On the Analysis of Robust Stability of Metabolic Pathways

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It is common to encounter proposed solutions to analysis problems posed in other fields that ignore the methodologies, results, and tools published in the control literature. Below is an example where a result adopted from a robust control paper shows that a proposed approach for analyzing a metabolic network can produce misleading results.

Many theoretical papers consider the stability analysis of biological networks (e.g., [1], [2], and the citations therein), including in the presence of parametric uncertainties [3]–[6]. One analysis problem is to assess whether a metabolic network described by the ordinary differential equations

$$\frac{d}{dt}x = f(x, y) \tag{1}$$

is locally asymptotically stable to specified variations in metabolic concentrations x and enzyme activities y [6], where f is a vector whose elements are polynomial functions of the elements of the vectors x and y. (Robust nonlinear control theorists would rightfully argue against the value of such a definition of robust stability, but such a discussion will not be given here due to space constraints; perhaps such a discussion will appear in a future education column.) The variations in x and y are specified in terms of polynomial equality constraints and box constraints. From an undergraduate control textbook, a sufficient condition for local asymptotic stability is that all of the eigenvalues of the Jacobian

$$A = \left.\frac{\partial f}{\partial x}\right|_{(x,y)=(x_{ss},y_{ss})} \tag{2}$$

have negative real parts for all steady-state values of (x_{ss}, y_{ss}) that satisfy the constraints on x and y, where

$$0 = f(x_{ss}, y_{ss}). \tag{3}$$

Since the elements of f are polynomial functions, all elements of the matrix A are polynomial functions of the x_i and y_j, and the coefficients of the characteristic polynomial

$$p(s) \equiv \det(sI - A) = 0, \tag{4}$$

are also polynomial functions of the x_i and y_j. This approach transforms the robust local asymptotic stability analysis problem into the determination of whether the roots of a characteristic equation (4) are in the open left-half plane for specified variations in (x_{ss}, y_{ss}). An approach [6] proposed for analyzing this latter problem is to first compute tight lower and upper bounds on each coefficient of $p(s)$ and then apply a well-known analytical test (the Kharitonov theorem [7]) to assess whether the characteristic polynomial with independent variations in its coefficients is robustly stable. Below is an example that shows that this proposed approach for the robust stability analysis of metabolic networks can be highly misleading. This example is followed by a discussion of its construction, and some broader comments on such constructions.

A METABOLIC NETWORK EXAMPLE

Consider a metabolic network described by

$$\frac{dx_1}{dt} = x_2 - y_2 \tag{5}$$

$$\frac{dx_2}{dt} = x_3 - y_3 \tag{6}$$

$$\frac{dx_3}{dt} = x_4 - y_4 \tag{7}$$

$$\frac{dx_4}{dt} = -y_1x_1 - x_2 - 2y_1x_3 - x_4 + y_5 \tag{8}$$

where the x_i are metabolite concentrations and the y_j are enzyme activities. Each of (5)–(8) represents a conservation equation for a metabolite. For example, (5) indicates that the accumulation of metabolite 1 (x_1) is increased by the concentration of metabolite 2 (x_2) and decreased by enzyme 2 (y_2). Similar competing effects are seen in all four conservation equations (5)–(8). At steady state, the variables must satisfy

$$x_{2,ss} - y_{2,ss} = 0 \tag{9}$$

$$x_{3,ss} - y_{3,ss} = 0 \tag{10}$$

$$x_{4,ss} - y_{4,ss} = 0 \tag{11}$$

$$-y_{1,ss}x_{1,ss} - x_{2,ss} - 2y_{1,ss}x_{3,ss} - x_{4,ss} + y_{5,ss} = 0. \tag{12}$$

Assume that physiological considerations require that the metabolic concentrations and enzyme activities stay within the region

Digital Object Identifier 10.1109/MCS.2012.2196323
Date of publication: 12 July 2012

