Instability and Mitotic Patterns in Tissue Growth

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A mathematical model which reproduces several qualitative features of cancerous tumor growth is proposed. The onset of unstable growth is characterized by a dimensionless number, which is defined in terms of the parameters describing the system. Patterns of mitotic activity of the model system are compared with experimentally observed patterns of mitotic activity in cancerous tissues.

I Introduction

LATERALLY spreading inhibitory and excitatory interactions play important roles in the development and stabilization of spatial patterns in a wide variety of biological systems. For example, inhibitory synaptic connections in neural networks can lead to the observation of edge effects and Mach bands in visual perception [1, 2];[2] regular spatial patterns of structures in a homogeneous field (hairs on insects, stomata on leaves, cacti in deserts) can arise as a result of inhibition of new structure formation in the region of a structure already present [3–5]; gradients of diffusing metabolites might determine developmental fields in simple organisms [6, 7] and embryogenesis [8, 9]. In the following we show how diffusible mitotic inhibitors can act to regulate tissue size in normal and cancerous growth.

There is good evidence that control of cellular replication in a number of mammalian tissues is at least partially determined by a negative feedback from the tissue itself [10–12]. The agents of negative feedback, called "chalones," are tissue specific, mitotic inhibitors produced by the tissues. Chalones have been isolated (but not in pure form) [13–15] and have been found to inhibit mitosis via a sigmoidal curve [16]. For low chalone concentrations the mitotic rate is high, but as the chalone concentration is increased through some critical range, the mitotic rate rapidly decreases and remains at a comparatively low basal rate for high chalone concentrations. It has been suggested that a breakdown in the normal functioning of this chalone mechanism may be responsible for the uncontrolled (but potentially controllable) tissue growth in at least some cancers [12]. Strong supporting evidence for this hypothesis comes from a study of the pattern of tumor growth [17], as well as experimentally induced regression of tumors in rodents following treatment with solutions believed to contain tissue specific chalones [18–20]. In Section II we propose a theoretical model for tissue size regulation via negative feedback from the tissue itself. The model, which is schematic and confined to one dimension, illustrates the qualitative properties of the chalone mechanism. In Section III, the observed patterns of growth for the model system and for cancerous tumors are compared. The results are discussed in Section IV.

II Instability in the Chalone Control Mechanism

We assume the target tissue is of length, L, and extends $x = -L/2$ to $x = L/2$. The tissue produces chalone at a rate $P(l^{-1}t^{-1})$, the chalone diffuses with a diffusion coefficient $D(l^2t^{-1})$, and decays at a rate $\lambda(t^{-1})$, proportional to its concentration. Beyond the target tissue, the chalone continues to diffuse and decay but is not produced. This system may be represented by the diffusion equation

$$\frac{\partial C}{\partial t} = D\frac{\partial^2 C}{\partial x^2} - \lambda C + PS(x) \tag{1}$$

where the source term, $S(x)$, is given by

$$S(x) = 1 \quad |x| \leq \frac{L}{2}$$

$$S(x) = 0 \quad |x| > \frac{L}{2} \tag{2}$$

We assume that tissue growth is regulated by the concentration of the chalone by a switch mechanism [16]. If $M(l^{-1}t^{-1})$ is the mitotic rate, we have

$$M = 0 \quad C \geq \theta$$

$$M = M_0 \quad C < \theta \tag{3}$$

If the concentration of chalone is lower than threshold at some place in the tissue, mitosis and subsequent growth will occur. This will result in a larger tissue and a new distribution of chalone. The growth process, however, takes place very slowly, with rapidly growing tumors enlarging at a rate of approximately 0.1–0.5 mm/day [17], in comparison with the transport mechanisms which rapidly establish new distributions of chalone in perturbed tissues [16]. Consequently the size of the tissue may be considered stationary and the distribution of chalone computed. If a tissue grows until it reaches a size in which the distribution of chalone is everywhere greater than θ, growth will cease.

