STUDIES ON REFLEX CONTROL OF BREATHING IN PIGS AND BABOONS

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Abstract. In 8 pigs and 4 baboons, spontaneously breathing, anaesthetised with halothane, Hering-Breuer reflex was tested by means of a total obstruction of the airway preventing either inspiration or expiration. Subsequently animals were paralysed and maintained on phrenic nerve driven servo-respirator. The response of phrenic motoneurone output to various degree of lung inflation, introduced for one breath only, was then carefully studied. This was achieved by varying the gain of servo-respirator. Additionally in baboons, identical series of gain manoeuvres was performed against a background of different levels of the initial gain setting. Changes in both inspiratory time and peak amplitude of phrenic signal were monoexponentially dependent on gain of servo-respirator and linearly dependent on tidal volume (all negatively correlated). The relationship between inspiratory time $T_I$ and subsequent expiratory duration $T_E$ existed only within a range of growing $T_I$. Vagal positive feedback phenomenon was apparent in pigs and negligible in baboons. It is postulated that inspiratory cut-off mechanism terminates inspiration when excitatory function are outbalanced by their integral.

INTRODUCTION

Bartoli et al. (2) were the first to demonstrate with nonocclusive methods the quantitative dependence of phrenic motoneurone output on the degree of lung inflation in the dog. Although this dependence
was undoubtedly of a negative feedback character, they provided, however, a clear cut evidence for the existence of a dynamic positive feedback phenomenon originating from pulmonary vagal receptors.

The nature of the existing vagal feedback mechanism in the reflex control of breathing has created a great deal of controversy and it is being still disputed. Two most likely hypotheses should be considered: (i) the excitatory component of the vagal feedback is not easily detectable, (ii) the species difference underlies discrepancy in experimental data.

In an attempt to answer these questions we have decided to follow part of the experimental procedure of Bartoli et al. (2) on rather less common species in the field, namely pigs and monkeys.

METHODS

Experiments were performed with 8 healthy piglets weighing 16–26 kg and 4 baboons weighing 7–26 kg. Baboons were not quite healthy since they were taken over from surgery department where they were subjected to chronic experiments on neurotrophic ulcer of lower limbs. All animals were initially anaesthetised; pigs with Nembutal (ABBOTT), baboons with Ketamine Hydrochloride (PARKE–DAVIS) in order to expose the pharynx and to introduce through a cutting on thyroid membrane (membrana thyrohyoidea) an endotracheal tube (size 6.5–9). To the other end of this tube pneumotachograph head (Fleisch No 1) was connected so that tidal volume could be measured with GODART pneumotachograph. Anaesthesia was maintained with 0.5–2% halothane in air, always adjusted to the lowest concentration that could keep animal still while surgical procedures were carried out. Right marginal ear vein (pigs) or right elbow vein (baboons) were cannulated for further injections. Left femoral artery was cannulated and polyvinyl catheter advanced some 20 cm for systemic blood pressure monitoring and blood samples collection. Both cervical vagi and C₄ or C₅ root of right phrenic nerve were exposed and dissected from surrounding tissues. Vagi were then looped with cotton bands and immersed in saline. The superficial sheath from the cut central end of the phrenic nerve root was removed and its bioelectrical activity amplified with classical methods (5A22N preamplifier—TEKTRONIX) to the level suitable for further processing in the input circuits of the servo–respirator (MEDIPAN). The processing was of a leaky integration character and the resulting analog signal was closely resembling the transpulmonary pressure shape (for details see Bartoli et al. (2)). This signal called “integrated” phrenic signal was continuously monitored and, when required, it could drive bellows
of the servo-respirator so that the positive pressure ventilation could be achieved.

In this instance endotracheal tube with pneumotachograph head had to be connected to the respirator while halothane supply was diverted into its inlet port. Airway pressure was monitored with pressure transducer (UP-1, PYE ETHER).

Arterial blood gases were measured at appropriate stages of experiment using RADIOMETER BMS-3 assembly. In experiments on baboons end-tidal CO₂ was also recorded using infrared analyser (MIJN-HARDT) ¹.

After reaching steady state with respect to blood gas tensions, depth of anaesthesia and ventilation, a series of tests of Hering-Breuer reflex was performed by clamping the endotracheal tube either at the peak of inspiration or during late expiratory pause. It was repeated a number of times (from three to six) at intervals of about 1 min and the resulting prolongation of expiratory pause or inspiratory time, that respectively followed particular occlusions, were measured.

