A Mathematical Analysis of Rebound in a Target-Mediated Drug Disposition Model. II. With Feedback

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Abstract

We consider the possibility of free receptor (antigen/cytokine) levels rebounding to higher than the baseline level after the application of an antibody drug using a target-mediated drug disposition model. It is assumed that the receptor synthesis rate experiences homeostatic feedback from the receptor levels. It is shown for a very fast feedback response, that the occurrence of rebound is determined by the ratio of the elimination rates, in a very similar way as for no feedback. However, for a slow feedback response, there will always be rebound. This result is illustrated with an example involving the drug efalizumab for patients with psoriasis. It is shown that slow feedback can be a plausible explanation for the observed rebound in this example.

1 Introduction

In this paper, we continue our investigation into the phenomenon of receptor rebound, i.e., a post-dose rise in receptor levels to higher than pre-dose (baseline). In our previous paper [2], we showed that if no homeostatic feedback is present, rebound will occur if and only if the elimination rate of the target-drug complex is slower than both the elimination rate of the drug and the elimination rate of the target. Binding to cell-surface receptors typically results in accelerated turnover of the anti-body and hence complex, so it can be expected that rebound will occur in rare cases only. However, Ng *et al.* [15] describe a treatment for psoriasis patients with efalizumab in which rebound is observed. They also derive a model that shows good agreement with the experimental observations. A main difference between this model and standard TMDD models is that the model includes receptor feedback for the synthesis rate. In the basic TMDD model, reduction of free target levels does not have any impact on the endogenous production or elimination rate of the free target, i.e., no endogenous feedback control exists to compensate for the antibody effect on target. The model in [15] includes such feedback control via an additional differential equation for the synthesis rate. Another indicator of the importance of feedback in the synthesis rate is the recent paper by Kristensen *et al.* [11], in which it is postulated that the protein synthesis rate is the predominant regulator of protein expression during differentiation.

In this paper we investigate how receptor feedback influences the occurrence of rebound in the TMDD model. The receptor feedback is usually a dynamical process via some moderators and we modify the TMDD model to include feedback by adding an additional differential equation. If the feedback is very fast, a quasi-equilibrium approach can be used and the feedback can be included in the synthesis term itself. This approach is similar to the one leading to the Michaelis-Menten approximation or the Quasi-Steady-State approximation. We will use the term "direct feedback" for the quasi-equilibrium approximation. The modification to include feedback is presented in full detail

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in Section 3, after a review of the main results of our first paper [2] on rebound in the basic TMDD model without feedback in Section 2. A wide class of feedback functions is considered with some natural assumptions on their action.

The TMDD model with the direct feedback approximation is analysed in Section 4. This analysis shows that the existence or non-existence of rebound is still linked to the elimination rates, though rebound can be expected in a larger region in the elimination parameter plane, see the left plot in Fig. 1. The general case of feedback via a moderator is analysed in Section 5. Now the response speed of the feedback moderator plays an important role as well as the elimination rates. It is shown that there is a similar region in the elimination rate plane as for the basic TMDD model for which rebound will occur for any response speed. Furthermore, if the feedback responds slowly to a change in the receptor levels, rebound will occur for any value of the elimination rates. A schematic overview of this result is in the right plot of Fig. 1.
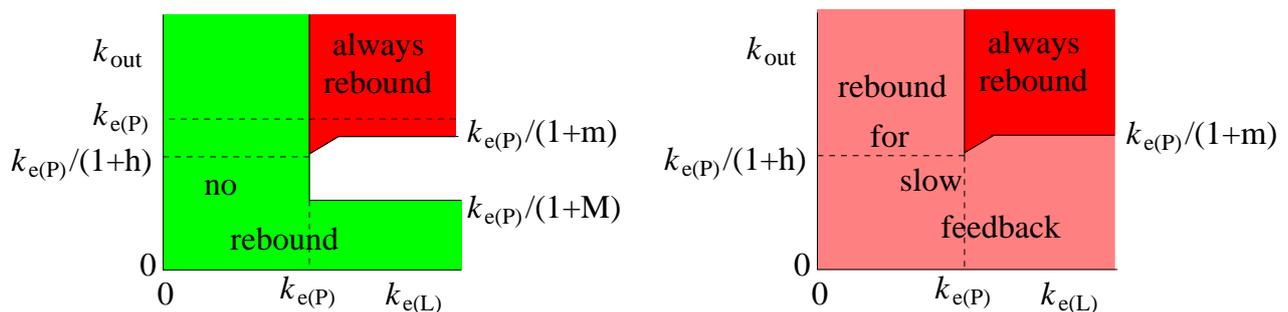

Figure 1: A schematic overview of the elimination plane. On the horizontal axis is the elimination rate of the ligand (antibody/drug), denoted by $k_{e(L)}$, and on the vertical axis is the elimination rate of the receptor (antigen/target), denoted by k_{out}. The elimination rate of the antibody-antigen complex (drug-target product) is denoted by $k_{e(P)}$. On the left is an overview of the occurrence of rebound in the case of the direct feedback approximation. In the red region, rebound will occur and in the green region no rebound can occur. The symbols h, m and M are related to the feedback function and will be defined in Section 4. In the case where there is no feedback, $m = h = M = 0$ and the region in which there is rebound is the red region above the line $k_{out} = k_{e(P)}$.

On the right is an overview of the occurrence of feedback, depending on the speed of the feedback response. There is a red region in the elimination plane, that equals the direct feedback rebound region, for which rebound will occur for any speed of the feedback response. In addition, if the response is slow, rebound will occur for any elimination values. The precise response speed for which the rebound stops occuring will vary across the pink region.

The TMDD model is a one-compartment model. In Section 6, we consider briefly two classes of more general models with feedback (one class is related to multiple compartments, the other to more general feedback mechanisms) and show that rebound will generically occur in such models for slow feedback moderator response. In this paper, the word generic refers to the assumptions that the linearisation about the baseline state has no degenerate eigenvalues and that trajectories approaching the attracting baseline state do so tangent to the eigenvector associated with the least attracting eigenvalue. Section 7 illustrates the theory for two examples. The first example is a standard TMDD model for the IgE mAb omalizumab. We discuss what the effect of the two types of feedback mechanisms would be in this case . The second example is the model in [15] which describes the efficacy of efalizumab for patients with psoriasis. Simulations show that the model does not lead to rebound if the feedback is turned off, but a significant rebound (about 140% of baseline) will occur if feedback is included. These results can be predicted with our analysis and underpin the observations of rebound in some patients. Finally, Section 8 contains a discussion of the results obtained in this paper and poses some open questions. The proofs of some of the technical results in the paper are
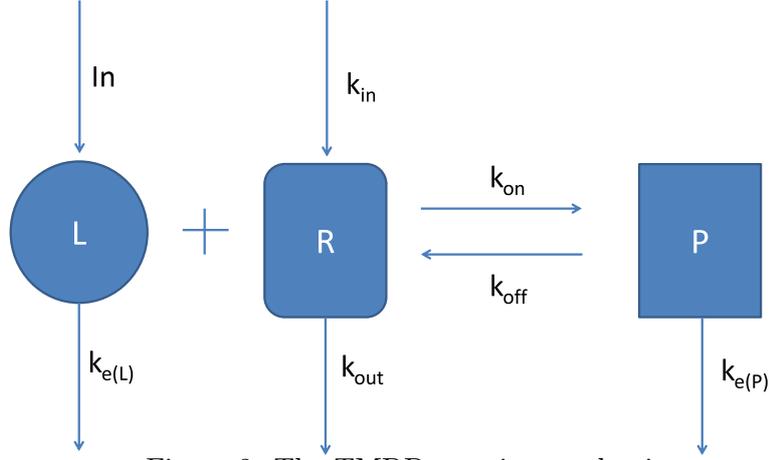
Figure 2: The TMDD reaction mechanism

given in the Appendix.

2 Review of Previous Results

In our previous paper [2], we considered a one-compartment TMDD model based on the original work of Levy [13] where the ligand L binds reversibly with the receptor R to form a receptor-ligand complex P as shown in Fig. 2. The TMDD model assumes a mechanism-based reaction to explain the ligand-receptor interaction. The parameters of the model are the binding rate constants k_{on} and k_{off}, the receptor turnover and elimination rates k_{in} and k_{out}, and the elimination rates of the ligand and complex $k_{e(L)}$ and $k_{e(P)}$. The system is assumed to be initially at steady state, into which a single bolus infusion L_0 of the ligand into the central (plasma) compartment is made (represented in Fig. 2 by 'In'). The differential equations that comprise the mathematical model for this system are given by

$$\frac{dL}{dt} = -k_{e(L)}L - k_{on}LR + k_{off}P \tag{1}$$

$$\frac{dR}{dt} = k_{in} - k_{out}R - k_{on}LR + k_{off}P \tag{2}$$

$$\frac{dP}{dt} = k_{on}LR - k_{off}P - k_{e(P)}P \tag{3}$$

A steady state of this system is given by $L = P = 0$, $R = k_{in}/k_{out}$. Adding the bolus injection L_0 gives the initial conditions

$$L(0) = L_0, \quad R(0) = R_0 = \frac{k_{in}}{k_{out}}, \quad P(0) = 0. \tag{4}$$

After the ligand is added to the system in its baseline state, initially the receptor level decreases, but after a while it goes up again and returns to its baseline value, since the steady state is globally asymptotically stable [2]. Rebound occurs if, in the return to the baseline value, the receptor level increases to values above the baseline R_0. In [2], we determined precise conditions for the existence and non-existence of rebound. Our main result was the following.

