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THE PATTERN OF SYMPATHETIC NEURONE ACTIVITY DURING EXPIRATION IN THE CAT

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SUMMARY

1. The properties of sympathetic preganglionic neurone activity during expiration were studied in pentobarbitone-anaesthetized ($n = 26$) and in non-anaesthetized, mid-collicular decerebrate ($n = 5$), paralysed, artificially ventilated cats in which the electrical activity of the phrenic nerve and of the cervical sympathetic trunk was recorded.

2. In control conditions (end-tidal P_{CO_2} between 35 and 40 mmHg, zero end-expiratory pressure) sympathetic activity during expiration was either steady at a low level ($n = 11$) or showed a modest progressive increase from a low level in early expiration ($n = 17$). Very infrequently ($n = 3$), it showed a transient increase during the second half of expiration.

3. Artificial ventilation with positive end-expiratory pressures in the range from $2{\cdot}1 \pm 0{\cdot}4$ (mean $\pm$ S.D.) to $6{\cdot}7 \pm 0{\cdot}6$ cmH_2O caused, in cats with intact vagus nerves, an increase in sympathetic neurone activity during the second half of expiration. Within this range of pressures, the magnitude of the increase was related to the magnitude of the positive end-expiratory pressure. This effect reversed at higher positive end-expiratory pressures. Pressures in excess of $10{\cdot}2 \pm 1{\cdot}8$ cmH_2O caused inhibition of sympathetic activity.

4. The sympatho-excitatory effect of positive end-expiratory pressure disappeared after bilateral cervical vagotomy. With intact vagus nerves, it also disappeared at levels of systemic hypocapnia (end-tidal $P_{CO_2} \leqslant 15$ mmHg) which abolished phrenic nerve activity. In hypocapnia, artificial ventilation with peak tracheal pressures greater than $7{\cdot}2 \pm 1{\cdot}1$ cmH_2O caused inhibition of sympathetic activity, while ventilation with lower end-expiratory pressures had no effect on sympathetic activity. It may be concluded that the sympatho-excitatory effect of positive end-expiratory pressure is mediated by vagal afferents and requires a certain level of brain-stem respiratory neurone activity.

5. Sympatho-excitation during expiration was also observed, in normocapnic conditions, during short-duration static lung inflation with tracheal pressures in the range from $2{\cdot}5 \pm 0{\cdot}3$ to $7{\cdot}0 \pm 0{\cdot}8$ cmH_2O as well as during artificial ventilation with zero end-expiratory pressure when lung inflation occurred in expiration. These responses were abolished by bilateral cervical vagotomy and during systemic hypocapnia.

6. Sympatho-excitation during expiration was also observed when systemic

hypercapnia was produced in vagotomized cats by artificial ventilation with gas mixtures containing 5 or 10% CO_2.

7. These results can be explained by the hypothesis that some brain-stem expiratory neurones are a source of facilitatory synaptic input to sympathetic neurones. The activity of brain-stem expiratory neurones is known to be enhanced by moderate degrees of lung inflation and by increased chemical drive. Under these conditions sympathetic neurone activity would be expected to increase during expiration.

INTRODUCTION

Previous studies have firmly established that the firing pattern of single sympathetic preganglionic units, or of whole sympathetic nerves, has a rhythmic component, which is synchronous with the inspiratory phase of the respiratory cycle at normal respiratory frequencies and has properties similar to those of the phrenic nerve burst (Cohen & Gootman, 1970; Preiss, Kirchner & Polosa, 1975; Barman & Gebber, 1976; Preiss & Polosa, 1977; Gerber & Polosa, 1978, 1979; Polosa, Gerber & Schondorf, 1980). In contrast there are few observations on the properties of the sympathetic discharge which occurs during the expiratory phase of the respiratory cycle. Published records of the electrical activity of whole sympathetic nerves in vagotomized animals show that the level of activity increases from early to late expiration (Cohen & Gootman, 1970; Barman & Gebber, 1976). Some of these records show a monotonic increase in activity throughout expiration, while others show an early increase followed by a plateau. Various interpretations of these incrementing patterns of sympathetic activity during expiration are possible, but none has been subjected to experimental test as yet. The incrementing pattern may represent a recovery of activity from an early expiratory depression or an incrementing excitation synchronous with the phase of activity of brain-stem expiratory neurones (Bainton, Richter, Seller, Ballantyne & Klein, 1985) or of phase-spanning respiratory neurones (Gootman, Cohen, Piercey & Wolotsky, 1975).

The present experiments were directed to the question of whether the background discharge of a sympathetic preganglionic neurone contains rhythmic components related to the activity of brain-stem expiratory neurones. The existence of such components would suggest the existence of synaptic connexions between these two sets of neurones. The activity level of brain-stem expiratory neurones is known to be enhanced during moderate lung inflation (Sears, 1964; Bishop, 1967) and during systemic hypercapnia (Bainton, Kirkwood & Sears, 1978; Bainton & Kirkwood, 1979). Therefore in the present experiments we have investigated the effect of these expiratory facilitating manoeuvres on the pattern of firing of the cervical sympathetic trunk in expiration.

