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CARDIOVASCULAR RESPONSES TO STIMULATION OF CARDIAC RECEPTORS IN THE CAT AND THEIR MODIFICATION BY CHANGES IN RESPIRATION

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SUMMARY

1. In cats anaesthetized with a mixture of chloralose and urethane, stimulation of cardiac receptors by left atrial injections of veratridine had little or no effect on pulmonary ventilation but caused bradycardia, systemic hypotension and hindlimb vasodilatation with a latency of 3·3 s.

2. The hindlimb vasodilatation was due largely, if not entirely, to a reduction in sympathetic vasoconstrictor activity.

3. Similar cardiovascular responses occurred when the arterial blood pressure was maintained constant and also in artificially ventilated animals.

4. When the cardiac receptors were excited during a period of apnoea which was induced reflexly by electrical stimulation of the central cut end of a superior laryngeal nerve, the cardio-inhibitory response to left atrial injections of veratridine was enhanced but the size of the vasodilator response was unaffected.

5. In contrast, the cardiovascular effects of stimulation of the carotid body chemoreceptors, bradycardia and hindlimb vasoconstriction were enhanced by the laryngeal input.

6. The possible central mechanism responsible for the differential modulation of cardiac receptor and carotid chemoreceptor reflexes by respiration are discussed.

INTRODUCTION

Respiratory modulation can occur in the cardiac and vascular responses elicited by stimulation of specific groups of receptors. For example, the reflex bradycardia elicited by stimulation of the carotid baroreceptors or chemoreceptors is greater when the stimuli are delivered during the expiratory phase of the respiratory cycle than during the inspiratory phase, due to the cardiac vagal motoneurones being more excitable (see Spyer, 1981, 1982, 1984; Daly, 1983, 1986; Jordan & Spyer, 1986). The vasoconstrictor reflex response resulting from stimulation of the carotid chemo-

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receptors is enhanced when the test is carried out during the expiratory phase of respiration which was reflexly induced by upper-airways stimulation (Daly, Korner, Angell-James & Oliver, 1978*c*; Daly & Kirkman, 1988*a*).

The question arises as to whether the effectiveness of inputs to the nervous system from other cardiovascular afferents is similarly modified by changes in respiration. Daly & Kirkman (1988*a*) showed recently that in contrast to the modulation of the cardiac and vascular components of the carotid chemoreceptor reflex by apnoea which was induced reflexly by excitation of a superior laryngeal nerve, the cardio-inhibitory and vasodilator effects of stimulation of pulmonary C fibres were unaffected.

In the present paper we have extended this study to an examination of the respiratory modulation of the cardiac and hindlimb vascular responses elicited by stimulation of cardiac receptors by veratridine. Previous work has shown that excitation of these receptors, while having little effect on respiration, evokes a brisk bradycardia and systemic hypotension (Bezold & Hirt, 1867; Dawes, 1947), and in the dog at least, a reduction in vascular resistance (McGregor, Hainsworth & Ford, 1986).

In the first part of this paper, we describe experiments to establish the nature of the vasodilator effect in the cat in response to stimulation of cardiac receptors by veratridine injected into the left atrium. In the second part we present the results of a study designed to establish whether the cardiovascular responses to stimulation of cardiac receptors are modified by a change in respiration. For this purpose, a comparison was made of the control responses to cardiac receptor excitation with those elicited during a period of apnoea which was reflexly induced by electrical stimulation of the central cut end of a superior laryngeal nerve.

Some of our results have been reported briefly elsewhere (Daly, Kirkman & Wood, 1987).

METHODS

Cats of either sex and varying in weight from 2·83 to 4·87 kg (for mean weights, see Table 1) were anaesthetized with an intraperitoneal injection of a mixture of 2% α-chloralose (Kuhlmann, Paris; 52 mg kg^{-1}) and 20% urethane (British Drug Houses Ltd; 520 mg kg^{-1}) dissolved in a solution containing 85 parts of sodium chloride (154 mM) and 15 parts polyethylene glycol ('Carbowax', Union Carbide Ltd).

Injections of veratridine were made into the left atrium. Two catheters were inserted through the atrial appendage which was approached via a thoracotomy in the fourth left intercostal space, during positive-pressure ventilation. The chest was then closed, the pneumothorax reduced and spontaneous respiration re-established. One catheter was used to inject veratridine, the other for the injection of vehicle alone.

Other methods used were essentially those described in detail elsewhere (Daly & Kirkman, 1988*a*). Briefly, respiration was recorded quantitatively by the bag-in-the-box technique in combination with heated respiratory valves (Bacon, Daly, Ead & Scott, 1982) and a Fleisch pneumotachograph (Godart, N.V.). The bag was filled with 100% O_2. In some experiments intermittent positive-pressure respiration was applied by means of a Starling 'Ideal' pump, the chest was opened by a medial sternotomy and the lungs collapsed passively against a resistance of up to 3 cm H_2O pressure. Chest movements were measured by a linear displacement transducer. The inspired gas mixture was approximately 35% O_2 in N_2. In some experiments the lungs were selectively denervated by the method of Daly & Scott (1958). In these the vagosympathetic nerves were divided at the level of the diaphragm before recording began to exclude abdominal afferents.

