# Design Principles of Transcriptional Logic Circuits

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**Abstract**

Using a set of genetic logic gates (AND, OR and XOR), we constructed a binary full-adder. The optimality analysis of the full-adder showed that, based on the position of the regulation threshold, the system displays different optimal configurations for speed and accuracy under fixed metabolic cost. In addition, the analysis identified an optimal trade-off curve bounded by these two optimal configurations. Any configuration outside this optimal trade-off curve is sub-optimal in both speed and accuracy. This type of analysis represents a useful tool for synthetic biologists to engineer faster, more accurate and cheaper genes.

## Introduction

The desire to control is a recurring theme of human nature and the control of biological systems represents the ultimate goal for synthetic biologists. Towards achieving this goal, researchers have modelled and engineered genes in bacterial cells that perform basic computational tasks. These tasks mainly mimic the behaviour of simple electronic components, such as logic gates, oscillators, toggle switches and counters (Gardner et al., 2000; Elowitz and Leibler, 2000; Guet et al., 2002). However, when attempting to increase the complexity of these engineered genetic systems, certain limitations of the components are likely to hamper their construction. Thus, there is an urgent need for an extensive analysis of the biophysical limits of the elementary components.

Synthetic biologists showed that binary logic gates can be engineered in living cells using transcriptional logic (Guet et al., 2002; Kramer et al., 2004; Yokobayashi et al., 2002; Cox III et al., 2007; Anderson et al., 2007; Sayut et al., 2009). Transcriptional logic gates are genes which can integrate multiple signals at the level of cis-regulatory transcription control using various binary logic functions (AND, OR, NAND, NOR, XOR, etc.). To implement binary logic, both the input and the output of these genes needs to have two abundance levels corresponding to the two logical levels, a high and a low abundance level. Biological modellers successfully identified and described various designs of these logic gates (Weiss et al., 2003; Buchler et al., 2003; Hermsen et al., 2006; Schilstra and Nehaniv, 2008; Silva-Rocha and deLorenzo, 2008). However, what is still missing is a complete analysis of how these logic gates can be used as building blocks for more complex logical systems and what are the parameters which ensure optimal design in terms of speed and accuracy under limited (constant) energetic resources.

There are three properties of a genetic system that we use in our analysis: speed, accuracy and cost. We define the *propagation time* as the time required by the output species in a logical system to reach the new steady state after an instantaneous change of the inputs. This is directly connected with *speed* in the sense that fast system are described by short propagation times and conversely. Due to low copy number and slow chemical reactions, genetic systems are stochastic and, thus, they are affected by *noise* (Kaern et al., 2005). The noise reduces the ability to distinguish between different logical outputs of a gate and, because of that, it reduces *accuracy*. Finally, the *metabolic cost* is usually measured as the required number of ATP molecules. We are interested in the scaling properties of this measure, rather than in the exact value. Hence, we measure cost as the maximum synthesis rate of a gene.

Recently we investigated speed and accuracy in the case of single binary genes (genes with two expression levels, high and low) (Zabet and Chu, 2010). The analysis revealed that these genes display a trade-off curve between switching time and noise under fix metabolic cost, i.e., lower noise is achieved at lower speeds and conversely. This trade-off is controlled by the decay rate, in the sense that higher decay rate means higher speed but also lower accuracy.

In this contribution, we extend this analysis to gene networks by considering a specific binary logic system, the full-adder. The full-adder is a system able to perform binary addition (to produce both the sum and the carry) for three binary inputs, two of which are the two operands and the third allows plugging in the carry from a previous full-adder module. We constructed the required logic gates by considering genes that can be regulated by two proteins in an independent fashion, i.e., binding of any of the inputs does not alter

the binding of the other input. Moreover, these logic gates need to ensure *interconnectivity*. Assuming that the two inputs that regulate a gene can have two possible abundance levels, high ($H_{in}$) and low ($L_{in}$), then, in order to connect an *arbitrary* number of logic gates, the output has to have two possible abundance levels ($H_{out}$ and $L_{out}$) with at least the same signal strength, $(H_{in} - L_{in}) \leq (H_{out} - L_{out})$ (Magnasco, 1997). Usually the output levels are identical with the input one or very close to them, $H_{out} \geq H_{in}$ and $L_{out} \leq L_{in}$. Based on these requirements, we found the set of parameters which ensures interconnectivity of the required logic gates and then we constructed the full-adder showing the correct functioning of the system.

