J. Physiol. (1978), **282**, *pp.* 353–364
With 1 plate and 4 text-figures
Printed in Great Britain

TWO TYPES OF VAGAL PREGANGLIONIC MOTONEURONES PROJECTING TO THE HEART AND LUNGS

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(*Received 8 February 1978*)

SUMMARY

1. A study has been made of eighty-four cells in the cat's nucleus ambiguus whose axons projected to the cardiac (seventy-four) and pulmonary (ten) branches of the right vagus. Their axonal conduction velocities were all in the range of B fibres (2·8–15·5 m/sec).

2. Pulmonary branch projecting neurones were usually spontaneously active (nine out of ten) and fired in phase with inspiration. Their activity showed no pulse modulation.

3. Ten cardiac branch projecting neurones had properties indistinguishable from those of pulmonary branch projecting neurones. Inspiratory-firing cells projecting to either branch are believed to be bronchoconstrictor in function.

4. The remaining sixty-four cells that projected to the cardiac branch had properties expected of cardioinhibitory neurones. Most (fifty-four) were silent until activated by ionophoresis of excitant amino acids. All showed an expiratory discharge when active, and of twenty-seven tested twenty-three showed a cardiac modulation of their discharge. When the aortic baroreceptors were denervated, the cardiac rhythm was always abolished reversibly by carotid occlusion.

5. Ionophoretic activation of expiratory firing (presumed cardioinhibitory) cells slowed the heart (fifteen out of eighteen neurones tested). Excited inspiratory-firing cells never had this effect (eleven tested).

6. Both types of neurones were found in the nucleus ambiguus, but presumed cardioinhibitory cells tended to be found more caudally and ventrally than presumed bronchoconstrictor neurones.

INTRODUCTION

In a previous paper (McAllen & Spyer, 1976) we described a population of neurones in the cat's nucleus ambiguus whose axons were found in the cardiac branches of the right vagus. From several lines of evidence, we concluded that they were likely to have a cardioinhibitory function. However, as few were spontaneously active under our experimental conditions, we could not often confirm that they behaved in a manner appropriate to cardioinhibitory neurones. Nor were we able to measure the prevalence in that population of bronchial efferent neurones, which we considered to be most likely contaminants of the sample.

In the present series of experiments, we have used excitant amino acids (applied

ionophoretically) to raise the excitability of vagal motoneurones. The resultant firing patterns have been used to infer their synaptic connexions and to provide a physiological basis on which to classify them. Most of the sample showed properties expected of cardioinhibitory neurones, which we describe in this and the accompanying paper (McAllen & Spyer, 1978). A minority of cells which we consider likely to be bronchoconstrictor in function, showed distinct properties that were apparently identical to those of other nucleus ambiguus cells whose axons projected to the pulmonary vagal branches.

Additional information has been sought on the efferent connexions of presumed cardioinhibitory cells. In the present experiments the cardiac branches were preserved intact, and changes in heart-rate could be measured while such neurones were ionophoretically activated. A preliminary report of part of this work has been published (McAllen & Spyer, 1977).

METHODS

Experiments were performed on thirty-two female cats (2–3 kg body wt.) anaesthetized with α-chloralose (BDH, 70 mg/kg I.V.) after induction with ethyl chloride and ether. Supplementary doses of anaesthetic were given, if and when necessary, as a small dose of either chloralose or sodium pentobarbitone (Sagatal, 2 mg/kg I.V.). In all experiments the trachea was cannulated low in the neck, and cannulae placed in a femoral artery and vein for the measurement of arterial blood pressure and the administration of anaesthetic respectively.

The right phrenic nerve was isolated in the neck and cut peripherally. Its activity was then recorded using bipolar electrodes. In six animals the aortic baroreceptors were denervated by cutting the right aortic nerve and left cervical vagus. In all experiments snares were placed around both common arteries and the external carotid arteries were ligated.

The animal was then placed in a stereotaxic head holder and laid on its left side. The chest was opened between the right fourth and fifth ribs and the animal was then ventilated by means of a respiratory pump (Harvard) which was connected also to an expiratory resistance of 0·5–1·5 cm H_2O to prevent complete collapse of the lungs. The ventilatory minute volume was adjusted to maintain the end-tidal CO_2 at approximately 4 %, this being monitored continuously using a Beckman (LB-1) CO_2 meter. Arterial blood samples were taken from time to time through a cannula in the second femoral artery for blood gas analysis, and arterial HCO_3^- concentration was maintained at around 23 m-equiv/100 ml. blood by I.V. injections of sodium bicarbonate solution. The animal's temperature was maintained between 37 and 38 °C using an electric blanket controlled by a feedback circuit.

