VAGAL INFLUENCES ON RESPIRATORY REFLEXES: INTERACTION OF P.S.R. AND R.A.R. ON THE INFLATION AND DEFLATION REFLEX, THEIR ROLE IN LINKING RESPIRATORY CYCLES; AND POSTVAGOTOMY EFFECT OF P.D.G.

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ABSTRACT

VAGAL INFLUENCES ON RESPIRATORY REFLEXES: INTERACTION OF P.S.R. AND R.A.R. ON THE INFLATION AND DEFLATION REFLEX, THEIR ROLE IN LINKING RESPIRATORY CYCLES; AND POSTVAGOTOMY EFFECT OF P.D.G.

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There is evidence that changes in one respiratory cycle may influence subsequent cycles by a central mechanism. Thus the influence of P.S.R. and R.A.R. activity from within one respiratory cycle on subsequent cycles, which we have called "memory", needed to be examined in the determination of duration of expiration (tE) and inspiration (tI).

This study was designed to investigate the relative roles of P.S.R. and R.A.R. stimulation in expiration influencing tI amd tE over several subsequent breaths. In particular to investigate their role in linking respiratory cycles.

In 14 anaesthetized spontaneously breathing rabbits we studied the response of tI and tE to +ve and -ve pressure pulses of 20KPa applied to the lung at various stages in expiration before and during P.S.R. block with SO2.

Before P.S.R. block, +ve pressure pulses early in expiration generally shortened tE containing the pulse, applied later +ve pressure pulses lengthened tE. Positive pressure pulses after P.S.R. block,

and -ve pressure pulses before and after block always shortened tE. Regardless of sign of pulse tE was shortened in subsequent breaths before and after block.

The inspiration after negative pulse application was usually lengthened. After effective block -ve pulses rarely lengthened tI. Large shortening of tE containing the pulse was usually followed by a shortened tI. Positive pulses did not significantly effect the duration of tI. Regardless of sign of pulse tI was not usually changed but occasional large shortening occured in subsequent breaths before and after P.S.R. block.

This indicates that the tE containing the stimulation is governed by a balance between P.S.R. and R.A.R. activity. The tI following the stimulus is governed by a balance between "memory" of P.S.R. and R.A.R. activity. In the breaths following both tE and tI were influenced by "memory" of R.A.R. activity only. However "memory" of strong R.A.R. activity is required to affect tI.

During this study it was intended to use phenyldiguanide (P.D.G.) to test J receptor patency. Intravenous injections of P.D.G. have been used to provoke respiratory reflexes, these have been considered to be due mainly to stimulation of type J receptors. However although most workers demonstrated that vagotomy abolished or reduced these reflexes, some still had significant response

to P.D.G. after vagotomy. A study was conducted to resolve this difference and demonstrate the sites at which P.D.G. acts in rabbits.

We measured tE and tI in 10 anaesthetized spontaneously breathing rabbits. 50 μg/kg P.D.G. was given intravenously (via a catheter with its tip close to the right atrium) to the intact rabbit; after blocking epicardial receptors; immediately after bilateral cervical vagotomy; 15 minutes after vagotomy; and after the glossopharyngeal nerves were cut near the base of the skull.

The respiratory reflex after injection of xylocaine, 15 minutes after vagotomy, and after cutting the glossopharyngeal nerves was as pronounced as in the intact state, and consisted of an increase in frequency almost totally due to a reduction in tE. With injections given up to 3 minutes after bilateral vagotomy the respiratory response was greatly attenuated and variable. We suggest this question of timing may contribute to the differences seen by different groups of workers. It is clear that intravenous injection of P.D.G. is not an adequate test of J receptor presence in the rabbit.

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INTRODUCTION

The introduction will review the material relevant to vagal influences on respiratory pattern. Firstly the control of breathing will be briefly reviewed in terms of:

1) central control,
2) efferent activity,
3) afferent activity.

Then the vagal influence of pulmonary receptors will be examined more closely in terms of:

1) non myelinated fibres,
2) slowly adapting receptors with myelinated fibres,
3) rapidly adapting receptors with myelinated fibres.

Next the regulation of duration of inspiration and expiration will be discussed. Then the evidence for linking of respiratory cycles will be examined. And finally a review of the conflicts in the literature on the effects of phenyldiguanide will be made.

CONTROL OF BREATHING

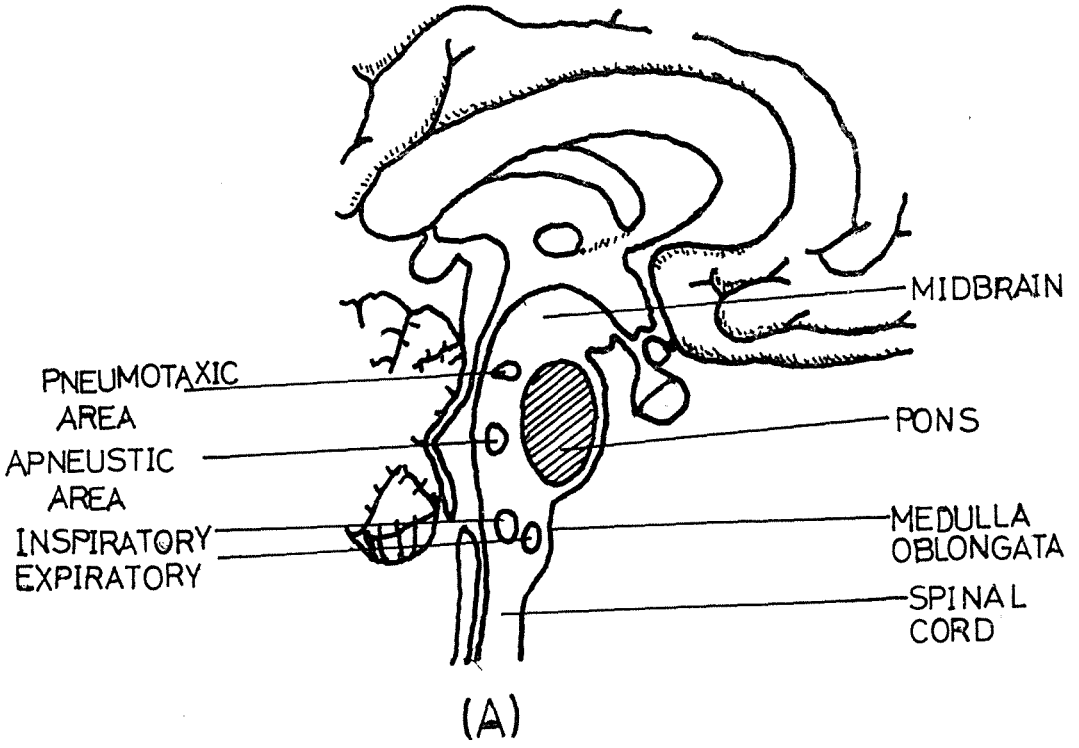
It is easy to take breathing for granted, we breathe approximately 16 times a minute without much thought until we over exert ourselves. And yet the pattern of breathing changes all the time

to keep maximum efficency despite altered oxygen needs and CO2 production with different activities.

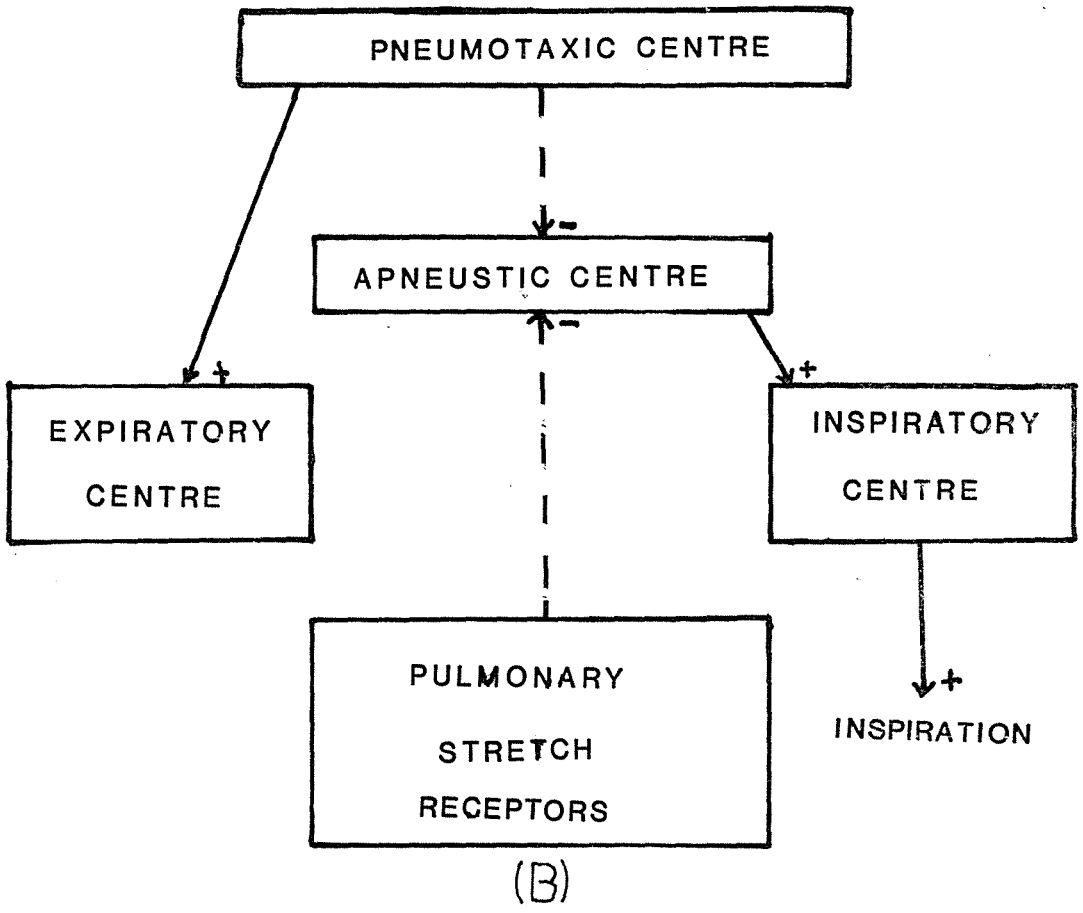
These changes in pattern are brought about by changes in the brainstem "respiratory centres" neural activity affecting efferent activity to the respiratory muscles. These changes are made in response to afferent information from receptors, located mainly in the thorax.

Central control

In 1812 LeGallois reported that if the medulla oblongata is isolated, cells within continue to generate a respiratory rhythm. Thus the medulla contains a respiratory pattern generator. Since Flourens 1851 described a "vital node" the concept of a small, bilateral, inherently rythmic centre has remained attractive. From early attempts to localize this centre this pattern generator was historically described as the "respiratory centres" (fig 1). These consisted of the inspiratory and expiratory centres which provided the oscillations from inspiration to expiration. The pattern from these was thought to be modified by the pneumotaxic centre which recieved vagal afferent information and the inhibitory apneustic centre. The pneumotaxic centre was considered to receive information on the onset of inspiration. After a delay on receiving this information the pneumotaxic centre was considered to inhibit the inspiratory



(A)



(B)

FIG 1. SCHEMATIC REPRESENTATION OF THE LOCATION (A) AND INFLUENCES (B) OF THE HISTORICAL "RESPIRATORY CENTRES".

FACILITATORY INFLUENCE

INHIBITORY INFLUENCE

neurons and terminate inspiration.

Pitts, Magoun & Ranson 1939 described the anatomical localization of overlapping inspiratory and expiratory neurons, dispelling the concept of discrete centres. Thus this concept of "respiratory centres" must now be redefined as a group of neuronsand synapses that influence the pattern of breathing. Cohen (1970 & 1976) proposed a model of interacting neuron systems which has replaced this model. This model differs from the historical mainly by describing functional rather than anatomical centres. Hence reference to the "respiratory centres" will refer to all the central neuron systems concerned with generating respiratory pattern.

Pitts (1946) considered the "respiratory centres" contain linked inspiratory neurons and linked expiratory neurons which synchronizes their activity. These inspiratory and expiratory neurons are mutually inhibitory which limits duration of activity during eupnoea (Comroe 1975). Although the expiratory neuron pool is active , in eupnoea this activity does not reach the threshold needed to activate expiratory motor neurons and therefore expiratory muscles. Thus in eupnoea expiration is passive (Comroe 1975). The "respiratory centres" may consist of inhibitory interaction between two groups of neurons to generate rhythm (Robson). This is a persistant idea with little evidence for it

(Mitchell & Berger 1975).

A model of the "respiratory centres" was proposed (von Euler & Trippenbach 1976; von Euler 1977; Cohen & Feldman 1977) consisting of functional pools of neurons generating the rhythm (fig 2). A pool of neurons generate the central inspiratory activity (CIA) which produces the basic pattern. This is terminated by another pool of neurons the inspiratory off-switch (O-S). A third pool of neurons is responsible for the interaction between the CIA pool and the activity from the pulmonary stretch receptors (P.S.R.). Once this pool reaches a threshold level of activity the O-S activity may rise quickly to terminate inspiration and CIA. The activity of the CIA-P.S.R. pool will die slowly with slow reduction of P.S.R. activity. Another pool of neurons may control rate of breathing with inspiratory and expiratory duration controlled by different sections of this pool. The inspiratory rate being inhibited by P.S.R. activity and the expiratory rate facilitating O-S activity.

All these pools are modified by afferent information (Trippenbach & Milic-emili 1977). The way in which afferent information modifies the "respiratory centres" will be discussed later. The CIA generator has efferents to the spinal respiratory motor neurons.

Efferent systems

The diaphragm is activated during inspiration

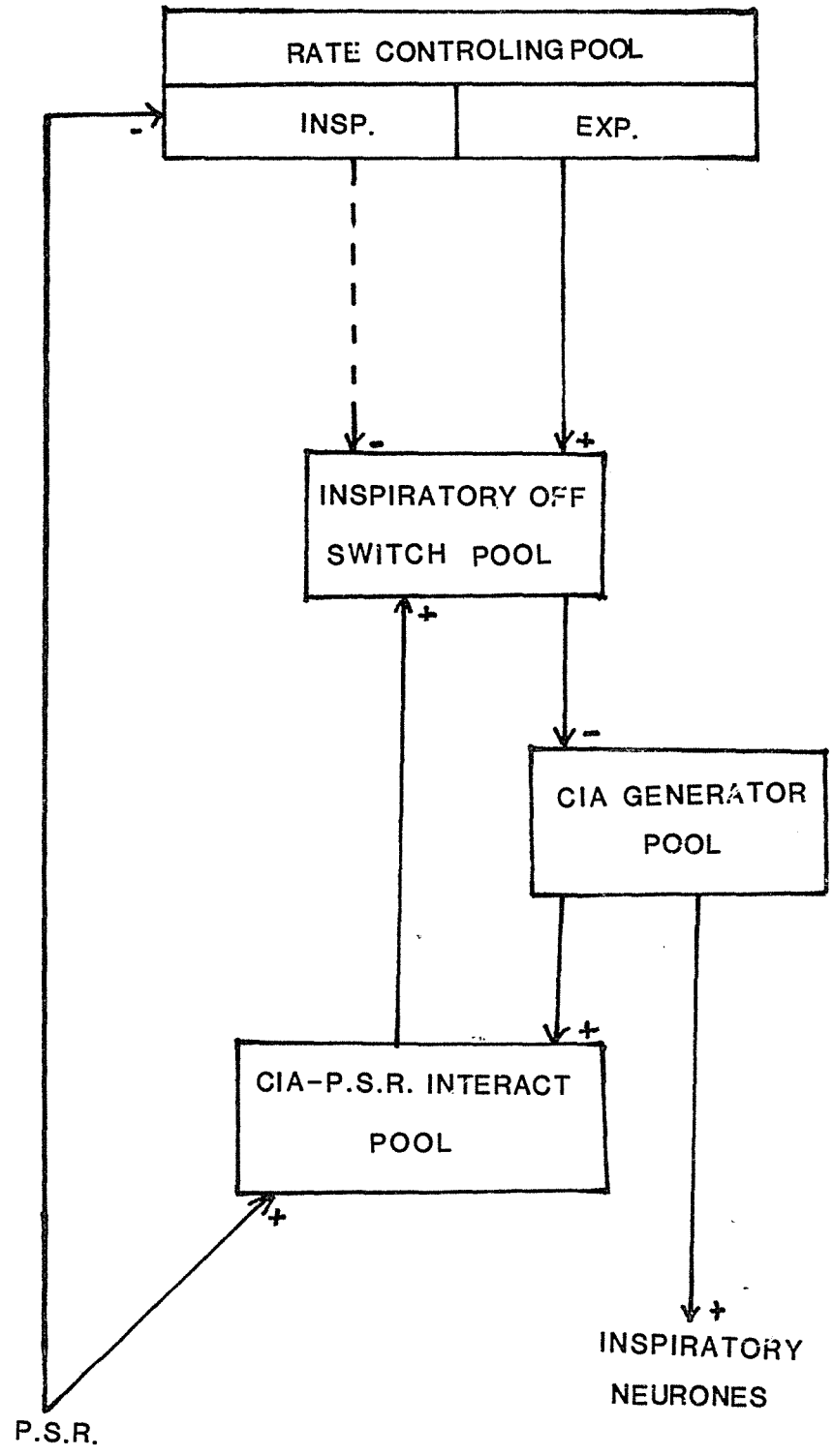


FIG 2. SCHEMATIC REPRESENTATION OF THE POOLS OF NEURONS FORMING THE CENTRAL CONTROL OF RESPIRATION.

+ → FACILITATORY

− → INHIBITORY

– – → ALTERNATIVES

by the phrenic nerve with neurons arising from cervical nerves C3, 4, and 5. These are efferents from the "rhythm generator" and are the only fibres in the phrenic nerve. The diaphragm has few muscle spindles thus the phrenic nerve is almost exclusively the source of diaphragmatic activity. The fibres of the phrenic nerve lack Renshaw cells with inhibitory feedback to prevent after discharge (Widdicmbe & Davies 1983).

Thus a recording of phrenic activity (fig 3) shows an accurate representation of the drive to inspire from the "respiratory centres", although some after discharge is seen. A recording of diapragmatic emg also records this drive to inspire. A trace of phrenic activity or diaphragmatic emg shows activity during inspiration and no activity during expiration.

The "respiratory centres" have efferent fibres to the other respiratory muscles, external and internal intercostal and abdominal muscles. However these muscles also have muscle spindles which produce reflex contraction (Widdicombe & Davies 1983).

Afferent systems

A major afferent influence on respiratory pattern is from chemoreceptors (fig 4). The main peripheral chemoreceptors are located in the carotid bodies and aortic bodies with afferents in

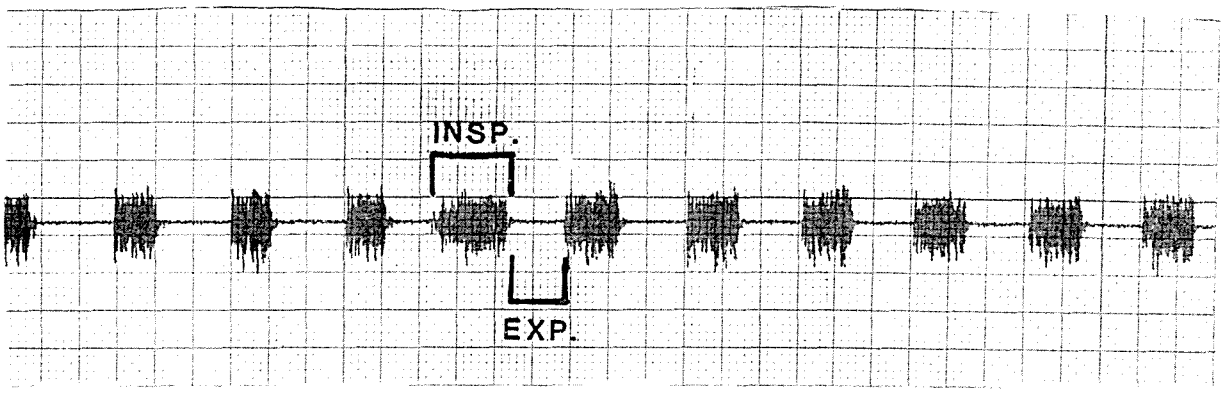


FIG 3. TRACE OF PHRENIC ACTIVITY.

INSP. SHOWS ACTIVITY OF PHRENIC DURING INSPIRATION (T_I).

EXP. SHOWS LACK OF PHRENIC ACTIVITY DURING EXPIRATION (T_E).

the glossopharyngeal and vagus nerves respectively. These receptors are stimulated by low arterial oxygen tension and increased levels of CO2 (Comroe 1975). However Guz, Noble, Widdicombe, Trenchard & Mushin 1966 showed block of these receptors did not influence pattern of breathing in eupnoea.

The central chemoreceptors are stimulated by an increase in hydrogen ion concentration in the cerebral spinal fluid. Hydrogen ion levels in the cerebral spinal fluid are related to CO2 levels. Thus stimulation of central chemoreceptors is due to raised levels of CO2 in the general circulation (Comroe 1975).

Stimulation of chemoreceptors increase frequency of breathing by inhibiting the inspiratory O-S pool of neurons and facilitating the CIA generator (von Euler 1977). There is a latency of 20-30 seconds after increasing CO2 levels before much change in breathing is seen and it may be 5-10 minutes before a new level of respiration is set (Widdicombe & Davies 1983). This illustrates that chemoreceptors have a long term influence on the pattern of breathing.

The pulmonary receptors, pulmonary stretch receptors (P.S.R.) and rapidly adapting receptors (R.A.R.), of the myelinated fibres of the vagus nerves are most important for breath by breath control of pattern of breathing. The R.A.R. are stimulated by inflation and deflation of the lungs

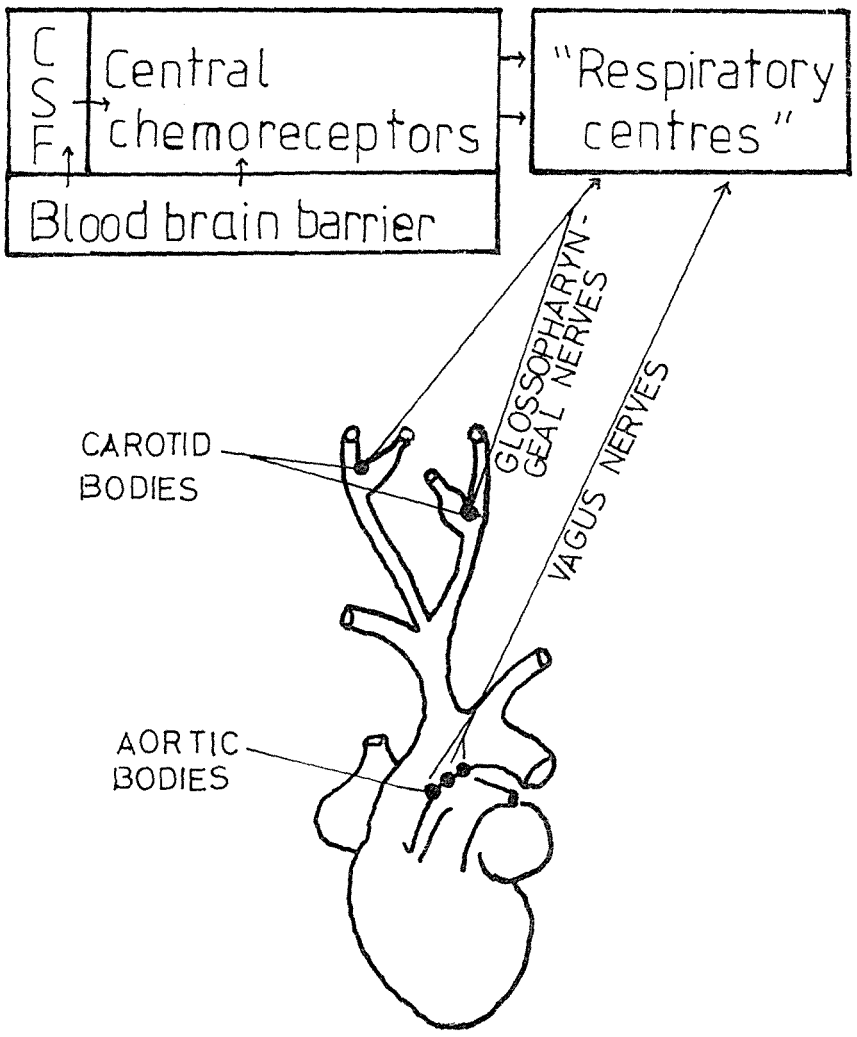


FIG 4. THE CHEMORECEPTORS.

to increase frequency of breathing (Widdicombe & Davies 1983). This is probably by stimulating the inspiratory O-S (von Euler 1977). Augmented breaths or sighs are also caused by R.A.R. stimulation (Davies & Roumy 1976), by large lung inflations and is probably by inhibition of the inspiratory O-S pool of neurons or by facilitating the inspiratory rate controlling pool (von Euler 1976).

The P.S.R. are stimulated by inflation of the lungs to terminate inspiration and initiate expiration (Widdicombe & Davies 1983). This is probably by interacting with the CIA-P.S.R. interaction pool of neurons to facillitate the O-S and by inhibition of the insiratory rate controlling pool (von Euler 1976).

The type J receptors (JR) have mainly nonmyelinated vagal fibres and are stimulated physiologically by pulmonary oedema to cause an increase in frequency of breathing (Paintal 1977). Their effect on the "respiratory centres" has not been established.

In unanaesthetized animals emotion and voluntary control may influence respiratory pattern. Voluntary control probably bypasses the respiratory centres and exerts its effect directly on the respiratory muscles (Widdicombe & Davies 1983). Emotion may influence the rate controlling pool in the pons (von Euler 1977).

Propioreceptors in the chest wall relay information about the position of the chest and allows efficient breathing regardless of body position. Activity from these fibres facillitate either the O-S pool or the CIA-P.S.R. interaction pool, thus shortening inspiration (von Euler 1977).

Hyperthermia affects the rate and growth of CIA and thus influences the CIA generator. It also interacts with the CIA-P.S.R. pool to activate the O-S (von Euler 1977).

Fig 5 is a diagragm of the afferent influences on the "respiratory centres".

PULMONARY RECEPTORS

The pulmonary receptors consist of type J receptors with nonmyelinatewd fibres, R.A.R.with myelinated fibres, P.S.R. which adapt slowly and have myelinated fibres (Paintal 1973b). In the cat there are 4 times as many non myelinated fibres as myelinated fibres from P.S.R. and R.A.R.(Agostoni, Chinnock, Daly & Murray 1957). The P.S.R. and R.A.R.are particularly important in breath by breath control of breathing, and help maintain an efficient breathing pattern.