Gene regulation is usually modelled by a Hill function (Ackers et al., 1982; Bintu et al., 2005; Chu et al., 2009). The Hill function is a sigmoid function described by two parameters: the threshold $K$ (which represents the input abundance required for half activation of the gene) and the Hill coefficient $l$ (which determines the steepness of the function). The results show that, for step-like regulation functions ($l \to \infty$), the system displays an optimal position of the threshold in terms of speed and accuracy, while, for finite Hill coefficients, there is a trade-off between these two properties and the trade-off is controlled by the position of the threshold.

## Model

We selected a design for the full-adder with five logic gates: two XOR gates, two AND gates, and one OR gate (see Fig. 1).

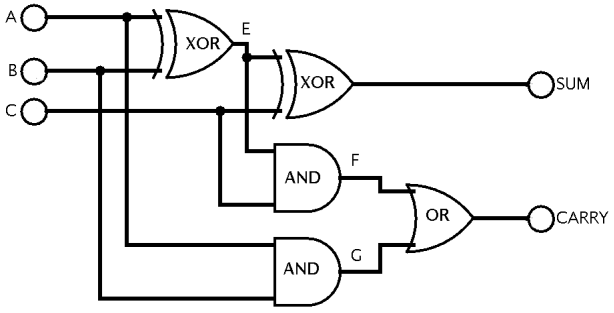



Figure 1: *Full-adder*. The logic gate diagram of the full adder.

To construct this full-adder from genes, we need first to construct transcriptional logic gates. We model a transcriptional logic gate as a gene $G_z$, which synthesises protein $z$, the output of the gate. This gene is regulated by two proteins $x$ and $y$, which are considered as the inputs of gate. Species $z$ is described by the following deterministic differential equation

$$\frac{dz}{dt} = \alpha + \beta f(x, y) - \mu z \qquad (1)$$

where $\alpha$ is the basal synthesis rate, $\alpha + \beta$ the maximum synthesis rate, $f(x, y)$ is the regulation function of gene $G_z$, and $\mu$ is the decay rate.

Although there are many scenarios for promoter regulation that mimic the behaviour of different logic gates, we selected independent binding (binding of one TF does not influence in any way the binding of the other TF). In this scenario there are two operator sites $O_x$ and $O_y$, each of them having $l$ binding sites. On each operator site only molecules of a specific transcription factor can bind, and they do this in a homo-cooperative maner. The probabilities that an operator site is full is described by a Hill function (Ackers et al., 1982; Bintu et al., 2005; Chu et al., 2009)

$$p_x(x) = \frac{x^l}{x^l + K^l}, \quad p_y(y) = \frac{y^l}{y^l + K^l} \qquad (2)$$

where $K$ is the regulation threshold (the required input value for half activation of the gene) and $l$ is the Hill coefficient (indicates steepness of the function). We assumed that the two operator sites ($O_x$ and $O_y$) have identical parameters ($K$ and $l$).

Assuming that the gene is turned on when any of the two TF are present, then the regulation function will mimic the behaviour of an OR gate. Analogously, assuming that a gene can be turned on only when both of the transcription factors are present, then the regulation function will mimic the behaviour of an AND gate. Finally, if the gene is turned on when any of the TF is present, but when both of them are present their effects cancels out and the gene is turned off, then the gene will behave as an XOR gate. The corresponding forms of the regulation functions are

$$\begin{aligned} f_{AND} &= \frac{(xy)^l}{(xy)^l + (Kx)^l + (Ky)^l + K^{2l}}, \\ f_{OR} &= \frac{(xy)^l + (xK)^l + (yK)^l}{(xy)^l + (Kx)^l + (Ky)^l + K^{2l}}, \\ f_{XOR} &= \frac{(Kx)^l + (Ky)^l}{(xy)^l + (Kx)^l + (Ky)^l + K^{2l}}. \end{aligned} \qquad (3)$$

Fig. 2 confirms that these regulation functions display the desired behaviour.

