Applying a Rigorous Quasi-Steady State Approximation Method for Proving the Absence of Oscillations in Models of Genetic Circuits

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Abstract. In this paper, we apply a rigorous quasi-steady state approximation method on a family of models describing a gene regulated by a polymer of its own protein. We study the absence of oscillations for this family of models and prove that Poincaré-Andronov-Hopf bifurcations arise if and only if the number of polymerizations is greater than 8. A result presented in a former paper at *Algebraic Biology 2007* is thereby generalized. The rigorous method is illustrated over the basic enzymatic reaction.

1 Introduction

In a former paper [1], we studied a simple family of models depending on an integer parameter n and featuring a negative feedback loop, one of the core ingredients for generating oscillations [2]. These abstract models are closely related to models studied by Goodwin and Griffith in the 60's [3–5]. Griffith considered a model of a gene regulated by a polymer formed of n copies of its own protein. We studied the same problem, but in a slightly more general case, where gene activation is not assumed to be fast. We eventually concluded with the absence of Poincaré-Andronov-Hopf bifurcation in our family of models for $n \leq 8$ and their existence for $n \geq 9$. The absence/presence of Poincaré-Andronov-Hopf bifurcation for $n \leq 8$ is a strong indicator for the absence/presence of oscillating trajectories. Extensive numerical experiments [6, 7] confirmed the absence of oscillations for $n \leq 8$ and their existence for $n \geq 9$.

In this paper, the models are designed by means of systems of parametric nonlinear ordinary differential equations (ODE) [8]. The approach applied in [1] consisted in two steps: first simplifying the initial system of $n + 2$ parametric ordinary differential equations as a reduced system of three ODE by means of a quasi-steady state approximation; second, studying the reduced model.

The idea of quasi-steady state approximation is simple: study the dynamics of the slow reactions, assuming that the fast ones are at quasi-equilibrium,

thereby removing from the ODE system, the differential equations which describe the evolution of the variables at quasi-equilibrium. Many authors [9–12] state that carrying out rigorously this approximation is far from straightforward. We would rather say that there are different ways to perform this approximation and that it is the problem of ascertaining the domain of validity of each kind of approximation which is not straightforward.

In another paper [13], the authors reformulated the methods of [9–12], which are equivalent, and made them fully algorithmic, by means of differential elimination methods [14–18]. An efficient implementation, based on [19], was developed by the third author.

In this paper, we show that the reduction method of [13] can be applied to our family of models. It yields a reduced model which contains that of [1] as a particular case: our new approximation is more precise. By a very concise proof, we show that the results obtained in [1] also hold for the new model. This paper gives us also the opportunity to widen the audience of our [13, DifferentialModelReduction] algorithm by recalling its principle.

2 Our Family of Models

2.1 The Initial Model

A schematic model describing a single gene regulated by an order n polymer of its own protein is provided in Fig. 1. It is borrowed from [1, page 68]. The variables G and H represent the state of the gene. The mRNA concentration and the concentration of the protein translated from the mRNA are represented by M and P. The n types of polymers of P are denoted by $P = P_1, P_2, \ldots, P_n$. Greek letters represent parameters.

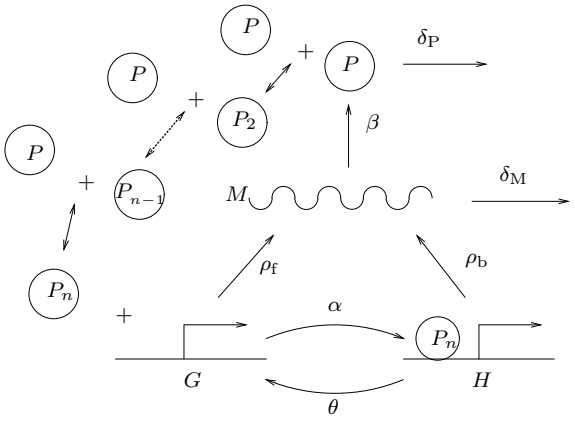

Fig. 1. A single gene regulated by a polymer of its protein

First, one translates the diagram as a system of generalized [20] chemical reactions (transcription and translation are not balanced reactions), introducing $2\,(n-1)$ extra parameters k_i^-, k_i^+ $(1 \leq i \leq n-1)$:

$$G + P_n \underset{\theta}{\overset{\alpha}{\rightleftharpoons}} H, \quad G \xrightarrow{\rho_\mathrm{f}} G + M, \quad H \xrightarrow{\rho_\mathrm{b}} H + M,$$
$$M \xrightarrow{\beta} M + P, \quad M \xrightarrow{\delta_\mathrm{M}} \emptyset, \quad P \xrightarrow{\delta_\mathrm{P}} \emptyset, \qquad P_i + P \underset{k_i^-}{\overset{k_i^+}{\rightleftharpoons}} P_{i+1} \quad (1 \leq i \leq n-1).$$

The initial model is obtained by translating the above chemical reactions system as the following system of parametric ordinary differential equations, denoting $A_i = (k_i^-\, P_{i+1} - k_i^+\, P_i\, P)$. Variables G, H, M, $P = P_1, \ldots, P_n$ are time varying functions. The dot appearing over the variables, in the equations left handsides, denotes the derivative w.r.t. the time.

$$\begin{aligned}
\dot{G} &= \theta\, H - \alpha\, G\, P_n, \\
\dot{H} &= -\theta\, H + \alpha\, G\, P_n, \\
\dot{M} &= \rho_\mathrm{f}\, G + \rho_\mathrm{b}\, H - \delta_\mathrm{M}\, M, \\
\dot{P} &= \beta\, M - \delta_\mathrm{P}\, P + 2\, A_1 + A_2 + \cdots + A_{n-1}, \\
\dot{P}_i &= -A_{i-1} + A_i \qquad (2 \leq i \leq n-1), \\
\dot{P}_n &= -A_{n-1} + \theta\, H - \alpha\, G\, P_n.
\end{aligned} \tag{1}$$

The variables G and H should be viewed as "random variables" instead of concentrations. They are bound by the relation $G + H = \gamma_0$ where γ_0 is a constant equal to the total quantity of the gene. In [1], the variable H was replaced by $\gamma_0 - G$.

2.2 The New Quasi-Steady State Approximation

The quasi-steady state approximation that is performed in this paper relies on the following hypotheses: the $n-1$ chemical reactions describing the polymerization of the protein are fast while the other ones are slow. This happens when the parameters k_i^+, k_i^- are much larger than the other parameters.

Given those hypotheses, one applies the algorithm [13, DifferentialModelReduction] on our model (Fig. 1). The principle of the method is recalled in Sect. 4.1. Applied on our model, the algorithm amounts to the following steps. First one replaces, in the initial model, the contributions of the fast reactions by new variables F_i $(1 \leq i \leq n-1)$. This just amounts to rewriting $A_i = F_i$ in system (1). Then one adds the following algebraic equations to the system, in order to express the pre-equilibrium conditions:

$$0 = k_i^+\, P\, P_i - k_i^-\, P_{i+1} \qquad (1 \leq i \leq n-1).$$

Then one eliminates the new variables F_i from the above differential-algebraic system.[3] One is led to the raw reduced model:

$$\begin{aligned}
\dot{G} &= \theta\, H - \alpha\, K_{n-1}\, P^n\, G,\\
\dot{H} &= -\theta\, H + \alpha\, K_{n-1}\, P^n\, G,\\
\dot{M} &= \rho_{\mathrm{b}}\, H + \rho_{\mathrm{f}}\, G - \delta_{\mathrm{M}}\, M,\\
\dot{P} &= \frac{n\,\theta\, H - n\,\alpha\, K_{n-1}\, P^n\, G - \delta_{\mathrm{P}}\, P + \beta\, M}{\displaystyle\sum_{i=0}^{n-1} (i+1)^2\, K_i\, P^i}
\end{aligned} \tag{2}$$

where $K_i = \dfrac{k_1^+ \cdots k_i^+}{k_1^- \cdots k_i^-}$ with the convention $K_0 = 1$.