The next experimental step was to establish an assisted ventilation. First, the gain of phrenic nerve driven servo-respirator was adjusted until the bellows displacement (monitored with a built-in transducer) closely corresponded to tidal volume of the animal. The respirator was then connected to animal's airway. After a while, if necessary, small gain corrections were done to restore previous level of "integrated" phrenic nerve signal. The final gain setting was read from a dial and from that moment on it was regarded as a control, i.e., 100% gain value. Subsequently animals were paralysed by i.v. administration of gallamine triethiodide (GERMED) 100 mg, so that their breathing was entirely dependent on the phrenic nerve-driven servo-respirator. Usually about 10 min later we proceeded to the basic stage of the experiment. A series of manoeuvres of various magnitude step changes in gain of the servo system was performed. Each change in gain setting was introduced for one breath only and repeated a number of times (six in average) with at least five control breaths separating subsequent manoeuvres. There was usually four gain reductions, i.e., ~70%, ~50%, ~30% and 0% (omission of one breath) and one or two increases up to ~160% depending on whether the particular gain setting change could be repeated a number of times with no augmented breath type of response (see 2). On baboons the number of gain changes to be introduced was reduced to G = 0%, ~50% and highest possible over 100% but they were

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repeated against a background of various levels of the initial gain setting. After completing a series of manoeuvres at \( G = 100\% \) a new control gain volume (denoted as \( G_0 \)) below and above \( 100\% \) were fixed and maintained for at least 10 min before and identical series of gain manoeuvres was performed. Arterial blood samples were collected just before and after each set of manoeuvres.

In both groups of animals the extreme (0\% and sometimes \( > 100\% \)) gain manoeuvres were performed after unilateral and bilateral vagotomy (on baboons at \( G_0 = 100\% \)).

Variables were recorded either from an oscilloscope (TEKTRONIX 5103ND13) on 70 mm photographic paper with camera (OK-3, MEDIPAN) or directly with six channel pen recorder (WATANABE).

RESULTS

General observations

The group of eight pigs maintained an average \( \text{PaCO}_2 \) of \( 47.9 \pm 5.6 \text{ mm Hg} \) at average \( \text{pH} \) of \( 7.389 \pm 0.065 \). In the group of four baboons these values were respectively \( 40.4 \pm 4.3 \text{ mm Hg} \) and \( 7.323 \pm 0.08 \). In pigs the pattern of breathing was extremely stable; in baboons there were some fluctuations with respect to phrenic nerve activity and tidal volume.

The strength of Hering–Breuer reflex was expressed in terms of coefficients \( i \) and \( e \). Coefficient \( i \) was determined as the ratio of the duration of the silent period which followed airway occlusion at the peak of inspiration to the duration of control expiratory pause. Average value of \( i \) found for pigs was \( 6.23 \pm 3.7 \) whereas for baboons it was \( 2.7 \pm 0.35 \). Coefficient \( e \) was determined as the ratio of the duration of the inspiratory effort following airway occlusion prior to the onset of inspiration to the duration of the preceding control inspiration. Average values of \( e \) were \( 1.85 \pm 0.4 \) for pigs and \( 1.54 \pm 0.15 \) for baboons.

Control values of respiratory variables at the basic stages of experiment were also averaged for both groups of animals and they are displayed in Table I.

In both groups of animals an augmented breath type of response was occasionally observed for \( G > 100\% \) manoeuvres. The incidence of this type of response grew for bigger gain increase so the gain values around 150–160\% were the highest we could study.

The other common feature of both groups studied was the existence of a very strong relationship between inspiratory time \( T_I \) and the subsequent expiratory duration \( T_E \) (Fig. 1). In the pigs this dependence was
Control values of respiratory variables at the basic stages of experiments for both groups of animals

<table>
<thead>
<tr>
<th>Number of animals</th>
<th>Spontaneous breathing</th>
<th>Paralysed, ventilated by means of phrenic driven respirator</th>
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<tr>
<td></td>
<td>$f$ (min$^{-1}$)</td>
<td>$V_T$ (ml)</td>
</tr>
<tr>
<td>Pig</td>
<td>8</td>
<td>25</td>
</tr>
<tr>
<td>SD±</td>
<td>9</td>
<td>38.8</td>
</tr>
<tr>
<td>Baboon</td>
<td>4</td>
<td>22.4</td>
</tr>
<tr>
<td>SD±</td>
<td>3.9</td>
<td>55.9</td>
</tr>
</tbody>
</table>

Fig. 1. The relationship between inspiratory time $T_I$ and the subsequent expiratory duration $T_E$ when $T_I$ varied in response to single breath gain manoeuvres. Vagi intact. Grouped data for pigs (dots) and baboons (squares).