Identification of cardiac branches

The cardiac branches of the right thoracic vagus were exposed and identified as described previously (McAllen & Spyer, 1976). Once their ability to slow the heart was confirmed, these nerves were placed in a sleeve electrode made from a slit polythene tube hooked through by a pair of silver wires which served as stimulating electrodes. This arrangement was left *in situ*, being attached to underlying tissues using contact adhesive (Permabond 102) after covering and isolating with petroleum jelly. The threshold for the efferent effect on heart rate was then measured by stimulating the cardiac branch (or branches) with 0·1 msec pulses at 75 Hz. The range of voltage over which additional cardioinhibitory fibres could be recruited was assessed by stimulating at 3 Hz with step increases in voltage. This range was assessed from time to time during the experiment and set the primary criterion for acceptance of a cardiac efferent responding antidromically to that branch (or branches). We could not test the effects of stimulating the cardiac branches on bronchomotor tone since (i) the nearest lung lobe was ligated and (ii) high frequency stimulation is required to elicit a measurable change in airway resistance and this would have stopped the heart.

Identification and isolation of pulmonary branches

It has been reported previously (McAllen & Spyer, 1976) that the majority of bronchoconstrictor fibres in the right vagus leave the main thoracic vagus below the azygos vein. However, it was often observed that small filaments projected from the cardiac branches towards the stump of the excised upper lobe of the right lung (see also Fig. 1. of McAllen & Spyer, 1976). Caudal to the azygos vein it was possible to identify the major pulmonary branches passing towards the lungs. Respiratory pressure was measured from a side tube attached to the tracheal cannula, respiratory airflow being monitored using a Fleisch Pneumotachograph head attached to a Grass PT5A differential pressure transducer. Changes in airway resistance evoked on stimulating pulmonary branches were seen as either changes in tracheal pressure or as a change in slope when tracheal pressure was plotted against airflow using an $X-Y$ plotter. The effects of stimulating these branches was also tested on oesophageal contractions (monitored from a saline-filled balloon attached to a pressure transducer and lowered through the mouth into the oesophagus) and changes in heart rate. Stimulation at 100 Hz with 0·1 msec pulses evoked changes in airway resistance but left the heart and oesophagus unaffected.

In a separate group of experiments we verified that the vagal efferent fibres responsible for bronchoconstriction had conduction velocities in the range of those of B fibres. The cervical vagus was cut at the level of the nodose ganglion, the distal portion being placed on two pairs of silver wire electrodes, one pair for stimulation, the other for monitoring the evoked neural volley. The electroneurogram to stimulation with 0·1 msec pulses at 2 Hz was monitored. As a new component was recruited, its effect on airway resistance was monitored as described above, stimulating then at 100 Hz. Bronchoconstriction was only produced when B fibres were activated, and became maximal before C fibres had been recruited. Similar results were obtained for oesophageal motor fibres.

A pulmonary projecting branch (or branches) identified as containing bronchomotor fibres, was then placed in a sleeve electrode of the type described above, the branch being crushed peripherally.

Recording experiments

In order to expose the medulla, the occipital bone was opened and the dura over the cerebellum excised and reflected. The caudal portions of the cerebellum were removed by suction.

The activity of medullary neurones was recorded using double-barrelled micro-pipettes, the recording barrel containing pontamine sky blue dye dissolved in 0·5 M-sodium acetate, the second barrel containing either L-glutamic acid (0·8 M, pH 8·0) or DL-homocysteic acid (DLH, 0·2 M, pH 8·5). Current controls, when needed, could be made by recording from the drug barrel, current being delivered via the pontamine blue barrel. Recordings were made from the region of the nucleus ambiguus in response to stimulation of the cardiac branches (thirty-two experiments) and pulmonary branches (six experiments). Unit activity together with phrenic nerve activity and arterial blood pressure were stored on tape (Ampex PR 500).

Pulse-triggered histograms were generated by feeding the discriminated spike into an Ortec time histogram generator, which was triggered on every second pulse. The femoral pulse wave was averaged on the same system and displayed on the same time scale.