The chalone concentration may be readily computed for this system by using Green's function techniques [21]. For equation (1) we find that for long times the concentration of chalone is given by

$$C(x) = P\int_{-\infty}^{\infty} dx' G(x - x')S(x') \tag{4}$$

where $S(x)$ is given in equation (2) and the Green's function for the diffusion equation is

$$G(x) = \frac{1}{2\sqrt{D\lambda}} \exp\left(-\sqrt{\frac{\lambda}{D}}|x|\right) \tag{5}$$

Equation (4) may be immediately integrated to find the chalone concentration. After some algebra and simplification we find

$$C(x) = \frac{P}{\lambda}\left\{1 - \left[\cosh\sqrt{\frac{\lambda}{D}}\,x\right]\left[\exp\left(-\sqrt{\frac{\lambda}{D}}\frac{L}{2}\right)\right]\right\} \quad \text{for } |x| \leq \frac{L}{2}$$

$$C(x) = \frac{P}{\lambda}\left[\sinh\sqrt{\frac{\lambda}{D}}\frac{L}{2}\right]\left[\exp\left(-\sqrt{\frac{\lambda}{D}}|x|\right)\right] \quad \text{for } |x| \geq \frac{L}{2} \tag{6}$$

For a tissue of any size, the chalone concentration is therefore maximum at the center of the tissue and is given by

$$C(0) = \frac{P}{\lambda}\left[1 - \exp\left(-\sqrt{\frac{\lambda}{D}}\frac{L}{2}\right)\right] \tag{7}$$

The chalone concentration monotonically decreases to a concentration

$$C\left(\left|\frac{L}{2}\right|\right) = \frac{P}{2\lambda}\left[1 - \exp\left(-\sqrt{\frac{\lambda}{D}}L\right)\right] \tag{8}$$

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[2]Numbers in brackets designate References at end of Brief.

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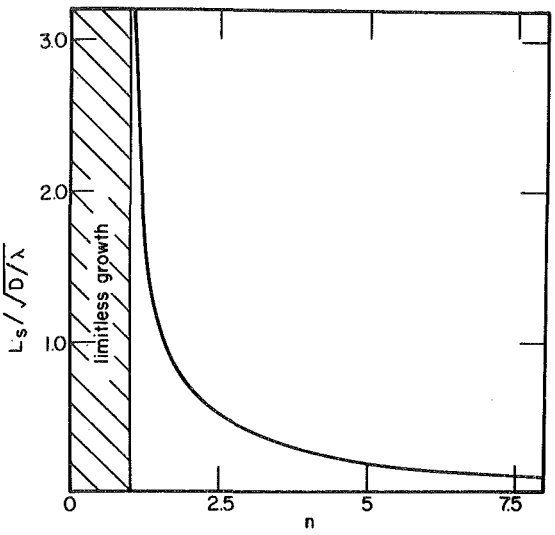

Fig. 1 The stable size of the model tissue described in the text, plotted in units of $\sqrt{D/\lambda}$, as a function of n. For $n<1$, the model is unstable and limitless growth will occur.

at the edge. As the size of the tissue increases, the concentration of the chalone reaches a limiting maximum value of P/λ at the center of the tissue and $P/2\lambda$ at the edge of the tissue. In order for growth to cease, the chalone concentration must everywhere in the tissue be greater than θ. Defining the dimensionless variable, n,

$$n = \frac{P}{2\lambda\theta} \tag{9}$$

we find that limited growth will occur for $n > 1$. If this condition does not hold the tissue will be unstable and limitless growth will ensue.

For tissues in which $n > 1$, the size of the tissue will be determined by the parameters governing the chalone production, decay and transport. Since the chalone concentration is a monotonically decreasing function of distance from the center of the tissue, the concentration of chalone will be everywhere greater than the threshold, θ, if it is equal to θ at the edge of the tissue. Substituting in equations (8) and (9) we compute the size of the stable tissue L_s, to be

$$L_s = \sqrt{\frac{D}{\lambda}} \ln\left(\frac{n}{n-1}\right) \tag{10}$$

for tissues in which $n > 1$. This dependence is plotted in Fig. 1. For this model, the singularity in equation (10) at $n = 1$ corresponds to the region of limitless tissue growth which is found in cancerous tumors. A small change in the parameters describing production and decay of the chalone, may be sufficient to switch a previously stable, well-regulated tissue to a system of limitless growth. Conversely, a system in the region of limitless growth can be brought under control by increasing chalone concentration artificially. In recent experimental work, remission of cancer has been achieved by administering chalone derived from the tissue of origin of the malignancy. Although further experimental verification and extension of these results is needed, remission has been achieved for melanomata tumors in mice and hamsters by treatment with melanocyte chalone [18], chloroma tumors in rats by treatment with granulocytic chalone [19], generalized granulocytic leukemia rats by treatment with granulocytic chalone [20]. These experimental results indicate that changes in the concentration of chalone can markedly alter the stability properties of growing malignant tissue, *in vivo*.

