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RESEARCH ARTICLE

Self-organization of signal transduction [v1; ref status: indexed, http://f1000r.es/zg]

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Abstract

We propose a model of parameter learning for signal transduction, where the objective function is defined by signal transmission efficiency. We apply this to learn kinetic rates as a form of evolutionary learning, and look for parameters which satisfy the objective. This is a novel approach compared to the usual technique of adjusting parameters only on the basis of experimental data. The resulting model is self-organizing, i.e. perturbations in protein concentrations or changes in extracellular signaling will automatically lead to adaptation. We systematically perturb protein concentrations and observe the response of the system. We find compensatory or co-regulation of protein expression levels. In a novel experiment, we alter the distribution of extracellular signaling, and observe adaptation based on optimizing signal transmission. We also discuss the relationship between signaling with and without transients. Signaling by transients may involve maximization of signal transmission efficiency for the peak response, but a minimization in steady-state responses. With an appropriate objective function, this can also be achieved by concentration adjustment. Self-organizing systems may be predictive of unwanted drug interference effects, since they aim to mimic complex cellular adaptation in a unified way.

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Introduction

Signal transduction systems are often modeled as networks of biochemical kinetic equations implemented as continuous-time dynamical models using differential equations[1,2]. If we regard a subset of species as inputs, and make sure that the system always converges to equilibrium values by using weakly reversible equations[3–5], we may transform these models into a set of matrices fulfilling the role of input-output transfer functions, i.e. a mapping from sustained input signal levels to steady-state concentrations for all target species[6]. Protein signaling functions (psfs) are a systemic generalization of individual dose-response functions, which are usually described by Hill equations[7]. In contrast to Hill equations, which are not available for enzymatic reactions, which only calculate relative concentrations, and which only work for one reaction in isolation, the psf system calculates enzymatic and complex formation reactions in a complex systemic environment using absolute concentrations[6]. In addition, the reaction times to equilibrium are calculated as delay values, and the dynamic shape ("transients") is also available for further analysis.

In this paper, we want to ask the question of optimality of signal transduction. From an evolutionary standpoint, we assume that any biological signal transduction system is constructed with optimized efficiency of signal transmission. Furthermore, we assume that cells have the ability to adapt to perturbations of protein concentrations and changes in extracellular signaling by reinstating signal transmission efficiency. In the following, we investigate this question using a biologically realistic system – beta-adrenergic signaling in a submembrane compartment of a mouse embryonic fibroblast for a single input scenario, focusing on a selected target species as relevant output or actuator of the system (Figure 1A).

Experimental analysis of signal transduction systems has shown fold-change responses to changes in input[8,9]. Accordingly, input-output transfer functions usually follow the shape of hyperbolic (saturating) curves, which are equivalent to sigmoids for logarithmically scaled input[6]. In Figure 1B we show the effect of a knock-out (KO) for a RGS protein in an experimental assay in yeast[10], and compare this with the effect in the model system[11]. We see that the effects of the RGS KO on dose-response signaling efficiency are robust across very different cellular systems.

Usually, when we use a computational model to investigate perturbations, we only study the effects as reflected in the simulation. By utilizing optimization in terms of signaling efficiency, we can make the system itself adjust to the perturbation. In this way, we are studying signal transduction as a self-organizing system, which uses objective functions to adapt. This basic idea is extremely powerful, and could be used with different kinds of constraints on parameters, reaction times, etc. and with different, multiple input scenarios for larger systems. To explore this question further is of significant importance in assessing cellular health and functioning.

