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THE EFFECTS OF STIMULATION OF THE CAROTID BODY CHEMORECEPTORS ON PULMONARY VASCULAR RESISTANCE IN THE DOG

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In a previous study we investigated the effects of carotid sinus baroreceptor reflexes on pulmonary vascular resistance (Daly & Daly, 1957*b*, *c*). An examination has now been made of the reflex effects of stimulation of the carotid body chemoreceptors on the lesser circulation and the results which have been briefly reported elsewhere (Daly & Daly, 1957*a*) are presented in this paper.

METHODS

Dogs of 14·7–19·6 kg body weight were anaesthetized with chloralose (0·1 g/kg intravenously) after premedication with morphine hydrochloride (1 mg/kg subcutaneously). The chest was opened in the mid-sternal line and the lungs ventilated by means of a Starling 'Ideal' pump at constant pressure, which varied from 8 to 15 cm water in different experiments. During expiration the lungs collapsed passively against a resistance of 2–2·5 cm water. Changes in tidal air volume were measured by the method of Konzett & Rössler (1940). Both phrenic and recurrent laryngeal nerves were crushed to minimize mechanical effects on the lungs.

Perfusion of the carotid bodies. Reflex effects from the carotid body chemoreceptors were elicited by temporarily changing the perfusate from arterial blood to venous. For this purpose both carotid bifurcation regions were isolated from the circulation by ligation of all branches of the common and external carotid arteries. The veins draining the carotid bodies were, however, carefully preserved. Perfusion of the carotid sinus and body on both sides was carried out through the common carotid arteries. The arrangement of the two Dale-Schuster pumps used for the perfusion is shown in Fig. 1. That portion of the blood perfusing the carotid bodies drained into the internal and external jugular veins (Chungcharoen, M. de B. Daly & Schweitzer, 1952) and by inference from experiments on the cat (M. de B. Daly, Lambertsen & Schweitzer, 1954) would constitute a very small proportion of the total flow passing through the carotid bifurcation. Pump *a* perfused the carotid bodies with arterial blood from a reservoir attached to the left auricle. A three-way tap *c* was inserted on the output side of the pump. The third limb of the tap was connected to the left auricle reservoir. The second pump *b* and three-way tap *d* was similarly arranged to perfuse the carotid bodies with venous blood from a reservoir connected to the right auricle. The third limb of tap *d* was connected to the right auricle reservoir. The direction of the

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taps was 180° out of phase, so that when arterial blood from the left auricle reservoir perfused the carotid bifurcations from pump *a*, the output of pump *b*, drawing its blood from the right auricle reservoir, was returned again to this reservoir. The purpose of this was to ensure that when pump *b* came to be used for perfusing venous blood through the carotid bodies it contained a representative sample of the blood in the right auricle reservoir. Furthermore, it had the advantage that blood stasis and hence the formation of vasoactive substances were minimized. The blood perfusing both carotid sinuses was returned to the circulation via the cannulated external carotid arteries and a femoral vein (Fig. 1). The carotid sinus pressure was controlled by the adjustable resistance inserted on the outflow side of the external carotid arteries (Fig. 1*e*). A change over of the perfusate from arterial to venous blood was achieved by simultaneously turning both taps *c* and *d* through 180°. When this took place, the output of the arterial pump *a* was returned to the left auricle reservoir. The taps were incorporated in a single barrel to facilitate this manipulation.