Receptors of non myelinated fibres

The regenerative region of nerve fibres myelinated or not are not protected by the diffusion barrier of the nerve sheath (Paintal

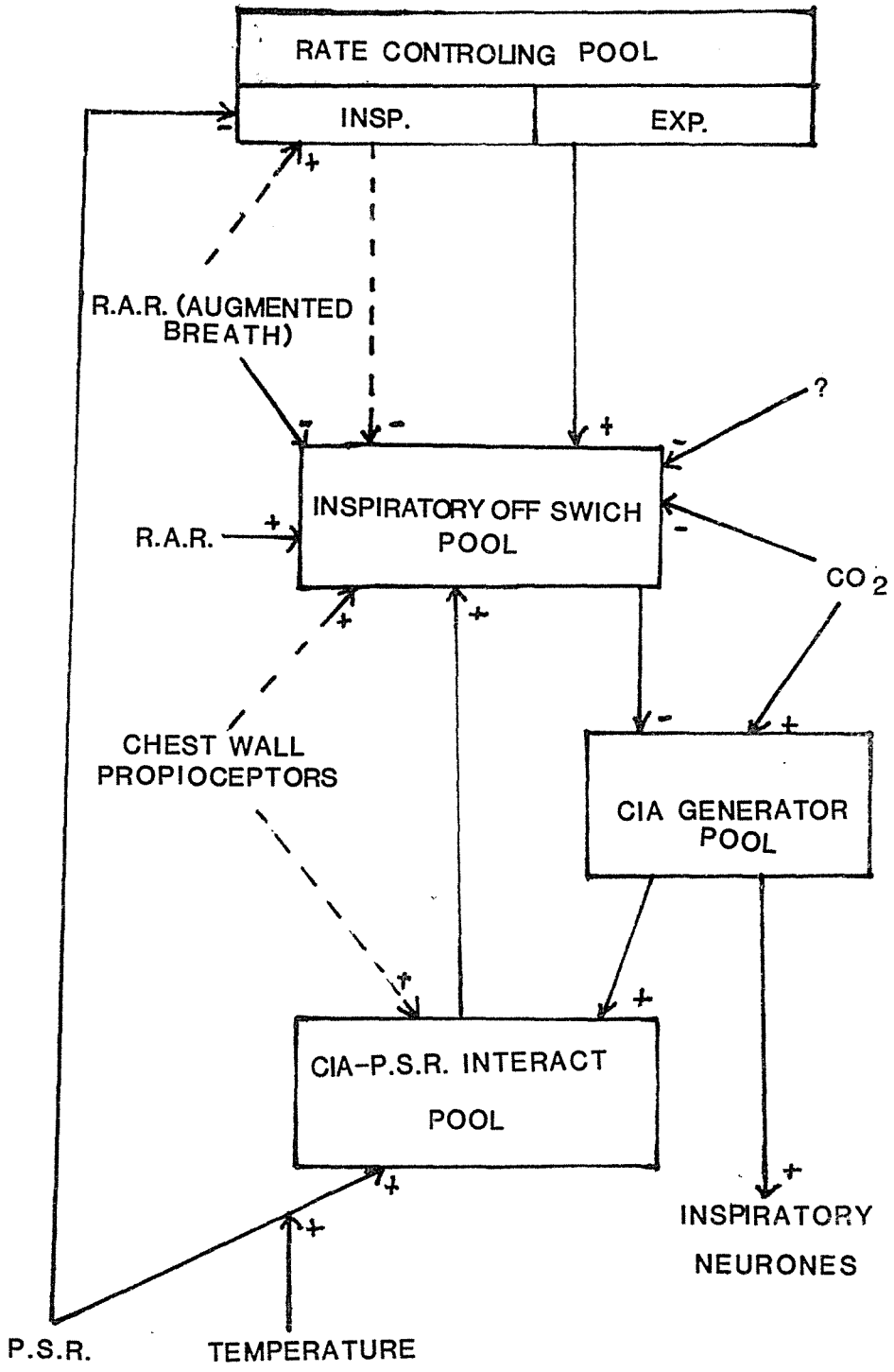


FIG 5. AFFERENT PATHWAYS TO THE "RESPIRATORY CENTRES".

+ FACILITATORY

− INHIBITORY

ALTERNATIVES

1973b). Thus some chemical types of chemical are able to stimulate or sensitize at this region examples of these are volatile anaesthetics and veratrum. Non myelinated fibres are susceptible to stimulation by a wider variety of drugs than myelinated fibres as the regenerative region is not protected by a myelin sheath (fig6). Acetylcholine, 5-hydroxytryptamine, phenyldiguanide, histamine and similar chemicals may only stimulate non myelinated fibres (Paintal 1973b).

Type J receptors were discovered accidentally while studying other nonmyelinated fibres using P.D.G. as a stimulant (Paintal 1953, 1954, 1973a). It was some time after discovery that the physiological stimulus of these receptors became known. Initially interest focussed on their response to forced deflation and collapse of the lung and hence were known as deflation receptors (Paintal 1973a). Sellick and Widdicombe 1970 found although most receptors were not stimulated by lung inflations or deflations some were stimulated by deflation produced by 50 ml pneumothorax.

Eventually it became known that these receptors were stimulated by pulmonary oedema, specifically the increase in interstitial volume consequent of rise in pulmonary capillary pressure (Paintal 1969). In keeping with this function it has been shown they lie close to the pulmonary

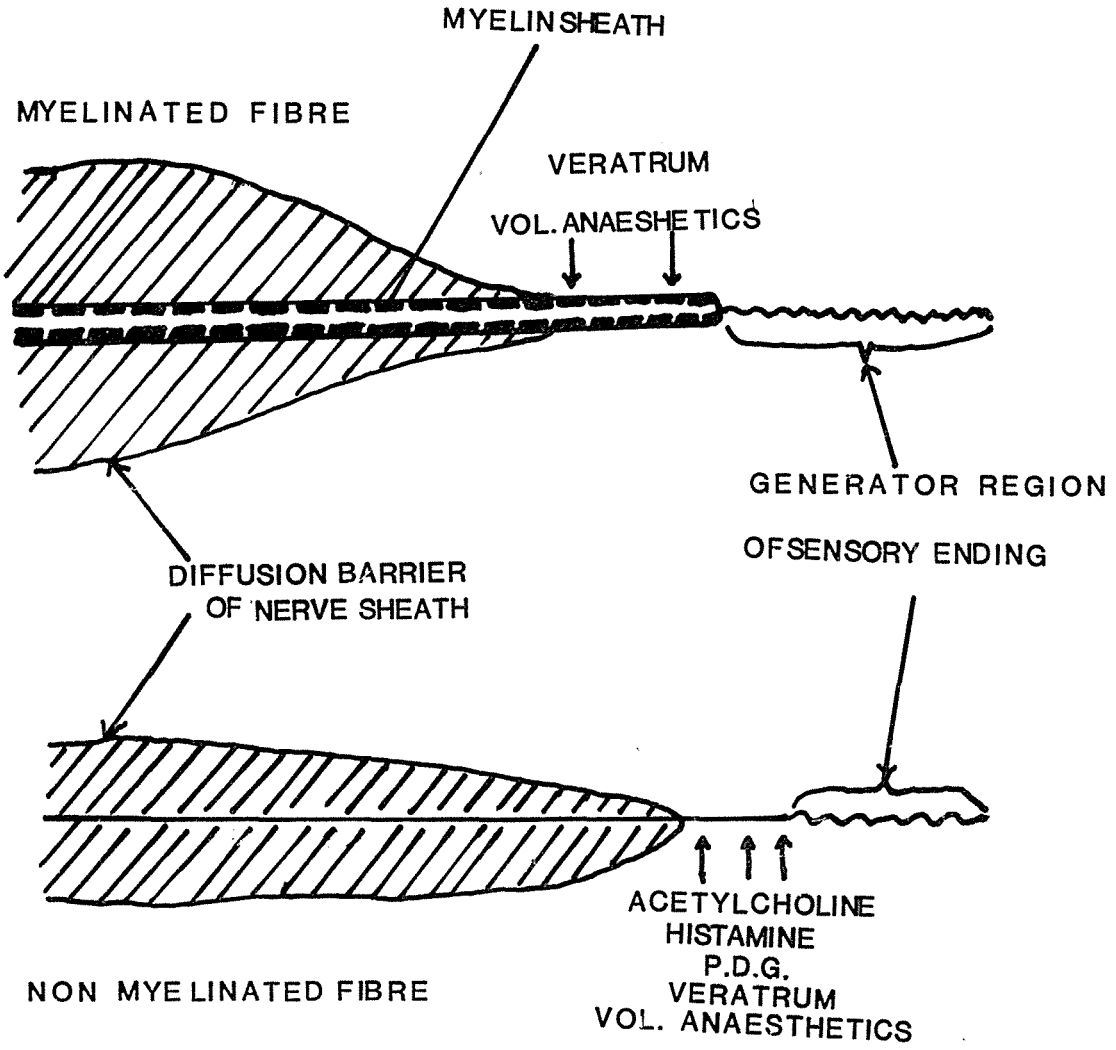


FIG 6. SCHEMATIC DIAGRAM OF MYELINATED AND NON MYELINATED FIBRES SHOWING THE RESPONSE TO CHEMICAL STIMULATION.

capillaries, giving rise to the name juxtapulmonary capillary receptors or type J receptors (Paintal 1973a).

Stimulation of J receptors accelerates breathing, and sometimes causes apnoea. It also causes hypotension, bradycardia and inhibition of somatic muscles (Paintal 1970, 1973b). Type J receptors are probably stimulated physiologically by a rise in pulmonary capillary pressure due to exercise. This may contribute to the acceleration of breathing and may be involved in the sensations of breathlessness (Paintal 1973a).

Substances such as 5-hydroxytryptamine stimulate these endings and as such may stimulate J receptors when released by pulmonary embolism. Other pathological conditions involving pulmonary circulation, suh as pulmonary oedema and embolism, stimulate J receptors, resulting in tachypnoea (Guz & Trenchard 1971). Sensations of dyspnoea in these diseases may be produced by J receptor stimulation (Paintal 1973a).

Slowly adapting receptors

Breuer and Hering in 1868 noted that inflation of the lungs in inspiration terminated inspiration while deflation terminated expiration, initiating inspiration. This effect was abolished by vagotomy. Breuer and Hering proposed that receptors which detected the state of the lung distension lay in

the lung tissue with the vagus as an afferent to the respiratory centres. They proposed that the respiratory centre which produced the normal respiratory pattern required vagal information about the state of the lung.

Adrian (1933) showed that when a sudden and maintained inflation is applied to the lungs, one type of vagal fibre discharges, the receptor of this fibre became known as the pulmonary stretch receptor. Davis, Fowler and Lambert (1956) suggested stretch from lung inflation was the primary stimulus with some responsiveness to the rate of change of stretch. Physiological and degeneration experiments along with histological evidence indicates they are located in the smooth muscle of the bronchi (Widdicombe 1954b). It has been noted these receptors are either slowly adapting or have varying adaption rates (Widdicombe & Davies 1983).

The main influence of P.S.R. is to shorten duration of inspiration accompanied by reduced tidal volume and lengthened duration of expiration (Widdicombe & Davies 1983). The P.S.R. are stimulated during the lung inflation of inspiration and activity rises till the O-S mechanism is activated, thus terminating inspiration (von Euler 1977). During the first part of expiration while the lungs are deflating, these receptors are still strongly active but discharge reduces as lungs empty (Widdicombe and Davies 1983). P.S.R. are

stimulated proportionally by mechanical deformation caused by transpulmonary pressure (Davis, Fowler, and Lambert 1956).

The inflation reflex exists in all mammals and operates qualitatively in the same manner in all of them. The strength of the reflex varies considerably, being strongest in rabbits and weakest in man (Widdicombe 1961). Since rhythmical breathing continues after bilateral vagotomy, the activity of lung stretch recetors is not essential for rhythm but modify the pattern. The advantage of this modification is to make breathing more efficient.

Although some receptors have superimposed cardiac rhythm (Paintal 1933) this is likely to be due to influence from local vessels rather than functional. This is supported by the observation that no cardiac rhythm is observed in these receptors when stimulated by inflation (Paintal 1973a). Recently it has been shown that P.S.R. can be inhibited by airway CO2 (Coleridge, Coleridge, & Banzett 1978), hypoxia does not similarly affect P.S.R. activity. P.S.R. also causes reflex bronchodilation, systemic vasodilation, tachycardia, and widening of the glottal apperture (Widdicombe 1982).

Rapidly adapting receptors

Impulses from endings which adapted rapidly to

maintained inflation or deflation of the lungs were recorded by Knowlton and Larrabee 1946. These endings were called rapidly adapting receptors. Widdicombe 1954a located many of these receptors in the trachea. Further investigation revealed endings in intrapulmonary airways (Mills, Sellick, & Widdicombe 1970) which lie beneath the respiratory tract epithelia. These have similar responses to those in the trachea. These endings have been named irritant receptors. The greatest concentration of these receptors are in the large airways (Sant'Ambrogio 1982).

There is much debate on the naming of these receptors. Although a natural stimulus to these receptors may be mechanical irritation, these receptors also play a role in determining pattern of breathing. I consider the term rapidly adapting receptor more appropriate in terms of their response to stimulus, I will thus refer to them as rapidly adapting receptors.

The physiological properties of extrapulmonary and intrapulmonary rapidly adapting receptors is identical (Paintal 1973a). In the cat R.A.R.are silent during eupnoea (Widdicombe 1954a, Knowlton & Larrabee 1946), even during moderate increase in tidal volume, while in the rabbit, R.A.R.show activity during spontaneous respiration (Sellick & Widdicombe 1970). Activity in the rabbit usually consists of a brief burst near the peak inspiration

(Sellick & Widdicombe 1970). Davies and Roumy 1982 recording R.A.R.activity, recorded greatest stimulation by deflation at functional reserve capacity and by deflation at peak tidal volume.

Stimulation of these receptors may cause tachypnoea, mainly shortening expiration

(Widdicombe & Davies 1983) and may shorten inspiration (Widdicombe & Winning 1976). The augmented breath may be initiated by stimulation of R.A.R.(Davies & Roumy 1982). Reflex bronchial and laryngeal constrictions are also seen. R.A.R.are stimulated by inflation and deflation, mechanical irritation (Widdicombe & Davies 1983), chemical irritation (Karczewski & Widdicombe 1969a) and some lung diseases, such as pneumonia, oedema and embolism (Mills, Sellick & Widdicombe 1969; Frankstein & Sergeeva 1966; Frankstein 1970).

REGULATION OF THE PHASES OF BREATHING

Clark and von Euler (1972) proposed that duration of inspiration was regulated by two mechanisms. At tidal volumes below the threshold for the Hering-Breuer reflex, duration of inspiration is governed by a central mechanism, thus holds a constant relationship with tidal volumes. The range of volumes at which this mechanism operates is called range 1. Duration of

inspiration becomes dependant on lung volume at tidal volumes above this threshold, at thse tidal volumes duration of inspiration has a hyperbolic relationship with tidal volume. The range of volumes over which this relationship holds is called range 2 (fig 7).

In humans, having a high threshold for the Hering-Breuer reflex there is a distinction between the two ranges. In laboratory animals, having a lower threshold for the Hering-Breuer reflex the two ranges merge, shown in the cat (Clark & von Euler 1972) and rat (Cragg & Drysdale 1983). This in terms of von Eulers model suggests that where two ranges exist, duration of inspiration is governed by CIA alone in range 1, P.S.R. activity and CIA govern durat-ion of inspiration in range 2. However in animals in which the two ranges merge tI is governed by P.S.R. and CIA activity.

Inspiration is initiated by P.S.R. (Paintal 1973a) and probably also by R.A.R.(Davies, Sant'Ambrogio, Sant'Ambrogio 1981). Termination of inspiration probably involves R.A.R.as well as P.S.R. (Davies, Nadal & Weinmann 1984) Pressure pulses of inflation and deflation influence the duration of inspiration by the activity of P.S.R. (Paintal 1973a). Activity of R.A.R.may be involved in the augmented breath. Augmented breaths are extra large lung inflations and may be involved in preventing local lung collapse during normal

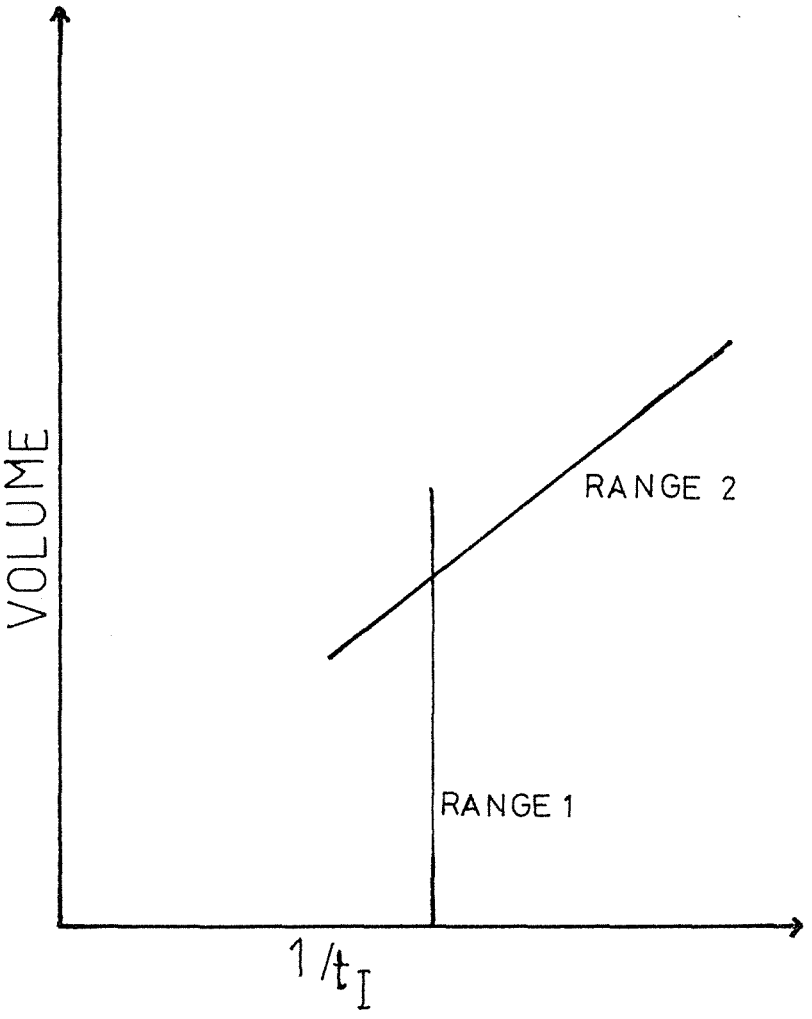


FIG 7. THE RELATIONSHIP BETWEEN VOLUME AND DURATION OF INSPIRATION IS CONSTANT IN RANE1 BUT HYPERBOLIC IN RANGE2.

breathing. These can be triggered by rapid lung inflation during inspiration and occur in an "all or none" way (Davies & Roumy 1982). Augmented breaths, whether spontaneous or triggered by inflation of the lungs, have been ascribed at least in part to the excitation of lung receptors, probably RAR.

Davies and Roumy 1982 suggest that an augmented breath is due to a normal inspiratory efferent discharge, followed immediately by a reflexly induced efferent discharge. An augmented breath can be triggered by lung inflation or deflation when R.A.R.are stimulated above threshold. A spontaneous augmented breath is due to sumation of inspiratoy drive with the reflex effects of R.A.R. early on in inspiration, at a time when receptors would be sensitized by collapse of the lungs.

It has been demonstrated that duration of expiration shortens hyperbolically with increasing tidal volumes over both ranges (Gardener 1977). Clark and von Euler 1972 showed a linear relationship beween tI and tE over a range of tidal volumes.

It has been considered that the P.S.R. are primarily responsible for the inflation reflex from the lungs (Guz, Noble, Eisele & Trenchard 1970; Paintal 1973a). Pulses of deflation shortens

expiration (Davies & Vizek 1977). This may be accounted for by a reduction in P.S.R. activity during deflation (Guz et al 1970; Paintal 1973a). However R.A.R.are stimulated and may contribute to this shortening (Davies & Roumy 1982). Sellick and Widdicombe 1970 point out that, although R.A.R.may initiate shortening of tE, R.A.R.activity is too short to maintain shortening of tE. Recent work has shown that the effects of receptor stimulation may remain for some time after stimulation (Eldridge 1973; Eldridge 1974; Karczeweski, Budinski, Gromysza, Herczynski & Romanuik 1976).

Knox 1973 using 200ms inflation pulses did not observe any shortening in expiration with pulses of inflation. However Davies and Roumy using 100ms inflation pulses, found these pulses when applied early in expiration occasionally shortened expiration. This lead Davies and Roumy 1982 to conclude that duration of expiration is controlled by a balance between P.S.R. and RAR.

From this it appears that duration of inspiration is governed by P.S.R. activity except in humans where in eupnoea and during augmented breaths CIA acts alone. Duration of expiration is controlled by a balance between P.S.R. and R.A.R. activity.

If tI is measured from a volume tracing, rather than inspiratory neuron discharge, it has been shown the period of decline in inspiratory

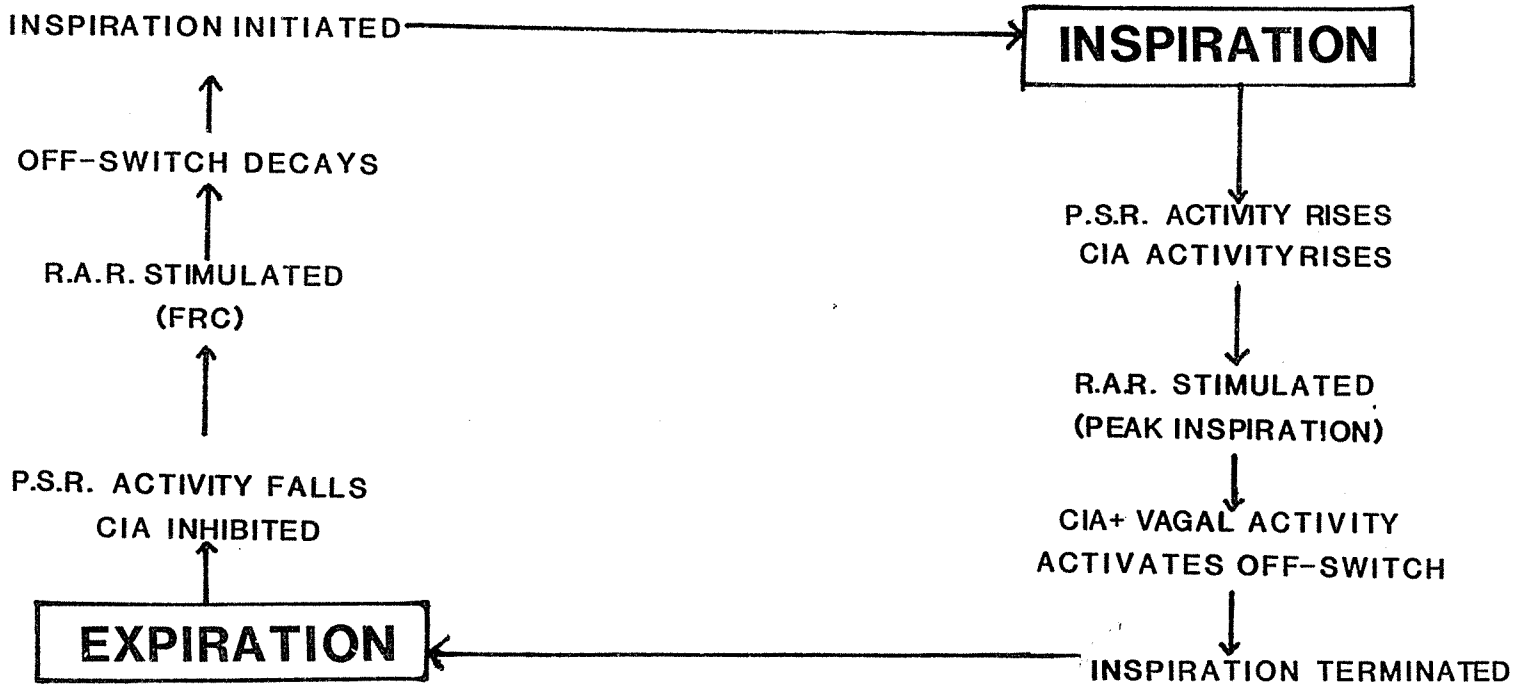


FIG 8. RECEPTOR INFLUENCE ON THE PHASES OF BREATHING. ALTERATION OF RECEPTOR ACTIVITY CHANGES DURATION OF INSPIRATION OR EXPIRATION. STIMULATION OF R.A.R. MAY INITIATE AN AUGMENTED BREATH.

discharge is incorporated into expiratory duration (Youners & Remmers 1981). Thus the method of measuring tE and tI must be taken into account when relating the two phases of breathing. We are interesred in measuring the effect on inspiratory discharge, as a measure of the response of respiratory centres to afferent information.

Fig 8 summarizes the receptor influence on initiation of inspiration, tI, tE and augmented breaths.

LINKING OF RESPIRATORY CYCLES

Clark and von Euler (1972), and Knox (1973) suggest that although duration of expiration may be dependant on the preceeding inspiration, one breath was not considered to influence the following breath. Karczewski et al (1976) reported that electrical stimulation of the vagi in bilaterally vagotomised rabbits produced smaller decrease in tI than tE and that the changes in tE preceeded the changes in tI by one respiratory cycle. Davies and Roumy 1982 using deflation pulses in expiration noted shortening of tI followed the shortening of tE. This shows that tI need not be independant of the previous tE.

The changes to tE produced by these workers were caused by short term stimulation of vagal afferents. The activity of these fibres would not

continue into the next breath. This suggests that there is some central mechanism linking inspiration to the previous expiration. Benchetrit and Bertrand 1975 recording regular respiration showed minor changes in depth and duration influence subsequent cycles. This illustrates this linking may opperate during eupnoea.

Eldridge (1973 & 1974) using induced hyperventilation, has shown that these changes to the ventilation pattern can remain for several breaths after cessation of stimulation. Similar posthyperventilation responses have been noted by other workers (Tawadrous & Eldridge 1974; Cunningham, Howson, Metias & Petersen 1983; Kumar, Nye & Torrance 1983; Jennet & Walker 1984), and in infants (Fleming, Goncalves, Levine & Wollard 1984). I have shown in preliminary experiments that duration of expiration is shortened for several breaths after a pressure pulse was applied to the lung (Jones 1984). This suggests that the linking of one breath to another may be part of longer term influence to pattern of breathing. This long term influence we have called "memory".

Eldridge (1973 & 1974) and Karczweski et al (1976) have proposed a central mechanism for "memory". Eldridge 1974 suggested that after discharge in respiratory afferents, activated by breathing and having a long decay time, may have a positive feedback into respiratory neurons.

Karczewski et al 1976 suggested the mesencephalic reticular formation may be involved.

The purpose of the present study was to investigate this linking of the phases of breathing, to investigate the longer term influences on breathing pattern, to investigate the relative roles of "memory" of P.S.R. and R.A.R.activity, and to investigate further their relative roles in the response inflation and deflation of the lungs in expiration.

The preliminary study demonstrated "memory" in breaths subsequent to pulse application. However the exact time of pulse application, or tE and tI was not recorded precisely enough to examine the effects of pressure pulses on tE containing the pulse or the subsequent tI. The results of this preliminary study were not included in this thesis and improvements made to the methods.

EFFECTS OF P.D.G.

During the course of these experiments it was intended to test the patency of J receptors after differential cold block. As pulmonary oedema is slowly developing and difficult stimulus to use the method used to study these receptors has been to inject P.D.G. into the right atrium. In the rabbit, injection of P.D.G. into the right atrium causes tachypnoea due to shortening mainly of expiration with slight shortening of inspiration.

This is accompanied by a reduction in tidal volume and on increase in functional residual capacity. Apnoea may occur in the end inspiratory position.

If these effects are solely due to J receptor stimulation then vagotomy should abolish this effect, as some workers have demonstated (Dawes & Mott,1950; Davies, Dixon, Callanan, Huszczuk, Widdicombe & Wise,1978). Other workers demonstated a weak or altered response to P.D.G. after vagotomy (Dawes& Fastier,1950; Dawes, Mott & Widdicombe, 1951; Karczweski & Widdicombe,1969b; Guz & Trenchard, 1971). Dawes, Mott, and Widdicombe demonstrated that this response could be abolished by carotid denervation. However Miserocchi, Trippenbach, Mazzerelli, Jaspar, and Huzucha (1978) demonstrated a large effect from P.D.G. could be obtained after vagotomy. Fig 9 is a summary of literature on the effectsof intravenous P.D.G. in the rabbit.