Theorem 2.1 *Rebound occurs in equations (1)–(3) if and only if*

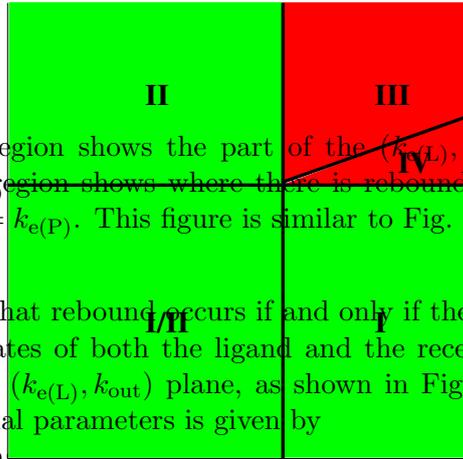$$k_{e(P)} < k_{e(L)} \quad and \quad k_{e(P)} < k_{out}.$$

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Figure 3: The green region shows the part of the $(k_{e(L)}, k_{out})$ parameter plane where there is no rebound, and the red region shows where there is rebound. The boundaries are given by the lines $k_{e(L)} = k_{e(P)}$ and $k_{out} = k_{e(P)}$. This figure is similar to Fig. 2 in [2].

This result shows that rebound occurs if and only if the elimination rate of the product is slower than the elimination rates of both the ligand and the receptor. This is represented graphically by different regions in the $(k_{e(L)}, k_{out})$ plane, as shown in Fig. 3. The above condition for rebound in terms of non-dimensional parameters is given by

$$k_4 < k_1 \quad \text{and} \quad k_4 < k_3,$$

where

$$k_1 = \frac{k_{e(L)}}{k_{on}R_0}, \quad k_3 = \frac{k_{out}}{k_{on}R_0}, \quad k_4 = \frac{k_{e(P)}}{k_{on}R_0}.$$

Intuitively, if the clearance of the complex is slow relative to the other two clearance rates, when there is a significant build-up of the complex due to the rapid binding of ligand and receptor, then this complex is only eliminated slowly. This results in an additional "production" of the receptor when the complex dissociates which is likely to lead to rebound. Our analysis quantifies this and we now show how these thresholds that we derived previously are affected if receptor feedback is added to the system.

3 Including Feedback in the TMDD Model

We now extend our previous work on rebound in the basic TMDD model by extending the model to include feedback on the synthesis rate. The feedback gives an increase in the production rate of the receptor if the receptor levels drop under the baseline. Similarly, if the receptor level exceeds the baseline value, then the feedback induces a decrease in the production rate. Thus, this mechanism can be classified as negative feedback.

We start with the TMDD equations (1)–(3) but now assume that the rate of production of R (k_{in}) is not constant but varies as R changes. Thus, equation (2) is now modified to have the form

$$\frac{dR}{dt} = k_{in}F - k_{out}R - k_{on}LR + k_{off}P. \tag{5}$$

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The new feedback variable F, which will be related to R, has the effect of varying the production rate of the receptor. The feedback is usually via some moderators and its dynamics is described with the differential equation

$$\frac{dF}{dt} = \alpha(H(R) - F), \quad F(0) = 1. \tag{6}$$

The constant $\alpha \geq 0$ determines the strength of the feedback and determines the time scale on which the feedback moderator responds to changes in the receptor levels. Clearly, if $\alpha = 0$, then F is a constant and the model reduces to the basic TMDD model with no feedback. If α is small, the feedback response is slow, while if α is larger, there is a very fast feedback response. Formally, dividing (6) through by α and taking the limit $\alpha \to \infty$, we see that the derivative term vanishes and the fixed relation

$$F = H(R) \tag{7}$$

is obtained. This motivates a quasi-equilibrium approximation in which the differential equation for F is replaced with the relation (7). We will call this the direct feedback approximation.

Next we consider the function $H(R)$. The function must be such that the model has the following features:

- the production rate of the receptor, denoted by $k_{\text{in}}F$, should be positive;

- the receptor concentration should have the baseline $R = R_0 = k_{\text{in}}/k_{\text{out}}$;

- the feedback mechanism should reduce the production rate of the receptor when the receptor level exceeds the baseline and should increase it when the receptor level is below the baseline.

This leads to the following general assumptions on the function $H(R)$:

(H1) $H(R) > 0$ for all $R \geq 0$;

(H2) $H(R_0) = 1$, where $R_0 = k_{\text{in}}/k_{\text{out}}$;

(H3) $H(R) > 1$ when $0 \leq R < R_0$ and $0 < H(R) < 1$ when $R > R_0$;

(H4) $H \in C^2([0, \infty))$.

We note that assumption (H1) ensures that in case of the direct feedback approximation (i.e, F is a function of R and (7) holds) the feedback $F > 0$, so that the production rate of R is always positive. Alternatively, if the dynamics of F is described by the differential equation (6), then this condition ensures that

$$\left.\frac{dF}{dt}\right|_{F=0} > 0$$

and so the line $F = 0$ cannot be crossed from above to below. This also ensures that $F > 0$ for all $t > 0$ and hence the production rate is always positive.

The basic TMMD model without feedback, equations (1)–(3), has a steady state solution given by $L = P = 0$, $R = R_0 = k_{\text{in}}/k_{\text{out}}$. This will still be a steady state of our modified equations in both the full model and the direct feedback approximation, provided that (H2) holds. For the full model, we observe that the steady state value for F is $F = 1$. Finally, the assumption (H3) ensures that the production of the receptor slows down when there is excess receptor, and increases when the receptor level is below the baseline and (H4) is a technical assumption assuring sufficient smoothness. Combining (H2)–(H4) implies that $H'(R_0) \leq 0$. We note that if H is a positive, strictly decreasing, smooth function with $H(R_0) = 1$, then (H1)–(H4) are satisfied. However, the stated conditions are more general and do not require H to be strictly decreasing.

We now have two modified TMDD models that incorporate feedback. For the direct feedback approximation, we use (7) to replace F with $H(R)$ which gives the differential equations

$$\frac{dL}{dt} = -k_{e(L)}L - k_{on}LR + k_{off}P \tag{8}$$

$$\frac{dP}{dt} = k_{on}LR - k_{off}P - k_{e(P)}P \tag{9}$$

$$\frac{dR}{dt} = k_{in}H(R) - k_{out}R - k_{on}LR + k_{off}P \tag{10}$$

Alternatively, the full feedback model involves an extra differential equation for the moderator F and the modified TMDD model is given by

$$\frac{dL}{dt} = -k_{e(L)}L - k_{on}LR + k_{off}P \tag{11}$$

$$\frac{dP}{dt} = k_{on}LR - k_{off}P - k_{e(P)}P \tag{12}$$

$$\frac{dR}{dt} = k_{in}F - k_{out}R - k_{on}LR + k_{off}P \tag{13}$$

$$\frac{dF}{dt} = \alpha(H(R) - F) \tag{14}$$

The initial conditions for both models are given by (4), with the extra condition $F(0) = 1$ for the feedback moderator.

We non-dimensionalise these equations in the same way as in [2]. We note that our new variable F is non-dimensional, and so we define the dimensionless variables

$$x = \frac{L}{L_0}, \quad y = \frac{R}{R_0}, \quad z = \frac{P}{R_0}, \quad w = F, \quad \tau = k_{on}R_0 t.$$

We also define a new function

$$h(y) = H(R_0 y).$$

The above assumptions on the function $H(R)$ translate into the following assumptions on $h(y)$:

(h1) $h(y) > 0$ for all $y \geq 0$;

(h2) $h(1) = 1$;

(h3) $h(y) > 1$ when $0 \leq y < 1$ and $0 < h(y) < 1$ when $y > 1$;

(h4) $h \in C^2([0, \infty))$.

For later use, we define

$$h_0 = -h'(1) \tag{15}$$

and note that the assumptions (h2)–(h4) imply that $h_0 \geq 0$.

