Relating Cross Gramians and Sensitivity Analysis in Systems Biology

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***Abstract*— One of the key challenges in systems biology is the analysis of often complex biochemical reaction networks which contain many uncertain parameters. Typically, the dynamics of these systems strongly depends on a significant amount of parameters, hampering the analysis significantly as even small changes in the value of parameters can have significant influences on the overall behaviour of the entire network. Thus, one of the key problems in systems biology is to analyse the influence of parameters on the steady state and transient behaviour. In the first part of this work we derive links between first order sensitivity analysis as typically employed in systems biology and the concepts of controllability and observability of systems theory. Specifically we establish a close connection between cross Gramians and the so called response coefficients as used in Metabolic Control Analysis. In a second part we outline an expansion of this approach using empirical cross Gramians, allowing to overcome some of the limitations of first order sensitivity methods such as local validity.**

I. Introduction

Dramatic advances in proteomics, genomics, and measurement technologies such as DNA arrays have lead to a significantly increased knowledge about biological organisms. Traditionally, biological research is pursuing a reductionistic approach and is thus interested in identifying individual genes, proteins and cells, and studying their specific functions. However, by now it has become clear that understanding biological organisms is not merely possible by collecting information about all involved components. Rather, a holistic understanding of biological organisms requires considering all involved components as well as the interactions between them, since the interactions are ultimately responsible for an organism's form and functions [1], [2].

Systems biology aims at obtaining a holistic understanding of biological systems such as a single cell, organ or even a whole living organism combining approaches from system sciences, life sciences, and information sciences. One of the problems in systems biology is the modelling and analysis of biochemical reaction networks governing metabolic pathways or signalling cascades. These networks are typically very complex involving many species and parameters. The dynamics of the overall reaction system often depends significantly on a rather large number of independent parameters such as reaction rate constants or initial conditions of single reactions. For example, even a small change of a single enzyme concentration can lead to significant changes in the behaviour of the entire network. Thus, analysing the influence of these independent parameters on the static and transient behaviour of the network is of paramount importance. One of the tools typically used in systems biology to obtain deeper understanding and insight is sensitivity analysis. One specific example is Metabolic Control Analysis. In the frame of this work, we outline relations between controllability and observability as well as sensitivity analysis typically used in systems biology. Most of these connections are based on linear observations, e.g. are based on sensitivities of first order. In comparison to earlier works [3] we outline an expansion of these methods using nonlinear empirical cross Gramian methods, which partially allow to overcome the limited validity of linear/first order methods.

Typically, first order sensitivity analysis methods are employed to analyse the influence of parameter changes on the system behaviour. A common methods for biochemical reaction networks is Metabolic Control Analysis, which explicitly takes into account the structure and invariances imposed on the system by the stoichiometry of the network. The basic framework of Metabolic Control Analysis can for example be found in [4]–[6]. Significant progress with respect to Metabolic Control Analysis has been made over the last decades [7]–[10].

Recently, deriving links between control theory and the analysis of biochemical reaction systems has received significant interest, see e.g. [11]–[14]. Deriving similarities and connections between control theory and the analysis of biochemical reaction systems, it is hoped that one can tackle problems which are so far not solvable, for example, with Metabolic Control Analysis such as nonlinearities, multi-parameter interactions, large-scale parameter alterations etc.

Specifically, we derive links between classical systems theory concepts, such as controllability and observability, and Metabolic Control Analysis. We establish a clear connection between the cross Gramian, which is closely related to observability and controllability Gramians, and the response coefficients of Metabolic Control Analysis. Conceptually these results are of interest, since they relate the analysis via the consideration of whole system trajectories (Gramians) to purely static considerations such as used in Metabolic Control Analysis. Furthermore, the derived results can be expanded to time varying systems

and higher order/nonlinear sensitivity analysis.