This effect could have been due to a difference in anaesthetic used. Miserocchi et al used a cocktail mixture, Davies et al 1978 used solely pentobarbitone. Dawes and Mott noted the response to P.D.G. was affected by the anaesthetic techniques used. In light of this, the effects of P.D.G. after vagotomy had to be reinvestigated, before P.D.G. could be used to test for the prescence of J receptors.

In a preliminary experiment, we injected

1950	Dawes & Fastier	"The effects in the rabbit were not completely abolished by vagotomy."
1950	Dawes & Mott.	"...the response is abolished by cutting the vagi ..."
1951	Dawes, Mott & Widdicombe.	"...cutting the vagi abolished these respiratory changes. In a minority of rabbits ...still caused some alteration in respiration. ....abolished by carotid denervation."
1969	Karczewski & Widdicombe	"Vagotomy abolised the respiratory changes."
1971	Guz &	"This ventilatory response disappeared when conduction in non-myelinated fibres was abolished by section. Occasionally ...the response to phenyl diguanide, mediated by receptors at the carotid bifurcation was seen after vagal section."
1978	Davies, Dixon et al.	"After vagotomy phenyl diguanide had no effect on pattern of breathing."
1978	Misserocchi, Trippenbach et al.	"P.D.G. after vagotomy still caused a significant shortening of the expiratory time."

Fig 9. Summary of literature on the effects of P.D.G. after vagotomy in the rabbit.

P.D.G. into the general circulation one hour after vagotomy. A large response to P.D.G. was elicited. As barbituates were the sole anaesthetic agent used in this experiment and we elicited a large response to P.D.G., differences in anaesthetic were not responsible for the lack of response after vagotomy Davies et al 1978 injected P.D.G. within 1-5 minutes after vagotomy while we conducted our experiment one hour after vagotomy. This lead us to believe that time of injection after vagotomy may influence the response. Thus we conducted a series of experiments to determine the nature of the response to P.D.G. after vagotomy.

METHODS

We used 24 New Zealand White Rabbits of either sex weighing between 2 and 3 kg, from the stock maintained by the University. The rabbits had free access to food and water. Fig 10 shows the experimental setup.

Anaesthesia

Anaesthesia was induced with 40mg/kg sodium pentobarbitone (Nembutal) injected into the marginal ear vein. The first 2/3 of the dose was injected rapidly for speedy induction and the remainder given slowly while carefully watching the rabbits breathing for indications of impending respiratory arrest. Anaesthesia was maintained with 0.6mg of pentobarbitone as needed. Depth of anaesthesia was assessed by respiratory rate which was maintained at approximately 60 breaths per minute.

Catheterization

The rabbit was placed supine and the hair clipped from the neck and inguinal regions. 2-3ml of 2% xylocaine was infused under the skin of the inguinal region and the femoral vein and artery exposed. 4-5cm of the vein and artery were cleared and saline filled vinyl catheters inserted several centimeters and tied into the blood vessels to enable us to inject drugs and measure blood pressure respectively.

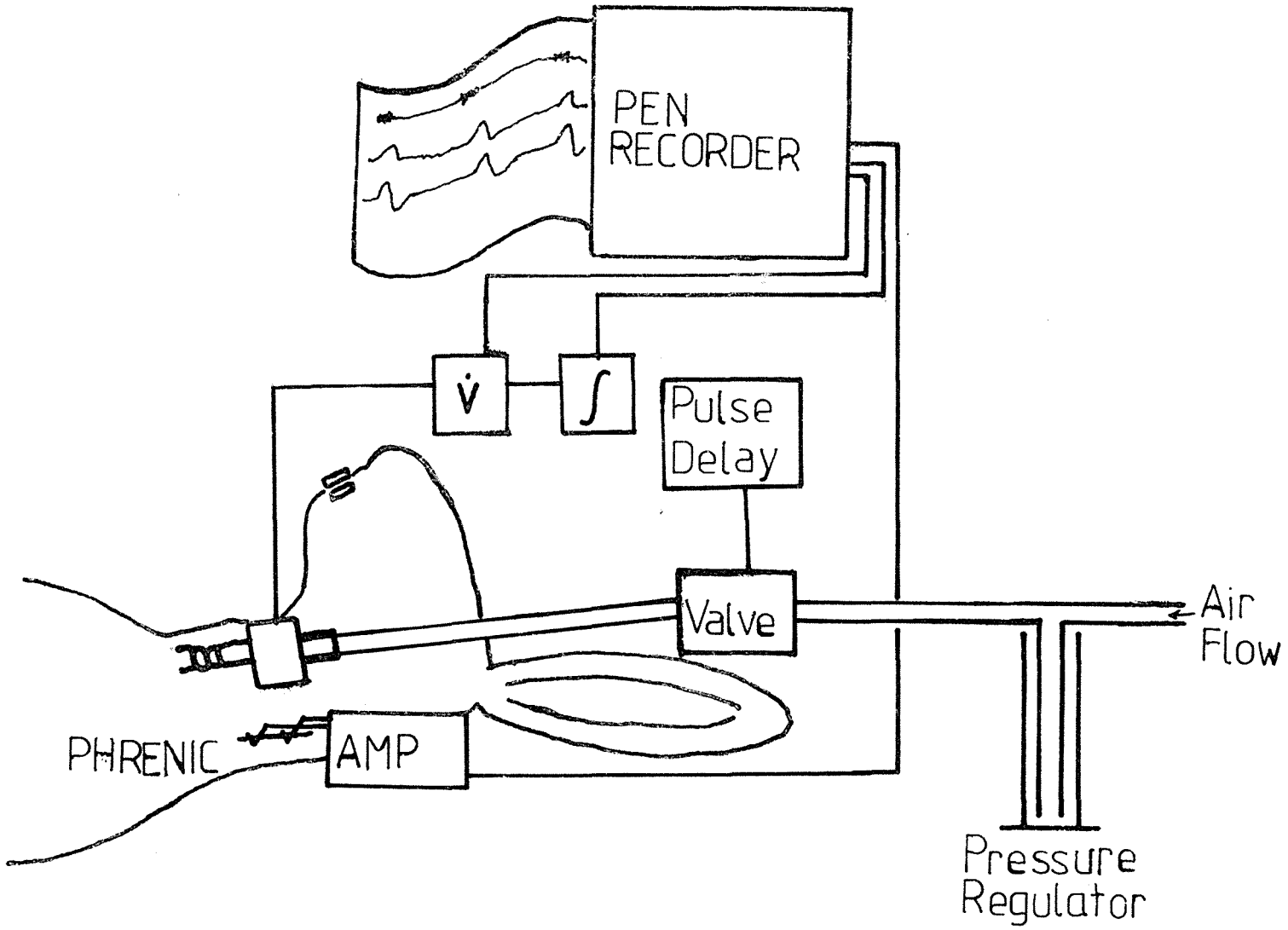


FIG 10. EXPERIMENTAL SET-UP.

Tracheostomy

A skin incision about 8-9cm. long was made along the midline of the throat region. Fascia was cleared by blunt dissection allowing access to the sternohyoidius muscle. This muscle was seperated by dissection along the linea alba (its midline) to expose the trachea.

The trachea was cleared for 3-4cm and a loose tie placed around it. A radial cut was made between the C rings of the trachea. This enabled a plastic tracheal cannula (3.5mm internal diameter) to be tied in place, forming an airtight seal around the cannula.

Phrenic Nerve Isolation

The sternocleidomastoideus muscle was removed after tying off and cutting its major vessels then crushing the muscle tissue at either end before cutting. The phrenic nerve was identified as a small nerve arising from the spinal cord at segments C 3, 4, and 5. It lay close to and parallel to the spinal cord before angling back towards the mid line at the level of the shoulder.

After identifying the nerve further surgery was carried out using a dissecting microscope. The most caudal end of the nerve was cut disturbing the nerve as little as possible. On cutting the nerve the rabbits diaphragm often twitched, confirming identification of the nerve. The nerve was gently freed for 2-3cm by lifting the caudal

end with fine forceps and clearing the tissue underneath with fine optical scissors. The nerve was disturbed as little as possible.

A paraffin filled nerve tray was moved into a position which allowed the nerve to be placed in the tray without stretching the nerve. The nerve was covered with paraffin at all times to prevent drying out.

The tissue around the nerve was carefully removed and the stripped nerve placed on the recording electrodes. This proceedure provided a low noise to signal ratio on the recording the nerves activity. Further clearing of accumulated blood was performed at various stages during the experiment to maintain a low noise to signal ratio.

PRESSURE PULSE SERIES

Recording equipment

The phrenic nerve was placed on bipolar silver electrodes which were lowered into the nerve tray filled with paraffin oil. To facilitate fine adjustments of electrode position the electrodes were held by micromanipulators (Prior). These electrodes were connected to a Neurolog recording system. This consisted of an A.C. preamplifier, filters, A.C. amplifier and an audio amplifier connected in sequence.

The A.C. preamplifier was used to provide a high input impedence (MΩ) to the phrenic signal. The preamplifier had a differential input which was balanced before each experiment giving the optimum common mode rejection ratio (CMRR). The preamplifier output was passed through filters which enabled choice of frequency band width, giving optimum signal to noise ratio. The A.C. amplifier then boosted the power of the signal to usable values. The signal fed to the pen recorder (Gould) to provide a permanent trace of phrenic activity. The audio amplifier converted the electrical signal from the phrenic to an audible signal via speakers allowing constant monitoring of respiration.

The tracheal cannula was attatched to a pneumotachograph (Fleish). This consisted of a cylinder interupted by a low air flow resistant

gauze. Air flow caused a pressure difference across the gauze which was measured by a sensitive pressure transducer mV/cm H2O (Honeywell). The changes in pressure difference over each breath was proportional to flow. As volume is the integral of flow, volume was calculated by the amplified output of the pressure transducer being fed to a respiratory integrator. This was connected to the pen recorder to provide a trace of tidal volume. The traces were calibrated at the end of the experiment.

The pressure inside the tracheal cannula could be measured by inserting a wide bore needle into the cannula lumen. This needle was attatched to a pressure transducer (Honeywell) and connected to the recorder. This gave a recording of the pressure changes in the tracheal cannula.

Pressure regulation

The free end of the pneumotachograph was atta-ched to a solenoid valve by a short length of rubber tubing. The valve was normally open to the atmosphere, allowing the rabbit to breathe normally. However when triggered the valve opened to positive or negative pressure reservoirs as required.

The valve was triggered electronically to allow a preset length pressure pulse to be applied after a preset delay from time of triggering

(Fig11A). This "delay after trigger" system consisted of a period generator, counter, interface card, digital width and pulse buffer connected in sequence. A switch to the counter allowed manual triggering. The pulse buffer was connected to the solenoid valve allowing the low voltage electronic pulse to to be converted to a power pulse sufficient to open the valve to the pressure system.

Positive pressure was generated from a blower. This was passed through a pressure reservoir and pressure regulator. This consisted of a tube interupted by a T-piece, one limb of which passed to the bottom of a 20cm column of water. The water was open to the atmosphere allowing excess pressure to be bubbled off. The other end of the T-piece controlled the airflow by a screw clamp to give a steady bubbling. This regulated the pressure in the reservoir to 20KPa (Fig11B).

Negative pressure was generated from a tap aspirator. This passed through a reservoir and a pressure regulator. The pressure regulator consisted of a tube into a cylinder closed to the atmosphere. A tube from the atmosphere passed through a 20cm column of water. Air was drawn through this tube to regulate the negative pressure. Flow from the aspirator was regulated to create a steady bubbling. This regulated the pressure in the reservoir to negative 20KPa

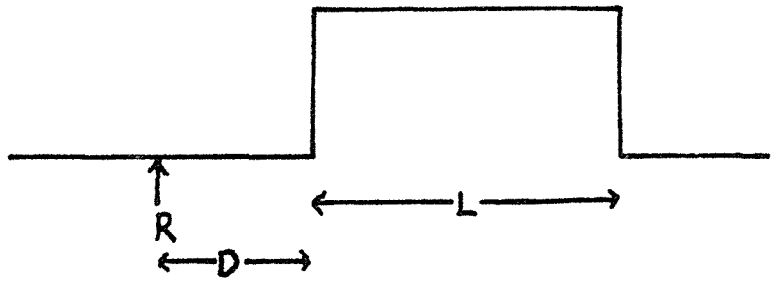


FIG 11A. DELAY ON TRIGGER SYSTEM. AFTER RELEASE OF THE SWITCH (R) AND A PRESETDELAY (D) A SQUARE PULSE OF PRESET LENGTH (L) OPENS THE VALVE.

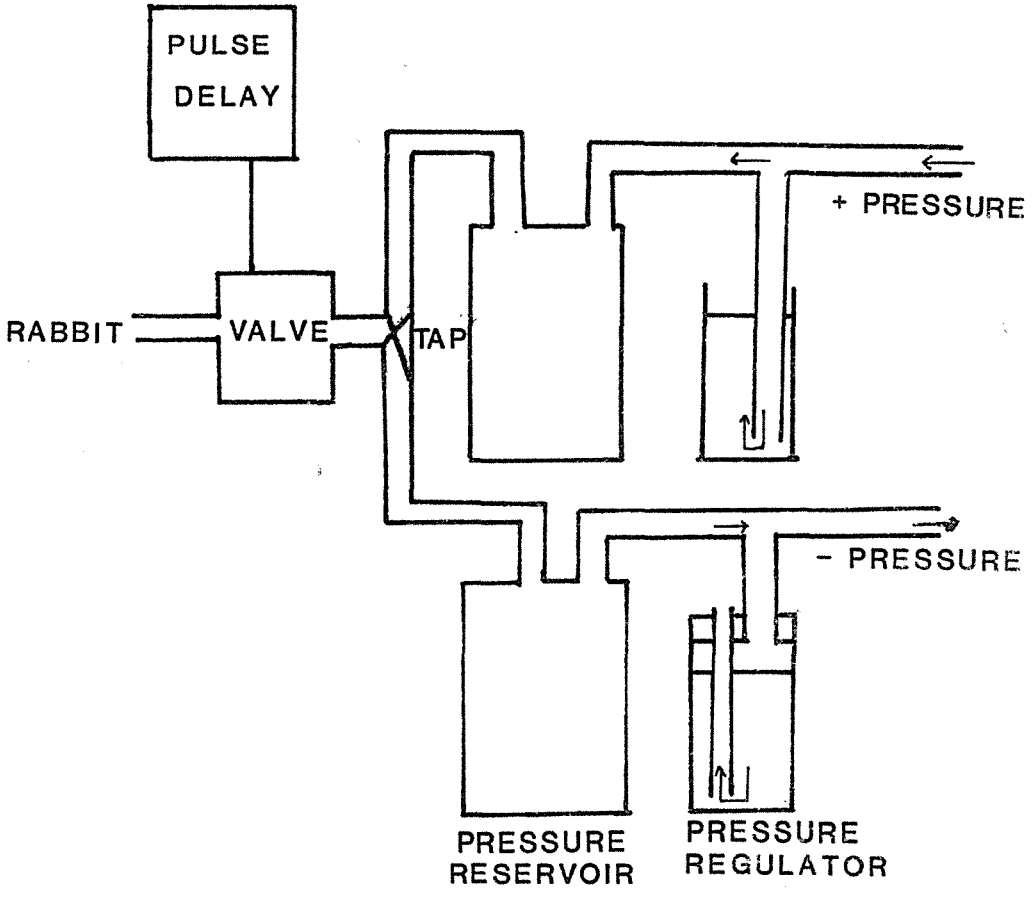


FIG11B. PRESSURE PULSE REGULATION.

relative to atmosphere (Fig11B).

Both pressure reservoirs were connected together leading to the solenoid valve. A tap on each connection from the reservoir allowed choice of positive or negative pressure to be applied to the rabbits lungs when the solenoid valve was opened (Fig11B).

PROTOCOL

Validation

Two rabbits were used to demonstrate that pressures of ±20KPa produce adequate lung volume changes. These rabbits were prepared as described earlier with provision for measuring the pressure changes in the tracheal cannula. A trace was taken of this and of tidal volume.

Positive and negative 20KPa pulses were applied. 37 pairs of values were obtained from the two rabbits. The mean change in volume was determined.

Vagi Intact

In each experiment one of twelve rabbits was weighed, sexed, anaethetized, and prepared as described earlier. Phrenic activity and volume traces were recorded for two breaths before each pulse was applied and three breaths after.

The latency between the end of inspiration and subsequent pulse application was set at four

different positions. These were delays of 0%, 20%, 35%, 55% of control duration of expiration calculated each run on average control values. Actual latencies differed from the calculated delays as the pulse was triggered manually when the end of inspiration was determined by eye.

Each run consisted of a positive and negative pulse at each calculated pulse position (table 1). The order of application of pulse position in the run was randomized. Two tables of pulse position were drawn from random number tables, using all possible order combinations and no two runs being the same. One table was used for odd numbered rabbits and the other table for even numbered rabbits.

Four runs were made each of rabbits one and two. Six runs were made of each of the other rabbits. In the odd numbered runs, positive pulses were applied before the negative. In the even numbered runs negative pulses preceeded positive.

At least two minutes elapsed between each pulse application. The pneumotachograph was disconnected from the solenoid valve between each treatment, to reduce dead space. It was reconnected one or two breaths before the control breaths were taken. When anaesthetic was administered two to three minutes was allowed to elapse, until respiration had restabilised, before recommencing with the experiment. Between each run

		Series 1 (odd rabbits)		Series 2 (even rabbits)	
Treatment	run	order of sign	order of positions	order of sign	order of positions
Intact	1	+ −	1,3,4,2	+ −	2,3,1,4
	2	− +	4,1,3,2	− +	3,1,4,2
	3	+ −	3,4,1,2	+ −	1,4,3,2
	4	− +	2,1,3,4	− +	4,1,2,3
	5	+ −	1,4,2,3	+ −	4,2,1,3
	6	− +	3,2,1,4	− +	2,3,4,1
H-B ratio SO2 admin					
	7	+ −	2,4,1,3	+ −	3,4,2,1
	8	− +	4,3,1,2	− +	2,1,4,3
	9	+ −	1,3,2,4	+ −	1,2,4,3
SO2 admin H-B ratio					
	10	− +	2,4,3,1	− +	3,2,4,1
	11	+ −	3,1,2,4	+ −	4,3,2,1
	12	− +	4,2,3,1	− +	1,2,3,4

Table 1. Protocol of pressure pulse series. Each run consists of a positive and negative pulse in each position.

the duration of expiration of the controls was checked and adjustments of actual latency of pulse made.

At the end of the runs with stretch receptors intact the Hering-Breuer ratio was measured, this was the ratio between control tE and tE while a constant 10KPa pressure is applied to the air in the lungs. The ratio is an index of stretch receptor activity. Stretch receptors were then blocked using sulphur dioxide.

Stretch Receptor Block

Sixty mls SO2 was drawn from a cylinder and injected into a stream of air filling a 10l douglas bag. After filling, the bag was shaken to ensure even distribution of sulphur dioxide. The concentration was tested using a commercial calirometric crystal test for sulphur dioxide (Dräger).The mixture was made fresh prior to each experiment.

The sulphur dioxide was administered to the rabbit via the tracheal cannula with gentle suction from a tap aspirator removing expired gas. This allowed the rabbit to breath the sulphur dioxide freely. As sulphur dioxide is corrosive the mixture was not passed through the pneumotachograph.

Sulphur dioxide was administered at 200 to 300 ppm for 20 to 30 minutes (Callanan et al 1975). The Hering-Breuer ratio was then measured. If this ratio was greater than 1.5 SO2 inhalation was

continued until adequate stretch receptor block was obtained.

When adequate stretch receptor block was obtained the protocol for vagi intact was followed for three runs. A further 10 minutes of SO2 was administered. The Hering-Breuer ratio was measured and further SO2 administered if a poor block was obtained. Three more runs were completed and degree of stretch receptor block assessed by the Hering-Breuer ratio at the end of the experiment. In one rabbit the block of stretch receptors wore off more quickly. In this rabbit SO2 was administered more frequently.

Vagotomy

Vagotomy was performed on three rabbits. The carotid sheath was identified and the vagus cleared from the surrounding sheath. Local anaesthetic (2% Xylocaine) was applied to the nerve on small cotton balls before cutting. This prevented stimulation of the nerve during the cut. A complete section of the nerve was made. This was performed to both vagi.

Pulses of positive and negative pressure were applied at various times in expiration. Eight treatments were performed in each rabbit. In one rabbit it became evident that complete vagotomy had not been performed. After vagotomy was completed a fresh set of results were obtained.

Cold Block

Differential block of myelinated fibres of the vagus can be achieved by localized cooling of the vagus (Paintal 1973). The tempreture at which this block occurs varies considerably between individuals. However in each individual there is a temperature at which there is a good block of myelinated fibres without block of unmyelinated (Douglas & Malcolm 1955). Thus the temperature must be lowered gradually until effective block of myelinated fibres occured. To differentially block activity in the vagus nerves each nerve was seperated from the carotid sheath and laid onto a copper thermode. This consisted of a hollow copper disc through which cooled water was circulated. A groove lay across the surface of the dic to contain the nerve. Over these discs another copper disc was laid to protect the nerve from heat and drying out (fig 12).

Differential cold block of the vagus was applied to three rabbits. The Hering-Breur ratio was determined with the vagus at room temperature. The response to deflation (of 20KPa) held over a few breaths was noted at room temperature. This tests the presence of rapidly adapting receptor activity.

Water cooled to 7 degrees Celcius was continuously passed through the thermodes from a holding reservoir to a collecting bucket. The

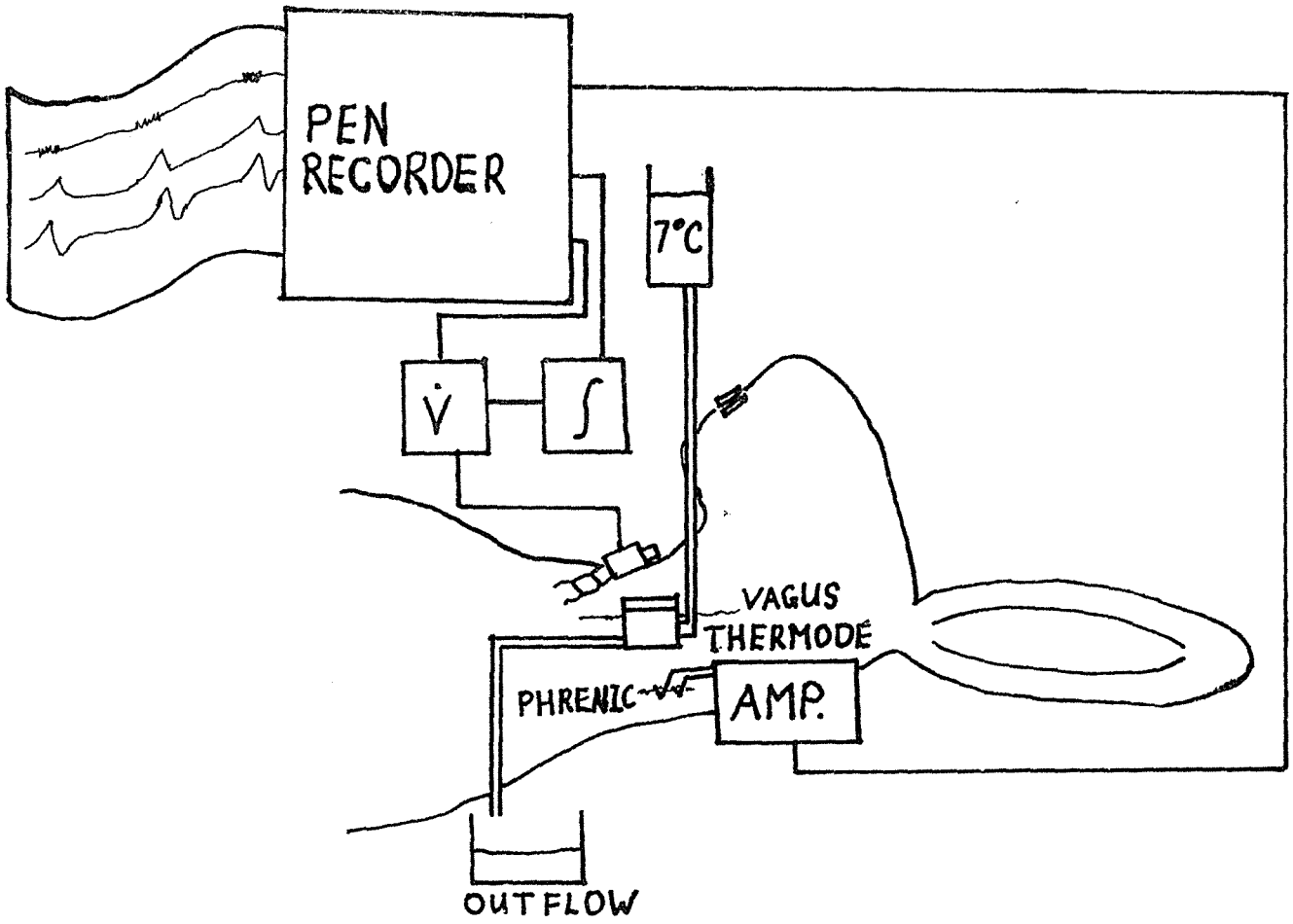


FIG 12. DIFFERENTIAL COLD BLOCK.

temperature of the water was measured at both the holding and collecting reservoirs. The degree of stretch and rapidly adapting receptor block was assessed by the Hering-Breuer ratio and the response to lung deflation after respiration stabilized. If block was not adequate the water was cooled further. Several positive and negative pressure pulses were applied in different positions of the respiratory cycle.

The vagus was then warmed with water at room temperature. When respiration had stabilized the Hering-Breuer ratio and response to deflation measured. A few more pressure pulses of either sign were then applied.

TREATMENT OF DATA

The duration of inspiration (tI) and expiration (tE) were measured directly from the trace. At the paper speed used of 25mm/sec the paper had divisions representing .02sec which could be read accurately to ± .01sec. Determination of begining and end of inspiration/expiration was repeatable ± .01sec. Thus the trace could be measured accurately to ± 0.02sec. The position of pulse application was usually identified by electrical spike caused by the release of the solenoid valve, marking the trace. Where this was not clear the sudden change in volume caused by the pressure changes in the lung assisted

identification. When volume traces determined the position of pulse application the accuracy repeatability was reduced to an accuracy of ± .02sec. Thus the overall accuracy of determining pulse position was reduced to ± 0.03sec.

There were differences between the calculated latency and actual delay from the end of inspiration to pulse application. This was partly due to delay between recognising the end of inspiration and triggering the valve. These differences were also due to small changes in control expiration within runs. The actual delay was calculated as percentage of control expiration. The data was grouped into four pulse positions. These groups were 0-20%, 20-35%, 35-55%, and greater than 55% of control expiration.

For each rabbit the data was grouped into +Intact, -Intact (± pressure with stretch receptors intact), +Blocked, -Blocked (± pressure with stretch receptors blocked by SO2), within each of these groups were the groups of the four pulse positions. Data which included an augmented breath was excluded from these groups and treated seperately. Vagotomy and cold block treatments were also treated seperately.

Two control values for duration of expiration and inspiration were taken. The mean of these values was taken as the control duration (ie tEc/tIc). The differences between the duration of

the phases of breathing of the experimental runs and the controls were measured.

The ratio of duration of expiration (tE) after the pulse divided by tEc was the normalized value. Similarly normalized values of inspiration can be obtained. A normalized value over 1 represents an increase in duration while a value below 1 represents a decrease. The duration of expiration containing the pulse (tE') was only altered from the time the pulse was applied. The change in duration of expiration containing the pulse could only occur after the pulse was applied. Thus the normalized value of an expiration was calculated as time of pulse application (tP) subtracted from duration of expiration containing the pulse (tE') divided by the control duration of expiration (tEc) minuse the time of pulse appliçation.