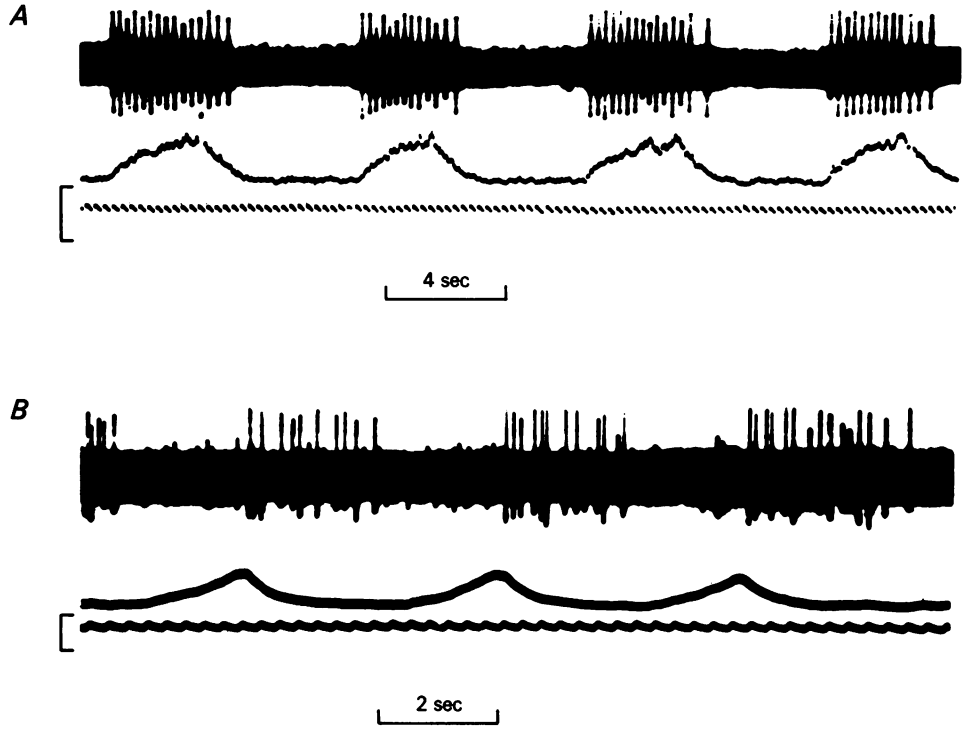
Histological location of recording sites

Responsive units were marked by the ejection of pontamine sky blue dye (Hellon, 1971). At the end of each experiment the brain was removed and fixed in 10 % formal saline. Histological preparations and the mapping procedures were as described in a previous paper (Lipski, McAllen & Spyer, 1975).

RESULTS

This paper describes the properties of eighty-four vagal neurones recorded from the area of the nucleus ambiguus. These were identified by their antidromic response to stimulation of the cardiac (seventy-four neurones) or pulmonary (ten neurones) branches of the right thoracic vagus. Their axons had conduction velocities between

2·8 and 15·5 m/sec (see Table 1 and Text-fig. 4*B* and *C*) and were thus considered to be B fibres. We will present evidence that these neurones are likely to belong to two distinct populations which are cardioinhibitory (sixty-four) or bronchoconstrictor (twenty) in function. The present data concern only those neurones which were either spontaneously active or induced to fire by ionophoretic application of either DLH or glutamate (i.e. a selected population).



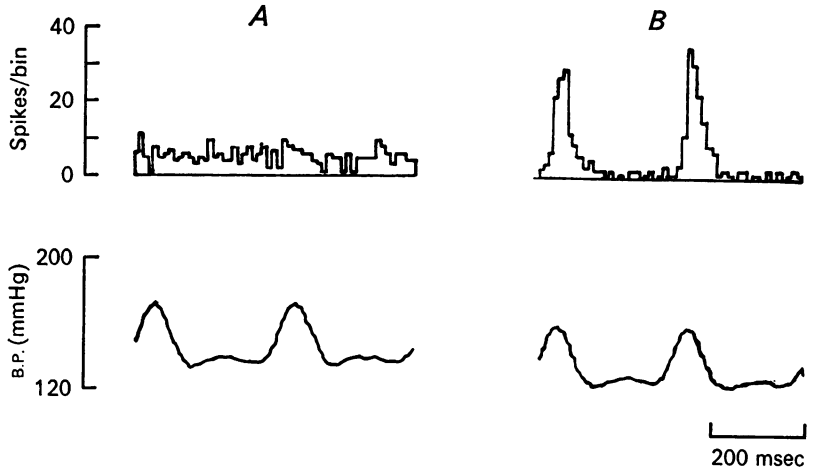
Text-fig. 1. Vagal efferent activity recorded from the nucleus ambiguus. Traces from top downwards: unit activity, rectified and smoothed phrenic activity, femoral arterial blood pressure (calibration bar 0–200 mmHg). *A*, inspiratory-firing neurone whose axon projected to a pulmonary branch of the right vagus (DLH, 2 nA backing current). *B*, expiratory-firing neurone projecting to a cardiac branch of the right thoracic vagus. Neurone only fired in the presence of DLH (40 nA in this case). (The neurone's activity started during the rapid decrease in phrenic discharge, somewhat obscured in this case by the long time constant of the integration.)

Pulmonary branch projecting neurones

Ten neurones were activated antidromically on stimulating the pulmonary branches of the vagus. They had B fibre axons (conduction velocities 5·4–15·5 m/sec, see Text-fig. 4*B* and *C*). Bronchoconstrictor fibres have conduction velocities in this range (see Methods). Other vagal efferents with B fibre axons include both cardiac and oesophageal efferents, but the pulmonary vagal branches identified in this study when stimulated produced an increase in respiratory resistance (see Methods) but no effect on heart rate or oesophageal tone. We consider that the pulmonary branch projecting neurones we have identified are likely to have a bronchoconstrictor function.

