

SELF-SUSTAINED OSCILLATIONS IN THE
JACOB-MONOD MODE OF GENE REGULATION

Z. Grossman and I. Gumowski
CERN, 1211 Geneva 23, Switzerland

A plausible macroscopic model of genetic regulation by feedback repression, evolved from the original formulation of Goodwin, is considered. This model has the form of a second-order non-linear dynamic system, $\dot{x} = f_1(x,y)$, $\dot{y} = f_2(x,y)$, where $x(t) \geq 0$ describes the mRNA which codes for the protein $y(t) \geq 0$, the latter acting as a repressor. Published analytical, analog, and digital studies of this model do not report the existence of any periodic solutions. A stochastic analysis has produced some irregular undamped oscillations, but these appear to be due to the stochastic elements so introduced.

The model $\dot{x} = f_1(x,y)$, $\dot{y} = f_2(x,y)$ is known to be approximate. In particular it neglects delays of synthesis and transport of x,y from the place of production to the place of effect. Periodic solutions are found when a constant delay is introduced into $y(t)$, or a variable one into $x(t)$. In the latter case, analytical expressions for the amplitude and period are determined by means of a Poincaré-type expansion. In the presence of delay, sustained oscillations of x and y are found to exist in a wide parameter range.

INTRODUCTION

Under the influence of classical thermodynamics and statistical mechanics it was believed for a long time that chemical, biochemical, and biological reactions, described by a system of non-linear ordinary differential equations, have only monotonically varying transients toward or away from a state of static equilibrium. This rather entrenched belief started to evolve with the discovery of sustained oscillations in a purely chemical reaction involving an inorganic catalyst (oxydation of malonic acid by KBrO_3 in the presence of Ce ions)^{1,2}). The presence of temporal oscillations resulted in spatial oscillations [photographs in Herschkowith-Kaufman³]. In due course many oscillatory phenomena were found in biochemistry [survey in Hess and Boiteux⁴]. Present interest appears to centre on glycolysis^{5,6}), which constitutes the next natural step in chemical (and mathematical) complexity. The present paper considers oscillations in a cellular system and, more specifically, the so-called epigenetic oscillations. Although the experimental evidence for the existence, and especially biological relevance of self-sustained epigenetic oscillations, is still a subject of debate⁷⁻⁹), the study of mathematical models of such oscillations has been pursued quite intensively¹⁰⁻¹³). This paper falls in the scope of the latter activity.

A plausible macroscopic model of genetic regulation by feedback repression, evolved from the original formulation of Goodwin¹⁰), is

$$\dot{x}(t) = a(1 + Ay(t))^{-1} - bx(t) \quad , \quad \dot{y}(t) = Bx(t) - Cy(t) \quad , \quad (1)$$

where $x(t) \geq 0$ is a mRNA which codes for the protein $y(t) \geq 0$, the latter acting as a repressor. All reaction constants in (1) are positive, and x, y represent instantaneous concentrations. This model is analogous to the Jacob-Monod system^{14,15}). The study of (1) did not disclose any self-sustained oscillations¹²), except when (1) was replaced by a roughly similar stochastic process¹³). Some rather irregular undamped oscillations were then found, but these oscillations appear to be due entirely to the stochastic elements so introduced. A modification of the non-linear term to the form

$$a y^m (1 + Ay^n)^{-1} \quad , \quad m, n > 0 \quad , \quad (2)$$

as well as an increase of the differential order from two to three, by introducing an intermediate metabolite, also failed to produce a qualitative change in the hoped-for direction¹²). Periodic solutions were reported in a rather narrow parameter range, when $m = 0$, $n = 2$ in (2), and the number of intermediate metabolites is increased to six or more (differential order increased to eight or more)¹⁶).

The situation is radically changed, as will be shown, when pure delay is introduced into (1) without any increase of differential order. Such a modification constitutes an improvement of the model because it permits us to take into account the finite times of transcription, synthesis, and transport (diffusion) between the place

of production and effect of x and y . The biological arguments behind an unambiguous definition of these times are still a subject of study¹⁷). Mathematically no loss of generality occurs when x , y , and t are normalized. Keeping the same names for the normalized variables, the modified version of (1) has two reaction constants and four delays:

$$\dot{x}(t) = [1 + y(t - \tau_1)]^{-1} - b x(t - \tau_3), \quad \dot{y}(t) = x(t - \tau_2) - c y(t - \tau_4). \quad (3)$$