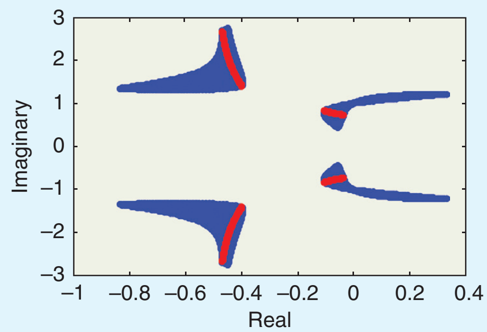

FIGURE 1 Eigenvalues for the characteristic equation (in red) and its interval polynomial (in blue) for $y_{1,ss} \in [1.5, 4]$. The plot for the characteristic equation is also the root locus of $(2s^2 + 1)/(s^4 + s^3 + s)$.

$$S = \begin{Bmatrix} 0.1 \le x_{1,ss} \le 1,\ 0.1 \le x_{2,ss} \le 1,\ 0.1 \le x_{3,ss} \le 1,\ 2 \le x_{4,ss} \le 4.15, \\ 0.1 \le y_{2,ss} \le 1,\ 0.1 \le y_{3,ss} \le 1, 2 \le y_{4,ss} \le 4.15, \\ 1.5 \le y_{1,ss} \le 4, \\ 2.5 \le y_{5,ss} \le 22 \end{Bmatrix}. \quad (13)$$

For this metabolic network, the local asymptotic robust stability condition is that all of the eigenvalues of the Jacobian of the right-hand side of (5)–(8),

$$A = \begin{pmatrix} 0 & 1 & 0 & 0 \\ 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 1 \\ -y_{1,ss} & -1 & -2y_{1,ss} & -1 \end{pmatrix}, \quad (14)$$

have negative real parts for all $\{x_{i,ss}, y_{j,ss}\} \in S$ that satisfy (9)–(12). These eigenvalues, which are the roots of the characteristic polynomial

$$p(s) = s^4 + s^3 + 2y_{1,ss}s^2 + s + y_{1,ss} = 0, \quad (15)$$

have negative real parts for all $y_{1,ss}$ satisfying the constraint (13), as seen in the root locus in Figure 1. In contrast, the corresponding interval polynomial [the additional constraints (9)–(13) do not restrict the range of $y_{1,ss}$]

$$\tilde{p}(\lambda) = s^4 + s^3 + [3, 8]s^2 + s + [1.5, 4], \quad (16)$$

includes eigenvalues with positive real parts (see Figure 1). The interval polynomial (16) is not stable whereas the characteristic polynomial (15) is stable for the full operating range of $\{x_{i,ss}, y_{j,ss}\} \in S$.

While the bounds on the coefficients for the interval polynomial (16) are the tightest possible, the discrepancy arises because the correlation between two coefficients of the characteristic polynomial (15) is not taken into account when constructing the interval polynomial (16). Figure 1 shows that the discrepancy in analyzing the linearized stability of metabolic networks using the interval polynomial (16) can be large.

TABLE 1 Matlab code for producing Figure 1.

```
clear all
y1 = linspace(1.5,4,100);
y2 = linspace(3,8,100);

for i = 1:100
    for j = 1:100
        c = roots([1 1 y2(i) 1 y1(j)]);
        plot(c,'.')
        hold on
    end
end

for i = 1:100
    c = roots([1 1 2*y1(i) 1 y1(i)]);
    plot(c,'r.')
end
xlabel('Real');
ylabel('Imaginary');
```

More generally, the set of variables $\{x_{i,ss}, y_{j,ss}\}$ satisfying the characteristic polynomial subject to polynomial and box constraints S is usually a strict subset of the interval polynomial. If the interval polynomial is robustly stable (that is, has all roots with negative real part), then the corresponding characteristic polynomial is also robustly stable. If the interval polynomial is unstable then the characteristic polynomial may or may not be unstable. A large discrepancy can arise when using an interval polynomial to determine the stability of a system in which the coefficients of the characteristic polynomial are correlated (see [8] for a further discussion of this issue).

HOW WAS THE EXAMPLE CONSTRUCTED?

