

**Cellular Control Models with Linked Positive and  
Negative Feedback and Delays.  
II. Linear Analysis and Local Stability**

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An analysis of local behavior is made of two nonlinear models which incorporate both an induction or positive feedback control mechanism and a repression or negative feedback control mechanism. The systems of differential equations with delays are linearized about their equilibria. The related characteristic equations which are exponential polynomials are studied to determine the local stability of the models. Computer studies are included to show the range of stability for different parameter values, and the biological significance is discussed briefly.

**1. Introduction**

In this paper we shall study the local behavior of the models developed in Mahaffy (1983). We shall concentrate our studies on the highly nonlinear model of induction and repression derived from the *lac* operon using enzyme kinetics and the simplified induction-repression model. The first is given by the system

$$\begin{aligned} \dot{z}_1(t) &= \frac{1}{(1 + (z_5(t - R_1))^{\gamma})} - b_1 z_1(t), \\ \dot{z}_2(t) &= \frac{z_1(t)(1 + K_2(z_4(t - R_2))^{\rho})}{(1 + K_3 z_1(t))(1 + K_4(z_4(t - R_2))^{\rho}) + K_5} - b_2 z_2(t), \\ \dot{z}_3(t) &= z_2(t) - b_3 z_3(t), \\ \dot{z}_4(t) &= z_3(t - R_3) - b_4 z_3(t) z_4(t). \\ \dot{z}_5(t) &= z_3(t) z_4(t) - b_5 z_5(t). \end{aligned} \tag{1}$$

The simplified induction-repression model is given by the system

$$\begin{aligned} \dot{z}_1(t) &= \frac{1}{1 + (z_5(t - T_1))^\gamma} - b_1 z_1(t), \\ \dot{z}_2(t) &= \frac{z_1(t)(1 + K_2(z_4(t - T_2))^\rho)}{1 + K_4(z_4(t - T_2))^\rho} - b_2 z_2(t), \\ \dot{z}_3(t) &= z_2(t) - b_3 z_3(t), \\ \dot{z}_4(t) &= z_3(t) - b_4 z_4(t), \\ \dot{z}_5(t) &= z_4(t) - b_5 z_5(t). \end{aligned} \tag{2}$$

To study local behavior the equilibria of equations (1) and (2) must be found. The systems of differential equations are then linearized about these equilibria. The local stability is then determined by an analysis of the characteristic equation, which is a complicated exponential polynomial. Several examples are analyzed with the aid of computer studies and the biological relevance is discussed.

## 2. Equilibria

In this section we shall determine the equilibria of the systems (1) and (2). Note that it can be easily shown for both equations (1) and (2) that the positive orthant is invariant, and in fact there is a bounded invariant region in the positive orthant. The latter follows from the nonlinear function in the first equation of each model being monotonically decreasing in  $z_5$ . Hence the important physical behavior is contained in a bounded region which includes all the equilibria.

The equilibria of the systems of delay equations are found by substituting the constant solution  $\bar{x} = (\bar{x}_1, \bar{x}_2, \bar{x}_3, \bar{x}_4, \bar{x}_5)^T$  into the equations with  $\dot{z}_i = 0$ . The resulting algebraic system of equations for (1) is given by the following:

$$\frac{1}{1 + (\bar{x}_5)^\gamma} = b_1 \bar{x}_1, \tag{3a}$$

$$\frac{\bar{x}_1(1 + K_2(\bar{x}_4)^\rho)}{(1 + K_3\bar{x}_1)(1 + K_4(\bar{x}_4)^\rho) + K_5} = b_2 \bar{x}_2, \tag{3b}$$

$$\bar{x}_2 = b_3 \bar{x}_3, \tag{3c}$$

$$\bar{x}_3 = b_4 \bar{x}_3 \bar{x}_4, \tag{3d}$$

$$\bar{x}_3 \bar{x}_4 = b_5 \bar{x}_5. \tag{3e}$$

From equation (3d) it is easily seen that either  $\bar{x}_3 = 0$  or  $\bar{x}_4 = 1/b_4$ . The former leads to a contradiction of  $\bar{x}$ s being an equilibrium solution, so  $\bar{x}_4 = 1/b_4$ . Hence,

$$\bar{x}_2 = b_3 b_4 b_5 \bar{x}_5 \quad \text{and} \quad \bar{x}_2 = \frac{\bar{x}_1 C_1}{b_2 (C_2 + C_3 \bar{x}_1)}$$

where  $C_1 = (1 + K_2(1/b_4)^\rho)$ ,  $C_2 = 1 + K_4(1/b_4)^\rho + K_5$  and  $C_3 = K_3(1 + K_4(1/b_4)^\rho)$ . From equation (3a) we see that  $\bar{x}_1 = 1/(b_1(1 + \bar{x}_5^\gamma))$ . Combining the above information we obtain

$$\bar{x}_5 = \frac{C_1}{b_1 b_2 b_3 b_4 b_5 (C_2 + (C_3/b_1) + C_2 \bar{x}_5^\gamma)}. \quad (4)$$

The right hand side of equation (4) is a function in  $x_5$  monotonically decreasing to zero, while the left hand side of equation (4) is the increasing function  $x_5$ ; hence there is a unique solution  $\bar{x}_5$  to equation (4). By substituting the unique value  $\bar{x}_5$  into the system (3), we can readily find the equilibrium solution  $\bar{x} = (\bar{x}_1, \bar{x}_2, \bar{x}_3, \bar{x}_4, \bar{x}_5)^T$ .

