

THE INTERACTION OF REFLEXES
ELICITED BY STIMULATION OF CAROTID BODY CHEMO-
RECEPTORS AND RECEPTORS IN THE NASAL MUCOSA
AFFECTING RESPIRATION AND PULSE
INTERVAL IN THE DOG

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SUMMARY

1. The effects on respiration and pulse interval of stimulation of the carotid body chemoreceptors before, during and after stimulation of receptors in the nose have been studied in the anaesthetized dog.

2. Stimulation of a carotid body by infusion of cyanide into the ipsilateral common carotid artery causes hyperpnoea and either an increase, decrease or no change in pulse interval.

3. Excitation of receptors in the nasal mucosa leads to reflex apnoea or a reduction in breathing, and an increase in pulse interval.

4. When the carotid bodies are excited by the same dose of cyanide during stimulation of the nasal mucosa, the chemoreceptor-respiratory response is abolished or reduced in size compared with the control effect. On the other hand, the chemoreceptor-cardio-inhibitory response is considerably enhanced.

5. The potentiated cardio-inhibitory response of combined chemoreceptor and nasal stimulation could not be accounted for by the change in pulmonary ventilation, arterial P_{O_2} or P_{CO_2} , or mean arterial blood pressure.

6. These results indicate that excitation of the nasal reflex inhibits the chemoreceptor-respiratory reflex response but facilitates the chemoreceptor-cardio-inhibitory reflex response. The possible sites of these interactions between the nasal and chemoreceptor reflexes are discussed.

INTRODUCTION

When marine mammals and aquatic birds submerge in water, apnoea occurs together with changes in the cardiovascular system which enable the organism to conserve oxygen and remain submerged for periods of time

longer than would otherwise be possible. These changes include bradycardia and vasoconstriction in skin, muscle, alimentary tract and kidneys (see Andersen, 1966). The cessation of breathing is reflexly engendered and is due to stimulation of receptors in and around the nose or on the face, the afferent pathway being the trigeminal nerves (Huxley, 1913; Andersen, 1963). A vagal reflex from receptors in the glottis may also be involved (Butler & Jones, 1968).

With regard to the initiation of the cardiovascular responses, at least two reflex mechanisms are responsible: First, a trigeminal nerve reflex (Andersen, 1963), and secondly, an arterial chemoreceptor reflex involving stimulation of the carotid bodies by progressive hypoxia and hypercapnia (Jones & Purves, 1970; Holm & Sorensen, 1972). These findings suggest that the pattern of the reflex responses to stimulation of the carotid bodies may be modified during diving since in this situation the chemoreceptors can apparently evoke bradycardia without at the same time stimulating breathing.

Since these trigeminal nerve and carotid body reflexes are also operative in terrestrial animals (Kratschmer, 1870; Heymans, Brouckaert & Dautrebande, 1931 *a, b*; Daly & Scott, 1958; Angell James & Daly, 1969 *a, b*, 1972 *a, b*) an attempt to throw light on this problem was made using dogs in which the respiratory and cardiac responses to stimulation of the carotid body chemoreceptors were elicited before, during and after excitation of receptors in the nasal mucosa. Some of our results have been published briefly elsewhere (Angell James & Daly, 1972 *c*).

METHODS

Dogs of either sex, varying in weight from 10.0 to 15.4 kg, were anaesthetized with morphine hydrochloride (1 mg/kg s.c.) followed 30 min later by an i.v. injection of a mixture of α -chloralose (Kuhlmann, Paris; 0.05 g/kg) and urethane (British Drug Houses Ltd.; 0.5 g/kg) dissolved to make a solution of 10 g α -chloralose and 100 g urethane in 100 ml. of a solution containing 85 parts sodium chloride solution (0.9 g/100 ml.) and 15 parts polyethylene glycol ('Carbowax', Union Carbide Ltd.).

Stimulation of the carotid body. The carotid body chemoreceptors were stimulated by sodium cyanide (concentration of solution 1 mg/ml.) infused into a common carotid artery. For this purpose a catheter (Nylon Intravenous Cannula, type 5 FG, o.d. 1.65 mm, length 7 cm; Portex Ltd., Hythe, Kent, England) was inserted through a superior thyroid artery into the common carotid artery. The tip of the catheter lay about 3 cm caudal to the origin of the superior thyroid artery.

To ensure that the same dose was given each time in any one series of observations in one animal the cyanide was infused by a micro-metering pump (MHRE/Delta, Watson-Marlow Ltd., Falmouth, Cornwall) connected to the catheter in the carotid artery via a three-way tap. The pump ran continuously, the cyanide normally being directed through the third limb of the tap into a small conical flask. An infusion into a carotid artery was made by turning the tap in such a way as to shut off the tube

to the flask and bring the pump into direct communication with the catheter. Infusions were made for 3 or 5 sec periods, the dose being adjusted by altering the speed of rotation of the pump.

To minimize capacity effects in the tubing between the pump and the tap through changes in pressure when the tap was turned, the pressure against which the pump operated was maintained relatively constant by fixing the conical flask about 160 cm above the level of the heart, that is at the same level as the mean arterial pressure.