In terms of the non-dimensional quantities, the direct feedback equations (8)–(10) become

$$\dot{x} = -k_1 x - xy + \mu k_2 z \tag{16}$$

$$\dot{z} = \frac{xy}{\mu} - (k_2 + k_4)z \tag{17}$$

$$\dot{y} = k_3(h(y) - y) - \frac{xy}{\mu} + k_2 z \tag{18}$$

with initial conditions

$$x(0) = 1, \quad z(0) = 0, \quad y(0) = 1, \tag{19}$$

where dot denotes differentiation with respect to τ and the dimensionless parameters are defined as in [2] by

$$\mu = \frac{R_0}{L_0}, \quad k_1 = \frac{k_{\mathrm{e(L)}}}{k_{\mathrm{on}} R_0}, \quad k_2 = \frac{k_{\mathrm{off}}}{k_{\mathrm{on}} R_0}, \quad k_3 = \frac{k_{\mathrm{in}}}{k_{\mathrm{on}} R_0^2} = \frac{k_{\mathrm{out}}}{k_{\mathrm{on}} R_0}, \quad k_4 = \frac{k_{\mathrm{e(P)}}}{k_{\mathrm{on}} R_0}.$$

Clearly, this choice of non-dimensionalisation requires that $k_{\mathrm{on}} \neq 0$ and $k_{\mathrm{in}} \neq 0$ (so that $R_0 \neq 0$). We note that all parameters must be non-negative due to their physical meaning, and additionally we will assume that they are in fact all strictly positive. The three variables x, y, and z are related to physical quantities and so must also be non-negative.

Similarly, the non-dimensional equations for the full feedback model (11)–(14) are given by

$$\dot{x} = -k_1 x - xy + \mu k_2 z \tag{20}$$

$$\dot{z} = \frac{xy}{\mu} - (k_2 + k_4) z \tag{21}$$

$$\dot{y} = k_3(w - y) - \frac{xy}{\mu} + k_2 z \tag{22}$$

$$\dot{w} = \epsilon(h(y) - w) \tag{23}$$

with initial conditions

$$x(0) = 1, \quad z(0) = 0, \quad y(0) = 1, \quad w(0) = 1. \tag{24}$$

Furthermore,

$$\epsilon = \frac{\alpha}{k_{\mathrm{on}} R_0} \tag{25}$$

is a non-dimensional measure for the response speed of the feedback moderator.

4 Analysis of the TMDD model with Direct Feedback

In this section, we will analyse the TMDD model with the direct feedback approximation, i.e., the equations (16)–(18), with initial conditions (19) and seek to determine conditions for the existence or non-existence of rebound in this model. However, we start by proving some basic results for the model equations.

4.1 Invariance, Steady States and Stability

For the basic TMDD model, we proved in [2] that the positive octant $x, y, z \geq 0$ is invariant; that there is a unique steady state in the positive octant; and that this steady state is a global attractor. In this section we show that the same properties hold for the TMDD model with direct feedback. The proofs of these results can be found in Appendix A. We start by stating invariance of the positive octant.

Lemma 4.1 *The octant of $\mathbb{R}^3$ defined by $x, y, z \geq 0$ is invariant under the flow of equations (16)–(18).*

This result shows that the equations are a good model in the sense that the concentrations of the ligand, complex and receptor can never go negative. Next we consider the steady states of equations (16)–(18).

Lemma 4.2 *In the region of $\mathbb{R}^3$ defined by $x, y, z \geq 0$, the equations (16)–(18) have a unique steady state given by*

$$x = 0, \quad y = 1, \quad z = 0. \tag{26}$$

This steady state is globally asymptotically stable.

This result shows that for all non-negative initial conditions and for all positive parameter values, each of the variables will converge to their unique steady state value.

It is also straightforward to show that the eigenvalues of the Jacobian of equations (16)–(18) evaluated at the steady state (26) are very similar to those given in [2]. In particular, the two eigenvalues λ_1 and λ_2 are the same as previously and are given by

$$\lambda_1 = \frac{1}{2}\left(-(1+k_1+k_2+k_4) + \sqrt{(1+k_1-k_2-k_4)^2+4k_2}\right) \tag{27}$$

$$\lambda_2 = \frac{1}{2}\left(-(1+k_1+k_2+k_4) - \sqrt{(1+k_1-k_2-k_4)^2+4k_2}\right) \tag{28}$$

while the third eigenvalue, which previously was $\lambda_3 = -k_3$, is now given by

$$\lambda_3 = -k_3(h_0+1), \tag{29}$$

where we recall that $h_0 = -h'(1)$. The corresponding eigenvectors are given by

$$v_i = (\mu(\lambda_i + k_3(1+h_0))(k_2+k_4+\lambda_i), \lambda_i + k_3(1+h_0), -(\lambda_i + k_4)), \quad i=1,2, \quad v_3 = (0,0,1) \tag{30}$$

4.2 Receptor Dynamics

Before considering the full model, we first consider the effect of feedback on the dynamics of y, the non-dimensional form of the receptor, in the absence of the ligand or product. As with the basic TMDD model, we note that $x = z = 0$ satisfies (16) and (17). Substituting $x = z = 0$ into (18), we obtain the single differential equation

$$\dot{y} = k_3(h(y) - y), \quad y(0) = y_0. \tag{31}$$

Assumption (h2) ensures that the previous steady state value $y = 1$ is also a steady state of this equation while assumption (h3) ensures that this is the only non-negative steady state solution and that $\dot{y} < 0$ if $y > 1$ and $\dot{y} > 0$ if $0 \leq y < 1$. The theory of scalar, autonomous differential equations [9] then tells us that if the initial condition y_0 is less than the steady state ($y_0 < 1$) then $y(\tau)$ will increase monotonically to the steady state, whereas if y_0 is greater than the steady state ($y_0 > 1$), then $y(\tau)$ will decrease monotonically to the steady state. As $y(\tau)$ is monotonic, there is no possibility of rebound occurring in the dynamics of the receptor on its own.

Assumptions (h2) and (h3) together with definition (15) imply that

$$h(y) - y = -(h_0+1)(y-1) + O((y-1)^2).$$

As $h_0 \geq 0$, the inclusion of the function $h(y)$ ensures that the asymptotic rate of convergence to the steady state (proportional to $e^{-k_3(h_0+1)t}$) will be faster than or equal to the rate of convergence which occurs when $h(y) = 1$ (proportional to e^{-k_3t}). Thus, qualitatively, the direct feedback has the effect of speeding up the monotonic convergence to the steady state in the absence of ligand and product, but there is no rebound in this case, i.e., if $y_0 < 1$, then $y(\tau) < 1$ for all $\tau \geq 0$.

4.3 Conditions for Rebound

Having established that feedback does not lead to rebound in the absence of the ligand, we now investigate the effect of adding the ligand. A local approximation of the feedback near the baseline will give a linear feedback function. So we first consider the special case that the feedback function $h(y)$ is linear for $y \leq 1$. It turns out to be straightforward to extend our results for the TMDD model without feedback to this case. The general nonlinear case will be analysed next and builds on the ideas of the linear analysis.

4.3.1 A Mainly Linear Feedback Function

We consider a feedback function function which is linear on a y-interval including $[0, 1]$:

$$h(y) = \begin{cases} 1 + h_0(1 - y), & 0 \leq y \leq 1 + \frac{\beta}{h_0}; \\ h_1(y), & y > 1 + \frac{\beta}{h_0} \end{cases} \tag{32}$$

where $h_0 > 0$, $0 < \beta < 1$, and $h_1 \in C^2\left(\left(1 + \frac{\beta}{h_0}, \infty\right)\right)$, $h_1(y) \in (0, 1)$, with

$$\lim_{y \downarrow 1 + \frac{\beta}{h_0}} h_1(y) = 1 - \beta, \quad \lim_{y \downarrow 1 + \frac{\beta}{h_0}} h_1'(y) = -h_0, \quad \text{and} \quad \lim_{y \downarrow 1 + \frac{\beta}{h_0}} h_1''(y) = 0.$$

This function satisfies all the conditions (h1)–(h4). For the linear part of the function h, we note that $h(y) - y = (1 + h_0)(1 - y)$ and so equation (18) becomes

$$\dot{y} = \begin{cases} k_3(1 + h_0)(1 - y) - \frac{xy}{\mu} + k_2 z, & 0 \leq y \leq 1 + \frac{\beta}{h_0}; \\ k_3(h_1(y) - y) - \frac{xy}{\mu} + k_2 z, & y > 1 + \frac{\beta}{h_0}. \end{cases} \tag{33}$$

We then have the following result.