Similarly, the constant solution  $\bar{x} = (\bar{x}_1, \bar{x}_2, \bar{x}_3, \bar{x}_4, \bar{x}_5)^T$  can be substituted into equation (2) to give the following algebraic system of equations:

$$\frac{1}{1 + \bar{x}_5^\gamma} = b_1 \bar{x}_1, \quad (5a)$$

$$\frac{\bar{x}_1 (1 + K_2 \bar{x}_4^\rho)}{1 + K_4 \bar{x}_4^\rho} = b_2 \bar{x}_2, \quad (5b)$$

$$\bar{x}_{i-1} = b_i \bar{x}_i, \quad i = 3, 4, 5. \quad (5c)$$

From equation (5c) it follows that  $\bar{x}_2 = b_3 b_4 b_5 \bar{x}_5$ . Let  $B = b_1 b_2 b_3 b_4 b_5$ , then using equation (5) we see that

$$B \bar{x}_5 = \frac{1 + K_2 (b_5 \bar{x}_5)^\rho}{(1 + \bar{x}_5^\gamma)(1 + K_4 (b_5 \bar{x}_5)^\rho)}. \quad (6)$$

The left hand side of equation (6) is a linear function of  $x_5$ . However, it is impossible to determine whether or not the right hand side of the equation increases as a function of  $x_5$ . It is clear that at  $x_5 = 0$  the right hand side is equal to one, while as  $x_5 \rightarrow \infty$  the right hand side of equation (6) tends to zero, implying that there is at least one solution to equation (6). There can be as many as three equilibria satisfying equation (6) as we shall show later by example. For  $\rho$  and  $\gamma$  integers Descartes' Rule shows that there are at most three values for  $\bar{x}_5$ . As  $K_2 > K_4$ , the quantity  $(1 + K_2 (b_5 \bar{x}_5)^\rho)/(1 + K_4 (b_5 \bar{x}_5)^\rho)$  represents the familiar S-shaped saturation curve found in induction models. The term  $1/(1 + \bar{x}_5^\gamma)$  forms the monotonically decreasing

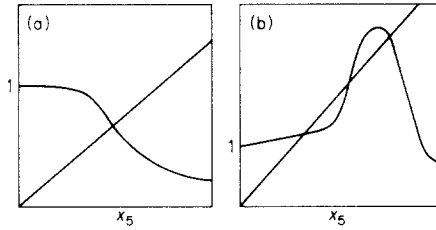


FIG. 1. Solution of equation (6).

repression term. The amount of induction compared to the amount of repression determines the number of equilibria (Fig. 1).

### 3. Linearization of the Models

In the previous section we found the equilibria of the models given by equations (1) and (2). In this section the linearization of the models will be developed and the corresponding characteristic equations shown. We shall begin with the analysis of system (1).

Let  $x_i = z_i - \bar{x}_i$ , where  $\bar{x} = (\bar{x}_1, \bar{x}_2, \bar{x}_3, \bar{x}_4, \bar{x}_5)^T$  is the equilibrium solution of equation (1), and define the following:

$$f(\xi) = \frac{1}{1 + (\xi + \bar{x}_5)^\gamma} - b_1 \bar{x}_1,$$

and

$$g(\xi, n) = \frac{(\xi + \bar{x}_1)(1 + K_2(\eta + \bar{x}_4)^\rho)}{(1 + K_3(\xi + \bar{x}_1))(1 + K_4(\eta + \bar{x}_4)^\rho) + K_5} - b_2 \bar{x}_2.$$

We can now write a system of differential equations equivalent to equations (1) in the following manner:

$$\begin{aligned} \dot{x}_1(t) &= f(x_5(t - R_1)) - b_1 x_1(t), \\ \dot{x}_2(t) &= g(x_1(t), x_4(t - R_2)) - b_2 x_2(t), \\ \dot{x}_3(t) &= x_2(t) - b_3 x_3(t), \\ \dot{x}_4(t) &= x_3(t - R_3) - b_4(x_3(t)\bar{x}_4 + \bar{x}_3 x_4(t) + x_3(t)x_4(t)), \\ \dot{x}_5(t) &= (x_3(t)\bar{x}_4 + \bar{x}_3 x_4(t) + x_3(t)x_4(t)) - b_5 x_5(t). \end{aligned} \tag{7}$$

In equation (7) the zero function is the unique equilibrium solution.