All but one of these neurones were spontaneously active, the otherwise silent cell requiring only a small expulsion current of DLH (2 nA) to excite it. The spontaneously active neurones were also powerfully excited by DLH. Small currents ($\leqslant$ 20 nA) potentiated their normal inspiratory discharge and initiated firing at a lower rate during expiration. An example of the spontaneous activity of such a neurone is shown in Text-fig. 1*A*.

Under our experimental conditions their activity was not obviously modulated by the respiratory pump, but the rapid shallow ventilation used in these experiments would have been unfavourable for showing such an influence. We have looked for an influence of the carotid sinus baroreceptors on their discharge by plotting pulse-triggered histograms of their activity. In the six cases studied in detail we saw no significant increases or decreases in the probability of firing at any phase of the cardiac cycle. An example is illustrated in Text-fig. 2*A*.



Text-fig. 2. Pulse-triggered histograms of vagal efferent activity (256 cycles superimposed, bin width 10 msec). *A*, inspiratory firing neurone relaying in pulmonary branch (2 nA DLH backing current). *B*, expiratory firing neurone relaying in cardiac branch (17 nA DLH ejection current). Lower trace is the averaged femoral pulse wave form (bin width 10 msec).

Cardiac branch projecting neurones

Although the cardiac branches which were stimulated innervated the heart, there were often small side branches coursing towards the stump of the excised upper lobe of the lung (see Methods). Therefore, it would not have been surprising if a small proportion of the neurones projecting to the cardiac branches were bronchomotor in function. Indeed ten out of the seventy-four neurones excited antidromically on stimulating cardiac branches had properties indistinguishable from those described for pulmonary branch projecting neurones, i.e. they fired spontaneously in inspiration and showed no pulse correlation (see Table 1). We consider that these, like the pulmonary branch projecting neurones, are probably bronchoconstrictor in function (bronchoconstrictor vagal motoneurones, b.v.m.).

Fifty of the remaining sixty-four cardiac branch projecting neurones were silent

until excited by expulsion of excitant amino acids (forty-five units by DLH, five units by glutamate). Both the spontaneously active cells and those excited by ionophoresis at expulsion currents close to threshold (range 2–40 nA) fired exclusively in *expiration* (see Text-fig. 1*B*). At higher currents they could sometimes be induced to fire (more slowly) in inspiration. This distinct difference in firing pattern compared to that of b.v.m.s provided the primary criterion for dividing the two classes of efferents, but other properties correlated with this division. In particular, expiratory-firing neurones showed obvious pulse modulation when pulse-triggered histograms were constructed (Text-fig. 2*B*). This was so for twenty-three of the twenty-seven units tested (fifteen

TABLE 1. The properties of cardiac branch projecting and pulmonary branch projecting vagal neurones of the nucleus ambiguus. *P* values for significantly different population. *In cases of failure the cardiac branches were shown not to be conducting centrifugally

	Cardiac branch projecting		Pulmonary branch projecting		
	(1)	(2)	(3)		
No.	64	10	10		
Firing pattern	Expiratory	Inspiratory	Inspiratory		
Spontaneous activity	14	8	9	(1)–(2)	$P < 0.01$
				(1)–(2+3)	$P < 0.001$
Conduction velocity	2·8–12·8 m/sec (mean 7·1 m/sec)	5·2–11·5 m/sec (mean 8·3 m/sec)	5·4–15·5 m/sec (mean 8·4 m/sec)		
Cardiac rhythm	23/27	0/6	0/6	(1)–(2)	$P < 0.05$
				(1)–(2+3)	$P < 0.01$
Bradycardia* to activation	15/18	0/5	0/6	(1)–(2)	$P < 0.05$
				(1)–(2+3)	$P < 0.01$