Aortic blood pressure was measured via a catheter inserted into the right femoral artery by means of a strain gauge manometer. The frequency response of the catheter manometer system was flat (±5%) up to 8 Hz. Mean blood pressure was obtained electrically. In some tests the blood pressure in the arterial baroreceptor regions was prevented from falling by controlled inflations of a balloon (Swan–Ganz monitoring double-lumen catheter, size 4F; American Edwards Laboratories, Irvine, U.S.A.), inserted into the thoracic aorta via the left femoral artery. Aortic pressure was then measured via this catheter. Mean inferior vena caval pressure was measured via a catheter inserted through the left femoral vein.

Pulse interval was measured in milliseconds from the fast-moving blood pressure record or from an electrocardiogram with the aid of a magnified graticule. Values were taken of the peak pulse interval.

The right hindlimb was vascularly isolated and perfused at constant blood flow through the right femoral artery with blood from the aorta, as described previously (Daly & Kirkman, 1988*a*). Changes in vascular resistance are proportional to the changes in (mean arterial perfusion pressure minus the mean inferior vena caval pressure). Since the vena caval pressure did not alter, the change in perfusion pressure was used as the index of a change in vascular resistance, and was taken as the difference between the control value and the peak of the response.

The carotid body chemoreceptors were stimulated in two ways. In some experiments small doses of sodium cyanide (0·01%–0·1%, w/v) in sodium chloride solution, 154 mM, were injected into a common carotid artery via a catheter inserted into the lingual artery in volumes less than 0·2 ml. Control injections of the same volume of sodium chloride solution (154 mM) had no respiratory or cardiovascular effect. In other experiments, the carotid bodies were stimulated with hypoxic hypercapnic blood. For this purpose, both carotid bifurcation regions were vascularly isolated by ligation of the occipital and ascending pharyngeal arteries and the vestigial remnants of the internal carotid arteries. The venous drainage from the carotid bodies was preserved. Pulsatile flow perfusion with blood from a carotid artery was carried out through the rostral ends of the common carotid arteries using a micropump described previously (Daly, Ead & Scott, 1978*a*). Blood was returned to the right atrium from the carotid bifurcation regions via the cannulated external carotid arteries and a Starling-type resistance by which the carotid sinus pressure was controlled and maintained constant. The perfusion system was similar to that described previously (Daly, Korner, Angell-James & Oliver, 1978*b*) except that the carotid bodies were stimulated by temporarily substituting venous blood obtained from the inferior vena cava instead of from a disc equilibrator. The extracorporeal perfusion circuit, volume about 5 ml, was primed with heparinized blood obtained from a second animal.

A superior laryngeal nerve on one side was dissected, cut close to the larynx, and its central end mounted on platinum wire electrodes. It was stimulated electrically with pulses of 2–4 V, 2 ms duration at a frequency of 30–50 Hz (Grass S88 stimulator), through a stimulus isolation unit.

In all experiments Penicillin (Crystapen, Glaxo Laboratories Ltd; 500000 units, I.V.) was given routinely at the beginning of each experiment, and the urinary bladder was catheterized suprapubically and continually drained to prevent reflexes arising from this organ (Daly, Ward & Wood, 1986). When the surgical procedures were completed, heparin (Monoparin, C. P. Pharmaceuticals Ltd, 2000 i.u. kg^{-1} and supplementary doses of 500 i.u. kg^{-1} hourly, intravenously) was given to prevent clotting of the blood.

All variables were recorded on a direct-writing ultraviolet light recorder (S. E. Laboratories Ltd, Feltham, Middlesex, U.K.). Zero pressures were established post-mortem by exposing to air the tips of the catheters *in situ*.

Blood gas analysis. Samples of arterial blood and of the venous blood used to stimulate the carotid bodies were drawn anaerobically into oiled glass syringes and analysed immediately. The P_{O_2}, P_{CO_2} and pH of the blood were determined using a calibrated electrode system (Model 413, Instrumentation Laboratories (U.K.) Ltd). Estimations were made at a temperature of 37·5 °C. Metabolic acidosis was corrected by an infusion of a mixture of 1 M-sodium bicarbonate solution (1 part) and sodium chloride (154 mM; 6 parts). Packed cell volume was determined by centrifuging samples of blood in capillary haematocrit tubes for 5 min at 10000 *g*.

Drugs. The following drugs were used: veratridine free base (Sigma Chemical Co.), atropine sulphate (BDH Chemicals Ltd), propranolol hydrochloride (Inderal, Imperial Chemical Industries Ltd), guanethidine (Ismelin, Ciba Laboratories Ltd), Val-angiotension II (Sigma Chemical Co. Inc.) and isoprenaline sulphate (Martindale Samoore Ltd).

Statistical analysis. Data for controls and experimental values are expressed as means ± S.E.M. unless otherwise stated. Where appropriate, Student's t test was used to evaluate the significance of the differences between sets of paired data. Values of $P < 0.05$ were taken as significant.