METHODS

Thirty-one cats of both sexes were used (2·3–4·0 kg). Five of the cats were decerebrated at mid-collicular level under ether anaesthesia. In the remainder, anaesthesia was obtained with I.P. sodium pentobarbitone (35 mg/kg initial dose, followed by a maintenance dose of 9 mg/kg I.V. every 3 h). With this dose the withdrawal reflex on pinching forepaw or hind paw was suppressed for the duration of the experiment. The trachea was cannulated and the animals were artificially ventilated, while continuously monitoring tidal CO_2 concentration with an infra-red gas analyser

and tracheal pressure with a strain gauge. All cats were paralysed with pancuronium bromide (initial dose 200 μg/kg followed by maintenance doses of 100 μg/kg which were given every 2–3 h, when the effect of the previous dose had worn off, as evidenced by the appearance of spontaneous breathing movements, and after testing for adequacy of the level of anaesthesia). Frequency and tidal volume of the respiratory pump were adjusted to obtain, in control conditions, an end-tidal P_{CO_2} of between 35 and 40 mmHg. In ten experiments a phrenic-triggered respiratory pump was used (Remmers & Gauthier, 1976). Central respiratory cycle is defined as the interval between two successive phrenic nerve bursts. Inspiration is defined as the time between onset of phrenic nerve activity and beginning of rapid decline, expiration as the remainder of the cycle. Inflation is defined as the increase in tracheal pressure caused by the respiratory pump, deflation as the return of tracheal pressure to pre-inflation level from peak inflation. For consistency with the terminology used in respiratory physiology (Grippi, Pack, Davies & Fishman, 1985) the value of tracheal pressure or of lung volume at the end of the deflation phase of the ventilation cycle is referred to as end-expiratory level. A variable load to passive deflation (a water column of various heights), which produced graded increases in end-expiratory lung volume (functional residual capacity; F.R.C.), was introduced in the circuit when required (Bishop, 1967, Russell & Bishop, 1976). Systemic hypercapnia was produced by ventilation with gas mixtures containing various CO_2 concentrations in O_2, systemic hypocapnia by hyperventilation in room air. Catheters were placed in a femoral artery and vein for continuous recording of systemic arterial pressure and for administration of drugs, respectively. Rectal temperature was maintained at 37–38 °C using an infra-red lamp controlled by a feed-back circuit.

The electrical activity of the phrenic nerve, of the cervical sympathetic trunk and, in three cats, of the recurrent laryngeal nerve was recorded monophasically with silver hook electrodes, amplified (bandpass 10 Hz–10 KHz) and stored on magnetic tape. After half-wave rectification and low-pass filtering (time constant 100 ms) these signals were also displayed on a storage oscilloscope and pen recorder. These rectified, low-pass filtered, records of neural activity are usually referred to in the literature as 'integrated' activity. The level of 'zero' activity for the recording of cervical sympathetic trunk activity was obtained by applying procaine to the nerve or by crushing the nerve proximal to the recording electrodes.

The carotid bifurcation was exposed bilaterally in all animals, after resection of a segment of the oesophagus and trachea, and both carotid sinus nerves were identified at their junction with the IX cranial nerve and cut. The aortic nerves were also bilaterally cut when they were identified as separate from the cervical vagus nerves. The vagus nerves were cut in some cases before the beginning of the experiments. In the majority of cases they were prepared for section during the experiment. In two cats, after a left thoracotomy, a catheter was inserted into the pericardial sac for injection of procaine (2 ml of a 2% solution) (Arndt, Pasch, Samodelov & Wiebe, 1981) at the appropriate time during an experiment.

RESULTS

Patterns of activity during expiration of the cervical sympathetic trunk at normal F.R.C.

The pattern of sympathetic activity during expiration was examined in nine vagotomized cats and in twenty-two cats with intact vagus nerves. In the latter group the observations were made in the absence of artificial ventilation during a period equal to two respiratory pump cycles. All animals were normocapnic and showed a pronounced inspiration-synchronous sympathetic discharge. In eleven of the cats the mean level of sympathetic activity was approximately constant throughout expiration. This will be referred to as a 'tonic' pattern (Fig. 1*A*). In the majority of cats (seventeen) the mean level of activity increased from early expiration to middle or late expiration. This will be referred to as an 'incrementing' pattern. In some cats showing an incrementing pattern the increase was continuous throughout expiration, in others a plateau was formed in late expiration (Fig. 1*B*). These differences in the pattern of expiratory firing between animals were not related to differences in the

duration of the respiratory cycle or of its phases, nor to presence or absence of the vagus nerve. Each pattern was invariant for a given set of experimental conditions. Three cats showed a distinct burst of activity in late expiration (Fig. 1*C*). Thus, in these cases sympathetic activity showed two peaks for each respiratory cycle. Similar observations have been recently described by Bainton *et al.* (1985). Peak amplitude and rate of rise of this expiratory burst, in the low-pass filtered record, were lower than for the inspiration-synchronous sympathetic burst. Since these animals were

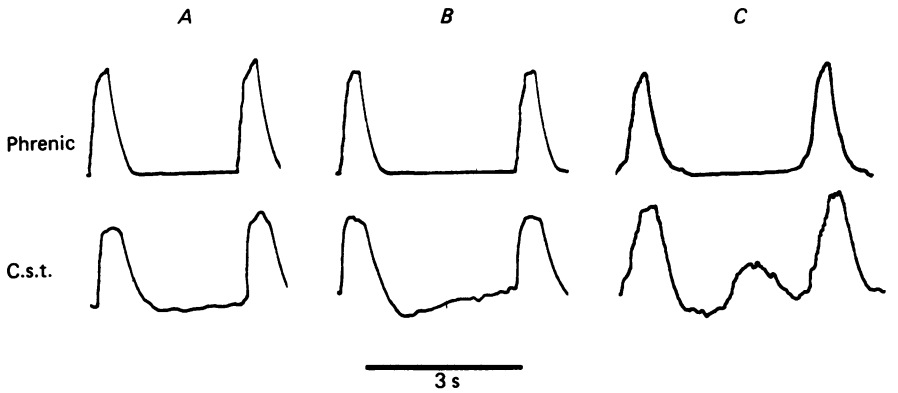

Fig. 1. Patterns of sympathetic activity during expiration in anaesthetized, vagotomized, paralysed, normocapnic cats. From above, averaged (twenty sweeps) integrated phrenic nerve and cervical sympathetic trunk (c.s.t.) activity. In this and all subsequent Figures all averages shown were triggered from the onset of phrenic nerve discharge. An upward deflexion indicates increased discharge. *A*, *B* and *C*, recordings from three different cats, all showing a pronounced inspiratory sympathetic discharge. *A*, mean level of sympathetic activity remained constant throughout expiration (tonic type). *B*, mean level of sympathetic activity increased continuously during expiration (incrementing type). *C*, a distinct wave-like discharge of sympathetic activity appeared during the second half of expiration.