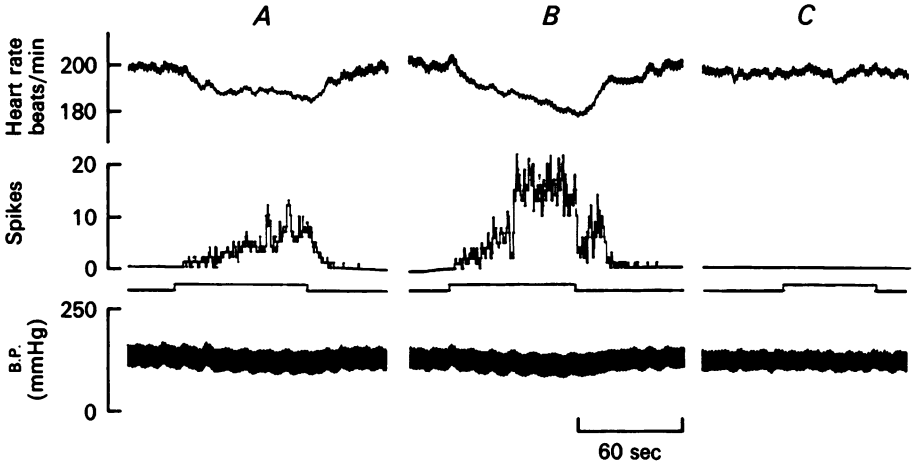
units with all buffer nerves intact, eight units with only sinus nerves intact; see Table 1). In two of the four cases where no pulse correlation was seen, carotid occlusion had no effect on the heart rate or blood pressure, so the carotid baroreceptors were probably not functioning. That the pulse correlation was of baroreceptor origin could be clearly shown in animals with aortic nerves sectioned, where it disappeared reversibly on bilateral carotid occlusion regardless of the magnitude of the reflex changes in blood pressure (see Fig. 1 of the following paper, McAllen & Spyer, 1978). In those cases with all buffer nerves intact, a small residual effect, presumably of aortic origin, remained after carotid occlusion in five of fifteen units tested. The population of expiratory-firing neurones thus have properties that suggest that they are cardioinhibitory vagal motoneurones (c.v.m.).

Heart rate changes during amino acid application

During preliminary experiments, it became apparent that stimulation of cardiac branches of the right vagus containing very few B fibres could have a marked effect on heart rate. This favourable situation led us to look for the possibility that activating individual cardiac vagal neurones might produce a measurable bradycardia. Not without some surprise we were able to observe a small but reproducible fall in heart rate, which correlated strikingly with the neurone's discharge, when a c.v.m. was activated with DLH or glutamate (Text-fig. 3). Current controls had no effect on

the neurone or heart rate. This phenomenon was observed with fifteen of eighteen such neurones tested. On two occasions where the test proved negative we verified that there was a conduction block in the branch between the stimulating electrodes and the heart. Heart rate slowed by 1 beat per min for every 2–8 spikes/sec discharge in individual units. In ten units studied in detail in nine experiments, the heart rate fell by 5–30 beats/min in response to DLH at currents below 40 nA.

The heart rate response to amino acid was extremely localized around a c.v.m. (Pl. 1). Applications elsewhere within the nucleus ambiguus, including experiments on eleven units previously classified as b.v.m. (five units relaying in the cardiac



Text-fig. 3. Traces from top downwards: heart rate derived from the arterial pulse, c.v.m. activity plotted as spikes in 200 msec bins, event marker showing application of DLH, and lastly femoral arterial blood pressure.

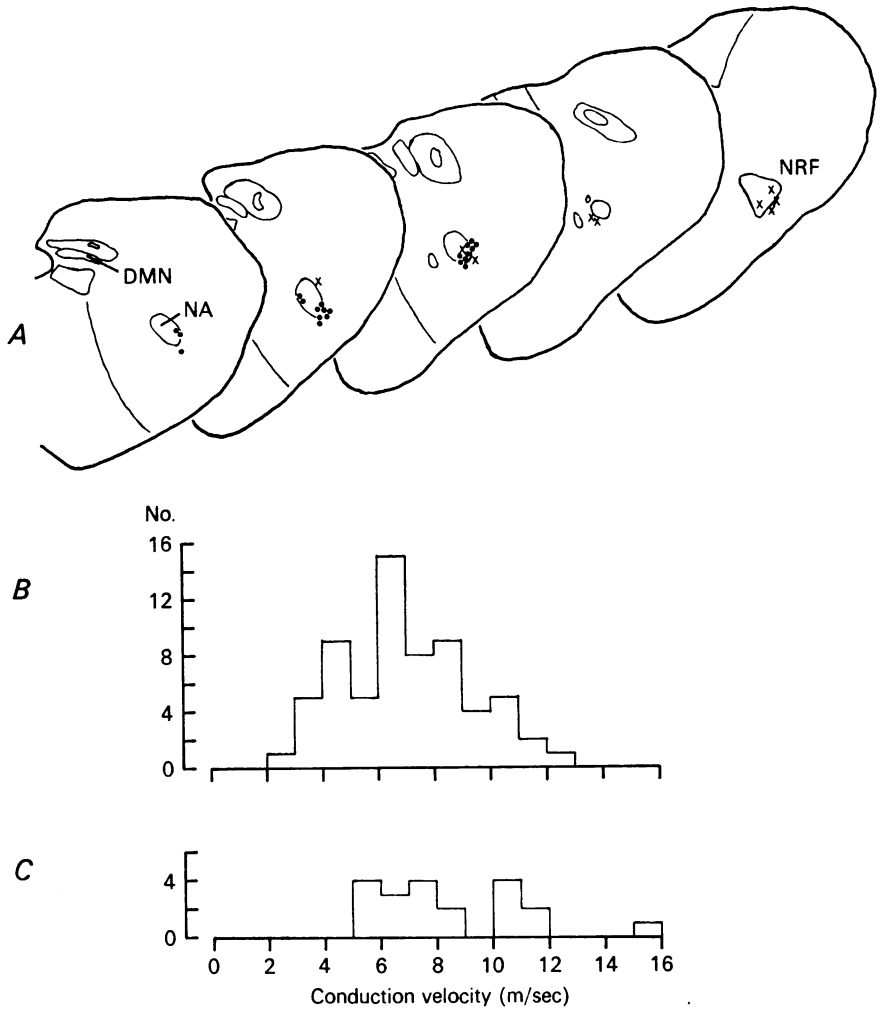
The response of c.v.m. to application of DLH at 60 nA (*A*) and 80 nA (*B*). Note the accompanying fall in heart rate and the similarity in time course of the two effects. The effects can be attributed to the influence of DLH, since passing 80 nA of direct current (through pontamine blue barrel, recording through the DLH barrel) affected neither the c.v.m. nor heart-rate (*C*).

