Chapter 8
Quasi-Steady State: Searching for and Utilizing Small Parameters

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Dedicated to Jürgen Scheurle on the occasion of his 60th birthday

Abstract We present an outline of quasi-steady state methods (QSS) in ordinary differential equations which model systems of chemical reactions, and its application to reduction of dimension. Special attention is given to the relation between QSS and singular perturbations including, as a new result, a general explicit reduction formula. Moreover, we describe and discuss heuristics which convert a QSS assumption to conditions restricting the parameters of the differential equation.

2010 Mathematics Subject Classification: 92C45, 80A30, 34D05, 34D23.

8.1 Introduction

Quasi-steady state (QSS) reduction is frequently employed to reduce the dimension of differential equations for chemical and biochemical reactions, in particular as a preliminary step in parameter identification problems. While QSS has been used by biologists, chemists, and also by application-oriented mathematicians since the early twentieth century, a precise mathematical description and analysis was achieved only in the late 1980s, and some aspects are still not completely resolved. The issue is complicated by the fact that different groups of scientists (including different groups of mathematicians) have different notions of, and different approaches to, QSS assumptions and reductions. Another critical point concerns the role, the applicability, and the application of singular perturbation theory.

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A. Johann et al. (eds.), *Recent Trends in Dynamical Systems*, Springer Proceedings in Mathematics & Statistics 35, DOI 10.1007/978-3-0348-0451-6_8,

Most of this paper collects some classical and recent results on QSS and QSS reduction. We also present a few new results and aspects, including a general explicit reduction formula for mass action kinetics, given a singular perturbation setting. In Sect. 8.2 we review definitions of QSS (including a working definition we will adopt), give a short historical outline, and describe some problems and applications, including standard examples. Motivated by applications, it seems advisable to distinguish two notions of QSS (which also appear under different names in the literature). On one hand, there is QSS for reactions, where certain (forward–backward) reactions are assumed to reach equilibrium quickly. On the other hand, there is QSS for concentrations of certain chemical species, which goes back to Michaelis and Menten. Analyzing these different QSS assumptions leads to different mathematical problems. Section 8.3 is about reduction of dimension in the classical Tikhonov–Fenichel setting of singular perturbations. We present a general reduction formula, sketch its derivation, and give several examples. Moreover we show that, in the scenario of slow–fast reactions, Tikhonov–Fenichel theory is applicable in rather general circumstances. Section 8.4 is about various heuristics—including scaling methods—for finding "small parameters" from QSS assumptions. While these heuristics provide satisfactory results in many cases, identification of small parameters for QSS—which is closely tied to the chosen definition—still seems unfinished. Most of the examples we give are presented for the purpose of illustration and have been discussed in other publications. One exception is a somewhat larger example to demonstrate the feasibility of the reduction procedure.

8.2 Background and Statement of Problem

8.2.1 Chemical Reactions and ODEs

Systems of chemical reactions are frequently modeled with the help of differential equations. In this paper we will concentrate on systems that can be modeled by ordinary differential equations, which is justified in the following scenario:

- Reactions take place in a closed vessel, and there is no spatial inhomogeneity.
- Thermodynamical parameters such as temperature and pressure are (being kept) constant.
- There are explicit expressions for the reaction rates (usually mass action kinetics).

Given these conditions, there is a standard procedure to transfer a chemical reaction scheme to a system of ordinary differential equations and there is a number of strong theoretical results on the properties of such equations. The procedure was formalized and the class of resulting equations was discussed by several authors in the 1960s and 1970s, with fundamental contributions, in particular with regard to convergence to equilibrium, due to Feinberg [7], and Horn and Jackson [15]. One important ingredient of this procedure is stoichiometry: Molecules do not vanish into nothing and are not created out of nothing. Thus in a reaction like $A+B\rightleftharpoons C$, for every C-molecule that is created, an A and a B vanish. Hence stoichiometry implies the existence of linear first integrals for the differential equations.

Example 8.1. The Michaelis–Menten system (Michaelis and Menten [20], see also Briggs and Haldane [5] and many textbooks and monographs such as Atkins and de Paula [2]; Berg et al. [3]; Keener and Sneyd [18]; Murray [22]) is a basic model reaction for an enzyme E catalyzing the transformation of substrate S to product P via an intermediate complex C. The reaction scheme

$$E+S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} E+P$$

by way of the above-mentioned procedure with mass action kinetics yields a differential equation system for the concentrations:

$$\begin{aligned}
\dot{e} &= -k_1 es + (k_{-1}+k_2)c - k_{-2}ep,\\
\dot{s} &= -k_1 es + k_{-1}c,\\
\dot{c} &= k_1 es - (k_{-1}+k_2)c + k_{-2}ep,\\
\dot{p} &= \phantom{-k_1 es +{}} k_2 c \phantom{{}+ k_{-1}} - k_{-2}ep.
\end{aligned}$$

The relevant initial values are $s(0)=s_0>0$, $c(0)=0$, $e(0)=e_0>0$, and $p(0)=0$. All rate constants k_i are assumed to be >0, with the possible exception $k_{-2}\geq 0$. In case $k_{-2}=0$ one speaks of the irreversible Michaelis–Menten reaction, the case $k_{-2}>0$ is called reversible. The irreversible system is usually presented and investigated in monographs and research articles.

From stoichiometry one obtains the linear first integrals $e+c$ and $s+c+p$, which may be used to reduce the differential equation to dimension two. The standard procedure leads to the following equation:

$$\begin{aligned}
\dot{s} &= -\,k_1 e_0 s + (k_1 s + k_{-1})c,\\
\dot{c} &= k_1 e_0 s - (k_1 s + k_{-1} + k_2)c + k_{-2}(e_0 - c)(s_0 - s - c).
\end{aligned} \tag{8.1}$$

Example 8.2. A cooperative enzyme-catalyzed reaction is described by the reaction scheme (see e.g. Keener and Sneyd [10, 18, 25]):

$$\begin{aligned}
S+E &\underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C_1 \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} E+P,\\
S+C_1 &\underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} C_2 \underset{k_{-4}}{\overset{k_4}{\rightleftharpoons}} C_1+P.
\end{aligned}$$

Here substrate and enzyme react to form a complex C_1, and moreover substrate and C_1 react to form a complex C_2. In the reversible scenario, enzyme and product may also combine to form C_1 with rate constant $k_{-2} > 0$, and C_1 and P may combine to form C_2 with rate constant $k_{-4} > 0$. Similar to the Michaelis–Menten system one also considers the irreversible case with $k_{-2} = k_{-4} = 0$; all other rate constants are assumed > 0 throughout. From mass action kinetics, stoichiometry, and the initial values $s(0) = s_0 > 0$, $c_1(0) = c_2(0) = 0$, $e(0) = e_0 > 0$, and $p(0) = 0$, one obtains the differential equation

$$\begin{aligned} \dot{s} &= -k_1 e_0 s + (k_{-1} + k_1 s - k_3 s)c_1 + (k_1 s + k_{-3})c_2, \\ \dot{c}_1 &= k_1(e_0 - c_1 - c_2)s - (k_{-1} + k_2)c_1 + k_{-2}(e_0 - c_1 - c_2)(s_0 - s - c_1 - 2c_2) \\ &\quad - k_3 c_1 s + (k_{-3} + k_4)c_2 - k_{-4} c_1 (s_0 - s - c_1 - 2c_2), \\ \dot{c}_2 &= k_3 c_1 s - (k_{-3} + k_4)c_2 + k_{-4} c_1 (s_0 - s - c_1 - 2c_2). \end{aligned} \tag{8.2}$$

8.2.2 *Quasi-Steady State*

It seems much harder to precisely define QSS, as well as the corresponding QSS assumption, than to illustrate the use of QSS to reduce the dimension of the system. Some authors use a (relatively straightforward) notion of QSS for reactions, which we will consider in Sect. 8.3.3 below. However, the notion of QSS for chemical species, which will be in the focus of this paper, seems more delicate. (The distinction has also been noticed and investigated in detail by Goussis [11]. One also speaks of partial equilibrium instead of QSS for reactions.) It should be emphasized that the choice of a definition for QSS critically influences its translation to mathematical terms, and that various notions exist in the literature. The following characterization (taken from [24]) may be the least common denominator of all definitions:

Working Definition. A reacting system is in *QSS*, or *quasi-stationary*, with respect to certain species, if the rates of change of their concentrations are negligibly small compared to the overall rate of reaction, during some relevant time interval.

A *QSS assumption* amounts to the hypothesis that a reaction is in QSS with respect to certain components.

The source of a QSS assumption generally lies outside mathematics. Usually experimental observations or biological or chemical intuition are invoked. Generally QSS corresponds to restrictions on certain parameters, such as rate constants or initial concentrations.