The existence of a constant steady state (static equilibrium) $x_0 = c y_0$, $y_0 = -\frac{1}{2} + \sqrt{\frac{1}{4} + 1/(bc)}$, is unaffected by the delays, but the effect on stability may become strong. For constant delays the variational and characteristic equations of (3) at (x_0, y_0) are

$$L(\tau, y) = \dot{y}(t) + b \dot{y}(t - \tau_3) + c \dot{y}(t - \tau_4) + \beta y(t - \tau_1 - \tau_2) + b c y(t - \tau_3 - \tau_4) = 0, \quad (4)$$

$$\lambda^2 + \lambda (b e^{-\lambda \tau_3} + c e^{-\lambda \tau_4}) + \beta e^{-\lambda(\tau_1 + \tau_2)} + b c e^{-\lambda(\tau_3 + \tau_4)} = 0, \quad (5)$$

where $\beta = (1 + y_0)^{-2}$.

In general, (5) admits an infinity of roots s_i [eigenvalues of $L(\tau, y)$]. Let $\theta(t)$ be a sufficiently smooth function and

$$y(t) = \theta(t), \quad -\tau \leq t \leq 0, \quad \tau = \max(\tau_1 + \tau_2, \tau_3 + \tau_4) \quad (6)$$

an initial condition assuring the existence of unique solutions of (3) and (4). It is known that in general the eigenvalues s_i are enumerable and can be ordered according to their moduli, i.e. so that $|s_{i+1}| \geq |s_i|$. Suppose for simplicity that all s_i are non-degenerate. The solution of (4) and (6) can then be written in the form

$$y(t) = \sum_{i=-\infty}^{\infty} C_i e^{s_i t}, \quad C_i = \text{complex constants} \quad (7)$$

where the C_i , $i < 0$ are so chosen that $y(t)$ is real-valued. When $\theta(t)$ is sufficiently smooth (as assumed), the sequence $\{C_i\}$ will not cause a divergence of the series in (7). A sufficient condition of stability of the steady state (x_0, y_0) is therefore $\text{Re } s_i < 0$ for all i , whereas a sufficient condition of instability is $\text{Re } s_i > 0$ for one i . The case $\text{Re } s_i = 0$ for some i and $\text{Re } s_i < 0$ for all others is a critical one (in the sense of Liapunov). The s_i depend continuously on τ_1, \dots, τ_4 . If a critical case exists in the permissible range of delays, a bifurcation of the form

$$\text{stable } (x_0, y_0) \rightarrow \text{unstable } (x_0, y_0) + \text{stable periodic solution} \quad (8)$$

becomes possible. Whether this bifurcation actually occurs depends on the form of the non-linearity in (3). The objective of this paper consists in ascertaining the conditions of such an occurrence.

PERIODIC SOLUTIONS ATTRIBUTABLE TO PURE DELAY

A preliminary step in the study of periodic solutions of (3) is the determination of purely imaginary roots s_1 of (5). Since (5) contains six parameters, it is expedient to examine a few special cases before drawing any general conclusions.

Consider first the case $\tau_3 = \tau_4 = 0$. The delays τ_1 and τ_2 appearing only in the combination $\tau = \tau_1 + \tau_2$, one of them may be omitted and the other replaced by τ . The requirement $s = i\omega$, $\omega = \text{real}$, leads to the condition that the two algebraic equations

$$-\omega^2 + bc + (1+y_0)^{-2} \cos \omega\tau = 0, \quad (b+c)\omega - (1+y_0)^{-2} \sin \omega\tau = 0 \quad (9)$$

resulting from (5), should admit at least one real root τ, ω . Such a root can only exist when $bcy_0^2 > 1$. This inequality is never satisfied, because (3) implies $1 = bcy_0(1+y_0) > bcy_0^2$. Constant delays τ_1 and τ_2 are therefore not a primary cause of periodic solution of (3) via the bifurcation (8), so τ_1, τ_2 can be omitted in what follows.

As a second special case consider $\tau_4 = 0, \tau_3 = \tau$. The algebraic equations analogous to (9) admit a real root $\omega = \omega_0, \tau = \tau_0$,

$$\tau_0 = \frac{\pi}{2\omega_0}, \quad \omega_0^2 = \alpha + \beta + [(\alpha + \beta)^2 + b^2c^2 - \beta^2]^{1/2}, \quad \alpha = \frac{1}{2}(c^2 - b^2), \quad (10)$$

for all b, c . An analysis of the critical case shows, however, that the bifurcation (8) does not take place, the equilibrium (x_0, y_0) becoming simply unstable when $\tau > \tau_0$. The bifurcation (8) is also absent when the non-linearity in (3) is given by (2), $m = 1, 2$, and $n = 0, 1, 2, \frac{1}{2}$. Hence $\tau_3 = \text{const}$ is also not a primary cause of periodic solutions of (3).

