Engineering transcription-based digital logic devices

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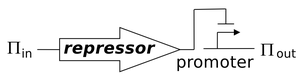
Abstract

The goal of Synthetic Biology is to engineer systems from biological parts. One class of systems are those whose purpose is to process information. My work seeks to build transcription-based devices for use in combinational digital logic. Preliminary characterization experiments show that existing devices fall short of desired device behavior. I propose to develop a novel implementation of transcription-based logic by designing synthetic transcription factors from well-characterized DNA binding and dimerization domains. Initial modeling work serves to inform design of these devices.

I Background

Goal

Implement *in vivo* combinational digital logic using transcription-based devices.



Transcriptional inverter

Desirable device features
- well-characterized
- composable
- engineerable
- numerous

Figure 1: An inverter is a simple digital logic device. The work presented here focuses on characterizing and modeling a transcriptional inverter.

II Device characterization

To engineer good devices, we need device performance metrics and measurement methods.

Device behavior

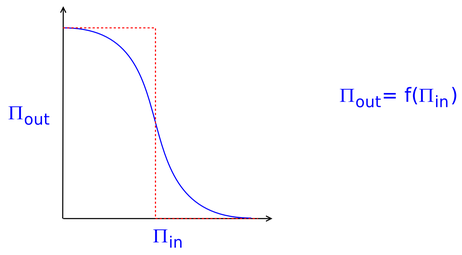


Figure 2: A transfer characteristic is a plot of device output as a function of device input. It describes static device performance.

Device performance

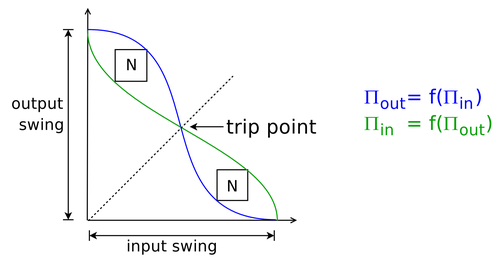


Figure 3: The swing, noise margin and trip point are quantitative measures of the quality of the transfer characteristic. Ideal devices maximize the noise margin and have a trip point close to half the device swing.

Existing devices fail

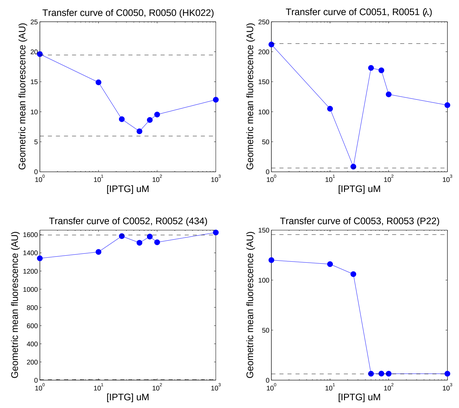


Figure 4: Experimentally-determined transfer characteristics for 4 transcriptional inverter prototypes.

One inverter has good performance characteristics; however, the other devices fail to function properly.

III Device modeling

Modeling informs design of synthetic transcription factors for digital logic.

Simplifying assumptions

1. Use reaction rate equations
2. Neglect cell growth and DNA replication
3. Ignore details of synthesis and degradation reactions
4. All or none repression
5. Binding reactions at equilibrium
6. Steady-state

Model formulation

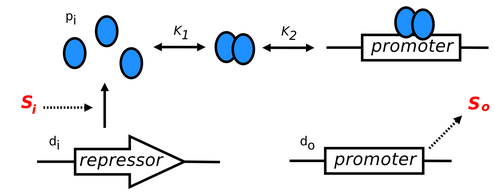


Figure 5: Model includes synthesis, degradation and binding reactions.

Model equations

$$S_o = \frac{K_1 K_2}{K_1 K_2 + [p_i]^2}$$

$$[p_i]^T = S_i \alpha_i = [p_i] + 2K_1^{-1}[p_i]^2 + \frac{2[d_o]^T [p_i]^2}{K_1 K_2 + [p_i]^2}$$

$$\alpha_i \equiv \frac{k_{sp_i} k_{sm_i}}{k_{dp_i} k_{dm_i}} [d_i]^T$$

Figure 6: Key parameters that affect device behavior are the steady-state maximum protein level α_i, promoter copy number $[d_o]^T$ and dissociation constants K_1 and K_2.

Target parameter values

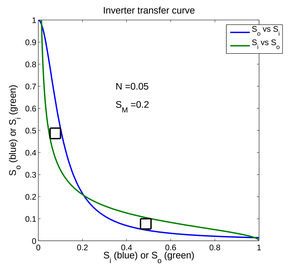


Figure 7: These parameter values lead to acceptable device behavior.

$$K_1 \approx \alpha_i * 100 \approx 10\ mM \text{ (dimerization)}$$
$$K_2 \approx \alpha_i / 10000 \approx 10\ nM \text{ (DNA binding)}$$
(assuming $\alpha_i \approx 50,000\ proteins/cell$)

- **Steady-state maximum input protein concentration α_i is the input protein swing.**
- **Value of dissociation constants relative to α_i determines noise margin.**
- **Inverter skew may be a problem.**

References

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Sensitivity analysis

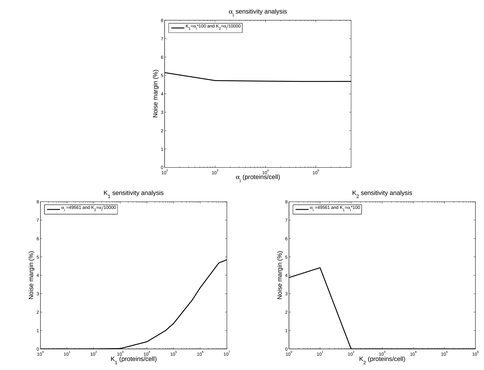


Figure 8: Noise margin as a function of parameter value for α_i, K_1 and K_2.

Device performance is sensitive to K_1 and K_2 but not α_i.

Alternative design

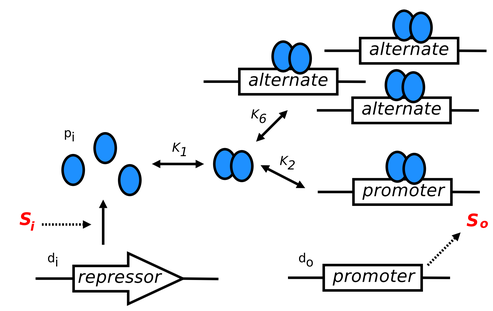
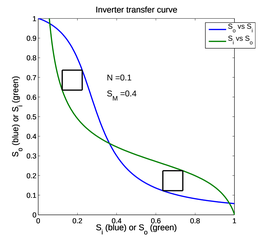


Figure 9: A possible alternate device design includes additional nonfunctional protein binding sites on the DNA.



$[d_a]^T = 0.1\alpha_i$ and $K_6 = K_2/10$

Figure 10: This alternate design can lead to enhanced device performance.

Alternate device designs may lead to improved transfer characteristics.

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