In each experiment initial trials were carried out to find the dose of cyanide which gave brief respiratory and cardiac responses. This dose varied from 2.25–8.7 $\mu\text{g}/\text{kg}$ in different animals.

Stimulation of receptors in the nose. Tap water or sodium chloride solution (0.9 g/100 ml.) was drawn over the nasal mucous membrane as described previously (Angell James & Daly, 1969b, 1972a).

Respiration. Tidal volume was measured by a balanced spirometer connected to a closed-circuit respiratory system (Bacon, Daly & Scott, 1962). The counter-weight for the spirometer formed the core of a linear displacement transducer (S.E. Laboratories Ltd., Feltham, Middlesex) which was connected to a carrier amplifier (S.E. Laboratories Ltd.). The animals breathed 100% oxygen throughout each experiment.

Calculations of respiratory minute volume were made from measurements of tidal volume and respiratory frequency, and were corrected for body surface area using Meeh's formula, where $k = 0.112$. The control values for respiratory minute volume were computed from the sum of the values for tidal volume for each breath, measured at a.t.p.s., and the number of breaths over a period of 1 min. During stimulation of the nasal mucosa, the period taken was that of the duration of the stimulus. The responses to stimulation of the carotid chemoreceptors were analysed in a similar way, the measurements of accumulated tidal volume being made only over the period of the increased respiration and expressed in terms of ventilatory volume (l./min. m^2). The chemoreceptor responses which occurred on a background of apnoea while the nasal mucosa was being stimulated, were often small and consisted of only one or two breaths. In these cases the accumulated tidal volume was measured over a period of time of similar duration to that measured during the control chemoreceptor response, on the assumption that the duration of the carotid body 'drive' was the same in the two conditions.

In some experiments pulmonary ventilation was maintained constant by means of a Starling 'Ideal' pump. A bilateral open pneumothorax was created and the lungs collapsed against an expiratory pressure of 2–4 cm H_2O in different experiments.

Measurement of pressures. Arterial blood pressure was measured from a brachial or femoral artery by way of a flexible nylon catheter connected to a Statham strain-gauge (model P23Gb). The undamped natural frequency response was greater than 130 Hz the degree of damping being 0.2. This gave an estimated amplitude distortion or less than 5% up to about 40 Hz. Mean arterial pressure was obtained electrically by passing the output of the amplifier through a simple R-C network with a time constant of 1 or 2 sec, and was recorded separately. The manometers were calibrated with a mercury manometer and zero reference pressures were obtained post mortem and taken as those recorded when the tips of the catheters were exposed to air.

Pulse interval. Measurements of pulse interval were made between the beginning of the upstrokes of the arterial pressure wave. In the control periods the average length of all the cardiac cycles occurring during the expiratory phase of five consecutive breaths was determined, the intervals during inspiration being excluded. During stimulation of the nose, the average pulse interval during the first 15 sec of the apnoeic period was taken, but in those tests in which apnoea did not occur, the breathing being diminished, the pulse interval was calculated in the same way as for

the control periods. In the case of the brief cardiac responses produced by stimulation of the carotid body, the length of the cardiac cycle coinciding with the beginning of the expiratory pause was chosen, because in many instances the pause was of such short duration. During the apnoeic period produced by stimulation of the nose, the peak chemoreceptor response was measured.

Heart rate was recorded continuously using a pulse frequency meter (J. F. Tonnies, Frieberg im Breisgau, Western Germany) which was triggered by the output signal of the Statham strain-gauge measuring arterial blood pressure.

Regional blood flow. Measurements of mean blood flow were made in the carotid artery into which cyanide was infused, using an electromagnetic flowmeter (Nycotron, Oslo, Norway). A non-cannulating transducer was applied to the artery about 2 cm caudal to the tip of the indwelling catheter. In some experiments mean blood flow was measured simultaneously in a femoral artery.

Zero reference flow was obtained at intervals during each experiment by temporary occlusion of the vessel distal to the transducer. At the end of each experiment the probe was calibrated *in situ*. The vessel was cannulated distally and the animal's own blood was run into a measuring cylinder at different rates which were timed with a stop-watch. In all cases there was a linear relation between blood flow and galvanometer deflexion, the line passing through the point of origin.

Vascular resistance was calculated as mean arterial pressure (mm Hg)/blood flow (ml./min) and expressed in convenient units representing the pressure necessary to force blood at 1 ml./min through the vascular bed under test. Changes in venous pressure were relatively small (Angell James & Daly, 1972*a*) and were therefore ignored.

Blood gas analysis. Samples of arterial blood were drawn anaerobically into oiled syringes and analysed immediately. The P_{O_2} , P_{CO_2} and pH of the blood was determined using a Blood Microsystem (Type BMS3, Radiometer, Copenhagen, Denmark) in conjunction with a digital acid-base analyser (Type PHM72, Radiometer). Estimations were made at a temperature corresponding to that measured in the rectum of the animal (37.0–38.5° C).