The third special case $\tau_3 = 0, \tau_4 = \tau$ is more favourable. A real root $\omega = \omega_0, \tau = \tau_0$ exists for all b, c , but instead of $\tau_0 = \pi/(2\omega_0)$ in (10), one has

$$\tau_0 = \frac{1}{\omega_0} \arctan \left[\frac{\omega_0}{bc} (\beta - b^2 - \omega_0^2) \right] \quad \text{modulo } \frac{\pi}{\omega_0} \quad (11)$$

For example, $b = 0.5, c = 0.1$ yields $\omega_0 = 0.074, \tau_0 = 33.4$. The bifurcation (8) occurs for a wide range of b, c , and $\tau \geq \tau_0$. Illustrative forms of the resulting periodic solutions are shown in Fig. 1 [a) in the phase plane $x-y$, b) in the phase plane $\dot{x}-\dot{y}$, and c) x and y as a function of t]. When $\tau/\tau_0 - 1 \ll 1$, the amplitude of these periodic solutions is a rather irregular function of the excess delay $\tau - \tau_0$.

The occurrence of the bifurcation (8) is strongly favoured by the presence of a variable delay depending on x, y . For example, when $\tau_4 = 0, \tau_3 = \tau, \tau_0$ given by (10) and

$$\tau = \tau_0 + \bar{\delta}_0 - \bar{\delta} x^2(t), \quad \bar{\delta}_0, \bar{\delta} > 0, \quad (12)$$

periodic solutions of (3) exist for a wide range of $\bar{\delta}_0, \bar{\delta}$. Illustrative forms are shown in Fig. 2 (the same representation as in Fig. 1 is used). For the same values

of b and c the oscillation periods in Fig. 2 are, however, much shorter than those in Fig. 1. When $\bar{\delta}_0, \delta \ll \tau_0$, the periodic solutions are almost sinusoidal. The bifurcation (8) occurs also when x^2 in (12) is replaced by y^2 or $x^2 + y^2$. In fact, any smooth function $g(x, y)$ will do, provided it assures a finite value of $\max \tau$ as x, y increase. Terms of an odd degree in the expansion of $g(x, y)$, linear ones included, have no effect on the amplitude limitation of the resulting periodic solutions. They merely cause some dissymmetry in the form of $x(t), y(t)$. In the context of reaction dynamics the presence of a variable delay component, such as $g(x, y)$ in (3), implies a transport time between the place of production and effect which depends on the product concentrations.

In the case of small $g(x, y)$ the amplitude and period of periodic solution of (3) can be expressed analytically by means of a Poincaré-type expansion. Consider, for example, $\tau_1 = \tau_2 = \tau_4 = 0, \tau_3 = \tau$ given by (12), and let $\mu > 0$ be a small parameter. The periodic solution is sought in the form

$$\left. \begin{aligned} \tau - \tau_0 &= \mu [\delta_0 - \delta x^2(t)] , & x(t) &= x_0 + \sum_{i=0}^{\infty} x_i(\bar{t}) \mu^i , & y(t) &= y_0 + \sum_{i=0}^{\infty} y_i(\bar{t}) \mu^i \\ \bar{t} &= \omega_0 t \sum_{i=0}^{\infty} h_i \mu^i , & \dot{y}(0) &= 0 , & L(\tau, y) &= \mu N(\tau, x, y) \end{aligned} \right\} (13)$$

where $N(\tau, x, y)$ is the non-linear part of (3), $L(\tau, y)$ is given by (4), h_i are undetermined constants, and $x_i(\bar{t}), y_i(\bar{t})$ undetermined periodic functions of period 2π . The substitution of (13) into (3), followed by a series expansion in powers of μ , leads to the usual linear recursive system

$$L(\tau_0, y_i) = f_i(y_0(\bar{t}), \dots, y_{i-1}(\bar{t})) , \quad i = 1, 2, \dots , \quad (14)$$

where the functions f_i are unambiguously defined. For $i = 0$ one obtains

$$y_0(\bar{t}) = A_0 \cos \bar{t} , \quad h_0 = 1 , \quad (15)$$

which is simply the eigenfunction of (4) corresponding to the critical eigenvalue (the so-called generating solution). The absence of secular terms in (14) yields, after some lengthy algebra,