2.3 Parameters Reduction

The raw reduced model (2) can now be simplified by rescaling all parameters and variables. The following equations express the old variables as functions of the new ones, which are overlined:

$$\theta = \overline{\theta}\,\overline{\delta_{\mathrm{M}}}, \quad \beta = \overline{\beta}\,\overline{\delta_{\mathrm{M}}}, \quad \delta_{\mathrm{P}} = \overline{\delta}\,\overline{\delta_{\mathrm{M}}}, \quad \rho_{\mathrm{b}} = \frac{\overline{\mu}\,\overline{\delta}\,\overline{\theta}\,\overline{\delta_{\mathrm{M}}}}{\overline{\alpha}\,\overline{\beta}}, \quad \rho_{\mathrm{f}} = \frac{\overline{\delta}\,\overline{\theta}\,(\overline{\mu}+\overline{\lambda})\,\overline{\delta_{\mathrm{M}}}}{\overline{\alpha}\,\overline{\beta}},$$

$$\delta_{\mathrm{M}} = \overline{\delta_{\mathrm{M}}}, \quad \alpha = \frac{\overline{\alpha}\,\overline{\delta_{\mathrm{M}}}}{\overline{K}_{n-1}}, \quad K_i = \frac{\overline{K}_i\,\overline{\alpha}^i}{\overline{\theta}^i} \quad (1 \le i \le n-1),$$

$$G = \overline{G}, \quad H = \overline{\gamma_0} - \overline{G}, \quad M = \frac{\overline{M}\,\overline{\delta}\,\overline{\theta}}{\overline{\alpha}\,\overline{\beta}}, \quad P = \frac{\overline{\theta}\,\overline{P}}{\overline{\alpha}}, \quad t = \frac{\overline{t}}{\overline{\delta_{\mathrm{M}}}}.$$

Performing these substitutions in the raw reduced model (2), discarding the redundant ODE which expresses the evolution of H and removing the bars for legibility, one gets the reduced model (3):

$$\begin{aligned}
\dot{G} &= \theta\,(\gamma_0 - G - G\,P^n),\\
\dot{M} &= \lambda\, G + \gamma_0\,\mu - M,\\
\dot{P} &= \frac{n\,\alpha\,(\gamma_0 - G - G\,P^n) + \delta\,(M - P)}{\displaystyle\sum_{i=0}^{n-1} (i+1)^2\, K_i\, P^i}.
\end{aligned} \tag{3}$$

Remark 1. In order to recover [1, system (1)], it is sufficient to replace the right handside of the last equation of system (3) by its numerator.

[3] Our algorithm does not handle the generic system with a symbolic n. We computed the reduced system for many different values of n, inferred the general formula and, checked afterwards that the inferred formula is correct.

Last observe that one more parameter could be removed from the above system, the software [21] shows. The above reduction is however more convenient in this paper for it permits us to directly apply the results of [1].

3 On the Existence of Poincaré-Andronov-Hopf Bifurcations

We prove in this section that no Poincaré-Andronov-Hopf bifurcation [22] arises in system (3) for meaningful[4] values of the parameters and the variables if and only if $n \leq 8$. Our proof essentially amounts to reducing the study of system (3) to that of [1, system (1)] and then applying the main result of [1].