The paper is structured as follows: In Section II we introduce the considered biochemical reaction networks and problem setup. Section III contains a brief outline of first order sensitivity analysis, including Metabolic Control Analysis which is typically used in the analysis of biochemical reaction networks. Furthermore we derive in this Section a link between between Metabolic Control Analysis and cross Gramians as known from control theory. In Section IV we derive an "nonlinear" expansion of the derived results using empirical Gramian methods. These allow to obtain less local results. The derived result is exemplified in Section V considering a small example system, before we conclude in Section VI.

II. Considered System Class

We consider in this paper the analysis of dynamical systems describing the behaviour of biochemical networks. Typically biochemical reaction networks are modelled in terms of the evolution of the n-dimensional vector $\boldsymbol{x}$ of concentrations

$$\dot{\boldsymbol{x}} = N\boldsymbol{v}\big(\boldsymbol{x}, \boldsymbol{p}(t)\big), \quad \boldsymbol{x}(t_0) = \boldsymbol{x}_0. \tag{1}$$

The rate vector $\boldsymbol{v}$ is an m-dimensional function of the state and the l-dimensional parameters $\boldsymbol{p}$. Parameters are typically reaction rate constants or enzyme concentrations, but can be initial conditions. The stoichiometry $N \in \mathbb{R}^{n\times m}$ relates the rate vector to the rate of change of the states.

A. Conserved Moieties

A common feature of many biochemical reaction systems is the existence of conserved moieties. These correspond to linear dependent states, often restricted to linear dependencies with non-negative coefficients.

In the presence of linear dependent states, the state space can be partitioned into independent states $\tilde{\boldsymbol{x}}$ and dependent states $\tilde{\boldsymbol{z}}$:

$$\boldsymbol{x} = \begin{bmatrix}\tilde{\boldsymbol{x}} \\ \tilde{\boldsymbol{z}}\end{bmatrix}. \tag{2}$$

With a suitably chosen link matrix L, (1) can be rewritten as

$$\frac{d}{dt}\begin{bmatrix}\tilde{\boldsymbol{x}} \\ \tilde{\boldsymbol{z}}\end{bmatrix} = \begin{bmatrix}I \\ L\end{bmatrix}\tilde{N}\boldsymbol{v}(\boldsymbol{x}, \boldsymbol{p}(t)). \tag{3}$$

where $\tilde{N}$ has full row rank. It directly follows that the evolution of $\tilde{\boldsymbol{z}}$ can be described by pure algebraic equations:

$$\tilde{\boldsymbol{z}}(t) = L\tilde{\boldsymbol{x}}(t). \tag{4}$$

Thus, only the differential equation for $\tilde{\boldsymbol{x}}$ is necessary for describing the systems dynamics. In the following, we assume that the reduction via conserved moieties has already been performed, i.e. for simplicity of notation $\boldsymbol{x}$ corresponds to the reduced state vector $\tilde{\boldsymbol{x}}$.

B. First Order considerations

Around a steady-state $(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}})$, the system (1) can be approximated by its linearisation:

$$\Delta\dot{\boldsymbol{x}} = A_{\text{ss}}\,\Delta\boldsymbol{x} + B_{\text{ss}}\,\Delta\boldsymbol{p}, \tag{5}$$

where

$$\begin{aligned}\Delta\boldsymbol{x} &= \boldsymbol{x} - \boldsymbol{x}_{\text{ss}},\\ \Delta\boldsymbol{p} &= \boldsymbol{p} - \boldsymbol{p}_{\text{ss}},\\ A_{\text{ss}} &= N\frac{\partial\boldsymbol{v}}{\partial\boldsymbol{x}}(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}}),\\ B_{\text{ss}} &= N\frac{\partial\boldsymbol{v}}{\partial\boldsymbol{p}}(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}}).\end{aligned}$$

For an asymptotically stable steady-state, straightforward calculation shows that a constant parameter deviation of $\Delta\boldsymbol{p}_{\text{ss}}$ results in a steady-state shift of

$$\Delta\boldsymbol{x}_{\text{ss}} = -A_{\text{ss}}^{-1}B_{\text{ss}}\,\Delta\boldsymbol{p}_{\text{ss}}. \tag{6}$$

The asymptotic stability of the steady-state directly implies that A_{ss} is invertible and therefore a unique solution of (6) exists.