While the surgical procedures were being carried out, an infusion of a mixture of four parts of Dextran ('Macrodex 6%' in sodium chloride solution (0.9%, w/v), Pharmacia, Uppsala) and one part of sodium bicarbonate solution (2 mM) was administered intravenously at a rate of 2–3 ml./kg. hr to maintain a normal acid-base balance.

The initial arterial blood P_{O_2} was always greater than 305 mm Hg; the mean value (\pm s.e. of mean) for the P_{CO_2} was 43.8 ± 1.1 mm Hg, and the pH 7.324 ± 0.01 (eight experiments).

Experimental procedure. In each series of observations, four tests were carried out: (1) excitation of the nasal mucosa by water; (2) stimulation of the carotid body chemoreceptors by the intracarotid injection of cyanide; (3) stimulation of the chemoreceptors during excitation of the nose, and (4) repeat stimulation of the chemoreceptors alone. The values for respiratory minute volume and pulse interval obtained for the two control injections of cyanide were averaged.

RESULTS

Stimulation of the nasal mucous membrane

Stimulation of the nasal mucosa by water or sodium chloride solution caused inhibition of breathing in the expiratory position or a reduction of breathing, and usually slowing of the heart. In seven of the eight animals,

apnoea occurred in twenty one of twenty-two tests (Fig. 1A); in the remaining test, there was a reduction in breathing. In the eighth animal no apnoea occurred, but there was a reduction in breathing in each of the five tests carried out. Individual responses for all twenty-seven tests in the eight animals are shown in Fig. 2. The resting control value for respiratory minute volume of 2.92 ± 0.16 l./min. m^2 (range 1.73–5.12) fell to 0.43 ± 0.17 l./min. m^2 (range 0–2.63) during stimulation of the nose.

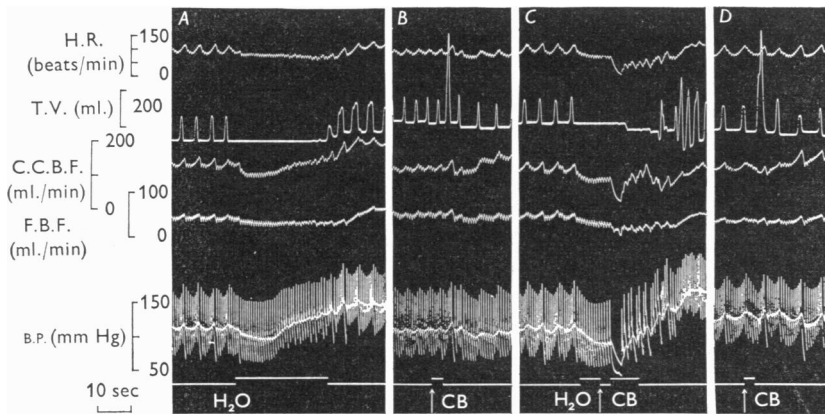


Fig. 1. The effects of stimulation of the carotid body chemoreceptors by cyanide ($3 \mu\text{g}/\text{kg}$) before, during and after stimulation of the nasal mucous membrane by water. Dog, male, 13.5 kg. *A*, nasal stimulation alone (H_2O). *B*, carotid body stimulation alone (CB). *C*, carotid body stimulation during stimulation of the nose. *D*, carotid body stimulation alone. H.R., heart rate; T.V., tidal volume (inspiration upwards); C.C.B.F., common carotid artery mean blood flow; F.B.F., femoral artery mean blood flow; B.P., mean and phasic blood flow. Time calibration, 10 sec.

In twenty-four out of twenty-seven tests stimulation of the nose increased the pulse interval; in the remaining three tests, it decreased. Taking all tests into consideration the pulse interval increased by 135.5 ± 18.6 msec (range -54 to $+330$), the control value being 582.3 ± 25.6 msec (range 359–800). This represents an increase of 23.3%.

Stimulation of the carotid body

In twenty-seven tests in eight experiments the infusion of sodium cyanide (2.25 – $8.7 \mu\text{g}/\text{kg}$) into a common carotid artery caused, after a latency of 1–2.5 sec, an increase in respiratory minute volume of 3.10 ± 0.25 l./min. m^2 (range 1.11–5.68), the initial control value being 2.92 ± 0.16 l./min. m^2 (range 1.73–5.12). This represents an average increase in minute volume of 106.2%. Typical responses are shown in Fig. 1*B, D*, and results of individual responses for all experiments are summarized in Fig. 2 (●—).

Sodium cyanide had variable effects on pulse interval. The predominant peak response during chemoreceptor stimulation was an increase in pulse interval which occurred in seventeen tests in eight animals (Fig. 1*B, D*). In three of these animals, however, additional tests produced either no change (six tests) or a reduction in pulse interval (four tests). Considering all twenty-seven tests together, there was an increase in pulse interval of 154.3 ± 29.1 msec (range -109 to $+577$), or 26.5%, the control value being 582.3 ± 25.6 msec (range 359–800).