Theorem 4.3 *Rebound occurs in equations (16), (17), (33) if and only if*

$$k_4 < k_1 \quad \text{and} \quad k_4 < k_3(1 + h_0).$$

Proof We note that for $0 \leq y \leq 1$, equations (16), (17) and (33) are the same as the basic TMDD model that we studied in [2] except that k_3 has been replaced by $k_3(1 + h_0)$. We claim that the results of Theorem 3.1 of [2] apply in this case also after making this parameter replacement. To see this, we note that when proving the absence of rebound the only relevant values of y are $0 \leq y \leq 1$ and so the matching point $y = 1 + \beta/h_0$ in (33) is not reached and the feedback function for $y > 1$ is therefore not relevant. Moreover, the proof of rebound in [2] involves only the asymptotic behaviour near to the globally stable steady state $(x, y, z) = (0, 1, 0)$ and the dynamics for $0 \leq y \leq 1 + \frac{\beta}{h_0}$ will give all the necessary information to determine this asymptotic behaviour. Hence, we can conclude that Theorem 3.1 in [2] can be applied in this situation with k_3 replaced by $k_3(1 + h_0)$. $\quad\square$

Converting back to the dimensional parameters gives the following result.

Corollary 4.4 *Consider the mainly linear feedback function*

$$H(R) = \begin{cases} 1 + H_0(R_0 - R), & 0 \leq R \leq R_0 + \frac{\beta}{H_0} \\ H_1(R), & R > R_0 + \frac{\beta}{H_0} \end{cases} \tag{34}$$

where $H_0 > 0$, $0 < \beta < 1$, $H_1(R) \in C^2\left(\left(R_0 + \frac{\beta}{H_0}, \infty\right)\right)$, $H_1(R) \in (0, 1)$ and H_1 and its first and second derivatives match the linear function at $R = R_0 + \beta/H_0$.

Rebound occurs in the model given by equations (8)–(10) with this feedback function if and only if

$$k_{\text{e(P)}} < k_{\text{e(L)}} \quad \text{and} \quad k_{\text{e(P)}} < k_{\text{out}}(1 + R_0 H_0).$$

The rebound region in this case is shown in Fig. 4. We note by comparison with Fig. 3 that this mainly linear feedback function has enlarged the region in which rebound occurs compared to the case with no feedback.
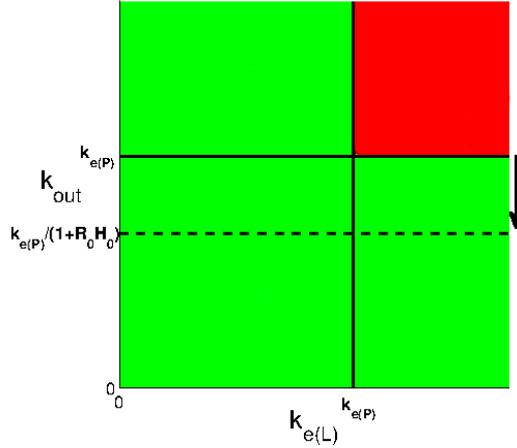
Figure 4: The green region shows the part of the $(k_{e(L)}, k_{out})$ parameter plane where there is no rebound, and the red region shows where there is rebound for $H(R)$ given by (34). The solid line $k_{out} = k_{e(P)}$ denotes the boundary of the rebound region when no feedback is present and the dashed line $k_{out} = k_{e(P)}/(1 + R_0 H_0)$ denotes the boundary of the rebound region when feedback is present.

4.3.2 A General Nonlinear Feedback Function

We now consider the more general case where the feedback function h is nonlinear. We express the function $h(y)$ in the form

$$h(y) = 1 + (1 - y)\tilde{h}(y). \tag{35}$$

Differentiating (35), evaluating at $y = 1$ and using (15), we see that

$$\tilde{h}(1) = h_0. \tag{36}$$

Condition (h3) implies that

$$\tilde{h}(y) > 0, \quad \text{when } 0 \leq y < 1, \text{ and } 0 < \tilde{h}(y) < \frac{1}{y-1}, \quad \text{when } y > 1. \tag{37}$$

With this form of h, equation (18) becomes

$$\dot{y} = k_3(1-y)(1+\tilde{h}(y)) - \frac{xy}{\mu} + k_2 z \tag{38}$$

Remark We note that expanding $h(y)$ in a Taylor series with remainder gives that $\tilde{h}(y) = -h'(\xi(y))$ where $\xi(y)$ lies between y and 1. However, the fact that $\xi(y)$ is an unknown function is not helpful, which is why we use the form of h given in (35).

We now consider regions similar to the ones defined for the model without feedback in [2]. First we consider region II and show that no rebound occurs in this region.

Theorem 4.5 *Rebound does not occur if $k_1 \leq k_4$.*

Proof Following through the proof of Theorem 3.4 of [2] for these modified equations, the only difference is that

$$\dot{y}|_{y=0} = k_3(1 + \tilde{h}(0)) + k_2 z.$$

Now $\tilde{h}(0) > 0$ by (37) and so $\dot{y}|_{y=0} > 0$ when $z \geq 0$, which is the condition required on this plane in the proof of Theorem 3.4 of [2]. Thus, the result holds in this case also. □

The next result considers a region similar to region III and we show that rebound does occur.

Theorem 4.6 *Rebound occurs if $k_1 > k_4$ and $k_3(1 + h_0) > -\lambda_1(k_1, k_2, k_4)$, where λ_1 is given by (27).*

Proof The Jacobian matrix for equations (16), (17) and (38) evaluated at the steady state is the same as that for the simple TMDD model given in [2] but with k_3 replaced by $k_3(1 + h_0)$. The proof of Theorem 3.5 of [2] (region III) is entirely based on the eigenvalues and eigenvectors of this matrix, and so the result holds in this case also, with the appropriate replacement of k_3. Since λ_1 does not involve k_3, there is no change to this eigenvalue. □

The proofs in regions I and IV of the (k_1, k_3) parameter plane in [2] both made use of the fact that for the model without feedback the plane $v = y + z = 1$ is invariant when $k_3 = k_4$. However, this is no longer the case for our modified equations which include feedback. The differential equation for the dimensionless variable associated with the total amount of receptor $v = y + z$ is now given by

$$\dot{v} = (1 - y)\left(k_3(1 + \tilde{h}(y)) - k_4\right) - k_4(v - 1) \tag{39}$$

and so

$$\dot{v}|_{v=1} = (1 - y)\left(k_3(1 + \tilde{h}(y)) - k_4\right). \tag{40}$$

Clearly no condition on the parameters will give this derivative to be zero (unless $\tilde{h}$ is constant, the case considered in the previous section), and so we see, as already stated, that the plane $v = 1$ is no longer invariant. However, the function $h(y)$ has a continuous derivative by (h4) and this implies that $\tilde{h}(y)$ is continuous. Thus the Extreme Value Theorem for continuous functions gives that $\tilde{h}(y)$ has a minimum and a maximum for $y \in [0, 1]$. So we define

$$m = \min_{y \in [0,1]} \tilde{h}(y), \quad M = \max_{y \in [0,1]} \tilde{h}(y). \tag{41}$$

Using (36), this immediately implies that

$$m \leq h_0 \leq M. \tag{42}$$

Also, since $\tilde{h}(y) > 0$ for all $0 \leq y < 1$ by (37), we get

$$m \geq 0. \tag{43}$$

Since m and M are the minimum and maximum of $\tilde{h}(y)$, then clearly

$$m \leq \tilde{h}(y) \leq M \quad \text{for all } 0 \leq y \leq 1,$$

and substituting for $\tilde{h}(y)$ from (35) and rearranging gives

$$1 + m(1 - y) \leq h(y) \leq 1 + M(1 - y) \quad \text{for all } 0 \leq y \leq 1. \tag{44}$$

Graphically, this means that on the interval $y \in [0, 1]$, the function $h(y)$ is bounded by two straight lines, both of which pass through the point $(1, 1)$, and which have slopes $-m$ and $-M$. This situation is illustrated in Fig. 5. Note that the Mean Value Theorem gives that $-M \geq \min\{h'(y) \mid 0 \leq y \leq 1\}$ and $-m \leq \max\{h'(y) \mid 0 \leq y \leq 1\}$.

With the bounds m and M, we can give a slightly weaker version of Lemma 2.3 in [2] and get regions in the (k_3, k_4)-plane for which the dimensionless variable v associated with the total amount of receptor stays on one side of the plane $v = 1$.
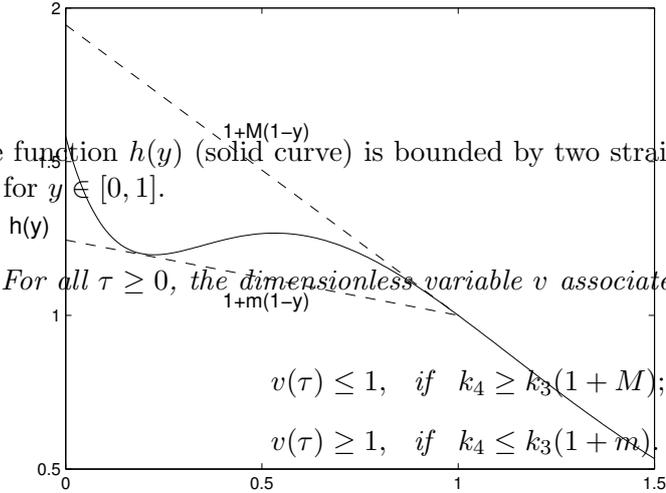
Figure 5: The function $h(y)$ (solid curve) is bounded by two straight lines (dashed lines) with slopes $-m$ and $-M$ for $y \in [0,1]$.