sino-aortic denervated and vagotomized or at constant lung volume, it may be assumed that this pattern of sympathetic activity in expiration was not generated reflexly but by mechanisms within the C.N.S. A possible explanation for this pattern is that it results from excitatory input from brain-stem expiratory neurones to sympathetic preganglionic neurones. If this was the case, it should be possible to evoke an increase of sympathetic activity during expiration by performing manoeuvres which are known to increase the level of activity of brain-stem expiratory neurones.

Patterns of activity in expiration of the cervical sympathetic trunk during ventilation with positive end-expiratory pressure (increased F.R.C.)

Positive end-expiratory pressure within the range from $2{\cdot}1 \pm 0{\cdot}4$ to $6{\cdot}7 \pm 0{\cdot}6$ cmH_2O caused an increase in the level of activity during expiration of the cervical sympathetic trunk in nineteen of twenty-two cats with intact vagus nerves tested. In addition, expiratory loading caused attenuation of the inspiration-synchronous sympathetic discharge. Fig. 2 shows the results of such an experiment. At zero end-expiratory pressure the pattern of expiratory activity of the cervical sympathetic trunk was tonic

(Fig. 2*A*). Fig. 2*B* and *C* shows the effect of ventilation with end-expiratory pressures of 2·5 and 5·0 cmH_2O, respectively. An expiratory wave appeared, reminiscent of that shown in Fig. 1*C*. The expiratory wave appeared with some delay with respect to the onset of expiration, incremented first and then decremented somewhat before the onset of the following inspiration. The amplitude of this expiratory wave of

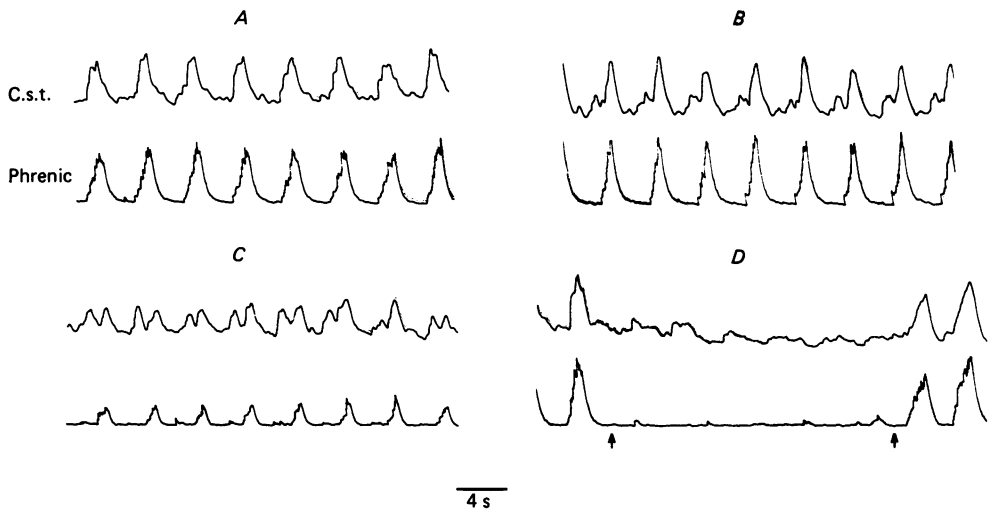

Fig. 2. Effect of positive end-expiratory pressure on the pattern of cervical sympathetic trunk activity in expiration. Each panel shows integrated cervical sympathetic trunk (c.s.t.; above) and phrenic nerve activity. *A*, control records at zero end-expiratory pressure. Sympathetic activity in expiration is of the tonic type. *B*, *C* and *D* show the effects of end-expiratory pressures of 2·5, 5·0 and 10·0 cmH_2O respectively. In *B*, a small wave-like increase in sympathetic activity appears late in expiration. In *C*, the wave of sympathetic activity in expiration increases in size, while the phrenic burst and inspiration-synchronous sympathetic discharge are markedly reduced. In *D*, after one cycle of ventilation at zero end-expiratory pressure, end-expiratory pressure of 10 cmH_2O is applied between the arrows. The phrenic nerve burst is completely suppressed. Cervical sympathetic trunk activity shows a progressive slow decline, on which are superimposed waves of depression, presumably synchronous with each lung inflation. Returning to zero end-expiratory pressure (second arrow) causes the reappearance of the phrenic burst and of the inspiration-synchronous sympathetic discharge.