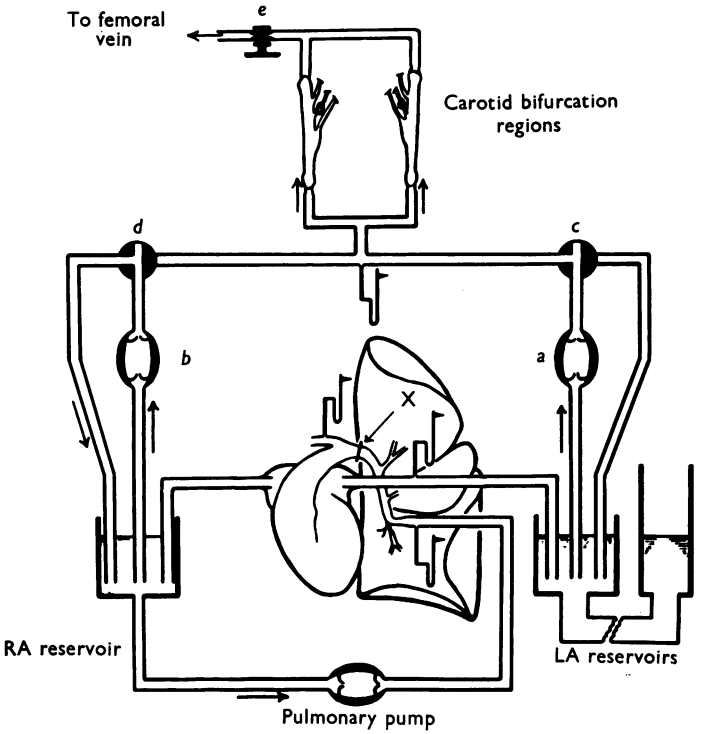

Fig. 1. Diagram showing the methods for pulmonary lobar perfusion (Daly & Daly, 1957*c*) and for perfusion of the carotid bodies with either arterial or venous blood. The right lung which receives the whole of the right ventricular output is not shown. The apical and cardiac lobes were perfused at constant volume inflow. The bronchial circulation to all lobes is normal. All lobes of both lungs are ventilated by the method of Konzett & Rössler (1940). The left pulmonary artery is ligated at X. Both carotid bifurcation regions are perfused through the common carotid arteries with either arterial blood from the left auricle reservoir by pump *a* through tap *c* or venous blood from the right auricle reservoir by pump *b* through tap *d*. When either pump is not being used for this purpose, it returns its blood to the reservoir. Blood leaving the carotid sinuses by the external carotid arteries is returned to the animal via a femoral vein after passing through the adjustable resistance *e*. For further details see text.

The output of the two pumps, each of which was approximately 150 ml./min, was initially balanced and the variable resistance in the external carotid output (Fig. 1*e*) set at a constant value so that there was no appreciable change in mean carotid sinus perfusion pressure on changing from arterial to venous perfusion of the carotid bodies. All pumps and reservoirs were water-jacketed or electrically heated to maintain the blood temperature at 36–38° C.

In some experiments reflex changes in systemic arterial pressure were compensated by connecting the central ends of the common carotid arteries to a reservoir. The pressure in this reservoir could be varied at will for the purpose of maintaining the recorded femoral arterial blood pressure constant.

Measurement of the pulmonary vascular response. Perfusion of the apical and cardiac lobes of the left lung by a Dale-Schuster pump delivering a constant blood volume inflow was carried out by the method described by Daly & Daly (1957*c*). Changes in pulmonary vascular resistance were indicated by alterations in pulmonary arterial perfusion pressure. In one experiment the technique was modified slightly in that these two lobes were perfused at a constant head of pressure. The change in pulmonary blood volume inflow during a test was measured by the pulmonary arterial overflow method of I. de B. Daly, Duke, Hebb & Weatherall (1948). It was found that venous blood stimulated the carotid bodies and caused changes in left auricular pressure. These were accurately compensated for by inserting a second reservoir connected to the left auricle reservoir (Fig. 1). This eliminated back-pressure effects on the lungs.

The pulmonary arterial perfusion pressure was measured by means of a Marey tambour and the left auricular pressure by a conventional pressure-volume recorder. Systemic arterial pressure in the femoral artery and the carotid sinus perfusion pressure were measured by mercury manometers.

In all experiments, electrical stimulation of nerves containing efferent pulmonary vasomotor fibres was carried out to show that these fibres were functionally active. For this purpose shielded platinum wire electrodes and an Attree (1950) electronic stimulator were used. To prevent current spread, nerves for stimulation were 'air-borne' and thus out of contact with adjacent tissue.