Lemma 4.7 *For all $\tau \geq 0$, the dimensionless variable v associated with the total amount of receptor satisfies*

$$v(\tau) \leq 1, \quad if \quad k_4 \geq k_3(1+M);$$

$$v(\tau) \geq 1, \quad if \quad k_4 \leq k_3(1+m).$$

Proof We consider the vector field at the plane $v = 1$. Equation (40) gives

$$\dot{v}|_{v=1} = (1-y)\left(k_3(1+\tilde{h}(y)) - k_4\right).$$

Since we have restricted attention to the plane $v = 1$, this implies that $y + z = 1$ and so $y \leq 1$ as z must be non-negative. As $1 - y$ is non-negative, we can use the bounds on $\tilde{h}(y)$ given in (41) to get

$$(1-y)\left(k_3(1+m) - k_4\right) \leq \dot{v}|_{v=1} \leq (1-y)\left(k_3(1+M) - k_4\right) \tag{45}$$

First we consider the case $k_4 \geq k_3(1+M)$. In this case, (45) gives $\dot{v}|_{v=1} \leq (1-y)\left(k_3(1+M) - k_4\right) \leq 0$, hence the vector field on the plane $v = 1$ is either tangent to $v = 1$ or points towards $v < 1$. This implies that the region of phase space defined by $x, y, z \geq 0$, $v = y + z \leq 1$ is invariant in forward time since the vector field on the four boundary planes is either tangent to the plane or points into the region [18]. This was proved for the planes $x = 0$, $y = 0$ and $z = 0$ in Lemma 4.1 and for the plane $v = 1$ above. Since $v(0) = 1$ and this region is invariant, we conclude that $v(\tau) \leq 1$ for all $\tau \geq 0$.

Next we consider the case $k_4 \leq k_3(1+m)$. Now the other inequality in (45) gives $\dot{v}|_{v=1} \geq (1-y)\left(k_3(1+m) - k_4\right) \geq 0$, hence the vector field on the plane $v = 1$ is either tangent to $v = 1$ or points towards $v > 1$. This implies that the region of phase space defined by $x, y, z \geq 0$, $v = y + z \geq 1$ is invariant in forward time. Since $v(0) = 1$, this implies that $v(\tau) \geq 1$ for all $\tau \geq 0$. $\qquad \square$

With this observation, we can now derive a result on rebound for regions similar to regions I and IV in [2].

Theorem 4.8 *Define m and M as in (41), i.e., the function $h(y)$ satisfies (44).*

(i) *Rebound does not occur if*

$$k_3 \leq \frac{k_4}{1 + M} \tag{46}$$

(ii) *Rebound does occur if*

$$\frac{k_4}{1 + m} < k_3 < -\frac{\lambda_1}{1 + h_0} \tag{47}$$

Using (42) and (43), we note that

$$0 < \frac{k_4}{1 + M} \leq \frac{k_4}{1 + h_0} \leq \frac{k_4}{1 + m} \leq k_4.$$

This illustrates in particular that there is no overlap between the region for which we have shown the existence of rebound and the region for which we have shown that there is no rebound.

Proof of Theorem 4.8

(i) The case when (46) holds is similar to region I in [2] and has the property that $v(\tau) \leq 1$ for all $\tau \geq 0$ by Lemma 4.7. Thus for all $\tau \geq 0$, we have $y(\tau) \leq v(\tau) \leq 1$ and so rebound cannot occur in this case.

(ii) When (47) holds (a condition similar to the one for region IV in [2]), Lemma 4.7 ensures that $v(\tau) \geq 1$ for all $\tau \geq 0$. The right hand inequality in (47) implies that $\lambda_1 < \lambda_3 = -k_3(1 + h_0)$ and so the eigenvalue closest to zero is λ_3, as occurred in region IV previously. Thus, trajectories will generically approach the steady state tangent to the y-axis, since the eigenvector associated with the eigenvalue λ_3 points along this axis. As the trajectory always satisfies $v \geq 1$ for all $\tau \geq 0$ then we again conclude that generically, almost all orbits will approach $y = 1$ from above and hence rebound occurs.

We must again eliminate the possibility that an orbit might approach the steady state tangent to one of the other eigenvectors for a particular choice of parameters. As previously, the linearised manifold in the direction of the eigenvector v_2 is outside of the phase space and so an orbit cannot approach tangent to this one-dimensional manifold.

The one-dimensional linearised manifold in the direction of the eigenvector v_1 given in (30) is

$$\begin{bmatrix} x \\ z \\ y \end{bmatrix} = \begin{bmatrix} 0 \\ 0 \\ 1 \end{bmatrix} + a \begin{bmatrix} \mu(k_2 + k_4 + \lambda_1)(\lambda_1 + k_3(1 + h_0)) \\ \lambda_1 + k_3(1 + h_0) \\ -(\lambda_1 + k_4) \end{bmatrix}.$$

We note that $\lambda_1 + k_3(1 + h_0) < 0$ from the right hand inequality in (47). Thus, we must take $a < 0$ to ensure that this linearised manifold gives positive values of x and z (since $k_2 + k_4 + \lambda_1 > 0$ by Lemma 2.7 of [2]). The linearised manifold for $v = y + z$ is then given by

$$v = 1 + a\left(k_3(1 + h_0) - k_4\right).$$

Using (42) and the left hand inequality of (47) gives $k_3(1 + h_0) - k_4 \geq k_3(1 + m) - k_4 > 0$. Thus, this one-dimensional linearised manifold has a negative slope and so occurs for $v < 1$. Since the trajectory in this case must satisfy $v \geq 1$, it is clearly not possible for it to approach the steady state tangent to this manifold. Thus, we conclude that all trajectories must approach the steady state tangent to the y-axis from above and hence rebound will occur. $\quad\square$

In Theorems 4.6 and 4.8, we have proved that rebound occurs on both sides of the line $\lambda_1 = \lambda_3$ with $k_3 > k_4/(1 + m)$. We now show that there is also rebound along this line.

Theorem 4.9 *Rebound occurs if $k_3 = -\lambda_1/(1 + h_0)$ and $k_3 > k_4/(1 + m)$.*

Proof The proof uses exactly the same arguments as the proof of Theorem 3.7 in [2]. Along the curve $k_3 = -\lambda_1/(1 + h_0)$, the eigenvalues λ_1 and λ_3 collide and have one eigenvector, which is along the y-axis, and one generalised eigenvector. Thus all solutions will asymptotically align with the y-axis. Since $k_4/(1 + m) < k_3$, Lemma 4.7 gives that $v(\tau) \geq 1$ and the intersection of the y-axis and the region $v(\tau) \geq 1$ is the part of the y-axis with $y \geq 1$. Hence all orbits must approach the steady state $(x, y, z) = (0, 1, 0)$ from above and rebound will occur. $\quad\Box$

We summarise the results of Theorems 4.5, 4.6, 4.8 and 4.9 as follows.

Theorem 4.10 *Define*

$$m = \inf\left\{ \frac{h(y) - 1}{1 - y} \,\middle|\, 0 \leq y < 1 \right\} \quad and \quad M = \sup\left\{ \frac{h(y) - 1}{1 - y} \,\middle|\, 0 \leq y < 1 \right\}. \tag{48}$$

For the model equations (16)–(18) and for any function $h(y)$ satisfying the assumptions (h1)–(h4)

- *there is no rebound if $k_1 \leq k_4$ or $k_1 > k_4$ and $k_3 \leq k_4/(1 + M)$;*

- *rebound does occur if $k_1 > k_4$ and $k_3 > \min\left(k_4/(1 + m), -\lambda_1/(1 + h_0)\right)$.*

We can also express these results in the terms of the dimensional parameters. Let $H(R)$ be a function satisfying the assumptions (H1)–(H4) and define

$$m = \inf\left\{ \frac{(H(R) - 1)R_0}{R_0 - R} \,\middle|\, 0 \leq R < R_0 \right\} \quad and \quad M = \sup\left\{ \frac{(H(R) - 1)R_0}{R_0 - R} \,\middle|\, 0 \leq R < R_0 \right\}.$$

For the model equations (8)–(10) with $H_0 = -H'(R_0)$

- *there is no rebound if $k_{e(L)} \leq k_{e(P)}$ or $k_{e(L)} > k_{e(P)}$ and $k_{out} < k_{e(P)}/(1 + M)$;*

- *rebound does occur if $k_{e(L)} > k_{e(P)}$ and $k_{out} > \min\left(k_{e(P)}/(1 + m), -\lambda_1 k_{on} R_0/(1 + R_0 H_0)\right)$.*

These results are illustrated in Fig. 6.