Thus

$$tE' \text{ normalized} = \frac{tE'-tP}{tEc-tP}$$

other tE values

$$tE \text{ normalized} = \frac{tE}{tEc}$$

and

$$tI \text{ normalized} = \frac{tI}{tIc}$$

The Hering-Breuer ratio is the duration of expiration before inflating the lungs divided by duration of expiration afterwards. Thus the amount of lengthening of expiration by inflating the lungs is the Hering-Breuer ratio minus one. Stretch

receptor block was assessed by dividing the Hering-Breuer ratio minus 1 by the Hering-Breuer ratio minus 1 before block. This gave the fraction the Hering-Breuer ratio was shortened by SO2 administration. Values close to one were poorly blocked with a value of 1 being totally unblocked. Values close to 0 being well blocked with a value of 0 for total block. The degree of block was assessed for each rabbit, for the three Hering-Breuer ratio's taken after block. The rabbits were ranked on the degree of block and the consistancy of block.

Means, standard deviation, standard error of the mean, were calculated for controls, tI, tE, normalised values, and differences from controls under the different conditions. For individual rabbits, rabbits pooled and the eight rabbits with the best block pooled. A programme (Appendix A p#) was written in BASIC to perform these calculations.

Significance of differences from controls was tested by paired t test. Significance of variation of effect between positions and between treatments was tested by two way analysis of variance. See appendix A p122 for a more complete discussion of statistical analysis.

P.D.G. SERIES

Ten rabbits were anaesthetised, and prepared as described previously. The catheter in the femoral vein was pushed up towards the heart so that the tip lay close to the right atrium. In two rabbits phrenic activity was not recorded due to electronic failure. In these animals diaphragmatic e.m.g. was recorded.

P.D.G. (ICI) was dissolved in saline to concentrations of 100µg/ml. P.D.G. was injected into the venous catheter and washed in with saline. Ten minutes was allowed between each injection to prevent tachyphylaxis occuring (Karczewski & Widdicombe 1969b). The peak frequency of breathing was assessed over 2-3 breaths after apnoea, and always measured within 12 seconds of injection.

Epicardial Receptors Intact

In three rabbits doses of P.D.G. ranged from 25µg/kg to 300µg/kg. P.D.G. was administered two or three times before vagotomy (table 2). Both vagi were seperated from the carotid sheath, local anaesthetic applied, then cut. P.D.G. was then administered immediately, 15, 25, 60, and 70 minutes after vagotomy.

Both glossopharyngeal nerves were identified local anaesthetic applied and the nerves cut. P.D.G. was administered 15 and 25 minutes after the nerves were cut. In one rabbit a catheter was

EPICARDIAL RECEPTORS INTACT (3 rabbits)		EPICARDIAL RECEPTORS BLOCKED (6 rabbits)	
Treatment	P.D.G. injection	Treatment	P.D.G. injection
Control	begining of experiment	Control	begining of experiment
	10 min into experiment		10 min into experiment
	20 min into experiment		20 min into experiment
Vagotomy	immediately after vag.	Xylocaine	15 min after xylocaine
	15 min after vagotomy		25 min after xylocaine
	25 min after vagotomy		
	60 min after vagotomy	Vagotomy	15 min after vagotomy
	70 min after vagotomy		25 min after vagotomy
Glossopharyn- geal section	15 min after section	Glossopharyn- geal section	15 min after section
	25 min after section		25 min after section

Table 2. Protocol of P.D.G. series.

placed in the left carotid vein so its tip lay close to the right atrium. P.D.G. was administered via the femoral and carotid catheters alternatively for each treatment.

Epicardial Receptors Blocked

Six rabbits were given 50µg/kg doses of P.D.G. In one rabbit the dose was raised due to lack of response to the lower dose. In all rabbits the abdomen was opened just below the xiphisternal process to expose the diaphragm. P.D.G. was administered two or three times.

The Hering-Breuer ratio and response to deflation was measured. This assessed the degree of stretch receptor and rapidly adapting receptor activity.

Rather than subject the rabbit to the trauma of thoracotomy to install the catheter for injecting xylocaine, used by Anand and Paintal to block epicardial receptors we used the method shown in Fig 13. Gently moving the abdominal contents with the blade of a laryngoscope the heart beating inside the pericardium could be seen. The needle was gently inserted into the pericardium to allow injection of xylocaine.

In three rabbits the syringe and needle for injecting xylocaine was fitted with a three way tap to enable the needle to be connected to a pressure transducer which was connected to an oscilliscope. The needle was gently inserted into the pericardial

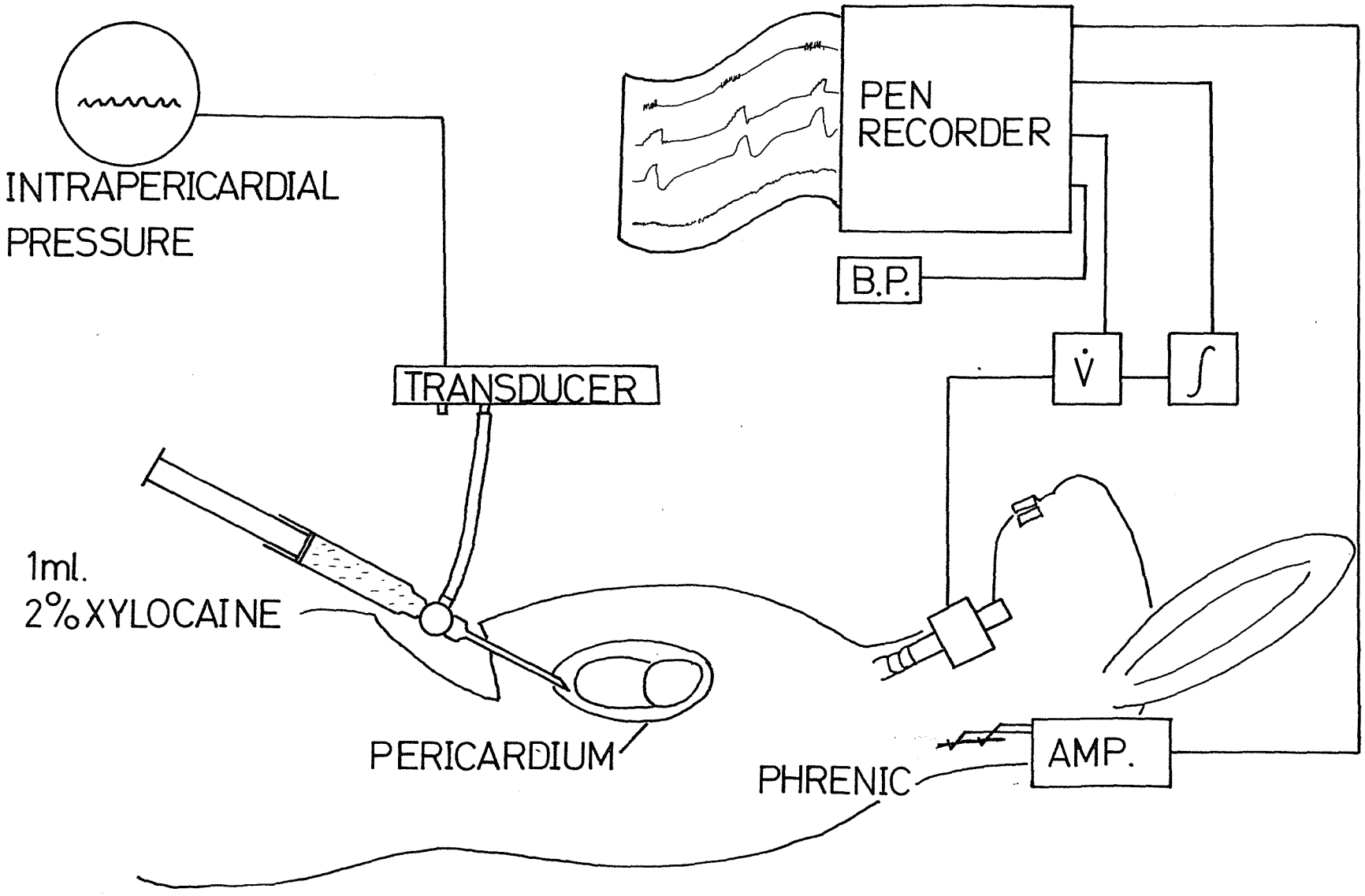


FIG 14. XYLOCAINE ADMINISTRATION.

sac until a cardiac rhythm was detected. Thus injection site could be confirmed by these pressure changes. One ml of 2% xylocaine was injected into the pericardial sac to block epicardial receptors. The Hering-Breuer ratio and response to deflation was measured. P.D.G. was administered 5 and 15 minutes after xylocaine injection.

Both vagi were seperated from the carotid sheath, local anaesthetic applied and cut. More xylocaine was reinjected into the pericardial sac. P.D.G. was administered 15 and 25 minutes after the vagi were cut.

Both glossopharyngeal nerves were identified, local anaesthetic applied and cut. More xylocaine was injected into the pericardial sac. P.D.G. was administered 15 and 25 minutes after the vagi were cut.

Dye included in the local anaesthetic was injected into the pericardial sac to check the site of the injections. One rabbit had the Hering-Breuer ratio and the response to deflation measured. Xylocaine was then deliberately injected outside the pericardial sac into the intrapleural space. The Hering-Breuer ratio and response to deflation was measured.

Treatment of data

Peak frequency was measured directly from the trace over 2 or 3 breaths. An estimate of the

onset of the effect was taken as the begining of the first expiration shortened as determined by eye. Occurence of apnoea was noted. See appendix A p134 for statistical analysis.

RESULTS

PRESSURE PULSE SERIES

The normal value is the fraction tE or tI is of the control and is thus an indication of extent of change in duration. Normal values above one indicate an increase from controls, values below one indicate a decrease. Normalization of the data transforms the data into a form which is easier to interpret than absolute values. However it is invalid to use standard errors of the mean of fractions. Thus although the tables give normal values, all statistics were performed on absolute values.

EXPIRATION CONTAINING THE PULSE

Positive pressure pulses with P.S.R. intact

See table 3 for effect of positive pressure pulses applied in expiration on tE containing the pulse with P.S.R. intact. Table 4 shows number of times tE containing the pulse is shortened, lengthened or no effect after a positive pressure pulse.

Positive pressure pulses in expiration either increased or decreased the duration of the expiration (tE) containing the pulse. Pulses applied early in expiration (Pos 1) often shortened tE containing the pulse (34 out of 72 runs). This shortening was significant at 5% level (t=2.38, 71degrees freedom).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.4975	7	.5578	2	.6876	5	1.108	2
2	.9474	4	.8168	2	.6177	4	1.736	8
3	.8874	6	.8113	5	.9451	6	5.596	6
4	.9173	7	1.082	6	1.501	3	2.775	7
5	1.329	6	1.506	12	1.715	5	2.841	1
6	.7530	4	.9816	10	1.227	7	1.371	3
7	1.018	4	1.189	4	1.451	3	1.167	8
8	.9426	7	1.029	8	1.199	9	.5498	0
9	1.014	7	1.120	8	1.955	3	1.807	3
10	1.090	4	1.257	7	1.530	7	3.164	3
11	1.172	4	1.552	0	1.987	5	3.124	9
12	1.028	6	1.354	1	1.779	3	2.943	4
Mean	.9652	66	1.110	65	1.431	60	2.656	54

Table 3. Normalized values of tE containing the pulse after a positive pressure pulse was applied to P.S.R. intact animals.

Pulses applied later in expiration (Pos 3 and 4) often lengthened tE containing pulse (51 out of 62 runs and 51 out of 63 runs respectively). This lengthening was significant at 1% level (t=3.65, 61df and t=6.25, 62df). Pulses applied in Pos2 did not significantly affect tE containing the pulse (48 out of 73 runs increased, t=.621, 72df).

RB	POS1			POS2			POS3			POS4		
	+	−	=	+	−	=	+	−	=	+	−	=
1	0	6	0	0	5	0	1	2	0	2	1	0
2	0	1	0	0	5	0	1	4	0	4	0	0
3	1	3	1	0	4	0	1	3	0	7	1	0
4	1	2	0	5	3	0	5	0	0	6	0	0
5	6	1	0	5	0	0	5	1	0	5	0	0
6	0	6	0	2	6	0	6	0	0	3	1	0
7	6	5	0	7	0	0	4	0	0	1	0	1
8	1	3	1	4	2	0	5	1	0	1	6	0
9	4	3	0	9	0	0	3	0	0	3	0	0
10	4	3	0	6	0	0	5	0	0	5	0	0
11	1	0	0	6	0	0	7	0	0	8	0	0
12	2	1	0	4	0	0	8	0	0	6	2	0
Mean	26	34	2	48	25	0	51	11	0	51	11	1

Table 4. Numbers of times duration of expiration increased(+),decreased(−),or remained unchanged(=) with positive pressure pulses in intact rabbits.

Negative pressure pulses with P.S.R. intact

See table 5 for effect of negative pressure pulses applied in expiration on tE containing the pulse with P.S.R. intact.

Negative pressure pulses applied in expiration always shortened tE containing the pulse. This shortening was significant at the 1% level regardless of the position the pulse was applied in (t=17.4, 57df; t=19.1, 65df; t=15.8, 61df; t=8.53, 49df; positions 1 to 4 respectively).

The degree of shortening was significantly greater (F=4.79 3&190 df) at the 1% level for pulses applied earlier in expiration (-.746 and -.628 ,pos1 and 2 respectively) than those applied later (-.352 and -.2 ,pos3 and 4 respectively).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.1995	4	.1851	7	.1579	5	.5285	0
2	---	2	.1823	0	.3922	6	.5140	8
3	.2061	5	.2530	10	.4067	4	.8785	4
4	.1006	6	.1816	5	.2270	5	.4059	7
5	.1617	8	.3795	10	.4370	4	.3572	1
6	.0705	11	.1102	6	.1329	4	.1445	3
7	.1004	6	.1704	5	.2174	6	.4076	7
8	.0711	13	.0495	9	.0823	2	.1254	0
9	.1105	9	.0565	5	.0639	5	.1011	5
10	.0745	5	.2505	11	.1853	4	.2296	4
11	.1342	2	.1327	4	.1594	8	.5244	8
12	.5106	1	.0923	9	.1676	7	.3679	5
Mean	.1254	72	.1620	81	.2272	60	.4228	52

Table 5. Normalized values of tE containing the pulse after a negative pressure pulse was applied to P.S.R. intact animals.

Degree of stretch receptor block

See table 6 for assessment of degree of stretch receptor block after SO2 administration and end of experiment.

P.S.R. block after the first SO2 administration was quite complete (Hering-Breuer ratio less than 10% of control) in rabbits 1 and 5 to 12. Only rabbits 2, 4 and 6 did not block fully after the second administration of SO2, although no measurement was taken for rabbits 7 and 8. However at the end of the experiment only rabbits 1, 3 to 5, and 9 had Hering Breuer ratios less than 10% of control, and rabbits 2 and 7 had Hering-Breuer ratios between 10 and 15%.

From this it can be seen only three rabbits 1, 9, and 5 had Hering-Breuer ratios of less than 10% throughout the period of block. The four rabbits with the most effective block in order of rank are rabbits 5, 9, 1, and 7. The four rabbits with the least effective block in order of rank are rabbits 4, 11, 12, and 8.

RB	SO2 ADMINISTRATION 1ST	2ND	END EXPT.	RANK
1	.0866	.0626	.0759	3
2	.100	.143*	.129	7
3	.156	.0694	.0184	6
4	.652	.609	.0435	12
5	.0102	.0139	.0139	1
6	.0185	.130	.241	8
7	.0134	---	.134	4
8	.0909	---	.242	9
9	.0265	.0132	.0132	2
10	.0050	.0050	.243	5
11	.0393	.0169	.388	11
12	.0438	.0219	.372	10

Table 6. Assessment of degree of stretch receptor block after SO2 and end of experiment, Hering-Breuer ratio after block as fraction of control. One indicates no block while zero indicates total block. Rank is from 1 with best block to 12 with poorest block.

Positive pressure pulses with P.S.R. blocked

See table 7 for effect of positive pressure pulses applied in expiration on tE containing the pulse with P.S.R. blocked.

Positive pressure pulses applied in expiration after P.S.R. were blocked often shortened tE containing the pulse. This shortening was significant at the 1% level pos 1 to 3 (t=7.42, 64df; t=4.45, 65df; t=4.06, 59df), a slight non significant lengthening of tE occured in pos4 (t=1.24, 53df).

The degree of shortening of tE containing the pulse was significantly greater (F=2.60 3&200df) at the 1% level for pulses applied earlier in expiration (-.234, pos1) than those applied later (-.172 & -.156 pos 2 & 3) with slight lengthening in pos 4 (.143). After effective P.S.R. block (rabbits 5, 9, 1, and 7) the degree of shortening of tE containing the pulse was not significantly different between pulse positions (F=.123 3&64df; -.128, -.172, -.141 & -.019, pos 1 to 4 respectively).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.8737	2	.7428	7	.6362	5	.7409	2
2	.7222	2	.6590	4	.6590	4	2.674	8
3	.2633	5	.3265	6	.4971	6	.7924	6
4	.6697	6	.7599	7	.7193	3	1.154	7
5	.7779	12	.8702	6	.8338	5	.6667	1
6	.7209	10	.6938	4	.8028	7	.8372	3
7	.6134	4	.7014	4	.7014	3	2.828	8
8	.4486	8	.6068	7	.4673	9	---	0
9	.9157	8	.7747	7	.9704	3	1.112	3
10	.9016	7	.8437	4	.7830	7	1.064	3
11	---	0	1.003	4	.8815	5	.7216	9
12	.8871	1	1.057	6	1.180	3	1.384	4
Mean	.7017	65	.7468	66	.7795	60	1.483	54

Table 7. Normalized values of tE containing the pulse after a positive pressure pulse was applied to P.S.R. blocked animals.

Negative pressure pulses with P.S.R. blocked

See table 8 for effect of negative pressure pulses applied in expiration on tE containing the pulse with P.S.R. blocked.

Negative pressure pulses applied in expiration after P.S.R. were blocked always shortened tE containing the pulse. This shortening was significant at the 1% level regardless of the position the pulse was applied in (t=11.7, 71df; t=13.6, 80df; t=11.0, 59df; t=6.39, 51df; positions 1 to 4 respectively).

The degree of shortening of tE containing the pulse was significantly greater (F=2.24 3&221df) at the 1% level for pulses applied earlier in expiration (-.359, -.374 ,& -.291 pos1 to 3 respectively) than those applied later (-.116 pos4). After effective P.S.R. block (rabbits 5, 9, 1, and 7) the degree of shortening of tE containing the pulse was not significantly different between pulse applications (F=.090 3&70df; -.120, -.150, -.143, and -.076 pos 1 to 4 respectively). The degree of shortening of the tE containing the pulse between positive and negative pulses was significantly different at the 1% level in pos 2 to 4 (F=2.56 1&127df, F=1.85 1&99df, & F=2.32 1&88df) but not pos 1. After effective P.S.R. block (rabbits 5, & 9) the difference was significant at the 1 & 5% level in positions 1 & 4 (F=9.18 1&34df, F=3.19 1&7df) but not at positions 2 and 3 (F=.26 1&25df, F=.84 1&14df).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.8161	4	.9118	7	.9164	5	---	0
2	.2304	2	---	0	.3879	6	1.775	8
3	.1612	5	.2487	10	.3966	4	.5671	4
4	.3446	6	.3213	5	.3905	5	.4845	7
5	.7782	8	.5958	10	.4924	4	.3448	1
6	.7515	11	.3517	6	.2143	4	.2531	3
7	.8260	6	.9013	5	.8900	6	1.371	7
8	.1847	13	.1811	9	.2067	2	---	0
9	.8829	9	.7194	5	.3857	5	.4362	5
10	.3364	5	.1917	11	.2177	4	.3420	4
11	.3685	2	.2059	4	.2459	8	.6962	8
12	.5806	1	.2053	9	.2026	7	.5074	5
Mean	.5471	72	.4051	81	.4202	60	.8119	52

Table 8. Normalized values of tE containing the pulse after a negative pressure pulse was applied to P.S.R. blocked animals.

EXPIRATION SUBSEQUENT TO PULSE

See tables 9 to 12 for the effect of pressure pulses applied in expiration on the second and third tE following the pulse with P.S.R. intact and blocked.

Pressure pulses shorten the second and third breath after pulse application regardless of sign of pulse, block of PSR, or position pulse was applied in. This shortening was significant to the 1% level in all cases.

t values		2nd breath	3rd breath	df
+Intact	pos1	12.8	5.40	61
	pos2	11.3	8.34	72
	pos3	7.92	6.88	61
	pos4	13.3	7.67	62
-Intact	pos1	8.16	6.33	57
	pos2	7.16	5.53	65
	pos3	5.52	8.42	61
	pos4	4.92	5.25	59
+Blocked	pos1	5.48	3.69	64
	pos2	6.13	3.82	65
	pos3	9.76	6.61	59
	pos4	5.20	2.91	53
-Blocked	pos1	9.59	8.65	71
	pos2	12.8	10.4	80
	pos3	10.0	9.04	59
	pos4	10.9	8.66	51

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	.8235	.7859	.8356	.8381	.9721	.8842	.9454	1.007	7	2	5	2
2	.8615	.9661	.8512	.7120	.9846	.9022	.9473	.9042	4	2	4	8
3	.8219	.8450	.7838	.6268	.9751	.9755	.8857	.9235	6	5	6	6
4	.8561	.7559	.8479	.8126	1.004	.9508	.9115	.8760	7	6	3	7
5	.8345	.7867	.8110	.8117	.8961	.9037	.9095	.9705	6	12	5	1
6	.8939	.8692	.8871	.7623	.9951	.9729	.9016	.8616	4	10	7	3
7	.8424	.8426	.8988	.8022	.9147	.9351	.9103	.9404	4	4	3	8
8	.8875	.8931	.8862	.8318	.9465	.8956	.9452	.9281	7	8	9	0
9	.8264	.8913	.8156	.8055	.8516	.8582	.8882	.8207	7	8	3	3
10	.7872	.7758	.7786	.8224	.8324	.7988	.8239	.8405	4	7	7	3
11	.6970	.7973	.8019	.8061	.8788	.8654	.8297	.9208	4	0	5	9
12	.8385	.8765	.8725	.8147	.8865	.9389	.8805	.8976	6	1	3	4
Mean	.8370	.8308	.8414	.7823	.9205	.9058	.8950	.9052	66	65	60	54

Table 9. Normalized values of duration of expiration of 2nd & 3rd breaths after a positive pressure pulse was applied to P.S.R. intact animals.

RB	2ND BREATH				3rd BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	.8609	1.007	.7143	.8782	.8904	.9587	.8000	.9734	4	7	5	0
2	---	1.051	1.203	1.248	---	.9895	1.029	.8686	2	0	6	8
3	.5944	.7890	.7417	.8012	.6550	.9426	1.000	.9367	5	10	4	4
4	.7792	.8318	.8060	.8116	.9667	.9098	.9373	.9595	6	5	5	7
5	.8789	.9029	.8249	.8281	.9157	.9640	.9101	.9065	8	10	4	1
6	.8743	.8253	.6849	.8832	.9822	.9306	.8496	.8666	11	6	4	3
7	.8676	.9073	.8532	.8899	.9200	.9158	.8729	.9021	6	5	6	7
8	.9517	.8995	.9611	.8892	.9481	.8965	.8709	.8740	13	9	2	0
9	.8513	.8563	.8730	.9051	.8464	.8360	.8730	.8203	9	5	5	5
10	.8582	.6890	.8246	.7974	.8081	.7652	.8604	.7465	5	11	4	4
11	.9097	.8658	.8329	.7878	.8401	.8593	.8846	.8286	2	4	8	8
12	.7170	.8525	.7955	.8877	.8680	.8999	.8769	.9507	1	9	7	5
Mean	.8596	.8733	.8365	.8940	.8913	.9055	.9004	.8960	72	81	60	52

Table 10. Normalized values of duration of expiration of 2nd & 3rd breaths after a negative pressure pulse was applied to P.S.R. intact animals.

There was no significant differences in effect between the pulse positions in either breath.

Fvalues	2nd Breath	3rd Breath	df
+ Intact	.59	.05	3&242
- Intact	.25	.24	3&190
+ Blocked	.03	.04	3&200
- Blocked	.36	.04	3&221

There was no significant differences in effect between the treatments in either breath.

Fvalues	2nd Breath	3rd Breath	df
pos 1	.13	1.02	3&212
pos 2	.17	.06	3&240
pos 3	.13	.29	3&198
pos 4	.65	.39	3&175

The shortening of the second tE was significantly less than the shortening of tE containing the pulse after P.S.R. were effectively blocked (rabbits 5, 9, 1, and 7). The shortening of the third tE was significantly less than the second tE.

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	1.048	1.037	.9956	1.001	.9415	.9375	.9855	1.017	2	7	5	2
2	.8274	.9098	.9098	.8446	.9524	.9385	.9335	.9704	2	4	4	8
3	.5920	.7001	.7007	.7924	.8937	.9377	.9377	.9654	5	6	6	6
4	.7093	.6272	.6756	.6606	.8661	.9363	.8144	.9712	6	7	3	7
5	.8763	.8877	.8705	.9211	.9609	1.001	.9649	.9474	12	6	5	1
6	.8839	.8748	.8788	.7805	.9598	.9146	.9747	.9239	10	4	7	3
7	.4395	.6542	.6542	.5769	.6016	.7747	.7747	.7764	4	4	3	8
8	.8150	.8558	.8582	---	.8964	.8527	.9130	---	8	7	9	0
9	1.055	.8945	.9311	.9636	1.038	.8884	.9391	.8789	8	7	3	3
10	.8667	.8275	.8273	.8835	.9063	.8631	.8834	.9706	7	4	7	3
11	---	.7573	.7500	.7019	---	.9633	.8661	.8333	0	4	5	9
12	.8386	.7655	.7957	.8009	.9677	.8649	.9396	.9191	1	6	3	4
Mean	.8299	.8199	.8183	.7520	.9196	.9079	.9122	.9082	65	66	60	54

Table 11. Normalized values of duration of expiration of 2nd & 3rd breaths after a positive pressure pulse was applied to P.S.R. blocked animals.

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	.9842	.9827	.9692	---	.9339	.9720	.9281	---	4	7	5	0
2	.8883	---	.8379	.8543	.8666	---	.9260	.9102	2	0	6	8
3	.7964	.7658	.7713	.7755	.8901	.8706	.9480	.9140	5	10	4	4
4	.7745	.6930	.7036	.6278	.8877	.9327	.8222	.8060	6	5	5	7
5	.9394	.9215	.9411	.9589	.9810	.9667	.9328	.9863	8	10	4	1
6	.9245	.8864	.8627	.7863	.9138	.9136	.8722	.8739	11	6	4	3
7	.6195	.6254	.6330	.6274	.7240	.7867	.8293	.7751	6	5	6	7
8	.8482	.8606	.8728	---	.8084	.8606	.9337	---	13	9	2	0
9	.9210	.8697	.8803	.9319	.9639	.9200	.8713	.9017	9	5	5	5
10	.7100	.7584	.7003	.7385	.7428	.7922	.7550	.7497	5	11	4	4
11	.7094	.5562	.6635	.6498	.7051	.7569	.7209	.7492	2	4	8	8
12	.7647	.7965	.8041	.7164	.8824	.8257	.8181	.7693	1	9	7	5
Mean	.8444	.8085	.7896	.7392	.8706	.8738	.8505	.8262	72	81	60	52

Table 12. Normalized values of duration of expiration of 2nd & 3rd breaths after a negative pressure pulse was applied to P.S.R. blocked animals.