According to remark 1, the steady point equations of system (3) are exactly those of [1, Sect. 4]. They write:

$$\gamma_0 = G + G\,P^n, \quad M = P, \quad \lambda = \frac{P - \mu\,G - \mu\,G\,P^n}{G}\cdot$$

The Jacobian matrix of system (3), evaluated at the steady points[5] writes:

$$J = \begin{pmatrix} -\theta\,(1+P^n) & -n\,\theta\,G\,P^{n-1} & 0 \\ \dfrac{P - \mu\,G - \mu\,G\,P^n}{G} & 0 & -1 \\ -\dfrac{n\,\alpha\,(1+P^n)}{B} & -\dfrac{n^2\,\alpha\,G\,P^{n-1} + \delta}{B} & \dfrac{\delta}{B} \end{pmatrix}$$

where $B = \displaystyle\sum_{i=0}^{n-1} (i+1)^2\,K_i\,P^i$.

If one clears the denominators of the last row of the Jacobian matrix J, i.e. if one lets $B = 1$, then one exactly gets the Jacobian matrix of [1, page 73].

Remark 2. The matrix J is invariant under the following transformation, where ℓ denotes a nonzero arbitrary constant:

$$B \to \ell\,B, \quad \delta \to \ell\,\delta, \quad \alpha \to \ell\,\alpha.$$

Remark 3. The parameters K_i only occur in the denominator B of the Jacobian matrix J.

Proposition 1. *A Poincaré-Andronov-Hopf bifurcation arises for [1, system (1)] if and only if such a bifurcation arises for the system (3), for meaningful values of the systems variables and parameters.*

[4] Following [1], all variables and parameters are required to be positive apart λ, which may be negative but must anyway be greater than $-\mu$,

[5] Note that the derivative of B w.r.t. to P which appears in the Jacobian of the system (3) disappears after the evaluation at the steady points since it is multiplied by a term that cancels it.

Proof. Assume that a Poincaré-Andronov-Hopf bifurcation occurs for system (3), for some *meaningful*[6] values of the parameters and variables ($1 \leq i \leq n-1$):

$$(G,\, P,\, M,\, \lambda,\, \alpha,\, \theta,\, \delta,\, \gamma_0,\, \mu,\, K_i) = (G^0,\, P^0,\, M^0,\, \lambda^0,\, \alpha^0,\, \theta^0,\, \delta^0,\, \gamma_0^0,\, \mu^0,\, K_i^0).$$

Then, by introducing $B^0 = \sum_{i=0}^{n-1}(i+1)^2\, K_i^0\, (P^0)^i$, and according to the two remarks above, a bifurcation of [1, system (1)] occurs for the meaningful values:

$$(G,\, P,\, M,\, \lambda,\, \alpha,\, \theta,\, \delta,\, \gamma_0,\, \mu,) = (G^0,\, P^0,\, M^0,\, \lambda^0,\, \alpha^0/B^0,\, \theta^0,\, \delta^0/B^0,\, \gamma_0^0,\, \mu^0).$$

Conversely, suppose that a birfurcation of [1, system (1)] arises for some meaningful values

$$(G,\, P,\, M,\, \lambda,\, \alpha,\, \theta,\, \delta,\, \gamma_0,\, \mu) = (G^0,\, P^0,\, M^0,\, \lambda^0,\, \alpha^0,\, \theta^0,\, \delta^0,\, \gamma_0^0,\, \mu^0).$$

Using the two remarks above and taking $\ell = 2$, one can easily find some positive values for $K_1^0, \ldots, K_{n-1}^0$ such that $B_0 = \sum_{i=0}^{n-1}(i+1)^2\, K_i^0\, (P^0)^i = 2$. Thus a bifurcation for the system (3) occurs when

$$(G,\, P,\, M,\, \lambda,\, \alpha,\, \theta,\, \delta,\, \gamma_0,\, \mu,\, K_i) = (G^0,\, P^0,\, M^0,\, \lambda^0,\, 2\alpha^0,\, \theta,\, 2\delta^0,\, \gamma_0^0,\, \mu^0,\, K_i^0)$$

which are meaningful values. □

The next proposition follows from the results of [1] and Prop. 1:

Proposition 2. *For meaningful values of the parameters and the variables, no Poincaré-Andronov-Hopf bifurcation arises in system (3) if and only if* $n \leq 8$.