sympathetic activity was greater at 5·0 (Fig. 2*C*) than at 2·5 (Fig. 2*B*) cmH_2O. At 5·0 cmH_2O end-expiratory pressure a decrease in peak amplitude of the phrenic nerve burst and of the inspiration-synchronous wave of sympathetic discharge was also observed. The inspiratory and expiratory waves were of similar amplitude at this value of end-expiratory pressure. With a pressure of 10 cmH_2O (Fig. 2*D*), phrenic nerve activity disappeared, together with the inspiratory and expiratory components of sympathetic discharge. At this value of end-expiratory pressure the sympathetic record showed waves of depression which were related in time to each inflation phase of the respiratory pump cycle and which appeared to summate, resulting in a progressively decreasing level of sympathetic activity. Positive end-expiratory pressure produced the expected (Cohen, 1975) excitation of expiratory neurones: a burst of activity in late expiration appeared in the record of recurrent laryngeal nerve

activity during ventilation with end-expiratory pressures of 2·5 and 5·0 cmH_2O. All these effects on sympathetic, recurrent laryngeal and phrenic nerve activity disappeared after bilateral cervical vagotomy. Effects similar to those just described were obtained in cats in which procaine was injected in the pericardial sac. This result rules out the possibility that sensory receptors in the heart, responsive to changes

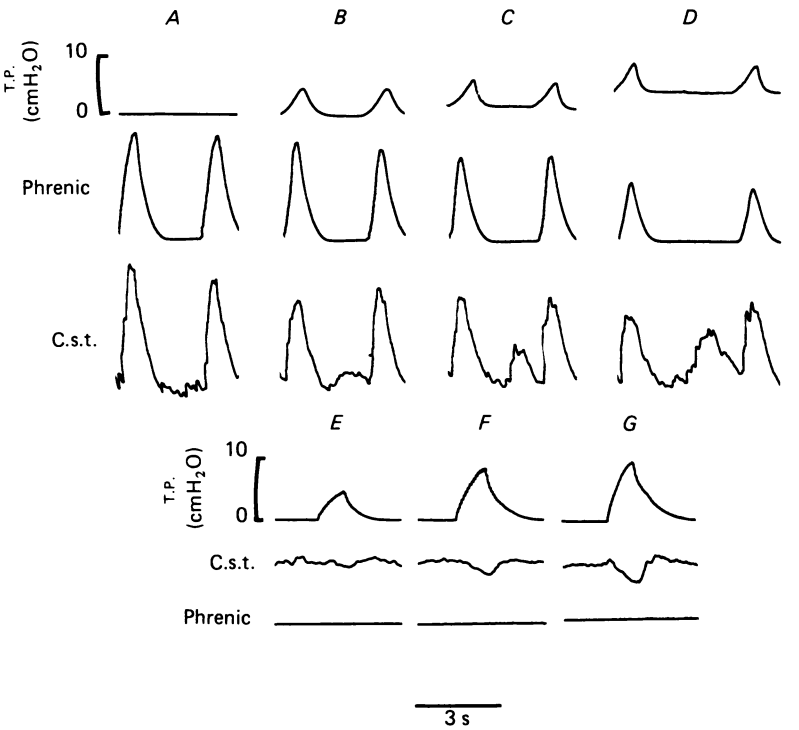

Fig. 3. Effect of increasing F.R.C. on cervical sympathetic trunk activity in expiration. Ventilation with phrenic-triggered pump. In each panel, from top tracheal pressure (T.P.), integrated phrenic nerve and cervical sympathetic trunk (c.s.t.) activity (averages of twenty sweeps). *A–D*, normocapnia (end-tidal P_{CO_2} 40 mmHg). *E–G*, hypocapnia (end-tidal P_{CO_2} 15 mmHg). *A*, respiratory pump off. *B*, ventilation with zero end-expiratory pressure. Notice small expiratory wave of sympathetic activity. *C* and *D*, ventilation with positive end-expiratory pressure of 2 and 6 cmH_2O. Notice graded increase in the expiratory wave of sympathetic activity, together with depression of phrenic burst and inspiration-synchronous sympathetic discharge and prolongation of expiration. *E–G* show the effect of lung inflation on sympathetic activity in the absence of rhythmic phrenic nerve activity.

in transmural pressure and/or shape of the cardiac chambers resulting from positive end-expiratory pressure ventilation (Cassidy & Mitchell, 1981), could be the source of the observed excitation in expiration of sympathetic neurones. In these experiments, in which ventilation with positive end-expiratory pressure was used, lung inflation started in inspiration and terminated in early expiration, and inspiration was terminated by the inflation, i.e. phrenic nerve activity was entrained to the respiratory pump cycle. This is the most commonly observed phase relation during entrainment of phrenic nerve activity to lung inflation in cats with intact vagus nerves artificially ventilated at inflation frequencies close to, or higher than, the spontaneous

frequency of the respiratory pattern generator (Cohen, 1969; Vibert, Caille & Segundo, 1981; Petrillo, Glass & Trippenbach, 1983).

Results similar to those shown in Fig. 2 were obtained during ventilation with positive end-expiratory pressure when lung inflation was limited to inspiration by using a phrenic-triggered respiratory pump, thus simulating the phase relation of inflation to central respiratory activity existing during spontaneous breathing. Onset

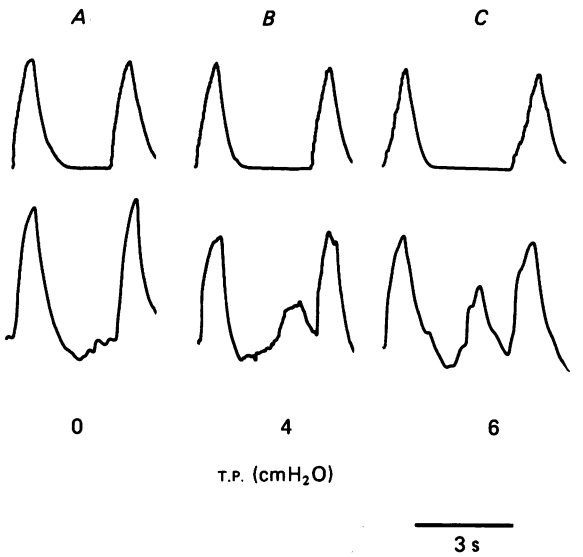

Fig. 4. Effect of static lung inflation on activity pattern of cervical sympathetic trunk in expiration. From above, integrated phrenic nerve and cervical sympathetic trunk activity (averages of eight sweeps). *A*, control at constant lung volume and zero end-expiratory pressure. Incrementing pattern of cervical sympathetic trunk activity in expiration. *B* and *C*, an increase in static lung volume (tracheal pressure (T.P.) of 4 and 6 cmH_2O) caused the appearance of a wave-like discharge during the second half of expiration, which was greater at 6 than at 4 cmH_2O tracheal pressure.