clearly of a curvilinear character whereas in the baboons better correlation was obtained when a straight line was fitted, although lower number of experiments might be responsible here for bigger scatter and, in consequence, certain distortion of the real dependence. Nevertheless it is worth mentioning that in both species studied the $T_I$–$T_E$ relationship exists practically only for growing $T_I$. 
Gain manoeuvres in pigs

Almost all basic observations and results obtained in pigs fully confirmed those found in dogs by Bartoli et al. (2). Figure 2 displays records taken from one pig at different stages of an experiment. On the right
hand side of each record (except the last one) superimposition of the phrenic nerve signals corresponding to the control and gain change states demonstrates the magnitude of both positive (dynamic change of slopes) and negative (peak levels) feedback type of the response. This could also be seen after unilateral vagotomy and practically no response was observed after both vagi were cut (see Table II). All “integrated”

<table>
<thead>
<tr>
<th>State</th>
<th>Parameters</th>
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<tbody>
<tr>
<td></td>
<td>$G$</td>
</tr>
<tr>
<td>Vagi intact</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>32</td>
</tr>
<tr>
<td></td>
<td>48</td>
</tr>
<tr>
<td></td>
<td>71</td>
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<tr>
<td></td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>143</td>
</tr>
<tr>
<td>Unilateral vagotomy</td>
<td>100</td>
</tr>
<tr>
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<td>0</td>
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<td></td>
<td>100</td>
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<td></td>
<td>0</td>
</tr>
</tbody>
</table>

phrenic signals corresponding to $G = 0$, $\sim 50\%$ and $\geq 100\%$ manoeuvres were traced. For overall number of 160 tracings only in five cases no change in the ascending slope of the phrenic signal was found in comparison to the control slope. After initial unchangeable period ($320 \pm 150$ ms on average) the slope of the “integrated” phrenic signal always changed in the same direction as the change of gain (see Fig. 2). In consequence, $T_I$ always changed more than PHR. The magnitude of the average responses obtained from eight pigs concerning both peak phrenic activity level (PHR) and inspiratory time duration ($T_I$) reached

Fig. 2. Pig 14. Responses of “integrated” phrenic activity (INT PHR) to gain manoeuvres at different stages of experiment. Percentual values of gain settings reduced or increased from control value of 100% are indicated between arrows. On each record from top to bottom: BP, blood pressure; INT PHR, “integrated” phrenic signal; $P_{TR}$ airway pressure; $V_T$, inspired tidal volume. Records A, B, C, vagi intact; D, E, after unilateral vagotomy; F, both vagi cut. Superimposition of phrenic signals and corresponding tidal volumes is displayed on the right hand side of each record.
higher levels than in those observed on dogs (see 2) but the mathematical form of their dependence on gain (G) was the same, i.e.:

\[
\text{PHR}^{\%} = 150 \exp (-0.0038 \text{G}^{\%}) \quad (r = 0.99),
\]

\[
T_1^{\%} = 253 \exp (-0.0086 \text{G}^{\%}) \quad (r = 0.989).
\]

The gain manoeuvres could be also expressed in terms of corresponding tidal volume (V_T) changes, thus the regression lines, fitted to experimental data were of the form:

\[
\text{PHR}^{\%} = -0.5 \quad V_T^{\%} + 153 \quad (r = 0.988),
\]

\[
T_1^{\%} = -1.5 \quad V_T^{\%} + 258 \quad (r = 0.99).
\]

Equation 3 describes in a mathematical form the negative feedback phenomenon.

When instead of terminal values of PHR vs. V_T their dynamic changes in time \(\Delta\text{PHR}/\Delta t\) vs. \(\Delta V_T/\Delta t\) were plotted the following equation was found:

\[
\frac{\Delta\text{PHR}^{\%}}{\Delta t} = 0.52 \quad \frac{\Delta V_T^{\%}}{\Delta t} + 64 \quad (r = 0.99).
\]

Thus the existence of a dynamic positive feedback can be fully demonstrated and, furthermore, the strength of this mechanism is of the order of the negative one (virtually the same steepness of lines).