Using these three logic gates, the full-adder, can be constructed as a set of chemical reactions. Since the full-adder contains five logic gates, then we need five species to implement this system ($e$, $f$, $g$, sum and carry). The chemical reactions which describe all these species are given by

$$\emptyset \underset{\mu_e}{\overset{\alpha_e + \beta_e f_{XOR}(a,b)}{\rightleftharpoons}} e, \quad \emptyset \underset{\mu_f}{\overset{\alpha_f + \beta_f f_{AND}(c,e)}{\rightleftharpoons}} f,$$

$$\emptyset \underset{\mu_g}{\overset{\alpha_g + \beta_g f_{AND}(a,b)}{\rightleftharpoons}} g,$$

$$\emptyset \underset{\mu_s}{\overset{\alpha_s + \beta_s f_{XOR}(e,c)}{\rightleftharpoons}} \text{sum}, \quad \emptyset \underset{\mu_{co}}{\overset{\alpha_{co} + \beta_{co} f_{OR}(f,g)}{\rightleftharpoons}} \text{carry}$$

where $a$, $b$ and $c$ are three input species.

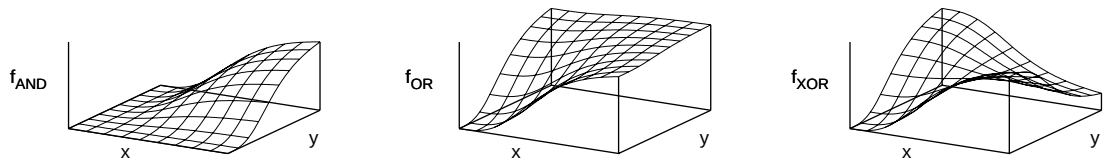



Figure 2: *Regulation functions that mimic logic gate behaviour*. The threshold was set to $K = 0.5\ [\mu M]$ and we considered a Hill coefficient of $h = 3$.

## Results

First we need to identify the sets of parameters which allow interconnection of gates and then we need to identify the sub-set of parameters which allows optimal functioning of the full-adder in terms of speed and accuracy under fixed metabolic cost. We will apply these two analyses for two cases: ($i$) step-like regulation functions ($l \to \infty$) and ($ii$) finite Hill coefficients.

To keep the mathematics tractable, and without losing too much generality, we consider identical gates, i.e., all genes are affected by the same decay rate ($\mu$), have the same synthesis rates ($\alpha$ and $\beta$) and the same Hill parameters ($l$ and $K$). The only thing that differentiates the gates is the regulation function, which, in the case case of the full-adder, can be $f_{AND}$, $f_{OR}$ or $f_{XOR}$.

### Step Regulation Functions

We start our analysis by considering the ideal case, the system where the regulation functions have infinite Hill coefficient.

The interconnectivity property can be met by considering the output signal strength to be kept constant, $H_{out} = H_{in} = H$ and $L_{out} = L_{in} = L$. In the case of the OR gate, the system has the following steady state behaviour

$$\begin{aligned} L &= \frac{1}{\mu}\left[\alpha + \beta f_{OR}(L, L)\right], \\ H &= \frac{1}{\mu}\left[\alpha + \beta f_{OR}(L, H)\right], \\ H &= \frac{1}{\mu}\left[\alpha + \beta f_{OR}(H, H)\right]. \end{aligned} \tag{4}$$

For infinite Hill coefficient the solution is given by $\alpha = L$ and $\beta = (H - L)$. Analogously, it can be shown that the solution is the same for all gates. This synthesis rates ensure a correct steady-state behaviour of the full-adder (see Fig. 3(a)).

**System Performance** We investigate two properties of a logic system, namely speed and accuracy, under the constraint of fix metabolic cost. The metabolic cost of a gene $Z$ can be defined as the maximum synthesis rate of that gene, $\zeta_z = \alpha + \beta f_z^H$, where $f_z^H$ is the highest value which $f(x, y)$ takes. Thus, by keeping the synthesis rate fixed the metabolic cost is kept constant. Note that this is just an approximation to the actual metabolic cost, and that the metabolic cost of the maintenance of the entire machinery was not included in it. However, this measure indicates how the metabolic costs scales with different parameters.