branch, six in the pulmonary branch), had no effect on the heart rate. We only observed the effect during *focal* c.v.m. recordings, (we could not activate them otherwise) and only then when the cell was excited by amino acid. This probably limits the effective spread of amino acid to less than 100 μm (see Discussion). Higher expulsion currents ($>$ 80 nA) might have spread further in effective concentration but these also tended to produce falls in arterial blood pressure and sometimes respiratory effects. The heart rate changes (but not the neurones' activity) could be abolished by crushing the relevant cardiac branch.

Location

All the neurones in this study were recorded from the region of the nucleus ambiguus at levels rostral to the obex. More details of their location were provided by marking a proportion of recording sites by ejecting pontamine sky blue from the

electrode tip. The locations of 21 c.v.m.s are shown in Text-fig. 4*A* (see also Pl. 1*A*). They were found associated with, and usually slightly ventral to, the lateral concentration of inspiratory neurones. Their distribution corresponds to that shown in



Text-fig. 4. *A*, location. The positions of twenty-one c.v.m.s (●) and nine b.v.m.s (×) are shown on five standard sections of the medulla taken at obex level and 1, 2, 3, and 4 mm rostral to the obex. DMN = dorsal motor nucleus of vagus, NA = nucleus ambiguus, NRF-nucleus retrofacialis. *B* and *C* show histograms of the conduction velocities of cardiac and bronchomotor units, respectively.

a previous paper (McAllen & Spyer, 1976) but is probably more accurate since in the present study focal recordings were selected, and contamination of this sample with b.v.m.s was avoided by the use of physiological criteria.

We also marked the positions of a small number of b.v.m.s (see Text-fig. 4). They were concentrated in more rostral areas of the nucleus ambiguus than the cardiac

group, encroaching on the nucleus retrofacialis, although some were found more caudally, when they were usually dorsal to the cardiac group. The last statement is based mainly on observations on unmarked cells.

DISCUSSION

In this paper we have described two distinct populations of motoneurones in the nucleus ambiguus, both of whose axons were B fibres which relayed in the thoracic branches of the right vagus. On the basis of their properties they have been presumed to be cardioinhibitory or bronchoconstrictor in function. C.v.m.s were *expiratory*-firing although they usually needed excitant amino acids to activate them. They showed a cardiac rhythm in their discharge and were capable, when activated, of slowing the heart. B.v.m.s (which relayed either in the pulmonary or cardiac branches) were *inspiratory*-firing and usually spontaneously active. They showed no cardiac rhythm and were not capable of slowing the heart. The two sets of properties will be discussed in more detail below.

Cardiac slowing in response to amino acids

This is perhaps the most surprising finding reported here and hence merits discussion first. Given that it is a reproducible effect, does the bradycardia result from the activity of a single preganglionic neurone? While we cannot answer the question unequivocally, several lines of evidence suggest that this can happen. Certainly the time course of the bradycardia correlated closely with the activity evoked in the neurone under study. This was particularly striking with DLH at currents close to threshold, when the bradycardia only started when the c.v.m. fired, though this was considerably delayed from the onset of amino acid application and its excitant effect on any non-cardiac neurones in the background.

