# Mechanism for signal adaptation in cells

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**Abstract**: Cells perceive different stimuli and respond through a variety of dynamic behaviors. One of these interesting responses is perfect adaption, where the cell can detect a change in a stimulus and give a response. Nevertheless, this response is only transient and the cell returns to basal levels of activity after a certain time. Our objective is to describe different circuits that exhibit this behavior. Using numerical methods, we characterize the common and distinct features exhibited by these different systems.

## I. INTRODUCTION

Understanding the response mechanisms of cells to external and internal stimuli is a problem that involves an enormous number of variables. The complex networks that are created by interacting genes and proteins make it impossible to describe their dynamical properties by intuitive reasoning alone. Nonetheless, some recent theoretical and computational studies have shown that these networks can be properly modeled in mathematical terms [1]. This has led to the identification of a variety of circuits (i.e., networks of few genes and proteins) that are able to account for the dynamics observed experimentally [2]. These circuits can involve both spatial and temporal coordinates, which can be analyzed independently. The main focus will be the temporal dynamics of the systems but we will also do some simplifications and considerations to be able to do an analysis of the spatial dynamics.

Adaptation is a key feature of cellular signaling and regulatory pathways. Adaptation is defined as a process where a system responds to a stimulus, but the response is temporal, and the system returns to basal or near-basal levels after a certain time. If this return is complete and the system ends at the exact same level as initially, the behavior of the system is called perfect adaptation. Mathematically, this could be described as the steady state of the response element of a system being independent of the stimuli.

A common example of perfect adaptation is sensory signal transduction. After a certain time smelling a new scent, we stop perceiving it even though is still there. But if we leave and come back later, we will again sense it. This example could be described as an incoherent feedforward system. More specific examples are bacterial chemotaxis for the negative feedback loop and ion channel activation and inactivation for the state-dependent inactivation system.

Our objective is to characterize and analyze three different circuits that can achieve perfect or near-perfect adaptation. These are incoherent feedforward systems, negative feedback loops, and state-dependent inactivation systems. The description of the dynamics of systems will be done through the mass-action law, following the review article [2].

Regarding the units used in this work, we will be taking the following considerations. For the time variable, seconds or minutes will be used depending on the timescale associated with the biological processes involved in every type of circuit. For the concentration variable, adimensional units will be used for simplification purposes. This could be interpreted as every concentration value is divided by a characteristic constant concentration $C_o$ = 1 μM and therefore the obtained values have no unit associated with them.

## II. INCOHERENT FEEDFORWARD SYSTEMS

We will start with a circuit that exhibits perfect adaptation and was introduced in [3]. Our system is made of two molecular species, A and B, that are dependent on an input [I]. Molecule A regulates positively the response (or output) while B inhibits A, and thereby the output. All these processes could be thought of as the synthesis and degradation of these two protein species. The entire mechanism is described by these two equations:

$$\frac{d[A]}{dt} = k_1[I] - k_2[A][B] \quad (1)$$

$$\frac{d[B]}{dt} = k_3[I] - k_4[B] \quad (2)$$

In this case, we will use minutes as the time unit because the timescale of the biological processes of synthesis and degradation described by these equations is in the range of a few minutes to hours. Here $k_1$, $k_2$, $k_3$, and $k_4$ correspond to rate constants with units of $min^{-1}$. The first term of the first equation, dependent on the input, stimulates the synthesis of the specie A and the second term, dependent on the concentration of species A and B stimulates the degradation of A. In the second equation, the first term, dependent on the input, stimulates the synthesis of B, and the second term, dependent on B, stimulates the degradation of B.

Figure 1 shows the results from simulations of the dynamics over time of [A] and [B] as the input increases as step functions. From Fig. 1 we see that a sudden increase in the input causes an abrupt increase in the output [A] that returns toward the baseline monotonically. This is the perfect

adaption. Moreover, every subsequent increase in the input produces weaker and weaker responses. This is due to the increasing amount of inhibitor [B] in the system after every increase. The inhibitory response is every time stronger and the response of the output is smaller.