Methods

Example system: GPCR signaling in a submembrane compartment

Figure 1A shows the example system, a submembrane compartment with a GPCR (G-protein coupled receptor) pathway from a mouse embryonic fibroblast, with ISO as input (extracellular ligand to $\beta(2)$-adrenergic receptors) and the phosphorylation of a protein VASP as output. This model was implemented as an ODE model with 23 reactions and 27 molecular species, derived from 12 initial concentrations (cf. Table 1, Table 2). The parameters were adapted to experimental biological data (not shown[11]). In this subsystem, the central cAMP response often follows a plateau curve, i.e. a rise to steady-state, but cAMP transients which are typically observed in cytoplasm may also occur[11]. The dose-response transfer functions were derived as in[6].
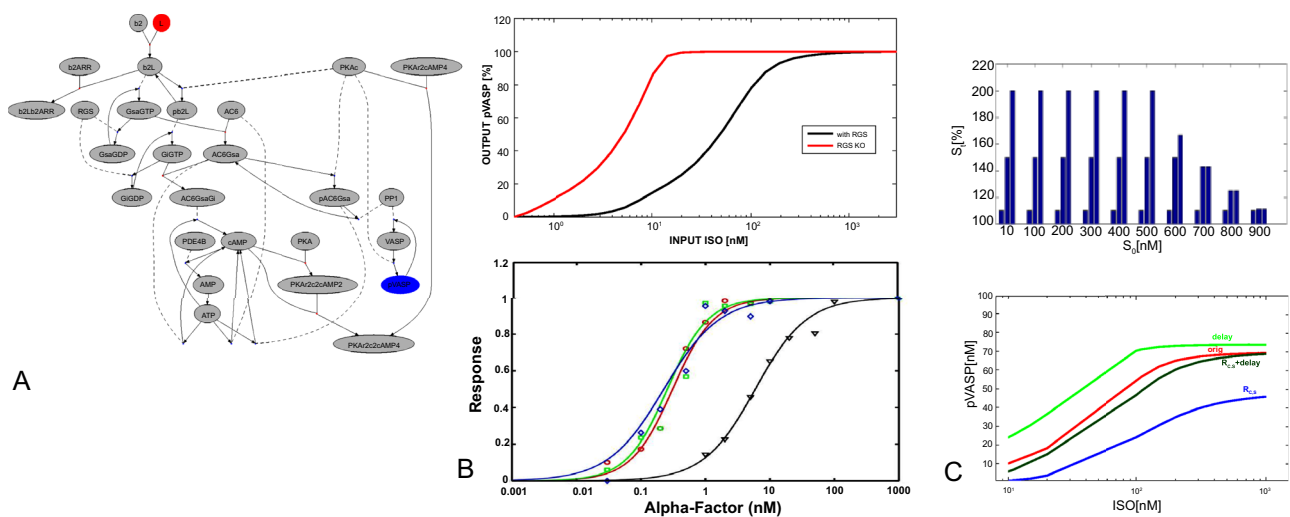
Figure 1. A. Biochemical Reaction System with Selected Input (red) and Output (blue) **B**. (Top) ISO-pVASP transfer function (with RGS KO, red) in the MEF model[11] (Bottom) Experimental Response to RGS KO for GPCR signal-response in a yeast model[10] **C**. (Top) Distribution of Extracellular Signals Used to Calculate Signal Transmission. From a baseline signal S_0 [10nM–900nM] the extracellular signaling level S_t rises to 110%,150% and 200%, but not above 1μM . (Bottom) Transfer Functions for ISO → pVASP. Shown is the original model, optimization by delay, by $R_{C,S}$, and by both.

Table 1. Kinetic rates of the sample system, adapted from Sabio-Rk[25] and Brenda[26], adjusted to experimental cAMP time-series data[11].

	k_{on}	k_{off}	k_{cat}
$b2 + L \leftrightarrow b2L$	0.0003	0.1	
$b2L + PKAc \leftrightarrow b2LPKAc \rightarrow pb2L + PKAc$	0.00026	1	5.4
$pb2L \rightarrow b2L$			0.1
$GsaGDP + b2L \leftrightarrow b2LGsaGDP \rightarrow GsaGTP + b2L$	0.006	0.8	0.2
$GsaGTP + RGS \leftrightarrow RGSGsaGTP \rightarrow GsaGDP + RGS$	0.0008	1.2	16
$GiGDP + pb2L \leftrightarrow pb2LGiGDP \rightarrow GiGTP + pb2L$	1.2	0.8	16
$GiGTP + RGS \leftrightarrow RGSGiGTP \rightarrow GiGDP + RGS$	1.2	0.8	16
$GsaGTP + AC6 \leftrightarrow AC6Gsa$	0.00385	3	
$GiGTP + AC6Gsa \leftrightarrow AC6GsaGi$	0.00385	10	
$AC6Gsa + PKAc \leftrightarrow AC6Gsa\_PKAc \rightarrow pAC6Gsa + PKAc$	0.00026	1.5	30.4
$pAC6Gsa + PP1 \leftrightarrow pAC6PP1 \rightarrow AC6Gsa + PP1$	0.0026	3	54
$ATP + AC6Gsa \leftrightarrow AC6Gsa\_ATP \rightarrow cAMP + AC6Gsa$	6e-05	10	80.42
$ATP + pAC6Gsa \leftrightarrow pAC6Gsa\_ATP \rightarrow cAMP + pAC6Gsa$	6e-05	10	8.042
$ATP + AC6 \leftrightarrow AC6\_ATP \rightarrow cAMP + AC6$	0.0001	120	0.142
$cAMP + PDE4B \leftrightarrow PDE4BcAMP \rightarrow AMP + PDE4B$	0.03	77.44	19.36
$1PKA + 2cAMP \leftrightarrow 1PKAr2c2cAMP2$	3.5e-08	0.06	
$1PKAr2c2cAMP2 + 2cAMP \leftrightarrow 1PKAr2c2cAMP4$	2.7e-07	0.28	
$1PKAr2cAMP4 + 2PKAc \leftrightarrow 1PKAr2c2cAMP4$	8.5e-08	0.05	
$V\,ASP + PKAc \leftrightarrow V\,ASP\,PKAc \rightarrow pV\,ASP + PKAc$	0.00026	1.5	30.4
$pV\,ASP + PP1 \leftrightarrow pV\,ASPPP1 \rightarrow V\,ASP + PP1$	0.0026	3	54
$AMP \rightarrow ATP$			1
$ATP + AC6GsaGi \leftrightarrow AC6GsaGi\_ATP \rightarrow cAMP + AC6GsaGi$	6e-05	10	1
$b2L + bARR \leftrightarrow b2LbARR$	0.0006	0.1	

