Arsenic biosensor modelling: simple alternative formulation

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1 Introduction

This report presents an alternative simplified model formulation for the arsenic (As(III)) biosensor system [1], in particular to explore relationships between binding activities of arsR and arsD to their corresponding promoters and arsenate respectively. The report structured as follows. First, short background to the biological system being modeled is provided. Second, modeling assumptions and computational approaches are described. Finally,results of model simulation are described.

2 Biological System

The model is a simplified representation of the arsenateal resistance (ars) operon of resistance plasmids R773 and R46 that encodes a pump that produces resistance to arsenite [2]. The operon has five genes, ArsR, -D, -A, -B, and -C. ArsR and ArsD are both trans-acting repressor proteins that regulate the levels of ars transcript ArsR is an As(III) -responsive repressor with high affinity for its operator site that controls the basal level of expression of the operon . Binding of arsenate produces dissociation of ArsR from the operator site, permitting transcription. As the levels of transcript rise, synthesis of the integral membrane ArsB protein becomes toxic, limiting

growth. ArsD is a second regulator that controls the upper level of expression of the operon, preventing overexpression of ArsB [3]. ArsD is also a homodimer of two 120-residue subunits that binds to the same operator site as ArsR but with an affinity that is 2 orders of magnitude less than that of ArsR [4]. Consequently, ArsD only binds to the ars operon when produced in high concentrations, such as after prolonged stimulation of transcription of the arsD gene following induction by arsenite or antimonite. Together, ArsR and ArsD form a regulatory circuit that controls the basal and maximal levels of expression of the ars operon.

3 Model

In the model we tried to harness the property of different binding affinities of ArsD and ArsR to the arsenate and corresponding operator site to see how well they can account for different responses of our pH based arsenate biosensor. As a recollection, in our model urease is expressed from a hybrid promoter repressed by both lambda CI repressor and LacI repressor. In the presence of lactose, but absence of arsenate, urease is induced and leads to rise in pH. When low amount of arsenate is present, an ArsR-repressed promoter is induced, leading to expression of lambda lCI repressor, switching off urease production. Thus the pH remains neutral. If higher amounts of arsenate are present, lacZ expression is induced through an ArsD-responsive promoter, leading to a fall in pH. By using multiple promoters in this way, a high sensitivity and high dynamic range are achieved. We sought to address the question of interplay between different promoters and transcription factors for arsenate with simpler model, which is truncated to only three major types of equations and is provided below. The form of equations was adapted from [5].All the modeling was done using COPASI software.

$$\frac{d(LacZ)}{dt} = \frac{\beta_l acZ}{1 + ARSD_T/K_{arsd}/(1 + (ARS/K_{ars-d})^n)} - K_{lacz}[LacZ]$$

$$\frac{d(lCI)}{dt} = \frac{\beta_l CI}{1 + ARSR_T/K_{arsr}/(1 + (ARS/K_{ars-r})^n)} - K_{lCI}[lCI]$$

$$\frac{d(urease)}{dt} = \frac{\beta_u rease}{1 + (lCI/K_{lCI})} - K_{urease}[Urease]$$

Where ARS - arsenic concentration, Kars-d and Kars-r - affinity of ArsD and ArsR to arsenic, ARSRt and ARSDt - total amount of ArsR and ArsD, Karsd

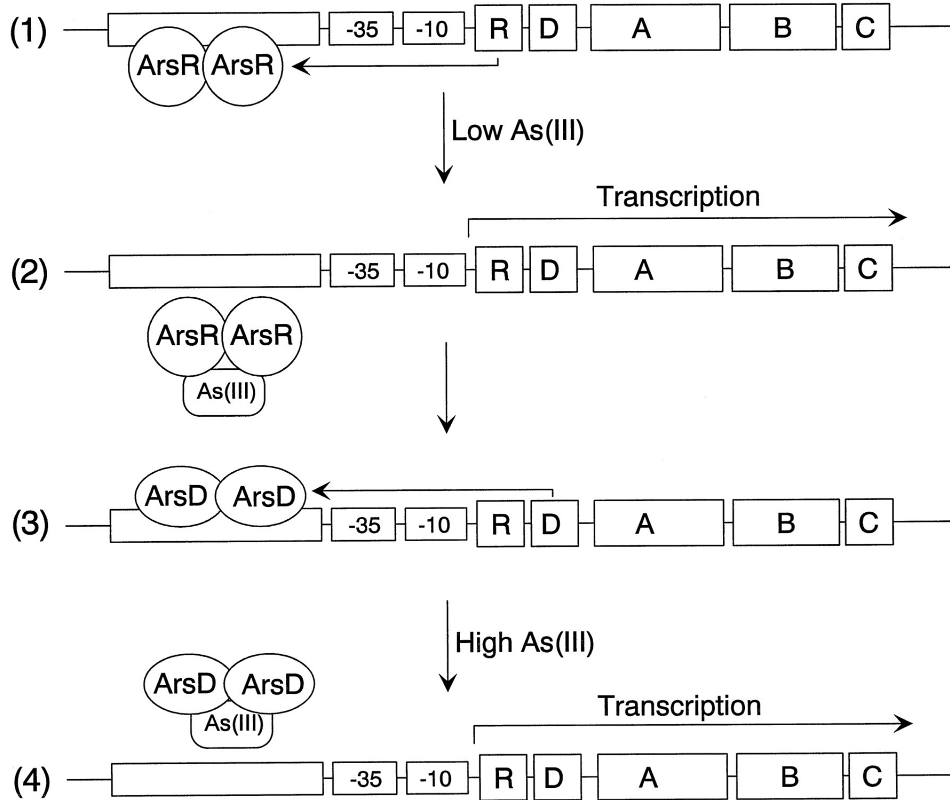

Figure 1: A model of the ArsR-ArsD metalloregulatory circuit. Step 1, in the absence of arenite transcription is repressed by a basal level of ArsR synthesis. Step 2, in the presence of low to moderate concentrations of inducer, the ArsR-inducer complex dissociates from the DNA, resulting in transcription of the ars operon. Step 3, when the concentration of ArsD increases sufficiently to allow this low affinity DNA binding protein to bind to the operator site, transcription is again repressed. Step 4, in the presence of high concentrations of inducer, the ArsD-inducer complex dissociates from the DNA, resulting in a further increase in ars transcription (reproduced from [3])