4 On the New Quasi-Steady State Approximation

4.1 Principle of the Method

Our method is based on similar ideas as in [11, 9, 12] (see [13, Sect. 4.2] for more details). The main interest of our method is that it is fully algorithmic. One first builds a system of differential equations involving some one extra variable F_i for each fast reaction. Those extra variables are then eliminated by performing elimination. Our original implementation is based on the DIFFALG [23] package available in the standard library of MAPLE. A more recent implementation is based on the RegularChains [19] package.

This section aims at summarizing [13, Sect. 2]. Consider the classical system of chemical reactions (4) and (5) describing the transformation of a substrate S into a product P under the action of the enzyme E (an intermediate complex C is produced):

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} C \tag{4}$$

$$C \xrightarrow{k_2} E + P \tag{5}$$

[6] In the sense precised above.

Assume that the reaction (4) is fast. Our method consists in introducing the following system:

$$\left[\dot{E} = -F_1 + k_2\,C, \quad \dot{S} = -F_1, \quad \dot{C} = F_1 - k_2\,C, \quad \dot{P} = k_2\,C, \quad k_1\,E\,S = k_{-1}\,C\right].$$

In this system, the new variable F_1 denotes the rate of reaction (4). This variable introduces one degree of freedom. However, this freedom is contrained by the the quasi-steady state equilibrium of reaction (4) which is $k_1\,E\,S = k_{-1}\,C$.

The value of F_1 can be computed by performing an elimination process which yield:

$$\left[F_1 = \frac{k_2\,E\,S\,(S+K)}{K\,(S+E+K)}, \quad \dot{E} = \frac{k_2\,E^2\,S}{K\,(S+E+K)}, \quad \dot{P} = \frac{k_2\,E\,S}{K}, \quad \dot{S} = -\frac{k_2\,E\,S\,(S+K)}{K\,(S+E+K)}, \quad C = \frac{E\,S}{K}\right]$$

where $K = k_{-1}/k_1$.

Using the conservation laws $E + C = E_0 + C_0$ and $S + C + P = S_0 + C_0 + P_0$ (where the subscript 0 indicates the initial concentration), assuming that $C_0 = P_0 = 0$ and introducing $V_m = k_2\,E_0$, further computations yield:

$$\dot{S} = -\frac{V_m\,S\,(K+S)}{K\,E_0 + (K+S)^2} \tag{6}$$

which differs from the Henri-Michaëlis-Menten and Briggs-Haldane formulae[7]:

$$\dot{S}(t) = -\frac{V_m\,S(t)}{K + S(t)}. \tag{7}$$

With this easy example, the benefits are clear since the reduction is automatic and yields the formula (6) which seems more accurate than the classical reductions, especially when the condition $S \gg E_0$ is not fulfilled. Observe that formula (7) is recovered from (6) by assuming $S \gg E_0$.

4.2 A Better New Reduced Model

The reduced system (2) appears to be more precise than that of [1], as numerical simulations show. Figure 2 shows two different numerical simulations of the variable $G(t)$ for the same parameters and initial conditions values except for the k_i^-'s (hence for the K_i's). In both cases, three curves are displayed: one for the initial model (1) in solid line, one for the raw reduced model (2) in diamond dots and one for [1, last system page 69] in dotted line. In both cases, diamond dots are almost superimposed on the solid line.

The reduction performed in [1] is clearly less precise than the new one. As the K_i's parameters tend towards zero, the reduction performed in [1] becomes more accurate (the rightmost picture is obtained with values of K_i's smaller than that of the leftmost one). Thus the domain of validity of the reduction performed in [1, last system page 69] is narrower than that of our new reduction: the K_i's need to be small.

[7] $K = \frac{k_{-1}}{k_1}$ in Henri-Michaëlis-Menten's case, $K = \frac{k_{-1}+k_2}{k_1}$ in Briggs-Haldane's.