RESULTS

Effects of stimulation of cardiac receptors by veratridine

The initial values for respiratory, cardiovascular and arterial blood gas variables are shown in Table 1 for spontaneously breathing animals and animals which were artificially ventilated.

TABLE 1. Initial control values for respiration and cardiovascular variables

	Spontaneous ventilation	Positive-pressure ventilation
No. of animals	13	11
Body weight (kg)	3·92 ± 0·71	3·62 ± 0·43
$\dot{V}_I$ (l min^{-1} kg^{-1})	0·183 ± 0·04	—
Heart rate (beats min^{-1})	198·4 ± 31	184·1 ± 27·1
Mean BP (mmHg)	125·8 ± 21·7	120·5 ± 18·8
P_{limb} (mmHg)	115·6 ± 17·6	110·2 ± 11·6
P_{IVC} (mmHg)	4 ± 2	5 ± 1
Arterial blood		
P_{O_2} (mmHg)	230·6 ± 86·7	129·5 ± 13·6
P_{CO_2} (mmHg)	35·3 ± 4·5	34·2 ± 1·7
pH	7·435 ± 0·055	7·478 ± 0·057
Hct (%)	40·9 ± 5	39·9 ± 2·7
Rectal temperature (°C)	37·6 ± 0·8	37·5 ± 0·6

Values are means ± S.D. $\dot{V}_I$, respiratory minute volume; BP, arterial blood pressure; P_{limb}, hindlimb perfusion pressure; P_{IVC}, inferior vena caval pressure.

The dose of veratridine that was chosen in each experiment was the one that approximately doubled the pulse interval at the peak of the cardiac response.

The results are summarized in Fig. 1. In seven spontaneously breathing animals, sixteen injections of 1·42–5·73 μg kg^{-1} (mean 2·68 ± 0·36 μg kg^{-1}) into the left atrium caused a small reduction in respiration ($P < 0.005$), bradycardia ($P < 0.0001$; onset latency, 3·3 ± 0·2 s), a fall in arterial blood pressure ($P < 0.0001$, latency 3·6 ± 0·2 s) and in hindlimb perfusion pressure ($P < 0.0001$; latency 4·2 ± 0·2 s) (Fig. 1, left-hand panel). Since the hindlimb blood flow was maintained constant and the inferior vena caval pressure did not change, the fall in perfusion pressure indicates vasodilatation (Fig. 1, left-hand panel). Qualitatively similar responses occurred in animals in which the arterial blood pressure was maintained constant to control the baroreceptor input (Fig. 1, right-hand panel) and artificially ventilated animals (middle panel). The results of the three groups of experiments depicted in Fig. 1 should not be compared quantitatively since different doses of veratridine were used.

The receptors stimulated by veratridine lie in the heart because the responses were unaffected by cutting the aortic nerves and by selective denervation of the lungs (two animals), but were subsequently abolished by division of the cervical vago-sympathetic nerves (six animals).

The cardio-inhibitory responses to veratridine were vagal in origin, being unaffected by propranolol (1 mg kg^{-1} I.V.; three animals) and abolished by atropine (1 mg I.V.; one animal).

The vasodilator responses were due largely to withdrawal of sympathetic vasoconstrictor activity since they still occurred after the administration of atropine

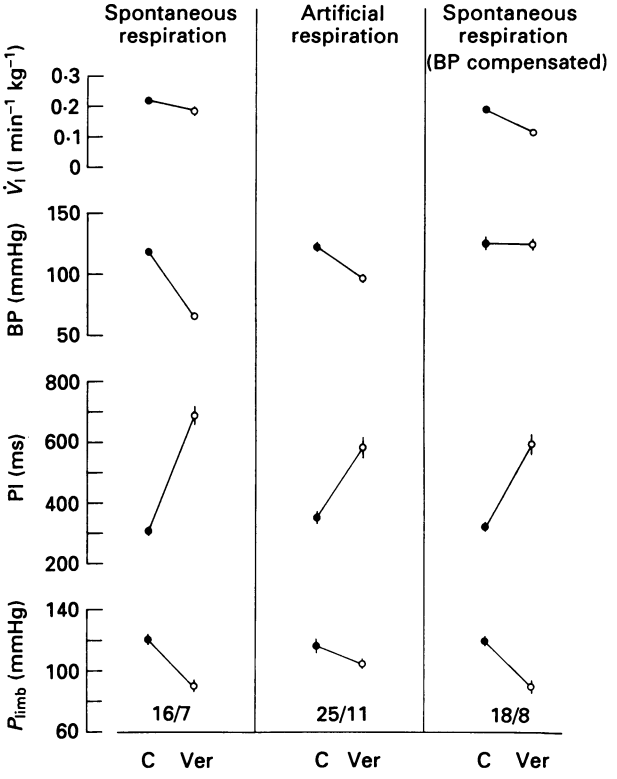

Fig. 1. Summary of the effect of left atrial injections of veratridine on respiratory minute volume ($\dot{V}_I$), mean arterial blood pressure (BP), pulse interval (PI) and hindlimb perfusion pressure (P_{limb}) under three experimental conditions: spontaneously breathing animals, artificially ventilated animals, and spontaneously breathing animals with arterial blood pressure compensated. ●, control values (C); ○, peak values after injection of veratridine (Ver). Values are means ± S.E.M. Where there is no standard error bar it is smaller than the size of the symbol. The figures at the foot of each panel are the numbers of tests/animals.