III. First Order Sensitivity Analysis in Systems Biology

Equation (6) relates small parameter change towards changes in the steady-state. More generally, sensitivity analysis describes the influence of the parameters on the states. The linear sensitivity is defined as the partial derivative of the state vector

$$S = \frac{\partial\boldsymbol{x}}{\partial\boldsymbol{p}}, \qquad S(t) \in \mathbb{R}^{n\times l} \tag{7}$$

where, for mathematical convenience, we assume constant parameters. These linear sensitivities can be calculated as the solution of the following equation:

$$\begin{aligned}\dot{S} &= N\frac{\partial\boldsymbol{v}}{\partial\boldsymbol{x}}(\boldsymbol{x}, \boldsymbol{p}) + N\frac{\partial\boldsymbol{v}}{\partial\boldsymbol{p}}(\boldsymbol{x}, \boldsymbol{p}),\\ S(0) &= \big[\boldsymbol{S}_1(0), \quad \cdots, \quad \boldsymbol{S}_l(0)\big],\end{aligned} \tag{8}$$

with

$$\boldsymbol{S}_i(0) = \begin{cases}\boldsymbol{e}_i & \text{if } p_i \text{ is an initial condition,}\\ \boldsymbol{0} & \text{otherwise,}\end{cases} \tag{9}$$

where $\boldsymbol{e}_i$ is the Euclidean basis vector whose i-th entry is equal to one. Equation (8) follows from (1) by taking the partial derivatives with respect to the parameters on both sides and swapping the order of differentiation.

Around an asymptotically stable steady-state $(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}})$, the solution of (8) converges to

$$S_{\text{ss}} = -A_{\text{ss}}^{-1}B_{\text{ss}}. \tag{10}$$

This is equivalent to (6) as

$$S_{\text{ss}} = \lim_{\Delta\boldsymbol{p}_{\text{ss}}\to\boldsymbol{0}}\frac{\Delta\boldsymbol{x}_{\text{ss}}}{\Delta\boldsymbol{p}_{\text{ss}}}. \tag{11}$$

In metabolic engineering, a classical approach for sensitivity analysis is Metabolic Control Analysis (MCA), see e.g. [15] for a review. Two sensitivities are commonly used in MCA. First, the *concentration response coefficient* which is equivalent to the steady-state sensitivity as given in (10):

$$R_{\boldsymbol{p}}^{\boldsymbol{x}} = -A_{\text{ss}}^{-1} B_{\text{ss}}. \tag{12}$$

Second, the *flux response coefficient* measures the linear sensitivity of the parameters with respect to the rate vector, also at steady state, and is given by

$$R_{\boldsymbol{p}}^{\boldsymbol{v}} = \frac{\partial \boldsymbol{v}}{\partial \boldsymbol{x}}(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}}) R_{\boldsymbol{p}}^{\boldsymbol{x}} + \frac{\partial \boldsymbol{v}}{\partial \boldsymbol{p}}(\boldsymbol{x}_{\text{ss}}, \boldsymbol{p}_{\text{ss}}). \tag{13}$$

Remark 1 *Common in MCA is the use of relative sensitivities, sometimes also called scaled sensitivities, measuring the impact of a relative change of parameter. To simplify the presentation, this paper discusses only the unscaled case. Other sensitivity approaches also not discussed here are higher-order sensitivities and bifurcation analyses.*

In this section, we will restrict the system class to linear systems with an asymptotically stable steady state:

$$\begin{aligned} \dot{\boldsymbol{x}} &= A\,\boldsymbol{x} + B\,\boldsymbol{p} \\ \boldsymbol{y} &= C\,\boldsymbol{x}, \end{aligned} \tag{14}$$

where the parameter $\boldsymbol{p}$ is used as input.