We shall consider the linearization of equation (7) about the solution  $x(t) = 0$ . With the following notation:

$$f'(0) = - \frac{\gamma \bar{x}_5^{\gamma-1}}{(1 + K_1(\bar{x}_5)^\gamma)^2},$$

$$g_1(0, 0) = \frac{\partial g}{\partial x_1}(0, 0) = \frac{(1 + K_5 + K_4 \bar{x}_4^\rho)(1 + K_2 \bar{x}_4^\rho)}{[(1 + K_3 \bar{x}_1)(1 + K_4 \bar{x}_4^\rho) + K_5]^2}$$

and

$$g_2(0, 0) = \frac{\partial g}{\partial x_4}(0, 0) = \frac{\rho \bar{x}_1 \bar{x}_4^{\rho-1} [(1 + K_3 \bar{x}_1)(K_2 - K_4) + K_2 K_5]}{[(1 + K_3 \bar{x}_1)(1 + K_4 \bar{x}_4^\rho) + K_5]^2}$$

we obtain the following characteristic equation for the linearization of equations (7) about the solution  $x(t) \equiv 0$ :

$$\det \begin{vmatrix} -b_1 - \lambda & 0 & 0 & 0 & f'(0)e^{-\lambda R_1} \\ g_1(0, 0) & -b_2 - \lambda & 0 & g_2(0, 0)e^{-\lambda R_2} & 0 \\ 0 & 1 & -b_3 - \lambda & 0 & 0 \\ 0 & 0 & -b_4 \bar{x}_4 + e^{-\lambda R_3} & -b_4 \bar{x}_3 - \lambda & 0 \\ 0 & 0 & \bar{x}_4 & \bar{x}_3 & -b_5 - \lambda \end{vmatrix} = 0$$

To evaluate this determinant we expand by the fourth row first. The remaining  $4 \times 4$  determinants can be expanded in any manner the reader chooses. The resulting characteristic equation can be expressed as follows:

$$(b_4 \bar{x}_4 - e^{-\lambda R_3})P_1(\lambda) - (b_4 \bar{x}_3 + \lambda)P_2(\lambda) = 0, \tag{8}$$

where  $P_1(\lambda) = -\{(b_1 + \lambda)(b_5 + \lambda)g_2(0, 0)e^{-\lambda R_2} + \bar{x}_3 g_1(0, 0)f'(0)e^{-\lambda R_1}\}$ , and  $P_2(\lambda) = (b_1 + \lambda)(b_2 + \lambda)(b_3 + \lambda)(b_5 + \lambda) - f'(0)g_1(0, 0)\bar{x}_4 e^{-\lambda R_1}$ . Equation (8) is a complicated exponential polynomial.

Now we shall consider the system (2). Let

$$h(\xi, n) = \frac{(\xi + \bar{x}_1)(1 + K_2(\eta + \bar{x}_4)^\rho)}{(1 + K_4(\eta + \bar{x}_4)^\rho)} - b_2 \bar{x}_2$$

and

$$x_i = z_i - \bar{x}_i,$$

where  $\bar{x} = (\bar{x}_1, \bar{x}_2, \bar{x}_3, \bar{x}_4, \bar{x}_5)^T$  is an equilibrium solution of equation (2). Recall that in this case there may be as many as three equilibria. A system of differential equations equivalent to equations (2) can be written as

follows:

$$\begin{aligned}
 \dot{x}_1(t) &= f(x_5(t - T_1)) - b_1 x_1(t), \\
 \dot{x}_2(t) &= h(x_1(t), x_4(t - T_2)) - b_2 x_2(t), \\
 \dot{x}_3(t) &= x_2(t) - b_3 x_3(t), \\
 \dot{x}_4(t) &= x_3(t) - b_4 x_4(t), \\
 \dot{x}_5(t) &= x_4(t) - b_5 x_5(t).
 \end{aligned}
 \tag{9}$$

In equation (9) the zero function is an equilibrium.

Linearizing equation (9) about the solution  $x \equiv 0$ , we obtain the following characteristic equation:

$$\det \begin{vmatrix} -b_1 - \lambda & 0 & 0 & 0 & f'(0) e^{-\lambda T_1} \\ h_1(0, 0) & -b_2 - \lambda & 0 & h_2(0, 0) e^{-\lambda T_2} & 0 \\ 0 & 1 & -b_3 - \lambda & 0 & 0 \\ 0 & 0 & 1 & -b_4 - \lambda & 0 \\ 0 & 0 & 0 & 1 & -b_5 - \lambda \end{vmatrix} = 0,$$

where

$$h_1(0, 0) = \frac{\partial h}{\partial x_1}(0, 0) = \frac{1 + K_2 \bar{x}_4^\rho}{1 + K_4 \bar{x}_4^\rho}$$

and

$$h_2(0, 0) = \frac{\partial h}{\partial x_4}(0, 0) = \frac{\rho \bar{x}_1 \bar{x}_4^{\rho-1} (K_2 - K_4)}{(1 + K_4 \bar{x}_4^\rho)^2}.$$

By expanding the determinant the characteristic equation can be written as follows:

$$\prod_{i=1}^5 (b_i + \lambda) - h_1(0, 0) f'(0) e^{-\lambda T_1} - (b_1 + \lambda)(b_5 + \lambda) h_2(0, 0) e^{-\lambda T_2} = 0.
 \tag{10}$$

Local stability of the systems (7) and (9) can be analyzed by determining whether any of the eigenvalues  $\lambda$  which satisfy equation (8) or (10) lie in the right half plane.