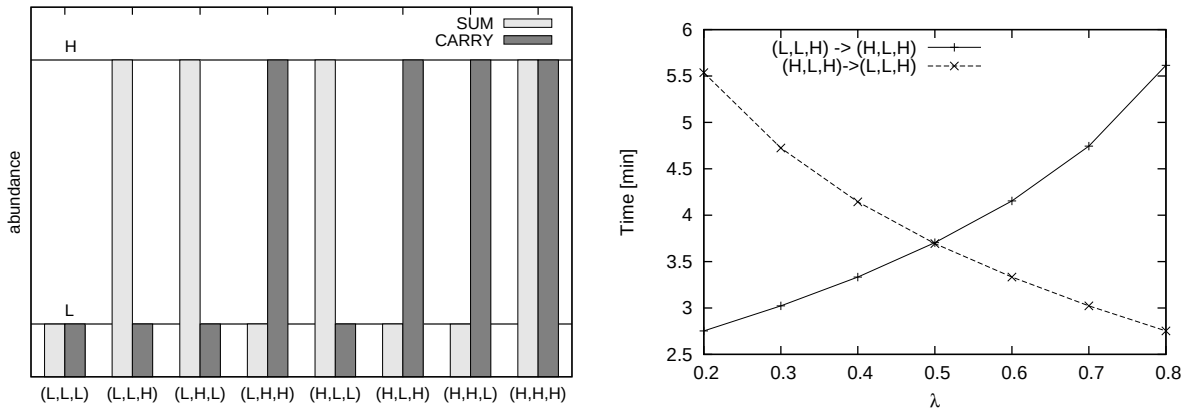
The *propagation time*, $T_{gene}$, of a gene is the time required to reach the steady state to within a fraction $\theta$ of $H - L$. Assuming instant change of the input, Eq. (1) can be solved analytically and the time to reach $L + (H - L)\theta$ or $H - (L - H)\theta$ can be computed as

$$T_i = \tau \cdot ln\left(\frac{1}{1-\theta}\right) \tag{5}$$

where $\tau = 1/\mu$ represents the average life time of the species.

The propagation time through a single gate can only be reduced by reducing the average life time of the protein ($\tau$). In the case when the two logical steady states are kept constant (so the signal strength is not reduced) and the synthesis rate is kept constant (so we do not increase the metabolic cost) then also the decay rate is kept constant. Thus, there is no optimization that one could attempt to perform on individual gates under fix metabolic cost without reducing signal strength. However in the case of logic gates systems, like the case of the full-adder, the input is not changed instantaneously in all gates and the position of the threshold influences the propagation time.

The threshold is located between the low and the high state, $K = L + (H - L)\lambda$, ($\lambda \in [0, 1]$). $\lambda$ indicates the position of the threshold; for $\lambda < 0.5$, $K$ is closer to $L$ and for $\lambda > 0.5$, $K$ is closer to $H$. Note that by considering $K$ to be outside the interval $[L, H]$ the regulation is removed, i.e., the gene is always in the same state no matter whether the input is $L$ or $H$. In order for a gene to change state, one of the inputs, has to cross over or under $K$. Using Eq. (5) one can compute the time it takes one species to move from low state to the threshold ($L \to K$) and from the high state




(a) steady state behaviour (b) optimum $K$ for speed

Figure 3: *Full-adder with step-like regulation function.* (a) The output abundance based on the input abundance for step-like regulation functions. (b) We plotted the propagation time when switching between $(L, L, H)$ to $(H, L, H)$. The following set of parameters have been used: $\mu = 1\ min^{-1}$, $l = 50$, $L = 0.2\ \mu M$, $H = 1.2\ \mu M$, $K = 0.7\ \mu M$, $\alpha = 0.2\ \mu M \cdot min^{-1}$, $\beta = 1.0\ \mu M \cdot min^{-1}$ and $\theta = 0.9$.

to the threshold ($H \rightarrow K$) as

$$t_{LK} = \tau \cdot ln\left(\frac{1}{1-\lambda}\right), \quad t_{HK} = \tau \cdot ln\left(\frac{1}{\lambda}\right). \quad (6)$$

Assuming that the longest cascade in the system has $n$ gates, then a general formula for the propagation time is given by

$$T = \sum_{i=1}^{n-1} t_{iK} + T_n \quad (7)$$

where $t_{iK}$ is equal to $t_{LK}$ if species $i$th was in low state before changing the input in the system, and $t_{iK}$ is equal to $t_{HK}$ if species $i$th was in high state before changing the input in the system. Hence, the propagation time in a cascade equals a sum of $t_{LK}$ and $t_{HK}$ terms and a fix time representing the last gene in the cascade $T_n$.