We give a brief sketch of the history of QSS and mention some contributors to its theory and practice, with no claim to completeness. Michaelis and Menten [20] stated and applied a certain equilibrium assumption, which they did not justify further. Briggs and Haldane [5] seem to be the first who discussed the QSS assumption for the complex C (now sometimes called the *standard QSS* *assumption*) in the Michaelis–Menten system (8.1), and moreover they justified this assumption by referring to smallness of certain parameters in the differential equation. Atkins and de Paula's popular introductory text on Physical Chemistry (see [2, p. 812 ff.]) reflects a frequently used notion of QSS in a reacting system: "(. . .) after an initial induction period (. . .), and during the major part of the reaction, the rates of change of concentrations of all reaction intermediates are negligibly small." The biochemistry text by Stryer et al. (see [3]) seems to make direct use of QSS, with no discussion of underlying assumptions. In the contribution by Rubinow and Segel to the collection [31] (see p. 3 ff.), one finds the following description for (irreversible) Michaelis–Menten: under suitable experimental conditions "one expects that after an initial short transient period there will be a balance between the formation of complex by the union of enzyme and substrate and the breaking apart of complex (. . .)." From a mathematical perspective, the (explicit or implicit) involvement of two different time regimes (initial phase vs. major part of the reaction, to paraphrase Atkins et al.) suggests a singular perturbation approach. One of the earliest papers on QSS from the perspective of Tikhonov's theorem is due to Heineken, Tsuchiya, and Aris [12], with "small parameter" e_0/s_0. Segel [29], and Segel and Slemrod [30] performed a careful analysis of QSS and conditions on parameters. These papers seem to be the starting point for time scale arguments in QSS considerations. Among the many who continued and extended this approach, with varying emphasis on mathematical rigor, we mention Ignetik and Deakin [17]; Ignetik et al. [16]; Borghans et al. [4]; Schnell and Maini [28]; and Tzafriri and Edelman [35]. An approach by Schauer and Heinrich [26] to the Michaelis–Menten system, on the other hand, could be seen as emphasizing the slow manifold in a singular perturbation setting, but their reasoning is essentially based on the assumption that the concentration of the complex is almost constant (more precisely, that $\dot{c} \approx 0$).

In Sect. 8.4 we will review some of these arguments and their use in heuristics for finding small parameters.

8.2.3 *The Ad Hoc Reduction from QSS*

The following reduction method (which we call the ad hoc reduction) is directly related to a QSS assumption: In the differential equation, set the negligible rates of change equal to zero, and use the subsequent algebraic relations to obtain a reduced system.

Example 8.3. QSS for the complex C in the Michaelis–Menten system.

In the irreversible case ($k_{-2} = 0$) one has

$$0\,(=\dot{c}) = k_1 e_0 s - (k_1 s + k_{-1} + k_2)c, \quad \text{thus } c = \frac{k_1 e_0 s}{k_1 s + k_{-1} + k_2}.$$

By substitution one obtains the reduced equation

$$\dot{s} = -\frac{k_2 e_0 s}{(s + (k_{-1} + k_2)/k_1)}$$

which can be found in virtually all books and papers which mention Michaelis–Menten. Note that this approach does not explicitly make use of small parameters, although in its justification in the literature (e.g., [4, 12, 29, 30]) small parameters are frequently invoked. We will discuss a different approach in Example 8.5 below.

In the reversible case the ad hoc method leads to a quadratic equation

$$0\,(=\dot{c}) = k_1 e_0 s - (k_1 s + k_{-1} + k_2)c + k_{-2}(e_0 - c)(s_0 - s - c),$$

which yields

$$c = \frac{1}{2k_{-2}}\left(k_1 s + k_{-1} + k_2 + k_{-2}(e_0 + s_0 - s) \pm \sqrt{(\cdots)}\right)$$

and a relatively cumbersome reduced equation, which is not frequently used. (There are discussions, e.g., in Miller and Alberty [21]; Seshadri and Fritsch [32].)

Example 8.4. QSS in the cooperative system.

Consider the system from Eq. (8.2), and assume QSS for both complexes. In the irreversible case ($k_{-2} = k_{-4} = 0$), solving "$\dot{c}_1 = \dot{c}_2 = 0$," which is a linear parameter-dependent system for c_1 and c_2, provides a nice reduced equation for s; see Keener and Sneyd [18]. But the reversible case leads to a system of quadratic equations for c_1 and c_2, which in turn leads to a reduced equation for s which is intractable, for all practical purposes. See [10, 25] for more details.

Thus the ad hoc reduction, although conceptually straightforward, may become quite inconvenient even in rather simple settings. And, more fundamentally, there remains the question: How, if at all, can a reduction procedure be justified mathematically?

8.3 Reduction in the Presence of Small Parameters

In this section we consider an analytic ordinary differential equation depending on a "small parameter" $\varepsilon \geq 0$. Thus we have

$$\dot{x} = h(x,\varepsilon) = h^{(0)}(x) + \varepsilon h^{(1)}(x) + \ldots, \quad x \in U \subset \mathbb{R}^{n+m}, \tag{8.3}$$

with both n and m positive integers (to be specified below), and we will be interested in the behavior of the solutions as $\varepsilon \to 0$. Our primary focus is on differential equations modeling chemical reactions, and the small parameter may stem either from a separation of fast and slow reactions, or (by some yet-to-be-discussed reasoning; see Sect. 8.4) from a QSS assumption. (For the examples introduced in Sect. 8.2, $\varepsilon = e_0$ works.) But once a small parameter is given, the natural starting point is to try singular perturbation theory.

Due to our focus on chemical reactions, we will impose additional conditions on the right-hand side, which go beyond what is necessary from the perspective of singular perturbation theory. Thus we assume that $h^{(0)}$ and $h^{(1)}$ are polynomials. These assumptions are natural in our setting, since we start from polynomial differential equations of mass action kinetics.

We will mostly rely only on the classical results of singular perturbation theory, see Tikhonov [34], Vasil'eva [36], Fenichel [8], and Hoppensteadt [14]. The monograph [37] by Verhulst, in particular Chapter 8, is an appropriate source for most of the relevant material. The principal new result in this section will be an explicit expression for a reduction of (8.3), given the special assumptions on the right-hand side. We obtain a QSS reduction which is both on solid mathematical ground and relatively simple to compute.

8.3.1 Singular Perturbations

The usual scenario for Tikhonov's and Fenichel's theorems starts with a system in what we call *Tikhonov standard form*:

$$\begin{aligned} \dot{y}_1 &= \qquad\qquad\qquad\quad \varepsilon f^{(1)}(y_1, y_2) + \dots, \quad y_1(0) = y_{1,0}, \\ \dot{y}_2 &= g^{(0)}(y_1, y_2) + \varepsilon g^{(1)}(y_1, y_2) + \dots, \quad y_2(0) = y_{2,0}, \end{aligned} \tag{8.4}$$

with $(y_1,\, y_2) \in D \subseteq \mathbb{R}^n \times \mathbb{R}^m$, D open, and (in our setting) analytic right-hand side.

We obtain the system in "slow time" by rescaling $\tau = \varepsilon t$:

$$\begin{aligned} y_1' &= \qquad\qquad\qquad\qquad f^{(1)}(y_1, y_2) + \dots, \quad y_1(0) = y_{1,0}, \\ y_2' &= \varepsilon^{-1} g^{(0)}(y_1, y_2) + g^{(1)}(y_1, y_2) + \dots, \quad y_2(0) = y_{2,0}. \end{aligned} \tag{8.5}$$

A fundamental result of Tikhonov's theory can be stated as follows. (See Verhulst [37], Theorem 8.1 for a more general theorem under less restrictive hypotheses.)

Theorem 8.1. *Let system* (8.5) *be given. Assume that:*

(i) The zero set $\tilde{Y}$ of $g^{(0)}$ is nonempty.
(ii) There exist a nonempty relatively open subset $\tilde{M}_0 \subseteq \tilde{Y}$ and $\rho > 0$ such that every eigenvalue of $D_2 g^{(0)}(y_1, y_2)$, with $(y_1, y_2) \in \tilde{M}_0$, has real part $\leq -\rho$.

Then there exists $t_1 > 0$ such that for every $t_0 \in (0,\, t_1)$ and for every point sufficiently close to $\tilde{M}_0$, the solution of (8.5) *with initial condition $(y_{1,0},\, y_{2,0})$ approaches the solution of the degenerate system*

$$\begin{aligned} y_1' &= f^{(1)}(y_1, y_2, 0), \quad y_1(0) = y_{1,0}\,, \\ 0 &= g^{(0)}(y_1, y_2, 0) \end{aligned}$$

uniformly on $[t_0,\, t_1]$ *as* $\varepsilon \to 0$.

A priori a system (8.3) derived from a chemical reaction network may not be given in Tikhonov standard form, which raises two questions. First, under what conditions does a transformation to standard form exist? Second, assuming the existence of a transformation, how can a reduced equation be computed?