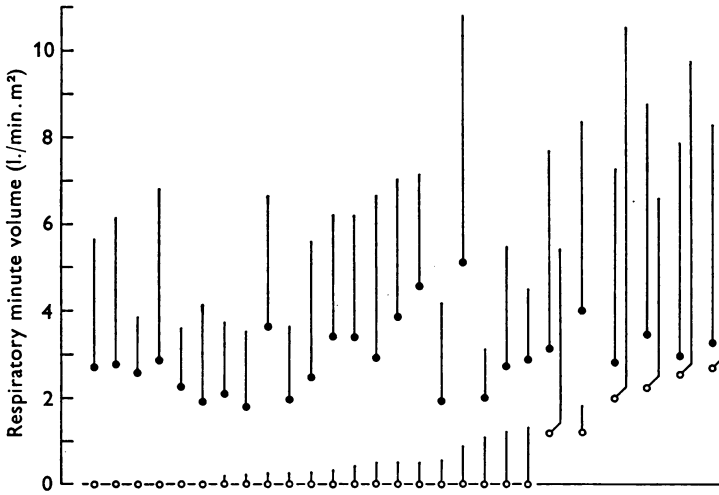


Fig. 2. The effects on respiration of stimulation of the carotid body chemoreceptors alone and during excitation of receptors in the nasal mucosa. Each vertical series of symbols represents a single series of observations (total twenty-seven).

Filled circles (●), values for the control respiratory minute volume; *open circles* (○), those for respiratory minute volume during stimulation of the nasal mucosa with water. The vertical lines are the responses to stimulation of the carotid body chemoreceptors alone (●—) and during excitation of the nasal reflex (○—). The observations from eight animals have been arranged in order according to the size of the chemoreceptor-respiratory response during the apnoeic period produced by stimulation of the nose.

The observed respiratory and cardiac responses to cyanide infusions were abolished by division of the carotid sinus nerve and are therefore reflex in nature. The maximum rate of infusion of cyanide corresponded to an injection of fluid of 0.17 ml./min.kg. In preparations in which the ipsilateral carotid sinus nerve was cut, this volume had no effect on carotid sinus pressure and would not therefore be expected to elicit a baroreceptor reflex in innervated preparations.

*Stimulation of the carotid body during excitation
of the nasal mucuous membrane*

In these tests the carotid body was stimulated during the period of excitation of the nasal mucous membrane, the infusion of cyanide being started 6–21 sec after commencement of the stimulus to the nose.

Respiration. In seven of eight animals, the hyperpnoea which occurred with control infusions of cyanide was either abolished (six tests) or reduced in size (sixteen tests) when the infusions were repeated during stimulation of the nose. Records from one such experiment are illustrated by Fig. 1. Whereas the control stimulations of the carotid body alone (*B*, *D*) caused increases in respiratory minute volume, infusion of the same dose of cyanide in the same time (3 sec) during stimulation of the nasal mucous membrane had no effect on respiration, except for a small reduction in the expiratory level of the lungs (*C*). This latter effect occurred 8 sec after the beginning of the infusion of cyanide and 5.5 sec after the onset of the cardiac response, and is not thought therefore to be the result of a primary reflex from the chemoreceptors. It was observed in other tests but its cause was not investigated further.

Slightly different results were obtained in five tests in the eighth animal. It was found that stimulation of the nose alone reduced the respiratory minute volume by only 32.4%. But whereas in three of the tests the respiratory response produced by carotid body stimulation during nasal stimulation was diminished compared with the control response, it was increased in the remaining two.

Comparison of all the control tests of stimulation of the carotid body alone with those during excitation of the nasal mucous membrane shows that the respiratory minute volume increased by an average of 3.10 ± 0.25 l./min.m² and 1.30 ± 0.43 l./min.m² respectively. A paired analysis of the data indicates that this difference is statistically highly significant ($P < 0.001$).

If the eighth experiment is excluded from the analysis because of the poor responses obtained on stimulation of the nose, the difference between the two values becomes more conspicuous, 2.7 ± 0.24 and 0.40 ± 0.08 l./min.m² respectively. The averaged responses in these seven experiments are shown in Fig. 3.

The individual responses to infusions of sodium cyanide are summarized in Fig. 2. It will be noted that in all those tests in which water to the nose caused inhibition of breathing, the respiratory response to cyanide when combined with stimulation of the nose, was absent or considerably reduced compared to that observed when the carotid body was stimulated alone. On the other hand when water to the nose did not cause apnoea but only

reduced breathing, there was in general, less inhibition of the respiratory response to cyanide; indeed in two tests, the response increased. It would appear from these results that the greater the reduction of breathing produced by stimulation of the nose, the greater the inhibition of the chemoreceptor respiratory response.

Pulse interval. In seventeen of twenty-seven tests in which stimulation of the carotid body alone caused an increase in pulse interval, the response was enhanced when the test was repeated during stimulation of the nose.