How easily the input can influence the state is quantified by the controllability Gramian W_c, the minimal input energy necessary for steering the system from $\mathbf{0}$ at time $-\infty$ to $\boldsymbol{x}_0$ at time 0

$$\|\boldsymbol{p}_{opt}(\cdot)\|^2 = \int_{-\infty}^{0} \boldsymbol{p}(\tau)^T \boldsymbol{p}(\tau)\, d\tau = \boldsymbol{x}_0^T W_c^{-1} \boldsymbol{x}_0. \tag{15}$$

The controllability Gramian W_c is defined by:

$$W_c = \int_{-\infty}^{0} e^{-A\tau} B B^T e^{-A^T\tau}\, d\tau \tag{16}$$

and is the solution of the Lyapunov equation [16]:

$$-BB^T = AW_c + W_c A^T. \tag{17}$$

The Gramian based energy $\|\boldsymbol{p}_{opt}(\cdot)\|$ is proportional to the "norm" of $\boldsymbol{x}_0$. To gain insight into how controllable specific directions in the state-space are, we therefore scale $\boldsymbol{p}_{opt}$ by the Euclidean length of $\boldsymbol{x}_0$:

$$\frac{\|\boldsymbol{p}_{opt}(\cdot)\|^2}{\|\boldsymbol{x}_0\|^2} = \frac{\boldsymbol{x}_0^T W_c^{-1} \boldsymbol{x}_0}{\boldsymbol{x}_0^T \boldsymbol{x}_0}, \qquad \boldsymbol{x}_0 \neq \mathbf{0}. \tag{18}$$

An analysis of the Eigenvalues and corresponding Eigenvectors of the controllability Gramian reveals which directions the system can be easily steered to, and which are more energy-demanding. In particular, the direction of the Eigenvector corresponding to an Eigenvector zero cannot be reached. In other words, a controllable system has a positive definite controllability Gramian.

The best controllable directions are spanned by the Eigenvectors with the smallest Eigenvalues of W_c^{-1}. These are equivalent to the span of the Eigenvectors to the largest Eigenvalues of the controllability Gramian W_c.

Equivalently to the controllability case, observability can be analysed quantitatively using the observability Gramian W_o defined by

$$W_o = \int_0^{\infty} e^{A^T\tau} C^T C e^{A\tau}\, d\tau \tag{19}$$

or by

$$-C^T C = A^T W_o + W_o A. \tag{20}$$

The observability Gramian is a measure of the energy visible in the output signal when letting the system freely evolve from $\boldsymbol{x}_0$ at time 0 towards the steady state $\mathbf{0}$:

$$\|\boldsymbol{y}(\cdot)\|^2 = \int_0^{\infty} \boldsymbol{y}^T(\tau)\boldsymbol{y}(\tau)\ \mathrm{d}\tau = \boldsymbol{x}_0^T W_o \boldsymbol{x}_0. \tag{21}$$

The observability Gramian is also symmetric and positive semi-definite. Scaling the output energy (21) by $\boldsymbol{x}_0$,

$$\frac{\|\boldsymbol{y}(\cdot))\|^2}{\|\boldsymbol{x}_0\|^2} = \frac{\boldsymbol{x}_0^T W_o \boldsymbol{x}_0}{\boldsymbol{x}_0^T \boldsymbol{x}_0} \qquad \boldsymbol{x}_0 \neq \mathbf{0}. \tag{22}$$

reveals that also here the directions in the span of the Eigenvectors to the largest Eigenvalues are best observable. In particular, states in the kernel of the observability Gramian are unobservable.

Therefore, in both controllability and observability analysis, the direction corresponding to large Eigenvalues are most sensitive to perturbations of the state $\boldsymbol{x}_0$.

A. Cross Gramians and Sensitivity Analysis

Analysing controllability and observability gives distinct views about the importance of directions in the state space. For an input-output analysis, a combination of both is required. Moore [16] showed that a straightforward combination can be misleading: For example, the least observable states could be very good controllable. Thus a small input signal could result in a non-negligible output signal.

Balancing allows to overcome this problem, as it transforms the states in such a way that the controllability and observability Gramian are diagonal and identical (see e.g. [17]). Then, the well controllable states are also well observable.