$$A_0^2 = \frac{4}{3} \frac{\delta_0 + h_1 \tau_0}{\delta + \varepsilon_1} , \quad h_1 = \frac{2b \delta_0 \cos \omega_0 \tau_0 + \varepsilon_2}{3 - 2b \tau_0 \cos \omega_0 \tau_0 + \varepsilon_3} , \quad (16)$$

where $\varepsilon_1, \varepsilon_2, \varepsilon_3$ are some formally complicated but numerically small expressions. The equations (15) and (16) possess a qualitatively correct dependence on b, c , and τ , and for small $\bar{\delta}_0/\tau_0, \bar{\delta}/\tau_0$ they agree quite well with the directly computed periodic solutions of (3). For example, when $b = 0.5, c = 0.1, \delta_0 = 0.01 \tau_0, \delta = \sim 1.34$, the computed amplitudes of $y(t)$ differ from A_0 by less than 0.1%.

When other delays are introduced into (3) together with τ_3 , there is generally no qualitative change, the sole effect being a weak deformation of the periodic solutions. When both τ_3 and τ_4 are non-zero, there exists a small region in parameter space where (5) simultaneously admits two critical roots. After the "composite bifurcation" the solution of (3) is still oscillatory, but apparently no longer periodic.

CONCLUSION

When pure delays are introduced into the Goodwin model of the Jacob-Monod mode of gene regulation, self-sustained stable periodic oscillations are found to exist in a wide parameter range, the parameters characterizing reaction rates, product synthesis, and transport times. Constant and concentration-dependent delays are both found to be primary causes of periodic oscillations.

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Figure captions

Fig. 1: Periodic oscillations for the case of a constant delay

- a) in the phase plane $x-y$
- b) in the phase plane $\dot{x}-\dot{y}$
- c) x and y as a function of t .

Fig. 2: Periodic oscillations for the case of a variable delay

- a) in the phase plane $x-y$
- b) in the phase plane $\dot{x}-\dot{y}$
- c) x and y as a function of t .

Fig. 1 Periodic solution of (3), $\tau_1 = \tau_2 = \tau_3 = 0$, $\tau_4 = \tau$.

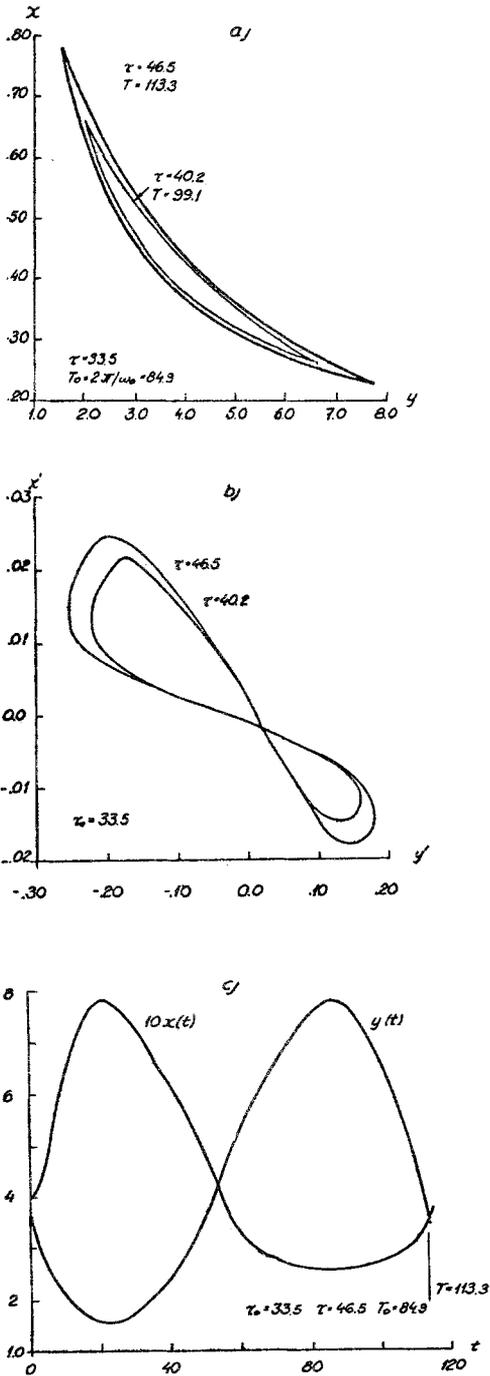
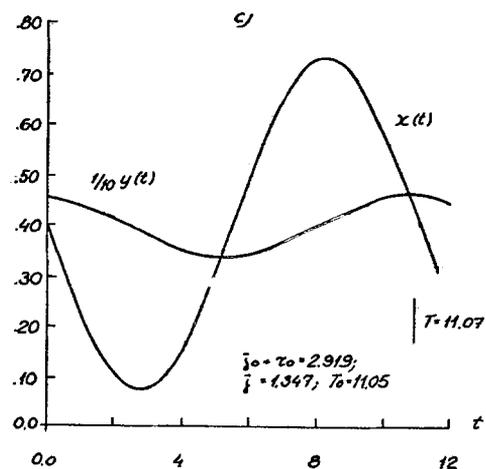
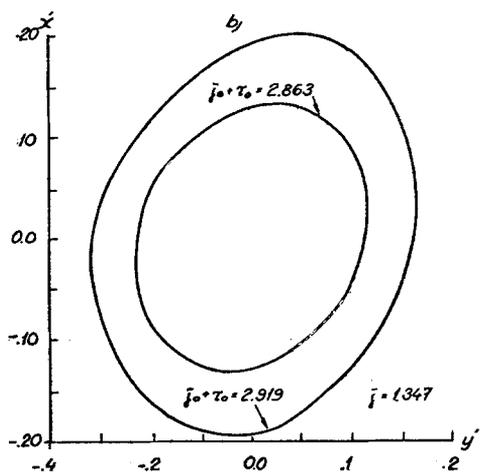
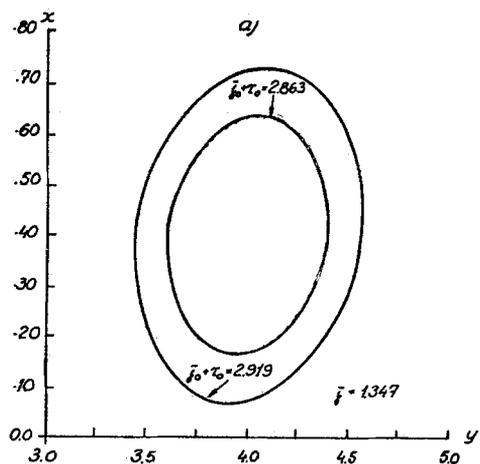