The blood was rendered incoagulable with heparin (Liquemin, Roche Products, 35–40 mg/kg). A second dog was bled from a femoral artery under local anaesthesia after premedication with morphine hydrochloride (1 mg/kg). This blood was heparinized (20–30 mg/100 ml.) and used to fill the perfusion apparatus.

RESULTS

It was found that stimulation of the carotid body chemoreceptors by the change to venous blood caused bradycardia, an increase in the rhythmic respiratory movements of the thorax and variable but usually small changes in systemic arterial pressure. Associated with these changes there was a decrease in pulmonary arterial perfusion pressure in five experiments and an increase in pulmonary arterial blood volume inflow amounting to 20% in the one experiment in which the lung lobes were perfused at constant head of pressure (Fig. 4*A*). Thus in all experiments there was a decrease in inflow resistance. These effects, shown in Figs. 2*B*, 3*A*, 4*A* and 5, were reversed when arterial perfusion of the carotid bodies was restored. In several tests the reduction in pulmonary arterial perfusion pressure occurred in the absence of any appreciable change in systemic arterial pressure (Fig. 3*A*). It will be noted that every time the perfusate was changed by turning the taps (Fig. 1*c*, *d*), there was a momentary fall in carotid sinus perfusion pressure. This was in no way responsible for the observed cardiovascular responses, for it was found

that turning the taps rapidly through 360°, thereby causing two momentary falls in carotid sinus perfusion pressure, had no effect.

Elimination of passive mechanisms. There are several mechanisms passively affecting the pulmonary vascular bed which might be the cause of the decrease in pulmonary arterial inflow resistance on stimulation of the carotid bodies. One of these is a change in left auricular pressure. This can be excluded in our

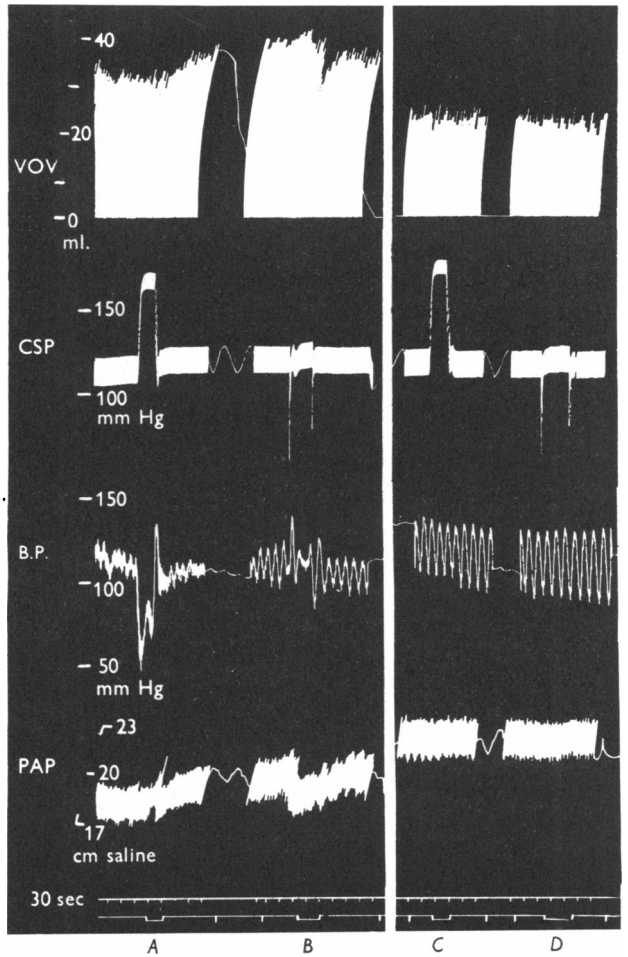

Fig. 2. Dog, ♂, 19·6 kg. Morphine-chloralose. Perfusion of the left apical and cardiac lobes at constant volume inflow. At *A* and *C*, the mean carotid sinus perfusion pressure was raised. At *B* and *D*, the carotid body chemoreceptors were stimulated by venous blood. Between *B* and *C*, the carotid sinus nerves were divided. In this and in subsequent figures: VOV = ventilation overflow volume; LAP = left auricular pressure; PAP = pulmonary arterial pressure of perfused lobes; PA overflow = pulmonary arterial overflow volume; CSP = carotid sinus perfusion pressure; B.P. = systemic arterial pressure.