#### 4. A Brief Discussion of the Parameters

The model given by equations (1) has a large number of parameters. The system of equations (1) is a nondimensional model based on the highly nonlinear induction-repression model in Mahaffy (1983). The parameters  $\gamma$  and  $\rho$  are the Hill coefficients for their respective reactions. For the

example of the *lac* operon the parameter  $\rho$  represents the degree of cooperativity between allolactose and the repressor protein. The repressor protein is inactivated by two to four molecules of allolactose. Experiments of Zubay & Lederman (1969) and Yagil & Yagil (1971) give the value of  $\rho$  near two. The parameter  $\gamma$  is the degree of cooperativity of glucose binding to a repressor protein in the unknown repression or inhibition of c-AMP. Many known repression processes in prokaryotic cells involve a degree of cooperativity between one and two (Yagil & Yagil, 1971).

The  $b_i$ s are associated with the decay of the various chemical species. The nondimensionalized parameters  $b_i$  are inversely proportional to the half-lives of the particular chemical species, several of which are known for the *lac* operon. The half-life of the mRNA is known to be about two minutes (Beckwith & Zipser, 1979). The  $\beta$ -galactosidase and  $\beta$ -galactoside permease are stable (though there appears to be some loss of activity with age (Rotman, 1970)), which implies the decrease in concentration of these enzymes is mainly due to dilution by cell growth. The half-life is then approximately equal to the cell doubling time which is about 50 minutes. The value for the parameter  $b_4$  is more difficult to determine because of the second order kinetics; however, it should be relatively large because of the rapid turnover rate of lactose inside the cell.  $b_5$  is proportional to the rate of utilization of glucose which should have a large value as glucose has a rapid turnover rate. The linear decay term used in the model could be improved by using a nonlinear Michaelis-Menten velocity term. Recently there have been investigations of the Goodwin repression models with the endproduct satisfying this type of decay (Painter & Tyson, 1982); however, we shall not investigate this effect here.

The decay  $R_2$  can be estimated as seen in Banks & Mahaffy (1979).  $R_2$  is a delay representing the sum of the delays for transcription and translation. Recall that translation in prokaryotes begins before the process of transcription is completed. The delay for transcription and translation with coupling considered is between one and two minutes.  $R_3$  is a delay accounting for transport across the cellular membrane. The delay  $R_1$  is more involved as it includes the sum of the delays for transcription and translation and a delay for the catabolite repression of c-AMP which is more difficult to estimate.

Finally, we must consider the  $K_i$ s. These parameter values are unknown. The  $K_i$ s determine the magnitude of the effects of induction and repression, so they are sensitive to any scaling. The only information we have is that  $K_2 > K_4$  in order that there be induction. The induced cell produces enzymes at a rate 1000 times greater than the basal rate (Gilbert & Muller-Hill, 1970).

The above discussion applies to the simplified induction-repression model given by equations (2), except for the part concerning  $b_4$ . Also, the delays are given by  $T_1 = R_1 + R_3$  and  $T_2 = R_2 + R_3$ .

### 5. Numerical Studies and Local Stability

In Section 2 the equilibria were determined for the models (1) and (2), then in Section 3 the models were linearized about their equilibria. Model (1) has a unique equilibrium. The local stability of the equilibrium can be studied by analyzing the characteristic equation (8). For model (2) there is a possibility of more than one equilibrium. The local stability of each equilibrium can be studied by an analysis of the characteristic equation (10). Below we shall present stability results concerning the different models and discuss the biological significance of these results.

In the model given by equation (7) the parameter  $R_3$  is a very small delay, so as a first approximation we shall let  $R_3 = 0$ . With this assumption the characteristic equation reduces to  $(b_4 \bar{x}_3 + \lambda)P_2(\lambda) = 0$ .  $P_2(\lambda)$  is of the form given in Theorem 1 of Mahaffy (1982). In the proof of this theorem a technique is given to determine the critical value  $r_0$  at which the eigenvalues cross the imaginary axis. For  $R_1 > r_0$ , the system of equations (7) is locally unstable. Table 1 gives examples of how different values of the parameters affect the stability of equations (7). In Table 1 the following values of  $b_2 = 0.5$ , and  $b_3 = 0.02$  are kept constant.

The values of  $r_0$  given in Table 1 represents how large the parameter  $R_1$  must be to destabilize the equilibrium of equations (7) for a particular set of parameter values. From Table 1 it is easily seen that the parameter that most affects the stability is  $\gamma$  which occurs in the part of the biological problem that is least understood. The  $b_i$ s, especially  $b_4$ , appear to have the second largest effect on the stability of the system (7). The stability of the system (7) is not as sensitive to changes in the parameters  $K_i$  and  $\rho$ . Numerical integration of the model given by equations (7) with parameter values such that the equilibrium is locally unstable indicate the existence of periodic solutions. A rigorous proof of their existence remains an open questions. Damped oscillations have been observed in *E. coli* by Knorre (1968, 1973).