We now focus on a few aspects of the theorem and compare it with the case where there is no feedback ($F = 1$, and so $h_0 = 0 = H_0$).

- In the equations without feedback, rebound occurred in the region $k_{e(P)} < k_{e(L)}$ and $k_{e(P)} < k_{out}$ (regions III and IV) and with feedback, rebound also occurs in this region.

- In the equations without feedback, there was no rebound in the region $k_{e(L)} \leq k_{e(P)}$ (region II) and with feedback, there is also no rebound in this region.

- In the equations without feedback, there was no rebound in the region $k_{out} < k_{e(P)} < k_{e(L)}$ (the part of region I that lies outside region II), but with feedback, there is rebound in some parts of this region but not in others, the precise details depend on the function h.

- In the region $k_{e(L)} > k_{e(P)}$, $k_{e(P)}/(1 + M) \leq k_{out} \leq \min\left(k_{e(P)}/(1 + m), -\lambda_1/(1 + h_0)\right)$, we have no general results, and the existence or otherwise of rebound will depend on the particular function h.

- If H is mainly linear, i.e. $F = H(R) = 1 + H_0(R_0 - R)$, for $0 < R < R_0$, then $m = M = R_0 H_0$. In this case, the region of uncertainly disappears and we get that rebound will occur if and only if $k_{e(P)} < k_{e(L)}$ and $k_{e(P)} < k_{out}(1 + H_0 R_0)$.
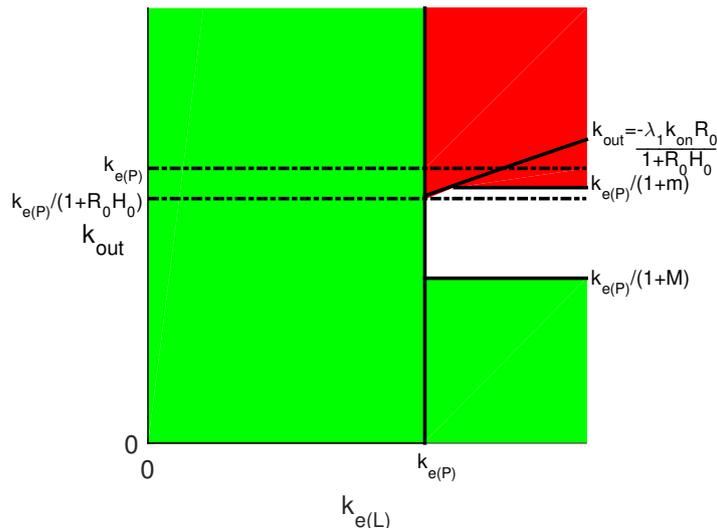
Figure 6: Regions in the $(k_{e(L)}, k_{out})$ plane where rebound does and does not occur for the nonlinear feedback function $H(R)$. Red indicates regions where rebound will occur and green indicates regions where rebound does not occur. In the white region, rebound may or may not occur depending on the particular feedback function H.

- If H is nonlinear but with $m = R_0 H_0$ (in which case the lower bounding curve for $H(R)$ is the tangent to the curve at $R = R_0$), then the red region where there is rebound for $k_{out} > k_{e(P)}/(1+R_0H_0)$ in Fig. 4 is preserved and the region of uncertainty is below this line. Conversely, if $M = R_0 H_0$ (so that the upper bounding curve for $H(R)$ is the tangent at $R = R_0$), then the green region where there is no rebound for $k_{out} \leq k_{e(P)}/(1 + R_0H_0)$ in Fig. 4 is preserved and the region of uncertainty is above this line.

- We note that when $k_{e(L)} < k_{e(P)}$ (the left green region in Fig. 6), then there is no rebound in the direct feedback model *for any feedback function $H(R)$*. As discussed above, when $k_{e(L)} > k_{e(P)}$, then the form of the feedback function determines whether or not rebound occurs.

In summary, the region where rebound occurs increases slightly to include some more values with $k_{out} < k_{e(P)} < k_{e(L)}$ when direct feedback is introduced into the basic TMDD model.

5 Analysis of the Full TMDD with Feedback Model

In the full TMMD with feedback model, we have an extra differential equation that governs the dynamics of the moderator w and so our equations are now (20)–(23) with initial condition (24). As mentioned previously, the parameter ϵ influences the time scale for the response dynamics. If ϵ is small, the feedback responds slowly and we will show that this always leads to rebound. On the other hand, if ϵ is very large, the feedback responds very fast and we will show that rebound will occur in the same region as found with the direct feedback approximation.

5.1 Dynamics of the Receptor with Feedback, without Ligand

Before considering the full model with feedback, we first consider the effect of feedback on the dynamics of the receptor y (or R in dimensional form) in the absence of the ligand or product. We substitute

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$x = z = 0$ into (20)–(23) to give the two equations

$$\dot{y} = k_3(w - y), \qquad y(0) = y_0 \tag{49}$$
$$\dot{w} = \epsilon(h(y) - w), \qquad w(0) = w_0 \tag{50}$$

Again, it is easily shown, using (h2) and (h3), that these equations have the unique steady state $y = w = 1$, i.e. the baseline values. Below we will prove the following rebound result.

Theorem 5.1 *For any $\epsilon \in (0, \infty)$, there are always some initial conditions with $y(0) < 1$ which give rise to trajectories with rebound.*

First we consider the linear stability of the baseline state as this gives the local behaviour near to the baseline. The Jacobian matrix evaluated at this steady state is given by

$$J_0 = \begin{bmatrix} -k_3 & k_3 \\ -\epsilon h_0 & -\epsilon \end{bmatrix} \tag{51}$$

(recalling that $h_0 = -h'(1) \geq 0$). The eigenvalues of J_0 can be found as solutions of the quadratic characteristic polynomial derived from this matrix. The discriminant of this quadratic equation is

$$D := \epsilon^2 - 2k_3(1 + 2h_0)\epsilon + k_3^2.$$

and the eigenvalues are

$$\lambda_3 = \frac{1}{2}\left(-(k_3 + \epsilon) - \sqrt{D}\right) = \frac{1}{2}\left(-(k_3 + \epsilon) - \sqrt{(k_3 - \epsilon)^2 - 4\epsilon k_3 h_0}\right), \tag{52}$$
$$\lambda_4 = \frac{1}{2}\left(-(k_3 + \epsilon) + \sqrt{D}\right) = \frac{1}{2}\left(-(k_3 + \epsilon) + \sqrt{(k_3 - \epsilon)^2 - 4\epsilon k_3 h_0}\right), \tag{53}$$

while the eigenvectors are of the form

$$v_i = (k_3, k_3 + \lambda_i), \quad \text{for} \quad i = 3, 4. \tag{54}$$

Note that in this section, the definition of the eigenvalue λ_3 is different from that used in the previous section. The discriminant D is itself a quadratic function of ϵ which has two positive roots given by

$$\epsilon_1^{\pm} = k_3\left(1 + 2h_0 \pm \sqrt{4h_0(1 + h_0)}\right). \tag{55}$$

Clearly the discriminant is negative between these roots and positive elsewhere, and so we have the following result.

Lemma 5.2 *The equations (49)–(50) have a unique steady state at $y = w = 1$ and the eigenvalues λ_3 and λ_4 of the Jacobian matrix J_0 are*

- *both real and negative if $0 < \epsilon \leq \epsilon_1^-$ or if $\epsilon \geq \epsilon_1^+$;*

- *a complex conjugate pair with negative real part if $\epsilon_1^- < \epsilon < \epsilon_1^+$.*

Furthermore, when $\epsilon \to \infty$, $\lambda_3 \to -\infty$ and $\lambda_4 \to \lambda_\infty = -k_3(1 + h_0)$.