The results can also be readily understood by a simple analysis of the equations. We first find the stationary state, by equating both equations to 0. From the second equation, we can find directly the stationary value of [B], [$B_{ss}$], then we substitute this value in the first equation and find the stationary value of [A], [$A_{ss}$]. Finally, we find the values for [$A_{ss}$] and [$B_{ss}$]:

$$[A_{ss}] = \frac{k_1 k_4}{k_2 k_3} \ , \ \ [B_{ss}] = \frac{k_3 [I]}{k_4} \qquad (3)$$

We see that the steady state of the output $[A_{ss}]$ doesn't depend on the input [I]. This explains why [A] returns to its basal level in Fig.1. A stationary output that is independent of the input is required for perfect adaptation.

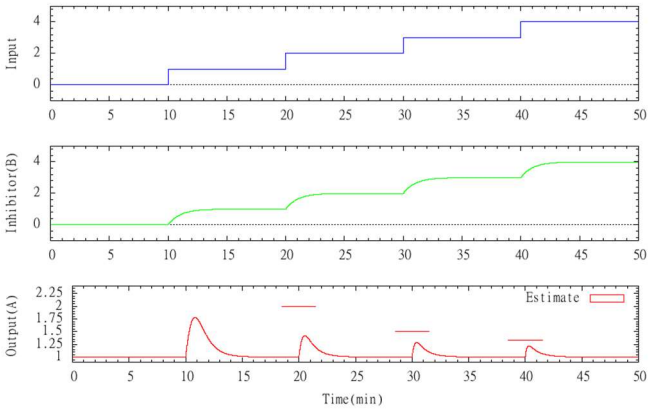


FIG. 1: Dynamics of the incoherent feedforward system for synthesis and degradation processes. Evolution of the output ([A], red) with the estimated value obtained from Eq. 5 (horizontal bars in bottom panel) for every increase and the inhibitor ([B], green) when the concentration of the input ([I], blue) increases an amount [ΔI] equal to 1 every 10 minutes. Parameter values: $k_1 = 2$ min$^{-1}$, $k_2 = 2$ min$^{-1}$, $k_3 = 1$ min$^{-1}$, $k_4 = 1$ min$^{-1}$. Adimensional concentrations are used.

We can also predict the maximum height of the output for an increase $\Delta I$ in the input. For that, we analyze the dynamics right after the increase in the input. We assume that A and B follow very different time scale dynamics, being A very fast and B very slow. Thus, when the maximum value of [A] is reached, the value of [B] remains the same as before the increase of the input, i.e., $[B_0] = [B_{ss}(I_0)]$. So, we have the following changes in our variables:

$$[A] \rightarrow [A_0] + [\Delta A], \quad [B] \rightarrow [B_0], \quad [I] \rightarrow [I_0] + [\Delta I] \qquad (4)$$

Then, we substitute these changes in our system of equations and equate the equation of [A] to zero (i.e. imposing it is a fast variable). We find the variation of the output with respect to the basal level:

$$[\Delta A] = \frac{k_1 k_4}{k_2 k_3} \frac{[\Delta I]}{[I_0]} \qquad (5)$$

From this equation, we see that the variation of the output depends on a relation between the parameter values and is proportional to the ratio between the increase of input, ΔI, and the last input value, $I_o$. In this case, $I_o$ changes after every increase and takes the value of the last input value and not only the initial one. For the first increase, we can't use this expression because $I_0$=0 and the equation diverges. The results of this prediction are plotted in Fig. 1 (bottom panel).

An interesting case is when we make $k_3$ and $k_4$ a lot smaller, so the response of molecule B is a lot slower (Fig. 2). We chose to keep the ratio $k_4/k_3$ fixed so that the predictions of Eq. (5) are the same values as for faster dynamics of B. As expected, when B has slower dynamics, the predictions match better computational results.