	F values	df	significance
+Intact	6.32	1&512	5%
-Intact	12.6	1&464	1%
+Blocked	4.21	1&482	5%
-Blocked	5.59	1&522	1%

FIRST INSPIRATION AFTER THE PRESSURE PULSES

Positive pressure pulses with P.S.R. intact

See table 13 for effect of positive pressure pulses applied in expiration on tI following the pulse with P.S.R. intact. Table 17 shows number of times tI following the pulse is shortened, lengthened or no effect after pressure pulses.

A non significant increase in tI following the pulse was seen after positive pressure pulses were applied in expiration with P.S.R. intact.

	mean diff	t value	df
Pos 1	.011	1.57	64
Pos 2	.023	1.50	72
Pos 3	.028	1.84	61
Pos 4	.024	1.87	62

There was no significant difference in effect between positions (F=.357 3&260df).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.785	6	.733	5	.810	3	1.08	3
2	.909	1	.924	5	.857	5	.757	4
3	1.28	5	1.46	4	1.46	4	1.06	8
4	1.17	3	1.12	8	1.33	5	1.35	6
5	1.06	7	1.06	5	1.00	6	1.09	5
6	1.02	6	1.05	8	.978	6	1.03	4
7	1.05	11	1.05	7	1.00	4	.949	2
8	1.06	5	1.03	6	1.00	6	.948	7
9	1.02	7	1.05	9	1.01	3	1.02	3
10	1.06	7	1.08	6	1.14	5	1.10	5
11	1.12	1	1.12	6	1.10	7	1.07	8
12	1.05	3	1.07	4	1.11	8	1.06	8
Mean	1.05	62	1.06	73	1.07	62	1.06	63

Table 13. Normalized values of the 1st tI after the pulse when a positive pressure pulse was applied to P.S.R. intact animals.

Negative pressure pulses with P.S.R. intact

See table 14 for effect of negative pressure pulses applied in expiration on tI following the pulse with P.S.R. intact. Table 17 shows number of times tI following the pulse is shortened, lengthened or no effect after pressure pulses.

Negative pressure pulses applied early in expiration did not significantly affect the tI following the pulse with P.S.R. intact (mean -.003, t=.227 57df, pos 1). When negative pulses were applied later in expiration a significant decrease at the 1% level in tI following the pulse occured.

	mean diff	t value	df
Pos 2	.069	5.76	64
Pos 3	.092	9.15	61
Pos 4	.107	6.27	55

This difference was greater in the later positions (F=1.94 3&238df).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.874	5	1.01	6	1.09	1	.899	3
2	---	0	1.16	4	1.02	3	1.09	6
3	1.32	3	1.38	5	1.27	6	1.30	8
4	1.35	3	1.26	7	1.31	6	1.36	7
5	1.00	9	1.02	6	1.15	6	1.26	3
6	.895	7	1.10	4	1.07	4	1.17	5
7	1.03	5	1.03	7	1.09	7	1.10	2
8	.915	8	1.05	5	1.11	5	1.28	3
9	.979	9	1.13	7	1.20	2	1.10	4
10	1.10	4	1.29	3	1.22	7	1.23	3
11	.982	4	1.10	5	1.19	9	1.15	2
12	1.31	1	1.04	6	1.16	6	1.15	4
Mean	1.01	58	1.12	65	1.17	62	1.19	50

Table 14. Normalized values of the 1st tI after the pulse when a negative pressure pulse was applied to P.S.R. intact animals.

Positive pressure pulses with P.S.R. blocked

See table 15 for effect of positive pressure pulses applied in expiration on tI following the pulse with P.S.R. blocked. Table 17 shows number of times tI following the pulse is shortened, lengthened or no effect after pressure pulses.

A non significant increase in tI following the pulse was seen after positive pressure pulses were applied in expiration with P.S.R. blocked.

	mean diff	t value	df
Pos 1	.018	1.26	55
Pos 2	.020	1.62	63
Pos 3	.008	.965	81
Pos 4	.026	.163	53

There was no significant difference in effect between positions (F=.232 3&253df).

Effective P.S.R. block (Rb 5, 9, 1, & 7) had significantly more shortening than poor P.S.R. block at the 1% level (F=6.93 1&131df).

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	.951	2	1.01	7	.914	5	.889	2
2	1.03	2	.950	2	.886	4	.873	8
3	1.06	5	1.14	6	1.07	6	1.07	6
4	1.13	6	1.02	7	1.07	3	1.10	7
5	1.00	12	1.01	6	.964	5	.964	1
6	1.02	10	1.03	4	1.06	7	1.07	3
7	1.04	4	.999	4	.997	3	1.05	8
8	.992	8	1.02	7	.994	9	---	0
9	.967	8	1.00	7	1.05	3	1.03	3
10	1.01	7	1.06	4	1.06	7	1.07	3
11	---	0	1.04	4	1.01	5	1.01	9
12	1.04	1	1.05	6	1.04	3	1.02	4
Mean	1.02	65	1.03	64	1.02	66	1.02	54

Table 15. Normalized values of the 1st tI after the pulse when a positive pressure pulse was applied to P.S.R. blocked animals.

Negative pressure pulses with P.S.R. blocked

See table 16 for effect of negative pressure pulses applied in expiration on tI following the pulse with P.S.R. blocked. Table 17 shows number of times tI following the pulse is shortened, lengthened or no effect after pressure pulses.

Negative pressure pulses applied early in expiration did not significantly affect the tI following the pulse with P.S.R. blocked (mean -.009, t=.880 71df, pos 1). When negative pulses were applied later in expiration a significant decrease at the 1% level in tI following the pulse occured.

	mean diff	t value	df
Pos 2	.022	3.12	80
Pos 3	.036	3.29	59
Pos 4	.055	4.24	51

This difference was not significantly greater in the later positions (F=1.29 3&261df).

After effective block (Rb 5, 9, 1, and 7) of P.S.R. negative pressure pulses caused a decrease in tI following the pulse in all positions This is significant to the 1% level in pos 1, but not significant in later positions.

	mean diff	t value	df
Pos 1	-.024	3.87	26
Pos 2	-.013	.172	26
Pos 3	-.0006	.059	19
Pos 4	-.012	.082	12

RB	POS1	N	POS2	N	POS3	N	POS4	N
1	1.04	4	1.01	7	1.03	5	---	0
2	.854	2	---	0	.984	6	.970	8
3	1.20	5	1.14	10	1.08	4	1.20	4
4	1.23	6	1.16	5	1.10	5	1.19	7
5	.950	8	.993	10	1.00	4	.975	1
6	1.01	11	.970	6	1.11	4	1.17	3
7	.937	6	.957	5	1.00	6	.970	7
8	1.00	13	1.03	9	1.05	2	---	0
9	.981	9	.975	5	.990	5	1.02	5
10	1.00	5	1.09	11	1.03	4	1.07	4
11	1.03	2	1.08	4	1.11	8	1.11	8
12	1.08	1	1.02	9	1.09	7	1.16	5
Mean	1.02	80	1.04	81	1.05	60	1.08	52

Table 16. Normalized values of the 1st tI after the pulse when a negative pressure pulse was applied to P.S.R. blocked animals.

This effect was not significantly different between postions (F=.149 3&78df).

In the rabbits with least effective block of P.S.R. (Rb 4, 11, 8, and 12) negative pressure pulses caused a significant increase (1% level) in the tI following pulse.

	mean diff	t value	df
Pos 1	.044	3.46	21
Pos 2	.040	6.42	26
Pos 3	.064	21.4	21
Pos 4	.102	36.5	19

This was not significantly different between pulse positions (F=.290 3&83df).

	+ Intact		− Intact		+ Blocked		− Blocked	
	+	tot	+	tot	+	tot	+	tot
1	4	17	3	15	5	15	7	16
2	1	17	8	14	2	16	6	16
3	18	21	17	24	15	23	19	24
4	17	22	19	23	13	24	19	24
5	15	23	13	24	9	24	5	22
6	15	24	14	20	16	24	12	24
7	8	24	13	21	12	24	6	23
8	12	24	15	21	11	24	13	24
9	14	22	15	20	9	22	8	23
10	17	23	16	17	21	24	21	24
11	21	22	19	20	8	19	20	24
12	21	23	15	17	13	21	19	23
All	163	260	177	235	140	249	154	173

Table 17. Numbers of times duration of inspiration increased (+) total number of runs (tot) with pressure pulses in all treatments.

2ND AND 3RD INSPIRATION AFTER PRESSURE PULSE

See tables 18 and 21 for the effect of pressure pulses applied in expiration on the second and third tI following the pulse with P.S.R. intact and blocked.

Pressure pulses usually do not change the second and third tI after pulse application regardless of sign of pulse, block of PSR, or position pulse was applied in. However occasional large shortening occurs in all treatments. This shortening was significant in some positions with some treatments.

t values	2nd tI	sig	3rd tI	sig	df
+Intact	3.39	1%	.365	ns	260
-Intact	1.07	ns	1.81	ns	241
+Blocked	3.93	1%	2.66	1%	265
-Blocked	.016	ns	4.26	1%	273

The sign test shows these large shortening of tI was significant to the 5% and 1% levels.

2nd breath	number shortened	number lengthened	sig
+Intact	127	88	1%
-Intact	110	97	5%
+Blocked	147	92	1%
-Blocked	121	108	5%
3rd breath			
+Intact	135	77	1%
-Intact	107	93	5%
+Blocked	145	87	1%
-Blocked	151	86	1%

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	.978	.963	.942	1.03	.940	.929	.865	1.02	6	5	3	3
2	1.09	.967	.800	.866	.909	.992	.935	1.02	1	5	5	4
3	.920	.959	.959	.962	1.03	.984	.984	.993	5	4	4	8
4	1.00	1.00	.917	.973	.944	1.03	.957	.993	3	8	5	6
5	.974	.938	.984	.979	.982	.967	.944	,965	7	5	6	5
6	.961	.960	.983	1.02	.978	.966	.936	1.01	6	8	6	4
7	.995	.976	.977	1.05	.997	.934	.997	.994	11	7	4	2
8	.993	1.02	1.06	1.02	.938	.990	.994	.993	5	6	6	7
9	.990	.982	.951	.999	.994	.982	.960	1.04	7	9	3	3
10	.948	.957	1.08	.990	.962	1.01	1.03	1.00	7	6	5	5
11	1.00	1.04	1.02	1.04	1.00	1.02	1.02	1.01	1	6	7	8
12	1.01	.983	1.01	.992	1.03	.991	1.00	.976	3	4	8	8
Mean	.976	.980	.981	.992	.983	.984	.974	.997	62	73	62	63

Table 18. Normalized values of duration of inspiration of 2nd & 3rd breaths after a positive pressure pulse was applied to P.S.R. intact animals.

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	PO
S4												
1	.958	.978	.857	.867	1.02	.938	1.03	.914	5	6	1	3
2	---	.944	.886	.992	---	1.04	.967	.984	0	4	3	6
3	.900	1.02	1.04	1.01	1.00	1.02	.990	1.01	3	5	6	8
4	1.03	1.02	.883	.950	.970	1.03	.952	.951	3	7	6	7
5	.968	1.00	.990	1.11	1.03	1.04	.978	1.05	9	6	6	3
6	.962	.980	1.07	.986	.923	.986	1.03	.965	7	4	4	5
7	1.00	1.04	1.01	.961	.986	.981	1.01	.999	5	7	7	2
8	1.02	1.05	1.09	1.10	.997	.964	.992	1.03	8	5	5	3
9	.966	.997	.986	.971	.953	.970	.955	.987	9	7	2	4
10	1.04	1.10	1.01	1.00	1.04	1.16	1.03	1.03	4	3	7	3
11	1.03	.988	1.03	1.02	1.02	.979	1.00	1.00	4	5	9	2
12	.923	1.02	1.00	1.01	.962	1.02	1.00	1.01	1	6	6	4
Mean O	.984	1.00	1.00	.990	.989	1.00	.997	.990	58	65	62	5

Table 19. Normalized values of duration of inspiration of 2nd & 3rd breaths after a negative pressure pulse was applied to P.S.R. intact animals.

There was no significant differences in effect between the pulse positions in either breath.

F values	2nd Breath	3rd Breath	df
+ Intact	.357	2.08	3&242
- Intact	.619	.167	.3&190
+ Blocked	.103	.114	3&200
- Blocked	.143	.346	3&221

There was no significant differences in effect between the treatments in either breath.

Fvalues	2nd Breath	3rd Breath	df
pos 1	.327	.091	3&212
pos 2	.688	2.13	3&240
pos 3	.333	.500	3&198
pos 4	.444	.900	3&175

The shortening of the third tI was significantly less than the second tI at the 1% level.

	Fvalues	df	level of significance
+Intact	8.87	1&512	5%
-Intact	3.75	1&464	1%
+Blocked	6.93	1&482	5%
-Blocked	30.3	1&522	1%

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	1.07	1.02	.982	.889	.967	1.05	.954	.889	2	7	5	2
2	.952	.924	.989	.964	1.05	1.08	.944	.995	2	2	4	8
3	.944	1.00	.901	.910	.991	1.01	.938	.937	5	6	6	6
4	.900	.952	.870	.971	.994	.894	.955	.877	6	7	3	7
5	.957	.960	.962	.940	.985	.947	.961	1.01	12	6	5	1
6	.996	1.02	.991	1.06	.979	1.02	.999	1.02	10	4	7	3
7	1.02	1.03	.955	.971	1.15	1.00	1.01	1.06	4	4	3	8
8	.981	1.04	.996	---	.978	.997	.980	---	8	7	9	0
9	.962	.964	.976	1.01	.969	.969	.987	.986	8	7	3	3
10	1.02	.990	1.01	.961	1.00	1.01	1.01	.975	7	4	7	3
11	---	.915	.985	1.03	---	.980	.965	.977	0	4	5	9
12	.928	.976	1.02	1.01	.928	.986	.998	.987	1	6	3	4
Mean	.974	.987	.978	.979	.995	.987	.981	.975	65	64	66	54

Table 20. Normalized values of duration of inspiration of 2nd & 3rd breaths after a positive pressure pulse was applied to P.S.R. blocked animals.

RB	2ND BREATH				3RD BREATH				NUMBER OF RUNS			
	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4	POS1	POS2	POS3	POS4
1	1.00	1.04	.985	---	.963	.999	.969	---	4	7	5	0
2	.999	---	1.05	.995	.955	---	.988	1.00	2	0	6	8
3	1.02	1.01	.962	.986	.957	1.01	.962	1.09	5	10	4	4
4	.917	1.03	.929	.962	.997	.975	.926	.946	6	5	5	7
5	1.02	.969	.995	1.00	1.01	.976	.983	.975	8	10	4	1
6	1.01	.989	1.03	1.10	1.02	.997	1.01	.945	11	6	4	3
7	1.07	1.02	1.01	1.02	1.00	1.02	1.02	.978	6	5	6	7
8	1.02	1.05	.988	---	.969	1.04	.988	---	13	9	2	0
9	.961	.932	.980	1.02	.968	.969	.950	.968	9	5	5	5
10	.997	1.03	.991	1.02	.986	1.02	.985	1.02	5	11	4	4
11	1.04	.947	.973	.964	1.02	.942	.965	.977	2	4	8	8
12	.917	.975	.969	.980	.917	.993	.988	.983	1	9	7	5
Mean	1.00	1.00	.988	.998	.986	.999	.978	.987	80	81	60	52

Table 21. Normalized values of duration of inspiration of 2nd & 3rd breaths after a negative pressure pulse was applied to P.S.R. blocked animals.

Relationship to previous tE

In all treatments tI was greatly shortened the preceeding tE was significantly shorter than the mean.

1st breath	mean tE when tI shortened	mean of all tE	F value	df	sig
+Intact	-.124	.066	4.75	3&327	1%
-Intact	-.706	-.516	6.54	3&275	1%
+Blocked	-.154	-.107	3.09	3&341	5%
-Blocked	-.369	-.301	1.45	3&354	ns
2nd breath					
+Intact	-.170	-.154	3.39	3&390	5%
-Intact	-.145	-.130	3.32	3&339	5%
+Blocked	-.165	-.139	1.85	3&368	ns
-Blocked	-.185	-.153	2.50	3&382	ns
3rd breath					
+Intact	-.101	-.083	3.36	3&395	5%
-Intact	-.129	-.093	3.33	3&344	5%
+Blocked	-.142	-.067	4.40	3&364	1%
-Blocked	-.140	-.117	2.91	3&375	5%

PRESSURE PULSES AFTER VAGOTOMY

Pressure pulses of either sign after vagotomy did not significantly affect tE or tI subsequent to pulse (table 22).

	tE of pulse	2nd tE	3rd tE	df
-Pulses				
means	-.028	-.003	.02	
t value	.538	1.06	.952	5
-Pulses				
means	-.047	.003	0	
t value	1.74	.200	.94	5
	1st tI	2nd tI	3rd tI	df
+Pulses				
means	.015	-.012	.01	
t value	.938	.414	.385	5
-Pulses				
means	-.035	-.013	0	
t value	1.67	.59	0	5

PRESSURE PULSES AFTER DIFFERENTIAL COLD BLOCK

Pressure pulses of either sign after differential cold block did not significantly affect tE or tI subsequent to pulse (table 23).

	tE of pulse	2nd tE	3rd tE	df
-Pulses				
means	-.005	-.002	.02	
t value	.625	.111	.167	13
-Pulses				
means	-.005	0	-.009	
t value	.510	0	.612	13
	1st tI	2nd tI	3rd tI	df
+Pulses				
means	-.001	-.0007	-.001	
t value	.091	1	.07	13
-Pulses				
means	-.005	-.005	-.001	
t value	.278	.547	.189	13

AUGMENTED BREATHS

Augmented breaths occur significantly more frequently with pressure pulses applied later in expiration, in all treatments.

	+Intact	-Intact	+Blocked	-Blocked
p value	.047	.014	.009	.025

Augmented breaths occur significantly more frequently with negative pressure pulses applied in expiration, before P.S.R. block.

	pos 1	pos 2	pos 3	pos 4
p value	.002	.100	.002	0

Augmented breaths occur significantly more frequently (at the 1% level) the 1st breath after the pressure pulse was applied (F=9.73 2&301df).

Positive pulses

	1st breath after pulse		2nd breath after pulse		3rd breath after pulse	
	tE	tI	tE	tI	tE	tI
Rabbit 5	.957	.989	1.02	1.00	1.01	.977
	1.05	1.00	1.03	.978	1.01	1.01
Rabbit 6	.866	1.03	1.00	1.07	.968	1.00
	.866	.944	.963	.915	1.03	1.07
Rabbit 7	.885	1.01	.933	1.00	.903	.917
	1.30	1.02	1.02	.988	.957	1.07
Means	.991	1.10	.994	.992	.980	1.01

Negative pulses

	1st breath after pulse		2nd breath after pulse		3rd breath after pulse	
	tE	tI	tE	tI	tE	tI
Rabbit 5	.957	.977	.965	1.00	.964	.978
	.972	.946	.952	.989	1.03	1.00
Rabbit 6	1.10	1.05	1.01	.916	1.01	.966
	.899	1.02	1.03	1.05	.870	1.04
Rabbit 7	.971	.980	.974	1.00	.974	.941
	1.20	1.07	1.04	.978	1.00	1.03
Means	1.02	1.01	.995	.989	.975	.993

Table 22. Effectsof vagotomy. Nomalized values (fraction of control).

	1st breath after pulse		2nd breath after pulse		3rd breath after pulse		
	tE	tI	tE	tI	tE	tI	N
Positive pulses							
Rabbit 12	.960	1.00	.900	.953	.963	1.01	6
Rabbit 13	1.01	.992	1.03	.995	1.02	.965	4
Rabbit 14	.998	1.04	.991	1.04	.982	1.04	4
Means	.985	1.01	.963	.986	.985	1.01	14
Negative pulses							
Rabbit 12	.923	.965	.975	.919	.989	1.00	6
Rabbit 13	.982	1.04	.998	1.04	.985	1.02	4
Rabbit 14	1.01	1.02	.989	.995	1.03	.976	4
Means	.965	1.00	.986	.975	1.00	.999	14

Table 23. Effects of differential cold block.
Normalized values (fraction of control).

		Pos 1	Pos 2	Pos 3	Pos 4
+ Intact	1st breath	0	4	2	2
	2nd breath	0	0	0	0
	3rd breath	0	0	0	0
	no. of runs	62	73	62	64
- Intact	1st breath	1	5	3	4
	2nd breath	2	0	0	4
	3rd breath	1	2	3	4
	no. of runs	58	66	62	50
+Blocked	1st breath	0	0	1	3
	2nd breath	0	0	0	0
	3rd breath	0	1	0	0
	no. of runs	65	66	60	54
-Blocked	1st breath	0	3	1	0
	2nd breath	0	0	0	0
	3rd breath	0	0	0	1
	no. of runs	72	81	60	52

Table 24. Number of augmented breaths which occured.

P.D.G. SERIES

Table 25 shows the effects of PDG injection.

PDG injected into the right atrium at least 15 minutes after treatment caused tachypnoea with all treatments. In all treatments this was due to a significant shortening in tE at the 1% level.

Treatment		mean change	t values	df
Noxylo-	Intact	-.619	9.83	16
	Vagot.	-1.09	9.08	15
	Gloss.	-1.46	4.82	4
Xylo-	Intact	-.237	3.54	8
	Vagot.	-1.11	3.76	6
	Gloss.	-1.50	5.36	3

In all treatments tI was shortened but this was only significant in the intact xylo state at the 1% level.

Treatment		mean change	t values	df
noxylo-	Intact	.045	2.14	16
	Vagot.	.041	.683	15
	Gloss.	.095	.528	4
Xylo-	Intact	.051	1.19	8
	Vagot.	.029	.362	6
	Gloss.	.214	.535	3

After xylocaine was injected into the pericardial sac the response of tE to PDG was significantly less at 1% than the control before xylocaine (t=16, 25df) but response of tI to PDG was not was not significantly different from the control before xylocaine (t=.265, 25df). The

NO XYLOCAINE

	INTACT		POST VAG.		POST IX SECTION	
	t_I	t_E	t_I	t_E	t_I	t_E
CONTROL	0.568 ± .03	0.96 ± .05	1.16 ± .7	1.50 ± .09	1.19 ± .04	1.88 ± .15
P.D.G.	0.50 ± .03	0.40 ± .06	1.13 ± .7	0.47 ± .1	1.10 ± .2	0.42 ± .20
	n = 19		n = 18		n = 5	

AFTER XYLOCAINE

CONTROL	0.63 ± .07	0.49 ± .06	0.87 ± .10	1.85 ± .33	1.20 ± .3	2.57 ± .37
P.D.G.	0.58 ± .07	0.24 ± .04	0.85 ± .20	0.747 ± .23	0.99 ± .2	1.19 ± .17
	n = 9		n = 7		n = 5	

TABLE 25. EFFECTS OF P.D.G. ON T_I AND T_E, ABSOLUTE VALUES IN SECS.

response of tE and tI to PDG at least 10minutes after vagotomy was significantly greater at the 5 & 10% level than the control before vagotomy (t=2.23, 13df; t=1.96, 13df for tE &tI respectively) regardless of xylocaine administration. The response of tE to PDG at least 15 min after the glossopharyngeal nerve was cut was significantly less at the 5% level from control before nerve section (t=2.92, 9df) regadless of xylocaine administration. The response of tI at least 15 min after the glossopharyngeal nerve ws cut was greater than the control before the nerve was cut was greater than control before nerve section (t=.635, 9df) regardless of xylocaine administration.

PDG injection caused apnoea 19 out of 44 runs before xylocaine was administered in all three treatments. After xylocaine was injected into the pericardial sac apnoea never occured in 20 runs. this lack of apnoea after xylocaine was significant to the 1% level (p=.00016).

PDG injected immediately after vagotomy did not significantly shorten tE (mean=-.013, t=.18, 3df) or tI (mean=.005, t=.093, 3df). The shortening of tE & tI when PDG was injected 60 & 70 min after vagotomy was significantly greater at the 1% level than when injected 15 to 25 min after vagotomy.

	mean diff tE	mean diff tI
15-25 min	-.992	.021
60-70 min	-1.38	-.10
t value	3.36	2.00
df	5	5

DISCUSSION

THE TRANSIENT EFFECT ON EXPIRATION OF PRESSURE PULSES IN EXPIRATION

Pulses of inflation and deflation administered to the lungs in expiration affect the duration of the expiration containing the pulse (Davies & Vizek 1982). This effect will be referred to as the transient effect of the pressure pulse. Vagotomy abolishes this effect suggesting that receptor activity of vagal fibres is involved.

There are three known lung receptors with vagal afferents which are considered to influence respiration (Paintal 1973a). The rapidly adapting receptor (R.A.R.) and the pulmonary stretch receptor (P.S.R.) both have myelinated vagal fibres. There are also the type J receptors (J.R.) which have nonmyelinated vagal fibres (Paintal 1973a). As mentioned later (p99) there may be other nonmyelinated fibres which influence respiration.

Large inflations and deflations are required to stimulate J.R (Paintal 1969; Sellick & Widdicombe 1970). The pulses of 20KPa used did not cause such large inflations and deflations. They were of similar in size to those used by Davies and Roumy (1982). That they were unlikely to be large enough to stimulate J.R. suggests that J.R. were unlikely to be involved. Differential cold block

of myelinated fibres was used to determine the involvement of nonmyelinated fibres. The transient effect of pressure pulses was abolished at temperatures which block myelinated fibres. Thus J.R. are not responsible for this effect.

As mentioned later (p 104) in respect to P.D.G. the effects of vagotomy may be time dependant. This may also be true of selective blocking of vagal fibres for example by cold block. In these experiments some of pulses were applied at least 10 minutes after vagotomy and cold block. In all cases vagotomy and cold block abolished the effect. Thus time dependence is unlikely to have obscured the results.

Thus the receptors mediating the transient effects of pressure pulses were unlikely to be J.R. or other nonmyelinated fibres. The majority of P.S.R. and all their effects can be blocked by inhalation of SO2 in the rabbit (Callanan, Dixon,& Widdicombe, 1975;Davies, 1976). Blocking P.S.R. with SO2 altered but did not abolish this effect. This suggests that both P.S.R. and R.A.R. were involved. This is supported by Davies and Roumy (1982) who suggest that expiration is controlled by a balance of P.S.R. and R.A.R. activity.

The von Euler model

von Euler 1977 proposed a model of respiratory control is based on spontaneously

increasing central inspiratory activity (C.I.A) giving the drive to inspire. This activity is terminated when it reaches a level that operates an off-switch (O-S). The rising level of C.I.A. activity and (P.S.R.) activity contributes to off switch activity while (R.A.R.) inhibit it. Other receptors may modify C.I.A. or O-S activity (see fig 2).

Duration of expiration is considered to be related to the previous inspiration, and may be modified if O-S or C.I.A. is modified. If the activity of the O-S is reduced then C.I.A. may begin earlier thus shortening expiration. If O-S activity is increased then C.I.A. may be inhibited for longer thus lengthening expiration.

Pulmonary stretch receptors have a spontaneous discharge (Paintal 1973a) at the end of expiration, the level of discharge being dependant on the degree of stretch of the lung. With inflation the receptors increase the rate of firing (Paintal 1973a). This in terms of von Eulers model would stimulate the off switch thus switching off the C.I.A. and lengthening duration of expiration. With deflation the receptors have a reduced level of firing (Paintal 1973a). This would reduce the stimulation of the off switch. Thus C.I.A. begin earlier and duration of expiration shortened.

R.A.R. do not have a spontaneous discharge, they discharge when stimulated by mechanical

deformation of the airways, this occurs with both inflation and deflation (Paintal 1973a). In terms of von Eulers model both inflation and deflation would suppress the off switch thus C.I.A. would be switched off later and duration of expiration would be shortened. However Sellick and Widdicombe point out that R.A.R. activity is too short in duration to maintain a shortening.