experiments because the pulmonary vascular response occurred whether the left auricular pressure increased or showed no change (Fig. 4*A*). The observed decrease in pulmonary arterial perfusion pressure or increase in pulmonary arterial inflow volume must therefore be the result of a diminution in pulmonary vascular resistance.

It had been shown previously by M. de B. Daly & Schweitzer (1951) that bronchodilator effects may occur reflexly as a result of stimulation of the carotid bodies, and it was necessary therefore to make certain that passive

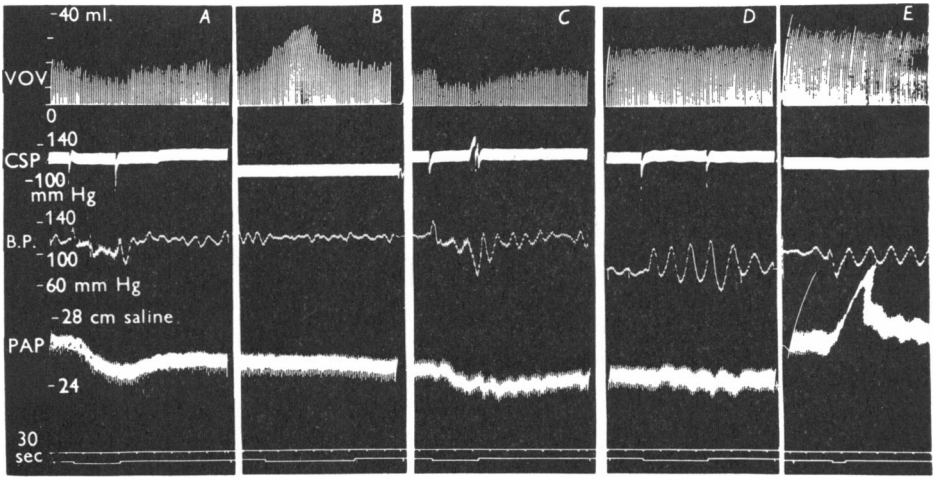

Fig. 3. Dog, ♂, 12·6 kg. Morphine-chloralose. Perfusion of the left apical and cardiac lobes at constant blood volume inflow. Respiratory pump stroke, 240 ml. *A*, *C* and *D* show the effects of stimulation of the carotid body chemoreceptors with venous blood. In *B* the peak lung inflationary pressure was reduced from 11 to 9·6 cm H_2O and then increased to 11 cm H_2O. In *E*, electrical stimulation of the caudal cut end of the left thoracic vagosympathetic nerve, 15 V, 10 msec, 50 c/s. Between *B* and *C*, decamethonium 3 mg intravenously. Between *C* and *D*, atropine 2 mg intravenously.

bronchomotor effects were not responsible for the observed changes in pulmonary vascular resistance. In the present experiments small changes (less than $\pm 3\%$) in tidal air volume sometimes occurred in response to carotid body stimulation, as indicated by the ventilation overflow volume record (Figs. 2*B*, 3*A*, 4*A*, 5). When, however, deliberate changes in peak inflationary pressure were made to produce this order of change in tidal air volume in either direction, they were found to have no passive effects on the pulmonary arterial perfusion pressure or pulmonary arterial inflow volume (Figs. 3*B*, 4*B*). Such a passive bronchomotor mechanism can, therefore, be ruled out as a cause of the changes in pulmonary vascular resistance.