Table 2. Initial concentrations (in nM) of species in the sample system.

PDE4B	200	b2	100
bARR	500	PKA	500
cAMP	100	AC6	1000
GsaGDP	200	GiGDP	200
RGS	100	VASP	200
PP1	100	ATP	1e+06

Objective function

A biological signal transduction system is defined by its state variable vector x, the set of all kinetic rate and initial concentration parameters.

We hypothesize that an efficient signal transmission would maximize the response coefficient $R_{C,S}$ (the response of species C to input S) defined as

$$R_{C,S} = \frac{\frac{C_t}{C_0} - 1}{\frac{S_t}{S_0} - 1}$$

with concentration change of target C and input S from baseline (t=0) to signal time t. For $R_{C,S}$, values < 1 show signal loss, with 1 for perfect transmission, and values > 1 showing signal amplification. We may also optimize for the slope s of the sigmoid at half-maximum concentration. This is equivalent to maximizing $R_{C,S}$, provided that the input signal remains entirely between the upper and lower boundaries of the sigmoid (Figure 1C). By optimizing for s, additional to $R_{C,S}$, we may force the system to implement a switch-like function instead of a more linear function. However, shifting the sigmoid function to the left or to the right is more important as the slope in our models.

In addition, we optimize for reaction time (delay to steady-state). Steady-state is defined, pragmatically, as relative change of less than 2% over 100s. The delay (d_S) is computed for 90% (EC90) of steady-state. We may now define an aggregate objective function:

$$f(x) = \max_x \left[R_{C,S}(x), -d_s(x) \right]$$

to select the system state variables that minimize delay and maximize response.

In addition to signal transmission from extracellular concentration changes onto steady-state concentrations, such as they typically occur for temporally integrating proteins like transcription factors,

we also look at signal transmission by transients, i.e. peak concentration, in response to extracellular signals. In this case, we minimize the delay to peak value, maximize the response at peak value, and minimize the response at steady-state value.

$$f_{transient}(x) = \max_x \left[R_{C,S}^p(x), -d_S^p(x), -R_{C,S}(x), d_S(x) \right]$$

Results

Delay vs. efficiency trade-off

The computation of signal transmission efficiency will be explained here for a single input-output pathway of cAMP/PKA-mediated transmission in a cellular membrane compartment. It is clear that a complex signaling system may have several inputs, and a large number of outputs or target proteins, and this is especially the case for cAMP-mediated signaling. Nonetheless we will focus on the simple case here to explain the basic principle. The input is a membrane receptor, a G-protein coupled receptor (GPCR), $\beta(2)$AR, which is activated by an extracellular pharmacological agonist ISO (isoproterenol); the output is a membrane protein, VASP, which promotes actin filament elongation. Activation of the receptor selectively inhibits VASP by phosphorylating VASP to pVASP via protein kinase A (PKA) activation. In Figure 1B the original ISO/pVASP transfer function is shown, which we use here for further optimization. The system was trained for a signal distribution between 10nM and 1 μM ISO adjusting both kinetic rate and concentration parameters (Figure 1A). The optimization method used is a simplex algorithm[12]. Figure 1B shows the transfer function before and after adjustment, maximizing for $R_{C,S}$, d_S or the combined function f. The results are summarized in Table 3. We see that optimizing for the response coefficient alone shifts the function to the right to better cover the input range. Optimizing for the delay alone shifts it to the left, speeding up signals in the lower range (which are slower). Both objectives together (equally weighted) are fairly close to the original, biologically validated curve, with an improved $R_{C,S}$.