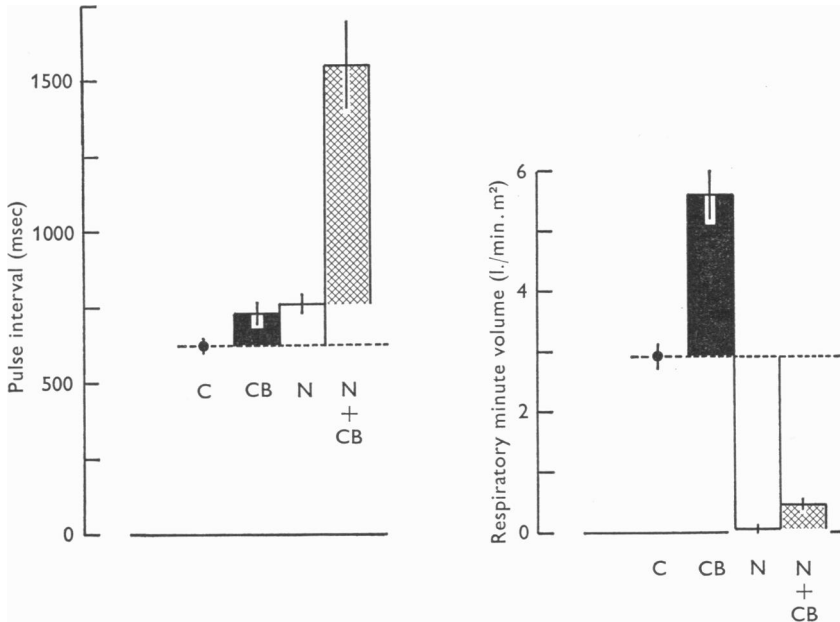


Fig. 3. The effects of stimulation of the carotid body chemoreceptors alone (CB, filled blocks) and during excitation of receptors in the nose (N + CB, cross-hatched blocks) on pulse interval and respiratory minute volume. Open blocks (N), stimulation of the nose alone. The filled circles (●), C, are the control values. The values represent the means \pm s.e. of mean from twenty-two tests in seven animals.

In six control tests in which there was no change in pulse interval, an increase occurred when each test was repeated during nasal excitation. In the remaining four control tests of chemoreceptor excitation which led to a decrease in pulse interval, an increase now occurred when the carotid body and nose were stimulated in combination. In all twenty-seven tests therefore stimulation of the carotid body during nasal stimulation led to an increase in pulse interval. The absolute value during water stimulation of the nose alone was 717.8 ± 36.9 msec (range 429–1000) and when the stimulus to the carotid chemoreceptors was superimposed, this increased

by 803.7 ± 112.1 msec (range 132–2779) to 1521.5 ± 123.9 msec (range 732–3529). This represents an average increase of pulse interval of 112.0%, compared with the value of 26.5% when the chemoreceptors were stimulated alone. This absolute value for the increase in pulse interval was considerably greater than that of 154.3 ± 29.1 msec obtained by stimulation of the carotid body without stimulation of the nose and a paired analysis of this data indicates that the difference is statistically highly significant ($P < 0.001$). It may be noted that the enhanced chemoreceptor-cardiac response observed during nasal stimulation occurred on a background heart rate which was slower than that without stimulation of the nose, the values being 717.8 ± 36.9 and 582.3 ± 25.6 msec respectively.

When compared with the control value for pulse interval, it was found in twenty-five of the twenty-seven series of observations that the cardiac responses to combined stimulation of the chemoreceptor and nasal reflexes were greater than the sum of their separate effects. This is also evident from the averaged results of the seven experiments shown in Fig. 3.

Denervation of the sympathetic supply to the carotid body. Stimulation of the receptors in the nasal mucous membrane by water causes an increase in resistance in a number of vascular territories through an increase in sympathetic adrenergic fibre activity (Angell James & Daly, 1972*a*). Such activity might also affect the carotid body vascular resistance and blood flow (Daly, Lambertsen & Schweitzer, 1954) and by producing local stagnant hypoxia, contribute to the enhanced cardiac effect observed when the carotid bodies are stimulated during nasal stimulation. In three experiments, therefore, the effects on pulse interval of stimulation of the nose and carotid chemoreceptors separately and in combination were studied before and after dividing the ipsilateral cervical vagosympathetic nerve at a level about 2 cm caudal to the origin of the superior thyroid artery.

It was found that, although the magnitude of the cardiac responses was diminished after vagosympathectomy, which could be attributed to the fact that some cardiac efferent fibres were cut as well, the increase in pulse interval elicited by the two reflexes stimulated in combination was greater than the sum of their separate effects.

Controlled pulmonary ventilation

The effects of stimulation of the carotid bodies on heart rate are determined to a large extent by the concomitant changes in respiration (Daly & Scott, 1958; Angell James & Daly, 1969*a*; Daly, 1972). To find out whether the difference in the cardiac responses observed when the carotid body is stimulated separately and combined with stimulation of the nose is determined by the different pattern of breathing in the two states, experiments

were carried out in which pulmonary ventilation was maintained constant throughout.