(1 mg I.V.; two animals) or propranolol (1 mg kg^{-1} I.V.; three animals). Furthermore they were abolished by guanethidine (1·6 mg kg^{-1} I.V.; three animals) and considerably reduced by division of the right sciatic, femoral and obturator nerves.

The reduction, or abolition, of the responses to veratridine were not due to loss of vascular 'tone'. Guanethidine itself did not alter the background vascular tone as demonstrated by close arterial injections of 5 ng isoprenaline. Following denervation of the limb, the perfusion pressure fell and was restored to its original level by a close arterial infusion of angiotensin II, 6–40 ng min^{-1}; the averaged fall in perfusion

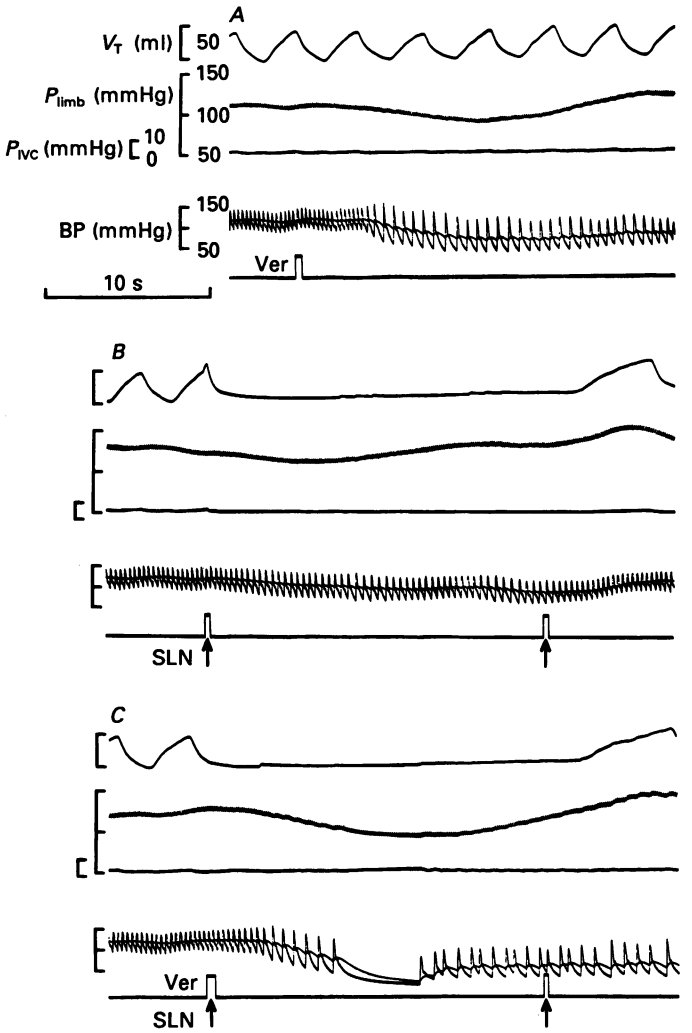

Fig. 2. The effects of a reflexly induced apnoea on the cardiac and vascular responses to veratridine, 4·3 μg kg^{-1}, injected into the left atrium of the spontaneously breathing cat. *A*, control injection of veratridine (Ver); *B*, electrical stimulation of the right superior laryngeal nerve (SLN), 3 V, 2 ms, 30 Hz, between arrows; *C*, first signal and arrow, simultaneous injection of veratridine and start of electrical stimulus to superior laryngeal nerve. Second signal and arrow, cessation of electrical stimulus. Records from above downwards: V_T, tidal volume (inspiration upwards); P_{limb}, hindlimb perfusion pressure; P_{IVC}, inferior vena caval pressure; BP, phasic and mean arterial blood pressure. Time calibration, 10 s. The values for the calibrations in *B* and *C* are the same as those in *A*.

pressure elicited by 5 ng isoprenaline was 31·7 mmHg compared to 25·0 mmHg before denervation (two animals).

Stimulation of cardiac receptors during excitation of a superior laryngeal nerve

In seven animals the respiratory and cardiovascular effects of excitation of cardiac receptors were compared with those elicited during a 20 s period of apnoea produced