and Karsr- affinity (Dissociation constant) of ArsD and ArsR to their corresponding promoters, Klacz and Klci - degradation constants for LacZ and lCI respectively, Blacz and Blci - maximal expression value of corresponding transcription factors. Burease and Kurease are maximal expression and degradation constant for Urease, and Klci - lCI affinity (Dissociation constant) for urease (inhibitory effect of LacI is absorbed into Burease). The model uses um units for arsenate. The following conversion table - 1 to 10,000 ppb equal approximately 0.01 to 100 um - taken from (Whole-Cell Bacterial Biosensors and the Detection of Bio available Arsenic [1]) was used in the modeling.The model was built using followings assumption and kinetic parameters:

1. Since every model is only an approximation of the real systems behavior, it is limited in its predictive ability to the questions it is addressed to answer. In current model we make an assumption that arsD and arsR bind non-overlapping regions of the operator and there is no *combinatorial effect* between to different transcription factors and sites they bind. Based on this assumption we consider a set of parameters that can model response to different levels of arsenic.

2. Transcription and translation reactions are combined, there fore promoter activity increases much faster (20 min) than if translation and transcription were separate processes.

3. Dissociation constant of arsenic (Karsd and Karsr) for arsR is 0.03 and for arsD - 10 (approx. 100 fold difference).

4. LacI is presumed to be in saturation; therefore mass action kinetics is applied.

5. It is also presumed that Dissociation constant of LCI for urease promoter is low - 0.01, which presumes strong binding.

Species	Initial Concentration
arsD total	0.1
arsR total	0.0001
LacZ	15.00
LCI	0.0001
Urease	0.001
Arsenic	VARIED
LacI	0.0001

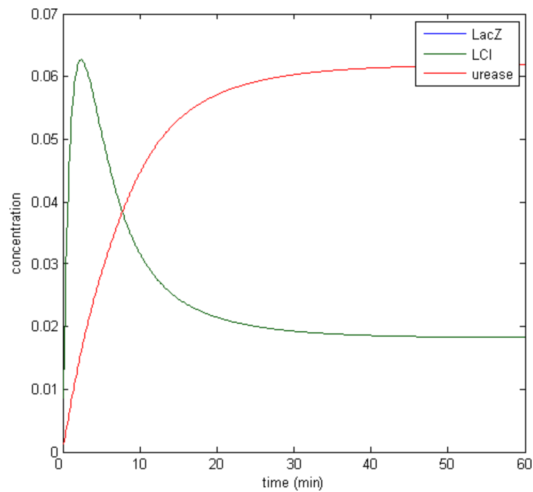

Figure 2: ODE simulation with no arsenic, deterministic, step size 0.1. The strong rise of the LCI in the beginning is explained by the assumption that LCI is initially available in the model and has some very small basal rate, which is surpassed with time by degradation rate.

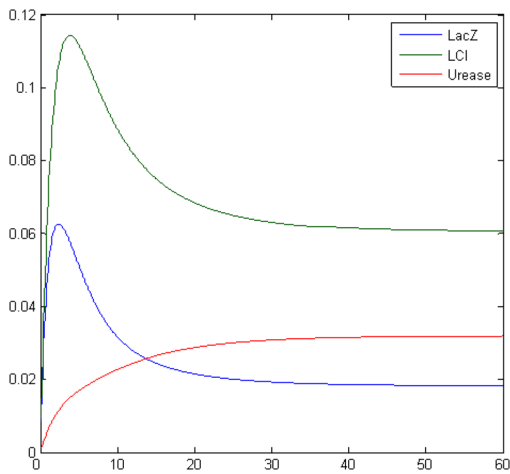

Figure 3: ODE simulation, deterministic, step size 0.1, 0.1 um of arsenic

4 Model response to arsenic

Here we present results of simulation which agrees with the other model formulation and biological system of arsenic biosensor (Fig. 2-4).

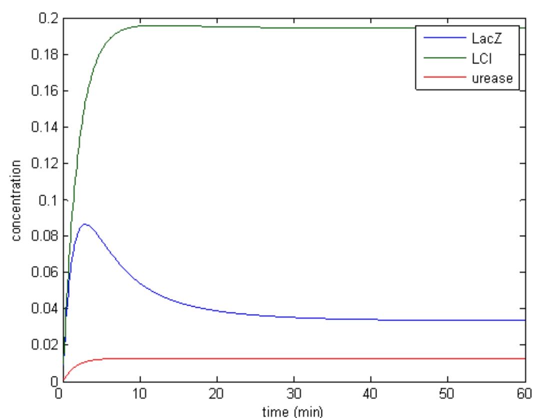

Figure 4: ODE simulation, deterministic, step size 0.1, 10 um of arsenic. As we can see LCI reaches saturation level, because it is controlled by arsR that have strong binding to arsenic. However, with this concentration of arsenic LacZ is also synthesized (cf figure 2)

5 References

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