In general, biochemical reactions are faster at higher substrate concentrations, but the relative concentration change in response to an increase in the enzyme or the binding partner is less. This is a fundamental trade-off between delay and transmission efficiency that may define an optimal operating range for a signaling system and be of relevance in disease processes[6]. The results obtained with this experiment are simple, intuitive and encourage continuing to explore the basic idea.

Evolutionary learning of kinetic rate parameters

In principle, we may use all parameters in a system, concentrations or kinetic rates, to maximize signal transmission. But the evolution

of protein structure and interactions shows that it is fine-tuning of molecular kinetic parameters which is subject to evolutionary learning, while concentrations are often regulated adaptively in each cell. Mass-action kinetics approximate molecular kinetic parameters, even though there are significant sources of uncertainty, such as the stochastics of molecular interactions. In the following, we explore the idea that kinetic rate learning operates on evolutionary timescales, and that biologically attested signal transduction pathways contain reaction rates which are optimal in terms of signal transmission efficiency. We use known concentration ranges, specific by cell type, together with kinetic rate optimization.

To explore the parameter space, we drew 1662 values for all kinetic rate parameters (kon, koff, kcat) from a distribution of 20% to 500% of the original parameters, and calculated $R_{C,S}$ and d_S for the corresponding models, relative to an improved signal distribution from 1nM to 1μM (Figure 2A). We find that there are parameter combinations which greatly improve efficiency of the signal transmission function. The basic distribution of a uniform low value of $R_{C,S}$ for fast d_S and a wide variability in $R_{C,S}$ above a certain threshold in d_S is robust against different types of signaling input (cf. also Figure 3A). This may, however, be highly dependent on the reaction network that underlies the transfer function. We have not further explored this question. To test for robustness of these systems against variability of concentration, we repeated experiments for 100 systems with 20% variation of original concentrations, which corresponds to generally accepted noise levels (cf.[13,14]). As expected, this low variation did not significantly affect the quality of a set of kinetic rates (Supplementary table).

We further analysed the parameter combinations with different signal transmission efficiency. In Figure 2B and C, we distinguish low and high efficiency signal transmission. Interestingly, we find, with respect to signal efficiency of the transfer function, that all parameters are "sloppy" (allow a wide variation), there are no "stiff" (low variation) parameters[15]. Standard deviation for all parameters in our case is similar (Figure 2B). Since it has been argued that optimization to experimental data yields reactions which allow more variance than others, as an indication of their influence on the signal transmission pathway, this analysis seems to contradict this effect. Possibly, these results pertain mostly to parameter variation that results from matching a networked system with many species to selected time-series data for only few species, which may behave differently from general optimization.

Co-regulation of protein concentration as an adaptive response to perturbation

Co-regulation of protein expression in cellular systems is important in disease progression and often a problem in targeted interventions. Here we are exploring the question of self-organization of protein concentration after a perturbation that reduces one protein to only 10% of its previous concentration. Keeping kinetic rates fixed, all concentrations in the system are allowed to adjust until optimality of signal transmission and delay is reinstated. There is a number of interesting observations here (cf. Figure 4A), which relate to the biological reality. For instance, reducing PDE4B causes much regulation in other proteins, but it is almost never targeted. In contrast, reducing PP1 has little effect, but PP1 is frequently responsive to other proteins. Reducing PKA, RGS and AC6 leads to widespread