The effect is very localized, even within the nucleus ambiguus. Bradycardia could only be evoked from an area where a c.v.m. was recorded; it was never seen in response to ionophoretic activation of b.v.m.s or inspiratory neurones. The graph in Pl. 1*C* indicates electrode displacements at which *no* response was evoked; it thus overestimates the distances over which the drug spreads in effective concentration. In practice, we have only seen bradycardia in response to DLH (or glutamate) when a c.v.m. was focally recorded, and only then when it was activated. With moderate currents this was only possible within distances of less than 100 μm from the point where the spike was largest. Our estimate of a restricted effective spread of DLH and glutamate on ionophoresis is borne out by studies of others in the central nervous system using similar current ranges for DLH (Biscoe & Curtis, 1967; Crawford, Curtis, Voerhoeve & Wilson, 1966) and for glutamate (Herz, Zieglgänsberger & Färber, 1969; Zieglgänsberger & Puil, 1973). Accepting, then, that the spread of the drug is limited, is it nevertheless able to activate other cardiac efferents or interneurones that then excite them? When the recording electrode is amongst a 'nest' of c.v.m.s, the antidromic potential may sometimes include two or even three units, but it is rare for more than one to be recorded focally and to be excitable by DLH (they are usually at least 200 μm apart). It is unlikely that the antidromic stimulus would fail to excite many cardioinhibitory axons, since we stimulated at voltages

supramaximal for heart rate effects, and the stimulating electrodes were normally placed around both caudovagal and craniovagal branches i.e. certainly most of the cardiac efferents of the right vagus were activated. If the bradycardia were mediated either by other cardioinhibitory neurones or by interneurones that excite them, we should expect DLH to produce activity in non-focally recorded c.v.m.s. At the moderate currents used, we know by testing for collisions that it did not.

Additional support for the contention that a single preganglionic neurone may slow the heart is provided by four separate observations of a transient bradycardia accompanying the injury discharge of accidentally penetrated single c.v.m.s. In summary, while we cannot exclude the participation of other cells in producing the bradycardia, it seems plausible that with moderate currents ($\leqslant 40$ nA) the effect may be produced by a single preganglionic neurone whose function we can thus confirm as cardioinhibitory.

Firing patterns; cardioinhibitory neurones

The population described here has just the properties one would expect of cardioinhibitory neurones. Our identification method allowed us to sample from virtually the whole range of preganglionic neurones rather than just those that happen to be spontaneously active, as is the case for fibre recordings. We found that relatively few fired spontaneously, which was not unexpected in view of the low vagal tone in anaesthetized cats with open chests (see also Kunze, 1972). Their ionophoretically-induced or spontaneous activity, however, showed that they do indeed form a homogeneous population with properties equivalent to those of presumed cardioinhibitory fibres in the cat (Kunze, 1972), and dog (Iruichijima & Kumada, 1964; Katona, Poitras, Barnett & Terry, 1970; Katona, Lipson & Dauchot, 1977).

In a recent report Schwaber & Schneiderman (1975) described five neurones in the dorsal motor nucleus of the rabbit which they considered to be cardioinhibitory. This was based on the observation that they had B fibre axons in the cervical vagus and were excited at short latency by stimulation of the aortic nerve. While we cannot exclude a species difference in the location of the c.v.m.s (in cats they are not present in the dorsal motor nucleus, McAllen & Spyer, 1976), we feel it is premature to comment on such a limited sample, especially since no details were given of their firing patterns and their projection to the heart was not demonstrated.

There is a central respiratory modulation of the discharge of the neurones described above, and this follows the pauses in phrenic activity, not the degree of inflation of the lungs. It is quite possible that pulmonary receptors can also modulate their activity (cf. Anrep, Pascual & Rössler, 1936), but our experimental conditions (rapid shallow inflations) were not favourable for detection of this. Further, c.v.m.s are evidently influenced by arterial baroreceptors in the expected manner. These are dealt with in more detail in the following paper (McAllen & Spyer, 1978). They can also be activated by carotid chemoreceptor stimulation (R. M. McAllen & K. M. Spyer, unpublished observations).

Bronchoconstrictor neurones

The evidence that b.v.m.s with axons in the cardiac branch belong to the same population as those of the pulmonary branch rests on their apparently identical firing patterns. The cardiac branches did include small offshoots going towards the lungs, and we can be reasonably sure that the pulmonary branches chosen were

destined for the lungs, and included bronchoconstrictor fibres but not cardiac or oesophageal efferents (see Methods). Bronchoconstriction is produced by vagal B fibres (see Methods) and this range includes the axons of all b.v.m.s. They appear to be the same as the population of vagal efferents which Widdicombe (1961, 1966), concluded from a variety of arguments were probably bronchoconstrictor. Our experimental conditions (under which they all fired predominantly in inspiration) correspond to those of Widdicombe when he used open-chested, artificially-ventilated animals and found eight out of nine to fire predominantly in inspiration. With the chest closed and spontaneous respiration, however, a number of different firing patterns were manifest (Widdicombe, 1961, 1966). The principal criterion he used to define the population and confirm their probable bronchoconstrictor function was a prolonged excitation by stimulation of the larynx. This provided us with great technical difficulties (holding the unit during a cough) and we have not made a systematic investigation, but on two occasions we have been able to produce clear excitatory responses in b.v.m.s by laryngeal stimulation.