For a proof of the last statement, see Appendix A. Lemma 5.2 is summarised in Fig. 7 with the direction of movement of the eigenvalues indicated corresponding to increasing ϵ. When $\epsilon = 0$, we see that $\lambda_3 = -k_3$ and $\lambda_4 = 0$. As ϵ increases from zero, the two eigenvalues move towards each other and collide on the negative real axis when $\epsilon = \epsilon_1^-$. After this collision, the eigenvalues become complex with $\text{Re}(\lambda_{3,4}) = -(k_3 + \epsilon)/2$, which decreases monotonically with ϵ. As ϵ increases further, the two complex eigenvalues collide on the real axis when $\epsilon = \epsilon_1^+$ and become real again. As ϵ increases from
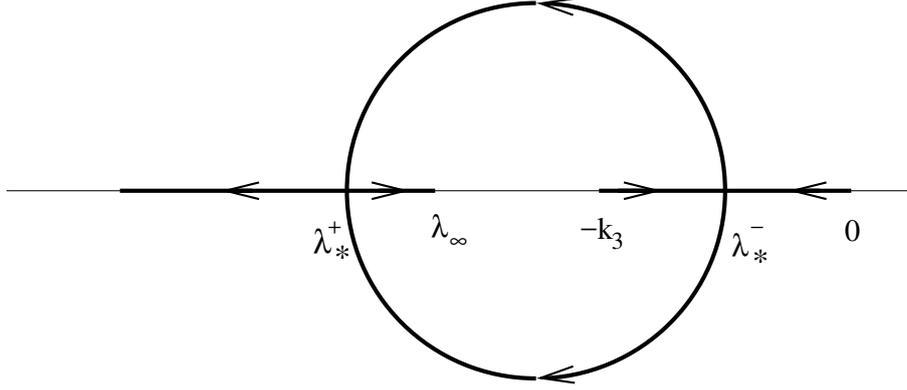
Figure 7: Movement of the eigenvalues λ_3 and λ_4 in the complex plane for increasing ϵ (indicated by the direction of the arrows).

ϵ_1^+ to ∞, λ_4 increases from the point of collision to $-k_3(1 + h_0)$ and λ_3 decreases to $-\infty$ from this point. At the collision values $\epsilon = \epsilon_1^\pm$, the eigenvalues $\lambda_3 = \lambda_4$ are given by

$$\lambda_*^\pm = -\frac{1}{2}(k_3 + \epsilon_1^\pm) = -k_3\left(1 + h_0 \pm \sqrt{h_0(1 + h_0)}\right). \tag{56}$$

Note that this implies that $\lambda_*^+ < \lambda_\infty < \lambda_*^-$, as $\lambda_\infty = -k_3(1 + h_0)$.

Lemma 5.2 implies linear stability, but a stronger stability result can be proved. The proof of this result uses a Lyapunov function and is given in Appendix A.

Lemma 5.3 *In the region of $\mathbb{R}^2$ defined by $y, w \geq 0$, the steady state $y = w = 1$ of equations (49)–(50) is globally asymptotically stable when $\epsilon > 0$.*

Thus the trajectory for all non-negative initial conditions will eventually approach the steady state $(y, w) = (1, 1)$ and Theorem 5.1 can be proved by analysing the solutions close to the steady states. The following lemma gives the detailed results, with the proof again in Appendix A.

Lemma 5.4

- If $0 < \epsilon \leq \epsilon_1^-$ and $y(0) < 1$, then rebound in equations (49)–(50) will occur for all $w(0) > 1$ and for some values of $w(0) \leq 1$.

- If $\epsilon_1^- < \epsilon_1 < \epsilon_1^+$ then rebound occurs for any initial condition with $y(0) < 1$.

- If $\epsilon_1^+ \leq \epsilon < \infty$ and $y(0) < 1$, then rebound in equations (49)–(50) will occur for some initial conditions which satisfy $w(0) > 1$.

Having established the effect of the feedback on the behaviour of the receptor without ligand and product, we next consider the effect of adding the ligand.

5.2   Invariance, Steady States and Stability

Before considering rebound, we first state invariance and stability results for the full TMDD system with feedback, similar to the results in Section 4.1 for the TMDD system with direct feedback. The proofs of the statements in this section can be found in Appendix A.

The first three of our variables are related to physical quantities and so must be non-negative and we have also assumed that the feedback variable w is non-negative. Therefore, we now establish the invariance of the region with $x, y, z, w \geq 0$ for equations (20)–(23).

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Lemma 5.5 *The region of $\mathbb{R}^4$ defined by $x, y, z, w \geq 0$ is invariant under the flow of equations (20)–(23) when $\epsilon > 0$.*

As with our previous model, this again shows that none of the variables can go negative, which is an essential property of a good model. We now consider the steady states of equations (20)–(23).

Lemma 5.6 *In the region of $\mathbb{R}^4$ defined by $x, y, z, w \geq 0$, there is a unique steady state of equations (20)–(23), given by*

$$x = 0, \quad z = 0, \quad y = 1, \quad w = 1. \tag{57}$$

The Jacobian matrix obtained from equations (20)–(23) evaluated at the steady state solution (57) is given by

$$\begin{pmatrix} -(k_1 + 1) & \mu k_2 & 0 & 0 \\ \frac{1}{\mu} & -(k_2 + k_4) & 0 & 0 \\ -\frac{1}{\mu} & k_2 & -k_3 & k_3 \\ 0 & 0 & -\epsilon h_0 & -\epsilon \end{pmatrix} \tag{58}$$

(recalling that $h_0 = -h'(1)$). The eigenvalues of this matrix can be found from the top left 2×2 matrix and the bottom right 2×2 matrix. From the top left matrix, we obtain the eigenvalues λ_1 and λ_2 that we had previously, given by (27) and (28). The bottom right matrix is the same as J_0, defined in (51), and so the eigenvalues of this matrix are λ_3 and λ_4 as defined in (52) and (53). The corresponding eigenvectors are given by

$$v_i = \begin{pmatrix} \mu(k_2 + k_4 + \lambda_i) \\ 1 \\ -\dfrac{(k_4 + \lambda_i)(\epsilon + \lambda_i)}{(k_3 + \lambda_i)(\epsilon + \lambda_i) + \epsilon k_3 h_0} \\ \dfrac{\epsilon h_0(k_4 + \lambda_i)}{(k_3 + \lambda_i)(\epsilon + \lambda_i) + \epsilon k_3 h_0} \end{pmatrix}, \quad i = 1, 2; \qquad v_i = \begin{pmatrix} 0 \\ 0 \\ k_3 \\ k_3 + \lambda_i \end{pmatrix}, \quad i = 3, 4. \tag{59}$$

We now immediately get the linear stability of the steady state solution as all eigenvalues λ_i are strictly negative.

Lemma 5.7 *The steady state (57) of equations (20)–(23) is linearly stable when $\epsilon > 0$.*

This result implies that all trajectories with initial conditions *sufficiently close* to the steady state will converge to it. On its own, it does not guarantee convergence for all initial conditions. However, for this model, a stronger stability result can again be proved.

Lemma 5.8 *The steady state $(x, z, y, w) = (0, 0, 1, 1)$ of equations (20)–(23) is globally asymptotically stable when $\epsilon > 0$.*

This result ensures that for any non-negative initial conditions and for all positive parameter values, each of the variables will converge to their unique steady state value, as with the previous model.

The proofs of these two results are again in Appendix A.

5.3 Rebound

In this section, we focus on the analysis of rebound in the later stages of the evolution, i.e., the approach to the steady state. The linearised system will play an important role in this, but first we consider the limiting case $\epsilon = 0$ and derive some estimates on the initial behaviour of the moderator.

For $\epsilon = 0$, the full TMDD equations with feedback via a moderator, i.e., (20)–(23), reduce to

$$
\begin{aligned}
\dot{x} &= -k_1 x - xy + \mu k_2 z \\
\dot{z} &= \frac{xy}{\mu} - (k_2 + k_4)\, z \\
\dot{y} &= k_3(w - y) - \frac{xy}{\mu} + k_2 z \\
\dot{w} &= 0
\end{aligned}
$$

Hence w is constant. With the initial condition (24), this implies $w = 1$ and the system reduces to the TMDD equation without feedback as studied in our previous paper [2]. However, if w has a different initial condition, say $w = w_0 \neq 1$, then the system will converge to the steady state $(x, z, y, w) = (0, 0, w_0, w_0)$. Within the hyperplane $w = w_0$, this equilibrium is an attractor. Thus if $w_0 > 1$, then the receptor will converge to a value above the original baseline. Note that in this case, we have $\lambda_4 = 0$, corresponding to the invariance of the moderator w and the line of steady states $(0, 0, w_0, w_0)$.

For $\epsilon > 0$ and the initial condition (24), we can show that initially the moderator w will increase to values above its baseline and hence the production of the receptor will be stimulated.

Lemma 5.9 *For $\epsilon > 0$ and $h_0 > 0$, there exists a $\tau^* > 0$ such that $w(\tau) > 1$ for all $\tau \in (0, \tau^*)$.*

Proof The initial condition is $w(0) = 1$ and since this is also the steady state value of w, we have $\dot{w}(0) = 0$. Differentiating (23) and evaluating at $\tau = 0$ gives

$$
\ddot{w}(0) = \frac{\epsilon h_0}{\mu} > 0
$$

and the result follows immediately from this. $\quad\square$

The initial conditions give that $\dot{y}(0) = -\frac{1}{\mu}$, hence initially y will decrease and take values smaller than its baseline value 1. The following lemma implies that if there is no rebound, then $w(\tau)$ must be larger than 1 for all time. This observation will be used several times later to derive a contradiction on the assumption that there is no rebound.