**Table 3. Reaction times (d_S) and response efficiency ($R_{C,S}$) in a biological system and under optimization.**

	$R_{C,S}$	d_S
original	0.272	423
optimized for $R_{C,S}$	0.41	594
optimized for d_S	0.11	169
optimized for f	0.34	481
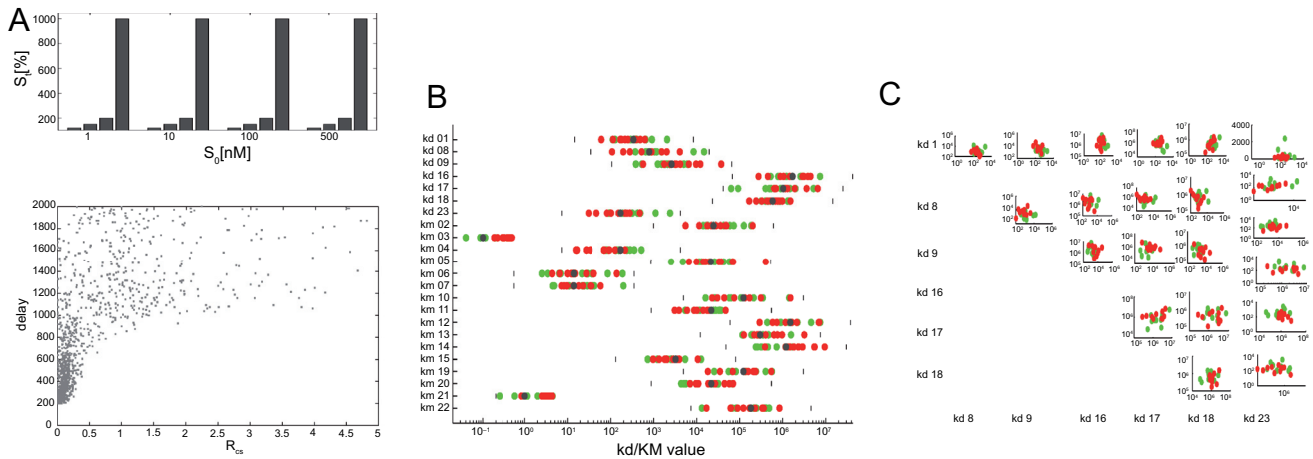
Figure 2. A. Distribution of systems according to R and d values. ~ 1600 different systems states were randomly generated with k-parameter variations (20–500%), $R_{C,S}$ and d values were measured with the signal set shown on top. **B**. Kd and KM parameters for high efficiency (red, $f < 2$) and low efficiency (green, $f > 5$) systems. **C**. Cross-correlation of Kd values.
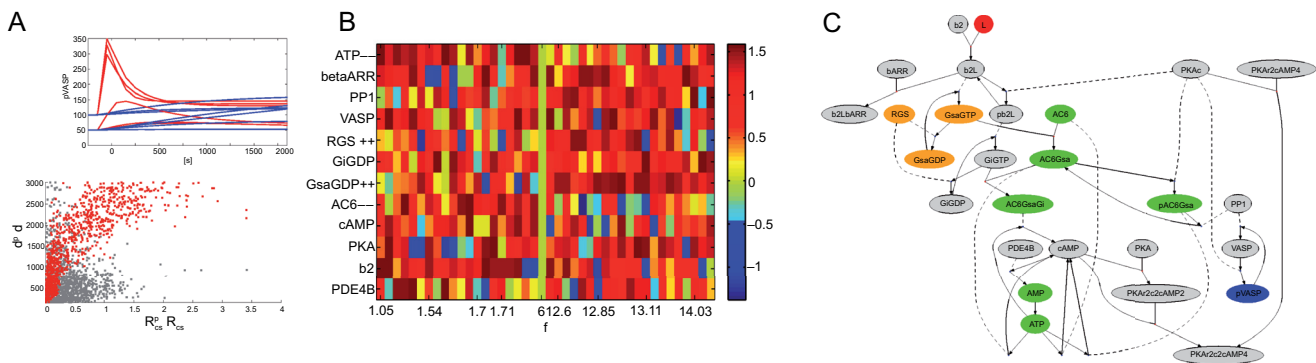
Figure 3. A. (top) Dynamics of selected systems with low ($f_{transient} < 3$, blue) or high ($f_{transient} > 10$, red) propensity for a transient response. (bottom) Distribution of concentration parameters according to d^p, $R_{C,S}^p$ (grey) or $R_{C,S}$, d (red). **B**. Concentration changes in response to optimizing for transients, with plateau signaling (left) and strong transients (right), sorted by $f_{transient}$. **C**. Concentration shifts (orange=high, green=low) in the biochemical reaction network for a high transient system, as averaged from **B**. Conforming to intuition, we see that the earlier, driver complex (G-protein) is high for transients, which increase the on-slope for cAMP production.
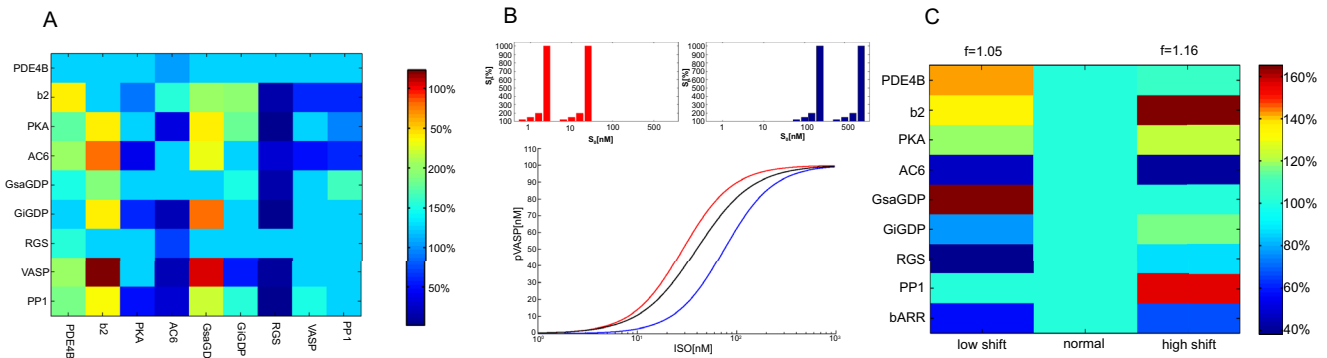
Figure 4. A. Concentration changes in response to total protein concentration reduction. Selected concentrations were reduced to 10% (shown on the x-axis). Learning was applied until f values were improved (shown on top for each experiment). Colors show the relative adjustment of all concentrations on the y-axis. **B**. Transfer function response to shifts in input signal range (shown on top). **C**. Concentrations changes in response to extracellular signal shift. Matrix is constructed as in **A**.