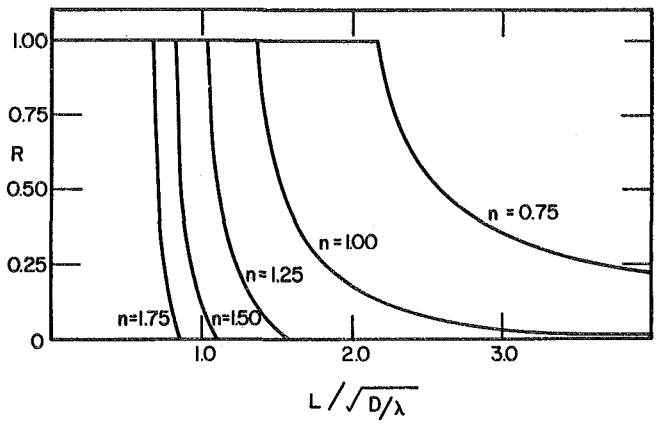

Fig. 2 The fraction of tissue mass engaged in active mitosis, R, as a function of the size of the system, plotted in units of $\sqrt{D/\lambda}$, for several values of the dimensionless variable, n

III Mitotic Patterns in Stable and Unstable Growth

For stable tissue growth ($n > 1$), the chalone concentration will everywhere be less than the mitotic threshold and mitosis will occur throughout the tissue when the tissue is small. As the tissue increases in size, the chalone concentration will exceed the mitotic threshold in the central tissue, resulting in a peripheral mitotic pattern. The growth zone will be confined to an increasingly narrow region at the periphery of the tissue and will eventually disappear at the stable tissue size (Fig. 2).

In constrast, for unstable tumor growth ($n < 1$), mitosis either occurs throughout the entire tissue for all stage of growth, or becomes confined to a peripheral zone which reaches a *constant* width as the tumor enlarges. The width of the zone of active mitosis can be computed for the unstable growth regime. Consider a point located a distance W, internal to the tissue from the edge of a growing tumor of size L. The chalone concentration at W can be found from equation (6) and is given by

$$C(W) = \frac{P}{\lambda}\left\{1 - \frac{\exp\left(-\sqrt{\frac{\lambda}{D}}W\right)}{2} - \frac{\exp\left[-\sqrt{\frac{\lambda}{D}}(L-W)\right]}{2}\right\} \tag{11}$$

If the chalone concentration at W is equal to θ, active mitosis will occur only in the regions lying peripheral to W. Further, for large tumors $\left(L >> \sqrt{\frac{D}{\lambda}}\right)$ the last term on the right-hand side of equation (11) makes a negligible contribution to the chalone concentration at W, for the region in which $W << L$. Equating the right-hand side of equation (11) to θ, and dropping the last term, we compute the width of the peripheral zone of active mitosis, W_s, to be

$$W_s = \sqrt{\frac{D}{\lambda}} \ln\left(\frac{n}{2n-1}\right) \tag{12}$$

In Fig. 3 we give W_s as a function of n. Three regimes are present. For $n > 1$, there is stable growth, equation (12) does not hold, and W_s is zero throughout the range. For $0.5 < n < 1$, there is a peripheral zone of constant finite width of active mitosis for large tumors. At $n = 0.5$ there is a singularity in equation (12), so that for $n \leq 0.5$ there is continuous active mitosis throughout the tumor for all tumor sizes.

Mitotic patterns in cancerous tumors have been studied. In early work, Mayneord observed a peripheral mitotic pattern in rapidly enlarging tumors, but attributed the existence of this zone to cellular degeneration (necrosis) in the central tumor mass [22]. In more recent work, mitotic figures were observed