**Gain manoeuvres in baboons**

Initial gain setting (G_0) had a strong influence on the level of the driving signal, i.e., the “integrated” phrenic signal (PHR). For steady states it was a hyperbolic relationship (Fig. 3). Steady states were reached within relatively short period of time (Fig. 4) after every permanent change to any new level of G_0. For G_0 \(\leq 100\%\) region very efficient regulation of pHCO\_2 and PACO\_2 was observed, whereas in the region of G_0 > 100\% gradual but moderate hypocapnia was developing. There was a little difference in the magnitude of PHR and T_1 responses to a single breath gain manoeuvres within G_0 \(\leq 100\%\) region, whereas in the region of G_0 > 100\% the magnitude of these responses was growing with the growth of G_0 (Fig. 5). For these reasons all experiment-
al data obtained from an almost identical series of a single breath gain manoeuvres performed against a background of various levels of the

![Graph of PHR% vs Go%](image)

**Fig. 3.** The influence of the initial gain setting (Go) on the peak level of the integrated phrenic activity (PHR) in four baboons. Regression line fitted:

\[
\text{PHR}\% = \frac{9600}{G_0\%} + 5.28 \quad (r = 0.98)
\]

initial gain setting (Go) were classified into two groups, namely: Go \(\leq 100\%\) — normocapnic and Go > 100\% — moderately hypocapnic.

Averaged results could be expressed in the following forms:

**group Go \(\leq 100\%\):**

\[
\text{PHR}\% = 144 \exp (-0.0035 \text{G}\%) \quad (r = 0.99),
\]

\[
T_1\% = 166 \exp (-0.0045 \text{G}\%) \quad (r = 0.99),
\]

or

\[
\text{PHR}\% = -0.45 V_T\% + 146 \quad (r = 0.99),
\]

\[
T_1\% = -0.63 V_T\% + 169 \quad (r = 0.99),
\]
Fig. 4. Baboon 2. The examples of responses to sudden increase (A) and decrease (B) of the initial gain setting ($G_0$). From top to bottom the traces are: $P_{TR}$, tracheal pressure; PHR INT, "integrated" phrenic activity; $ETCO_2$, end tidal $CO_2$; $VT$, tidal volume; BP, blood pressure.
Fig. 5. Baboon 3. Responses to omission of one breath (G = 0 manoeuvre) performed against a background of different initial gain settings (G₀). The traces are as in Fig. 4.
and group $G_0 \geq 100\%$: 

$$\text{PHR}^\circ = 170 \exp \left( -0.0051 \frac{G}{\%} \right) \quad (r = 0.99), \quad [10]$$

$$T_1^\circ = 186 \exp \left( -0.0057 \frac{G}{\%} \right) \quad (r = 0.98), \quad [11]$$

or

$$\text{PHR}^\circ = -0.74 \, V_T^\circ + 175 \quad (r = 0.99), \quad [12]$$

$$T_1^\circ = -0.91 \, V_T^\circ + 194 \quad (r = 0.99). \quad [13]$$

The striking difference between these results and those obtained in pigs and earlier in dogs (2) is that in baboons the responses of PHR and $T_1$ do not significantly differ in magnitude. Moreover the difference that matters exists only between the magnitude of $T_1$ responses since the PHR responses are practically the same (compare equations 1 and 6; 3 and 8) for pigs and baboons. In consequence no dynamic change in the slope of the “integrated” phrenic signal could be found when the appropriate signals were traced. Actually for all $G = 0$ manoeuvres studied only in about 20% of cases the tracings revealed small deviations of the slopes but even these failed tests for significance. The general impression was that on the contrary to pigs and dogs, in the baboons the regulation of the ascending slope of the phrenic signal during control conditions was very poor. Consecutive phrenic signals corresponding to control conditions ($G = 100\%$) always were showing considerable scatter of their slopes. Also mathematical test for the existence of a dynamic positive feedback (analogous to that applied for pigs — see equation 5) practically failed to detect such a phenomenon:

$$\frac{\Delta \text{PHR}^\circ}{\Delta t} = 0.09 \, \frac{\Delta V_T^\circ}{\Delta t} + 84.5 \quad (r = 0.92). \quad [14]$$

After unilateral vagotomy $G = 0$ manoeuvres performed at $G_0 = 100\%$ gave average responses of PHR and $T_1$ which in magnitude were rather tending to obey the equations found for $G_0 > 100\%$ (i.e., moderately hypocapnic group) in a proportion resembling that observed in pigs (see Table II). Bilateral vagotomy abolished all responses.

In order to verify the validity of the mathematical tests for the estimation of the strength of both negative (equations 3, 8 and 12) and positive vagal feedback (equations 5 and 14) we processed the experimental data obtained in dogs by Bartoli et al. (2).
The following was found:

\[
\text{PHR}^\% = -0.31 \, V_T^\% + 134 \quad (r = 0.979) \quad [15]
\]

and

\[
\frac{\Delta \text{PHR}^\%}{\Delta t} = 0.33 \frac{\Delta V_T^\%}{\Delta t} + 40 \quad (r = 0.99). \quad [16]
\]

Although lower in absolute values, as compared to those for pigs, the steepness of both lines seems to be identical, thus also in dogs both types of vagal feedback seem to be equally strong.