As for existence, one needs a diffeomorphism $\Psi = (\Psi_1,\, \Psi_2)^{\mathrm{tr}}$ (defined on some open subset of D) which sends solutions of (8.3) to solutions of a system (8.4) in standard form. A necessary and sufficient condition is the identity

$$D\Psi(x)\left(h^{(0)}(x) + \varepsilon h^{(1)}(x) + \dots\right) = \begin{pmatrix} \varepsilon \cdot f^{(1)}(x)(\Psi_1(x), \Psi_2(x)) + \dots \\ g^{(0)}(x)(\Psi_1(x), \Psi_2(x)) + \dots \end{pmatrix}.$$

For $\varepsilon = 0$ one obtains

$$D\Psi(x)h^{(0)}(x) = \begin{pmatrix} 0 \\ g^{(0)}(\Psi_1(x), \Psi_2(x)) \end{pmatrix},$$

and this implies the existence of n independent first integrals (viz., the entries of Ψ_1) for $\dot{x} = h^{(0)}(x)$. Recall that the existence of first integrals is not trivial near stationary points. Moreover, $h^{(0)}$ then admits an n-dimensional local manifold M_0 of stationary points. The following result is taken from [25], but it essentially goes back to Fenichel [8].

Proposition 8.1. *Given* $\dot{x} = h(x, \varepsilon)$*, there exists a transformation* Ψ*, defined on some open* $\tilde{U} \subseteq D$*, to Tikhonov standard form with the eigenvalue condition (ii) from Theorem 8.1, if and only if the following hold:*

The zero set Y *of* $h^{(0)}$ *in* $\tilde{U}$ *is nonempty. Moreover there exist a nonempty relatively open* $M_0 \subseteq Y$ *and* $\rho > 0$ *such that for every* $x_0 \in M_0$ *the derivative* $Dh^{(0)}(x_0)$ *admits the eigenvalue* 0 *with algebraic and geometric multiplicity* n*, and the remaining eigenvalues have real part* $\leq -\rho$*. (In particular* M_0 *is a local* n*-dimensional submanifold.)*

The condition given in Proposition 8.1 implies the existence of a direct sum decomposition

$$\mathbb{R}^{n+m} = \operatorname{Ker} Dh^{(0)}(x_0) \oplus \operatorname{Im} Dh^{(0)}(x_0) \tag{8.6}$$

for every $x_0 \in M_0$. Moreover, this condition implies locally the existence of n independent first integrals for $\dot{x} = h^{(0)}(x)$.

8.3.2 Computing a Reduction

First, we discuss the special case when the hypotheses of Proposition 8.1 are satisfied and a transformation Ψ to Tikhonov standard form (as well as its inverse) is explicitly known. Then determining a reduced system in original coordinates is relatively straightforward. Although Ψ cannot be directly applied to the reduced system in the version

$$y_1' = f^{(1)}(y_1, y_2), \quad g^{(0)}(y_1, y_2) = 0,$$

there is an equivalent version on the invariant manifold $\tilde{M}_0$ introduced in Theorem 8.1, viz.

$$\begin{pmatrix} y_1' \\ y_2' \end{pmatrix} = p(y) := \begin{pmatrix} f^{(1)}(y_1, y_2) \\ -D_2 g^{(0)}(y_1, y_2)^{-1} D_1 g^{(0)}(y_1, y_2) \cdot f^{(1)}(y_1, y_2) \end{pmatrix}, \quad (8.7)$$

which can be transported back via Ψ, to a differential equation with invariant manifold M_0, see [25].

Example 8.5. The reduced system for reversible Michaelis–Menten.

Here the small parameter is (assumed to be) e_0, and system (8.1) gives the function

$$h^{(0)} = \begin{pmatrix} (k_1 s + k_{-1})c \\ -(k_1 s + k_{-1} + k_2 + k_{-2}(s_0 - s - c))c \end{pmatrix}.$$

The differential equation with right-hand side $h^{(0)}$ is a scaled linear system, and a first integral (and therefore a transformation) can be found explicitly. Carrying out the program outlined above (see [25]), one obtains the reduced equation, in addition to $\dot{c} = 0$,

$$\dot{s} = -e_0 \cdot \frac{s(k_1 k_2 + k_{-1} k_{-2}) - k_{-1} k_{-2} s_0}{k_1 s + k_{-1} + k_2 + k_{-2}(s_0 - s)},$$

which is generally different from the ad hoc-reduced equation, and actually appears less complicated (no square roots). But note that the Tikhonov–Fenichel reduction coincides with the ad hoc reduction when $k_{-2} = 0$.

If an explicit transformation is known, it may provide an additional benefit because Theorem 8.1 in Verhulst [37] characterizes the admissible initial conditions. Moreover, for Michaelis–Menten one can verify Hoppensteadt's [14] criteria for convergence on the interval $[t_0, \infty)$ (notation from Theorem 8.1).

Generally, one cannot hope for an explicit construction of a transformation to Tikhonov standard form, but still it is possible to compute a reduced equation. If the slow manifold M_0 can be explicitly represented as the graph of some function, Fenichel ([8], Lemma 5.4) and Stiefenhofer ([33], Equation (2.13), with a different and quite short proof) gave expressions for the reduced equation.

One can carry Fenichel's observations further to obtain a reduced system in the general setting. A closer look at Eq. (8.7) shows that one gets $p(y)$ via the kernel-image decomposition of the derivative

$$\begin{pmatrix} 0 & 0 \\ D_1 g^{(0)}(y) & D_2 g^{(0)}(y) \end{pmatrix}$$

(compare Eq. (8.6)) by computing the kernel component of $(f^{(1)}(y), g^{(1)}(y))^{\mathrm{tr}}$. Since the kernel-image decomposition is preserved in coordinate transformations, one obtains (see [25]):

Proposition 8.2. *Given the eigenvalue condition from Proposition 8.1 for transformability to Tikhonov standard form, near a point x_0 with $h^{(0)}(x_0) = 0$, one obtains the reduced system of (8.3) by computing the kernel component of $h^{(1)}(x)$ with respect to the direct kernel-image decomposition of $Dh^{(0)}(x)$.*

As noted in [25], the projection onto the kernel can be found from the minimal polynomial of $Dh^{(0)}(x)$, and for some—relatively small—examples this approach is computationally feasible. For higher dimensions and many parameters, this procedure becomes prohibitively expensive. But in any case the argument shows that for polynomial (or rational) $h^{(0)}$ and $h^{(1)}$ the reduced system will have a rational right-hand side.

A practicable method to compute reduced systems was recently developed in [9]. It is based on an auxiliary result from classical Algebraic Geometry.

Lemma 8.1. *For system (8.3) with polynomials (or rational functions) $h^{(0)}$ and $h^{(1)}$, let x_0 be such that $h^{(0)}(x_0) = 0$ and that the eigenvalue condition from Proposition 8.1 hold for $Dh^{(0)}(x_0)$, with $m = \text{rank } Dh^{(0)}(x_0)$. Then there exist a $(n+m) \times m$ matrix P with rational entries, of rank m, and a vector valued polynomial μ with m entries, such that*

$$h^{(0)}(x) = P(x)\mu(x)$$

in some Zariski neighborhood of x_0. By appropriate choice of the neighborhood, one may assume that $h^{(0)}$ and μ have the same zero sets. The entries of μ may be taken as any m independent entries of $h^{(0)}$.

This Lemma, which is proved in [9], is almost trivial in the local analytic (or differentiable) setting, in view of the Implicit Function Theorem. But the point is that P is rational, and that there are constructive methods to determine P. With this auxiliary result, the reduction is relatively straightforward, as is shown in the next theorem.

Theorem 8.2. *For system (8.3), with assumptions as in Lemma 8.1, let x_0 be such that $h^{(0)}(x_0) = 0$. Then the reduced system, in a Zariski neighborhood M_0 of x_0 in the zero set Y of $h^{(0)}$, is given (in slow time) by*

$$x' = h^{(1)}(x) - P(x)\left(D\mu(x)\,P(x)\right)^{-1} D\mu(x)h^{(1)}(x).$$

Proof. The eigenvalue conditions at x_0 guarantee that Y is locally an n-dimensional manifold. Let M_0 be a relatively open subset of Y such that the eigenvalue conditions hold for all points of M_0. Denote the columns of P by $p_1, \ldots, p_m$. The Jacobian matrix of $h^{(0)}$ equals

$$Dh^{(0)}(x) = \sum_{i=1}^{m} \left(p_i(x)D\mu_i(x) + \mu_i(x)Dp_i(x)\right)$$

in a Zariski neighborhood of x_0, and therefore

$$Dh^{(0)}(x) = P(x) \cdot D\mu(x) \quad \text{for all } x \in M_0. \tag{8.8}$$

Now fix $x \in M_0$. Then

$$\text{Ker } Dh^{(0)}(x) = \text{Ker } D\mu(x), \tag{8.9}$$

due to the rank condition for $P(x)$. The condition $\operatorname{rank} P(x) = m$ also implies

$$\text{Im } Dh^{(0)}(x) = \text{Im } P(x).$$

From our basic hypothesis we have the direct sum decomposition (8.6). Set

$$A(x) := D\mu(x) \cdot P(x).$$

We first show that $A(x)$ is invertible. Thus let $\beta \in \mathbb{R}^m$ be a solution of the equation

$$D\mu(x)P(x)\beta = 0. \tag{8.10}$$

The direct sum decomposition and

$$P(x)\beta \in \text{Ker } Dh^{(0)}(x) \cap \text{Im } Dh^{(0)}(x)$$

show $P(x)\beta = 0$. Since $P(x)$ has full rank, we see $\beta = 0$. Thus Eq. (8.10) admits only the trivial solution, whence $A(x) = D\mu(x)P(x)$ is invertible.