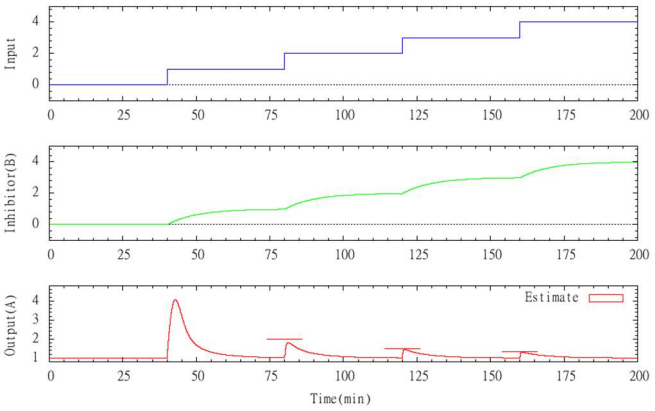


FIG. 2: Dynamics of the incoherent feedforward system for synthesis and degradation processes when the inhibitor is slow. Evolution of the output ([A], red) with the estimated value obtained from Eq. 5 (horizontal bars in bottom panel) for every increase and the inhibitor ([B], green) when there is an increase in the concentration of the input ([I], blue) an amount [ΔI] equal to 1 every 40 minutes. Parameter values: $k_1 = 2$ min$^{-1}$, $k_2 = 2$ min$^{-1}$, $k_3 = 0.1$ min$^{-1}$, $k_4 = 0.1$ min$^{-1}$.

We can now analyze an alternative description of an incoherent feedforward circuit, introduced in [2]. In this case, we can think of this process as the activation and inactivation of a protein A. Active A sets the output. The input [I] activates A and B, which inactivates A. Here, the appropriate biological timescale is in the order of seconds, so we are going to use seconds as our time unit. This system is described by the following equations:

$$\frac{d[A]}{dt} = k_1[I](1-[A]) - k_2[A][B] \quad (6)$$

$$\frac{d[B]}{dt} = k_3[I]\frac{1-[B]}{k_5+1-[B]} - k_4[B] \quad (7)$$

In this case, we have conserved the total amounts of each protein, which are normalized to 1. We can find the stationary state. If we equate both equations to 0 and solve the system as we have done before, we obtain that:

$$[A_{ss}] = \frac{k_1k_4}{k_1k_4+k_2k_3}\ ,\ [B_{ss}] = \frac{k_3[I]}{k_4} \quad (8)$$

To find this stationary state we have made the approximation that $k_5 \ll 1-[B]$. As before, we see that the output doesn't depend on the input. We can also apply the same proceeding as described before to predict the height of the output. In this case, we obtain the following relation:

$$[\Delta A] = \frac{k_1k_4}{k_1k_4+k_2k_3}\frac{[\Delta I]}{[I_0]}(1-[A_0]) \quad (9)$$

From this equation, we see that the variation of the output depends on a relation between the parameter values and is proportional to the ratio between the increase of input, $\Delta I$, and the last input value, $I_o$. The expression is also proportional to one minus the initial value of protein A, $A_0$. We see that the expression is like Eq. (5) with an additional term. For the first increase, we can't use this expression because $I_0=0$ and the equation diverges.

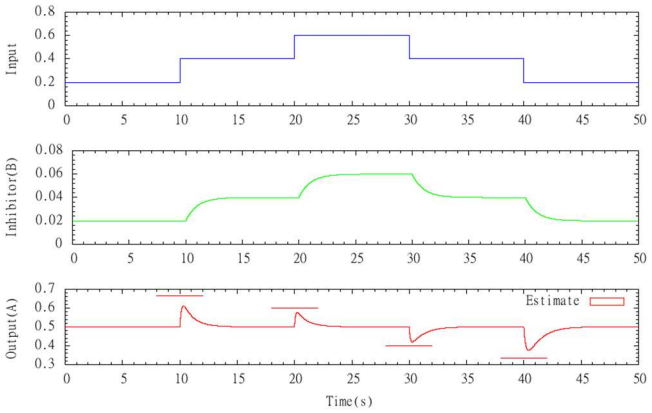


FIG. 3: Dynamics of the incoherent feedforward system for activation and inactivation processes. Evolution of the output ([A], red) with the predicted value obtained from Eq. 9 for every increase and the negative leg ([B], green) when there is a variation of the concentration of the input ([I], blue) an amount [ΔI] equal to 0.2 every 10 seconds. Parameter values: $k_1 = 10\ s^{-1}$, $k_2 = 100\ s^{-1}$, $k_3 = 0.1\ s^{-1}$, $k_4 = 1\ s^{-1}$, $k_5 = 0.001$.