and end of lung inflation coincided approximately with onset and peak of the phrenic nerve burst. The results of such an experiment are shown in Fig. 3 in which *A* shows the wave form of the cervical sympathetic trunk recorded with the respiratory pump turned off and *B–D* show the effect of lung inflation during inspiration at increasing F.R.C.S caused by increasing levels of positive end-expiratory pressure. Fig. 3*E–G* shows the effect of lung inflations, causing peak tracheal pressures in the same range (4·4–9·4 cmH_2O) as in Fig. 3*B–D* on cervical sympathetic trunk activity during systemic hypocapnia. In Fig. 3*A*, with the respiratory pump turned off and zero tracheal pressure, the activity of the cervical sympathetic trunk in expiration was of the tonic type and at a low level. When the respiratory pump was turned on (Fig. 3*B*) a small-amplitude expiration-synchronous sympathetic discharge appeared. At increasing F.R.C. values (Fig. 3*C* and *D*) the peak amplitude of the expiration-synchronous sympathetic discharge increased; in addition, there was a progressive decrease in the amplitude of the inspiration-synchronous sympathetic discharge and phrenic burst. Duration of expiration increased. All these effects disappeared after bilateral cervical vagotomy.

In the experiments shown in Figs. 2 and 3 (panel *B–D*), the deflation phase of the ventilation cycle occurred in expiration. This fact suggests the possibility that the appearance of the expiration-synchronous sympathetic discharge during ventilation with positive end-expiratory pressures in the range from $2{\cdot}1 \pm 0{\cdot}4$ to $6{\cdot}7 \pm 0{\cdot}6$ cmH_2O was the result of reflex excitation of the sympathetic preganglionic neurone by vagal afferents activated by lung deflation (de Burgh Daly, Hazzledine & Ungar, 1967). This possibility is made unlikely by two sets of observations. One is that for the range

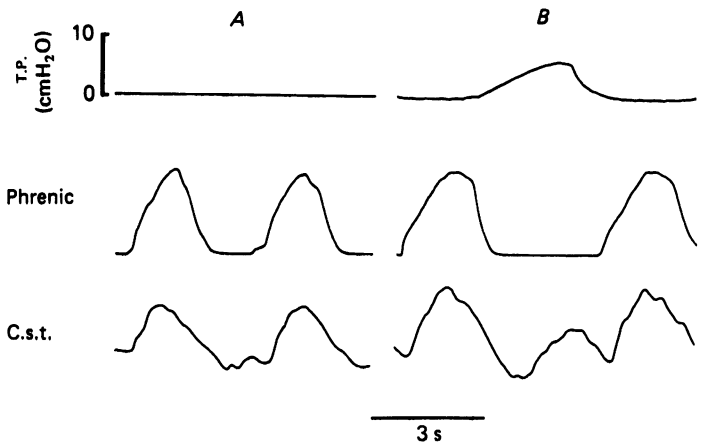

Fig. 5. Effect of phasic lung inflation in expiration on cervical sympathetic trunk activity in expiration. From above, tracheal pressure (T.P.), integrated phrenic nerve and cervical sympathetic trunk (C.S.T.) activities (averages of fifteen sweeps). *A* shows the cervical sympathetic trunk activity in expiration, in control conditions during apnoea. *B* shows that during ventilation with lung inflation occurring in expiration there is a wave-like sympathetic discharge, together with prolongation of the phase.

of lung volumes used in these experiments, when the rhythmic activity of the central respiratory pattern generator was eliminated by systemic hypocapnia (Fig. 3 *E–G*), no excitation of sympathetic activity on deflation, but only depression on inflation, was observed. The other observation is that at constant, elevated lung volume, in normocapnia, a similar sympatho-excitation in expiration was recorded (see next section). Both these observations are consistent with the expiration-synchronous discharge being an effect mediated by the rhythmical activity at brain-stem expiratory neurones.

Patterns of activity in expiration of the cervical sympathetic trunk at constant, elevated lung volume

In twenty-two cats with intact vagus nerves, at the end of the first pump cycle following the application of positive end-expiratory pressure, the respiratory pump was turned off for the duration of two to three respiratory cycles: the lungs remained inflated at the higher F.R.C. The results of a typical experiment are shown in Fig. 4. Fig. 4*A* shows the control sympathetic wave form at zero end-expiratory pressure and at constant lung volume: there was a prominent inspiration-synchronous sympathetic discharge while activity in expiration was of the 'incrementing' type.