Fig. 4 confirms that based on the threshold position, the system can be faster when switching in one direction and slower in the opposite direction. When the switching direction is not important, the problem of optimizing propagation time becomes a minimax problem, i.e., minimize the maximum time to switch. In the context of step-like regulation functions, the optimum threshold, according to Eq. (6), resides at the midpoint between high and low states, $\lambda_T = 0.5$ (see Fig. 4).

Analysing the circuit diagram of the full-adder 1 one can notice that the longest path through the circuit consists of three gates, and this is used when computing the carry . This path is followed, for example, when switching between $(L, L, H)$ and $(H, L, H)$. Fig. 3(b) confirms that the optimum threshold, in the case of step-like regulation function, resides at the midpoint between high and low state

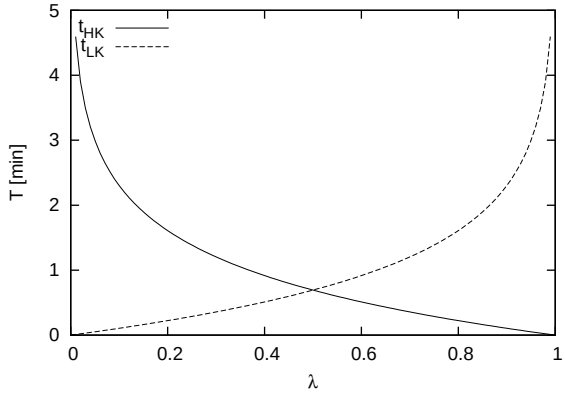



Figure 4: *The time to reach the threshold.* The protein average life time to $\tau = 1\ [min]$. The two steady states are $L = 0.2\ [\mu M]$ and $H = 0.8\ [\mu M]$, and the corresponding synthesis rates were considered. Both switching directions were consider.

($\lambda = 0.5$). Also note, that Eq. (7) and Eq. (6) correctly predict the propagation time in the full-adder in the case of high Hill coefficients.

Next, we need to investigate the accuracy of the system. At steady state the *variance* of the output $z$ of a logic gate, which has two inputs $x$ and $y$, can be written as (van Kampen, 2007; Elf and Ehrenberg, 2003; Paulsson, 2004)

$$\sigma_z^2 = \underbrace{z}_{\text{intrinsic}} + \underbrace{\overbrace{\left[\beta_z \frac{\partial f(x,y)}{\partial x}\tau_z\right]^2}^{\Gamma_{zx}} \overbrace{\frac{\tau_x}{\tau_x+\tau_z}}^{T_{zx}} \sigma_x^2}_{\text{upstream from } x} + \underbrace{\overbrace{\left[\beta_z \frac{\partial f(x,y)}{\partial y}\tau_z\right]^2}^{\Gamma_{zy}} \overbrace{\frac{\tau_y}{\tau_y+\tau_z}}^{T_{zy}} \sigma_y^2}_{\text{upstream from } y} \quad (8)$$

The intrinsic component is generated by the randomness in the birth-death processes and it can be approximated by a Poisson process (Bar-Even et al., 2006; Newman et al., 2006). The upstream component is the noise transmited from the upstream species (the species that regulate the gene) (Pedraza and van Oudenaarden, 2005). The upstream noise is composed of three terms: the regulation factor ($\Gamma_{zx}$ and $\Gamma_{zy}$), the time average factor ($T_{zx}$ and $T_{zy}$), and the variance of the upstream species ($\sigma_x^2$ and $\sigma_y^2$).

In this contribution, we are interested in how noise affects our ability to distinguish between the two known output states, $H$ and $L$. To get a meaningful measure of this, we will normalise the variance by the square of the signal strength, $\eta_z \doteq \sigma_z^2/(H-L)^2$, rather than by the square of the mean (which is often used as a definition of noise).

$$\eta_z = \frac{z}{(H-L)^2} + \left[\beta_z\tau_z\frac{\partial f(x,y)/\partial x}{(H-L)}\right]^2 T_{zx}\sigma_x^2 + \left[\beta_z\tau_z\frac{\partial f(x,y)/\partial y}{(H-L)}\right]^2 T_{zy}\sigma_y^2 \quad (9)$$

For step-like regulation function the derivatives in (9) will be zero, and the only contribution to the noise is the intrinsic component. Thus, the noise of the output depends only on the steady state abundance (high and low), but is independent of the number of gates in the system or of the threshold position. However, if the threshold is close enough to one of the steady states ($H$ or $L$), then small fluctuations in the input generates high fluctuations in the output and the analytical method is not accurate any-more. Assuming that the threshold is positioned at the midpoint (optimum position for speed) and the two steady states are far enough from each other, then the noise will be determined only by the intrinsic component. Hence, in the case of step-like regulation functions, the system displays an optimum threshold position ($\lambda = 0.5$) which ensures optimality both for speed and accuracy.