Location

The two groups of preganglionic neurones identified were located in the nucleus ambiguus, b.v.m.s being more rostral and dorsal than c.v.m.s. As yet we have no evidence on whether they appear histologically distinct, although the action potentials of b.v.m.s often appeared larger. Lawn (1966) showed that somatic motoneurones innervating different muscles of the pharynx and larynx are represented in distinct areas of the nucleus ambiguus. The present observations extend this principle to the preganglionic parasympathetic neurones supplying the heart and lungs.

This work was supported by a programme grant from the M.R.C. The technical assistance of D. Miller, S. Deeming and Mrs C. Loveland is gratefully acknowledged.

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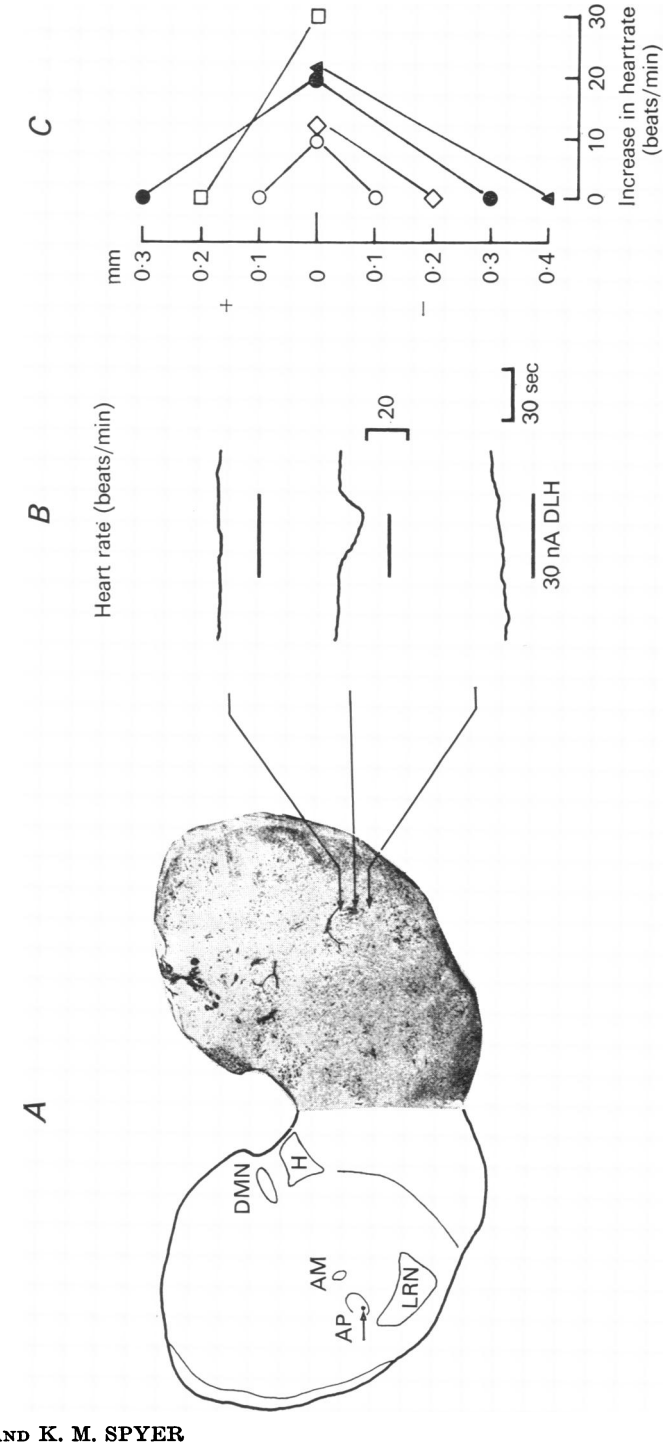
EXPLANATION OF PLATE

A, photomicrograph showing transverse hemisection of cat medulla and diagrammatic hemisection on the left side. The location of c.v.m. (marked by dye expulsion from the recording electrode) shown by the middle arrow on the right, and by the arrowed dot on the left. The effects on heart rate of 30 nA DLH delivered ionophoretically at the recording site shown by middle arrow (this activated the neurone) and at positions 300 μm above and below, marked by the other two arrows. Heart rate only fell during DLH application close to the c.v.m.s (see middle arrow). *C*, the effects on heart rate of activating five different c.v.m.s (each shown with a different symbol) using 30–40 nA DLH. The effects at the point of focal recording, and at points above, below or both are shown.

Abbreviations: AM, medial column of nucleus ambiguus; AP, principal column of nucleus ambiguus; CMN, dorsal motor nucleus of the vagus; H, hypoglossal nucleus; LRN, lateral reticular nucleus.



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