Positive end-expiratory pressure (4 cmH_2O, Fig. 4*B* and 6 cmH_2O, Fig. 4*C*) caused the appearance of an expiration-synchronous sympathetic discharge. In addition, there was an increase in duration of expiration and some attenuation of the peak amplitude of the phrenic nerve burst, as described by Bartoli, Bystrzycka, Guz, Jain, Noble & Trenchard (1973). The peak amplitude of the inspiration-synchronous

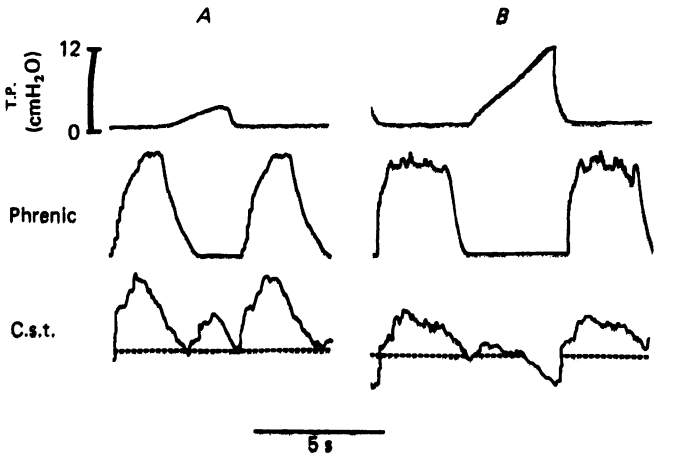

Fig. 6. The effect of lung inflation in expiration on cervical sympathetic trunk activity in this phase depends on lung volume. From above, tracheal pressure (T.P.), integrated phrenic and cervical sympathetic trunk (c.s.t.) activities (average of ten sweeps). Lung inflation triggered by phrenic burst. Dotted line marks the level of cervical sympathetic trunk activity at a value of systemic hypocapnia (end-tidal P_{CO_2} 15 mmHg) at which phrenic nerve activity was absent. *A*, lung inflation with low tidal volume (peak tracheal pressure 3·5 cmH_2O) caused an expiratory sympathetic discharge. *B*, lung inflation with large tidal volume (peak tracheal pressure 12 cmH_2O) produced an inhibition of cervical sympathetic trunk activity to a level lower than the hypocapnic level (i.e. below the level obtained by suppression of rhythmic respiratory activity). The increased duration and steeper rise of the integrated phrenic record in *B* are probably the result, respectively, of the dependence of inspiratory duration on the duration of the preceding expiration (Zuperku & Hopp, 1985) and of the reflex excitatory effect of large, rapid deflations on brain-stem inspiratory neurone activity (Sellick & Widdicombe, 1970).

sympathetic discharge was also somewhat attenuated. These effects were observed in nineteen out of twenty-two cats over the range of positive end-expiratory pressure values from $2·1 \pm 0·4$ to $6·7 \pm 0·6$ cmH_2O. Bilateral cervical vagotomy abolished all these effects, which were also lost at levels of systemic hypocapnia associated with abolition of phrenic nerve activity. With tracheal pressures in excess of 10·2 cmH_2O phrenic nerve activity disappeared together with the inspiration- and expiration-synchronous sympathetic discharge, and the level of sympathetic activity was markedly depressed.

Patterns of activity during expiration of the cervical sympathetic trunk at normal F.R.C. *when phasic lung inflations occur in expiration*

During the experiments of static lung inflation, described in the preceding section, the expiration-synchronous sympathetic discharge appeared at tracheal pressure

values well within the range of inflation pressures occurring during 'normal' artificial ventilation. It was of interest to test whether lung inflation with normal tidal volumes during expiration, in cats with intact vagus nerves, would cause an expiration-synchronous sympathetic discharge. This test was performed in fifteen cats with intact vagus nerves. Inflation in expiration was obtained in some of the cases by setting the frequency of the respiratory pump at a value lower than the intrinsic

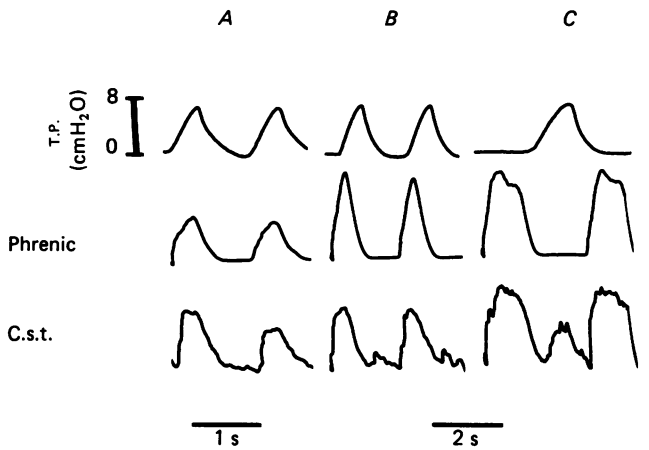

Fig. 7. Effect of lung inflation during various phases of the central respiratory cycle on cervical sympathetic trunk activity in expiration. Stable 1:1 locking of the lung inflation cycle to various phases of the respiratory cycle was produced by changing the frequency of the respiratory pump. From above, tracheal pressure (T.P.), integrated phrenic nerve and cervical sympathetic trunk (C.S.T.) activities (averages of twelve sweeps). *A*, lung inflation is synchronous with the phrenic nerve discharge. Sympathetic activity in expiration is of the tonic type and at a low level. *B*, lung inflation occurs during late inspiration and early expiration. Cervical sympathetic trunk discharge in expiration similar to *A*. *C*, lung inflation is entirely in expiration. A marked expiration-synchronous sympathetic discharge appears (absent when respiratory pump was turned off). Same explanation as in legend to Fig. 6 for the difference in phrenic burst shape between *B* and *C*. The difference in phrenic burst shape between *A* and *B* is likely to be due to the different phase relation of lung inflation to phrenic burst.