A further alternative is to combine observability and controllability using the so called cross Gramian [18], [19]. For systems with as many inputs as outputs, the cross Gramian is defined as

$$W_{co} = \int_0^{\infty} e^{A\tau} B C e^{A\tau}\ \mathrm{d}\tau, \tag{23}$$

and is the unique solution of the Sylvester equation

$$-BC = W_{co} A + A W_{co}. \tag{24}$$

The cross Gramian is not only related to the controllability and observability Gramians by its similar definition, it also holds that

$$W_{co}^2 = W_c W_o, \tag{25}$$

showing that the cross Gramian contains both the controllability and observability Gramian [20]. Furthermore, there is a direct relation between the cross Gramian and steady-state response as discussed next.

For single-input single-output systems, the steady-state system gain g with respect to a step input is given by

$$g = -CA^{-1}B. \tag{26}$$

This steady-state gain is closely related to the cross Gramian [18] as

$$\operatorname{trace} W_{co} = \frac{1}{2}g. \tag{27}$$

Note that the trace of a matrix is equivalent to the sum of its Eigenvalues. The steady-state gain is therefore related to the average of the Eigenvalues of the corresponding cross Gramian. Equation (27) thus allows to link between controllability and observability in a consistent way.

B. Linking Metabolic Control Analysis to Cross Gramians

This section provides a bridge between the system theoretic concepts of controllability and observability with Metabolic Control Analysis. Specifically, the the equivalence of the concentration flux coefficient commonly used in Metabolic Control Analysis and the cross Gramian is shown. The cross Gramian can be seen as combining quantitative controllability and observability properties of the system into a single matrix.

Theorem 1 *The concentration response coefficient of Metabolic Control Analysis can be expressed by an appropriately chosen cross Gramian:*

$$R_\pi^{x_i} := \frac{\partial x_{i,ss}}{\partial \pi} = 2 \operatorname{trace} W_{co}^{(\pi, x_i)} \tag{28}$$

in which the matrix $W_{co}^{(\pi,x_i)}$ is the cross Gramian for the input π and the output x_i.

Proof: The steady-state system gain (26) for the single-input single-output system with input $p = \pi$ and output $y = C\boldsymbol{x} = x_i$ is equivalent to the concentration response coefficient (12). Combining this with (27) result in:

$$R_\pi^{x_i} = CR_\pi^{\boldsymbol{x}} = -CA_{ss}^{-1}B_{ss} = g = 2 \operatorname{trace} W_{co}. \qquad \blacksquare$$

This theorem shows the relationship between Metabolic Control Analysis (a pure steady state consideration) and a joint controllability/observability consideration via the cross Gramian. With this relation, it is possible to quantify how the steady state response of a particular output is influenced by a step perturbation of a particular parameter.

Remark 2 *Further extensions of the provided result are possible considering frequency-weighting as done in model reduction, see for example [21], or [17]. With respect to frequency weighting, it is interesting to note that the Hankel norm of a linear system is equal to the largest singular value of the cross Gramian. This allows to investigate the sensitivity for certain frequency bands, see for instance [22].*

IV. "NONLINEAR" SENSITIVITIES USING EMPIRICAL NONLINEAR GRAMIANS

Several extensions to standard Metabolic Control Analysis have been proposed. Among these are extensions for non-equilibrium trajectories [23] or an approach that for large scale parameter alterations [24].

The link shown here between Metabolic Control Analysis on the one hand and controllability and observability on the other will allow a new viewpoint on non local sensitivity analysis. In the first part of this work we outlined a link between the system theoretic concepts of observability and controllability and first order sensitivity methods such as Metabolic Control Analysis as typically used in systems biology. Specifically we showed that there is a clear connection between cross Gramians known from control theory and concentration response coefficients from Metabolic Control Analysis. This connection is then used in a second part to outline a new sensitivity analysis approach considering empirical cross Gramians for nonlinear systems, allowing to overcome the local validity of first order sensitivity methods. This allows to directly taking nonlinearities as well as parameter-parameter-interactions into account [25].