Inflation and deflation pulses

As shown in Fig14 inflation shortened the expiration containing the pulse when the pulse was applied early in expiration. However inflation lengthened the expiration when the pulse was applied later. When the pulse was applied at an intermediate stage little effect was seen. This suggests that R.A.R. are being stimulated and in early expiration the shortening caused, is greater than the lengthening caused by P.S.R. stimulation. The converse being true with pulses applied later. This also suggests that either the stimulation of P.S.R. becomes greater with pulses applied later in expiration or stimulation of R.A.R. becomes weaker with later pulses.

As shown in Fig14 deflation shortened the expiration containing the pulse regardless of the time in which the pulse was applied. This shortening of expiration could be due to both reduction of P.S.R. activity and/or stimulation of R.A.R. The shortening was greater when the pulse

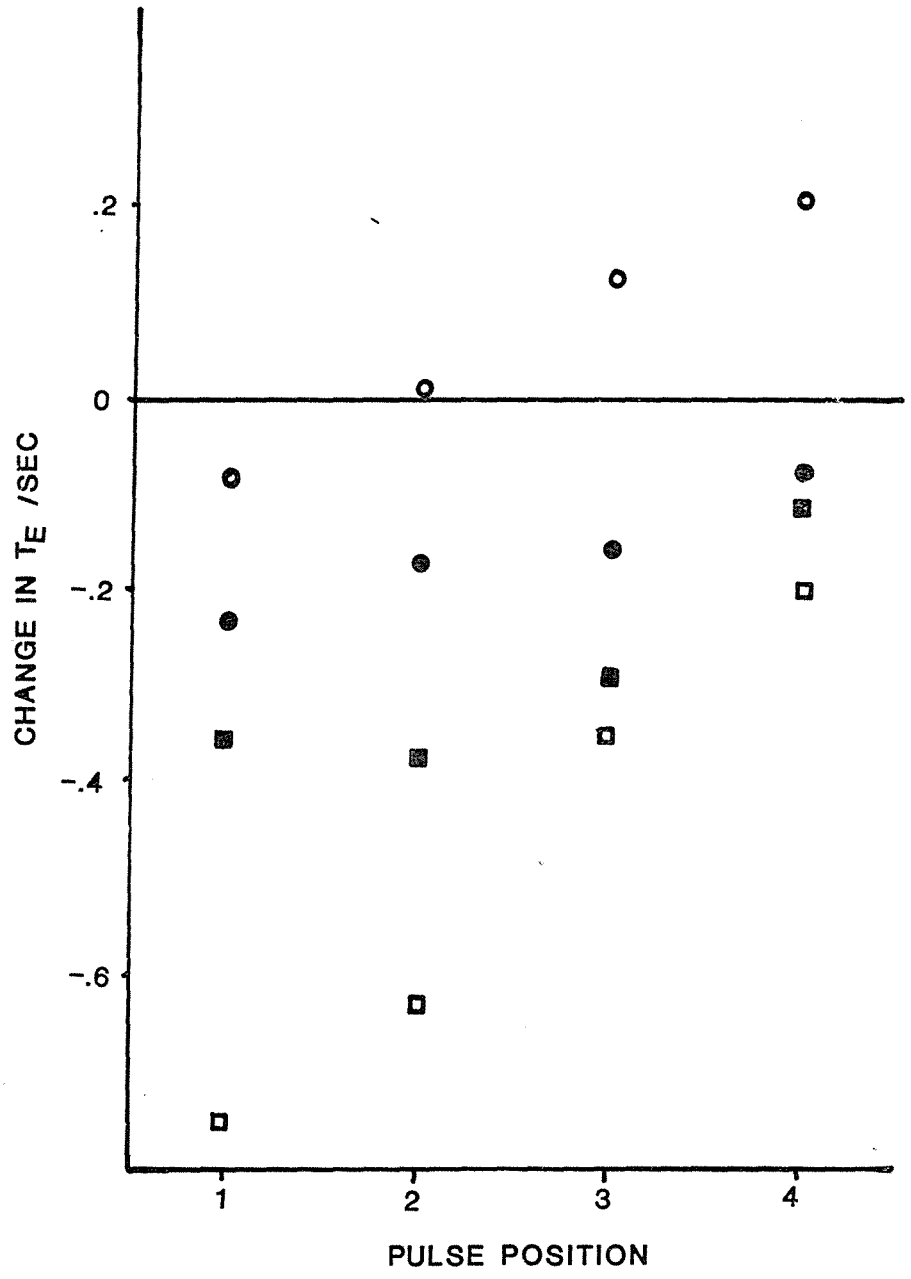


FIG 14. EFFECT OF ± PRESSURE PULSES ON T_E CONTAINING THE PULSE.

○ + INTACT ▫ - INTACT

● + BLOCKED ■ - BLOCKED

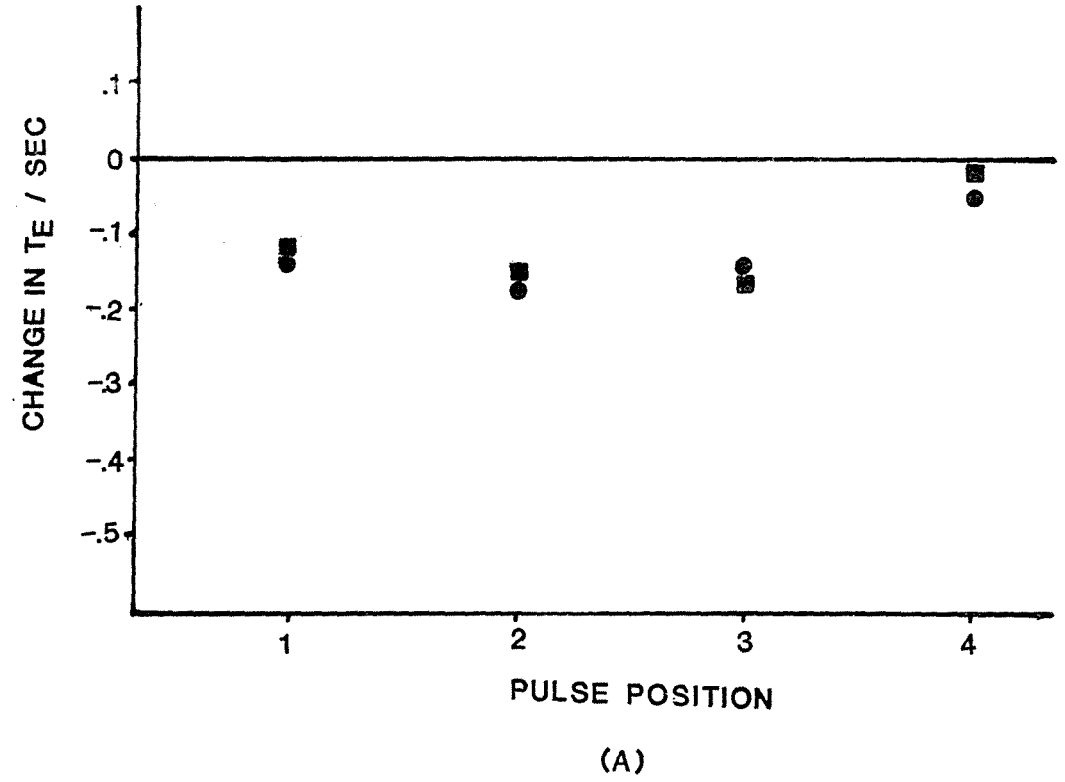
was applied early in expiration. This suggests that the strength of one or both of these receptors has activity dependant on time of pulse application.

Fig14 shows that blocking P.S.R. activity with SO2 shortens expiration regardless of whether the pulse inflation or deflation. This may represent shortening due to R.A.R. activity, however shortening is greater with negative pulses than positive. This difference may be due to incomplete blocking of P.S.R. activity.

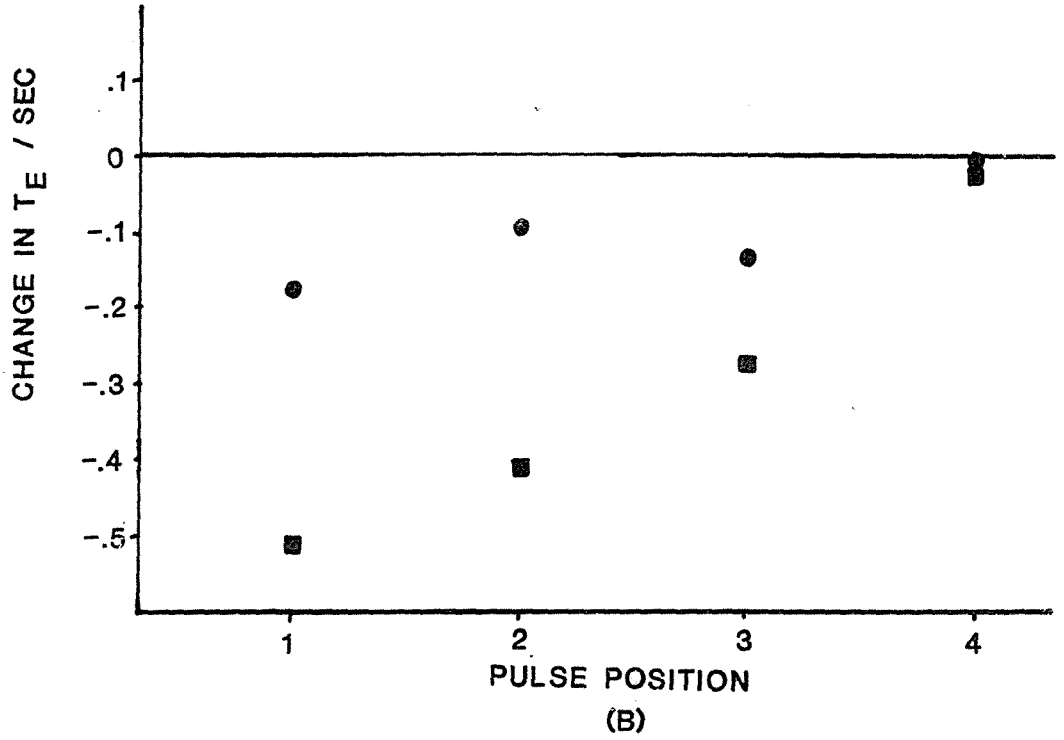
This is supported by comparing Fig15A&B. Fig15A shows positive and negative pulses after P.S.R. block in the rabbit with the most complete block (assessed by Hering-Breuer ratio). As can be seen there is no significant difference between the effects of positive and negative pulses in this rabbit. Fig15B shows that the rabbit with the least complete block has a large difference between positive and negative pulses.

Constant latency of shortening?

Davies and Roumy (1982) noted that deflation pulses and pulses after P.S.R. block shortened tE containing the pulse with approximately a constant latency. To test this with my results I tested differences in degree of shortening between the pulse positions. Deflation pulses in the intact rabbit shortened expiration significantly more with



(A)



(B)

FIG 15. EFFECT OF ± PULSES ON THE T_E CONTAINING THE PULSE WITH P.S.R. BLOCKED, (A) MOST EFFECTIVE BLOCK, (B) LEAST EFFECTIVE BLOCK.

● + BLOCKED ■ − BLOCKED

earlier pulse positions. In those rabbits with the most effective P.S.R. block the degree of shortening due to inflation or deflation pulses was not significantly different for the different pulse positions. Thus degree of stimulation of R.A.R. appears to vary with time of pulse application after effective P.S.R. block. Davies and Roumy (1982) recording R.A.R. activity, recorded greatest stimulation by deflation at FRC and by inflation at peak tidal volume. Thus they are stimulated greatest in late expiration by deflation and in early expiration by inflation. However R.A.R. activity is shortlived and "memory" of this activity must be involved to maintain the shortening of expiration. The strength of this "memory" decays with time, thus the relative strength between positions of the initial stimulus becomes less important and approximates constant latency of shortening.

The degree of difference between the various pulse positions is greater before P.S.R. block. Thus it appears that the effect on stimulation of the P.S.R. is dependant on time of pulse application. P.S.R. are stimulated proportionally by mechanical deformation caused by transpulmonary pressure (Davis,Fowler & Lambert 1956).

Thus changes in lung volume will change transpulmonary pressure and alter degree of P.S.R.

stimulation. Inflation early in expiration when the lungs are still inflated would stimulate less strongly than inflation when the lungs are deflated.

Similarly deflation early in expiration would produce a greater change in deformation than deflation later. However when P.S.R. activity drops below threshold further reduction in activity will not be more effective in reducing tE. As the change in R.A.R. stimulation depending on time of pulse application is relatively minor, deflation resulting in below threshold stimulation of P.S.R. would approximate a constant latency of shortening of tE as seen by Davies and Roumy (1982).

THE "MEMORY" EFFECT ON EXPIRATION OF PRESSURE PULSES IN EXPIRATION

Until relatively recently an individual breath has been considered to begin at the onset of one inspiration and end at the onset of the next expiration (Clark & von Euler 1972; Knox 1973). It was expected that inspiration may influence the following expiration, but one cycle has not been considered to influence the following cycle. This suggests that a pressure pulse applied in expiration will not affect the following breath, neither inspiration nor expiration.

Eldridge (1973 & 1974) showed that a disturbing stimulus within a respiratory cycle can alter the pattern of subsequent cycles. These results suggest that there is a "memory" linking respiratory cycles. A central mechanism for this was proposed by Eldridge (1974) and Karczweski et al (1976). The afferent impulses of receptor discharge cause a transient effect and allows reverberation of receptor discharge causing "memory" in subsequent breaths. Thus the "memory" effect of receptor discharge is likely to be similar in nature to the transient effect of the receptor.

The use of electrical stimulation may cause unusual transmitter persistance. However Davies and Kohl (1979) showed "memory" occurred when pressure pulses are the disturbing influence, which

may be more physiological. As pressure pulses stimulate P.S.R. and R.A.R. this "memory" may be of P.S.R. and/or R.A.R. activity. some "memory" of R.A.R. activity has been indicated by the ability of R.A.R. to maintain the deflation reflex after P.S.R. block, despite a short duration of receptor activity. As their receptor activity is shortlived any "memory" effect of these receptor activity continuing over into subsequent breaths must be central.

As "memory" is the reverberation of receptor activity, any "memory" of receptor activity is likely to affect the central "respiratory centres" in a manner similar to the direct effect of recptor activity. As discussed earlier , the transient effect of pressure pulses is likely to involve both P.S.R. and R.A.R. Thus if both P.S.R. and R.A.R. were involved the "memory" effect would be similar to the transient effect of pressure pulses. If P.S.R. only were involved positive and negative pulses would have opposite effects on duration of expiration. Block of P.S.R. by SO2 would abolish the "memory" effect. If R.A.R. only were involved pulses of opposite sign, blocked and intact states would have exactly the same effect of "memory". This should be similar to the transient effect after P.S.R. block.

Breaths subsequent to pressure pulses

As shown by Figs 16 & 17 pressure pulses significantly shorten the second and third expiration after the pulse is applied. This shortening occurs regardless of whether the pulse is inflation or deflation. Shortening occurs whether the expiration containing the pulse was shortened or lengthened.

The degree of shortening between inflation and deflation pulse is not significantly different. Block of P.S.R. shortens to the same degree as the shortening in the intact state. This suggests that the "memory" effect is due to R.A.R. discharge.

As can be seen in Figs 16 & 17 the shortening of the third breath is less than the shortening of the second breath after the pulse. After three to four breaths the effect is no longer significant. This suggests that the activity from the central "memory" diminishes with time.

von Eulers model of respiratory control is a functional model. Thus although the model does not include a basis for "memory", the model is compatible with it. In terms of this model R.A.R. facilitate the O-S and "memory" occurs by reverberation of R.A.R. activity in a "memory" pool of neurons. This "memory" pool would facilitate the O-S with diminishing activity as the reverberation dies away.

The shortening of duration of expiration was

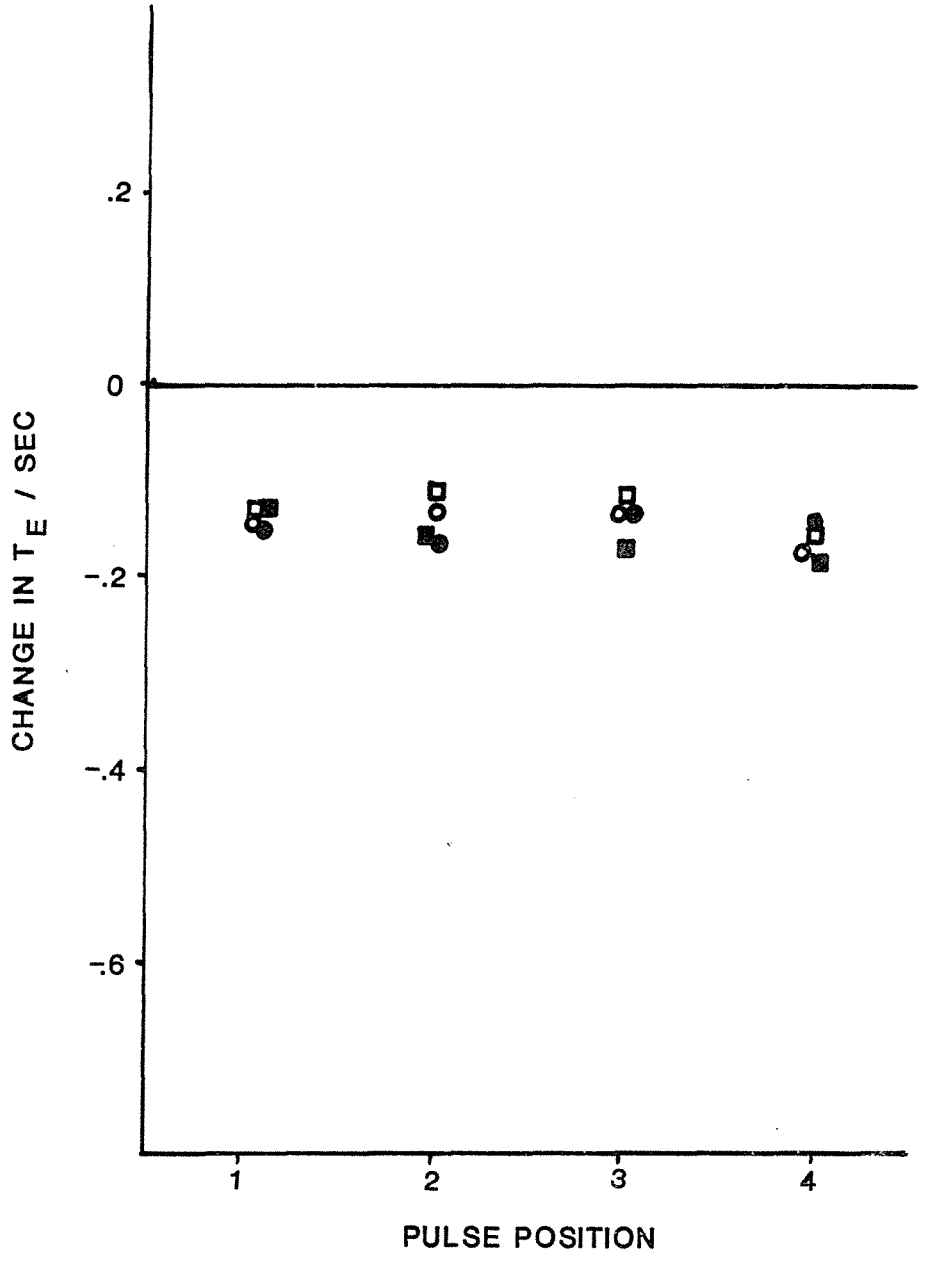


FIG 16. EFFECT OF ± PRESSURE PULSES ON THE SECOND T_E AFTER THE PULSE.

○ + INTACT ● + BLOCKED

□ - INTACT ■ - BLOCKED

reduced in the third breath after stimulation. There is no difference in degree of shortening between the different times of pulse application. Although the degree of R.A.R. activity varied with time of application of pulse, time has elapsed since the stimulus was applied, during this time activity dies away and strength of initial stimulus becomes less important. Thus the effect on shortening subsequent breaths is not significantly different over the different pulse positions.

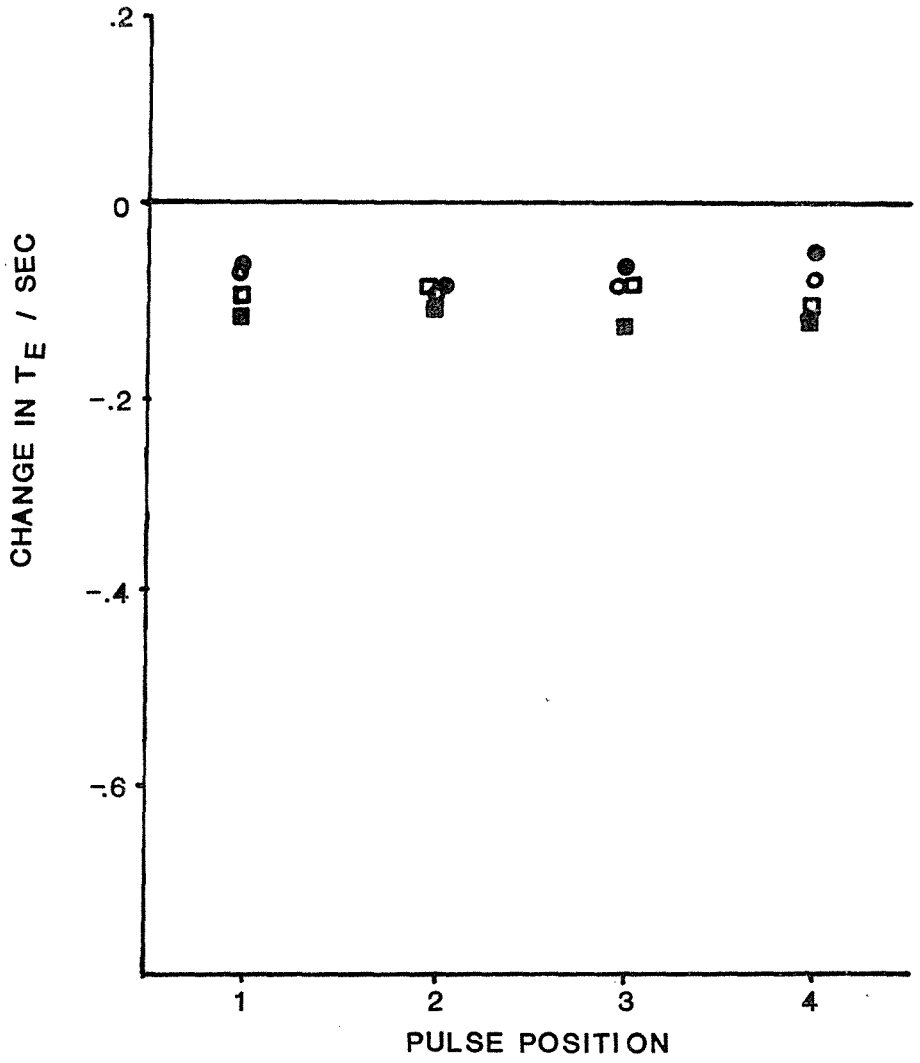


FIG 17. EFFECT OF ± PRESSURE PULSES ON THE THIRD T_E AFTER THE PULSE.

○ + INTACT ● - INTACT

□ + BLOCKED ■ - BLOCKED

THE EFFECTS ON DURATION OF INSPIRATION OF PRESSURE PULSES IN EXPIRATION

Karczewski et. al. (1976) reported that in the spontaneously breathing vagotomised rabbit electrical stimulation of the vagi produced decreases in tI and tE. The changes in tE lead tI. This suggests that a respiratory cycle is not independant of the preceeding cycle and there appears to be central linking of tE to the preceeding tI as well as tI to the preceeding tE.

Davies and Kohl 1979 using pressure pulses in expiration showed this link is not mandatory and suggested this may be associated with R.A.R. activity. They suggested that this linking occured only with large (greater than 50%) reduction of tE, when R.A.R. were stimulated strongly. Davies and Roumy (unpublished) used CO2 to alter tidal volume, the plot of tI/preceeding tE had three seperate populations of points intact, P.S.R. block, and vagotomy. This suggested that the relationship of tI to the preceeding tE involved R.A.R. activity.

Negative pressure pulses

As seen in fig 18 there was no significant effect of negative pressure pulses in expiration applied in position one on the following tI however with pulses applied in positions 2-4, the following tI was significantly increased. Mean change in tI increased from pos2 to 4.

Applying negative pressure pulses in expiration stimulates R.A.R. strongly. If linking of expiration to the following inspiration was due to R.A.R. activity deflation pulses would tend to shorten the inspiration following the pulse. However negative pulses reduce P.S.R. activity. "Memory" of this reduced activity would tend to lengthen the inspiration following the pulse. This effect would be greater with the pulses applied later, as less time has elapsed since the stimulus was applied.

This suggests that the increase in tI following deflation pulses applied in expiration is due to "memory" of reduced P.S.R. activity and R.A.R. are usually not stimulated enough to link tI to the preceeding tE. The mean tE preceeding a shortened tI is significantly less than the mean tE preceeding a lengthened tI. Strong stimulation of R.A.R. causes large shortening of tE containing pulse, giving support to the concept that linking of tI to the preceeding tE is due to strong stimulation of R.A.R.

As seen in fig 18 blocking stretch receptors with sulphur dioxide leaves the effect of rapidly adapting recptors only. Thus if linking of tI to the previous tE is due to R.A.R. the effect of negative pressure pulses in expiration after stretch receptors were blocked would be expected to shorten the following inspiration.

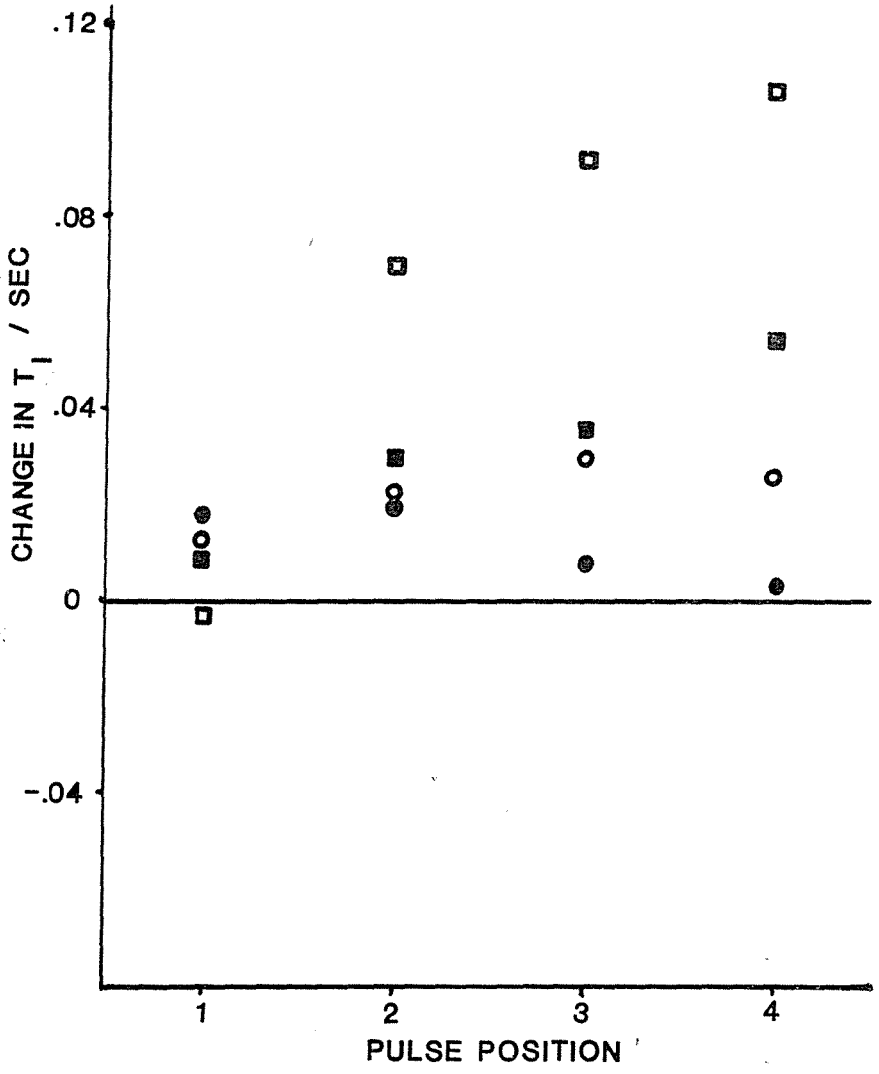


FIG 18. EFFECT OF ± PRESSURE PULSES ON THE FIRST T_1 AFTER THE PULSE.

