Lys-Pro-Lys/Arg sequences in a coiled structure near the amino acid terminal regions.

The amino acid compositions of the two groups of phosphoproteins are obviously different. This seems to rule out the possibility that the two groups evolved from a common ancestor. However, there remains the possibility that viral p12 evolved from H5 histones or related proteins through a genetic recombination process involving only the amino terminal end of the H5 histones. Alternatively, the sequence homology may be the result of convergent evolution. The finding of sequence homology between viral and cellular structural proteins is consistent with the emergence of infectious entities by a process of differentiation from preexisting cellular genes. Vestiges of these primordial origins might be more readily detected in retroviruses because of their inheritance in the cellular genome. Given recent information on messenger RNA splicing (*21*) and shifts in physical location of specific DNA segments during maturation of the immune system (*22*), it is not unreasonable to suggest that similar mechanisms might have contributed to the origin of viral structural proteins. Thus, several cellular genes of diverse function could have contributed to the origin of individual virion polypeptides. This speculation is readily derived from the protovirus hypothesis of Temin (*23*), in that the coding sequences assume their appropriate physical alignments by a series of rearrangements mediated by cellular reverse transcriptase. Further analyses of complete sequences of retrovirus polypeptides will be necessary to fully evaluate our speculations; however, this could provide a rigorous evaluation of current theories of retrovirus origin.

The sequence homologies between H5 histones and the viral p12 proteins indicate a conserved structural feature common to both groups of proteins and, in turn, a common function. As we pointed out above, both the H5 histones and the viral p12 proteins bind to nucleic acids through an interaction modulated by phosphorylation of the proteins. It has been suggested that the amino terminal portion of chicken H5 may participate in this binding (*7*). The amino terminal sequence homology between viral p12 proteins and H5 histones could suggest that the amino terminal regions of p12's also may be involved in nucleic acid binding or phosphorylation (or both).

Further studies may determine whether or not the common structure-function relationship implied here between proteins involved in erythroid differentiation and proteins isolated from leukemia viruses could be related to molecular mechanisms of viral leukemogenesis.

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15. Abbreviations for amino acid residues are Ala, alanine; Arg, arginine; Asn, asparagine; Asp, aspartic acid; Cys, cysteine; Gln, glutamine; Glu, glutamic acid; Gly, glycine; His, histidine; Ile, isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Phe, phenylalanine; Pro, proline; Ser, serine; Thr, threonine, Trp, tryptophan; Tyr, tyrosine; and Val, valine.
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Control of Oscillations in Hematopoiesis

Abstract. *Results of a mathematical analysis of models of hematopoietic systems introduced by Mackey and Glass are given. The models include a constant time lag, and it is shown that this lag has a critical value above which oscillations in blood cell concentration occur. To reduce the likelihood of disease associated with such oscillations, physiologists should seek to learn how to increase this critical value of the lag.*

Mackey and Glass (*1*) introduced mathematical models for blood cell production with a constant lag $\tau > 0$ between cell genesis in the bone marrow and release into the bloodstream. Their models have the form

$$\frac{dx(t)}{dt} = \Lambda(x(t - \tau)) - ax(t) \qquad (1)$$

where $x(t)$ is the cell density at time t, a is a constant death rate, and Λ is a smooth function describing the generation of cells of a particular kind, such as erythrocytes. We report the results of a mathematical analysis of these models, which leads to qualitative advice to the physiologist.

This analysis, which is essentially nonlinear, reveals that there exists a critical value of the lag, τ_c. For values of τ in an interval on one side of τ_c, the cell density exhibits stable oscillations with amplitude which increases linearly with $|\tau - \tau_c|^{1/2}$. For the nonlinear terms in Λ used in (*1*) (see Eq. 2 below) these oscillations, which are symptomatic of hematopoietic disease, occur for $\tau > \tau_c$. Thus our qualitative advice is to seek to affect the system so as to increase τ_c and, thereby, possibly suppress oscillatory behavior of the cell density.

To illustrate, consider the particular choice of the function introduced in (*1*) [see also (*2*)], namely

$$\Lambda(x(t - \tau)) = \frac{\beta_0 \theta^n x(t - \tau)}{\theta^n + x^n(t - \tau)} \qquad (2)$$

This corresponds to production being a single-humped function of the delayed density; for physiological details and ref-

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erences to the literature on hematologic mechanisms see (*1*-*3*). In Eq. 2, β_0, θ, and n are positive constants adjusted to fit experimental observations, and it is assumed here that $\beta_0 > a > 0$, $n\beta > 2$, and $6aB > \beta_0$, where $B = (\beta_0 - a)/\beta_0$. For the system given by Eqs. 1 and 2 there is a unique steady state $x_e = \theta(B\beta_0/a)^{1/n}$. About this equilibrium, the linear part of Eq. 1 takes the form

$$dz(t)/dt = -az(t) - bz(t - \tau)$$
$$z = x - x_e \qquad (3)$$

The solutions of this equation have a natural frequency $\omega = (b^2 - a^2)^{1/2}$ at a critical value of the lag given by

$$\tau_c = \frac{\arccos(-a/b)}{(b^2 - a^2)^{1/2}}$$
$$0 < \arccos(-a/b) < \pi \qquad (4)$$