Moreover, due to the direct sum decomposition (8.6), for any $y \in \mathbb{R}^{n+m}$ there exist $z \in \text{Ker } Dh^{(0)}(x) = \text{Ker } D\mu(x)$ and $\alpha \in \mathbb{R}^m$ such that

$$y = z + P(x)\alpha.$$

Since $z = y - P(x)\alpha \in \text{Ker } D\mu(x)$, one finds

$$D\mu(x)\left(y - P(x)\alpha\right) = 0,$$

which implies $\alpha = A(x)^{-1} D\mu(x)y$, and thus

$$z = y - P(x)A(x)^{-1} D\mu(x)y.$$

Apply this to $h^{(1)}(x)$ to obtain the assertion. □

Remark 8.1. (a) It may be appropriate to discuss invertibility of $A(x)$ and the eigenvalue condition in more detail. The zero set Y of $h^{(0)}$ is an algebraic variety in $\mathbb{R}^{n+m}$, and we are actually interested in an n-dimensional component M_0 of this variety. For $x \in Y$, $Dh^{(0)}(x)$ must therefore have eigenvalue 0 with geometric multiplicity n. If the geometric and algebraic multiplicity are equal to n then (and only then) the kernel-image decomposition (8.6) exists, and the latter is equivalent to invertibility of $A(x)$. Thus it is possible to write down the equation in Theorem 8.2. But additional conditions (for instance, all real parts of eigenvalues $\leq -\rho$) are necessary to make this a meaningful reduced system.

(b) The matrix $A(x)$ is of size $m \times m$, hence relatively small. One should also emphasize that inverting this matrix is not actually necessary to determine the reduced system: It suffices to solve a system of linear equations with this matrix.

(c) The eigenvalue condition (ii) from Theorem 8.1 on $Dh^{(0)}(x)$ is satisfied if and only if all eigenvalues of $A(x)$ have real part $\leq -\rho$. Indeed, by virtue of Eq. 8.8 and linear algebra the nonzero eigenvalues of these two matrices are the same.

Example 8.6. Irreversible Michaelis–Menten with slow product formation.
This is an example for a slow–fast reaction separation. One considers the familiar differential equation (8.1), but now with small parameter k_2. The underlying assumption is that product formation is much slower than formation of complex and degrading of complex back to enzyme and substrate. One has (with the arguments s, c suppressed)

$$h^{(0)} = \begin{pmatrix} -\mu \\ \mu \end{pmatrix}, \quad P = \begin{pmatrix} -1 \\ 1 \end{pmatrix}, \quad \mu = k_1 e_0 s - (k_1 s + k_{-1})c, \quad h^{(1)} = \begin{pmatrix} 0 \\ -c \end{pmatrix}.$$

Thus

$$A = (k_1(e_0 - c),\, -(k_1 s + k_{-1})) \begin{pmatrix} -1 \\ 1 \end{pmatrix} = -\left(k_1(e_0 - c) + k_1 s + k_{-1}\right)$$

and the reduction procedure yields the system

$$\begin{pmatrix} \dot{s} \\ \dot{c} \end{pmatrix} = -\frac{k_2}{k_1(e_0 - c) + k_1 s + k_{-1}} \begin{pmatrix} (k_1 s + k_{-1})c \\ k_1(e_0 - c)c \end{pmatrix}$$

on the invariant variety M_0^* defined by $\mu = 0$. Using the parametrization of M_0 one may obtain a reduced equation for substrate alone, viz.

$$\dot{s} = -\frac{k_2 k_1 e_0 s\,(k_1 s + k_{-1})}{k_1 k_{-1} e_0 + (k_1 s + k_{-1})^2}$$

This is different from the standard reduction based on QSS for complex. This example illustrates that different QSS assumptions (QSS for the species C, resp. QSS with slow product formation) will lead to different reductions.

Example 8.7. The cooperative system with small parameter e_0 (see Example 8.2 above) was originally discussed in [25], with the minimal polynomial approach (and serious reliance on a computer algebra system). Theorem 8.2 makes this computation feasible even by hand. With the abbreviations

$$\begin{aligned}\alpha &= -(k_1+k_3)s-(k_{-1}+k_2)-(k_{-2}+k_{-4})(s_0-s-c_1-2c_2)\\ \beta &= -k_1 s+k_{-3}+k_4-k_{-2}(s_0-s-c_1-2c_2)\end{aligned}$$

one has

$$h^{(0)} = \begin{pmatrix} (k_{-1}+k_1 s-k_3 s)c_1+(k_1 s+k_{-3})c_2 \\ c_1\alpha+c_2\beta \\ k_3 c_1 s-(k_{-3}+k_4)c_2+k_{-4}c_1(s_0-s-c_1-2c_2) \end{pmatrix},$$

$$h^{(1)} = \begin{pmatrix} -k_1 s \\ k_1 s+k_{-2}(s_0-s-c_1-2c_2) \\ 0 \end{pmatrix},$$

and the (relevant component of the) zero set of $h^{(0)}$ is given by $c_1 = c_2 = 0$. A decomposition according to Lemma 8.1 is given by

$$P = \begin{pmatrix} k_{-1}+k_1 s-k_3 s & k_1 s+k_{-3} \\ \alpha & \beta \\ k_3 s+k_{-4}(s_0-s-c_1-2c_2) & -k_{-3}-k_4 \end{pmatrix}$$

and

$$\mu = \begin{pmatrix} c_1 \\ c_2 \end{pmatrix}.$$

Note that such a decomposition is not unique. For $A = D\mu \cdot P$ one obtains

$$A(s,\,0,\,0) = \begin{pmatrix} a(s) & -k_1 s+k_{-3}+k_4-k_{-2}(s_0-s) \\ k_3 s+k_{-4}(s_0-s) & -k_{-3}-k_4 \end{pmatrix}$$

with abbreviation $a(s) = -(k_1+k_3)s-(k_{-1}+k_2)-(k_2+k_{-4})(s_0-s)$. The matrix A can easily be inverted, and the eigenvalue condition is readily checked. The final result for the reduced equation is, of course, identical to the one given in [25].

Example 8.8. To illustrate the feasibility of the approach via Theorem 8.2, we discuss a somewhat bigger example, from Stiefenhofer [33, Section 3], whose reduction was computed in [33] only for some special parameter choices. This system models communication between slime mold cells such as *Dictyostelium discoideum*. Communication is effected by $cAMP$, denoted by P, furthermore S stands for the substrate ATP, while D and R represent transmembrane receptors, with $\overline{D}$ and $\overline{R}$ representing the corresponding bound states. (See [33] for more details.) The reaction scheme, including production and decomposition of $cAMP$ with constant rates, can be written as follows.

$$\begin{array}{lll} D \underset{\varepsilon k_{-1}}{\overset{\varepsilon k_1}{\rightleftharpoons}} R & S+\overline{R} \overset{\varepsilon k_2}{\rightharpoondown} P+\overline{R} & \overset{\varepsilon k_3}{\rightharpoondown} P \overset{\varepsilon k_{-3}}{\rightharpoondown} \\ \overline{D} \underset{k_{-4}}{\overset{k_4}{\rightleftharpoons}} D+2P & \overline{R} \underset{k_{-5}}{\overset{k_5}{\rightleftharpoons}} R+2P, & \end{array}$$

with parameters $k_i > 0$, and the ε's indicating the slow reactions. We also adopt the further simplification $s(t) = S > 0$ from [33].

We use the first integral $\overline{d} = c - d - r - \overline{r}$, with some constant $c \geq 0$, to obtain the system

$$\begin{aligned} \dot{p} &= -2k_4dp^2 + 2k_{-4}(c-d-r-\overline{r}) - 2k_5rp^2 + 2k_{-5}\overline{r} + \varepsilon k_3 - \varepsilon k_{-3}p + \varepsilon k_2S\overline{r}, \\ \dot{d} &= -k_4dp^2 + k_{-4}(c-d-r-\overline{r}) - \varepsilon k_1d + \varepsilon k_{-1}r, \\ \dot{r} &= -k_5rp^2 + k_{-5}\overline{r} + \varepsilon k_1d - \varepsilon k_{-1}r, \\ \dot{\overline{r}} &= k_5rp^2 - k_{-5}\overline{r}. \end{aligned} \tag{8.11}$$

With

$$\mu(p,d,r,\overline{r}) := \begin{pmatrix} -k_5rp^2 + k_{-5}\overline{r} \\ -k_4dp^2 + k_{-4}(c-d-r-\overline{r}) \end{pmatrix}, \quad P(p,d,r,\overline{r}) := \begin{pmatrix} 2 & 2 \\ 0 & 1 \\ 1 & 0 \\ -1 & 0 \end{pmatrix},$$

and

$$h^{(1)}(p,d,r,\overline{r}) := \begin{pmatrix} k_3 - k_{-3}p + k_2S\overline{r} \\ -k_1d + k_{-1}r \\ k_1d - k_{-1}r \\ 0 \end{pmatrix},$$