### Finite Hill Coefficients

Due to the fact that Hill coefficients are bounded above by the number of regulatory binding sites (Chu et al., 2009), and genes have a small number of binding sites (Hermsen et al., 2006), biologically realistic Hill coefficients are finite and have low values.

For low Hill coefficients, Eq. (4) has only one solution, $H = L$. This is not a useful solution because it removes the binary logic. Therefore, we search for parameters which ensure that the signal strength is not reduced, $(H_{out} - L_{out}) \geq (H_{in} - L_{in})$, and this can be achieved by solving only the first two equations in Eq. (4):

$$\begin{aligned} \frac{\alpha_{OR}}{\mu} &= \frac{Lf_{OR}(L,H) - Hf_{OR}(L,L)}{[f_{OR}(L,H) - f_{OR}(L,L)]}, \\ \frac{\beta_{OR}}{\mu} &= \frac{H-L}{[f_{OR}(L,H) - f_{OR}(L,L)]}. \end{aligned} \quad (10)$$

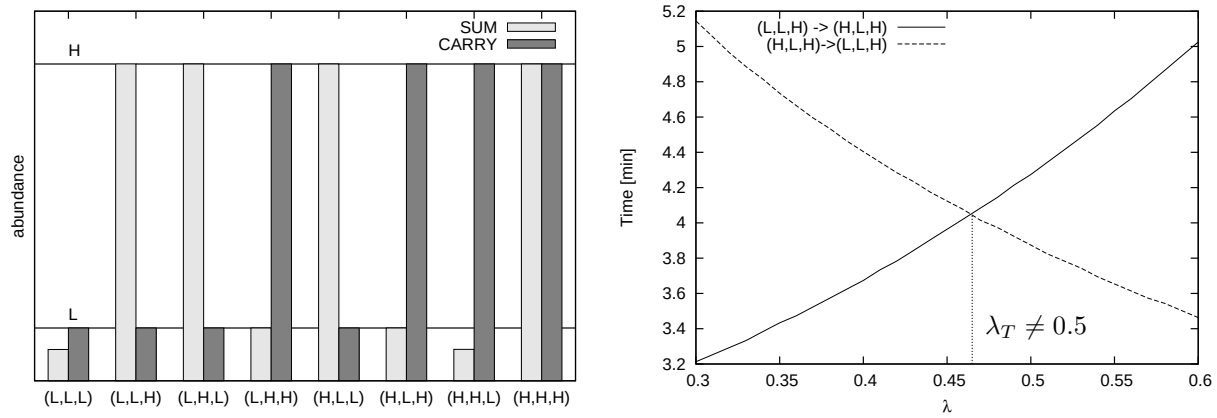
Note that not for all sets of parameters $(l, K, \mu, H, L)$ the synthesis rates will have positive values. Interestingly, increasing the Hill coefficient increases the space of allowed parameters, and in the limit case of a step function ($l \to \infty$) any values of the other parameters will generate positive synthesis rates. For Hill coefficient less than or equal to 1 there is no solution for this system. Analogously one could use the same mechanism to determine the synthesis rates for all the other gates. For AND and XOR gates the solution is given by

$$\begin{aligned} \frac{\alpha_{AND}}{\mu} &= \frac{Lf_{AND}(H,H) - Hf_{AND}(L,H)}{[f_{AND}(H,H) - f_{AND}(L,H)]} \\ \frac{\beta_{AND}}{\mu} &= \frac{H-L}{[f_{AND}(H,H) - f_{AND}(L,H)]} \end{aligned} \quad (11)$$

$$\begin{aligned} \frac{\alpha_{XOR}}{\mu} &= \frac{Lf_{XOR}(L,H) - Hf_{XOR}(H,H)}{[f_{XOR}(L,H) - f_{XOR}(H,H)]} \\ \frac{\beta_{XOR}}{\mu} &= \frac{H-L}{[f_{XOR}(L,H) - f_{XOR}(H,H)]} \end{aligned} \quad (12)$$

Fig. 5(a) confirms that the signal is not decreased and shows that in two cases the actual output low state ($L_{out}$) is lower than the desired one ($L$).