In each of four series of tests in three experiments the increase in pulse interval resulting from stimulation of the carotid body was greater when combined with excitation of the nasal mucosa. The mean increase in pulse interval produced by stimulation of the chemoreceptors alone was 235.5 ± 56.7 msec, or 49.6%, and when combined with excitation of the nasal receptors was 601.5 ± 189.0 msec, or 108.2%. Again the increase in pulse interval resulting from combined stimulation of the carotid body and the nose, 683.0 ± 190.5 msec, was greater than the sum of their individual effects, 235.5 ± 56.7 and 81.5 ± 32.6 msec respectively.

Relation to changes in blood pressure

There was no consistent relationship between the change in arterial blood pressure and the change in pulse interval on stimulation of the carotid chemoreceptors either alone or during stimulation of the nose (Fig. 4), the average control values for blood pressure under the two conditions being 125.1 ± 2.3 mm Hg (range 102–151) and 123.1 ± 2.3 mm Hg (range 104–146) respectively.

Relation to changes in carotid artery blood flow

Stimulation of the nasal mucosa causes variable changes in blood flow in the common carotid artery, which are determined largely by concomitant alterations in arterial blood pressure (Angell James & Daly, 1972*a*). It is possible therefore that the blood concentrations resulting from the intra-carotid injections of the same dose of cyanide with and without stimulation of the nose may be dissimilar and account, at least in part, for the differences in the respiratory and cardiac chemoreceptor responses under the two sets of conditions.

In twelve tests in four experiments the carotid flow at the beginning of the cyanide injection during stimulation of the nose was compared with the control value without nasal stimulation and was found to vary from 66.6 to 137.5% of the control value (mean 106%). In three further experiments in which the external carotid artery was ligated at the level of its bifurcation into the internal and external maxillary arteries, the common carotid blood flow varied from 60 to 153.3% of the control value (mean of twelve observations, 88.4%). Comparison of individual values for carotid artery blood flow corresponding to the time of injection of cyanide during the control state and during stimulation of the nose indicates that there is no consistent difference (Fig. 5). A similar conclusion was reached on the basis of analysis of similar data from experiments in which pulmonary ventilation was maintained constant.

In confirmation of the results of Angell James & Daly (1972a), stimulation of the nose had no significant effect on carotid artery vascular resistance in spontaneous breathing animals. The control and experimental values for vascular resistance were 1.63 ± 0.12 and 1.56 ± 0.18 mm Hg/ml. min respectively (twelve tests from four experiments; $P > 0.1$).

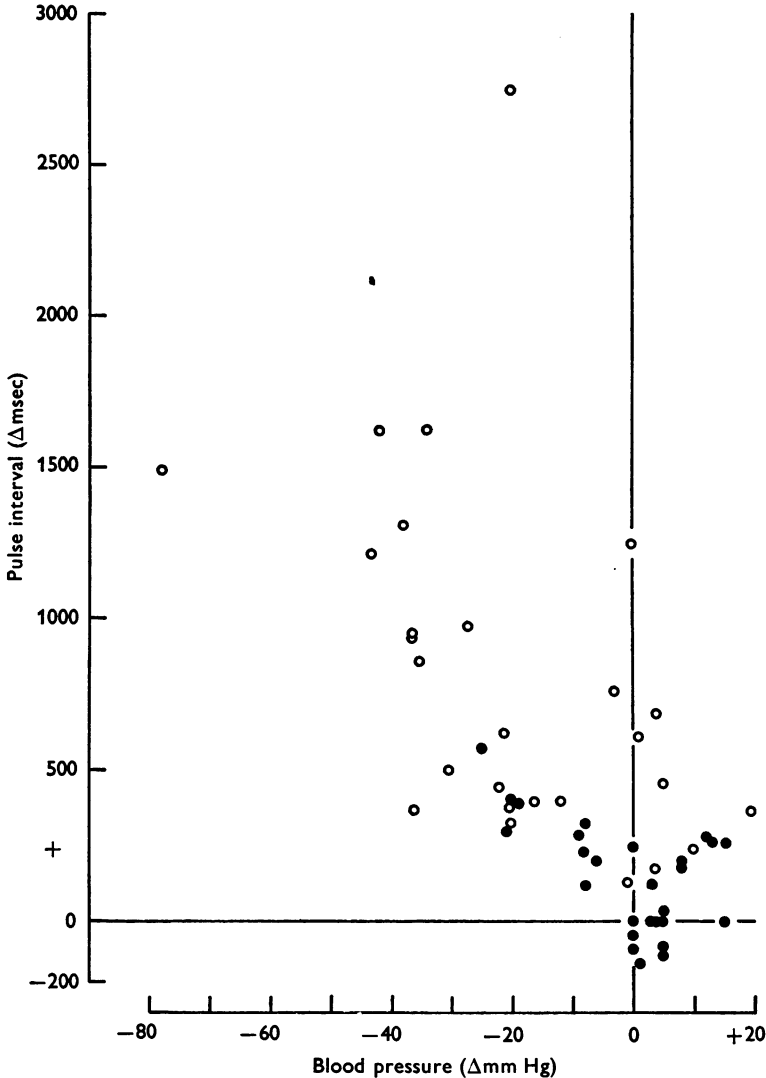


Fig. 4. The effects of stimulation of the carotid body chemoreceptors on the relationship between the change in mean arterial pressure and change in pulse interval. *Filled circles* (●), chemoreceptor stimulation alone; *open circles* (○), chemoreceptor stimulation during excitation of the reflex from the nose.