Equation (8.11) may be written as

$$\frac{d}{dt}\begin{pmatrix} p \\ d \\ r \\ \overline{r} \end{pmatrix} = P \cdot \mu + \varepsilon h^{(1)},$$

which is a representation according to Lemma 8.1. The variety Y may be taken as the zero set of μ, and one verifies that the choice

$$M_0 := Y \cap \operatorname{int} \mathbb{R}^4_+,$$

is possible. Straightforward computations show that the eigenvalue condition is at least generically satisfied, and the reduced system on the invariant set M_0 is given by

$$\frac{d}{dt}\begin{pmatrix} p \\ d \\ r \\ \overline{r} \end{pmatrix} = \frac{1}{Q}\begin{pmatrix} N_1 \\ N_2 \\ N_3 \\ N_4 \end{pmatrix}$$

on a suitable subset $W \subset \mathbb{R}^2$, which is determined from $M_0 \cap \mathbb{R}^4_+$ by this elimination, with

$$\begin{aligned}
N_1 :=\ & -k_{-3}p^5k_5k_4 + (k_2S\overline{r}\,k_5k_4 + k_3k_5k_4)\,p^4 - k_{-3}\,(k_{-5}k_4 + k_{-4}k_5)\,p^3 \\
& +\big(2\,k_1\,(-k_{-4}k_5 + k_{-5}k_4)\,d - 2\,k_{-1}\,(-k_{-4}k_5 + k_{-5}k_4)\,r \\
& +Sk_2\,(k_{-5}k_4 + k_{-4}k_5)\,\overline{r} + k_3\,(k_{-5}k_4 + k_{-4}k_5)\,\big)p^2 - k_{-3}pk_{-5}k_{-4} + k_3k_{-5}k_{-4} \\
& +k_2S\overline{r}\,k_{-5}k_{-4}, \\
N_2 :=\ & 2\,k_4dp^4k_5k_{-3} + (-2\,k_3k_5k_4 - 2\,k_2S\overline{r}\,k_5k_4)\,dp^3 + \big((2\,k_4k_{-5}k_{-3} - k_5k_{-4}k_1)\,d \\
& +k_5k_{-4}k_{-1}r\big)p^2 + \big(-4\,k_1d^2k_4k_{-5} + \big((-4\,k_5k_{-4}k_1 + 4\,k_{-1}k_4k_{-5})\,r \\
& -2\,k_4k_{-5}k_2S\overline{r} - 2\,k_4k_{-5}k_3\big)d + 4\,k_{-1}r^2k_{-4}k_5\big)p - k_1dk_{-5}k_{-4} + k_{-1}rk_{-5}k_{-4}, \\
N_3 :=\ & 2\,k_5rp^4k_4k_{-3} + (-2\,k_3k_5k_4 - 2\,k_2S\overline{r}\,k_5k_4)\,rp^3 + \big(k_{-5}k_4k_1d \\
& +(2\,k_5k_{-4}k_{-3} - k_{-1}k_4k_{-5})\,r\big)p^2 + \big(4\,k_1d^2k_4k_{-5} \\
& +(-4\,k_{-1}k_4k_{-5} + 4\,k_5k_{-4}k_1)\,rd - 4\,k_{-1}r^2k_{-4}k_5 + \big(-2\,k_5k_{-4}k_3 \\
& -2\,k_5k_{-4}k_2S\overline{r}\big)r\big)p + k_1dk_{-5}k_{-4} - k_{-1}rk_{-5}k_{-4}, \\
N_4 :=\ & (k_5k_4k_1d - k_5k_4\,(k_{-1} + 2\,k_{-3})\,r)\,p^4 + \big(4\,k_5k_4k_1d^2 + 4\,k_5k_4\,(-k_{-1} + k_1)\,rd \\
& -4\,k_5k_{-1}k_4r^2 + (2\,k_2S\overline{r}\,k_5k_4 + 2\,k_3k_5k_4)\,r\big)p^3 + \big(k_5k_{-4}k_1d - k_5k_{-4}\big(k_{-1} \\
& +2\,k_{-3}\big)r\big)p^2 + (2\,k_5k_{-4}k_2S\overline{r} + 2\,k_5k_{-4}k_3)\,rp
\end{aligned}$$

and

$$\begin{aligned}
Q :=\ & k_5p^4k_4 + (4\,k_4k_5r + 4\,k_4dk_5)\,p^3 + (k_{-5}k_4 + k_{-4}k_5)\,p^2 \\
& +\,(4\,k_5rk_{-4} + 4\,k_4dk_{-5})\,p + k_{-5}k_{-4}.
\end{aligned}$$

One may eliminate the variables r and $\overline{r}$ via

$$M_0 = \left\{(p,d,r,\overline{r})^{\mathrm{tr}};\, r = \frac{k_{-5}(-k_4dp^2 + k_{-4}(c-d))}{k_{-4}(k_{-5} + k_5p^2)}, \overline{r} = \frac{k_5p^2(-k_4dp^2 + k_{-4}(c-d))}{k_{-4}(k_{-5} + k_5p^2)}\right\},$$

and obtain the equivalent version

$$\frac{d}{dt}\begin{pmatrix} p \\ d \end{pmatrix} = \frac{1}{\tilde{Q}}\begin{pmatrix} \tilde{N}_1 \\ \tilde{N}_2 \end{pmatrix}$$

with

$$\begin{aligned}\tilde{N}_1 := {} & -p^{10}Sd{k_5}^3{k_4}^2k_2 - p^9{k_5}^3k_4\,k_{-4}\,k_{-3} + p^8(cStk_5{}^3k_4\,k_{-4}\,k_2 \\ & -2\,Sd{k_5}^3k_4\,k_{-4}\,k_2 - 2\,Sd{k_5}^2k_{-5}\,{k_4}^2k_2{k_5}^3k_4\,k_{-4}\,k_3) + p^7(-{k_5}^3{k_{-4}}^2k_{-3} \\ & -3\,{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_{-3}) + p^6(cS{k_5}^3{k_{-4}}^2k_2 + 2\,cS{k_5}^2k_{-5}\,k_4, k_{-4}\,k_2 - Sd{k_5}^3{k_{-4}}^2k_2 \\ & -4\,Sd{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_2 - Sdk_5\,{k_{-5}}^2{k_4}^2k_2 3\,{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_3 - 2\,d{k_5}^3{k_{-4}}^2k_1 \\ & +2\,d{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_1 - 2\,d{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_{-1} + 2\,dk_5\,{k_{-5}}^2{k_4}^2k_{-1} + {k_5}^3{k_{-4}}^2k_3) \\ & +p^5(-3\,{k_5}^2k_{-5}\,{k_{-4}}^2k_{-3} - 3\,k_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-3}) + p^4(2\,cS{k_5}^2k_{-5}\,{k_{-4}}^2k_2 \\ & +cSk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_2 - 2\,Sd{k_5}^2k_{-5}\,{k_{-4}}^2k_2 - 2\,Sdk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_2 \\ & +2\,c{k_5}^2k_{-5}\,{k_{-4}}^2k_{-1} - 2\,ck_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-1} - 4\,d{k_5}^2k_{-5}\,{k_{-4}}^2k_1 \\ & -2\,d{k_5}^2k_{-5}\,{k_{-4}}^2k_{-1} + 4\,dk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_1 + 2\,d{k_{-5}}^3{k_4}^2k_{-1} \\ & +3\,k_5\,{k_{-5}}^2k_4\,k_{-4}\,k_3) + p^3(-3\,k_5\,{k_{-5}}^2{k_{-4}}^2k_{-3} - {k_{-5}}^3k_4\,k_{-4}\,k_{-3}) \\ & +p^2(cSk_5\,{k_{-5}}^2{k_{-4}}^2k_2 + 3\,{k_5}^2k_{-5}\,{k_{-4}}^2k_3 - Sdk_5\,{k_{-5}}^2{k_{-4}}^2k_2 \\ & +2\,ck_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} - 2\,c{k_{-5}}^3k_4\,k_{-4}\,k_{-1} - 2\,dk_5\,{k_{-5}}^2{k_{-4}}^2k_1 \\ & -2\,dk_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} + 2\,d{k_{-5}}^3k_4\,k_{-4}\,k_1 + 2\,d{k_{-5}}^3k_4\,k_{-4}\,k_{-1} + 3\,k_5\,{k_{-5}}^2{k_{-4}}^2k_3 \\ & +{k_{-5}}^3k_4\,k_{-4}\,k_3) - p{k_{-5}}^3{k_{-4}}^2k_{-3} + {k_{-5}}^3{k_{-4}}^2k_3,\end{aligned}$$