down-regulations, to maintain sensitivity of signaling, but reducing the receptor beta-2 leads to up-regulation.

There are many individual adaptations which can be interpreted to maintain the sensitivity and responsivity of the small molecule cAMP. The results show that protein regulation is highly sensitive to positions and roles of individual proteins in the reaction network in transmitting the signal. This problem may also be amenable to a more principled mathematical analysis[16]. Another idea would be to rank reaction systems defined by kinetic parameters as in Figure 2 by how well they adapt to perturbations.

In our example, the quality of the readjustment is not always the same, but the simple optimization scheme that we use may easily produce suboptimal solutions (local minima). Concentration changes may be caused by genetic up- or down-regulation, secretion and re-uptake, increased degradation, RNA interference, etc. and are therefore not easy to model from the standpoint of mechanistic biological modeling, which would need modules for all biologically attested processes. A unified perspective by a set of constraints and a set of objectives, such as has been envisaged here, may lead to better predictive results and may also be used as a guiding principle in constructing mechanistic models. Since there are intricate biological processes of adjusting concentrations, we may assume that in the cell optimal solutions are more easily found.

Optimal signal transmission depends on the signaling level

From the standpoint of disease modeling, an unusual protein concentration may be an adaptive response where a still functioning cell in a dysfunctional external signaling environment struggles to keep signal transmission efficient to support cellular function. In such a case, targeting this protein by pharmacological intervention will lead to co-regulation on other proteins. In general, as well as in real biological signaling systems, it is the localization of the input signaling range and the distribution of signals that the system transmits which are important for optimization. If we select the signal set that we optimize for in the right way, the input range will move towards the loglinear range, i.e. between the lower and upper input boundaries (Figure 4B). As a result, a number of internal concentrations will change. Figure 4C shows the relative concentration changes that result from a shift in input range. Interestingly, the low shift requires a strong reduction in RGS, and upregulation for PDE4B, among other effects. GsaGDP (the activating G protein) is strongly increased, and GiGDP (the inhibitory G protein) is decreased. In the high shift, GiGDP and PP1 (the phosphatase which decreases pVASP) are stronger, and the beta-2 receptor density is much increased. In the future, it will be interesting not only to calculate these effects based on different optimization measures, and in multiple input-output scenarios, but also to compare this with attested cases of biological adaptation. By reverse engineering, we may infer an optimization measure from a sufficient number of attested co-regulations. For instance in addiction, protein co-regulation as a form of sensitization is well-attested[17], but also in cancer where intercellular signaling (e.g. by cytokines[18]) is affected. Gene expression data may then be mined not only for evidence of the mechanics of genetic regulatory pathways but also for evidence of shifts in extracellular signaling, which cause altered protein expression.