○ + INTACT □ - INTACT

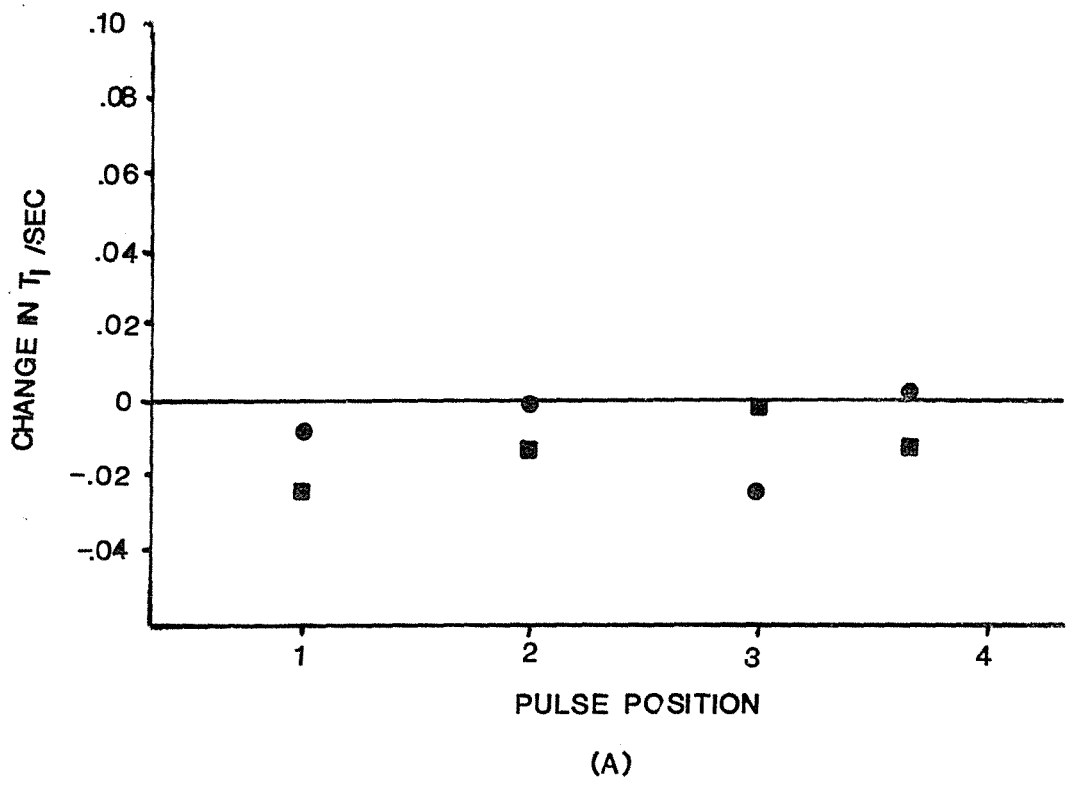
● + BLOCKED ■ - BLOCKED

However my results showed a significant lengthening. Incomplete stretch receptor block may have obscured results, due to "memory" of reduced P.S.R. activity. This was supported as the rabbits which had most complete block had significant shortening while those with least complete block had significant lengthening of inspiration following the pressure pulse (fig 19).

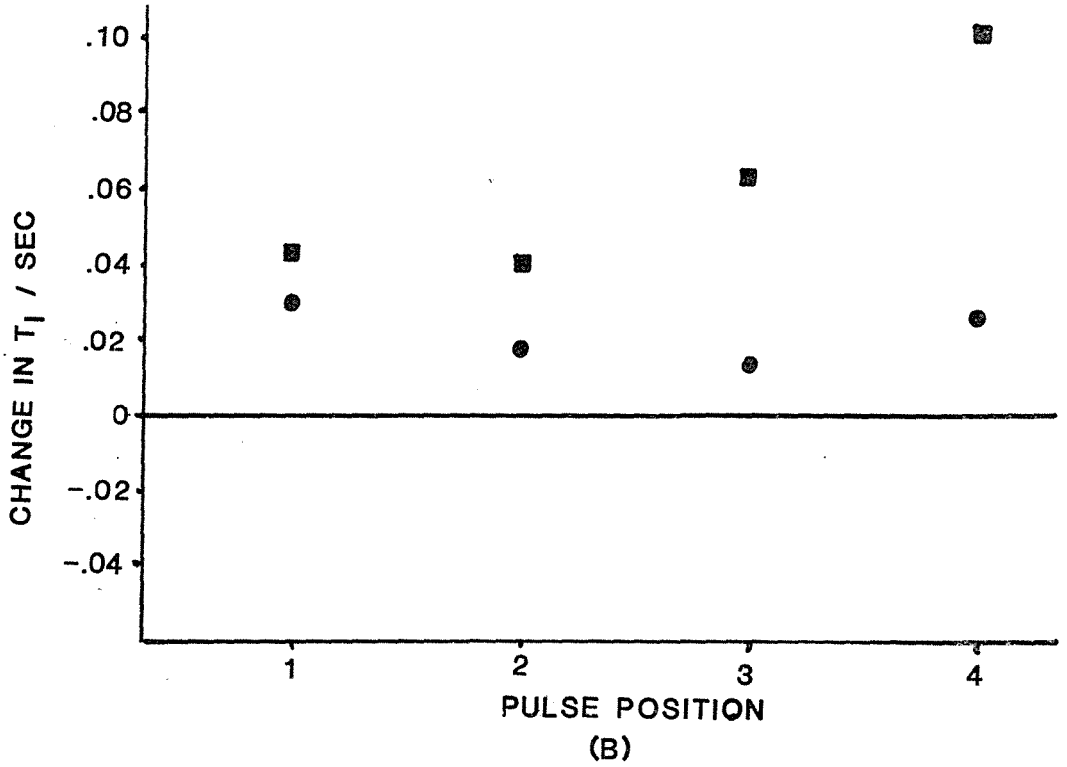
Positive pressure pulses

As seen in fig 18 positive pressure pulses in expiration did not significantly influence the following inspiration. A non significant lengthening of the following inspiration regardless of stretch receptor block occured. "Memory" of P.S.R. activity from a positive pressure pulse in expiration would tend to shorten the duration of the following inspiration as shown by greater shortening after effective P.S.R. block. This did not occur suggesting that "memory" of P.S.R. activity if it occurs is weak. This is despite strong stimulation of P.S.R. in the later pulse positions. "Memory" of reduced P.S.R. activity occured with negative pulses. This suggests that "memory" due to P.S.R. may be a dampening of reverberating circuits rather than P.S.R. activity setting off reverberation, leading to "memory" effects.

As R.A.R. are not strongly stimulated by positive pressure pulses the effect of "memory" of



(A)



(B)

FIG 19. EFFECT OF ± PULSES ON THE FIRST T_1 AFTER THE PULSE WITH P.S.R. BLOCKED, (A) MOST EFFECTIVE BLOCK, (B) LEAST EFFECTIVE BLOCK.

● +BLOCKED ■ - BLOCKED

R.A.R. activity from a positive pressure pulse given in expiration may have little influence on the following inspiration. However although R.A.R. activity is weak with positive pressure pulses it is strong enough to inflence tE of the 2nd and 3rd breath after stimulus and sometimes influence tI of these breaths.

Thus the slight lengthening seen suggests that there is some influence other than P.S.R. or R.A.R. activity. This is unlikely to be due to changes in blood gases, as discussed later (p93). This may be a response to changes in mechanics of breathing due to an inflation pulse. This response may obscure any "memory" of P.S.R. and R.A.R. activity after a positive pressure pulse. However any similar response due to a deflation pulse would be unlikely to affect tI in the same manner, rather it would tend to shorten tI. Thus it is unlikely that the "memory" response is merely due to changes in the mechanics of breathing.

Subsequent breaths

As seen in fig 20 & 21 duration of inspiration of subsequent breaths was not usually changed regardless of sign of pulse or whether P.S.R. were blocked occasionally a large shortening occured. This shortening was significant in several pulse positions with different treatments and significant in some treatments over all pulse positions. Asthe

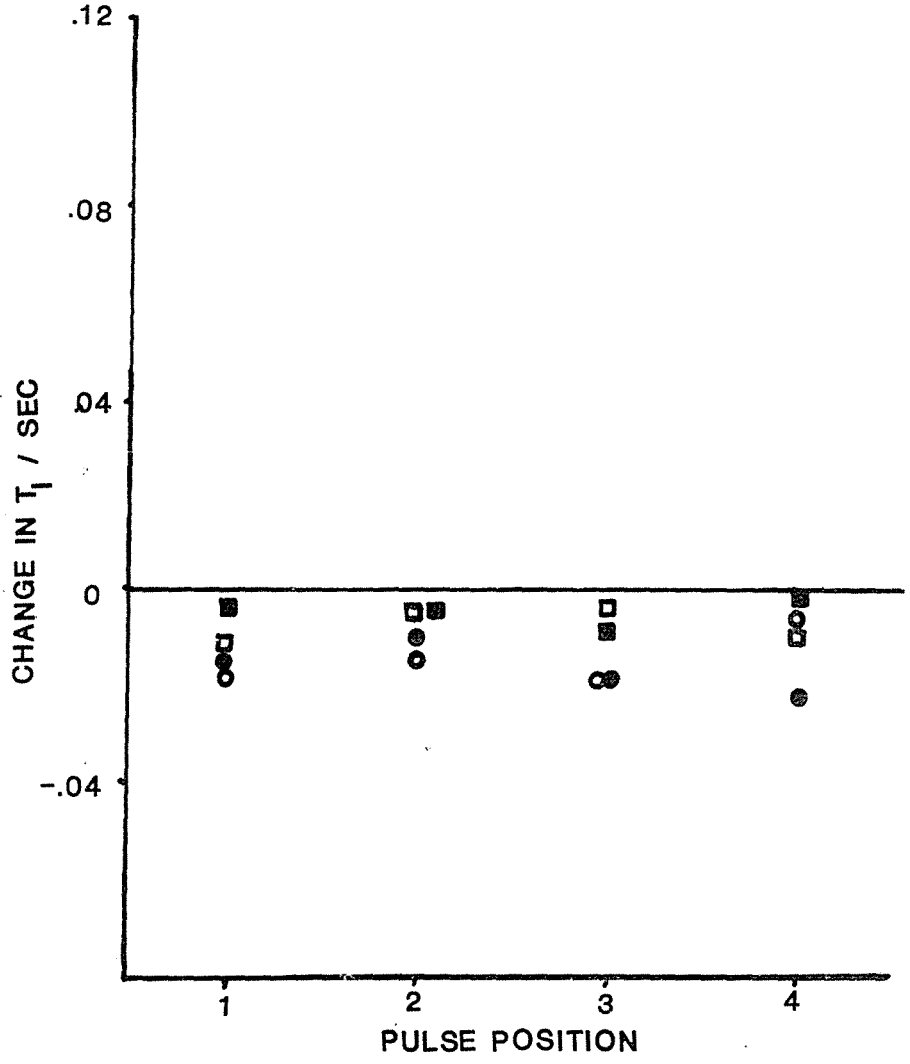


FIG 20. EFFECT OF ± PRESSURE PULSES ON THE SECOND T_1 AFTER THE PULSE.

○ + INTACT □ - INTACT

● + BLOCKED ■ - BLOCKED

shortening of tE was large but rarely occured the degree of significance of this shortening varies widely. This supports the concept that "memory" of R.A.R. activity continues to influence tI over several breaths. However the effect on tI was significantly less than the effect on tE and occured infrequently. This supports the requirement for a large R.A.R. activity to trigger the shortening of tI.

In subsequent breaths tI was not lengthened even after the large lengthening of negative pressure pulses. "Memory" of reduced P.S.R. activity has a shorter duration of activity than "memory" of R.A.R. activity. Also the response to changes in mechanics of breathing due to an inflation pulse did not continue into subsequent breaths. This further supports that "memory" is not merely a response to changes in the mechanics of breathing.

Augmented breaths

All runs which included augmented breaths were excluded from the main body of data. However a record was kept of the number augmented breaths which occured and in which breath they occured in. Many augmented breaths occured in the 1st breath after the pulse was applied , this was more frequent than occur spontaneously. Augmented breaths may be triggered by R.A.R. (Davies and Roumy 1982) which suggests that "memory" of R.A.R.

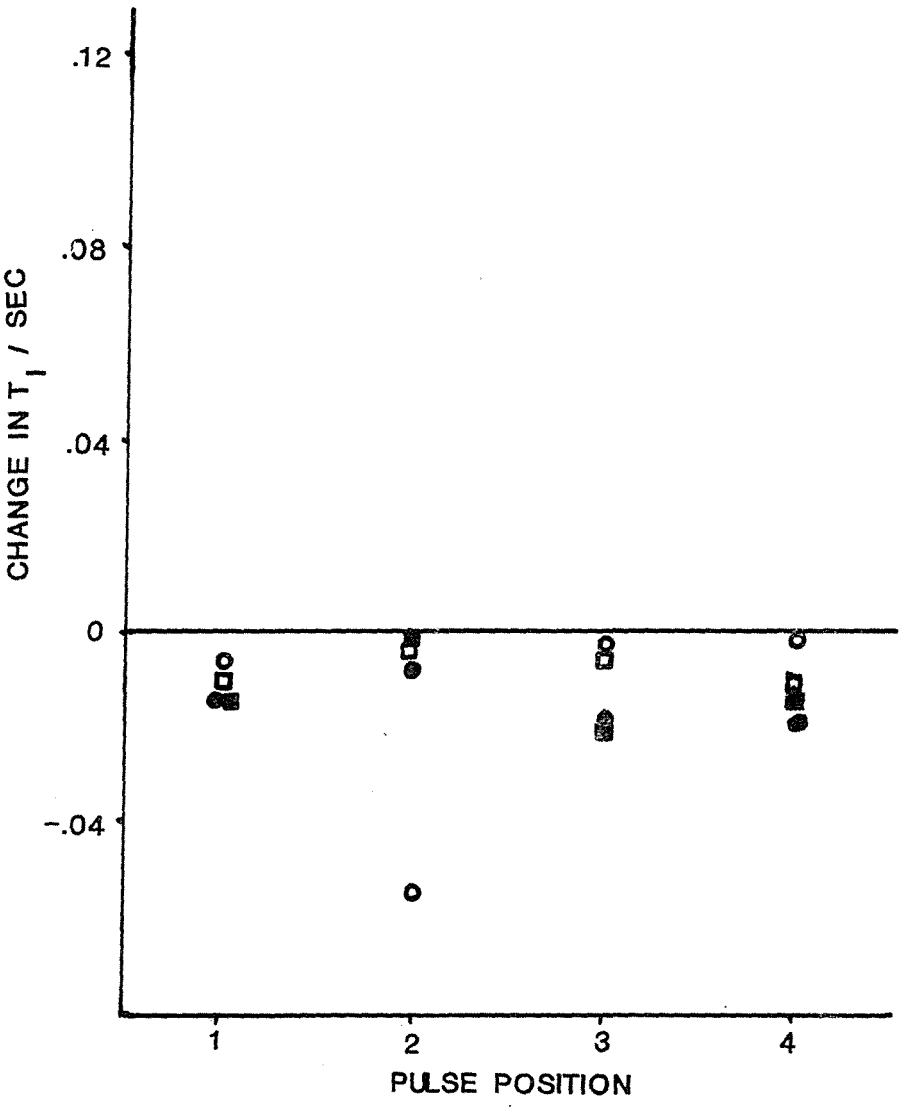


FIG 21. EFFECT OF ± PRESSURE PULSES ON THE THIRD T_1 AFTER THE PULSE.

○ + INTACT □ - INTACT

● + BLOCKED ■ - BLOCKED

activity must be sufficient in these breaths to trigger augmented breaths.

More augmented breaths occured after negative pressure pulses before P.S.R. block than with other treatments. More augmented breaths occured in the later positions. Negative pressure pulses stimulate R.A.R. strongly, particularly when applied later in expiration (Davies & Roumy 1982).

Augmented breaths did not occur as frequently after P.S.R. were blocked. After effective P.S.R. block negative pressure pulses shortened the following tI rather than lengthening as occurs before block. This supports the concept that an augmented breath is the summation of the inspiratory drive (Davies & Roumy 1982).

OTHER FACTORS INFLUENCING RESPONSE TO PRESSURE PULSES

Applying a pressure pulse to the lungs will cause changes to blood gas tensions, and the mechanics of breathing. The effects of these changes must be examined. As anaesthesia alters the function of many physiological systems, the role of anaesthesia in these preparation must also be examined.

Alteration of blood gas tensions

The absence of a method of rapidly following blood gas tensions prevented observation of any transient changes in these variables. Whether or not pulses of inflation and deflation caused alterations in blood gas tensions, the changes in arterial chemoreceptor activity that might result would not occur until one or two breaths later (Leitner, Pages, Puccinelli, & Dejours 1965) and could not have influenced the characteristic changes in duration of inspiration and expiration.

Increased respiratory frequency was seen 2-3 breaths after the pulse was applied. During this period of activity of arterial chemoreceptors was probably reduced by a transient increase in arterial pO2 (Biscoe and Purves 1967). However transient reduction in chemoreceptor activity by inhalation of a few breaths of oxygen decreases both tidal volume and respiratory frequency

(Leitner et. al. 1965) and cannot account for the acceleration reported here.

An increase of airways pCO2 due to a shortened tE may decrease the activity of chemorecptors, but this would be expected to lengthen the subsequent tE and tI rather than shorten. Positive pressure pulses which lengthen tE may decrease airways pCO2 increasing the activity of chemoreceptors, but this would be expected to lengthen the following tI rather than shorten. It is therefore unlikely that "memory" of R.A.R. is merely due to changes in airways or blood gases.

Influence of changes to the mechanics of breathing

Inflation and deflation pulses to the lungs will alter the mechanics of breathing. No paralysed animal preparations were used to test the influence of this. However although the mechanical changes from inflation pulses would differ from deflation pulses the effect on tE containing the pulse after P.S.R. block is shortening and the effect on the subsequent tI is generally lenthening. It is therefore unlikely that "memory" is merely a response to change in the mechanics of breathing.

The effects of anaesthesia

Surgical anaesthesia slows and deepens respiration to a degree dependant on the degree of anaesthesia (Lumb & Jones 1973). To minimise the

effects of anaesthesia on respiration, anaesthesia must be kept to a minimum while maintaining the state of anaesthesia. For adequate comparision of respiratory rates the degree of anaesthesia must remain constant.

Anaesthesia depresses the reticular activating system (RAS). As RAS facilitates CIA depression of RAS by anaesthesia depresses RAS. Anaesthetic agents depress the response of central chemorecptors to CO2. Volatile anaesthetic agents sensitize P.S.R., other agents may also affect receptor sensitivty (Lumb & Jones 1973).

During the experiment degree of anaesthesia was assessed between runs, if necessary anaesthetic was administered and respiration allowed to stabilize before proceeding with the next run. The order of position of application of the pulse was random and sign of pulse alternated. This regime helped reduce any bias of anaesthetic administration.

SO2 administration, vagotomy, and cold block of myelinated fibres all altered the respiratory pattern. The degree of anaesthesia was assessed on the initial stable pattern after treatment. These treatments necessarily came after the intact state. However the response to the treatment was noted the first runs after treatment making it unlikely that the response attributed to the treatment was due to the effects of anaesthesia.

THE RESPIRATORY EFFECTS OF P.D.G.

Injection of phenyldiguanide (P.D.G.) into the right atrium causes tachypnoea (Dawes & Mott 1950; Karczewski & Widdicombe 1969; Miserocchi, Trippenbach, Mazzarelli, Jaspar & Hazucha 1978). This was considered to be largely due to stimulation of type J receptors(Karczewski & Widdicombe 1969). J receptors have nonmyelinated fibres and are believed to be stimulated physiologically by pulmonary oedema (Paintal 1973a).

Non myelinated fibre endings are stimulated by a wide variety of chemical substances(Dawes & Mott 1950). The chemicals which stimulate non myelinated fibre endings are non specific and many different endings are stimulated by one chemical (Dawes & Mott 1950). Thus although P.D.G. stimulates type J receptors, other non myelinated fibres are also stimulated. P.D.G. is known to stimulate gastrointestinal receptors, epicardial receptors, carotid and aortic chemoreceptors (Paintal 1973a).

Doses of P.D.G. of less than 60 μg/kg stimulate J receptors but not gastrointestinal or aortic chemoreceptors (Anand & Paintal 1980). Injection of local anaesthetic into the pericardial sac blocks epicardial receptors (Anand & Paintal 1980). These receptors have vagal afferents thus their response to P.D.G. is abolished by vagotomy.

The carotid chemoreceptors should not be

stimulated by injection of P.D.G. into the right atrium (Anand & Paintal 1980). These receptors have effects mediated via the glossopharyngeal nerve (Widdicombe & Davies 1983) which are abolished when the nerve is cut.

As the effect of P.D.G. is reduced after vagotomy, P.D.G. is stimulating some vagally mediated receptor to increase the frequency of respiration and shorten expiration. This is not solely due to stimulation of epicardial receptors, as the effects are not totally blocked with block of these receptors with local anaesthetic. The response was seen with doses of much less than 60μg/kg so was not due to stimulation of gastrointestinal receptors or chemoreceptors of the aortic arch. The other non myelinated receptor with vagal fibres is the J receptor and is likely to be responsible for the major vagally mediated effects of P.D.G..

Vagotomy did not abolish the effects of P.D.G. in the rabbit. Also the effects were not abolished by cutting the glossopharyngeal nerve. Thus the carotid chemoreceptor is not the only non vagal receptor to cause tachypnoea on stimulation by P.D.G. This effect may be central. The effect of P.D.G. after section of the glossopharyngeal was less than the effect after vagotomy suggesting that some stimulation of the carotid chemoreceptors occurred.

From this it can be seen that in the rabbit P.D.G. stimulates many non myelinated receptors to cause tachypnoea. These include the type J receptor, the epicardial receptor, the carotid chemoreceptor and an unknown non vagal non glossopharyngeal receptor. The J receptor may be the most important receptor with vagal afferents to be stimulated. However in the rabbit the unknown non vagal receptor plays an important role in causing tachypnoea after P.D.G. stimulation.

These results suggest that intravenous injections of P.D.G. is not an adequate test of patency of J receptors in the rabbit. Glogowska & Widdicombe 1973 discuss the use of halothane as a J receptor stimulant, this may be a more appropriate method of testing patency of J receptors in the rabbit.

Response of different receptors to P.D.G.

The contribution made by various receptors to the tachypnoea of P.D.G. was assessed by injecting the drug sequentially

1. in the intact state,
2. after local anaesthetic was injected into the oericardial sac,
3. after vagotomy,
4. after cutting the glossopharyngeal nerve.

In the intact animal P.D.G. caused tachypneoa by shortening expiration but the duration of inspiration was not significantly altered. This is

the combined effect of many different receptors and the receptors do not necessarily act in the same manner.

Epicardial receptor block and cutting the vagus and glosssopharyngeal nerves alter frequency and duration of inspiration (tI) and expiration (tE). Thus comparison of effects must take into account these changes. Fig 22A shows the effects on duration of expiration before and after P.D.G. of these treatments. Fig 22B shows these effects on duration of inspiration. If the control before treatment is less than the effect of P.D.G. after treatment, the response to P.D.G. after treament is to lengthen tE and tI. This implies that the effect of the receptor, removed by the treatment, was to shorten tE/tI. If the control before treatment is more than the effect of P.D.G. after treatment, the response to P.D.G. after treatment is to shorten tE and tI. This implies that the effect of the receptor, removed by the treatment, was to lengthen tE/tI.

Local anaesthetic in the pericardial sac blocked epicardial receptors. The frequency increased due to a shortening of expiration despite a small lengthening of inspiration. This suggests that the effect of epicardial receptors on normal respiration is to reduce frequency due to a lengthening of expiration. Giving P.D.G. after epicardial block reduced the absolute frequency

response and reduced the shortening of expiration while increasing the shortening of inspiration compared to the response in the intact state.

The response of duration of expiration to P.D.G. after epicardial block was less than the control before block, thus block reduces shortening of expiration by P.D.G. However the response of duration of inspiration to P.D.G. after block was not significant (Fig 22). This supports the hypothesis of epicardial receptors lengthening expiration while not affecting inspiration.

After epicardial receptors were blocked and vagotomy performed the effect of stimulation of J receptors was abolished. This reduced the frequency response slightly despite further shortening of expiration. After the epicardial receptors were blocked and the vagi cut, carotid denervation removed the effects of the carotid chemoreceptors. Frequency was reduced despite a slight increase in shortening of inspiration. Response of duration of expiration was not significantly affected.

The response of duration of inspiration after both vagotomy and carotid denervation to P.D.G. was greater than the control before nerve section. This suggests that in the rabbit the tachypnoea from J receptor is due to a shortening of inspiration.

The response of duration of expiration to

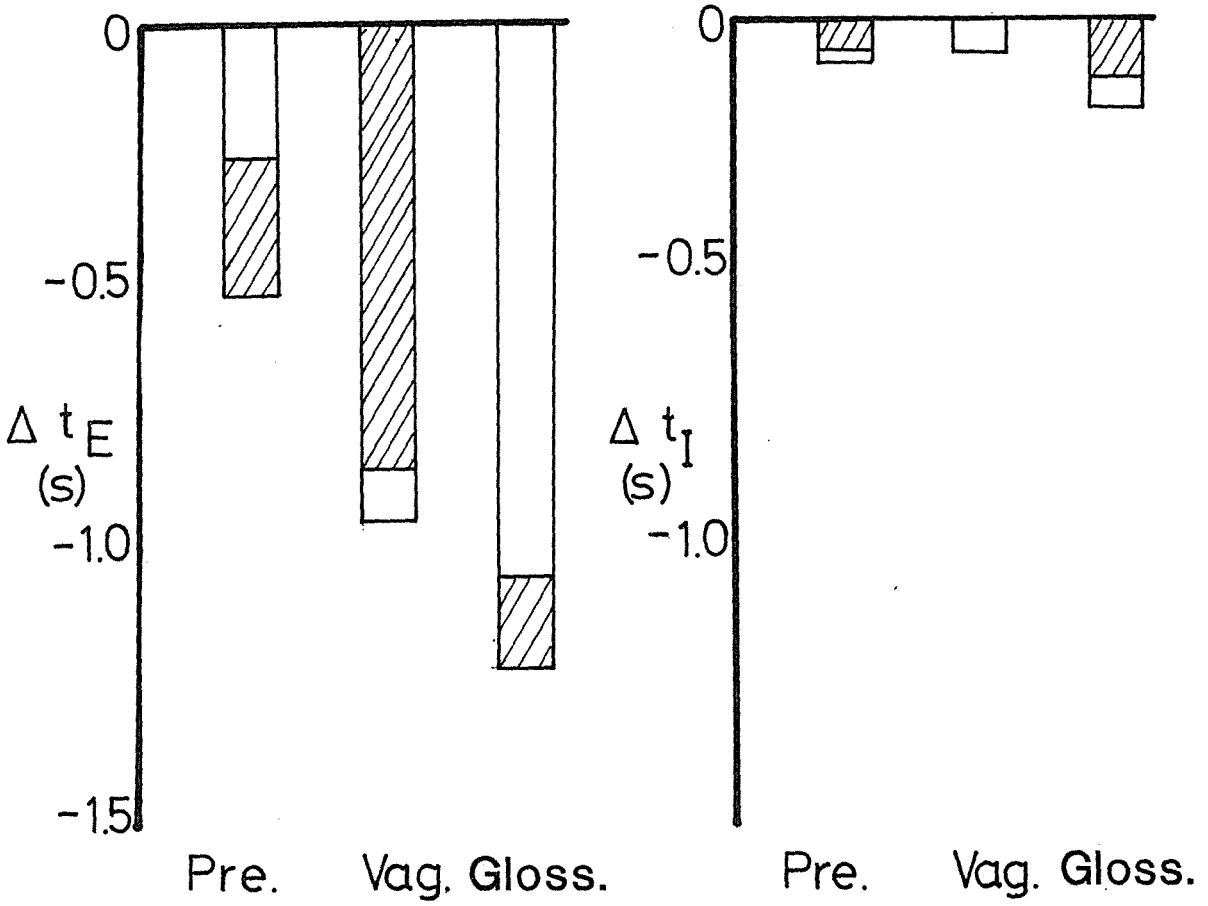


FIG 25. Effect of intravenous P.D.G. on rabbits before and after intrapericardial xylocaine.