$$\begin{aligned}\tilde{N}_2 := {} & p^9(2\,Sd^2{k_5}^3{k_4}^2k_2) + p^8(2\,d{k_5}^3k_4\,k_{-4}\,k_{-3}) + p^7(-2\,cSd{k_5}^3k_4\,k_{-4}\,k_2 \\ & +2\,Sd^2{k_5}^3k_4\,k_{-4}\,k_2 + 4\,Sd^2{k_5}^2k_{-5}\,{k_4}^2k_2 - 2\,d{k_5}^3k_4\,k_{-4}\,k_3) + p^6(-d{k_5}^3{k_{-4}}^2k_1 \\ & +6\,d{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_{-3} - d{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_{-1}) + p^5(-6\,d{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_3 \\ & -4\,cSd{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_2 + 4\,Sd^2{k_5}^2k_{-5}\,k_4\,k_{-4}\,k_2 + 2\,Sd^2k_5\,{k_{-5}}^2{k_4}^2k_2) \\ & +p^4(+c{k_5}^2k_{-5}\,{k_{-4}}^2k_{-1} - 3\,d{k_5}^2k_{-5}\,{k_{-4}}^2k_1 - d{k_5}^2k_{-5}\,{k_{-4}}^2k_{-1} \\ & +6\,dk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-3} - 2\,dk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-1}) + p^3(+4\,d^2{k_5}^2k_{-5}\,{k_{-4}}^2k_1 \\ & -4\,d^2k_5\,{k_{-5}}^2k_4\,k_{-4}\,k_1 + 4\,d^2k_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-1} - 4\,d^2{k_{-5}}^3{k_4}^2k_{-1} \\ & -6\,dk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_3 - 2\,cSdk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_2 + 2\,Sd^2k_5\,{k_{-5}}^2k_4\,k_{-4}\,k_2 \\ & -4\,cdk_5^2k_{-5}\,{k_{-4}}^2k_1 - 4\,cdk_5\,{k_{-5}}^2k_4\,k_{-4}\,k_{-1}) + p^2(2\,ck_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} \\ & -3\,dk_5\,{k_{-5}}^2{k_{-4}}^2k_1 - 2\,dk_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} + 2\,d{k_{-5}}^3k_4\,k_{-4}\,k_{-3} \\ & -d{k_{-5}}^3k_4\,k_{-4}\,k_{-1}) + p(4\,c^2k_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} - 4\,cdk_5\,{k_{-5}}^2{k_{-4}}^2k_1 \\ & -8\,cdk_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} + 4\,cd{k_{-5}}^3k_4\,k_{-4}\,k_{-1} + 4\,d^2k_5\,{k_{-5}}^2{k_{-4}}^2k_1 \\ & +4\,d^2k_5\,{k_{-5}}^2{k_{-4}}^2k_{-1} - 4\,d^2{k_{-5}}^3k_4\,k_{-4}\,k_1 - 4\,d^2{k_{-5}}^3k_4\,k_{-4}\,k_{-1} \\ & -2\,d{k_{-5}}^3k_4\,k_{-4}\,k_3) + c{k_{-5}}^3{k_{-4}}^2k_{-1} - d{k_{-5}}^3{k_{-4}}^2k_1 - d{k_{-5}}^3{k_{-4}}^2k_{-1},\end{aligned}$$

$$\begin{aligned}\tilde{Q} := {} & \big(p^6{k_5}^2k_4\,k_{-4} + p^5(4\,d{k_5}^2k_4\,k_{-4} - 4\,dk_5\,k_{-5}\,{k_4}^2) + p^4({k_5}^2{k_{-4}}^2 + 2\,k_5\,k_{-5}\,k_4\,k_{-4}) \\ & +p^3(4\,ck_5\,k_{-5}\,k_4\,k_{-4}) + p^2(2\,k_5\,k_{-5}\,{k_{-4}}^2 + {k_{-5}}^2k_4\,k_{-4}) + p(4\,ck_5\,k_{-5}\,{k_{-4}}^2 \\ & -4\,dk_5\,k_{-5}\,{k_{-4}}^2 + 4\,d{k_{-5}}^2k_4\,k_{-4}) + {k_{-5}}^2{k_{-4}}^2\big)\big(p^2k_5 + k_{-5}\big).\end{aligned}$$

Up to scaling of time, one therefore obtains a two-dimensional equation with polynomial right-hand side (of degree 11) on W. ($\tilde{Q} > 0$ on W follows from $Q > 0$ on $\mathbb{R}^4_+$.) In particular one has Poincaré–Bendixson theory available in the asymptotic limit.

8.3.3 Slow and Fast Reactions

For slow and fast reactions the reduction program via Tikhonov–Fenichel was carried out by Schauer and Heinrich [27] (who cited Vasil'eva [36] for singular perturbation results), and continued by Stiefenhofer [33]. (A recent paper by Lee and Othmer [19] reproduces several of these results.)

Before discussing slow and fast reactions in some detail, it will be necessary to give a more precise outline of the work by Feinberg [7], Horn and Jackson [15], and others. We mostly follow Feinberg's Lecture Notes [7]; a short overview can be found in Section 2 of Anderson's recent paper [1]. We sketch the formalism for chemical reaction networks and reaction systems with mass action kinetics, using the irreversible Michaelis–Menten system as an illustrating example. One starts with an ordered collection of q chemical *species*, which are identified with the standard basis of $\mathbb{R}^q$. Next one forms *complexes*, which formally speaking are nonnegative (integer) linear combinations of species (appearing as reactants or as reaction products). Then *reactions* are defined as ordered pairs of complexes, usually written in a notation with reaction arrows. (The notion of reversible reaction is obvious). For Michaelis–Menten the species are E, S, C, and P, which will in the following be identified with the standard basis vectors of $\mathbb{R}^4$. Moreover one has complexes $E+S$, C, $E+P$, and reactions

$$E+S \to C; \quad C \to E+S; \quad C \to E+P.$$

Using the identification of species and standard basis vectors, one assigns to each reaction a vector in $\mathbb{R}^q$, counting the reactants with negative sign, calls their span in $\mathbb{R}^q$ the *stoichiometric subspace* S, and collects these column vectors in a matrix Z which is related to the stoichiometric matrix as defined by Feinberg. For the Michaelis–Menten example one has, in the above order, column vectors

$$\begin{pmatrix} -1 \\ -1 \\ 1 \\ 0 \end{pmatrix}, \quad \begin{pmatrix} 1 \\ 1 \\ -1 \\ 0 \end{pmatrix}, \quad \begin{pmatrix} 1 \\ 0 \\ -1 \\ 1 \end{pmatrix}, \text{ thus } Z = \begin{pmatrix} -1 & 1 & 1 \\ -1 & 1 & 0 \\ 1 & -1 & -1 \\ 0 & 0 & 1 \end{pmatrix}.$$

The differential equation for the concentrations may now be written in the form

$$\frac{d}{dt}\begin{pmatrix} e \\ s \\ c \\ p \end{pmatrix} = \begin{pmatrix} -1 & 1 & 1 \\ -1 & 1 & 0 \\ 1 & -1 & -1 \\ 0 & 0 & 1 \end{pmatrix} \cdot \begin{pmatrix} k_1 es \\ k_{-1}c \\ k_2 c \end{pmatrix}$$

and generally for mass action kinetics one obtains a differential equation system of the form

$$\dot{x} = Z \cdot \Phi(x) \tag{8.12}$$

for the vector of concentrations. Φ can be characterized more precisely; see [7] for details.

There is a natural assignment of a directed graph to a reaction network: The nodes are the complexes, and there exists a directed edge from one complex to another if and only if there is a reaction from the former to the latter. The connected components of this graph are called *linkage classes*. The *deficiency* of a network is then defined by

$$\delta := \#\,\text{complexes} - \#\,\text{linkage classes} - \operatorname{rank} Z,$$

and one can show that $\delta \geq 0$. Finally, one calls the reaction network *weakly reversible* if, whenever there is a reaction from one complex to another, there is also a chain of reactions from the latter to the former.

Both $\mathbb{R}_+^q$ and its interior are positively invariant for the system (8.12). The cosets $x^* + S$, with S the stoichiometric subspace and $0 \neq x^* \in \mathbb{R}_+^q$, are called the *stoichiometric compatibility classes*, and are positively invariant sets for the differential equation. Now we can state one fundamental result of the theory.

Deficiency Zero Theorem (Horn and Jackson [15], Feinberg [7]). *Assume that* (8.12) *corresponds to a weakly reversible deficiency zero network. Then the following hold.*

(i) The intersection of every stoichiometric compatibility class with $\operatorname{int}\mathbb{R}_+^q$ *contains exactly one stationary point.*

(ii) This point is locally asymptotically stable within its compatibility class.

Remark 8.2. (a) The proof of part (ii) is based on an ingenious choice of a Lyapunov function. Linear asymptotic stability cannot be deduced from the inequalities in this argument.

(b) In Feinberg's Notes [7] a stronger claim is made, viz. global asymptotic stability within the intersection of the stoichiometric compatibility class and $\operatorname{int}\mathbb{R}_+^q$. Later a problem in the global stability argument was pointed out; and generally global stability is still a conjecture. Only recently Anderson [1] succeeded with a proof in the case of a single linkage class.

Now we turn to slow–fast systems of chemical reactions. These are usually described by

$$\dot{x} = h^{(0)}(x) + \varepsilon h^{(1)}(x), \quad x \in \mathbb{R}^q \tag{8.13}$$

with the fast subsystem $h^{(0)}$ (large rate constants) and the slow subsystem $\varepsilon h^{(1)}$ (small rate constants, symbolized by the factor ε). Thus both $h^{(0)}$ and $h^{(1)}$ admit a representation of the form (8.12). A substantial part of the following result is due to Schauer and Heinrich [27]. The transfer from Schauer and Heinrich's condition to weakly reversible deficiency zero systems in the statement and proof of the following Proposition is a first step, in ongoing work [9], toward a more comprehensive theorem.