If the parameter  $R_3$  is not equal to zero the exponential polynomial given by equation (8) is substantially more difficult to analyze. The theorems of Mahaffy (1982) do not apply; however, the contour  $C$  with  $\pm i\pi/R_1$  as the bounds of the imaginary part in that paper can be used for analyzing the stability of specific examples. Consider the model where the parameter values are given by the first line of Table 1. For  $R_3 = 0$  the eigenvalues



TABLE 1

The equilibrium value  $\bar{x}_5$  and the critical value of the delay  $R_1 = r_0$  where the eigenvalues cross the imaginary axis are tabulated for different parameter values for the system (7)

$b_1$	$b_4$	$b_5$	$K_2$	$K_3$	$K_4$	$K_5$	$\gamma$	$\rho$	$\bar{x}_5$	$r_0$
1	1	1	100	1	1	0	2	2	17.1	57.4
1	1	10	100	1	1	0	2	2	7.88	60.8
10	1	10	100	1	1	0	2	2	3.60	67.2
0.1	1	1	100	0.1	0.1	0	2	2	45.1	53.1
1	0.1	1	100	0.1	0.1	0	2	2	96.9	56.8
1	1	0.1	100	0.1	0.1	0	2	2	45.1	53.1
0.1	0.1	0.1	100	0.1	0.1	0	2	2	450	48.8
0.1	0.1	0.1	1	0.1	0.01	0	2	2	172	48.8
1	1	1	100	0.1	1	0	2	2	17.1	57.0
1	1	1	100	10	1	0	2	2	16.9	60.6
1	1	1	100	0.01	1	0	2	2	17.1	57.0
1	1	1	1000	0.1	1	0	2	2	36.8	56.8
1	1	1	100	0.1	0.1	0	2	2	20.9	57.0
1	10	1	100	0.1	0.1	0	2	2	2.58	74.9
1	100	1	100	0.1	0.1	0	2	2	0.659	$\infty$
1	10	10	100	0.1	0.1	0	2	2	0.975	$\infty$
1	1	1	100	0.1	0.1	0	2	4	20.9	57.0
1	1	1	100	0.1	0.1	0	4	4	6.20	20.1
1	1	1	100	0.1	0.1	0	4	2	6.20	20.1
1	1	1	100	0.1	0.1	0	1	2	95.3	$\infty$
1	1	1	100	0.1	0.1	10	2	2	9.66	57.8
1	1	1	100	0.1	0.1	-0.5	2	2	25.6	57.0
1	1	1	100	0.1	0.1	100	2	2	4.57	61.6

cross the imaginary axis near  $R_1 = 57.4$ . When  $R_3 = 0.5$  the critical value of  $R_1$  is 57.0 in order that the equilibrium becomes locally unstable. For  $R_3 = 1$  the critical value of  $R_1$  is 56.6 and for  $R_3 = 5$  the critical value of  $R_1$  is 54.1. For this analysis we assumed that  $R_2 = R_1$  and then considered the first part of equation (8) a perturbation of the exponential polynomial  $P_2(\lambda)$ . Our studies indicate that the delay  $R_3$  destabilizes the system (7), i.e., as the value of  $R_3$  increases the value of the delay  $R_1$  needed to have the eigenvalues in the right half plane decreases.

We have stated without proof that  $R_3 > 0$  destabilizes the system (7). A heuristic argument using the argument principle or Nyquist criterion is given below. Let  $\lambda$  traverse the contour  $C$  of Mahaffy (1982) in the counterclockwise direction with  $\pm i\pi/R_1$  as the bounds of the imaginary part and map  $(b_4\bar{x}_3 + \lambda)P_2(\lambda)$ . This case is analyzed in Mahaffy (1982). From equation

(8) we see that for stability the map

$$F(\lambda) = (b_4 \bar{x}_3 + \lambda) P_2(\lambda) - [1 - \exp(-\lambda R_3)] P_1(\lambda)$$

must be considered. The term  $-[1 - \exp(-\lambda R_3)] P_1(\lambda)$  is treated as a perturbation of  $F(\lambda)$ , assuming  $R_3$  is small. For  $\lambda = i\nu$  with  $\nu > 0$  small it can be shown that the above perturbation shifts the map initially to the right of the contour of the unperturbed or  $R_3 = 0$  system, i.e., initially the angle  $\theta$  between the positive real axis and the line segment  $\overline{F(0)F(i\nu)}$  is smaller for the perturbed system than the unperturbed system,  $0 < \theta < 2\pi$ . By applying the techniques of Mahaffy (1982) one can compare the maps of the unperturbed and perturbed systems and see that the shift changes the relative position of the map  $F(\lambda)$  so as to destabilize the system (7).

The model given by the system of equations (9) has the possibility of multiple equilibria. For this study the parameters  $b_2$  and  $b_3$  were fixed at 5 and 0.2 respectively. Table 2 shows how the equilibria and the stability of the equilibria change as the values of the parameters change. From Table 2 it is seen that there are five basic types of behavior. Figures 2-5 illustrate four of these basic types. The figures are obtained by integrating the system of equations (9) with a fourth order scheme for delay equations and a stepsize  $h = 0.2$ . Figures 2 and 3 use the parameter values given in line 4 of Table 2. The trajectories were formed by starting near each of the equilibria with constant initial functions. The initial values for each of the four trajectories starting from the bottom and working up are  $x_5 = x_2 = x_4 = 0.04, 0.236, 0.238$ , and  $1.0$ ,  $x_1 = 0.1995, 0.1894, 0.1914$ , and  $0.1$ , and  $x_3 = 0.25, 1.18, 1.182$ , and  $5$ . In Fig. 2 the delay  $T_1 = 2$  which from Table 2 gives two stable equilibria and one unstable equilibria. (Note that the middle equilibrium point is always unstable.) In this case all solution trajectories of equation (9) approach the two stable equilibria. In Fig. 3 the delay  $T_1 = 9$  which from Table 2 gives one stable equilibrium and two unstable equilibria. In this case the solution trajectories either asymptotically approach the lower stable equilibrium or they oscillate about the upper unstable equilibrium. It appears that in the latter case the trajectories asymptotically approach a stable periodic solution though this has not been proved. A third type of behavior could be illustrated by considering equations (9) with parameter values from line 2 of Table 2 and  $T_1 = 750$ . For technical reasons this case was not integrated though the expected behavior is two attracting periodic trajectories about the upper and lower unstable equilibria with the upper trajectory having a much greater amplitude.