All discussed expressions which deal with either negative or positive vagal feedback are graphically demonstrated on Fig. 6.

**DISCUSSION**

The importance of feedback mechanism in the regulation of breathing has been demonstrated for the first time by Breuer (4) and Hering (15) but at present one cannot expect much more from experiments in which the sustained inflations or deflations are being performed, for there is no physiological situation where either the state of distention of the lungs is associated with complete inhibition of the inspiratory activity or the lung volume would fall below EEV during enhancement of this activity.

In the course of physiological breathing the inspiratory activity as
a motor function is a primal factor that is being closely followed by changes in the physical state of the lungs which in turn exerts certain influence on the pattern of inspiration and this sequence of natural events should be unconditionally observed during an experiment. Classical experimental procedure of Breuer and Hering demonstrates, therefore, only the existence of a negative feedback phenomenon and can at most provide some information about the strength of this mechanism.

Other classical experiments of Gad (9) and Head (14) supported later by work of Wyss (32) who demonstrated that threshold electrical stimulation of the rapidly conducting fibres of the afferent vagus at low pulse frequencies enhanced inspiratory activity brought into the matter qualitatively new aspect. Their observations supplemented by evidence of Paintal (24) for “frequency dependence” of vagal cold block (explaining mechanism of Head’s paradoxical reflex) should be considered as a ponderable evidence for the existence of the vagal positive feedback phenomenon. Bartoli et al. (2) fully confirmed this conclusion. Bystrzycka et al. (5) have discussed possibility of interaction of the excitatory and inhibitory mechanisms of the vagal feedback.

The phenomenon of a positive feedback can not be identified only with its extreme manifestation, i.e., the uncontrolled burst up to potentially possible limits. The continuous predominance of a negative feedback in every regulatory system prevents excessive fluctuations at its output. The introduction of a certain component of a positive feedback (generally with much shorter time constant of operation) into that system enables precise modulation of the output function according to the nature of the variables at the input. As the symptoms of such a positive feedback should be recognized all temporarily recorded changes in an output function that reflect the direction of changes of a variable at the input. Thus, when the output function is being triggered, prolonged, enhanced or changes its derivative in response to the appropriate states of the input function, most likely the presence of a positive feedback component is responsible for all these symptoms.

It should be stressed, however, that still holds its strong position the view of Clark and von Euler (6) that the exclusive effect of the vagal afferentation from pulmonary stretch receptors is to cut off inspiration after reaching certain preset threshold. They based their concept of the regulation of depth and rate of breathing on two graphs, namely hyperbolically shaped correlation between $T_1$ and $V_T$ and linear relationship between $T_1$ and $T_E$, both depending upon integrity of the vagi. These plots have been obtained with experimental technique combining pulse and step inflations with rebreathing, thus mainly the region of decreasing $T_1$ was covered which is disputable since the loaded breathing
as a much more appropriate technique for studies on reflex control of breathing reveals for $T_1$ tendency to grow. Furthermore hypercapnia as a stimulus activating various peripheral and central feedback systems should be excluded from experimental techniques aiming at studies on reflex control of breathing. Besides obvious interference of hypercapnia at the level of respiratory “centre” the vagal feedback is also being affected through direct action of $CO_2$ on pulmonary stretch receptors. Experimental evidence of Schoener and Frankel (29) and Mustafa and Purves (22) recently supported by Bradley et al. (3), Trenchard et al. (31) and Huszczuk et al. (17) points to $CO_2$ sensitivity of pulmonary stretch receptors as a very important factor modulating vagal feedback from the lungs.

In the light of the foregoing the plots of Clark and v. Euler which underlie their theory represent only a graphical illustration of a general study on very complex links between some respiratory variables. Their time dependent volume threshold for inspiratory cut off has been also observed with only some shift to the right and upwards after vagotomy by many authors, e.g., Karczewski et al. (19), Gautier (10) and recently by Feldman and Gautier (8) and Glebovsky and Gizatullina (11). Since the key argument for discontinuity of the vagal feedback mechanism, i.e., the invariability of the “integrated” phrenic activity slope appears to be also disputable (see Discussion in Bartoli et al. (2) it seems, therefore, justified to conclude that the problem of vagal control of breathing still remains open.