central respiratory frequency determined with the respiratory pump off. With intact vagus nerves, one-to-one phase-locking of the central respiratory cycle to the slower respiratory pump cycle occurs, within a limited range of frequencies, with inflation occurring in expiration (Petrillo *et al.* 1983). In other cases the experiment was done with the phrenic-triggered pump by triggering inflation from the offset of the phrenic burst. Inflation pressures of 5–6 cmH_2O were used. Fig. 5 shows the results of such an experiment. An expiration-synchronous sympathetic discharge, of shape and time course comparable to those of the expiration-synchronous sympathetic discharge shown in Figs. 2–4, was observed in thirteen out of fifteen cases tested. In addition, a prolongation of expiration over the control value in apnoea was observed. It must be mentioned that when inflation pressures in excess of 10·2 cmH_2O were used, inhibition, rather than excitation, of sympathetic activity in expiration occurred (Fig. 6), as previously shown by Gootman, Feldman & Cohen (1980). When the animal was ventilated with peak tracheal pressure of 7 cmH_2O at various repetition rates,

such that the inflation occurred entirely in inspiration (Fig. 7*A*), in late inspiration and early expiration (Fig. 7*B*) or entirely during expiration (Fig. 7*C*), marked expiration-synchronous discharge was only observed when inflation was entirely in expiration. This discharge, as well as the phase-locking between respiratory and pump cycle, disappeared after bilateral cervical vagotomy. When central rhythmic respiratory activity was abolished by hyperventilation in air (end-tidal P_{CO_2} 15 mmHg)

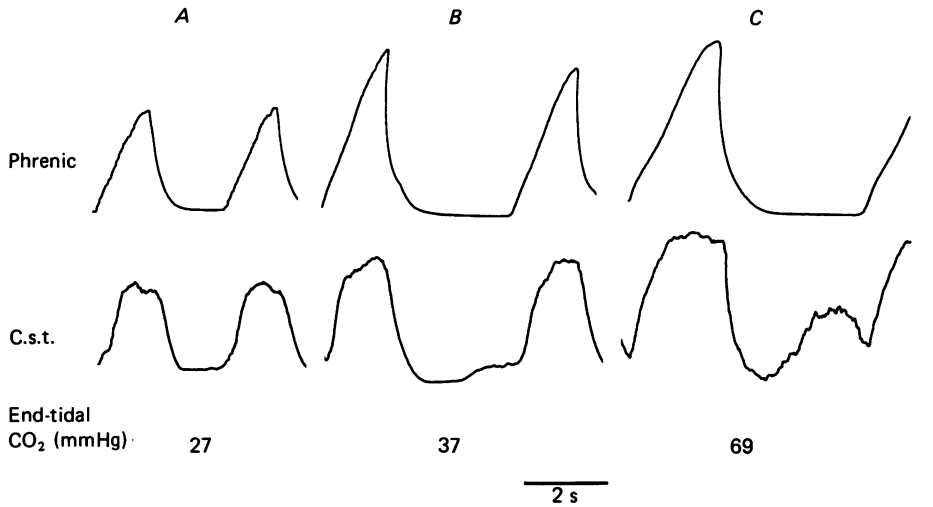

Fig. 8. Effect of systemic hypercapnia on the discharge of the cervical sympathetic trunk in expiration. Decerebrate sino-aortic denervated, vagotomized cat. From above, integrated phrenic nerve and cervical sympathetic trunk (c.s.t.) activities (average of twenty sweeps). *A*, in normocapnia, cervical sympathetic trunk activity in expiration is of the tonic type. During progressive hypercapnia, the level of cervical sympathetic trunk activity in expiration increases in *B* and assumes a wave-like shape at the highest end-tidal P_{CO_2} values (*C*). Slowing of the central respiratory rhythm in hypercapnia (as in *B* and *C*) in a similar preparation has been described before (St. John, 1979).

lung inflation at the same repetition rate and with the same peak tracheal pressure caused a small depression of sympathetic discharge.

Patterns of cervical sympathetic trunk activity in expiration during systemic hypercapnia

In eleven vagotomized, sino-aortic-denervated cats, the pattern of cervical sympathetic trunk activity was studied at various levels of arterial P_{CO_2}. Records from such an experiment in a decerebrate cat are shown in Fig. 8. With increasing CO_2 levels, in addition to the progressive increase of the peak amplitude of the inspiration-synchronous component of sympathetic discharge, previously described (Preiss & Polosa, 1977), there was also a progressive increase of the discharge in expiration, which had a wave-like appearance as in Figs. 2–7.

DISCUSSION

This paper reports the observation that the discharge, recorded in the cervical sympathetic trunk, has a component, time-locked to the respiratory cycle, which

occurs in expiration. This component is detected infrequently in control conditions but is consistently observed during moderate increases in F.R.C. and during hypercapnia.

Concerning the mechanism by which the increase in F.R.C. evokes the expiratory sympathetic discharge, the range of effective tracheal pressures and the abolition of this discharge by bilateral cervical vagotomy suggest that these sympatho-excitatory effects of increased F.R.C. are mediated by pulmonary stretch receptors (Sant'Ambrogio, 1982). The fact that systemic hypocapnia, which has negligible influence on pulmonary stretch receptor discharge (Kunz, Kawashiro & Scheid, 1976; Bradley, Noble & Trenchard, 1976), eliminates the expiratory sympathetic discharge caused by an increase in F.R.C., suggests that not only the discharge of pulmonary stretch receptors, but also a certain level of activity of brain-stem respiratory neurones is necessary for the increase in F.R.C. to cause the expiration-synchronous sympathetic discharge. The observation of rhythmic expiratory sympathetic discharge during static lung inflation in normocapnic animals with intact vagus nerves shows that the facilitation of sympathetic discharge in expiration caused by an increase in lung volume is locked to the central respiratory cycle rather than to the pump cycle. This set of observations suggests the inference that the expiratory sympathetic discharge results from the activity of neurones, with the rhythmicity of respiration, which are activated by sensory input from the lungs (presumably originating from pulmonary stretch receptors) and, in addition, by CO_2.