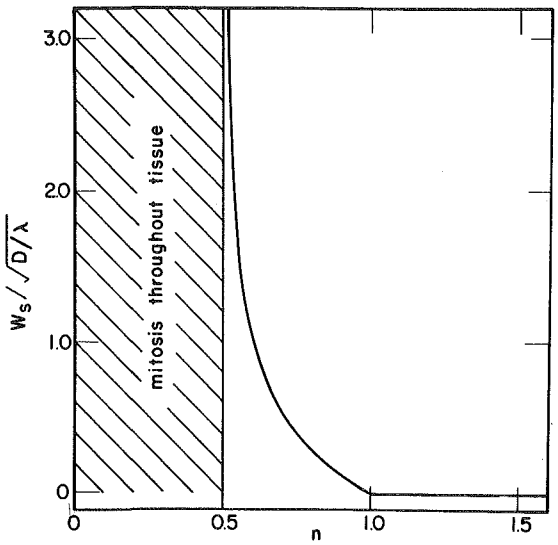

Fig. 3 The width of the zone of active mitosis for large tissues ($L >> \sqrt{D/\lambda}$) plotted in units of $\sqrt{D/\lambda}$, as a function of n

throughout the tissue for small tumors, while in larger tumors a peripheral mitotic zone was observed. For some specimens, close examination of the central mass of the growing tumors with peripheral mitotic zones revealed no evidence of necrosis, and it is possible that gradients of mitotic inhibitors were responsible for the observed growth pattern [17]. It is interesting to note that a direct consequence of having an active mitotic zone of constant width is that tumors will grow at a constant linear rate (or equivalently, at a rate proportional to the cube root of tumor volume) [22]. In studies of the dynamics of tumor growth, this simple functional dependence has been observed [23, 24]. However, in more recent work it has been argued that a better description of tumor growth is given by the Gompertz growth curve, and that the simple linear growth occurs over only a limited region of tumor growth [25].

IV Discussion

The model presented here is schematic. Important factors in the systems regulating tissue size have been ignored in an attempt to represent in simple mathematical form an instability which can lead to cancerous growth of tumors. It is important to determine whether the qualitative features discussed will be preserved for more realistic mathematical models of growing tissues. For two and three dimensional systems, both the instability and the peripheral growth zone will be observed. Of more critical importance is the effect of substituting experimentally observed continuous mitotic control functions for the discontinuous switch assumed in equation (3). In recent studies of the dynamics of biochemical control networks, it was observed that important qualitative features of the dynamics of these networks remained invariant to modifications of control functions of just the sort encountered here [26, 27]. Similarly, it is anticipated that the qualitative properties of the mitotic control network will also be preserved. However, for continuous control functions, the zone in which mitosis is actively occurring will not have a sharp boundary, but there will be a transition zone between regions of high and low mitosis. The width of the transition zone will depend on the slope of the mitotic control function, and will increase as the control function becomes less steep. Other modifications are needed if mitosis is never completely inhibited in the target tissue, but occurs continuously throughout the life of the organism. When this occurs, as it does for example in epidermal tissue, the rates of cell aging and cell death will be important and must also be considered in determining the tissue size [28]. For other tissues, in which the ability to replicate is almost completely lost, (for example, neural tissue and striated muscle) or for tissues in which replication is negligible after an initial growth phase *unless* there is some perturbation to the tissue such as partial removal of tissue (for example, liver tissue or renal tubules) [29], the aforementioned analysis will apply. For these systems, there will exist interrelationship between the geometry of the growing tissue, mitotic patterns, and stability characteristics of the mature tissue after alteration of its geometry.

I have analyzed a mathematical instability which will lead to uncontrolled growth. In order to determine whether this theoretical model is appropriate for studying cancer, careful experimental investigation of the parameters involved is necessary. Of primary importance is the isolation and characterization of pure chalone, so that accurate determinations of mitotic response to varied extracellular and intracellular chalone concentrations may be undertaken. An understanding of the control systems regulating cellular proliferation should lead to the development of successful therapies for malignant growth.

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References

1 Ratliff, F., *Mach Bands: Quantitative Studies on Neural Networks in the Retina*, Holden-Day, San Francisco, 1965.

2 Wilson, H. R., and Cowan, J. D., "A Mathematical Theory of the Functional Dynamics of Cortical and Thalamic Nervous Tissue," in Press, *Proceedings of the Royal Society*, London, Series B.

3 Wigglesworth, V. B., "Local and General Factors in the Development of Pattern in *Rhodnius Prolixis* (Hemiptera)," *Journal of Experimental Biology*, Vol. 17, 1940, p. 201.

4 Lawrence, P. A., "Cellular Differentiation and Pattern Formation during Metamorphosis of the Milkweed Bug *Oncopeltus*," *Development Biology*, Vol. 19, 1969, p. 12.

