Pis'ma v ZhETF, vol. 88, iss. 4, pp. 321 – 325

Amplification of kinetic oscillations in gene expression

V. P. Zhdanov[1)]

Department of Applied Physics, Chalmers University of Technology, S-412 96 Göteborg, Sweden

Boreskov Institute of Catalysis, Siberian Branch RAS, 630090 Novosibirsk, Russia

Submitted 7 May 2008

Resubmitted 7 June 2008

Because of the feedbacks between the DNA transcription and mRNA translation, the gene expression in cells may exhibit bistability and oscillations. The deterministic and stochastic calculations presented illustrate how the bistable kinetics of expression of one gene in a cell can be influenced by kinetic oscillations in expression of another gene. Due to stability of the states of the bistable kinetics of gene 1 and relatively small difference between the maximum and minimum protein amounts during oscillations of gene 2, the induced oscillations of gene 1 are found to typically be related either to the low- or high-reactive state of this gene. The quality of the induced oscillations may be appreciably better than that of inducing oscillations. This means that gene 1 can serve as an amplifier of kinetic oscillations of gene 2.

PACS: 05.40.−a, 87.16.−b

The expression of the information encoded in genes is known to occur via a templated polymerization called transcription, in which the genes are used as templates to guide the synthesis of shorter molecules of RNA [1]. Later on, many of RNAs, or, more specifically, messenger RNAs (mRNA) serve to direct the synthesis of proteins by ribosomes. Another large class of RNA includes nonprotein coding RNAs (ncRNA) [2]. The functions of the latter RNAs are based on their abilities to bind to and modulate the activity of mRNAs and/or proteins [2]. The whole process of gene expression can be regulated at all the steps. Specifically, the gene transcription, performed by RNA polymerase (RNAP) during its association with DNA, is often controlled by master regulatory proteins. Such proteins associate with DNA and either facilitate or suppress the RNA synthesis.

The positive and negative feedbacks between mRNA and protein production related to the same gene or different genes may result in complex kinetic behaviour including bistability [3 – 6] and oscillations [7 – 9]. Mathematically, these phenomena can be described by using conventional mean-field (MF) kinetic equations. In cells, most genes exist at single or low copy numbers, the number of mRNA and regulatory protein copies is often low [10], and accordingly the gene-transcription kinetics may exhibit stochastic features, e.g., transcriptional bursts related to bistability [3 – 5]. In experiments, the mRNA and protein expression has traditionally been characterized by using average data for cell populations and accordingly the stochastic and oscillatory features have often been smeared. At present, the stochastic bursts can however be directly observed (see e.g. experiments with mRNA [11] and proteins [12]).

The understanding of the general mechanistic principles behind temporal bistable and oscillatory kinetics of gene expression resulting in the formation of mRNAs and proteins is now relatively complete (for temporal oscillatory kinetics including ncRNA, see Ref. [9]; for spatio-temporal simulations, see Ref. [13]). The interplay between these two kinetic phenomena has however not been analyzed. In this Letter, we show how the bistable kinetics can be influenced by kinetic oscillations. This problem is of considerable intrinsic interest and may also be important for understanding the biological functions of oscillations (at present, this subject is open for debate [14]).

To motivate our study in more detail, we note that kinetic oscillations are possible in the case of transcription of a single gene provided that the feedback between mRNA and protein production is negative and the suppression of the mRNA production is delayed due to a few steps of protein conversion [7, 8]. Taking into account that the feedbacks between mRNA and protein are abundant and the protent conversion (e.g., phosphorylation) is quite common as well, this mechanism of oscillations is often considered to be the most likely. Although in this case the role of the protei-conversion-related delay in oscillations is constructive, it simultaneously damps the feedbacks between different steps and the response of a system to perturbations. For this reason, during the oscillations under consideration, the relative changes of the numbers of mRNA and protein copies are usually

[1)] e-mail: zhdanov@fy.chalmers.se

relatively small. In particular, the ratio of the minimum and maximum numbers is typically ≥ 0.5. For bistable kinetics, in contrast, this ratio is often close to zero. One of the likely ways to amplify oscillations is to regulate bistable kinetics of expression of one gene (gene 1) by protein formed during the oscillatory regime of expression of another gene (gene 2). Our goal is to explore the feasibility of this scenario. First, we recall typical bistable and oscillatory kinetics and then show the effect of oscillations on bistability. Basically, we study the induction of oscillations in the kinetics of expression of gene 1 by gene 2. Our analysis below does not include the effect of gene 1 on gene 2. With this effect, the oscillations would of course be improved as well. The physics behind oscillations in the expression of gene 2 will however be changed, and the phenomenon can hardly be qualified as amplification.