The method for the approximation of the empirical nonlinear cross-Gramian relies on the use of (simulated) data, similarly to the method for the approximation of the empirical controllability and observability Gramian [25]. The data is collected from system trajectories which result from simultaneous perturbations of the investigated input and the initial conditions. The perturbations are defined by the following sets:

$$\begin{aligned}
\mathcal{R}^\rho &= \left\{ R_1, \ldots, R_\rho; R_i \in \mathbb{R}^{n \times n}, R_i^T R_i = I, i = 1, \ldots, \rho \right\} \\
\mathcal{P} &= \{P_1 = 1, P_2 = -1\} \\
\mathcal{K}^\kappa &= \{c_1, \ldots, c_\kappa; c_i \in \mathbb{R}, c_i > 0, i = 1, \ldots, \kappa\} \\
\mathcal{S}^\sigma &= \{d_1, \ldots, d_\sigma; d_i \in \mathbb{R}, d_i > 0, i = 1, \ldots, \sigma\},
\end{aligned}$$

where n is the number of states of the nonlinear system (1). $\mathcal{R}^\rho$ is a set of ρ orthogonal matrices. This set should be chosen in such a way that it covers the simultaneous perturbations of the initial conditions which are of interest. The set $\mathcal{P}$ is associated with the input perturbations, P_1 for positive deviations from the nominal value, and P_2 for negative deviations. $\mathcal{K}^\kappa$ and $\mathcal{S}^\sigma$ are sets of positive constants c_i and d_i, respectively. $\mathcal{K}^\kappa$ should cover the amplitudes of interest for the perturbations of the initial conditions, and $\mathcal{S}^\sigma$ those for the inputs. The cross Gramian can now be approximated for linear and nonlinear systems by the empirical cross Gramian defined as follows:

Definition 1 (Empirical Cross Gramian) *Let $\mathcal{R}^\rho$, $\mathcal{P}$, $\mathcal{K}^\kappa$, and $\mathcal{S}^\sigma$ be given sets as described above. For the system* (1)*, define the empirical cross Gramian $\hat{W}_{co}$ around an exponentially stable steady-state $\boldsymbol{x}_{nom}$ with corresponding nominal parameter p_{nom} by*

$$\hat{W}_{co} = \sum_{p=1}^{2}\sum_{s=1}^{\sigma}\sum_{r=1}^{\rho}\sum_{k=1}^{\kappa} \frac{\int_0^\infty R_r \Psi^{psrk}(t) R_r^T dt}{2\sigma d_s \rho \kappa c_k} \tag{29}$$

where the entries of the $n \times n$-matrix $\Psi^{psrk}(t)$ are given for all $i = 1, \ldots, n$ and $j = 1, \ldots, n$ by

$$\Psi_{ij}^{psrk}(t) = \boldsymbol{e}_i^T R_r^T \Delta \boldsymbol{x}^{psrkj}(t; d_s) P_p^T C \Delta \boldsymbol{x}^{psrkj}(t; c_\kappa) \tag{30}$$

with

$$\Delta \boldsymbol{x}^{psrkj}(t;\xi) = \boldsymbol{x}^{psrkj}(t) - \bar{\boldsymbol{x}}^{psrkj} - \left(\boldsymbol{x}^{psrkj}\big|_{\xi=0}(t) - \bar{\boldsymbol{x}}^{psrkj}\big|_{\xi=0} \right),$$

where $\boldsymbol{x}^{psrkj}(\cdot)$ is the state of the system (1) *corresponding to the impulsive input $u^{sp}(t) = p_{nom} + d_s P_p \delta(t)$ and initial condition $\boldsymbol{x}_0^{rkj} = \boldsymbol{x}_{nom} + c_k R_r \boldsymbol{e}_j$ and $\bar{\boldsymbol{x}}^{psrkj}$ denotes its steady-state.*

Albeit the approximation method given by Definition 1 requires the collection of a large amount of data, it allows to calculate a cross Gramian for nonlinear systems that falls back to the classical definition (23) in the case of linear systems, as shown next.