Role of transients

The appearance of a transient vs. a plateau signal (or even a dampened oscillation) in response to sustained signaling depends on the construction of the biochemical reaction network (negative feedback interactions) and its parameters. We show that we can also train a system for the appearance of a transient response to a sustained signal. This means to search for high response at peak, low delay to peak, but also a low response at steady-state (see Section "Objective function" and Figure 3A). This requirement of invariance for the steady-state is sometimes considered a form of "robustness" or "homeostatic regulation" of the cellular response[19,20], and signaling by transients is widely regarded as an important mechanism. Here we found that concentration adjustment is quite sufficient to acquire a switch from transient to plateau response, with a high variability of response shape dependent on concentration, but also on the size of the signal (Figure 3A). In Figure 3B we show the distribution of concentrations that achieve high or low propensity for transients, as indicated by $f_{transient}$. We focus on concentrations with fairly uniform up- or down-regulation to create an experimental graph in Figure 3C. The simplicity of Figure 3C undermines the notion, as proven by the random search optimization, that deviations from up- or down-regulation for individual concentration may not be irrelevant noise, but rather part of a guided adaptation process that has many different solutions. We may have to consider this complexity to understand concentration adjustments in biological cells.

By using different objective functions for each target, it will be quite possible to combine different goals for targets in a multiple-output signaling system. Interestingly, a self-organizing cellular signaling system may also incorporate adaptive controls as objective functions, in particular when ion channels or membrane transporters are targeted. In this case, the goal of the system is to respond to an input (e.g. ISO at beta-2 receptors) such that the magnitude of another input such as calcium (ion channels as targets) is controlled. Signal transmission efficiency for such a target is not to be optimized to a maximum value, but instead to the appropriate ratio between the inputs. We have not followed up on this idea, but it may be worth further investigation as an example of natural computation for adaptive control.

Discussion

The idea to look for parametric optimization as the basis of realistic cellular properties has been applied with success to metabolic fluxes[21]. In that case, optimization of growth is usually regarded as the single objective function. Here we use another objective function, signal transmission efficiency, to study the adaptive response of a signaling system to perturbation in concentrations. This equals maximization of concentration change in a target species in response to input signal. Optimization of growth and optimization of signal transmission are therefore related.

Signal transmission in a cellular system may have multiple functions: transcription factor activation, which may be related to the cell cycle, to morphological change or to adjustment of protein concentrations, cytosolic kinase/phosphatase activation with multiple cellular targets, membrane protein activation such as ion channels or receptors, etc. We assume that signal transduction has been

optimized by evolution, and that concentration adjustment exists to maintain effective signal transmission. We have shown how to optimize a single-input single-output system for both speed and signal efficiency due to the basic properties of kinetic equations[6]. It is easy to extend the present discussion to optimize for multiple outputs in parallel, and a system could also be optimized for a number of I/O functions. In that case, other measures, such as mutual information, may also be employed as objectives. We used optimization in a two-step process: (a) in evolutionary learning, in order to find kinetic rates for estimated concentrations (b) in cellular adaptation, in order to re-calibrate the model in response to perturbations in concentrations, changes in extracellular signaling, or to effect a transient vs. plateau-like response. This also means that we have transitioned from a biologically defined system that is built bottom-up from available parameters and data to a self-organizing, learning system that adjusts to changes on evolutionary or individual time-scales.

Matching models to experimental time-series data is an under-constrained problem that often yields large ranges of suitable parameters[15,22]. Analysing signals and targets to find optimal transmission parameters may be used to further constrain and investigate the parametric space. Signal transmission efficiency in a biological system can be measured directly[23,24] and be compared to what is theoretically possible given a set of equations. During evolution, new protein subtypes develop with a different set of interactions and regulations. This corresponds to an adjustment of the available set of reactions, a type of structural learning to overcome the bottlenecks that are a result of tightly specified molecular kinetics and limited adjustment of concentrations.

Competing interests

No relevant competing interests disclosed.

Grant information

Supplementary table

Table 4. For 21 systems with $R_{c,s} > 0.5$ and $d_s < 800$, we re-calculate $R_{c,s}$ and $d_s < 800$ from 20 variations on concentration values within a 20% interval. Shown are the original, the mean and std values.

System no	Basic $R_{c,s}$	Mean $R_{c,s}$	Std $R_{c,s}$	Basic d_s	Mean d_s	Std d_s
1	0.578	0.582	0.0311	757	779	41.8
2	0.623	0.623	0.0419	723	722	35.4
3	0.575	0.575	0.0483	732	740	34.89
4	0.782	0.792	0.0571	749	768	50.45
5	0.609	0.622	0.099	578	590	46.82
6	0.609	0.615	0.033	728	737	37.05
7	0.612	0.628	0.0636	659	663	18.43
8	0.672	0.666	0.061	658	662	25.36
9	0.703	0.715	0.049	782	812	44.66
10	0.655	0.651	0.0511	761	760	29.3
11	0.576	0.551	0.0698	649	638	44.94
12	0.54	0.56	0.108	664	669	20.91
13	0.694	0.695	0.043	692	695	22.5
14	0.504	0.501	0.0312	726	731	35.70
15	0.522	0.526	0.0594	620	621	24.49
16	0.8	0.806	0.0369	726	742	50
17	0.693	0.7	0.0341	717	724	24.03
18	1.24	1.29	0.221	761	779	59.1
19	0.651	0.63	0.0512	668	666	11.64
20	0.529	0.5	0.045	777	762	43.25
21	0.537	0.534	0.044	752	754	33.42