The simplest models predicting bistability in gene expression include (i) a single gene with positive feedback between mRNA and regulatory-protein production or (ii) two genes with mutually negative feedbacks between mRNAs and regulatory-protein production. In our analysis, we use a model of the former type. In particular, we assume that the mRNA (R) production is activated if n regulatory sites of the gene are occupied by protein (P), and the P-gene association and dissociation are rapid and close to equilibrium. In this case, the conventional MF kinetic equations for the R and P numbers in a cell are as follows [5, 6]

$$\frac{dN_R}{dt} = k_0 + k_1\left(\frac{N_P}{K_P + N_P}\right)^n - k_R N_R, \qquad (1)$$

$$\frac{dN_P}{dt} = k_s N_R - k_P N_P. \qquad (2)$$

The first two terms in the right-hand part of Eq. (1) represent the R-production rate (k_0 and k_1 are the rate constants of the basal and P-regulated gene transcription, $(N_P/(K_P + N_P))^n$ is the probability that all the regulatory sites are occupied by P, due to this factor the feedback between the R and P production is positive, and K_P is the P association-dissociation constant). The third term in the right-hand part of Eq. (1) takes the R degradation into account (k_R is the degradation rate constant). The two terms in the right-hand part of Eq. (2) describe the P synthesis and degradation, respectively (k_s and k_R are the corresponding rate constants).

In cells, the intracellular biochemical reactions, e.g., protein and RNA degradation, often occur on the time scale from 1 to 10 min or faster [3]. This time scale is much shorter than the duration of the cell cycle. For this reason, one can often consider that the intracellular biochemical reactions in general and the gene transcription in particular occur under steady-state conditions. In our context, this means that all the parameters in Eqs. (1) and (2) can be considered to be independent of time and in the absence of perturbations one can use $dN_R/dt = dN_P/dt = 0$. In this case, Eq. (2) yields $N_P = (k_s/k_P)N_R$. Substituting this expression into Eq. (1), one obtains a non-liner equation for N_R. With appropriate kinetic parameters (in our examples below we employ typical biologically reasonable parameters chosen in line with Ref. [3]), it predicts bistability (Fig.1) provided that $n \geq 2$, and accordingly, with

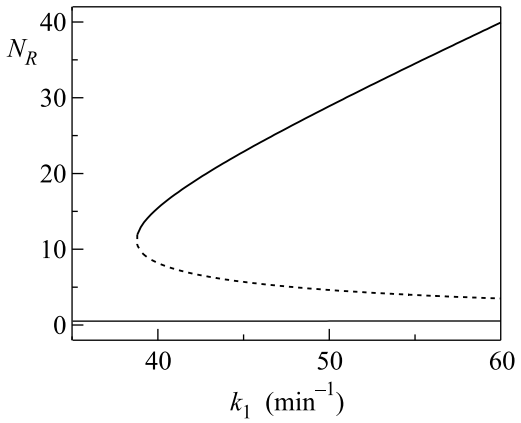

Fig.1. Number of R as a function of k_1 according to Eqs. (1) and (2) under steady-state conditions for $n = 3$, $K_P = 30$, $k_0 = 0.5\,\mathrm{min}^{-1}$, $k_R = 1\,\mathrm{min}^{-1}$, $k_s = 10\,\mathrm{min}^{-1}$, and $k_P = 2\,\mathrm{min}^{-1}$. The thick solid and dashed lines correspond respectively to the stable and unstable steady states. For comparison, note that in this case the P population is $N_P = (k_s/k_P)N_R = 5N_R$ (see Eq. (2))

increasing and subsequent decreasing one of the parameters, one can observe a hysteresis loop with stepwise jumps from one stable solution to another. Mathematically, these jumps represent a saddle-node bifurcation. (For discussion of the effect of the cell growth on such kinetics, see Ref. [6].)