5 Glass, L., "Stochastic Generation of Regular Distributions," *Science*, Vol. 180, 1973, p. 1061.

6 Rose, S. M., "Polarized Inhibitory Control of Regional Differentiation During Regeneration in *Tubularia*," *Growth*, Vol. 31, 1967, p. 149.

7 Macklin, M., "Analysis of Growth Factor Gradients in Hydra," *Journal of Cellular Physiology*, Vol. 72, 1968, p.1

8 Wolpert, L., "Positional Information and Pattern Formation," *Towards a Theoretical Biology*, Vol. 3, *Drafts*, Waddington, C. H., ed., Aldine, Chicago, 1970, p. 198.

9 Crick, F., "Diffusion in Embryogenesis," *Nature* (London) Vol. 225, 1970, p. 420.

10 Weiss, P., "Specificity in Growth Control," *Biological Specificity and Growth*, Butler, E. G., ed., University Press, Princeton, 1955, p. 195.

11 Osgood, E. E., "A Unifying Concept of the Etiology of the Leukemias, Lymphomas, and Cancers," *Journal of the National Cancer Institute*, Vol. 18, 1957, p. 155.

12 Bullough, W. S., "Mitotic and Functional Homeostasis: A Speculative Review," *Cancer Research*, Vol. 25, 1965, p. 1638.

13 Bullough, W. S., and Laurence, E. B., "Mitotic Control by Internal Secretion: The Role of the Chalone-Adrenalin Complex," *Experimental Cell Research*, Vol. 33, 1964, p. 176.

14 Bullough, W. S., and Laurence, E. B., "Control of Cancer in Mouse and Hampster Melanomata by Means of the Melanocyte Chalone," *European Journal of Cancer*, Vol. 4, 1968, p. 607.

15 Rytomaa, T., and Kiviniemi, K., "Control of DNA Replication in Rat Chloroleukaemia by Means of the Granulocytic Chalone," *European Journal of Cancer*, Vol. 4, 1968, p. 595.

16 Rytomaa, T., and Kiviniemi, K., "Control of Granulocyte Production. II. Mode of Action of Chalone and Antichalone," *Cell and Tissue Kinetics*, Vol. 1, 1968, p. 341.

17 Bullough, W. S., and Deol, J. U. R., "The Pattern of Tumour Growth," *Symposium of the Society for Experimental Biology*, Vol. XXV, 1971, p. 255.

18 Mohr, U., Althoff, J., Kinzel, V., Suss, R., and Volm, M., "Melanoma Regression Induced by 'Chalone': A New Tumour Inhibiting Principle Acting *in vivo*," *Nature* (London), Vol. 220, 1968, p. 138.

19 Rytomäa, T., and Kiviniemi, K., "Chloroma Regression Induced by the Granulocytic Chalone," *Nature* (London), Vol. 222, 1969, p. 995.
20 Rytomäa, T., and Kiviniemi, K., "Regression of Generalized Leukaemia in Rat Induced by the Granulocytic Chalone," *European Journal of Cancer*, Vol. 6, 1970, p. 401.
21 Morse, P. M., and Feshbach, H., *Methods of Theoretical Physics*, Part II, McGraw-Hill, New York, 1953, Chapter 12.
22 Mayneord, W. V., "On a Law of Growth of Jensen's Rat Sarcoma," *American Journal of Cancer*, Vol. 16, 1932, p. 841.
23 Klein, G., and Révész, L., "Quantitative Studies on the Multiplication of Neoplastic Cells *in vivo*. I. Growth Curves of the Ehrlich and MC1M Ascites Tumors," *Journal of the National Cancer Institute*, Vol. 14, 1953, p. 229.
24 Patt, H. M., and Blackford, M. E., "Quantitative Studies of the Growth Response of the Krebs Ascites Tumor," *Cancer Research*, Vol. 14, 1954, p. 391.
25 Laird, A. K., "Dynamics of Tumor Growth," *British Journal of Cancer*, Vol. 18, 1964, p. 490.
26 Glass, L., and Kauffman, S. A., "Co-operative Components, Spatial Localization and Oscillatory Cellular Dynamics," *Journal of Theoretical Biology*, Vol. 34, 1972, p. 219.
27 Glass, L., and Kauffman, S. A., "The Logical Analysis of Continuous, Nonlinear Biochemical Control Networks," *Journal of Theoretical Biology*, Vol. 39, 1973, p. 103.
28 Bullough, W. S., "The Control of Epidermal Thickness," *British Journal of Dermatology*, Vol. 87, 1972, p. 181 (Part 1), p. 347 (Part 2).
29 Post, J., and Hoffman, J., "Cell Renewal Patterns," *New England Journal of Medicine*, Vol. 279, 1968, p. 248.