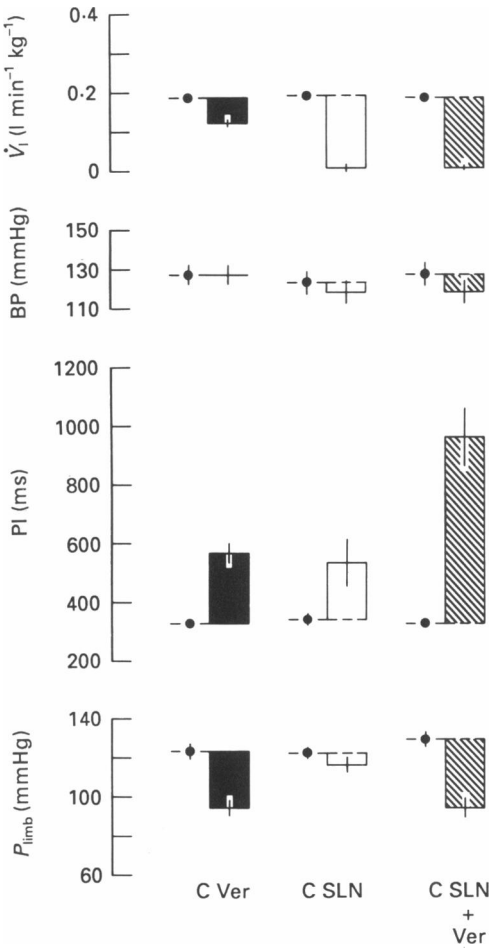

Fig. 3. The respiratory and cardiovascular responses to left atrial injections of $2{\cdot}42 \pm 0{\cdot}21$ μg kg^{-1} veratridine alone and during an apnoeic period produced by electrical stimulation of a superior laryngeal nerve, 3 V, 2 ms duration, 30 Hz. Arterial blood pressure maintained constant. C, control values before injection or stimulation. Left panel, responses to veratridine (Ver) alone; middle panel, stimulation of a superior laryngeal nerve (SLN) alone; right panel, combination of stimulation of a superior laryngeal nerve and injection of veratridine. $\dot{V}_I$, respiratory minute volume; BP, mean arterial blood pressure; PI, pulse interval; P_{limb}, hindlimb perfusion pressure. Values are means $\pm$ S.E.M. (thirteen series of tests in seven animals).

reflexly by electrical stimulation of the central cut end of a superior laryngeal nerve. In thirteen series of tests in seven animals the arterial pressure was maintained constant to exclude the participation of baroreceptor reflexes. All data refer to time-matched points. An example of one typical series of responses is shown in Fig. 2 and the averaged results are summarized in Fig. 3.

Cardiac receptors. Left atrial injections of veratridine, 1·17–3·25 μg kg^{-1} (mean $2{\cdot}42 \pm 0{\cdot}21$ μg kg^{-1}), caused a small reduction in respiratory minute volume of $0{\cdot}063 \pm 0{\cdot}01$ l min^{-1} kg^{-1} from a control value of $0{\cdot}183 \pm 0{\cdot}008$ l min^{-1} kg^{-1} ($P < 0{\cdot}0005$). Pulse interval increased by 242 ± 38 ms from a control value of 328 ± 17 ms

($P < 0{\cdot}0005$), while the hindlimb perfusion pressure fell $29{\cdot}6 \pm 4{\cdot}3$ mmHg from a control value of $123{\cdot}3 \pm 3{\cdot}7$ mmHg ($P < 0{\cdot}0001$) (Figs 2 and 3).

Superior laryngeal nerve. When the superior laryngeal nerve was stimulated, apnoea occurred in all tests accompanied by an increase in pulse interval of 197 ± 75 ms, control value 339 ± 19 ms, and a small reduction in hindlimb perfusion pressure of $6{\cdot}3 \pm 3{\cdot}0$ mmHg, the control value being $122{\cdot}3 \pm 3{\cdot}0$ mmHg ($P < 0{\cdot}05$)(Figs 2 and 3).

Cardiac receptors and superior laryngeal nerve in combination. The results are summarized in Fig. 3 (right-hand panel). When the same dose of veratridine was injected during the period of apnoea elicited by electrical stimulation of a superior laryngeal nerve, the apnoea persisted while the size of the cardio-inhibitory response was increased compared with that occurring on injection of veratridine alone. The pulse interval during stimulation of a superior laryngeal nerve was 534 ± 82 ms and the superimposed stimulus provided by the left atrial injections of veratridine increased this value to 962 ± 105 ms ($P < 0{\cdot}0005$). This represents a veratridine-induced increase in pulse interval of 428 ± 890 ms and compares with the value of 242 ± 38 ms when veratridine was injected alone. A paired statistical analysis of these data indicates this difference is significant ($P < 0{\cdot}05$).

In six of the thirteen series of paired observations the cardio-inhibitory response to veratridine was potentiated by the laryngeal stimulus without change in the control levels of pulse interval, indicating an interaction between the two inputs (Fig. 4). In ten of them the cardio-inhibitory response to combined stimulation of the cardiac receptor and laryngeal inputs was greater than the sum of their separate effects (Fig. 4).

During stimulation of the superior laryngeal nerve, veratridine caused a fall in hindlimb perfusion pressure of $22{\cdot}2 \pm 4{\cdot}4$ mmHg, from $116{\cdot}0 \pm 3{\cdot}7$ mmHg ($P < 0{\cdot}001$), which is not significantly different from the value of $29{\cdot}6 \pm 4{\cdot}3$ mmHg elicited by veratridine injections alone ($0{\cdot}1 > P > 0{\cdot}05$) (Fig. 3). The results of individual tests are shown in Fig. 4 which indicates that stimulation of the superior laryngeal nerve reduced the vasodilator response to veratridine in eight of thirteen tests. This result could not be attributed to the inability of the vascular bed to dilate further as demonstrated by using larger doses of veratridine. In three tests the size of the vasodilator response was increased, while in the remaining two tests, it was unchanged.