Before After

P.D.G. after vagotomy was greater than the control before vagotomy. This suggests that in the rabbit J receptors also shorten expiration. The response of duration of expiration to P.D.G. after carotid denervation was less than the control before denervation. This suggests that expiration is lengthened by P.D.G. stimulation of carotid chemoreceptors.

After the glossopharyngeal nerves were cut the response to P.D.G. was due to an unknown receptor. The frequency response was increased with a shortening of inspiration and expiration. This suggests that the tachypnoea in the rabbit due to this unknown receptor is due to shortening of both expiration and inspiration.

From this it can be seen that the tachypnoea caused by P.D.G. stimulating non myelinated receptors may be due to shortening of inspiration or expiration. With the other phase of breathing quite differently. Different receptors may exert their effect differently. This may be due to differences in the manner they affect the "respiratory centres".

Species differences

Intravenous injections of P.D.G. in the cat causes tachypnoea due to shortening of both inspiration and expiration. Tachypnoea from P.D.G. in the rabbit is due mainly to shortening of expiration. In the cat apnoea occurs in the end expiratory position while in the rabbit it occurs in the end inspiratory position. In both species P.D.G. reduces the tidal volume before vagotomy but some workers report a small increase in tidal volume after vagotomy in the rabbit. In the rabbit P.D.G. increases the functional residual capacity. These differences may be due to one or more receptors responding differently in the different species. Or these differences may be due to an absence of one of these receptors in one of these species. There have been no reports in cats of the effects of P.D.G. not being completely abolished by vagotomy. This suggests that the receptor involved in the postvagotomy response to P.D.G. in the rabbit is not present in the cat.

After blocking epicardial receptors with local anaesthetic in the rabbit apnoea was not elicited by P.D.G. Anand & Paintal (1980) showed apnoea occurred in the cat after epicardial receptor block. This suggests the genesis of apnoea in reponse to P.D.G. differs in rabbits from cats.

Thus the differences in response to P.D.G. in cats and rabbits may be due to differences in

response of epicardial receptors and also due to the absence in cats of the unknown receptor responding to P.D.G. in the rabbit.

Time dependance of vagotomy

P.D.G. injected immediately after vagotomy did not cause tachypnoea. The effects of P.D.G. one hour after vagotomy were greater than the effects 15 minutes after vagotomy. This suggests that vagotomy causes a disturbance which wears off with time (fig 23).

This may explain the conflicts in the literature regarding the effects of P.D.G. after vagotomy in the rabbit. Those workers who found that the effects of P.D.G. was abolished by vagotomy (Dawes & Mott 1950; Davies et al 1978) may have been injecting P.D.G. immediately after vagotomy. Those workers who found that the effects of P.D.G. was reduced by vagotomy (Dawes & Fastier 1950; Mott & Widdicombe 1951; Karczewski & Widdicombe 1969b; Guz & Trenchard 1971; Miserocchi et al 1978) may have giving a longer recovery time after vagotomy but not long enough for the complete response.

If vagotomy creates a disturbance on the effects of P.D.G. it is likely that vagotomy may disturb other aspects of respiratory control in a similar manner. This suggests that the effects of vagotomy must be divided into two categories. The first being the immediate effects of vagotomy, those effects which occur in the first few minutes after the vagus is cut. The second category being the permanent effects after vagotomy, those

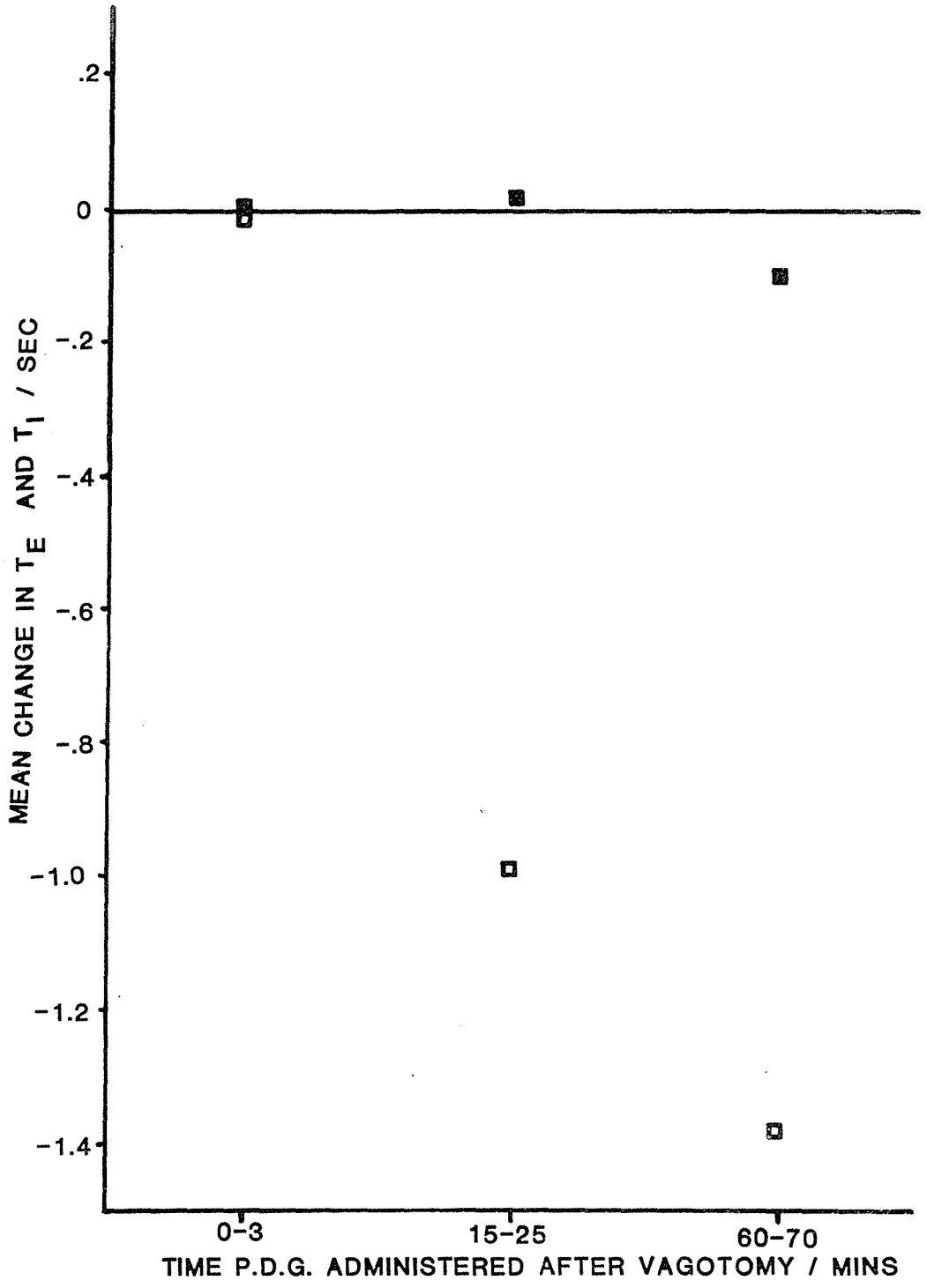


FIG 23. TIME DEPENDANCY OF THE EFFECT OF P.D.G. AFTER VAGOTOMY.

T_E T_I

effects which remain long after the vagus was cut.

This disturbance after vagotomy may occur as an immediate response of the "respiratory centres" to the lack of afferent information from the vagus, or to the synchronous discharge of damage potentials in all the afferent fibres of the vagi. If this is the case then a disturbance may occur when afferent information is cut off by other means eg.block of stretch receptors by SO2 inhalation or block of stretch and rapidly adapting receptors by cold or anodal block.

SUMMARY

PRESSURE PULSE SERIES

1. Positive pressure pulses applied early in expiration shorten tE containing the pulse, while those applied later lengthen it. After effective P.S.R. block positive pressure pulses in expiration always shorten tE containing the pulse. Negative pressure pulses in expiration always shorten tE containing the pulse.

2. Negative pressure pulses in expiration usually lengthen the following tI, after effective P.S.R. block tI was usually shortened. Positive pressure pulses in expiration do not significantly affect the following tI regardless of P.S.R. block.

3. Negative pressure pulses applied in expiration always shorten tE and occasionally shorten tI 2 or 3 breaths after the pulse regardless of sign of pulse or P.S.R. block.

4. R.A.R. appear to have a role in the deflation reflex and in reducing the inflation reflex. Some form of "memory" of R.A.R. activity is required to do this. Thus tE containing a pressure pulse is governed by a balance of P.S.R. and R.A.R. activity.

5. "Memory" of P.S.R. and R.A.R. activity may influence tE and tI of subsequent breaths.

6. The duration of P.S.R. "memory" is shortlived and does not exert influence beyond the 1st inspiration following the pulse. "Memory" of

R.A.R. has a longer duration of effect and significantly influences tE in the third breath after the pulse.

7. "Memory" of strong R.A.R. activity is required to shorten tI. Weaker R.A.R. activity will shorten tE.

8. "Memory of P.S.R. is "memory" of reduced activity. This may be in response to dampend reverberating neuron links, while "memory" of R.A.R. activity is probably in response to reberation of neuron links.

PDG SERIES

1. Vagotomy does not abolish response to PDG in the rabbit. Carotid denervation after vagotomy does not abolish this response.

2. No response to PDG can be elicited with injections immediately after vagotomy. A recovery period is required after vagotomy to obtain response to PDG.

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APPENDIX A

TREATMENT OF DATA OF PRESSURE PULSE SERIES

A computer programme to process data ideally should have the facility for retrieval of the data. Data can then be reprocessed without reentry. Installation problems of the computer prevented this being accomplished at the time of data processing.

Instead a programme was written to simplify and prepare the raw data for inspection and statistical analysis. This found the sums, means, sums of squares, variance and standard error of the mean for raw data, normalized data (see p47), and differences from control.

These values were found for each run of each treatment for the individual rabbits, all the rabbits pooled, and a pool of the eight rabbits which had the most effective PSR block. The programme had facility to estimate missing values based on the means of that run.

The programme

```
10 !////////////////////////////////
20 !/       STATS.BAS              /

30 !////////////////////////////////
40 PRINT
50 PRINT "This programme is designigned to prepare
```

```
    raw data for statistical analysis, values are
    normalized and corrected for treatment, means,
    standard deviation, and standard error of the
    means are found for individuals and pooled
    data."
60 PRINT
70 OPTION BASE 1
80 DIM A(15,12,13)
90 INPUT "INTACT/BLOCKED";A$
100 INPUT "+/-";D$
110 INPUT "POSITION NUMBER";E$
120 INPUT "INSPIRATION/EXPIRATION";B$
130 INPUT "ANY CORRECTIONS Y/N";Q$
140 IF Q$="Y" THEN GOTO 90
150 IF B$="INSPIRATION" THEN GOSUB 1540 ELSE GOSUB
     1690
160 GOSUB 560:!INPUT DATA////
170 PRINT %1
180  INPUT "ANY MORE RABBITS Y/N";Q$
190  IF Q$="Y"THEN GOTO 160
200  F$="ALL"
210  PRINT %1,"ALL RABBITS"
220  PRINT %1,"","MEANS","SUMS","SUM OF SQ","SQ OF
      SD","SEMS"
230  FOR K=C1 TO 12
240  SUMS=0:SUMOFSQ=0:A2=0
250  FOR J=1 TO 12
260   GOSUB 1330
270    NEXT J
```

```
280   GOSUB 1510
290  NEXT K
300   PRINT %1
310   PRINT %1,"8 RABBITS (1,3,5-10)"
320   PRINT %1,"","MEANS","SUMS","SUM OF SQ","SQ OF
       SD","SEMS"
330    FOR K=C1 TO 12
340     A2=0:SUMS=0:SUMOFSQ=0
350      J=1:GOSUB 1330
360      J=3:GOSUB 1330
370      FOR J=5 TO 10
380     GOSUB 1330
390      NEXT J
400     GOSUB 1510
410    NEXT K
420    PRINT %1
430  INPUT "ANY MORE DATA Y/N";Q$
440   IF Q$="Y" THEN GOTO 160
450  INPUT "DO YOU WANT TO PRINT MORE DATA Y/N";Q$
460   IF Q$="N" THEN GOTO 530
470    INPUT "RABBIT NUMBER";J
480     IF J>12 THEN GOTO 470
490    GOSUB 990
500   INPUT "DO YOU WANT ALL RABBITS Y/N";Q$
510    IF Q$="N" THEN GOTO 450
520   GOTO 200
530  GOSUB 2240
540  END
550 !////////////////////////////////INPUT
```

```
     DATA/////////
560 PRINT
570 INPUT "RABBIT NUMBER";J
580 PRINT "TO CORRECT RABBIT NUMBER ENTER RUNS=20 "
590  IF J>12 THEN PRINT "WRONG RABBIT":GOTO 570
600  IF J<1 THEN GOTO 570
610 INPUT "ENTER NUMBER OF RUNS";A1
620  A(J,1,13)=A1:!ALLOWS RUN NUMBER TO BE
       RETRIEVED FOR POOLING.
630   IF A1>0 THEN GOTO 680
640   PRINT
650   PRINT %1,"RABBIT "J" HAS NO DATA"
660   PRINT %1
670   PRINT %1:RETURN
680  PRINT %1
690  IF A1>15 THEN PRINT "TOO MANY RUNS":GOTO 560
700 FOR I=1 TO A1
710  PRINT "ENTER DATA,PRESS RETURN,CONTINUE ACROSS
      ROW"
720  PRINT "RUN NUMBER "I
730   FOR K=C1 TO 6
740    INPUT;A(I,K,J)
750   NEXT K
760  NEXT I
770 PRINT
780 PRINT "CHECKING DATA"
790 PRINT
800 FOR I=1 TO A1
810  PRINT "RUN NUMBER "I
```

```
820    FOR K=C1 TO 6
830     PRINT A(I,K,J)
840    NEXT K
850    INPUT "ANY CORRECTIONS Y/N";Q$
860    IF Q$="Y" THEN GOSUB 1850
870  NEXT I
880 INPUT "DO YOU WANT TO CHANGE RUNS Y/N";Q$
890 IF Q$<>"Y" THEN GOTO 1010
900  INPUT "NUMBER OF RUNS";N
910   IF N<=A1 THEN GOTO 990
920   N1=A1+1
930   FOR I=N1 TO N
940    PRINT "ENTER DATA, RUN "I
950     FOR K=C1 TO 6
960      INPUT ;A(I,K,J)

970     NEXT K
980   NEXT I
990   A1=N:A(J,1,13)=A1
1000  IF N>A1 THEN GOTO 780
1010 FOR I=1 TO A1
1020  IF A(I,6,J)=0 THEN MEANS=0:K=6:GOSUB
       2100:!MISSING DATA
1030  IF A(I,5,J)=0 THEN MEANS=0:K=5:GOSUB 2100
1040 !///NORMALIZING//
1050   U=2*A(I,4,J)-2*A(I,1,J)
1060   V=A(I,2,J)+A(I,3,J)
1070   W=V-2*A(I,1,J)
1080   IF C1=1 THEN A(I,7,J)=U/W ELSE
```

```
        A(I,7,J)=2*A(I,4,J)/V
1090    A(I,8,J)=2*A(I,5,J)/V
1100    A(I,9,J)=2*A(I,6,J)/V
1110 NEXT I
1120  PRINT %1,"RABBIT NUMBER "J
1130  PRINT %1,"NORMALIZED VALUES"
1140  PRINT %1,"","1ST","2ND","3RD"
1150 FOR I=1 TO A1
1160  PRINT %1,"RUN"I,A(I,7,J),A(I,8,J),A(I,9,J)
1170 NEXT I
1180  PRINT %1,"DIFFERENCES FROM CONTROLS"
1190  PRINT %1,"","1ST","2ND","3RD"
1200  FOR I=1 TO A1
1210   A(I,10,J)=A(I,4,J)-A(I,2,J)/2-A(I,3,J)/2
1220   A(I,11,J)=A(I,5,J)-A(I,2,J)/2-A(I,3,J)/2
1230   A(I,12,J)=A(I,6,J)-A(I,2,J)/2-A(I,3,J)/2
1240   PRINT
        %1,"RUN"I,A(I,10,J),A(I,11,J),A(I,12,J)
1250  NEXT I
1260  PRINT %1
1270 IF A1=1 THEN GOTO 2030!RABBIT HAS ONE RUN
1280 F$="ALONE"
1290 PRINT %1,"","MEANS","SUMS","SUM OF SQ","SQ OF
      SD","SEMS"
1300  FOR K=C1 TO 12
1310  SUMS=0:SUMOFSQ=0:A2=0
1320 !/////////////////////STATS////////
1330  A1=A(J,1,13)
1340  IF A1=0 THEN RETURN
```

```
1350  A2=A2+A1:!COMBINE RABBITS
1360   FOR I=1 TO A1
1370    SUMS=SUMS+A(I,K,J)
1380   NEXT I
1390  MEANS=SUMS/A2
1400   FOR I=1 TO A1
1410    IF K=5 AND A(I,5,J)=0 THEN GOTO 2100
1420    IF K=6 AND A(I,6,J)=0 THEN GOTO 2100
1430   NEXT I
1440   FOR I=1 TO A1
1450    SUMOFSQ=SUMOFSQ+A(I,K,J)*A(I,K,J)
1460   NEXT I
1470   SQOFSUM=SUMS*SUMS
1480   SQOFSD=ABS(SUMOFSQ-SQOFSUM/A2)/(A2-1)
1490   SEMS=SQR(SQOFSD/A2)
1500   IF F$="ALL" THEN RETURN
1510  PRINT
       %1,"#"K"=",MEANS,SUMS,SUMOFSQ,SQOFSD,SEMS
1520   IF F$="ALONE" THEN NEXT K
1530  RETURN
1540 !/////INSP PRINT OUT/////////
1550 PRINT %1,B$,D$" "A$,E$
1560 PRINT %1,"#2 IS 1ST CONTROL INSP DURATION"
1570 PRINT %1,"#3 IS 2ND CONTROL INSP DURATION"
1580 PRINT %1,"#4 IS 1ST INSP AFTER PULSE"
1590 PRINT %1,"#5 IS 2ND INSP AFTER PULSE"
1600 PRINT %1,"#6 IS 3RD INSP AFTER PULSE"
1610 PRINT %1,"#7 IS NORMALIZED 1ST INSP AFTER
      PULSE"
```

```
1620 PRINT %1,"#8 IS NORMALIZED 2ND INSP AFTER
     PULSE"
1630 PRINT %1,"#9 IS NORMALIZED 3RD INSP AFTER
     PULSE"
1640 PRINT %1,"#10 IS DIFFERENCE BETWEEN 1ST INSP
     AND CONTROLS"
1650 PRINT %1,"#11 IS DIFFERENCE BETWEEN 2ND INSP
     AND CONTROLS"
1660 PRINT %1,"#12 IS DIFFERENCE BETWEEN 3RD INSP
     AND CONTROLS"
1670 PRINT %1
1680 C1=2:RETURN:!REMOVES PULSE POSITION
1690 !/////EXP PRINT OUT/////////
1700 PRINT %1,B$,D$" "A$,E$
1710 PRINT %1,"#1 IS TIME OF PULSE APPLICATION"
1720 PRINT %1,"#2 IS 1ST CONTROL EXP DURATION"
1730 PRINT %1,"#3 IS 2ND CONTROL EXP DURATION"
1740 PRINT %1,"#4 IS EXP CONTAINING PULSE (1ST
     EXP)"
1750 PRINT %1,"#5 IS 2ND EXP AFTER PULSE"
1760 PRINT %1,"#6 IS 3RD EXP AFTER PULSE"
1770 PRINT %1,"#7 IS NORMALIZED AND CORRECTED EXP
     CONTAINING PULSE"
1780 PRINT %1,"#8 IS NORMALIZED 2ND EXP AFTER
     PULSE"
1790 PRINT %1,"#9 IS NORMALIZED 3RD EXP AFTER
     PULSE"
1800 PRINT %1,"#10 IS DIFFERNCE BETWEEN 1ST EXP AND
     CONTROLS"
```

```
1810 PRINT %1,"#11 IS DIFFERNCE BETWEEN 2ND EXP AND
      CONTROLS"
1820 PRINT %1,"#12 IS DIFFERNCE BETWEEN 3RD EXP AND
      CONTROLS"
1830 PRINT %1
1840 C1=1:RETURN:!LEAVES PULSE POSITION
1850 !////////ERROR CORRECTIONS//////
1860  PRINT
1870  Y=I
1880   INPUT "COLUMN NUMBER 1-6, (7=Whole Row,
        8=Global Change ";X
1890   IF X<7 THEN GOTO 1970
1900    IF X<8 THEN GOTO 1930
1910     INPUT "ENTER Run No.  ";Y
1920     GOTO 1880
1930    FOR K=C1 TO 6
1940     INPUT ;A(Y,K,J)
1950    NEXT K
1960   GOTO 2010
1970  INPUT "NEW DATA";Z
1980  PRINT
1990  IF C1=2 THEN X=X+1
2000  A(Y,X,J)=Z
2010  I=I-1
2020  RETURN
2030 PRINT %1,"RABBIT "J"HAS ONLY ONE RUN"
2040 PRINT %1,"CONTROLS
      ARE","1ST="A(1,2,J),"2ND="A(1,3,J)
2050 PRINT %1,"VALUES
```

```
      ARE","1ST="A(1,4,J),"2ND="A(1,5,J),"3RD="A(1,6,J)
2060 IF C1=1 THEN PRINT %1,"PULSE
      APPLICATION"A(1,1,J)
2070 PRINT %1
2080 PRINT %1
2090 GOTO 180
2100 !//////ESTIMATE MISSING VALUES//////
2110  A2=A1
2120  FOR L=1 TO A1
2130   IF A(L,K,J)=0 THEN A2=A1-1:GOTO 2190
2140   CON=A(L,2,J)+A(L,3,J)
2150   NORM=CON/A(L,K,J)
2160   DIFF=CON-A(L,K,J)
2170   EST2=CON/NORM+CON-DIFF
2180   MEANS=MEANS+EST2/(2*A2)
2190  NEXT L
2200  PRINT %1,"ESTIMATED VALUE FOR RABBIT "J" RUN
       "I" #"K " IS "MEANS
2210  A(I,K,J)=MEANS
2220  PRINT %1
2230  RETURN
2240 !/////////COUNTING NUMBERS SHORTENED, OR
      LENGTHENED///
2250 TU=0:TD=0:SU=0:SD=0:A2=0:A3=0
2260 PRINT %1,"","Norm tE'>1","Norm tE'<1","No. of
      runs"
2270  FOR J=1 TO 12
2280   NU=0:ND=0
2290   A1=A(J,1,13)
```

```
2300     IF A1=0 THEN PRINT %1,"RB
           "J,"0","0","0":GOTO 2390
2310      FOR I=1 TO A1
2320       IF A(I,7,J)>1 THEN NU=NU+1
2330       IF A(I,7,J)<1 THEN ND=ND+1
2340      NEXT I
2350     PRINT %1,"RB "J,NU,ND,A1
2360     A2=A2+A1:TU=TU+NU:TD=TD+ND
2370      IF J=2 OR J=4 OR J>10 THEN GOTO 2390
2380     A3=A3+A1:SU=SU+NU:SD=SD+ND
2390  NEXT J
2400 PRINT %1,"ALL RB",TU,TD,A2
2410 PRINT %1,"8 RB",SU,SD,A3
2420 RETURN
```

Statistical analysis

A paired t test was performed for each treatment and each pulse position to determine significance of any changes in tE and tI of three breaths after the pulse was applied.

A two way analysis of variance was performed as outlined below to determine significance difference of effect between pulse application or treatments. This test allowed for the differences of number of runs performed on each rabbit and the between rabbit variation of effect. These tests were performed using a spreadsheet programme (Calcstar, Micropro).

Fishers test of independence (see p134) was used to test significance of difference between

treatments of number of augmented breats occuring.

Two way analysis of variance

Pr= sum of tE/tI in all pos for rabbit r

Rp= sum of tE/tI in all rabbits for pos p

I = sum of tE/tI for each pos and rabbit

P = sum (Pr squared / runs of r) for all Rb

R = sum (Rp squared / runs of p) for all Rb

S = sum (I squared / runs of each pos and Rb)

E = total sum squared over total runs

degrees of freedom

pos dfP =(number of pos-1)*(number of rabbits-1)

rb dfR =(number of rabbits-1)

error dfE =number runs-(number pos * number rb)

standard error of means

pos SEMP =(P-E)/dfP

rb SEMR =(R-E)/dfR

error SEME =(total sums-S)/dfE

F values

pos FP =SEMP/SEME with dfP and dfE degrees freedom

rb FR =SEMR/SEME with dfR and dfE degrees freedom.

This tested significance of variation between positions (FP) and between rabbits (FR). Similar tests were used to test significance of variation between treatments.

Sign Test

The sign test was used to show the

significance of occasional large shortening of tI in the second and third breaths after the pulse was applied.

STATISTICAL ANALYSIS OF PDG SERIES

Significance of the difference of frequency, tI, and tE between before and after PDG administration was tested by the paired t test.

The significance of the difference of change in frequency, change in tE, and change in tI between the treatments was tested by one way analysis of variance.

Fishers test of independance was used to test the significance of the lack of apnoea ater injection of xylocaine into the pericardial sac.

Fishers test of independance

pre xylo	breaths(Bp)	apnoea(Ap)	no apnoea(Np)
post xylo	breaths(Bx)	apnoea(Ax)	no apnoea(Nx)
total	breaths(Bt)	apnoea(At)	no apnoea(Nt)

The probability of apnoea occuring independantly of xylocaine application is:

$$p=\frac{At!Nt!Bp!Bx!}{Bt!} * \frac{1}{Ap!Ax!Np!Nx!}$$

Three treatments were performed before and after xylocaine (intact, vagotomy, glossopharyngeal nerve section). The sum of the probability of independance for each treatment is the probability of independance for all treatments.

The validity of these tests was checked by Professor Munford.