Proposition 8.3. *Assume that the fast subsystem of* (8.13) *has deficiency zero and is weakly reversible. Assume moreover that every stationary point in* $\operatorname{int}\mathbb{R}_+^q$ *is linearly asymptotically stable for* $h^{(0)}$ *within its stoichiometric compatibility class. Then the following hold.*

(a) The eigenvalue conditions from Proposition 8.1 are satisfied for $Dh^{(0)}(x_0)$ *at all zeros* $x_0 \in \operatorname{int} \mathbb{R}_+^q$ *of* $h^{(0)}$.
(b) There exists a linear transformation of the system to Tikhonov standard form.

Proof. We write

$$h^{(0)}(x) = Z \cdot \Phi(x)$$

and let $x_0 \in \operatorname{int} \mathbb{R}_+^q$ be a stationary point. Let s be the dimension of the stoichiometric subspace, thus $\operatorname{rank} Z = s$. Then

$$Dh^{(0)}(x_0) = Z\, D\Phi(x_0)$$

admits the eigenvalue 0 with multiplicity $\geq q - s$ (due to the rank of Z) and has s eigenvalues with negative real part, due to the linear stability requirement. In particular

$$\operatorname{rank} Z = \operatorname{rank} Dh^{(0)}(x_0). \tag{8.14}$$

Therefore the zero set of $h^{(0)}$ is locally a manifold of dimension s. Moreover there are independent linear forms $\lambda_1, \ldots, \lambda_{q-s}$ such that $\lambda_i \circ Z = 0$, $1 \leq i \leq q - s$, and these are also first integrals of $h^{(0)}$. Completing these by suitable linear forms to a basis of the dual of $\mathbb{R}^q$ will produce the desired transformation to Tikhonov standard form. □

Remark 8.3. The importance of the rank condition (8.14) for the existence of a linear transformation to standard form was first noted by Schauer and Heinrich [27]. They also stated (with only a partial justification for some special cases, it seems; see [27, Section 4]) that the rank condition holds when every fast reaction is reversible with fast reverse reaction. It seems that linear stability conditions did not play a role in [27].

8.3.4 Why Does the Ad Hoc Method Persist?

As noted earlier, the ad hoc reduction produces the same result as Tikhonov–Fenichel in some relevant cases, but not in general. In [10] we provide a detailed investigation for several basic reaction schemes in biochemistry (including Michaelis–Menten), with the result that ad hoc and Tikhonov–Fenichel reduction coincide when certain product-forming reactions are irreversible, but differ in the fully reversible case. Since such reductions are actually used to determine rate constants and other reaction parameters, it is very likely that serious discrepancies between ad hoc reduction and reality would have been noticed in experimental verification. To explain this apparent lack of serious discrepancy, we note two possible reasons for good approximation by the irreversible reduced system.

First, some reversible reactions may be almost irreversible (for instance, k_{-2} may be very small in the Michaelis–Menten example). Since the reduced system in the irreversible case is the limit of the reversible case, the discrepancy may be hardly noticeable.

Second, continuous removal of product may be responsible, as noted in Heinrich and Schuster [13], Keener and Sneyd [18]. A thorough justification of this argument was also given in [10] for reversible Michaelis–Menten with product removal (rate $\alpha > 0$). Indeed, a Tikhonov–Fenichel reduction of the system

$$\begin{aligned}\dot{s} &= -k_1 e_0 + (k_1 s + k_{-1})c,\\ \dot{c} &= k_1 e_0 s - (k_1 s + k_{-1} + k_2)c + k_{-2}(e_0 - c)p,\\ \dot{p} &= k_2 c - k_{-2} p(e_0 - c) - \alpha p\end{aligned}$$

with "small parameter" e_0 yields the familiar "irreversible reduced equation" (with α vanishing along the way). See Examples 8.3 and 8.5 with $k_{-2} = 0$.

8.4 Finding Small Parameters

While the results in the previous section are based on a well-defined mathematical scenario, there is another facet of QSS which, in the present stage, is not so amenable to rigorous mathematics. The underlying problem is that the translation of a model assumption to mathematical terms is rarely straightforward, and it may depend on seemingly small details. Here we are concerned with translating certain assumptions on chemical reacting systems—in particular QSS assumptions—to mathematical terms.

8.4.1 Underlying Assumptions: QSS vs. Slow–Fast

Frequently QSS assumptions—directly or indirectly—amount to slow–fast hypotheses, and we briefly review some of these.

A direct slow–fast assumption (small and large rate constants) underlies the discussion of slow and fast reactions, as in Eq. (8.13). As noticed above, this is different from a QSS assumption for chemical species, which we discuss now. An indirect slow–fast assumption for species (based on the fact that the linearization of a system (8.4) in Tikhonov standard form necessarily has some very small eigenvalues near the slow manifold) works by seeking conditions to ensure a very small ratio of absolutely smallest to absolutely largest eigenvalue of the linearization (near some submanifold). This was used, for instance, by Duchêne and Rouchon [6], but the method frequently has to rely on numerical calculations, and general parameter conditions seem to be hard to derive. A different indirect slow–fast assumption proposed by Segel [29], Segel and Slemrod [30] starts from the observation that in singular perturbation scenarios there is a fast initial phase (a "boundary layer in time"; see Verhulst [37]), followed by a slower time regime. From time scale estimates for the initial and the later phase, and the requirement that their ratio should be very small, Segel and Slemrod [30] obtain conditions on the parameters in the Michaelis–Menten system. The approach by Schauer and Heinrich [26] may also be justified by singular perturbation arguments, but the line of reasoning is concerned not with time scales but rather with the presumed slow manifold, and derives parameter conditions from requiring closeness of a solution trajectory to this manifold.

Generally, all these approaches are (at least partly) of heuristic nature, and validity of QSS will have to be checked a posteriori. A potentially erroneous conclusion from the time scale comparisons in Segel and Slemrod [30] for so-called reverse QSS (QSS for substrate) is discussed in [10, Section 4]. Moreover it is easy to construct examples which satisfy the condition proposed by Schauer and Heinrich [26] but do not satisfy any initial phase requirement (as stated in Atkins and de Paula [2]; see quote in Sect. 8.2): consider systems with a first integral. We emphasize that, while QSS hypotheses frequently lead to singularly perturbed systems (with the benefit of a solid reduction theory), this does not seem to be the case in every relevant scenario. Again, much depends on the exact notion of QSS that is used.

8.4.2 The Role of Scaling

In the context of this chapter, a scaling transformation for an ordinary differential equation consists of multiplying the independent variable (time) and each dependent variable by positive numbers. In most mathematically oriented texts and research papers (see in particular Murray [22], Segel and Slemrod [30], Heineken et al. [12]) scaling is used, and frequently employed to find "small parameters." While there is no doubt that scaling is highly relevant for an appropriate analysis of differential equations modeling a real-life situation, in particular for concrete estimates, there may be some danger in the "lumping together" of several model parameters into one "small parameter" for asymptotic arguments.

We will briefly discuss the necessity, benefits, and limitations of scaling for irreversible Michaelis–Menten and the "small parameter" $\varepsilon^* = \frac{e_0}{s_0}$ from Heineken et al. [12]. (For the Segel–Slemrod "small parameter" $\varepsilon = \frac{k_1 e_0}{s_0 + k_{-1} + k_2}$—see [30]—similar remarks apply.) Note that ε^* tends to zero when $e_0 \to 0$, and this case has been resolved above in a satisfactory manner. But ε^* also tends to zero when $s_0 \to \infty$, and to properly analyze the latter case one should keep in mind that the relevant domain for the Michaelis–Menten system (8.1) is defined by the inequalities $0 \le c \le e_0$ and $0 \le s + c \le s_0$. Hence $s_0 \to \infty$ will blow up the region of interest. Since Tikhonov's theory applies to differential equations on fixed domains, scaling is necessary here. We scale (following Heineken et al. [12] in part, but not completely) by setting $\sigma = s/s_0$ and $\gamma = c/e_0$, and $\varepsilon = 1/s_0$, obtaining the system

$$\begin{aligned}\dot{\sigma} &= -k_1 e_0 \sigma(1-\gamma) + \varepsilon \cdot e_0 k_{-1}\gamma \\ \dot{\gamma} &= \varepsilon^{-1} k_1 \sigma\,(1-\gamma) - (k_{-1}+k_2)\gamma\end{aligned} \tag{8.15}$$

on the domain defined, e.g., by the inequalities $0 \le \sigma + e_0\gamma \le 1$, $0 \le \gamma \le 1$.