It is conceivable also that the pulmonary vascular resistance might have been influenced mechanically by the accompanying reflex increase in rhythmic

movements of the thoracic cage. In most experiments this possibility was also excluded by preventing such movements with decamethonium iodide (Geigy, 0·25 mg/kg intravenously). Under these conditions stimulation of the carotid bodies still caused a fall in pulmonary vascular resistance (Fig. 3*C*). The possibility that the pulmonary vascular response to stimulation of the carotid body chemoreceptors was the result of a nervous reflex was then examined.

Effects of nerve section and of atropine

The reduction in pulmonary vascular resistance caused by stimulation of the carotid body receptors with venous blood was abolished by section of the carotid sinus nerves (Fig. 2*B*, *D*) as were the cardiovascular responses to raising the mean carotid sinus perfusion pressure (Fig. 2*A*, *C*).

It was shown further that the pulmonary vascular response to carotid body stimulation was abolished by the addition of atropine to the blood (Fig. 3*D*) and by section of the cervical vagosympathetic nerves (Fig. 4*C*). It should be stressed, however, that these results were dependent upon there being no large reflex changes in systemic arterial pressure during the tests. In contrast to the small reflex changes in systemic arterial pressure which occurred on stimulation of the carotid bodies before giving atropine or before vagal section, there was now usually a considerable reflex rise in systemic arterial pressure without any obvious change in heart rate. This systemic pressure increase may cause a passive decrease in pulmonary vascular resistance due to an alteration in volume of blood traversing the communications between the bronchial and pulmonary vascular systems (Berry & I. de B. Daly, 1931). It was essential, therefore, to ensure that no passive effects on the pulmonary vascular bed of this kind became operative when testing the responses to stimulation of the carotid bodies. For these reasons any large changes in systemic arterial pressure were compensated by the technique described in 'Methods'.

An example of a test, in which changes in systemic arterial pressure were prevented by compensation during stimulation of the carotid bodies by venous blood, is shown in Fig. 4*C*. It will be seen that after section of the cervical vagosympathetic nerves chemoreceptor stimulation had no appreciable effect on the pulmonary arterial blood volume inflow. There was, however, a small reduction in volume inflow on changing the carotid body perfusate from venous back to arterial blood, owing to the passive effect of the fall in systemic arterial pressure. Again, when a similar stimulation of the carotid bodies was carried out and the reflex rise in systemic arterial blood pressure was not compensated, the expected passive rise in pulmonary blood volume inflow took place (Fig. 4*D*). Indeed it will be noted in this record that the double rise in systemic arterial pressure is accompanied by corresponding increases in pulmonary inflow.

With regard to the possible participation of the sympathetic nervous system

in the pulmonary vascular response to carotid body stimulation, we are unable to express a definite opinion, except to mention that in one experiment the response was still present after destruction of both upper thoracic sympathetic chains and ligation of the ventral and dorsal branches of the ansa subclavia on both sides (Fig. 5).

In all experiments we tested the integrity of the efferent nerve supply to the perfused lung lobes. It was found after atropinization that electrical stimulation of the caudal end of the cut left thoracic vagosympathetic nerve caused an increase in pulmonary vascular resistance; that is, the reverse of the effect produced by stimulation of the carotid bodies by venous blood (Figs. 3*E*, 4*E*). This confirms previous work, that electrical stimulation of