The other two types of behavior are shown in Figs 4 and 5. These figures use the parameter values given in line 8 of Table 2. The trajectories were formed by beginning at  $x_5 = x_2 = x_4 = 2.5$ ,  $x_1 = 0.1379$ , and  $x_3 = 12.5$  which

TABLE 2

The equilibria values  $\bar{x}_5$  and the critical values of the delay  $R_1 = r_0$  where the eigenvalues cross the imaginary axis are tabulated for different parameter values for the system (9)

$b_1$	$b_4$	$b_5$	$K_2$	$K_4$	$\gamma$	$\rho$	$\bar{x}_5$	$r_0$
5	3.5	1	100	1	2	2	1.33	6.36
5	3.92	1	100	1	2	2	{ 1.23 0.108 0.101	{ 6.52 0 726
5	4	1	100	1	2	2	{ 1.21 0.130 0.084	{ 6.6 0 $\infty$
5	5	1	100	1	2	2	{ 1.01 0.236 0.050	{ 7.13 0 $\infty$
5	6	1	100	1	2	2	{ 0.809 0.349 0.038	{ 14.3 0 $\infty$
5	6.695	1	100	1	2	2	{ 0.568 0.529 0.033	{ 82.9 0 $\infty$
5	7	1	100	1	2	2	0.0313	$\infty$
1	5	1	100	1	2	2	2.45	5.03
5	5	2	100	1	2	2	{ 0.862 0.110 0.025	{ 15.7 0 $\infty$
5	5	1	50	1	2	2	0.0436	$\infty$
5	5	1	200	1	2	2	1.61	5.94
5	5	1	100	0.5	2	2	{ 1.36 0.225 0.050	{ 8.42 0 $\infty$
5	5	1	100	2	2	2	{ 0.666 0.271 0.049	{ 15.7 0 $\infty$
5	5	1	100	1	1	2	{ 1.02 0.351 0.046	{ 16.1 0 $\infty$
5	5	1	100	1	1	4	{ 1.29 0.980 0.039	{ 17.9 0 $\infty$
5	5	1	100	1	2	4	{ 1.06 0.9309 0.040	{ 19.7 0 $\infty$

is near the unique equilibrium. In Fig. 4 the delay  $T_1 = 3$  and we see that the trajectory asymptotically approaches the equilibrium solution. In Fig. 5 the delay  $T_1 = 6$  which from Table 2 implies that the equilibrium is

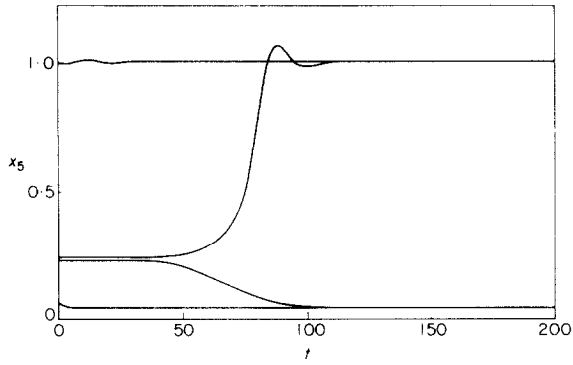


FIG. 2. Trajectories for the model with three equilibria and  $T_1 = 2$ .

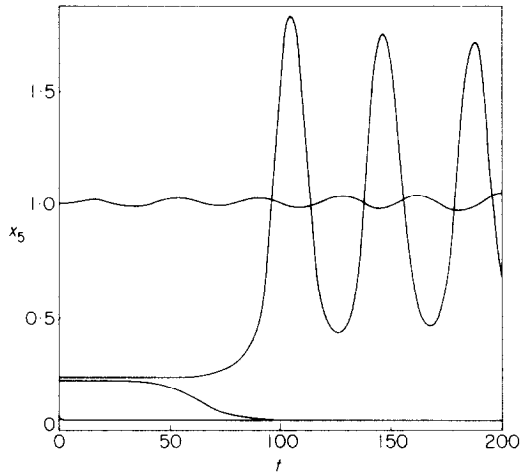


FIG. 3. Trajectories for the model with three equilibria and  $T_1 = 8$ .

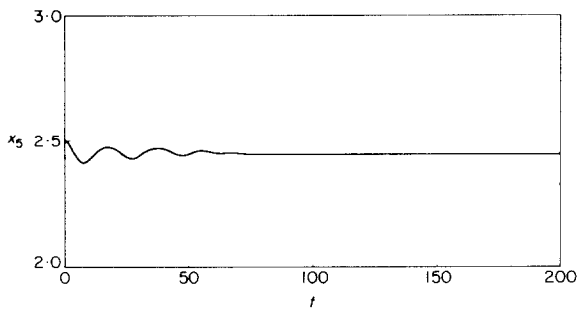


FIG. 4. Trajectory for the model with one equilibrium and  $T_1 = 3$ .