The above example took advantage of the rather large literature on robust stability analysis from the 1980s to 1990s. The search for a metabolic network example started with trying to locate a characteristic polynomial whose coefficients were simple polynomial functions of parameters while having a corresponding interval polynomial with very different robust stability characteristics. An ISI Web of Knowledge search of "stability" and "uncertain polynomials" located a paper by Wei and Yedavalli [9] whose Example 4.2 is (15)–(16). To quickly confirm that the characteristic equation (15) was robustly stable, (15) was rearranged to

$$\begin{aligned} p(s) &= s^4 + s^3 + 2y_{1,ss}s^2 + s + y_{1,ss} = 0 \\ &\Rightarrow (s^4 + s^3 + s) + y_{1,ss}(2s^2 + 1) = 0 \\ &\Rightarrow 1 + y_{1,ss}\frac{2s^2 + 1}{s^4 + s^3 + s} = 0, \end{aligned}$$

which showed that its robust stability could be quickly assessed by plotting the root locus of $(2s^2+1)/(s^4+s^3+s)$ for a gain ranging from 1.5 to 4. Then Figure 1 was plotted by calculating the roots of (15) and the interval polynomial (16) for a fine grid of its two uncertain coefficients (see Table 1). The matrix A in (14) is the controller-form realization of (15) that is described in any introductory state-space control textbook.

Equations (5)–(8) were then constructed to have the Jacobian (14). The allowable range for $y_{1,ss}$ was specified by [10, Ex. 4.2], all of the $\{x_{i,ss}, y_{j,ss}\}$ in (13) except for $y_{5,ss}$ were selected somewhat arbitrarily while having upper and lower bounds satisfying (9)–(11). The upper and lower bounds on $y_{5,ss}$ were then selected so that (12) would be satisfied within the other bounds in (13) without further constraining the value of $y_{1,ss}$, that is,

$$\begin{aligned} y_{5,ss} &= y_{1,ss}x_{1,ss} + x_{2,ss} + 2y_{1,ss}x_{3,ss} + x_{4,ss} \\ &\le 4(1) + 1 + 2(4)(1) + 4.15 < 22 \\ y_{5,ss} &= y_{1,ss}x_{1,ss} + x_{2,ss} + 2y_{1,ss}x_{3,ss} + x_{4,ss} \\ &\ge 1.5(0.1) + 0.1 + 2(1.5)(0.1) + 2 > 2.5 \end{aligned} \tag{17}$$

FINAL COMMENTS

This example illustrates the value of searching the control literature when encountering analysis problems posed in other fields (as well as in the control field!). After trying really simple examples [10], a good approach is to skim control books and highly cited papers on the most closely related control topic. In the above example, once the metabolic analysis problem in [6] was recognized for being a robust stability problem, [8]–[9] were located by searching "robust stability" and related terms using Google Scholar. Then an example from [9] that demonstrated the importance of taking into account parameter correlations was reformulated to be in the form of a metabolic analysis problem. As an example in another application area, robust control tools have been applied to solve an analysis problem posed in pharmaceutical manufacturing [11].

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MEMBER ACTIVITIES (continued from page 16)

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1) To ensure full consideration and timely approval of the application, we recommend that the Chapter chair contact the vice president of Member Activities at least a few months in advance of the proposed event.
2) Each application should contain the following info:
   - Chapter name
   - title of the proposed technical activity, date, and location
   - number of people involved in the activity (estimated)
   - number of IEEE members involved in the activity (estimated)
   - brief budget
   - proposed arrangements to promote IEEE/CSS membership
   - additional benefit made possible by LASS support if granted.
3) It is very important that CSS membership is promoted at the event (at the very least by brochures and a brief advertisement during the event), and we seek the organizers' cooperation to ensure this.
4) It will be highly valuable if the application can provide clear indications of the additional benefits made possible by LASS. In particular, we seek to know the extent to which LASS support can make a significant difference to the quality of the event, by covering expenses where the existing budget is unable to do so.
5) For successful recipients of LASS support, a brief report should be submitted after the event is completed.
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I hope that this information has opened up new possibilities for improving member activities. I am really interested in discussions on new ideas to enrich the activities of your chapters and ways to provide the best resources and opportunities for our CSS members!

Please apply as early as possible as applications are processed and approved on a first-come, first-serve basis until the annual budget is fully utilized. Together, we can make serve our community better.

Shuzhi Sam Ge