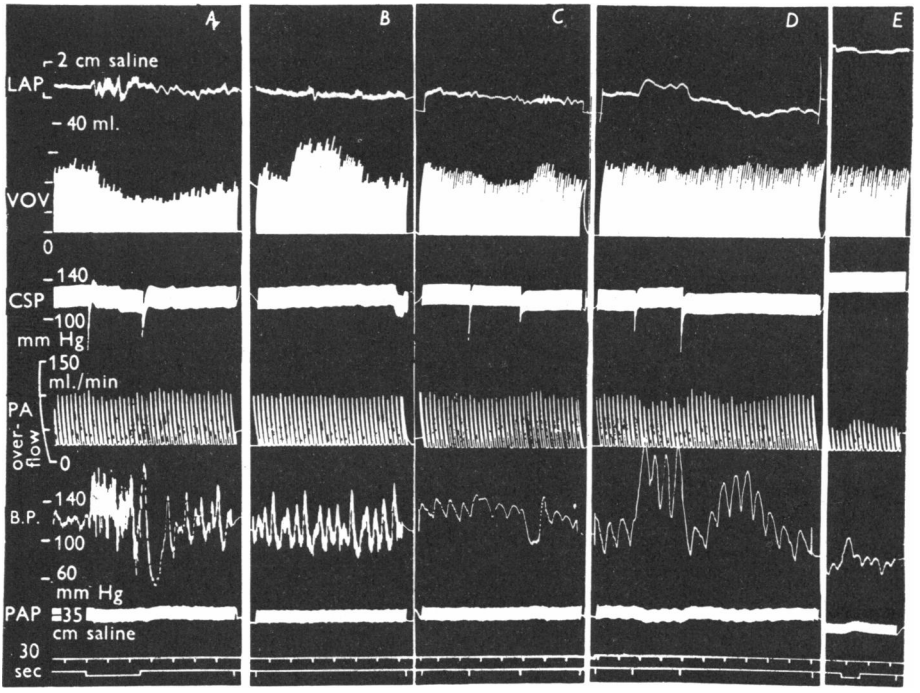

Fig. 4. Dog, ♂, 18·7 kg. Morphine-chloralose. Perfusion of the left apical and cardiac lobes at constant head of pressure. Pulmonary pump output, 200 ml./min. A reduction in pulmonary arterial overflow volume indicates an increase in pulmonary arterial inflow volume. Respiratory pump stroke, 300 ml. *A*, Stimulation of carotid chemoreceptors; left auricular pressure compensated. *B*, Respiratory pressure reduced from 12 to 11 cm H_2O and increased again to 12 cm H_2O. Between *B* and *C*, both cervical vagosympathetic nerves cut. *C*, Stimulation of carotid chemoreceptors; systemic arterial pressure and left auricular pressure compensated. *D*, Stimulation of carotid chemoreceptors, no compensation of systemic arterial pressure or left auricular pressure. *E*, Electrical stimulation of left thoracic vagosympathetic nerve, 10 V, 10 msec, 50 c/s. Note the left auricular pressure calibration represents a *change* of 2 cm saline.

nerves containing vasoconstrictor and dilator fibres to the lung blood vessels causes predominantly pulmonary vasopressor responses in atropinized lung preparations (I. de B. Daly *et al.* 1948).

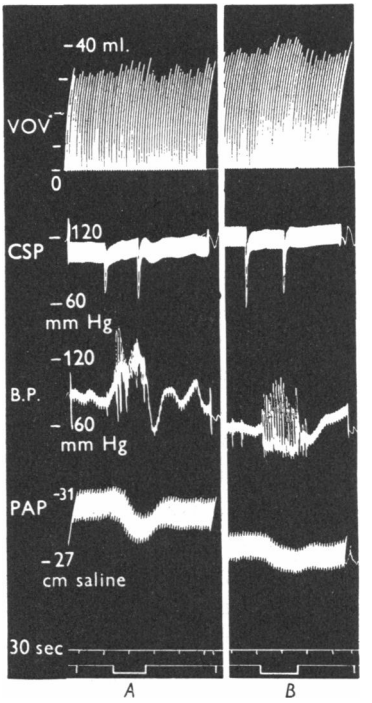

Fig. 5. Dog, ♂, 14·7 kg. Morphine-chloralose. Perfusion of left apical and cardiac lobes at constant blood volume inflow. Respiratory pump stroke, 350 ml. In *A* and *B*, the carotid body chemoreceptors were stimulated by venous blood. Between *A* and *B* the sympathetic nerves to the lungs were severed.

DISCUSSION

It has been found necessary in describing our results to enter into details of the various controls performed to eliminate passive effects on the pulmonary vascular bed. These include bronchomotor effects and those due to changes in left auricular pressure and in systemic arterial pressure. In our view we cannot overstress the importance of excluding these passive effects in examining reflexes affecting the lung blood vessels.