Fig. 2 Periodic solution of (3), $\tau_1 = \tau_2 = \tau_4 = 0$, $\tau_3 = \tau_0 + \bar{\delta}_0 - \bar{\delta}x^2(t)$



Note: The references cited are illustrative. No effort was made to assure completeness or a chronologically proper sequence.

- 1) B.P. Belousov, Sb. Ref. Radiat. Med. (1958), Moscow (1959), p. 145.
- 2) A.M. Zhabotinskii, Dokl. Akad. Nauk SSSR 157 (1964), p. 392.
- 3) M. Herschkowitz-Kaufman, CR Acad. Sci. C. 270 (1970), p. 1049.
- 4) B. Hess and A. Boiteux, Annu. Rev. Biochemistry 40 (1971), p. 237.
- 5) E.E. Selkov, Eur. J. Biochem. 4 (1968), p. 79.
- 6) J. Higgins, R. Frenkel, E. Hulme, A. Lucas and G. Rangazas, "Biological and Biochemical Oscillators", Academic Press (1973), p. 127.
- 7) W.A. Knorre, Biochem. Biophys. Res. Commun. 31 (1968), p. 812.
- 8) B.C. Goodwin, Eur. J. Biochem. 10 (1969), p. 515.
- 9) W.A. Knorre, "Biological and Biochemical Oscillators", Academic Press (1973), p. 425.
- 10) B.C. Goodwin, "Temporal organization in cells", Academic Press (1963).
- 11) B.C. Goodwin, Adv. Enz. Regul. 3 (1965), p. 425.
- 12) J.S. Griffith, J. Theoret. Biol. 20 (1968), p. 202 and 209.
- 13) J. Tiwari, A. Fraser and R. Beckman, J. Theoret. Biol. 39 (1973), p. 679, and 45 (1974), p. 311.
- 14) J. Monod and M. Cohen-Bazire, CR Acad. Sci. 236 (1953), p. 417 and p. 530.
- 15) J. Monod and F. Jacob, Cold Spring Harbour Symp. Quant. Biol. 26 (1961), p. 389.
- 16) P. Rapp, Bio. Systems 5 (1975), No 112.
- 17) Correspondence Ninio-Lodish, Nature 255 (29 May 1975), p. 429.