Proposition 1 *For any nonempty sets $\mathcal{R}_\rho$, $\mathcal{P}$, $\mathcal{K}^\kappa$ and $\mathcal{S}^\sigma$ the empirical cross Gramian $\hat{W}_{co}$ of the asymptotically stable linear system* (14) *is equal to the usual cross Gramian W_{co} (cf.* (23)*).*

Proof: Due to the linearity of (14), $\boldsymbol{x}_{\text{nom}} = 0$ and $p_{\text{nom}} = 0$. For the initial condition $\boldsymbol{x}_0^{rkj} = c_k R_r \boldsymbol{e}_j$ and the input $u^{sp}(t) = d_s P_p \delta(t)$, the trajectory of the state vector is

$$\boldsymbol{x}^{psrkj}(t) = e^{At} c_k R_r \boldsymbol{e}_j + e^{At} B d_s P_p,$$

and its steady-state is the origin, independently of p, s, r, k and j. Then, (30) simplifies to

$$\Psi_{ij}^{psrk}(t) = \boldsymbol{e}_i^T R_r^T e^{At} B d_s P_p P_p^T C e^{At} c_k R_r \boldsymbol{e}_j,$$

and

$$\Psi^{psrk}(t) = c_k d_s R_r^T e^{At} B P_p P_p^T C e^{At} R_r.$$

Hence,

$$\begin{aligned}\hat{W}_{co} &= \sum_{p,s,r,k} \frac{\int_0^\infty R_r c_k d_s R_r^T e^{At} B P_p P_p^T C e^{At} R_r R_r^T dt}{2\sigma d_s \rho \kappa c_k} \\ &= \sum_{p=1}^{2}\sum_{r=1}^{\rho} \frac{R_r R_r^T \int_0^\infty e^{At} B P_p P_p^T C e^{At} dt R_r R_r^T}{2\rho}.\end{aligned}$$

As R_r and P_p are orthonormal,

$$\hat{W}_{co} = \int_0^\infty e^{At} B C e^{At} dt = W_{co}$$

which is the desired result. ∎

V. Example

An enzymatic reaction pathway, see Figure 1, illustrates the use of empirical cross Gramians for sensitivity analysis The example consists of four states, S, E, C, P and the

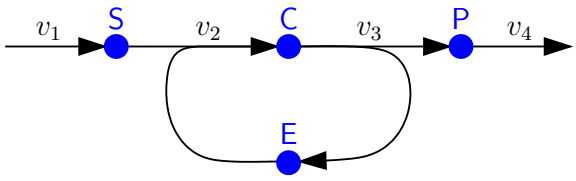

Fig. 1. Enzymatic reaction pathway. All reaction follow the law of mass action and are irreversible, except v_2.

four reactions

- v_1: $\rightarrow$ S: $v_1 = k_1$
- v_2: S+E $\leftrightarrow$ C: $v_2 = k_2 se - k_{-2} c$
- v_3: C$\rightarrow$ P+E: $v_3 = k_3 c$
- v_4: P $\rightarrow$: $v_4 = k_4 p$.

The corresponding system is described by

$$\dot{s} = k_1 - k_2 se + k_{-2} c \tag{31a}$$
$$\dot{e} = -k_2 se + k_{-2} c + k_3 c \tag{31b}$$
$$\dot{c} = k_2 se - k_{-2} c - k_3 c \tag{31c}$$
$$\dot{p} = k_3 c - k_4 p. \tag{31d}$$

Obviously, $e(t) + c(t)$ is constant, a so-called conserved moiety. Therefore, (31c) can be replaced by

$$c(t) = e_{\text{tot}} - e(t) \tag{32}$$

with e_{tot} the total amount of enzyme. We use the notation:

$$\begin{aligned}\boldsymbol{x} &= \begin{bmatrix} s, & e, & p \end{bmatrix}^T, \\ \boldsymbol{k} &= \begin{bmatrix} k_1, & k_2, & k_{-2}, & k_3, & k_4 \end{bmatrix}^T.\end{aligned}$$