The relevance of the experimental data presented in this paper depends on how strictly the methodological requirements for this kind of study have been actually fulfilled. A broad assessment of the experimental model adopted here has been undertaken by Bartoli et al. (2). Cardiopulmonary bypass technique has been deliberately dropped to avoid excessive stimulation of pulmonary stretch receptors by very low end-tidal $CO_2$ (generally below 1%) which is invariably associated with this technique. Single breath gain manoeuvres could only a little affect the gas exchange. The lung compliance, although not measured, was rather kept within a range typical for spontaneous breathing since on phrenic driven servo-respirator the incidence of deep breaths was always observed.

The use of halothane, however, requires some consideration. Recent work of Huszczuk et al. (17) quantitated halothane vapour effect on pulmonary stretch receptor endings. They found that global end expiratory stretch receptor activity in the rabbit and baboon (not published) was rapidly dropping by 35% in response to inhalation of 2% halothane vapour thus qualitatively resembling the effect of airway hypercapnia.
Peak inspiratory activity, however, changed only slightly. In the present study halothane concentration ranged between 0.5 and 2% thus the vagal feedback corresponding to EEV level could be to some extent affected and in consequence, according to Bartoli et al. (1), the respiratory rate was probably higher as a result of a shortened $T_E$.

The choice of the experimental animals appeared to be rather fortunate for this kind of study since classical tests for the strength of Hering-Breuer reflex revealed considerable species difference with respect to the values of the coefficient $i$. Reduction in respiratory rate and increase of tidal volume following vagotomy (see Table I) were also greater in pigs, thus in these animals the contribution of the vagal feedback to the regulation of respiratory pattern seems to be more pronounced. The values of the coefficients $e$ expressing the magnitude of $T_I$ increase in response to total obstruction of the airway for one inspiration were smaller than corresponding values of $T_I$ following $G = 0$ manoeuvre (see values of intercepts in the equations for $T_I$). Again for pigs this difference was greater.

This observation proves that the loaded breathing technique is not quite adequate for the analysis of vagal control mechanisms. Besides already documented by Richardson et al. (26) and Ryba (28) sensitivity of pulmonary stretch receptors to the negative pressure in the airway at constant EEV the extravagal reflexes tend also to shorten $T_I$ during airway occlusion as it has recently been described in vagotomised cats by Shimaraeva (30).

In both groups of animals strong $T_I - T_E$ relationship was observed for the region of growing $T_I$ only. Inspiratory time has been varied by altering the amount of the phasic vagal feedback as a factor concomitant only with inspiration thus not affecting tonic vagal feedback. Transient and very small degree of hypercapnia that could have developed might at most have an opposite effect on the subsequent expiratory duration, it seems therefore that there is a genuine centrally determined dependence of the expiratory pause on the inspiratory duration at least as strong as it has been found in this study. Similar techniques applied in dogs by Phillipson (25) and Bartoli et al. (2) did not however detect such a dependence. Short latency chemical feedback which seems to be more operative in dogs may be responsible for this discrepancy (see Discussion in Bartoli et al. (2). The lack of $T_I - T_E$ dependence for $T_I$ shorter or equal to control inspiratory duration conflicts with Clark and von Euler (6) finding. As it has already been mentioned their plot originated from multifactor study, i.e., at least three factors were active during an expiratory phase, namely: central action of hypercapnia, its peripheral action and a mechanical consequence of an excessive volume-
blast which is manifested by considerable relaxation of the airway smooth muscle tone followed by significant transient fall of a tonic activity of the pulmonary stretch receptors (28). All these factors tend to shorten an expiratory duration independently on the duration of inspiration. How seriously the $T_I - T_E$ relationship can be affected by the introduction of a factor spreading throughout the respiratory cycle may be deduced from the results of this study. Assuming that unilateral and bilateral vagotomy reduce vagal feedback to 50 and 0% respectively one can use the appropriate data from Table I and try to fit them to the plots in Fig. 1. It appears, however, that these data do not fit either of the plots. Actually for the pigs the relationship is much steeper whereas in the baboons it does not exist whatsoever.

Single breath gain manoeuvres carried out in pigs fully confirmed previous results obtained in dogs by Bartoli et al. (2) thus gain validity of their quantitation of the dependence of phrenic motoneurone output on the vagal feedback input. The existence of both negative and positive vagal feedback mechanisms was documented and confirmed by means of a mathematical analysis of the data obtained.

The plot of terminal values of the phrenic signal vs. corresponding tidal volume expressed by equation 3 determines the strength of a negative vagal feedback mechanism whereas the incremental form of this relationship (equation 5) reveals the excitatory component to be equally strong. The same was found for dogs (equations 15 and 16). Even the duration of the initial period of an unchangeable shape of the phrenic signal was found to be identical for both these species.