Adaptive Control of Respiratory Mechanics

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A hierarchial model is proposed to describe the performance principles of the adaptive system controlling respiratory mechanics. The decision criterion of the controller is modeled by a two-level minimization criterion, where the inspiratory and expiratory flow patterns are determined on the lower level and the overall pattern of breathing on the higher level. The objective functionals are related to the effectivity of breathing. An extension of the lower level model to include control of the rib cage and abdomen separately is also reported.

Introduction

HIERARCHY and adaptivity are typical characteristics of physiological control systems. In order to gain a deeper insight into the function of the regulators in man, the systemic approach and mathematical modeling have to be used. Large scale human systems are often divided into seemingly independent, in reality highly interconnected, blocks. When examining these blocks it is impossible to control all the relevant inputs and outputs simultaneously under experimental conditions. The only way to try to cope with the increasing number of variables in dynamical subsystems is to build a simulation model.

Simulation with black box models cannot, however, explain the ability of the system to learn, i.e., to adapt itself to new circumstances. To account for this adaptivity the model must include some kind of performance criterion. A natural choice is a minimization criterion. The system learns the new circumstances through a continuously active identification or self-optimizing process correcting the operation of the system so that it minimizes the objective functional in question subject to the possible constraints on the control variables.

The mechanical breathing process is a typically adaptive system whose main purpose is to meet the ventilatory demand determined by the chemical state of the blood. The desired ventilation level can, however, be achieved in many ways, thus a decision is needed to choose between the different alternative control strategies for the mechanical system. Our work concentrates on the adaptive nature of the system. Its performance principles are studied with the aid of mathematical modeling. The models we have developed will be dealt with in this paper.

Modeling Adaptivity

The early works by Rohrer [11][2] and Otis, et al. [8] were based on the minimum respiratory work rate criterion. They used a sinusoidal airflow shape and found that an optimal breathing frequency existed. Mead [7] showed that a minimum force amplitude criterion would give better results. Widdicombe and Nadel [13] included control of the airway dimensions in the former models and obtained an optimal dead space volume with respect to both criteria. There has been only one attempt [12] to find criteria that explain the shape of the respiratory airflow curve. The authors suggested minimum mean squared acceleration as the performance function for humans. In closed form their solution is a parabolic curve for both inspiration and expiration and it does not deviate much from a sine-curve.

None of the previous models have been able to predict the effects of increased ventilation or external loading on the individual respiratory variables of test subjects. In these papers the aim has merely been to prove that breathing is optimal with regard to minimum effort or maximum gas exchange. Our main hypothesis, on the contrary, is that the system is adaptive, and thus it is also assumed that it must have some criterion for decision making in adaption. A similar idea is presented in the qualitative model of Priban and Fincham [10].

In the case of such subprocesses as breathing the criterion is probably not so clear and general as the principle of minimum energy expenditure, although energy cost seems to be an essential component. It should be noted that in large scale systems partial optima need not necessarily yield the total optimum. Further, it is very difficult to define precisely all the tasks that different subsystems are performing, yet these tasks should be reflected in the corresponding performance criteria.

In the first stage we have tried to find a physiologically meaningful performance criterion expressed in mechanical entities, that successfully explains the spontaneous breathing of test subjects . To do this it has been necessary to extend the number of variables studied and to make the assumptions less restrictive and more detailed. In the earlier studies, for instance, the difference in the duration of inspiration and expiration and the active role of the muscles in the expiratory phase were neglected. Moreover, as far as we know this is the first time that a model has been used to study the control of the rib cage and abdomen with respect to adaptivity.

Nomenclature

The following symbols are used in the two models:

Common symbols

$\dot{V}_A$ = average alveolar ventilation, l/s
V_T = tidal volume, l
V_0 = change of end expiratory lung volume, l
t_1 = duration of inspiration, s
t_2 = duration of expiration, s
t_3 = duration of end expiratory pause, s
J = objective functional

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