With the usual notation we have

$$h^{(0)} = \begin{pmatrix} 0 \\ k_1\sigma(1-\gamma) \end{pmatrix}, \quad h^{(1)} = \begin{pmatrix} -k_1 e_0 \sigma(1-\gamma) \\ -(k_{-1}+k_2)\gamma \end{pmatrix}.$$

The zero set M_0 of $h^{(0)}$ has two components; the one defined by $\gamma = 1$ corresponds to the standard QSS assumption. The conditions for Tikhonov–Fenichel are satisfied, and a straightforward computation shows that the reduced equation is given by

$$\dot{\sigma} = 0, \quad \gamma = 1.$$

In other words, Tikhonov–Fenichel applies but it yields a degenerate reduced system. Including higher-order terms in ε (thus passing to a O'Malley-Vasil'eva expansion, see Verhulst [37]) one formally obtains the familiar reduced equation. The approach in [12, Equations (10) to (13)] encounters the same problem in the case $s_0 \to \infty$, because some of the scaled parameters approach zero in this limiting case. Taking this into account, the lowest-order reduction in [12] will also be trivial.

The point we want to emphasize here is the necessity to consider all possible ways in which a "small parameter" may approach zero. This also may be of some practical relevance, since $e_0 \to 0$ ("very little enzyme") and $s_0 \to \infty$ ("very high substrate concentration") represent different experimental settings. These cases require individual consideration, with one case not amenable to standard singular perturbation methods. However, other lines of reasoning, such as the phase plane arguments in [23], show that a QSS assumption is indeed justified for this scenario.

Finally, we note that the other component of M_0 for Eq. (8.15) is given by $\sigma = 0$, which would correspond to the reverse QSS assumption (with s approaching its equilibrium 0 very fast). In this case the Tikhonov–Fenichel reduction formalism is not applicable, due to a nilpotent Jacobian. (One may question whether reverse QSS is chemically sensible for very high s_0.)

8.4.3 Near-Invariance Heuristics

In [24] a proposal was made to generalize Schauer and Heinrich's [26] heuristics from Michaelis–Menten to general systems. We will present the heuristics here in a somewhat informal manner, referring for details to [24]. Thus we start with a parameter-dependent differential equation

$$\dot{x} = q(x,\, p) \tag{8.16}$$

with $q : U \times V \to \mathbb{R}^n$, where $U \subset \mathbb{R}^n$ is a neighborhood of a compact set K^* and V is some subset of $\mathbb{R}^d$ $(d \geq 1)$.

(i) We assume that certain functions $\psi_1(x,p), \dots, \psi_r(x,p)$, defined for all $x \in K^*$, are in QSS (according to the Working Definition) for some relevant solution. Thus their rates of change, given by the Lie derivatives

$$\phi_i(x,\, p) := L_q(\psi_i)(x,\, p) = \langle \operatorname{grad} \psi_i(x,\, p),\, q(x,\, p) \rangle\,, \quad 1 \leq i \leq r,$$

are small along this solution. (All derivatives are to be understood with respect to x only.) In applications, the ψ_i are frequently coordinate functions.

(ii) For $p \in V$ let

$$K = K_p = \{ y \in K^* : \phi_1(y) = \dots = \phi_r(y) = 0 \}\,.$$

Then, due to continuity and compactness arguments, the maximum of the terms $|\phi_1(x)|, \dots, |\phi_r(x)|$ tends to zero if and only if $\operatorname{dist}(x,\, K)$ tends to zero. Thus requiring QSS with higher and higher accuracy, one obtains invariance of the set K for the differential equation in the limiting case. This is one motivation for the following definition.

(iii) *Near-invariance* (see [24]): Let K^*, $\phi_1, \dots, \phi_r$ and K be as above, let the ϕ_i be sufficiently differentiable, and assume that the rank of $(D\phi_1, \dots, D\phi_r)$ on K is equal to r. Given $0 \leq \delta \leq 1$ we say that K is δ-nearly invariant for $\dot{x} = q(x,\, p)$ with respect to $\phi_1, \dots, \phi_r$ if for all $x \in K$ and $1 \leq j \leq r$ one has the inequality

$$|\, \langle \operatorname{grad} \phi_j(x,\, p),\, q(x,\, p) \rangle \,| \leq \delta \cdot \| \operatorname{grad} \phi_j(x,\, p) \| \cdot \| q(x,\, p) \|.$$

The inequality always holds for $\delta = 1$, due to Cauchy–Schwarz, and for $\delta = 0$ the condition implies invariance of K. Thus one may expect solutions to stay close to K when $\delta \ll 1$.

It should be emphasized that this is another heuristic approach, replacing slow–fast heuristics by "invariant set-heuristics." Also, the notion does not only depend on the set K but also on the defining functions.

(iv) Some properties of near-invariance.

- Locally the desired property from (ii) is a consequence of near-invariance (see [24]): Let K be δ-nearly invariant. Then locally in time $(|t| \leq \rho)$, solutions starting on K remain $(C \cdot \delta)$-close to K, with C and ρ independent of the starting point and of δ.
- In the limiting case $\delta \to 0$ one obtains an invariant set. Since one has a parameter-dependent system, and K may change with parameters, one has to take care that no degeneracies occur, as in scaling procedures, and one should avoid blowing up K^*.
- The notion is compatible (up to an error of order ε) with Tikhonov–Fenichel: While the asymptotic slow manifold M_0 in the singular

perturbation setting is not necessarily $(C \cdot \varepsilon)$-nearly invariant, by an order ε correction in the defining equations one will obtain order ε near-invariance (see [24]).

(v) Use in practice: Let a parameter-dependent system (8.16) be given on K^*. Let $\phi_1, \cdots, \phi_r$ define a desired or suspected nearly invariant set K. The near-invariance property cannot be expected to hold generally, but only for certain parameter combinations. Thus evaluation of the near-invariance condition in (iii) will produce (necessary) conditions for the parameters. Determine (or estimate)

$$\delta(p) := \max \left\{ \frac{|\langle (\mathrm{grad}\,\phi_j)(x,\,p),\, q(x,\,p)\rangle|}{\|\mathrm{grad}\,\phi_j(x,\,p)\| \cdot \|q(x,\,p)\|}; \quad x \in K, \quad 1 \le j \le r \right\}$$

as a function of the parameters. Requiring $\delta(p)$ to be small provides conditions on the parameter set p. Asymptotic conditions are obtained from the limiting case $\delta(p) \to 0$.
Again we emphasize that further analysis and verification is necessary.

Example 8.9. Reversible Michaelis–Menten.
Consider the reversible Michaelis–Menten reaction (8.1), which we restate as

$$\begin{aligned} \dot{s} &= - \ \phi(s,c) \ - k_2 c + k_{-2}(e_0 - c)(s_0 - s - c), \\ \dot{c} &= \quad \phi(s,c), \end{aligned} \tag{8.17}$$

(the right-hand side will be called q) with QSS for complex $\psi(s,\,c) = c$, and its Lie derivative

$$\phi(s,c) = k_1 e_0 s - (k_1 s + k_{-1} + k_2)c + k_{-2}(e_0 - c)(s_0 - s - c)$$

on the set $K^* \subseteq \mathbb{R}^2$ given by $0 \le c \le e_0^*$, $s \ge 0$, and $s + c \le s_0^*$. (Here e_0^* and s_0^* are upper bounds for the initial concentrations.)
This system was discussed in detail in [24], with attention to the range for which QSS is assumed to hold. (For instance, requiring QSS only when sufficient substrate is still present would amount to a different choice of K^*.) Here we focus on QSS for the whole course of the reaction (after some initial phase), and look at the asymptotic scenario. To obtain QSS conditions for $\psi = c$, evaluate

$$L_q(\phi)(s,\,c) = ((k_1 - k_{-2})(e_0 - c),\, *) \begin{pmatrix} -k_2 c + k_{-2}(e_0 - c)(s_0 - s - c) \\ 0 \end{pmatrix}$$

for $(s,\,c) \in K$ (taking into account $\phi = 0$). One has

$$\frac{|L_q(\phi)(s,\,c)|}{||q(s,\,c)||} = |k_1 - k_{-2}| \cdot (e_0 - c),$$

and a rough estimate yields

$$\frac{|L_q(\phi)(s,\, c)|}{||q(s,\, c)|| \cdot ||\mathrm{grad}\, \phi(s, c)||} \le \frac{|k_1 - k_{-2}| \cdot e_0}{k_{-1} + k_2} =: \delta^*$$

for all $(s,\, c) \in K$.

The particular case $k_1 = k_{-2}$ actually yields an invariant set (regardless of other parameters), as noted by Miller and Alberty [21]. For the irreversible case $k_{-2} = 0$, the expression for δ^* is equal to the one introduced by Seshadri and Fritzsch [32]; compare the discussion in [23].

It may be appropriate to clarify what has actually been gained. By design of the procedure, one is assured of an invariant set in the limiting case $e_0 \to 0$. This may be taken as a motivation for choosing the small parameter e_0 in the reversible Michaelis–Menten differential equation, which we did throughout this paper. One then verifies that the hypotheses for Tikhonov–Fenichel are satisfied, and one obtains a reduced system with a mathematically solid foundation. Finally (see [10]) one can check a posteriori that QSS does indeed hold for complex under the assumption of small e_0. Thus the circle closes.

Near-invariance heuristics like all the proposed heuristics leading from a QSS assumption (to a precisely stated QSS assumption) to finding small parameters should be seen as work in progress, but there seems to be more potential in this particular approach. One advantage is that the implementation of the procedure (see (v) above) is in principle straightforward.

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