**System Performance** For low Hill coefficients the optimum threshold in terms of speed in not positioned any more at the midpoint between high state and low state (see Fig. 5(b)). This is a consequence of the fact that for low Hill coefficient the Hill function loses the symmetry around the threshold. Hence, when designing a specific system, one could use numerical solutions to determine the




(a) steady state

(b) optimum $K$ for speed

Figure 5: *Full adder with low Hill coefficients*. (a) The output abundance based on the input abundance for low Hill coefficients. (b) We plotted the propagation time when switching between $(L, L, H)$ to $(H, L, H)$ for low Hill coefficient. The following set of parameters have been used: $\mu = 1\ min^{-1}$, $l = 6$, $L = 0.2\ \mu M$, $H = 1.2\ \mu M$, $K = 0.7\ \mu M$ and $\theta = 0.5$.

optimal threshold position for any specific set of parameters. Also, one can notice that decreasing the Hill coefficient increases the propagation time due to the fact that a gene is not instantly turned on/off when an input species crosses over/under the threshold (compare Fig. 3(b) and Fig. 5(b)). Increasing the Hill coefficient asymptotically reduces the propagation time to the one of the step-like regulation function and, thus, the optimal threshold asymptotically approaches the midpoint, $\lambda_T = 0.5$ (data not shown).

Next, we investigated the accuracy of the full-adder. The output sum for the input $(H, L, L)$ produces the highest noise levels independent of the threshold position. Considering this case we determined the dependence of noise on the threshold position. The mathematical formula of the noise is too complicated to give any information about the system, but we can use it to generate numerical solutions. Fig. 6(a) shows that there is an optimal position of the threshold in terms of noise which differs from the optimal position in terms of speed, $\lambda_\eta \neq \lambda_T$. However, around the optimal threshold position in terms of noise ($\lambda_\eta$) the noise does not vary significantly (see Fig. 6(a)).

The system displays two optimal threshold positions, one for speed ($\lambda_T$) and one for noise ($\lambda_\eta$). If these two positions coincide ($\lambda_T = \lambda_\eta$) then the system has on optimal set of parameters and the engineer needs to set up the threshold to this position.

However, it is most likely, that these two threshold positions will differ, as it is the case with our full-adder. In this case, there is an optimal trade-off curve when the threshold resides between these two optimal positions ($\lambda_T$ and $\lambda_\eta$). In addition any other trade-off curve is suboptimal comparing to this one.

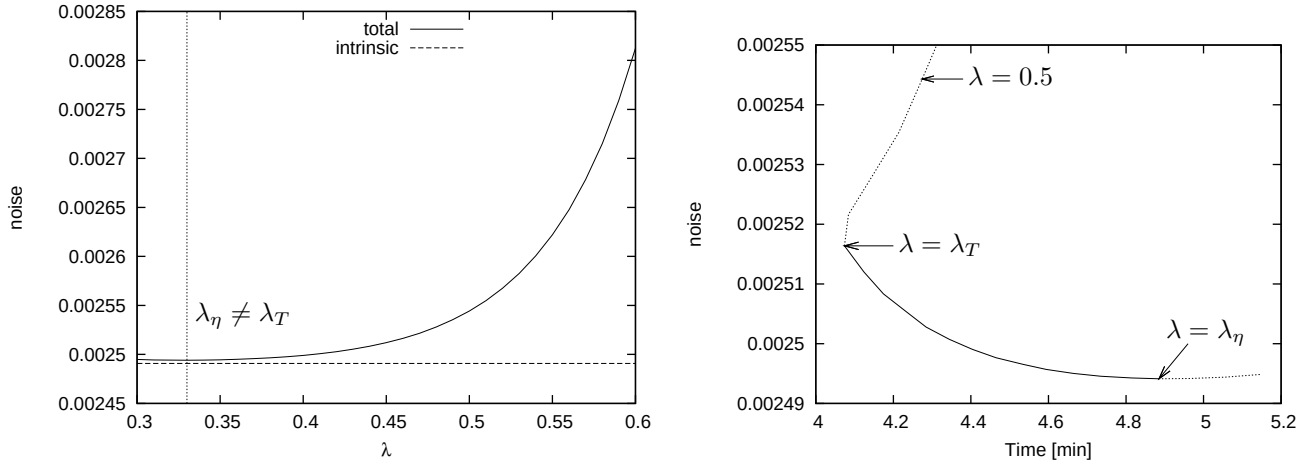
In our example of the full adder $0.5 \leq \lambda_\eta \leq \lambda_T$. Fig. 6(b) graphically represents the trade-off between noise and time based on the threshold position. We identified the optimal trade-off curve determined by $\lambda_\eta \leq \lambda \leq \lambda_T$. Any threshold in this interval can optimize the system either in speed or in accuracy, but never in both. However, for threshold positions outside this interval the system display sub-optimal trade-off curves; for $\lambda < \lambda_\eta$ or $\lambda > \lambda_T$ both the propagation time and the noise are worst compared to the ones in the optimal trade-off curve.