Thus, in the majority of tests there was a tendency for the vasodilator responses to veratridine to be reduced when superimposed on a reflex apnoea.

Stimulation of the carotid chemoreceptors during excitation of a superior laryngeal nerve

The present results are in contrast to previous findings involving reflexes from the carotid body chemoreceptors. In these experiments apnoea induced reflexly considerably enhanced both the cardio-inhibitory and vasoconstrictor responses resulting from excitation of the chemoreceptors in the cat, dog and monkey (Angell-James & Daly, 1973, 1978; Daly *et al.* 1978*c*; Daly & Kirkman, 1988*a*). It was therefore necessary to confirm these findings in the present experiments and to test the effects of the laryngeal input on the cardiovascular responses to stimulation of cardiac receptors in the same animals on a paired basis.

The results of six paired sequences of tests in the same five animals are shown in Fig. 5*A* and *B*. In *A* are depicted the results of left atrial injections of $2{\cdot}8 \pm 0{\cdot}16$ $\mu g\ kg^{-1}$ veratridine alone and during electrical stimulation of a superior laryngeal nerve, and are qualitatively similar to those described above (Fig. 3). In *B* are shown the comparable results of stimulations of the carotid body by intracarotid injections of sodium cyanide, $6{\cdot}3 \pm 0{\cdot}39$ $\mu g\ kg^{-1}$. Whereas stimulation of the carotid chemo-

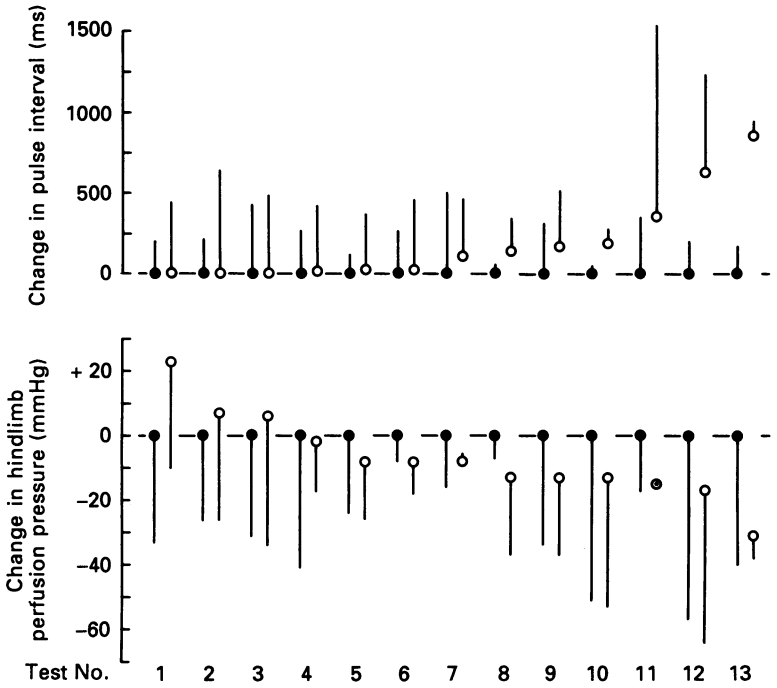

Fig. 4. The effects on pulse interval and hindlimb perfusion pressure of stimulation of cardiac receptors by veratridine alone (●, pre-injection control values; end of attached vertical lines, injection test values) and during electrical stimulation of the central end of a superior laryngeal nerve (○, values during laryngeal input alone; end of attached vertical lines, values during combination of superior laryngeal nerve stimulation and veratridine injection). The arterial blood pressure was maintained constant. The observations from seven animals are arranged in ascending order of magnitude of the pulse interval and hindlimb perfusion pressure responses to electrical stimulation of a superior laryngeal nerve.

receptors increased respiratory minute volume, the same stimulus applied during the apnoea produced by stimulation of a superior laryngeal nerve had no effect on respiration, confirming previous observations (Angell-James & Daly, 1973, 1978; Elsner, Angell-James & Daly, 1977; Daly *et al.* 1978*c*; Daly & Kirkman, 1988*a*). The laryngeal input itself caused only a small increase in pulse interval of 60 ± 23 ms from a control value of 290 ± 6 ms ($P < 0{\cdot}05$), and had no effect on hindlimb perfusion pressure but it enhanced in each of the six series of tests both the cardio-inhibitory ($P < 0{\cdot}005$) and the vasoconstrictor ($P < 0{\cdot}05$) response to stimulation of the carotid chemoreceptors.