One feature of these experiments was the variable latent period between turning the taps (*c* and *d* in Fig. 1) to perfuse the carotid bodies with venous blood and the onset of the pulmonary vascular response. This varied in different experiments from 3 to 15 sec, and must have depended in part upon the volume output of the carotid sinus perfusion pumps, the dead space

between the taps and the carotid sinuses, and the volume flow through the carotid bodies. The last named was perhaps the only variable factor in any one experiment. Because there was little difference between the latent period of the responses of the pulmonary vascular bed, the heart rate and the systemic arterial pressure, we attribute the variable delay in onset of all these responses to these factors.

Because of the variability in this latent period it was possible that the pulmonary vascular response, instead of being due to a reflex acting directly on lung blood vessels, might have been due to an alteration in the secretion of a hormone formed elsewhere in the body. This is excluded by the fact that the time necessary for a hormone to pass through the extracorporeal circulation to the perfused lungs in our experiments exceeded 20 sec (Daly & Daly, 1957*c*) which is greatly in excess of the observed minimum latency of the responses to carotid body stimulation.

Since in our opinion all passive effects have been eliminated, it is concluded that stimulation of the carotid body chemoreceptors causes a reflex decrease in pulmonary vascular resistance which is mediated by cervical vagosympathetic cholinergic nerve fibres. In this connexion, pulmonary vasodilator fibres of this kind have been shown to exist in the cervical vagosympathetic nerve of the dog (I. de B. Daly & Hebb, 1952). But it is not possible to say whether these fibres, originally demonstrated by means of electrical stimulation of the vagus, are the same as those activated reflexly by carotid body chemoreceptor stimulation.

Passive effects affecting pulmonary vascular resistance, in particular changes in systemic (bronchial) arterial pressure, having been excluded, it is evident that the observed change in pulmonary vascular resistance to stimulation of the carotid body chemoreceptors is the result of a reflex effect on intrapulmonary blood vessels. We are, however, unable to indicate the vascular territory involved in this response. One possible mechanism is that the response is due to dilatation of the pulmonary vascular bed proper and another, that it is the result of a reflex alteration in bronchial vasomotor tone causing a redistribution of blood between the bronchial and pulmonary vascular systems (Berry & I. de B. Daly, 1931). This latter explanation does not seem unreasonable in view of the finding that the vagus nerve functionally innervates the bronchial vascular bed (Bruner & Schmidt, 1947). The decision as to which of these two explanations is the correct one can only be decided by experiments in which chemoreceptor reflexes are tested under conditions which preclude the exchange of blood between the bronchial and pulmonary vascular systems (I. de B. Daly, 1956).

The demonstration of a lung blood vessel reflex appears to us to have an added significance in that a physiological stimulus to the carotid bodies was used in our experiments.

SUMMARY

1. An investigation has been made of the effects of stimulation of the carotid body chemoreceptors upon the pulmonary vascular resistance in the chloralosed dog. Pulmonary vascular responses were measured by controlled perfusion of part of the pulmonary vascular bed. Chemoreceptors were stimulated by changing the perfusate from arterial to venous blood.

2. Under conditions in which systemic arterial pressure changes did not participate, stimulation of the chemoreceptors caused a decrease in pulmonary vascular resistance as indicated by a reduction in the pressure gradient across the pulmonary vascular bed at constant pulmonary arterial blood flow, or by an increase in pulmonary arterial blood inflow volume at constant head of pressure.

3. This response occurred independently of changes in bronchomotor tone and in left auricular pressure.

4. The decrease in pulmonary vascular resistance persisted after destruction of the sympathetic nerves to the lungs, but was abolished by section of the carotid sinus nerves. It was also abolished by section of the cervical vago-sympathetic nerves or by atropinization.

5. It is concluded that carotid chemoreceptor stimulation causes a reflex decrease in pulmonary vascular resistance which is mediated by vagal cholinergic fibres. The possible intrapulmonary vascular mechanisms responsible for this reflex response are discussed.

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