## Discussion

In this contribution, we presented a general method for constructing arbitrarily large logical systems based on binary genes. For exemplification purpose, we designed a full-adder system formed of five genes. The approach modelled logic gates constructed using two cis-regulatory transcription control regions. This type of logic gates has been already synthetically engineered by synthetic biologists (Guet et al., 2002; Kramer et al., 2004; Yokobayashi et al., 2002; Cox III et al., 2007; Anderson et al., 2007; Sayut et al., 2009). We propose the tuning of the synthesis/decay rates in such a way that will permit interconnectivity of different gates/genes. This tuning represents basic requirement for a correct functioning of the logic system.

Recently we showed that leak free systems are optimal in terms of speed and noise (Zabet and Chu, 2010). However, Eq. (10) and Eq. (11) indicate that basal vanishing leak rates are very difficult to obtain. This suggests that leak free systems, although optimal in speed and noise are not always desirable, because they are likely to reduce the signal strength when thinking about interconnecting genes.

We also presented here an approach for selecting the set of parameters which optimizes the system in terms of speed




(a) optimum $K$ for noise

(b) trade-off curves

Figure 6: *Optimum $K$ for noise*. (a) The noise dependence on the threshold. The following set of parameters have been used: $V = 8 \times 10^{-16}\ l$, $\mu = 1\ min^{-1}$, $l = 6$, $L = 0.2\ \mu M$, $H = 1.2\ \mu M$, $K = 0.7\ \mu M$ and $\lambda = 0.5$. We assumed a Poisson noise of the three input species.

and accuracy under constant metabolic cost. Increasing the Hill coefficient will optimize both the speed and the accuracy, but this is not usually at the direct reach of synthetic biologists. However, the threshold can be altered by mutations of the regulatory binding sites (Buchler et al., 2005). We show that the threshold position, for a fixed Hill coefficient, influences both the speed (see Fig. 5(b)) and the noise (see Fig. 6(a)).

In an ideal system, a system with gates that display step-like regulation functions (infinite Hill coefficients), we found that the system has an optimal set of parameters (threshold positioned at the midpoint between the two steady states). This set of parameters maximizes both speed and accuracy for a fix cost. Moreover, the speed and the accuracy achieved in this type of system is the asymptotic limit that any biological real system can aim towards.

Real genes have finite low Hill coefficients and, in this case, a logic system will display two optimal sets of parameters: one in speed $\lambda_T$ and another one in noise $\lambda_\eta$. We found that there is a trade-off curve between speed and accuracy which is bounded by these optimal sets of parameters ($\lambda_T$ and $\lambda_\eta$) and any point between these two can optimize the system in either speed or accuracy. Nevertheless, any other set of parameters (the threshold outside this interval) is sub-optimal with respect to accuracy or speed.

This analysis showed that for finite low Hill coefficients there are two sets of parameters, one optimizing in terms of speed and the other on in terms of noise, when the metabolic cost is not increased. However, this analysis addressed only logic gates formed of individual genes. It was widely recognized, that network motifs can play a significant role in both speed and noise (Alon, 2007). Thus, further optimization can be achieved by considering logic gates built from more than one genes that form a network motif. Nevertheless, the details of this analysis need to be left for further research.

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