Dynamic Responses of LacI/GalR Chimera-Based Transcriptional Logic Gates

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***Short Abstract* — Genetically encoded logic is central to the function of living organisms. Synthetic biology has created many orthogonal genetic logic systems, but these systems are usually studied at steady state. We use microfluidic devices to study the dynamic responses of genetic logic gates. The specific gates in this work are constructed using chimeric transcriptional repressors, so logic can be created by controlling induction or production of these repressors. Controlling induction yields IMPLY or AND logic; controlling production yields NOT or NOR logic. We find that the speed at which gates reliably respond to environmental changes depends on their mechanism of induction.**

***Keywords* — Genetic Logic Gates, Microfluidics, Dynamics**

I. Purpose

In synthetic biology, genetic logic gates have been constructed with applications in mind such as biological computing and sensors in a dynamic environment. These logic gates may be integrated into larger networks, mediating response or coupling dynamics of modular genetic circuits based on environmental conditions. However, less work has been done to understand the limitations of transcriptional logic gates in these applications. It is clear that reliable logic gates should respond on a faster time-scale than time-varying environmental conditions. Therefore, we aim to measure the limitations on the dynamic range of transcriptional logic gates.

The focus of this study is chimera-based transcriptional logic gates. Previous work showed that chimeric proteins derived from the LacI/GalR family of transcriptional repressors can be used to create transcriptional AND gates *in vivo* [1]. These chimeric proteins have the same operator (DNA) binding domain but different ligand binding domains; hence, they will bind to the same operator site but are induced by different sugars. By changing the production of the chimeras to inducible promoters (from constitutive promoters), NOT and NOR logic is implemented.

II. Results

Two types of logic gates are described in this work – "ligand" gates (AND, IMPLY) and "inducible" gates (NOT, NOR). "Inducible" gates include an extra production step – that of the repressor – when compared to "ligand" gates. We find that transcriptional logic gates generally function as low-pass filters, responding faithfully to low frequency (long period) signals and unfaithfully to high frequency (short period) signals.

A. "Ligand" gates respond over a wide range of driving frequencies

Both AND and IMPLY gates were tested over a range of driving periods from 20 to 240 minutes. Both gates show robust output at periods from 40 to 240 minutes – that is, a clear threshold for ON and a clear threshold for OFF can be set in all these experiments. At a 20 minute driving period, the outputs of these gates do not reach a clear ON or OFF thresholds.

B. Responses in the NOT gate are delayed compared to "ligand" gates

While the AND and IMPLY gates are robust at periods greater than 40 minutes, the NOT gate tested here is only robust in its response at periods greater than 120 minutes. This difference in behavior is attributed to the extra time required to produce and degrade the repressor proteins, steps not required in the "ligand" gates.

III. Conclusion

While genetic logic can be implemented by controlling the production of a transcription factor, we show that robust responses are generated at faster time scales when the levels of regulatory proteins do not need to be controlled.

References

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