An alternative representation of the circuit dynamics can be done by plotting in the phase space ([B], [A]), as presented in [1]. The trajectory of [A] and [B] when the input changes from $I_0$ to $I_f$ is depicted (Fig. 4). In this phase space, the nullclines for each input value, $I_0$ and $I_f$, are also depicted. Each nullcline represents one of the system equations equalized to 0. The intersection of both defines the stationary solution. The nullclines for the original input $I_0$ are depicted with continuous lines while dashed lines correspond to the nullclines for the final input $I_f$. The stationary solution for each input value is highlighted with black and grey dots, respectively. The vector field is the vector of the time derivative for each variable taking the input value $I_f$. The color expresses the magnitude of the module of these derivatives. Finally, the blue line corresponds to the trajectory of the system variables when the input is changed from $I_0$ to $I_f$ and is tangent to the vector field.

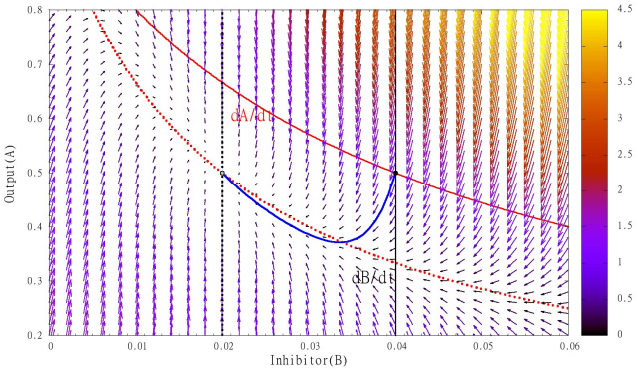


FIG. 4: Phase diagram of the incoherent feedforward system with activation and inactivation processes. The two input values are $I_0 = 0.4$ and $I_f = 0.2$. Red nullclines correspond to [A] and black nullclines correspond to [B], in each case, the solid one is for the initial input $I_0$ and the dashed one for the final input $I_f$. The vector field corresponds to the final input, its color represents the magnitude of the change, and it is graduated on the right-side scale. The blue line corresponds to the trajectory from the initial state (the stationary one for $I_0$) to the stationary state for $I_f$.

## III. NEGATIVE FEEDBACK LOOPS

The negative feedback loop consists of two proteins as in the previous one. In this case, the proposed circuit is slightly different than the previous one. An input stimulus activates output A which in turn activates B. Then, B mediates the inactivation of A. Thus, the delayed response from protein B returns the output to near a basal level. This return could be done monotonically or with damped oscillations depending on the particular conditions of the system. We use the circuit presented in [2]. The equations that describe this system are the following:

$$\frac{d[A]}{dt} = k_1[I](1-[A]) - k_2[A][B] \quad (10)$$

$$\frac{d[B]}{dt} = k_3[A]\frac{1-[B]}{k_5+1-[B]} - k_4\frac{[B]}{k_6+[B]} \quad (11)$$

As before, the total concentration of active and inactive forms of each protein is conserved and set to 1. Timescale for these activation and inactivation processes is in the range of a few seconds. The stationary state for this system follows the same mathematical procedure as before is:

$$[A_{ss}] = \frac{k_4}{k_3}\ ,\ [B_{ss}] = \frac{k_1k_3 + k_1k_4}{k_2k_4}[I] \quad (12)$$

In this case, we have made the approximation that $k_5 \ll 1-[B]$ and $k_6 \ll [B]$. Under this approximation, the output is independent of the input, as is required for perfect adaptation. But as we see in Fig. 5, where results from numerical simulations of the dynamics are shown, the system for finite values of $k_5$ and $k_6$ has a near-perfect adaptation behavior. So, the approximation that have been made do not capture the non-perfect adaptation behavior.

The expression that gives an estimation of the maximum height of the output following the previous procedures and applying the previous approximations is:

$$[\Delta A] = \frac{k_4[\Delta I](1-[A_0])}{k_4[\Delta I] + k_3[I_0]} \quad (13)$$

We see that this equation is different from the previous ones, (5) and (9). The numerator depends on the increase of input and initial value of protein A and the denominator depends on ΔI and the previous value of the input, $I_0$. In this case, we can obtain predictions for all of the increases as the equation doesn't diverge for any input value.