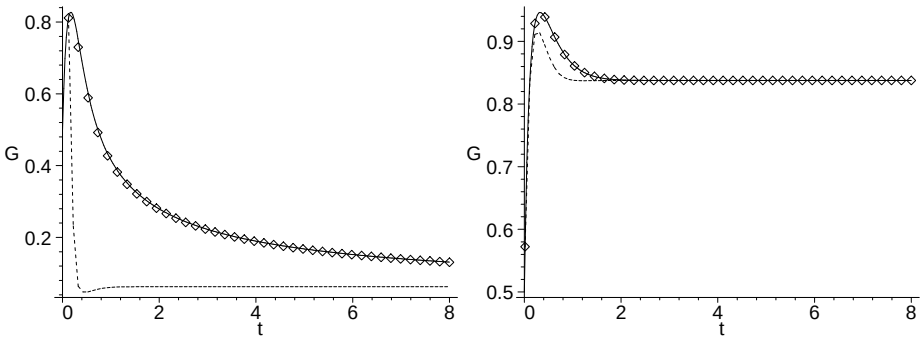

Fig. 2. Numerical simulations of the variable $G(t)$. The horizontal axis give the time t. The vertical one gives the value of G. For both simulations, $n = 3$, $G(0) = 0.5$, $M(0) = 1$, $P(0) = 1$, $\gamma_0 = 1$, $\alpha = 1$, $\theta = 10$, $\rho_{\mathrm{f}} = 10$, $\rho_{\mathrm{b}} = 5$, $\beta = 50$, $\delta_{\mathrm{M}} = 5$, $\delta_{\mathrm{P}} = 10$ and $k_i^+ = 100$ for each i. On the leftmost picture, $k_i^- = 100$ thus $K_i = (100/100)^i = 1$ for each i. On the rightmost one, $k_i^- = 2000$ thus $K_i = (100/2000)^i = 0.05^i$ for each i. The solid line (original model (1)) and the diamond dots (reduced model (2)) curves almost coincide. The dotted line [1, last system page 69] curve is the lower one on both pictures.

5 Conclusion

In this paper, the result presented in [1] is generalized by applying an algorithmic, accurate, quasi-steady state approximation method.

It is well-known that quasi-steady state approximation is useful for it permits to reduce the size of the differential system to study and the number of its parameters. But quasi-steady state approximation could also be viewed as a way to study the dynamical properties of gene regulatory networks which are invariant for a range of reactions mechanisms since the mechanisms involved in the fast reactions may not need to be known in detail. This issue is important [24]. This shows that the development of algorithmic and accurate quasi-steady state approximation methods is an important research domain in the field of algebraic biology.

References

1. Boulier, F., Lefranc, M., Lemaire, F., Morant, P.E., Ürgüplü, A.: On proving the absence of oscillations in models of genetic circuits. In Anai, H., Horimoto, K., Kutsia, T., eds.: Proceedings of Algebraic Biology 2007. Volume 4545 of LNCS., Springer Verlag Berlin Heidelberg (2007) 66–80 `http://hal.archives-ouvertes.fr/hal-00139667`.
2. Fall, C.P., Marland, E.S., Wagner, J.M., Tyson, J.J.: Computational Cell Biology. Volume 20 of Interdisciplinary Applied Mathematics. Springer Verlag (2002)
3. Goodwin, B.C.: Temporal Organization in Cells. Academic Press, London (1963)
4. Goodwin, B.C. In: Advances in Enzyme Regulation. Volume 3., Oxford, Pergamon Press (1965) 425