As already noted in the introduction, kinetic oscillations in the gene expression can be observed provided that the feedback between mRNA and protein synthesis is negative and the suppression of the mRNA production is delayed due to protein conversion from one form to another form [7, 8]. We analyze one of the simplest generic models of this type, including production of protein P_1 by mRNA (M), conversion of P_1 to P_2 and then to P_3, and suppression of the M production by P_3. In particular, the M production is assumed to occur provided that m regulatory sites are free of P_3. The corresponding MF

kinetic equations for the M, P_1, P_2 and P_3 populations in the cell are as follows [8]:

$$\frac{dN_M}{dt} = \kappa_t \left(\frac{K_{P3}}{K_{P3} + N_{P3}}\right)^m - \kappa_M N_{\mathcal{R}}, \qquad (3)$$

$$dN_{P1}/dt = \kappa_s N_M - (\kappa_{12} + \kappa_{P1}) N_{P1}, \qquad (4)$$

$$dN_{P2}/dt = \kappa_{12} N_{P1} - \kappa_{23} N_{P2}, \qquad (5)$$

$$dN_{P3}/dt = \kappa_{23} N_{P2} - \kappa_{P3} N_{P3}, \qquad (6)$$

where κ_t is the rate constant of the P_3-regulated gene transcription, $[K_{P3}/(K_{P3} + N_{P3})]^m$ is the probability that all the regulatory sites are free of P_3, K_{P3} is the P_3 association-dissociation constant, κ_s is the rate constant of P_1 synthesis, κ_{12} and κ_{23} are the P_1 and P_2 conversion rate constants, and κ_M, κ_{P1} and k_{P3} are the degradation rate constants (the P_2 degradation is neglected in order to reduce the number of model parameters). Typical oscillatory kinetics predicted by Eqs. (3)–(6) are shown in Fig.2.

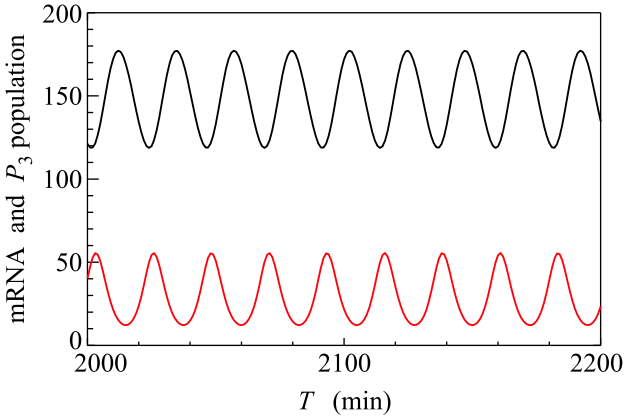

Fig.2. Numbers of M (lower curve) and P_3 (upper curve) as a function of time according to Eqs. (3), (4) with $m = 6$, $K_{P3} = 40$, $\kappa_t = 10^5\,\mathrm{min}^{-1}$, $\kappa_s = 2\,\mathrm{min}^{-1}$, $\kappa_{12} = \kappa_{23} = 0.2\,\mathrm{min}^{-1}$, $\kappa_M = 0.4\,\mathrm{min}^{-1}$, and $\kappa_{P1} = \kappa_{P3} = 0.2\,\mathrm{min}^{-1}$

Let us now consider the situation when the bistable kinetics of expression of gene 1 (like in the first example above) is regulated by protein P_3 produced due to oscillatory expression of gene 2 (like in the second example above). This situation can be described by introducing the P_3 association-dissociation factor into the second term of the right-hand part of Eq. (1). Assuming for example the existence of two P_3 regulatory sites and considering that the corresponding factor is simply proportional to N_{P3}^2, we have

$$\frac{dN_R}{dt} = k_0 + k_1 N_{P3}^2 \left(\frac{N_P}{K_P + N_P}\right)^n - k_R N_R. \qquad (7)$$