The excitatory action of the vagal feedback was not only attributed to the rate of rise of phrenic activity, it was also manifested by appreciable prolongation of the inspiratory time in response to $G = 0$ manoeuvres when compared to the duration of inspiration after vagotomy. These two states differed only by the amount of a tonic low frequency stretch receptors activity corresponding to the EEV level. The presence of this type of vagal feedback during $G = 0$ manoeuvre prolonged an inspiratory phase to 259% on average (258 from the regression line in the equation 4) of its control value whereas after bilateral vagotomy it was only 174% (see Table II and Fig. 2). Similar proportion can be found when the states corresponding to $G \approx 50\%$ and after unilateral vagotomy are considered. Both differences are highly significant and should not be overlooked. The linear regression line (equation 4) relating to the $T_I$ vs. $V_T$ dependence is most likely to be the key link in the task of a final learning of a concurrence of central and vagal mechanisms regulating the pattern of inspiration. Inspiratory cut-off hyperbola of Clark and von Euler although correlating the same variables considerably
deviates from a straight line probably due to the reasons mentioned above. Comparison of the equations 3 and 4 leads to the conclusion that the duration of inspiration is of major importance in the regulation of a breath volume during isocapnic conditions.

There are two basic factors determining tidal volume for a given value of lung compliance and airway resistance, namely the rate of rise of inspiratory activity and the duration of inspiration. The former is practically almost linear, hence the peak level of the inspiratory activity is of minor importance.

Integrated phrenic activity correlates very well with the shape of transpulmonary pressure during inspiration (2, 16). The rate of fall of transpulmonary pressure determines in turn the rate of inspiratory flow which is nearly constant throughout the most part of inspiration. The integral of flow gives volume. On the other hand, according to Davis et al. (7), Grotek et al. (13), Romaniuk et al. (27) and Ryba (28) the vagal feedback from pulmonary stretch receptors correlates very well with transpulmonary pressure. The great importance of the inspiratory time in the regulation of tidal volume implies that the mechanism of integration underlies the regulatory process terminating inspiration. What exactly is being integrated and how, remains still to be studied, nevertheless the most likely answers can be already postulated. There is a central mechanism perfectly capable of terminating inspiration in the absence of vagal feedback, except perhaps in guinea pig (12, 23) and it only shares the task of a precise volume control with the latter. It is also well known that in order to abolish the inspiratory cut-off it is necessary to perform pneumotaxic centre lesion as well as bilateral vagotomy (20, 21).

It seems justified to assume that there is one common mechanism (but not necessarily the same neuronal structure) taking into account all factors determining the pattern of inspiration. The inspiratory activity is therefore most likely to be subjected to a continuous integration and this would be either central inspiratory activity or the activity from pulmonary stretch receptors that is concomitant with inspiration. The existence of so called inspiratory time dependent volume threshold (shifted only) after either vagotomy or pneumotaxic centre lesion (8) supports this supposition.

The nature of a neural process of integration should not be identified with the essence of its mathematical meaning. Due to synaptic decay it must be an integration of a leaky character and as such would be possibly the simplest mechanism underlying the process of generation of a rhythmic activity. The rate of rise of the inspiratory activity is generally very well controlled so there must be some negative
feedback mechanism continuously regulating that rate. The presence of a positive feedback mechanism is a prerequisite for the process of generation of any activity. It seems therefore quite likely that the system as a whole is based upon the selfexcitatory action being inhibited by its integral. The excitatory mechanism boosting central inspiratory discharge is closely followed by inhibitory one controlling the rate of that boosting. In other words the excitatory function is being continuously integrated on synaptic links and the resulting inhibitory function holds the process of excitation within controllable limits, thus creating a sort of “push–pull” mechanism of control.

The inhibitory function as a resultant of integration of the excitatory one must at certain moment outgrow the latter and thus trigger the process of its extinction. The more vigorous is the development of the excitatory function, the faster is the growth of its integral and hence the sooner inspiratory cut-off. As it has been extensively discussed (2) the difficulty in inhibiting the progressing inspiration at its very beginning (very small value of integral) supports the concept that integration underlies the mechanism of inhibition. The leaky character of the process of inhibition implies that any function capable of inhibiting inspiration must develop at the rate higher than the rate of that leak otherwise it will impose only the excitatory action (e.g., Heads paradoxical reflex or experiments of Wyss). An excessive excitation, in turn, will result in much faster growth of an excitatory function (due to an inert property of integration) which will become uncontrolable (e.g., an augmented breath phenomenon).