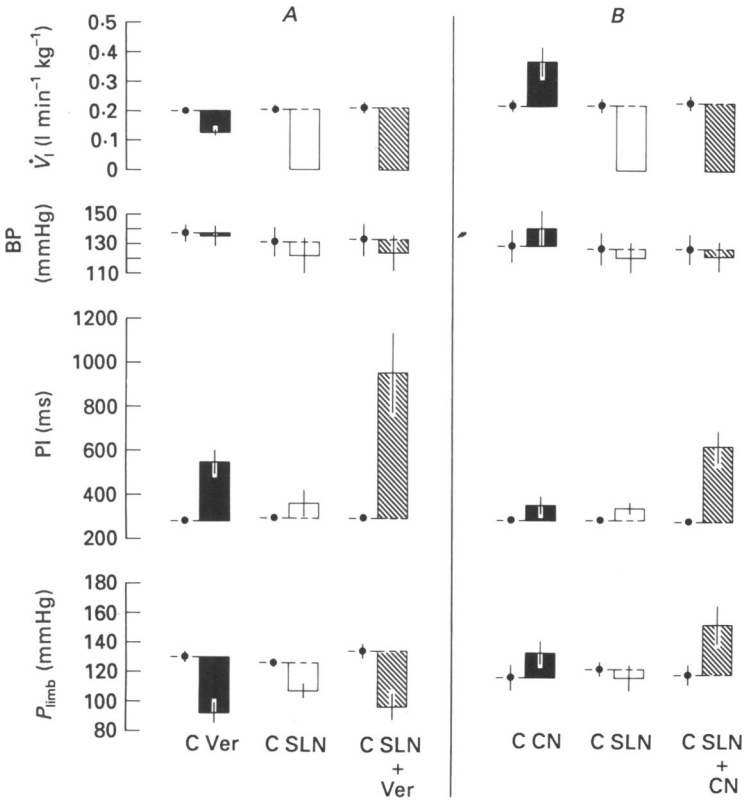

Fig. 5. Comparison of the effects of the laryngeal input on the respiratory and cardiovascular responses to stimulation of cardiac receptors and carotid body chemoreceptors. *A*, effects of left atrial injections of veratridine (Ver) alone ($2{\cdot}8 \pm 0{\cdot}16$ μg kg^{-1}), electrical stimulation of a superior laryngeal nerve (SLN) alone, and in combination (SLN + Ver). *B*, effects of intracarotid injections of sodium cyanide (CN) alone ($6{\cdot}33 \pm 0{\cdot}39$ μg kg^{-1}), electrical stimulation of a superior laryngeal nerve (SLN) alone, and in combination (SLN + CN). *C*, control values. Paired data (peak values, mean ± S.E.M.) from six series of tests in five animals. Where the standard error bar is absent, it is smaller than the size of the symbol. $\dot{V}_I$, respiratory minute volume; BP, mean arterial blood pressure; PI, pulse interval; P_{limb}, hindlimb perfusion pressure.

Thus whereas the laryngeal input potentiated the cardio-inhibitory responses to stimulation of the cardiac receptors and carotid chemoreceptors, the vascular responses were potentiated only during carotid chemoreceptor stimulation.

DISCUSSION

Our results confirm previous studies indicating that stimulation of cardiac receptors by veratridine reflexly causes a small reduction in respiration, hypotension and bradycardia (Bezold & Hirt, 1867; Dawes, 1947). We have also shown in the cat, as in the dog (McGregor *et al.* 1986), that these responses are accompanied by vasodilatation in the hindlimb, the efferent pathway being mediated largely, if not

entirely, by virtue of a reduction in activity in sympathetic vasoconstrictor fibres. Bergel & Makin (1967), on the other hand, found that the hindlimb vasodilator response to stimulation of epicardial receptors by injecting nicotine into the pericardial sac was much reduced by atropine and suggested a cholinergic mechanism in skeletal muscle was involved. Their Fig. 6, however, shows that after atropine epicardial receptor stimulation still caused an appreciable femoral vasodilatation as indicated by the fact that at the nadir of the 36 mmHg fall in arterial blood pressure, the level of femoral mean blood flow had not altered. In the present experiments there were small differences in the size of the vasodilator responses before and after atropine but the number of observations was too small to enable us to determine their significance. Thus, while we cannot rule out the participation of a cholinergic mechanism, it clearly cannot be the whole explanation. A change in the secretion of suprarenal catecholamines is unlikely to have made any important contribution to the vascular responses in view of their short latency of onset, 4·2 s, compared with the time of about 18 s required for the secreted hormones to reach the perfused hindlimb, and the fact that they were unaffected by propranolol.

With regard to the afferent pathway, all responses were abolished by division of the cervical vagus nerves, the aortic nerves having already been cut. Afferent fibres running in the sympathetic nerves (Malliani, 1982) do not constitute, therefore, an important pathway.