where $b = a(nB - 1)$ for Λ given by Eq. 2. The steady state of the nonlinear system is stable for $\tau < \tau_c$. Considering the nonlinear terms and using the Hopf bifurcation algorithm developed in (*4*), we obtain the mathematical result that as τ increases above τ_c, this state gives rise to asymptotically orbitally stable periodic solutions. These have periods longer than $2\pi/\omega$ and exist for τ in some interval $\tau > \tau_c$. Realistic values of the parameters can be substituted in Eq. 4 to give a numerical value of τ_c; for example, the parameters $a = 0.1$ per day, $\beta_0 = 0.2$ per day, and $n = 10$ used in (*1*) for numerical simulations yield $\tau_c = 4.71$ days. For lags greater than this value oscillations occur with periods greater than 16.22 days. In (*1*) the simulations are compared with observed periodic fluctuations in patients suffering from chronic granulocytic leukemia. For the choice of Λ in Eq. 2 it can be seen, for example, that τ_c increases if β_0 or n is decreased (while the other parameters are held fixed).

Results qualitatively the same as those stated above for the function Λ given by Eq. 2 are obtained for a wide class of such functions that depend only on the density at a lagged time. In particular, the onset of stable oscillations is insensitive to the algebraic sign of the second derivative of Λ at x_c. The algorithm used to derive the results presented here (*4*, *5*) gives an exact asymptotic formula for the period and amplitude of the bifurcating periodic solutions and enables their stability (or instability) to be determined. No recourse to numerical methods is required. The theory is, however, a local one; thus the sequence of bifurcations and aperiodic (chaotic) behavior, found numerically in (*1*) for some Λ when $\tau >> \tau_c$, is outside the range of the analytical results reported here.

Equation 1 may not be sufficiently general to model the blood cell control mechanism accurately. Mackey (*2*) considers a more general Λ that includes dependence on the present state. Mackey and Glass (*1*, *3*) consider an arterial CO_2 control system that also includes this form of Λ, which is explicitly

$$\Lambda(x(t), x(t - \tau)) = \lambda - \frac{\alpha V_m x(t) x^n(t - \tau)}{\theta^n + x^n(t - \tau)} \qquad (5)$$

where λ, α, V_m, θ, and n are constants, x is the arterial CO_2 concentration, and τ is the time between oxygenation of blood in the lungs and stimulation of receptors in the brainstem. For this Λ, with $a = 0$, τ_c is given in (*1*), and the algorithm of (*4*) shows that for normal ranges of parameter values stable periodic solutions occur in a range of τ for $\tau > \tau_c$ with period $> 4\tau_c$. Thus the advice for regulation of respiratory disease modeled by Eqs. 1 and 5 is again to seek to increase the critical lag.

More realistic models may require multiple lags or weighted lags. Such models can be analyzed by the algorithm in (*4*) [see also (*5*, *6*)]. Experiments are needed to establish the connection between the physiology of control processes subject to diseases with oscillatory symptoms (for instance, hematopoiesis) and the various model parameters; these could lead to means for control of the critical lag to cause less likelihood of stable or unstable oscillations.

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Sodium-Selective Liquid Ion-Exchanger Microelectrodes for Intracellular Measurements

Abstract. *The sodium-selective ligand 1,1,1-tris[1-(2-oxa-4-oxo-5-aza-5-methyl) dodecanyl]propane dissolved in 3-nitro-o-xylene containing a small amount of the lipophilic anion tetrachlorophenyl borate was used as a liquid ion-exchanger in sodium-selective microelectrodes. The microelectrodes gave rapid, stable responses that were linear functions of the logarithm of sodium activity. They were tested under conditions approximating those to be expected in the cell interior, and the results indicated that they can be used to measure intracellular sodium activity without significant interference from intracellular potassium.*

Intracellular ionic activity measurements with ion-selective microelectrodes are important in many areas of biophysics and physiology (*1*). The regulatory role of Na^+ in cellular function (*2*) makes the development of simple, accurate methods for measuring intracellular Na^+ activity (a^i_{Na}) of special interest. For several years, liquid ion-exchanger solutions (477317 and 477315, Corning Glass Works, Corning, N.Y.) selective to K^+ and Cl^-, respectively, have been commercially available, and they are widely used to make microelectrodes for measuring intracellular K^+ and Cl^- activities (*1*, *3*). So far, the development of liquid ion-exchanger microelectrodes, capable of measuring a^i_{Na} without significant interference from cellular K^+, has been frustrated by the lack of a ligand that, in liquid ion-exchanger form, discriminates at a sufficiently high level between these ions.

Palmer and Civan (*4*) used a 1.5 percent solution of potassium tetrachlorophenylborate in trihexylphosphate as the liquid ion-exchanger solution in Na^+-selective microelectrodes. Recently, we modified the Na^+-selective microelectrodes developed for extracellular Na^+ activity measurements by Kraig and Nicholson (*5*) and used them to measure steady-state a^i_{Na} values in epithelial cells of *Necturus* and bullfrog small intestine (*6*). The Na^+-sensing element in our microelectrodes was a 10 percent solution of the antibiotic monensin (*7*) in 3-nitro-*o*-xylene containing a small amount

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