A number of analogies between the properties of the expiratory sympathetic discharge on one hand and the properties of the activity of expiratory motoneurones and brain-stem expiratory neurones on the other suggest the brain-stem expiratory neurones as the source of the expiratory sympathetic discharge. Expiratory α-motoneurones show little or no activity during normal ventilation (Sears, 1964; Bishop, 1967). This is consistent with the low level of sympathetic activity during expiration in normocapnia (see Results). Sympathetic activity increases with moderate increases in F.R.C. which have been shown to recruit expiratory motoneurones (Bishop, 1967; Russell & Bishop, 1976; Dimarco, Stroml & Altose, 1984; the present study). At large values of F.R.C., which cause inhibition of expiratory motoneurones (Sommer, Feldman & Cohen, 1979), expiration-synchronous sympathetic discharge was never observed. In hypocapnia, expiratory motoneurones lose their rhythmicity and at very low values of arterial P_{CO_2} they become silent (Bainton *et al.* 1978; Bainton & Kirkwood, 1979). In this condition, the excitatory effect of moderate lung inflation on these neurones is lost (Barillot & Dussardier, 1976) as is on the sympathetic preganglionic neurones (see Results). Brain-stem expiratory neurones show responses similar to those of expiratory α-motoneurones. During lung inflation above F.R.C. brain-stem expiratory neurones show facilitation at transpulmonary pressures in the normal tidal range or slightly above it (Cohen, 1969; Bianchi & Barillot, 1975; Feldman & Cohen, 1978; Baker, Frazier, Hanley & Zechman, 1979) and inhibition at transpulmonary pressures markedly higher than normal (Koepchen, Klussendorf & Phillip, 1970; Bianchi & Barillot, 1975; Cohen, Feldman & Sommer, 1985). The firing rate and burst duration of these neurones increase during systemic hypercapnia (Cohen, 1968).

As stated above, the vagal afferents evoking the expiration-synchronous sympathetic discharge during the experiments of increased F.R.C. and of inflation in expiration are probably those associated with the low threshold, slowly adapting pulmonary stretch receptors which cause the Hering–Breuer expiratory-facilitatory reflex (Fishman, Phillipson & Nadel, 1973; Farber, 1982). These manoeuvres caused shortening of the phrenic nerve discharge and prolongation of expiration (see Results; also Figs. 3–7) consistent with the hypothesis that they were evoking the Hering–Breuer reflex (Bartoli *et al.* 1973).

The present results that large inflations in expiration (tracheal pressure in excess of 10 cmH_2O) cause sympathetic inhibition are consistent with those of Gootman *et al.* (1980). Thus, lung inflation, in the present experiments, had a dual effect on sympathetic preganglionic neurones in normocapnic cats with intact vagus nerves: it caused facilitation at low transpulmonary pressures, and inhibition at higher transpulmonary pressures. This dual effect can be explained with the hypothesis that the excitation, caused by inflation in expiration, is probably secondary to activation of expiratory neurones, as discussed above, whereas the inhibition is due, at least in part, to a primary reflex effect, independent of the respiratory neurones, since the inhibition is present in hypocapnia.

The observation, described in the present paper, that lung inflation may cause excitation of sympathetic neurones, leads to the prediction that under appropriate conditions, namely at some inflation volumes, the expiration-synchronous excitation may outweigh the depressant effect, with the net result that the activity level of these neurones may increase during lung inflation. Some observations in the literature (Hainsworth, 1974) are consistent with this prediction. In the dog, static inflation of an innervated lung, while the other lung was denervated and ventilated, caused a small but significant increase in hind-limb vascular resistance at tracheal pressure values of 5–20 cmH_2O, while at higher tracheal pressures (20–40 cmH_2O) hind-limb vascular resistance decreased. These data show, therefore, a reversal of the effects of lung inflation, reminiscent of the reversal observed in some of the present experiments (e.g. Figs. 2*D* and 6*B*). It must be pointed out, however, that no such reversal was obtained in the experiments of de Burgh Daly *et al.* (1967) who found in dogs that lung inflation with transpulmonary pressures up to 7 cmH_2O (estimated tracheal pressure 11 cmH_2O, Spells, 1970) had no effect on hind-limb vascular resistance, while lung inflation with transpulmonary pressures in excess of 7 cmH_2O caused vasodilation. While the mechanism of this reversal has not been investigated yet, the observation that in order to block the reflex vasodilator effect of large lung inflations requires vagal cooling to 1 °C (de Burgh Daly *et al.* 1967), while it is known that the vagal afferents causing the Hering–Breuer reflex are blocked at between 8 and 5 °C (Fishman *et al.* 1973) suggests that receptors associated with non-myelinated afferents could be responsible for the sympatho-inhibitory effect of lung hyperinflation. Pulmonary receptors associated with non-myelinated afferents have been shown in open-chest dogs to be excited by hyperinflation (threshold 7 cmH_2O transpulmonary pressure), to be non-adapting and to cause reflexly apnoea, hypotension and bradycardia (Coleridge, Coleridge & Luck, 1965; Green, Schmidt, Schultz, Roberts, Coleridge & Coleridge, 1984).

Finally, in the case of the CO_2-evoked increase of sympathetic discharge in expiration, its mechanism may include, in addition to an increased facilitatory input from brain-stem expiratory neurones, an increase in sympathetic preganglionic neurone excitability caused by CO_2 (Zhang, Rohlicek & Polosa, 1982) resulting in increased responsiveness to excitatory input.

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