The new findings reported in this paper concern the nature of the respiratory modulation of the cardiac and vascular responses elicited by stimulation of cardiac receptors. In the spontaneously breathing animal heart rate and the activity in cardiac vagal efferent fibres are subject to respiratory modulation provided there is a sufficient degree of cardiac vagal 'tone'. During the inspiratory phase of respiration there is tachycardia and a reduction or cessation of cardiac vagal efferent activity due to a combination of activity of the central inspiratory neurones and an increase in discharge in afferent fibres from intrapulmonary receptors excited by lung inflation (Anrep, Pascual & Rössler, 1936*a*, *b*; also Spyer, 1982, 1984; Daly, 1986). Through the same mechanisms the excitability of at least some cardiac vagal motoneurones alters in phase with respiration and in consequence so does the effectiveness of the excitatory inputs from baroreceptors and chemoreceptors onto these motoneurones. During the inspiratory phase of the respiratory cycle the cardiac vagal motoneurones are refractory to such inputs, the full expression of their reflex effects being evident only during the expiratory phase of the cycle (see Spyer, 1984; Daly, 1986). In keeping with these findings is the observation that the primary reflex bradycardia resulting from stimulation of the carotid bodies is considerably potentiated when the test is repeated during a reflexly induced apnoea in the expiratory position by stimulation of upper-airways receptors, including electrical stimulation of the central cut end of a superior laryngeal nerve (Angell-James & Daly, 1975; Elsner *et al.* 1977; Daly *et al.* 1978*c*; Daly & Kirkman, 1988*a*). The present experiments have not only confirmed these observations but have shown further that the cardio-inhibitory responses resulting from stimulation of cardiac receptors are similarly affected.

The cardiac vagal motoneurones can be activated synaptically by intravenous injections of veratridine (Lipski, McAllen & Spyer, 1975) and it would be reasonable

to assume therefore that the mechanisms whereby the cardio-inhibitory response to an excitatory input from cardiac receptors is respiratory modulated are similar to those involving inputs from chemoreceptors and baroreceptors. There is as yet, however, no information on the possible role of central inspiratory activity. With regard to the inhibitory effects of lung inflation, recent studies have shown that the cardio-inhibitory responses to stimulation of cardiac receptors are modulated by distension of the lungs to the same extent quantitatively as those from excitation of arterial baroreceptors (Daly & Kirkman, 1988*b*).

The results presented here on the respiratory modulation of the chronotropic response to stimulation of cardiac receptors might have been anticipated in view of the previous observations, cited above, showing that the excitatory inputs from chemoreceptors and baroreceptors are affected in a similar way by the laryngeal input. Nevertheless, it is not possible to draw a general conclusion that all such excitatory inputs from cardiovascular receptors are affected in a similar way. Daly & Kirkman (1988*a*) showed that under similar experimental conditions the reflex bradycardia resulting from stimulation of pulmonary C fibres was not consistently subject to respiratory modulation although in the same animals the excitatory input from carotid chemoreceptors was. Further studies by Daly & Kirkman (1988*b*) demonstrated that there was also a differential modulation of the cardio-inhibitory responses to stimulation of various cardiovascular receptors by a pulmonary input, driven by lung inflation. Whereas lung inflation with volumes of 24 ml kg^{-1} almost completely suppressed the bradycardia resulting from carotid chemoreceptor stimulation, it only partly inhibited the response to cardiac receptor and arterial baroreceptor excitation, and had no effect, or slightly increased, that elicited by stimulation of pulmonary C fibres. In this connection, Potter (1981) demonstrated that with the carotid sinus baroreceptor reflex there were differences in the mechanisms of inhibition by central inspiratory activity and lung inflation, and suggested that the latter exerted its influences earlier in the reflex pathway, although not directly on the baroreceptor afferent terminals (Jordan & Spyer, 1979). Further experiments are required to establish more clearly the complexities of the pathways whereby excitatory inputs from cardiovascular receptors are affected by central and reflex respiratory mechanisms.

We found here that the reflex vasodilatation elicited by veratridine was unaffected by apnoea induced by stimulation of the laryngeal input. This was not due to the vascular bed being fully dilated because larger doses of veratridine resulted in increased vasodilator responses. During normal respiration there is an oscillation of activity in sympathetic nerves in phase with the central respiratory cycle which has a maximum in the mid-inspiratory phase of the respiratory cycle and a minimum in the early expiratory phase (see Daly, 1986; also Koepchen & Thurau, 1958; Gilbey, Numao & Spyer, 1986), and is synchronous with oscillations in hindlimb (muscle) vascular resistance (Koepchen, Seller, Polsher & Langhorst, 1968). This pattern of firing is thought to be due to an excitatory synaptic input from brain stem inspiratory neurones (Preiss, Kischner & Polosa, 1975; Barman & Gebber, 1976; Gerber & Polosa, 1978) or to a common oscillator which drives both the phrenic and sympathetic discharges (Bachoo & Polosa, 1987); it can be suppressed by activation of pulmonary stretch receptors which inhibit inspiratory neurones (Gerber & Polosa,

1978; Gootman, Feldman & Cohen, 1980). How the respiratory integration of sympathetic effects of cardiac receptor and carotid chemoreceptor stimulation takes place is still unknown. Nevertheless, there is selectivity in the way inputs from chemoreceptors, baroreceptors and urinary bladder receptors are affected by pulmonary stretch receptor activity (Daly *et al.* 1986). Excitation of the laryngeal input, while leaving unaffected the hindlimb vasodilator response to stimulation of cardiac receptors (this paper) and pulmonary C fibres (Daly & Kirkman, 1988*a*), potentiates the vasoconstrictor effects of excitation of the carotid chemoreceptors (Daly & Kirkman, 1988*a*; this paper), indicating selectivity in the organization of the control of the sympathetic output.

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