Equation (7) should be solved in combination with Eqs. (2)–(6). In this case, due to oscillations of N_{P3} described by Eqs. (3)–(6), the R and P numbers given by Eqs. (2) and (7) oscillate as well. To illustrate quantitatively what happens in this case, we use k_1 as a governing parameter. Our calculations indicate that with increasing k_1 the model predicts a stepwise transition from the R and P oscillations with small amplitude (see e.g. Fig.3a) to the R and P oscillations with high amplitude

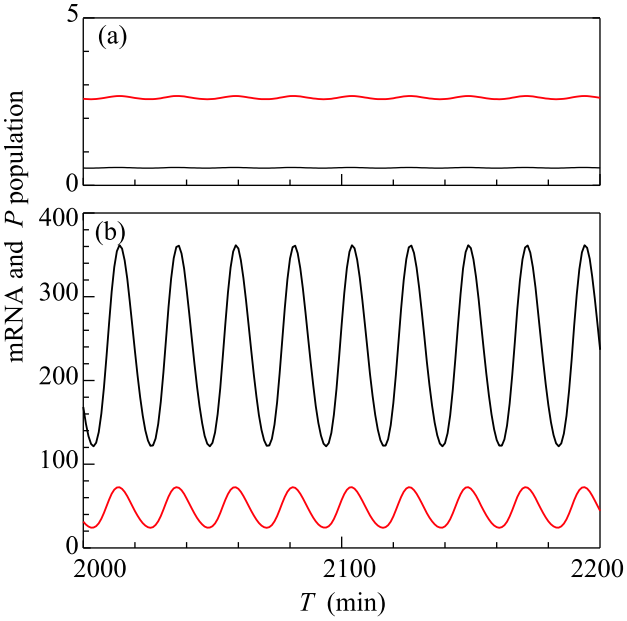

Fig.3. Numbers of R (lower curve) and P (upper curve) as a function time for gene 1 in the case when the expression of this gene is governed by protein P_3 as described by Eqs. (2)–(7) with $k_1 = 2 \cdot 10^{-3}$ (a) and $3 \cdot 10^{-3}\,\mathrm{min}^{-1}$ (b). The other parameters are as in Figs.1 and 2. For gene 2, the expression kinetics is the same as in Fig.2

(Fig.3b). These two regimes are related respectively to the low- and high-reactive state of the bistable kinetics. In particular, the minimum values of N_R and N_P during the latter oscillatory regime (Fig.3b) are higher than the maximum values of N_R and N_P during the former oscillatory regime. The R and P oscillations representing sequential transitions between the low- and high-reactive states of the bistable kinetics are not observed. This is explained by (i) stability of the states of the bistable kinetics and (ii) relatively small difference between the maximum and minimum values of N_{P3} during oscillations. For other values of the model parameters, the

situation is similar provided that they are selected so that the bistability is well manifested.

Despite the absence of the R and P oscillations representing sequential transitions between the low- and high-reactive states of the bistable kinetics, the R and P oscillations corresponding to the high-reactive state of the bistable kinetics are much more developed compared to the M, P_1, P_2 and P_3 oscillations (cf. Figs.2 and 3b). In particular, the ratio of the minimum and maximum values of N_R or N_P is appreciably higher than the corresponding ratios of N_M, N_{P1}, N_{P2} or N_{P3}. Thus, the high-reactive regime of expression of gene 1 is fairly sensitive to oscillations in the expression of gene 2. Basically, gene 1 amplifies oscillations of gene 2.

The gene-expression kinetics shown in Figs.1–3 have been calculated using the MF equations. Such kinetics, as noted in the introduction, can be influenced and modified due to fluctuations of the mRNA and protein numbers. To illustrate the role of fluctuations in the kinetics under consideration, we have performed Monte Carlo simulations by using the standard Gillespie algorithm [15] (for the model-specific details, see Ref. [6]).