The rate of leak attributed to the process of integration must also influence the net result of an excitatory and inhibitory action, i.e., the rate of rise of the inspiratory activity. CO₂ and high body temperature seem to considerably accelerate that leak. Both these factors decrease the strength of the Hering–Breuer inhibitory reflex and increase the rate of rise of the inspiratory activity. On the other hand hyperventilation apnoea reflects the result of a markedly reduced leak by hypocapnia and thus final predominance of an inhibitory influence (see also Discussion in Bystrzycka et al. (5)).

Summarizing, the rate of leak of the process of integration and the ammount of overall activity subjected to that integration determine the peak level of inspiratory activity and the duration of inspiratory phase. Inspiratory time dependent volume threshold should be, therefore regarded as a conventionary expression since tidal volume appears to be rather the net result of several factors and mechanisms. From the moment of inspiratory cut-off the inhibitory function (the value of integral) begins to fall with time constant determined by the rate
of leak and, in presence of vagal feedback, by the level of end expiratory activity from pulmonary stretch receptors. The higher level of this activity the slower decay of the inhibitory function. This would explain the role of the EEV level in vagal control of respiratory rate (see 1) and the role of vagal feedback in respiratory rate response to hypercapnia (CO\textsubscript{2} sensitivity of pulmonary stretch receptors).

In the absence of vagal feedback some respiratory rate response to CO\textsubscript{2} remains due to its acceleratory action on the rate of leak which would tend to shorten both inspiratory and expiratory duration. The \(T_1-T_E\) dependence is therefore an inherent property of central mechanisms generating rhythmic inspiratory activity. The existence of this dependence is not only limited to hypercapnic conditions, since it does not depend only on change in the rate of leak. At given rate of leak any factor affecting the inspiratory duration (e.g., reduced vagal feedback) must indirectly affect the duration of an expiratory pause. The lesser phasic vagal feedback the longer \(T_I\) and higher peak level of an inspiratory activity, thus the bigger value of its integral. The decay of an inhibitory function starts then from higher level so the next inspiration will be triggered later. Results of this study seem to confirm this supposition. When the vagal feedback is being reduced by unilateral vagotomy, both phasic and tonic activities from pulmonary stretch receptors are equally affected which, in turn, must affect the rate of decay thus the \(T_I-T_E\) relationship will be quantitatively different. The existence of this relationship as a general property of neural control of respiration can finally explain one of the most frustrating discrepancy among experimental observations, namely the contradiction between the evidence of Bartoli et al. (1) that the level of EEV has a strong vagally mediated influence on the rate of respiratory rhythm and the commonly known result of vagotomy.

In conclusion, the outline of the concept presented herein postulates the essence of the most likely mechanisms underlying the central and vagal control of breathing. The terms "integration", "leak", "decay" are of a conventionary character and express rather idea then proved mechanisms. Although this concept has been formulated on the ground of experimental results limited to "input–output" type of study one can find its confirmation in an experimental work focused on central organisation of respiratory complex. Experiments and idea of Kahn and Wang (18) have created solid base for the views expressed in this paper. They have provided strong evidence for both the continuity of the vagal inhibitory mechanism (see their Fig. 3 and 5) and an integrative character of this mechanism (their Fig. 6).

This study practically failed to answer one of the most important
questions, namely whether, due to the genuine species difference, there are conflicting views as to the existence of the positive vagal feedback. Indeed, baboons did not unequivocally display such a mechanism at least as far as the dynamic change of slopes was concerned. On the other hand, however, an augmented breath type of response was consistently observed.

Finally it is difficult to decide whether great variability in the pattern of inspiration can be regarded as specific for primates or it was just a result of a particular physiological condition of the animals. The absence or failure of certain vagal mechanisms responsible for precise control of the slope of raise of phrenic activity was apparent. Whichever was the case, the presence of the excitatory component of vagal feedback seems to be essential for the regulation of the pattern of inspiration.

The magnitude of a negative feedback type of response in baboons was higher for moderately hypocapnic state ($G_0 > 100\%$ group) than during normocapnic conditions. This would point out that the inhibitory mechanisms are reciprocally related to the level of CO$_2$ and, moreover, such a dependence is quite likely to play a predominant role in bridging a gap between reflex and chemical control of breathing.

The effect of a joint action of these two mechanisms was demonstrated by the hyperbolic relation shown in Fig. 3. Varying the level of the initial gain setting ($G_0$) was in fact a good simulation of hypo- and hyperefficiency of a motor function of the inspiratory muscles. The regulation of an arterial CO$_2$ within the range of hypoefficiency was nearly perfect. The lack of an identical study performed after vagotomy makes the quantitative assessment of the regulatory power of these two mechanisms practically impossible.

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