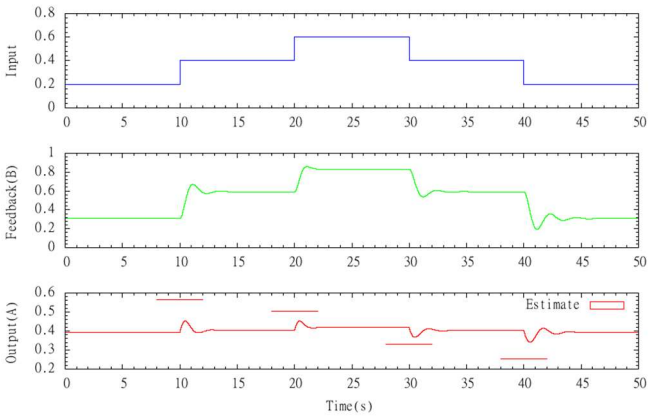


FIG. 5: Dynamics of the negative feedback loop. Evolution of the output ([A], red) with the predicted value obtained from Eq. 13 for every increase and the feedback ([B], green) when there is a variation of the concentration of the input ([I], blue) an amount [ΔI] equal to 0.2 every 10 seconds. Parameter values: $k_1 = 2\ s^{-1}$, $k_2 = 2\ s^{-1}$, $k_3 = 10\ s^{-1}$, $k_4 = 4\ s^{-1}$, $k_5 = 0.01$, $k_6 = 0.01$.

As we see (Fig. 5) the return to the basal level is done through damped oscillations. In Fig. 6 we will see the effects of these oscillations. Moreover, as we have already discussed, the system doesn't return exactly to the previous basal level (i.e., the output depends slightly on the input), in opposition to Eq. (12) which says that the output doesn't depend on the input. With the approximations we have done to obtain Eq. (12), we have lost this behavior.

A phase diagram for this system can also be represented (Fig. 6) and obtain a lot of information about the behavior of this system. First, we see that damped oscillations are represented by the multiple crossings of the nullclines that the trajectory of the stationary state, the blue line, does while reaching the final state. Second, we see that the initial stationary state and the final are not along a straight line parallel to the x-axis. This means that we only have a near-perfect adaptation and the system doesn't go back to exactly the same basal level after each variation of the input. As we have seen before the output depends slightly on the input.

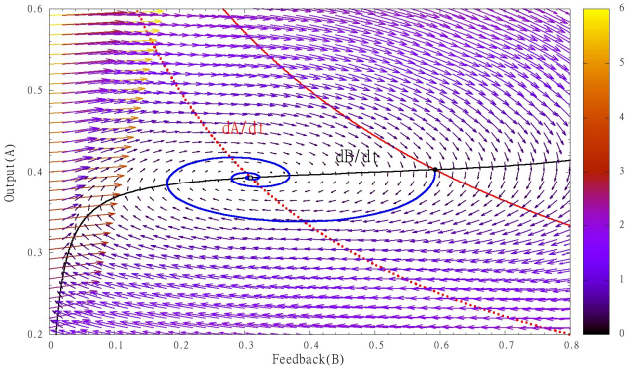


FIG. 6: Phase diagram for the negative feedback system. The two input values are $I_0 = 0.4$ and $I_f = 0.2$.

## IV. STATE-DEPENDENT INACTIVATION SYSTEMS

The last example is slightly different from the previous cases. It could be understood as a system of a protein that could transition between three states. In the first state, the off state ($A_{off}$), the proteins are waiting for an input that will trigger its activation. In the second state, $A_{on}$, the proteins are in the activated state after receiving an input and expressing an output. In the third state, $A_{in}$, the proteins are inactivated and aren't able to respond to any stimulus nor drive any output. As previously explained, the timescale involved in this type of process is of the order of seconds. The state-dependent inactivation mechanism for adaptation was introduced in [4]. Herein we use the approach reviewed in [2].