Typical stochastic kinetics of expression of single gene 1 are shown in Fig.4. With increasing k_1, as

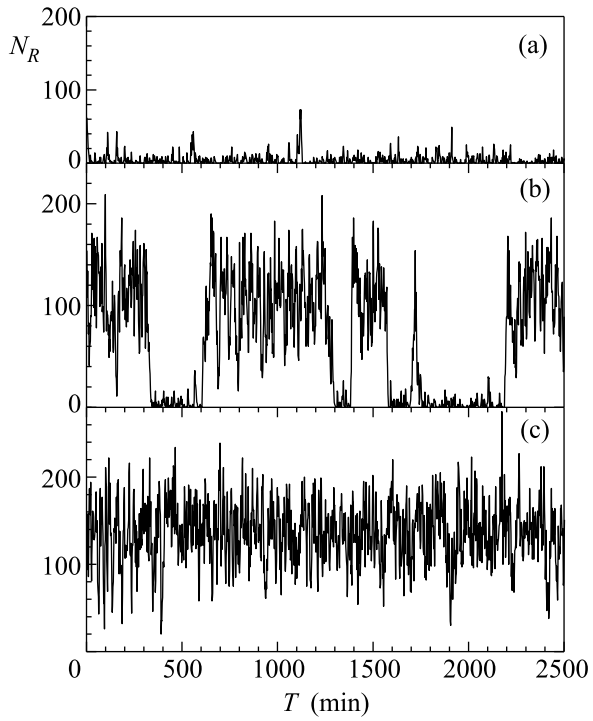

Fig.4. Stochastic kinetics of P expression for single gene 1 with $k_1 = 40$ (a), 45 (b), and $50\,\mathrm{min}^{-1}$ (c). The other parameters are as in Fig. 1. In this case, the R population (not shown) is $N_R \simeq (k_P/k_s)N_P = 0.2N_P$. The interval between data points is 2 min

expected [3–5], one can observe low-active regime (Fig.4a), stochastic bursts (Fig.4b), and high-active regime (Fig.4c).

Figure 5 exhibits stochastic kinetics in the situation when the expression of gene 1 is regulated by protein P_3

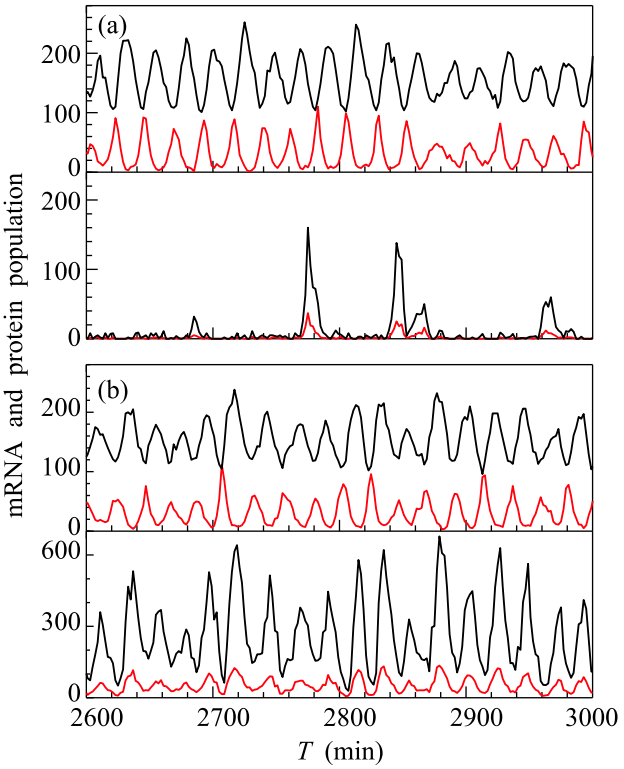

Fig.5. Stochastic kinetics of expression of genes 1 and 2 for $k_1 = 2 \cdot 10^{-3}$ (a) and $3 \cdot 10^{-3}\,\mathrm{min}^{-1}$ (b) (the other parameters are as in Figs.1 and 2). The upper panels show numbers of M (lower curve) and P_3 (upper curve). The lower panels exhibit numbers of R (lower curve) and P (upper curve). The interval between data points is 2 min

produced due to oscillatory expression of gene 2. The model parameters used in the simulations presented are the same as in Figs.2 and 3. For this reason, the stochastic oscillatory expression kinetics of gene 2 can be directly compared with the corresponding MF kinetics presented in Fig.2. In turn, the stochastic oscillatory expression kinetics of gene 1 can be compared with the MF kinetics presented in Fig.3. In both cases, except fluctuations, the stochastic and MF oscillations are seen to be basically the same. Thus, the stochastic effects do not change our main conclusions drawn above on the basis of the MF calculations.