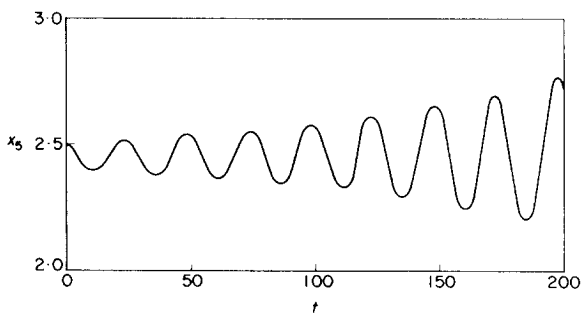


FIG. 5. Trajectory for the model with one equilibrium and  $T_1 = 6$ .

unstable. In this case the trajectory oscillates with increasing amplitude about the equilibrium solution.

In order to study the stability of this system the characteristic equation (10) is used. As in our study of the system (7) we shall first eliminate one of the delays in order to analyze equation (10). Let  $T_2 = 0$ . By making this simplification equation (10) reduces to the following equation:

$$\begin{aligned} H(\lambda, T_1) &= (b_1 + \lambda)(b_5 + \lambda)[(b_2 + \lambda)(b_3 + \lambda)(b_4 + \lambda) - h_2(0, 0)] \\ &\quad + A e^{-\lambda T_1} \\ &= P(\lambda) + A e^{-\lambda T_1} \end{aligned} \quad (11)$$

where  $A = -h_1(0, 0)f'(0)$ . If  $b_1 b_5 (b_2 b_3 b_4 - h_2(0, 0)) < A$ , then when  $b_2 b_3 b_4 - h_2(0, 0) > 0$ , the Routh-Hurwitz criterion (Lancaster, 1969) can be applied to  $P(\lambda)$  to show that  $H(\lambda, T_1)$  in equation (11) satisfies the conditions of Theorem 2 of Mahaffy (1982). In the proof of that theorem a technique is given for determining the bifurcation points (which may not be unique). In particular, the proof of the theorem gives a method for finding the bifurcation point  $r_0$  for which all values of  $T_1 > r_0$  result in local instability.

For the cases in Table 2 where  $b_2 b_3 b_4 - h_2(0, 0) > 0$  and  $b_1 b_5 (b_2 b_3 b_4 - h_2(0, 0)) > A$ , consider  $|P(-i\nu)|$  where  $\nu$  is real and  $P$  is defined as in equation (11). It can be shown that  $|P(-i\nu)|$  is monotonically increasing as a function of  $\nu$  for the examples in Table 2, so a proof similar to the one given for Theorem 1 of Mahaffy (1982) has all solutions of  $H(\lambda, T_1) = 0$  with  $\text{Re } \lambda < 0$ , i.e., the system is locally stable. In Table 2 these cases are denoted by  $r_0 = \infty$ .

When  $b_2 b_3 b_4 - h_2(0, 0) < 0$ , the Routh-Hurwitz criterion can be applied to  $P(\lambda)$  to see that  $P(\lambda)$  has one real positive  $\lambda$  satisfying  $P(\lambda) = 0$ . By differentiating the function  $|P(-i\nu)|$  with respect to  $\nu$ , it can be shown that  $|P(-i\nu)|$  is monotonically increasing in  $\nu$  ( $\nu \geq 0$ ). Again consider the contour  $C$  of Mahaffy (1982) with  $T_1 = r$ . If  $b_1 b_5 (h_2(0, 0) - b_2 b_3 b_4) > A$ , then as in

the proof of Theorem 1 of Mahaffy (1982) we see that  $H$  has the same orientation relative to the origin as  $P$  which implies  $H$  encircles the origin once. This implies  $H(\lambda, T_1) = 0$  has one root with  $\text{Re } \lambda > 0$  (actually,  $\lambda > 0$  real) independent of  $T_1$ . This is denoted  $r_0 = 0$  in Table 2. However, if  $b_1 b_5 h_2(0, 0) - b_2 b_3 b_4 < A$ , then initially  $P(0)$  and  $H(0, T_1)$  align with the origin in between. Since  $|P(-i\nu)|$  is monotonically increasing and tends to infinity as  $\nu \rightarrow \infty$ , there exists a value  $\nu_0$  such that  $|P(-i\nu_0)| = A$ . At  $\nu_0$  calculate the value of  $\arg P(-i\nu_0)$ . Note that  $\arg P(0) = \pi$ , and  $\arg P(-i\nu)$  initially increases monotonically to some maximum less than  $3\pi/2$  before decreasing monotonically to  $\pi/2$  as  $\nu \rightarrow \infty$ . If  $\arg P(-i\nu_0) \leq \pi$ , then using the contour  $C$  for any  $T_1 > 0$  will give at least two encirclements of the origin. (There are no examples of this case in Table 2.) If  $\arg P(-i\nu_0) > \pi$ , then by solving  $r_0 \nu_0 = \arg P(-i\nu_0) - \pi$  for  $r_0$  or

$$r_0 = \frac{\arg P(-i\nu_0) - \pi}{\nu_0},$$

we obtain the critical value  $r_0$  where two eigenvalues cross the imaginary axis, i.e.,  $H(\pm i\nu_0, r_0) = 0$ . For  $T_1 > r_0$ , a geometric argument similar to those in Mahaffy (1982) give two eigenvalues with  $\text{Re } \lambda > 0$ .