In this approach, the model only describes the transition between the $A_{off}$ and $A_{on}$ states and between $A_{on}$ and $A_{in}$ states. It could also be modeled an additional slow conversion back to the off state, with the addition of a third equation [4], but in the model studied is not considered. The total amount of the protein A, $A_{tot}$, is normalized to one and

remains constant throughout the process. The equations that regulate that simplified system are:

$$\frac{d[A_{on}]}{dt} = k_1[I](1 - [A_{on}] - [A_{in}]) - k_2[A_{on}] \quad (14)$$

$$\frac{d[A_{in}]}{dt} = k_2[A_{on}] \quad (15)$$

From the second equation, we see that the variation of $A_{in}$ is proportional to the amount of $A_{on}$. From the first equation, we see the normalization of the amount of protein A in the term between brackets and the variation of $A_{on}$ comes from a difference between $A_{off}$ and $A_{on}$. The stationary state for this system is:

$$[A_{on_{ss}}] = 0 \ , \ [A_{in_{ss}}] = 1 \quad (16)$$

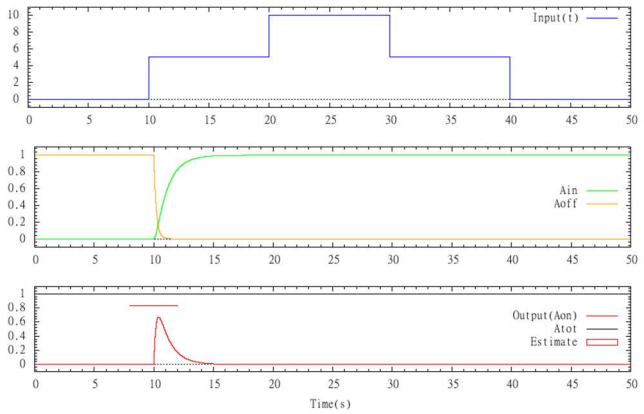


FIG. 7: Dynamics of the state-dependent inactivation system. Evolution of the output, i.e., the activated protein ($[A_{on}]$, red) with the estimated value obtained from Eq. 17, the protein in the inactivated state ($[A_{in}]$, green), the protein in the off state ($[A_{off}]$, yellow) and the total amount of protein A ($[A_{tot}]$, black) when there is a variation of the concentration of the input ([I], blue) an amount [ΔI] equal to 5 every 10 seconds. Parameter values: $k_1 = 1\ s^{-1}$, $k_2 = 1\ s^{-1}$.

Following the same procedure as before, we can find the expression that gives an estimation of the maximum height:

$$[\Delta A] = \frac{k_1[\Delta I](1 - [A_{on_0}] - [A_{in_0}])}{k_1([I_0] + [\Delta I]) + k_2} \quad (17)$$

We see that this equation is similar to the previous one, (13). The numerator depends on the increase of input and initial value of activated and inactivated protein and the denominator depends on ΔI and the previous value of the input, $I_0$.

From Fig. 7, we see that after the first increase, the system can't respond to any further increase of the input as all the proteins are in the inactivated state, $A_{in}$.

## V. CONCLUSIONS

- For all the circuits studied, we have reproduced the results in [2]. We have also incorporated the phase plane analysis done in [1] for the two first cases. Finally, we have introduced the estimations of the height of the output that isn't in any of the papers used.

- As we have seen all of the circuits respond with the behavior of perfect adaptation. However, the state-dependent inactivation system can't respond to multiple increases of the input, due to the exhaustion of $A_{off}$. After the first increase, there are no molecules remaining in the off state and the system can't produce more of them as there is no recovery process. On the other hand, the negative feedback loop is the only one that returns to the basal level through damped oscillations and has a near-perfect adaptation instead.

- All the circuits studied have one essential aspect in common. A fast time scale for activation and a slow one for inactivation to let the system produce the desired behavior.

### Acknowledgments

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[1] Ma, W., Trusina, A., El-Samad, H., Lim, W.A., and Tang, C. Defining network topologies that can achieve biochemical adaptation. Cell 138, pp. 760–773, 2009.

[2] Ferrell, James E. Perfect and Near-Perfect Adaptation in Cell Signaling. Cell systems, vol 2., pp. 62-67, 2016.

[3] Tyson, J. J., Chen, K. C., Novak, B. Sniffers, buzzers, toggles and blinkers: dynamics of regulatory and signaling pathways in the cell. Current Opinion in Cell Biology, vol 15., pp. 221-231, 2003.

[4] Friedlander, T., and Brenner, N. Adaptive response by state dependent inactivation. Proc. Natl. Acad. Sci. USA 106, 22558–22563, 2009.