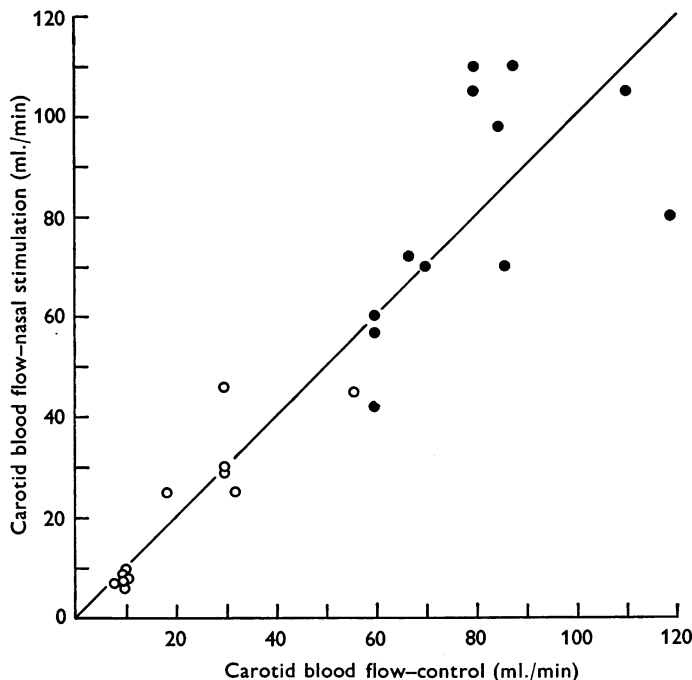


Fig. 5. Comparison of the values for common carotid artery blood flow at the time of injection of cyanide under control conditions (without stimulation of the nose) and during excitation of the nasal reflex. *Filled circles* (●) normal carotid circulation; *open circles* (○), external carotid artery ligated at level of bifurcation into the internal and external maxillary arteries.

Changes in femoral vascular resistance

In confirmation of the results of Angell James & Daly (1972*a*), stimulation of the nasal mucous membrane increased femoral vascular resistance in each of twenty-seven tests. The control value of 2.92 ± 0.25 mm Hg/ml.min (range 1.33–6.75) increased by 1.44 ± 0.31 mm Hg/ml.min (range 0.27–5.38), or by 45.6%.

In the majority of tests involving stimulation of the carotid body alone, no changes in vascular resistance occurred; in the remainder they were small and variable. During stimulation of the nose intracarotid infusions of cyanide caused large rapid fluctuations in arterial pressure and blood flow due to the profound cardiac chronotropic effects. The time constants of the instruments used were such that no meaningful values for mean pressure and mean flow could be obtained, and consequently no assessment of the changes in vascular resistance could be made.

DISCUSSION

We have confirmed previous observations that stimulation of the nasal mucous membrane reflexly causes apnoea in the expiratory position, or a reduction of breathing, and bradycardia (Kratschmer, 1870; Brodie & Russell, 1900; Dixon & Brodie, 1903; Angell James & Daly, 1972*a*), together with an increase in resistance in the femoral vascular bed (Angell James & Daly, 1972*a*). Our experiments have shown further that excitation of this reflex considerably modifies the pattern of the reflex responses produced by stimulation of the carotid chemoreceptors, in particular the respiratory and cardiac chronotropic effects.

Cyanide injected into a common carotid artery stimulates the carotid body chemoreceptors and has no effect on the nearby baroreceptors provided the carotid sinus blood pressure does not alter (Jacobs, Sampson & Comroe, 1971). As first shown by Heymans *et al.* (1931*a, b*) the reflex responses include hyperpnoea and bradycardia. These were the predominant effects seen in the present experiments but occasionally the heart rate remained unchanged or even increased. The probable reason for this is that although stimulation of the carotid bodies caused a primary reflex bradycardia, this response was masked or over-ridden by a secondary cardio-accelerator effect due to hyperventilation. The overall response therefore depends, amongst other factors, on a balance between these two mechanisms (Daly & Scott, 1958; Daly, 1972).

We did not observe consistent increases in femoral vascular resistance on stimulation of the carotid bodies, as in other studies (Bernthal, Motley, Schwind & Weeks, 1945; Daly & Scott, 1963; Daly & Ungar, 1966; Angell James & Daly, 1969*a*), and we attribute this to the use of small doses of cyanide acting to give a brief stimulus of sufficient intensity to excite only the respiratory and cardiac responses. Larger doses ($> 20 \mu\text{g}/\text{kg}$), however, regularly produced vasoconstrictor effects (J. E. Angell-James & M. de B. Daly, unpublished observations), as found by Calvelo, Abboud, Ballard & Abdel-Sayed (1970).