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Open Peer Review

Current Referee Status:

Referee Report 03 June 2014

Eugene Koonin

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This article reports a new approach to modeling biological signal transduction systems based on the simple but important assumption that such systems have been evolutionarily optimized for maximum signal transmission efficiency. By embedding evolutionary optimization into the equations, the author obtains a self-organizing, evolving model. The approach is not fundamentally novel as the idea comes from the well-developed field of flux balance analysis. Apparently, however, this methodology has not been previously applied to signal transduction.

The current article is somewhat too abstract to judge on the ultimate utility of the approach but certainly, this is a step in an interesting direction.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Competing Interests: No competing interests were disclosed.

Referee Report 06 November 2013

Reimer Kühn

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In this paper the author studies optimization of parameters in a network of bio-chemical reactions for signal-transduction in a one-input-one-output setting, seeking either to maximize a stationary response coefficient, or to maximize the slope at half-maximum of the (sigmoid) stationary response function, or to minimize the delay to steady state. Combinations of these individual criteria are also looked at. Optimization is performed over the space of kinetic rates and the initial concentrations of reactants of the set of kinetic equations making up the signal transduction system.

The paper is very interesting and addresses a valid problem. It takes a well tested tenet as its starting point, viz. that signal transduction - like many other biological processes - "has been optimized by evolution, and that concentration adjustment exists to maintain effective signal transmission."

Thus parameters are sought which would optimize a given signal transduction system. The example studied in the present paper consists of a set of 23 chemical reactions involving 27 molecular species, and one or a combination of several criteria as mentioned above are used to drive the optimization. This paradigm, it is argued would naturally give rise to a notion of self-organization of said system, as well as support adaptability under changing external conditions.

While I find this an attractive proposition in principle, and one that could carry quite far in further attempts to understand cellular processes, I am not entirely convinced that the way in which it is implemented in the present paper is the most appropriate.

My main concern is related to the fact that, kinetic coefficients are included in the space of parameters over which optimization of response coefficients is performed. Kinetic coefficients are, however, largely given by the chemistry, or rather the physical properties of reactants involved. There is thus only a restricted scope for varying these parameters; they may be varied to be sure (though not independently!) by changing temperature, or the pH or the salinity of the solute in which these reactions occur. Since neither of these conditions would change widely under natural in-vivo conditions, there is thus a question whether this form of optimization is actually a good model of the one chosen by nature in the evolutionary process. Differences between optimal and original values of kinetic coefficients are not reported in the paper. It would be interesting to know whether they are small (hence whether nature has exploited the given reactions to the full).

Indeed, using minor modifications of the present setup, which would allow to describe the co-variation of kinetic coefficients with temperature, pH and salinity, one might thus conceivably be able to verify that - given the set of reactions describing the signalling system - nature is indeed running them under optimal conditions.

An altogether more ambitious project would consist in implementing a more appropriate model of the optimization process likely to have been implemented by nature to find precisely these 23 reactions. This form of optimization would require something of a more combinatorial nature (changing or modifying reactants thus reactions themselves) rather than parameters describing given reactions. Genetic algorithms, in some sense also implemented by nature, could be the method of choice to go about it.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Competing Interests: No competing interests were disclosed.

Author Response 14 Jun 2014

Gabriele Scheler, Carl Correns Foundation for Mathematical Biology, USA

Concerning kinetic coefficients: Often many different variants of a protein, or an enzyme, exist, with different kinetic properties. Especially in the G-protein coupled pathway, variants for adenylate cyclase, phosphodiesterase, RGS proteins, G-coupled receptors are on the order of dozens or more (including splice variants). Accordingly, the actual kinetic rate e.g. for cAMP degradation can be fine-tuned by the expression level of the different PDE's involved. These protein variants also often have differentiated expression by cellular compartment and different regulatory interactions. Still, I believe the potential for kinetic rate learning by evolutionary evolvement of enzymes into subtypes is present.

I agree that learning of a set of reactions would be a more ambitious and worthwhile project.

Competing Interests: No competing interests were disclosed.