5. Griffith, J.S.: Mathematics of Cellular Control Processes. I. Negative Feedback to One Gene. Journal of Theoretical Biology **20** (1968) 202–208
6. Doedel, E.: AUTO software for continuation and bifurcation problems in ODEs. `http://indy.cs.concordia.ca/auto` (1996)
7. Ermentrout, B.: Simulating, Analyzing, and Animating Dynamical Systems: A Guide to XPPAUT for Researchers and Students. Volume 14 of Software, Environments, and Tools. SIAM (2002)
8. Conrad, E.D., Tyson, J.J.: Modeling Molecular Interaction Networks with Nonlinear Differential Equations. In Szallasi, Z., Stelling, J., Periwal, V., eds.: System Modeling in Cell Biology: From Concepts to Nuts and Bolts, Cambridge, Massachussets: The MIT Press (2006) 97–124
9. Van Breusegem, V., Bastin, G.: Reduced order dynamical modelling of reaction systems: a singular perturbation approach. In: Proceedings of the 30th IEEE Conference on Decision and Control, Brighton, England (December 1991) 1049–1054
10. Okino, M.S., Mavrovouniotis, M.L.: Simplification of Mathematical Models of Chemical Reaction Systems. Chemical Reviews **98**(2) (1998) 391–408
11. Vora, N., Daoutidis, P.: Nonlinear model reduction of chemical reaction systems. AIChE Journal **47**(10) (2001) 2320–2332
12. Bennet, M.R., Volfson, D., Tsimring, L., Hasty, J.: Transient Dynamics of Genetic Regulatory Networks. Biophysical Journal **92** (May 2007) 3501–3512
13. Boulier, F., Lefranc, M., Lemaire, F., Morant, P.E.: Model Reduction of Chemical Reaction Systems using Elimination (2007) Presented at the international conference MACIS 2007, `http://hal.archives-ouvertes.fr/hal-00184558`.
14. Ritt, J.F.: Differential Algebra. Dover Publications Inc., New York (1950) `http://www.ams.org/online_bks/coll33`.
15. Kolchin, E.R.: Differential Algebra and Algebraic Groups. Academic Press, New York (1973)
16. Boulier, F., Lazard, D., Ollivier, F., Petitot, M.: Representation for the radical of a finitely generated differential ideal. In: ISSAC'95: Proceedings of the 1995 international symposium on Symbolic and algebraic computation, New York, NY, USA, ACM Press (1995) 158–166 `http://hal.archives-ouvertes.fr/hal-00138020`.
17. Wang, D.: Elimination Practice: Software Tools and Applications. Imperial College Press, London (2003)
18. Boulier, F.: Differential Elimination and Biological Modelling. Radon Series on Computational and Applied Mathematics (Gröbner Bases in Symbolic Analysis) **2** (October 2007) 111–139 `http://hal.archives-ouvertes.fr/hal-00139364`.
19. Lemaire, F., Moreno Maza, M., Xie, Y.: The RegularChains library in MAPLE 10. In Kotsireas, I.S., ed.: The MAPLE conference. (2005) 355–368
20. F. Horn, R.J.: General mass action kinetics. Archive for Rational Mechanics and Analysis **47** (1972) 81–116
21. Sedoglavic, A., Ürgüplü, A.: Expanded Lie Point Symmetry (MAPLE package). `http://www.lifl.fr/~sedoglav/Software` (2007)
22. Hale, J.K., Koçak, H.: Dynamics and Bifurcations. Volume 3 of Texts in Applied Mathematics. Springer–Verlag, New York (1991)
23. Boulier, F., Lazard, D., Ollivier, F., Petitot, M.: Computing representations for radicals of finitely generated differential ideals. Technical report, Université Lille I, LIFL, 59655, Villeneuve d'Ascq, France (1997) Ref. IT306. December 1998 version published in the HDR memoir of Michel Petitot. `http://hal.archives-ouvertes.fr/hal-00139061`.

24. de Jong, H., Ropers, D.: Qualitative Approaches to the Analysis of Genetic Regulatory Networks. In Szallasi, Z., Stelling, J., Periwal, V., eds.: System Modeling in Cell Biology: From Concepts to Nuts and Bolts, Cambridge, Massachussets: The MIT Press (2006) 125–147