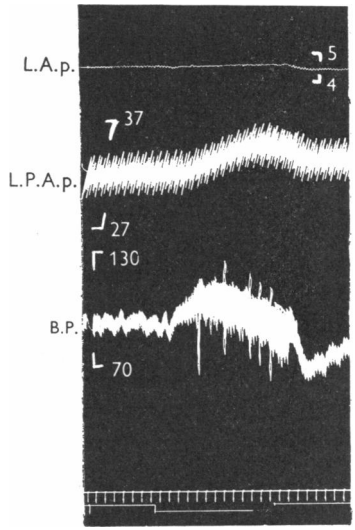

Fig. 5

Fig. 4. Cat, 3·0 kg. Left hind leg perfused in series with isolated lungs. B.P., left femoral arterial blood pressure; perfusion started at 11.03 a.m.; *A*, 11.50 a.m., ventilation changed from air to N_2 at signal; *B*, 11.55 a.m., ventilation with N_2 continued; changed to air at signal.

Fig. 5. Dog, 17·0 kg. Left lung perfusion started 2.24 p.m.; ventilation with air; 3.40 p.m., ventilation changed to 5 % O_2 in N_2 during signal; B.P., systemic blood pressure; L.P.A.p., left pulmonary arterial pressure.

DISCUSSION

The experiments now reported provide evidence that the pulmonary pressor effect of anoxia occurs in the dog under conditions in which changes of cardiac output and of left auricular pressure can be excluded as causes of pulmonary pressor responses, and they therefore support the observations of Hall (1953). The pulmonary pressor response to anoxia has been analysed more fully in cats. In this species it persists after removal of the adrenal glands so that change in the rate of secretions from the adrenal medulla cannot be a factor on which the response depends. If it is assumed that the anoxic rise of pulmonary arterial pressure is caused by similar mechanisms in the anaesthetized animal as in isolated lungs then neurohumoral factors are unlikely to be responsible. This hypothesis has been further tested by studying the effects of anoxia in isolated lung preparations made from animals which had been subjected to a chronic denervation of the lungs. In these preparations ventilation with N_2 had a pulmonary pressor effect and thus the activation of local

intra-pulmonary reflexes involving the sympathetic system is improbable. This conclusion is supported by the fact (Duke, 1951) that the anoxic pressure rise is not reversed or inhibited by dihydroergotamine. Assuming that all, or the majority, of the vagal efferent fibres were sectioned in the chronic experiments the possibility still exists that local reflexes involving the parasympathetic nervous system are involved but in this case they must be atropine-resistant, since pulmonary pressor responses to anoxia occur in isolated lungs after doses of atropine sufficient to block the effects of injected acetylcholine (Duke, 1951). A phenomenon which is still unexplained is the increasing sensitivity to N_2 found in most perfused lung preparations during the course of the experiment (Duke & Killick, 1952).

The direct dilator effect of anoxia on the blood vessels of the systemic circulation is well known (Fleisch, Sibul & Ponomarev, 1932). The contrast between this and the constrictor effect in the pulmonary circuit is striking. There is a possibility that a vasoconstrictor substance might be liberated from the lungs during ventilation with N_2, but no evidence for this was found since in the present experiments ventilation of the lungs with N_2 causes a pulmonary pressor but a femoral depressor response and in the experiments of Fleisch *et al.* the femoral blood flow increased at constant pressure inflow. Finally, the liberation of HT as a result of N_2 appears improbable since injection of LSD to block the response to injected HT has no effect on the response to N_2.

The site of action of N_2 on the pulmonary circulation can still only be postulated, although it has been suggested (Duke, 1954) that ventilation with N_2 may cause capillary constriction. The fall of left auricular pressure coincident with the rise of pulmonary arterial pressure, found in the experiments now reported, makes it appear that the vasoconstriction is indeed in the pulmonary vascular bed and that the pulmonary pressor response is not a mechanical effect caused from alterations in the amount of blood transferred between the pulmonary and bronchial blood vessels. It also appears probable that the dog and the cat behave similarly towards hypoxia in that a pulmonary pressor response occurs which is presumably due to pulmonary vasoconstriction. There can be no certainty that the rise in pulmonary arterial pressure in the dog is independent of changes in resistance of the lungs to inflation, although in the cat it is not dependent on such changes (Duke, 1954). These experiments do not exclude the possibility that passive or active vasodilatation of some parts of the pulmonary vascular bed occurs concurrently with vasoconstriction of other parts. In this connexion Nisell (1952) noted an increase in lung blood volume when isolated cats' lungs were perfused with partially deoxygenated blood, and Aviado *et al.* (1952) found an increase of lung blood volume in anaesthetized dogs.

SUMMARY

1. In isolated cats' lungs perfused through the pulmonary artery with the animal's own heparinized blood at constant volume inflow, ventilation of the

lungs with N_2 instead of air produces a rise of pulmonary arterial pressure and a fall of left auricular pressure.

2. The pressor response to N_2 occurs in preparations made from animals in which the lungs were chronically denervated: it is not inhibited by LSD.

3. No evidence of a circulating vasoconstrictor substance is found by perfusing the cat's denervated hind limb with blood which has been partially deoxygenated in the lungs.

4. Pulmonary pressor responses to hypoxia are found in isolated perfused dog lungs and in anaesthetized dogs with left lung perfusion.

5. In the anaesthetized cat with left lung perfusion the increase of left pulmonary arterial pressure in response to inhalation of 5% O_2 in N_2 is not prevented by removal of both adrenal glands.

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