The above arguments show how we arrived at the values for  $r_0$  in Table 2. For the system of equations (9) the approximation  $T_2 = 0$  is not a good approximation. Analysis of equation (10) with  $T_2 \neq 0$  has been studied only for specific cases. For comparison with Fig. 5 an example with parameter values from line 8 of Table 2 and the same initial data is considered where the delays  $T_1 = 6$  and  $T_2 = 3$ . The trajectory from integrating this example is shown in Fig. 6. Unlike Fig. 5, this example shows slowly damped oscillations and in fact the equilibrium is attracting.

A more thorough analysis of the regions of stability is given for the example with parameter values from line 8 of Table 2 and with different

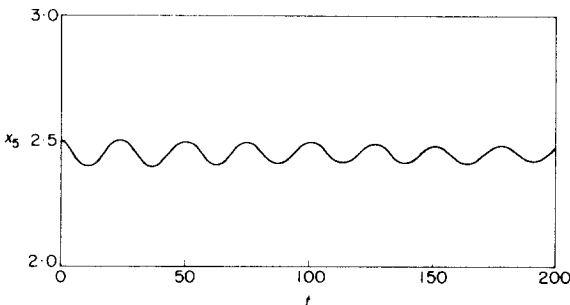


FIG. 6. Trajectory for the model with one equilibrium and  $T_1 = 6$ ,  $T_2 = 3$ .

values of the delay  $T_2$ . Again our analysis is based on work with the contour  $C$  of Mahaffy (1982) with  $T_1 = r$ . We find the value  $r_0$  in a similar manner to the proofs of the theorems in that paper with

$$P(\lambda) = \prod_{i=1}^5 (b_i + \lambda) - (b_1 + \lambda)(b_5 + \lambda)h_2(0, 0) e^{-\lambda T_2}$$

and

$$Q(\lambda) = h_1(0, 0)f'(0) e^{-\lambda T_1}.$$

With a fixed value of  $T_2$ , the critical value  $r_0$  of  $T_1$  can be determined where the eigenvalues from the characteristic equation (10) cross the imaginary axis. For example when  $T_2 = 0$ ,  $r_0 = 5.02$ , when  $T_2 = 3$ ,  $r_0 = 6.36$ , and when  $T_2 = 6$ ,  $r_0 = 7.88$ . A region of stability can be determined in the  $T_1 T_2$ -plane using the above information. Figure 7 shows the region of stability for the above example. Note that from the biological problem we must consider  $T_2 \leq T_1$  only.

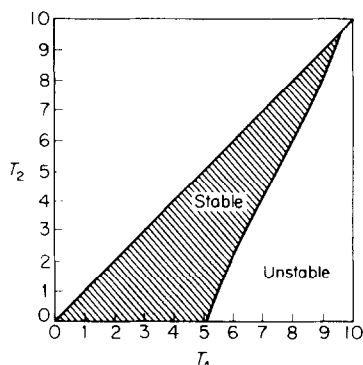


FIG. 7. Region of stability for equation (9) with respect to the delays  $T_1$  and  $T_2$ .

It is interesting to see that increasing values of  $T_2$  result in increasing critical values  $r_0$  for  $T_1$ . The perturbation caused by the delay  $T_2$  is thus a stabilizing one. From the biological argument one sees that an increase in the delay  $T_2$  increases the delay time for transcription and translation in the induction process but for constant  $T_1$  increasing  $T_2$  decreases the delay time for the repression of  $x_1$ (c-AMP) by  $x_5$  (glucose). This suggests that the unknown negative feedback process is perhaps the most important part of the model that contributes to the observed oscillations. Our study considered only a few examples and leaves room for further investigation.

Another case of particular interest from the biological point of view is when  $\gamma = 1$  for equation (9). From Table 2 line 14 we see that for  $T_1 > 16.1$

the system (9) becomes unstable. For this case the system has a degree of cooperativity of one in the repression part and can become locally unstable for sufficiently large delays. The main interest of this result is that without the induction term the model reduces to the Goodwin model with delays which was shown to be globally asymptotically stable independent of the delays (see Allwright, 1977; Banks & Mahaffy, 1978).

From our studies above we have shown that the induction-repression model (7) may have a locally unstable equilibrium for biologically significant parameter values, but that the model is highly sensitive to certain parameters involved in the unknown reactions leading to catabolite repression. In the simplified induction-repression model given by equation (9) there may exist three equilibria. Biologically, this could mean that a certain threshold of initial concentrations are needed to start the induction process with the smallest equilibrium being the intracellular steady-state at low concentrations. For some parameter values we have shown that the model (9) is locally unstable. The model (9) was shown to be locally unstable for some delays when  $\gamma = 1$ , which is significant considering several known repression processes in prokaryotic cells. The above information could be useful in explaining observed epigenetic oscillations in prokaryotic cells.

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