Since the reflex respiratory effects of excitation of the carotid body by a rapid injection of a stimulant depends on the phase of respiration the injection is made (Black & Torrance, 1971; Eldridge, 1972), the infusions were made in the present experiments over a period of time (3 or 5 sec) to cover more than one respiratory cycle. Thus repeated infusions of the same dose gave reproducible responses irrespective of the phase of respiration the infusion was started.

The blood concentration produced by infusions of cyanide of the same dose given over the same period of time will depend on the common carotid artery blood flow. Although the blood flow at the time of infusion

during stimulation of the nose was not always exactly the same as the control value, largely through concomitant changes in blood pressure (Angell James & Daly, 1972*a*), this did not materially affect the results. The modifications of the pattern of the chemoreceptor reflex responses produced by stimulation of the nose were the same irrespective of the small directional changes of blood flow and hence blood concentration of cyanide.

It is still a matter of conjecture the mechanisms by which the trigeminal nerve and chemoreceptor reflexes interact. Each reflex has as its efferent pathways the intercostal nerves and phrenic nerves for the respiratory responses, and the interactions could therefore be central or at a spinal level. The vagus nerves are largely responsible for their cardiac effects and presumably this interaction must be entirely central. However, two other possibilities must be mentioned. Firstly, the carotid body 'drive' during stimulation of the nose may be altered due either to increased sympathetic nerve activity, associated with the nasal reflex (Angell James & Daly, 1972*a*), acting on the carotid body (Daly *et al.* 1954; Purves, 1970) or to a change in activity of the efferent fibres from the brain stem that run in the carotid sinus nerves (Biscoe, Lall & Sampson, 1970; Neil & O'Regan 1971*a, b*). We have been able to exclude the first of these alternatives as a possible mechanism, but not the second. Secondly, the potentiation of the chemoreceptor-cardiac reflex by stimulation of the nose might be due to a peripheral vagal effect of increased secretions of supra-renal catecholamines engendered by the trigeminal nerve reflex (Allison & Powis, 1971). In this connexion adrenaline has been shown to potentiate the bradycardia produced by electrical stimulation of the vagus nerve (Kuroda & Kuno, 1915; MacWilliam, 1930). This mechanism is unlikely to be the sole cause of the enhanced chemoreceptor-cardiac response produced by nasal stimulation as it occurred when the two reflexes were excited within 6 sec, and before the increased secretions of catecholamines could have reached the coronary circulation.

There are a number of central mechanisms by which the trigeminal nerve-chemoreceptor interaction affecting the heart could take place: First, through direct connexions in the bulbar reticular formation between afferent fibres from the two types of receptor; secondly, by an effect occurring secondarily to changes in respiration; and thirdly, through a central modification of the reflex effects of the input from arterial baroreceptors. With regard to the second possibility, stimulation of the trigeminal nerve receptors inhibits the respiratory 'centres' and stops respiration with the lungs in the expiratory position. Mechanisms contributing to the bradycardia might, therefore, be brought about through a reduction in the 'irradiation' of impulses from the respiratory to the cardio-

inhibitory 'centres' (Anrep, Pascual & Rössler, 1936; Daly, 1972) and/or by a reduction in the input to the central nervous system from pulmonary inflation receptors (Daly & Scott, 1958; Daly, 1972). No detailed analysis of these possibilities has been made in the present experiments, except in so far as it has been shown that the potentiated cardiac effect occurring in response to combined stimulation of the trigeminal and chemoreceptor reflexes occurs under conditions in which ventilation, and presumably the input from the pulmonary inflation receptors, is maintained constant. However, too few observations have been carried out to enable a comparison to be made with the cardiac responses obtained in the spontaneously breathing animal, and consequently no conclusions can be drawn regarding the role of the pulmonary inflation receptors in this interaction. Again hypoxia and hypercapnia could not have participated in the cardiac responses in animals artificially ventilated, though their role during spontaneous breathing is unknown.

So far as the third mechanism is concerned, it is known that resetting of the arterial baroreceptor-cardiac reflex to a lower level of heart rate occurs in response to both stimulation of the carotid bodies (Angell James & Daly, 1969*a*) and the nasal mucosa (Angell James & Daly, 1972*a*). From the evidence presented in this paper, combined stimulation of the carotid bodies and nose leads to an enhanced cardio-inhibitory response at a similar or lower level of mean arterial blood pressure compared with that produced by either reflex evoked separately. This indicates that during combined stimulation of the two reflexes resetting of the baroreceptor-cardiac reflex occurs to a lower level of heart rate than is produced by stimulation of either reflex separately, on the assumption that there is little or no change in the sensitivity or gain of the reflexes. Although no quantitative data is yet available to support this contention it nevertheless points to the fact that the cardiac responses to separate and combined stimulation of the carotid bodies and nose can be interpreted and expressed in terms of changes in the set point of the baroreceptor-cardiac reflex.

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