Exemplarily, we choose the following numerical values for the nominal state and the parameters:

$$\begin{aligned}\boldsymbol{x}_{\text{nom}} &= \begin{bmatrix} 0.0534 & 9.6667 & 0.2500 \end{bmatrix}^T, \\ \boldsymbol{k}_{\text{nom}} &= \begin{bmatrix} 1, & 2, & 0.1, & 3, & 4 \end{bmatrix}^T, \\ e_{\text{tot}} &= 10.\end{aligned}$$

and analyse the sensitivity of k_1 on p. The set $\mathcal{P} = \{1, -1\}$, $\mathcal{R}^\rho$ is chosen as

$$\mathcal{R}^\rho = \{I, Q\}$$

where Q is a random orthonormal matrix, while the sets $\mathcal{S}^\sigma$ and $\mathcal{K}^\kappa$ are taken as logarithmically spaced intervals, two values per decade, from 10^{-4} up to a maximal value which is varied from 10^{-3} to at most 100. Finally, the systems is simulated over the time span $[0,\ 100]$ and the Dirac function is approximated as

$$\delta(t) = \begin{cases} \frac{2}{\epsilon}\left(1 - \frac{t}{\epsilon}\right) & t \in \begin{bmatrix} 0, & \epsilon \end{bmatrix} \\ 0 & \text{else} \end{cases}$$

with $\epsilon = 10^{-4}$. For large values of c_k or d_s, initial conditions or parameters can be negative. These cases have

been excluded from the corresponding sets to prevent nonsensical simulations. Figure 2 shows the empirical control coefficients calculated using (28) for different maximal values of the sets $\mathcal{K}^\kappa$ and $\mathcal{S}^\sigma$. Figure 2 shows that for

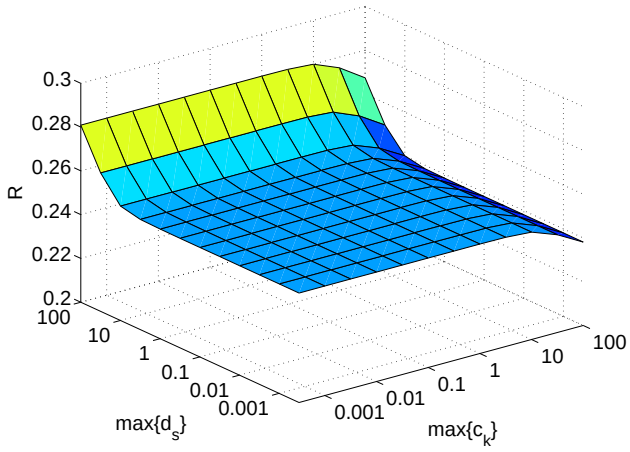

Fig. 2. Empirical cross Gramian for the exemplary pathway of Figure 1 from parameter k_1 to state x_4.

a large range of the parameter and initial conditions, the sensitivity is close to 0.25, the value obtained by calculating the sensitivity coefficient from the linearisation in the nominal case using (12).

This example shows that combining a sensitivity analysis such a Metabolic Control Analysis with system theoretic concepts such as controllability and observability may provide more insights into the systems.

VI. Summary

The dynamics of biochemical reaction networks as typically encountered in systems biology often significantly depends on a large amount of parameters such as maximum reaction rates. Since even small changes e.g. the kinetic properties of only one single enzyme can cause pronounced changes in the behaviour of the entire network, an analysis of the influence of these parameters on the steady states and transient behaviour is of paramount importance. In the first part of this work we outlined a link between the system theoretic concepts of observability and controllability and first order sensitivity methods such as Metabolic Control Analysis as typically used in systems biology. Specifically we showed that there is a clear connection between cross Gramians known from control theory and concentration response coefficients from Metabolic Control Analysis. This connection is then used in a second part to outline a new sensitivity analysis approach considering empirical cross Gramians for nonlinear systems, allowing to overcome the local validity of first order sensitivity methods.

This allows to directly taking nonlinearities as well as parameter-parameter-interactions into account.

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