In summary, we have presented MF calculations and MC simulations illustrating how the deterministic and stochastic bistable kinetics of expression of one gene can be influenced by kinetic oscillations in expression of another gene. Intuitively, one could expect that the

oscillations in expression of gene 2 would result in sequential transitions between the low- and high-reactive states of the bistable kinetics of gene 1. The results obtained indicate however that this is hardly possible. Due to stability of the states of the bistable kinetics of gene 1 and relatively small difference between the maximum and minimum numbers of protein copies during kinetic oscillations of gene 2, the induced oscillations of gene 1 are typically related either to the low- or to high-reactive state of the bistable kinetics of this gene. In the situations when the induced oscillations of gene 1 are related to the high-reactive state, their quality may be appreciably better than that of inducing oscillations. This means that gene 1 can serve as an amplifier of kinetic oscillations of gene 2.

Our findings clarify the basic principles of the interplay of complex kinetics of gene expression and may have important implications for the understanding and interpretation of real biological processes which are dependent of oscillations in gene expression. At present, there are only a few experimental observations of kinetic oscillations in genetic networks [14]. Such oscillations are believed to be behind, for example, circadian clocks and patterning of vertebrates [14]. Due to the complexity of real systems, the details of the mechanisms of oscillations and their functions are now open for debate. The experimental attempts to trace the effect of oscillations on bistability in gene expression are lacking. The current interest in the complex kinetics of gene expression is however high [3, 4]. For these reasons, our study is timely and the results obtained may guide the experiments.

This work was partially funded by the Swedish Science Council.

1. B. Alberts, A. Johnson, J. Lewis et al., *Molecular Biology of the Cell*, Garland, New York, 2002.
2. F. F. Costa, Gene **357**, 83 (2005); J. A. Goodrich and J. F. Kugel, Nat. Rev. Molec. Cell. Biol. **7**, 612 (2006).
3. M. Kaern, T. C. Elston, W. J. Blake, and J. J. Collins, Nature Rev. Genet. **6**, 451 (2005).
4. J. Paulsson, Phys. Life Sci. **2**, 157 (2005); S. Bornholdt, Science **310**, 449 (2005); B. B. Kaufmann and A. van Oudenaarden, Curr. Opin. Gen. Devel. **17**, 107 (2007).
5. L. Mariani, M. Löhning, A. Radbruch, and T. Höfer, Progr. Biophys. Mol. Biol. **86**, 45 (2004).
6. V. P. Zhdanov, Chem. Phys. Lett. **424**, 394 (2006); JETP Lett. **84**, 632 (2007).
7. D. Bratsun, D. Volfson, L. S. Tsimring, and J. Hasty, Proc. Nat. Acad. Sci. USA **102**, 14593 (2005); L. G. Morelli and F. Julicher, Phys. Rev. Lett. **98**, 228101 (2007); K. Rateitschak and O. Wolkenhauer, Math. Biosci. **205**, 163 (2007).
8. V. P. Zhdanov, JETP Lett. **86**, 492 (2007).
9. V. P. Zhdanov, Chem. Phys. Lett. **458**, 359 (2008).
10. S. Ghaemmaghami, W. Huh, K. Bower et al., Nature **425**, 737 (2003).
11. I. Golding and E. C. Cox, Proc. Nat. Acad. Sci. USA **101**, 11310 (2004).
12. L. Cai, N. Friedman, and X. S. Xie, Nature **440**, 358 (2006).
13. S. A. Isaakson and C. S. Peskin, SIAM J. Sci. Comp. **28**, 47 (2006); V. P. Zhdanov, JETP Lett. **85**, 302 (2007); Chem. Phys. Lett. **458**, 247 (2008).
14. G. Tiana, S. Krishna, S. Pigolotti et al., Phys. Biol. **4**, R1 (2007).
15. D. T. Gillespie, J. Phys. Chem. **81**, 2340 (1977).