Lemma 5.10 *If $\epsilon > 0$, $h_0 > 0$ and $0 \leq y(\tau) \leq 1$ for all $\tau \geq 0$, then $w(\tau) > 1$ for $\tau > 0$.*

Proof Assume that $\epsilon > 0$, $h_0 > 0$ and $0 \leq y(\tau) \leq 1$ for all $\tau \geq 0$. We evaluate $\dot{w}$ along the hyperplane $w = 1$ giving

$$
\dot{w}|_{w=1} = \epsilon(h(y) - 1).
$$

By hypothesis (h3), $h(y) > 1$ if $y < 1$ and so $\dot{w}|_{w=1} > 0$ in this case. Since $w(\tau) > 1$ for sufficiently small τ by Lemma 5.9, this result shows that $w(\tau)$ cannot touch or cross the line $w = 1$ when $y(\tau) < 1$.

Next we will show by contradiction that $y(\tau) = 1 = w(\tau)$ is impossible too. We assume that there is some $\tilde{\tau} > 0$ such that $w(\tilde{\tau}) = 1$ with $w(\tau) > 1$ for $\tau \in (0, \tilde{\tau})$, i.e. $\tilde{\tau}$ is the first moment that $w(\tau)$ equals one, and that $y(\tilde{\tau}) = 1$ also. Using the model equations (20)–(23), we obtain

$$
\dot{y}(\tilde{\tau}) = -\left(\frac{x(\tilde{\tau})}{\mu} - k_2 z(\tilde{\tau})\right), \quad \dot{w}(\tilde{\tau}) = 0, \quad \ddot{w}(\tilde{\tau}) = -\epsilon h_0 \dot{y}(\tilde{\tau}), \quad \dddot{w}(\tilde{\tau}) = \epsilon[h''(1)(\dot{y}(\tilde{\tau}))^2 - h_0 \ddot{y}(\tilde{\tau}) - \ddot{w}(\tilde{\tau})].
$$

Thus, using Taylor series, y and w near to $\tilde{\tau}$ are given by

$$
y(\tilde{\tau} + \hat{\tau}) = 1 + \dot{y}(\tilde{\tau})\,\hat{\tau} + \frac{\ddot{y}(\tilde{\tau})}{2}\,\hat{\tau}^2 + \mathcal{O}(\hat{\tau}^3), \quad w(\tilde{\tau} + \hat{\tau}) = 1 - \frac{\epsilon h_0}{2}\,\ddot{y}(\tilde{\tau})\,\hat{\tau}^2 - \frac{\epsilon h_0}{6}\,\ddot{y}(\tilde{\tau})\,\hat{\tau}^3 + \mathcal{O}(\hat{\tau}^4).
$$

The assumption $w(\tau) > 1$ for $\tau < \tilde{\tau}$ and $y(\tau) \leq 1$ for all τ together with $h_0 > 0$ imply that $\dot{y}(\tilde{\tau}) = 0$. Looking at the expressions for the derivatives at $\tilde{\tau}$, this gives $x(\tilde{\tau}) = \mu k_2 z(\tilde{\tau})$, $\ddot{w}(\tilde{\tau}) = 0$ and

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$\ddot{w}(\tilde{\tau}) = -\epsilon h_0 \ddot{y}(\tilde{\tau})$. Going to the next order in the Taylor series, since $y(\tau) \leq 1$, then we must have that $\ddot{y}(\tilde{\tau}) \leq 0$. If $\ddot{y}(\tilde{\tau}) < 0$ then this implies that $w(\tilde{\tau} + \hat{\tau}) < 1$ for $\hat{\tau} < 0$ and $|\hat{\tau}|$ sufficiently small, which contradicts our earlier assumption on w. Thus, the only possibility is that $\ddot{y}(\tilde{\tau}) = 0$. Using the relation $x(\tilde{\tau}) = \mu k_2 z(\tilde{\tau})$ we then find that

$$\ddot{y}(\tilde{\tau}) = k_2(k_1 - k_4)z(\tilde{\tau})$$

and so this second derivative is zero if either $z(\tilde{\tau}) = 0$ or $k_1 = k_4$. If $z(\tilde{\tau}) = 0$, then $x(\tilde{\tau}) = \mu k_2 z(\tilde{\tau}) = 0$ too. As we also have $y(\tilde{\tau}) = 1 = w(\tilde{\tau})$, this would imply that the solution is at the equilibrium at $t = \tilde{\tau}$. Uniqueness of solutions gives that this is not possible. With the alternative option $k_1 = k_4$, evaluating $\dot{x} - \mu k_2 \dot{z}$, $\dot{y}$ and $\dot{w}$ on the line

$$\{x = \mu k_2 z, \; y = 1, \; w = 1\}$$

shows that this line is invariant under the dynamics. At $t = 0$, $x(0) - \mu k_2 z(0) = 1$, hence initially the solution is not on this line and therefore it can never be on the line in finite time, which rules out the possibility that $k_1 = k_4$ and we have come to a contradiction. We therefore conclude that $w(\tilde{\tau}) \neq 1$ and so it follows that $w(\tau) > 1$ for all $\tau > 0$. $\square$

Corollary 5.11 *If $\epsilon > 0$, $h_0 > 0$ and $w(t^*) \leq 1$ for some $t^* > 0$, then rebound occurs.*

Proof This result is simply the converse of Lemma 5.10. $\square$

The observation that initially the moderator will be larger than 1, combined with the fact that for $\epsilon = 0$, the initial condition $w(0) > 1$ leads to rebound, makes it likely that rebound will happen for all small values of ϵ. This is indeed the case as will be shown below.

5.3.1 Relative position of the eigenvalues

To analyse rebound in the latter stages of the evolution, we focus on solutions near to the baseline state. We are especially interested in the eigenvalue of the Jacobian matrix (58) with real part closest to zero, since generically trajectories approach the steady state tangent to the eigenvector corresponding to this eigenvalue. We note that the eigenvalues λ_1 and λ_2 are always real, depend on k_1, k_2 and k_4, do not depend on ϵ and k_3, and that $\lambda_2 < \lambda_1$. Furthermore, for k_2 and k_4 fixed, the eigenvalue $\lambda_1(k_1, k_2, k_4)$ is monotonic decreasing in k_1 and

$$\lambda_1(0, k_2, k_4) = -\frac{1 + k_2 + k_4 - \sqrt{(1 + k_2 + k_4)^2 - 4k_4}}{2}; \quad \lambda_1(k_4, k_2, k_4) = -k_4; \quad \lim_{k_1 \to \infty} \lambda_1(k_1, k_2, k_4) = -(k_2 + k_4). \tag{60}$$

On the other hand, the eigenvalues λ_3 and λ_4 depend on k_3 and ϵ, do not depend on k_1, k_2 and k_4, and $\mathrm{Re}(\lambda_3) \leq \mathrm{Re}(\lambda_4)$. Thus, the eigenvalue with real part closest to zero will be either λ_1 or λ_4. As observed before, λ_1 does not depend on ϵ, and for small ϵ we have

$$\lambda_4 = -(1 + h_0)\epsilon + O(\epsilon^2).$$

Thus, for sufficiently small ϵ, the eigenvalue closest to zero will be λ_4. To describe how this changes when ϵ increases, we consider four λ_1 intervals that are defined in terms of $\lambda_*^{\pm}$ (see (56)) and λ_∞ (see Lemma 5.2) – a summary is sketched in Fig. 8:

- $\lambda_*^-(k_3) \leq \lambda_1(k_1, k_2, k_4) < 0$ (i.e., $k_3 \geq \frac{-\lambda_1}{1 + h_0 - \sqrt{h_0(1 + h_0)}}$):

  Now λ_1 is in the interval where λ_4 takes real values, hence the eigenvalues λ_4 and λ_1 cross at the point where $\lambda_4 = \lambda_1$ and solving this equation for ϵ gives

  $$\epsilon = \epsilon_2(\lambda_1, k_3) := -\lambda_1\left(\frac{\lambda_1 + k_3}{\lambda_1 + k_3(1 + h_0)}\right) = \epsilon_1^-(k_3) - \frac{(\lambda_1 - \lambda_*^-)^2}{\lambda_1 - \lambda_\infty} = \epsilon_1^+(k_3) + \frac{(\lambda_1 - \lambda_*^+)^2}{\lambda_\infty - \lambda_1}. \tag{61}$$
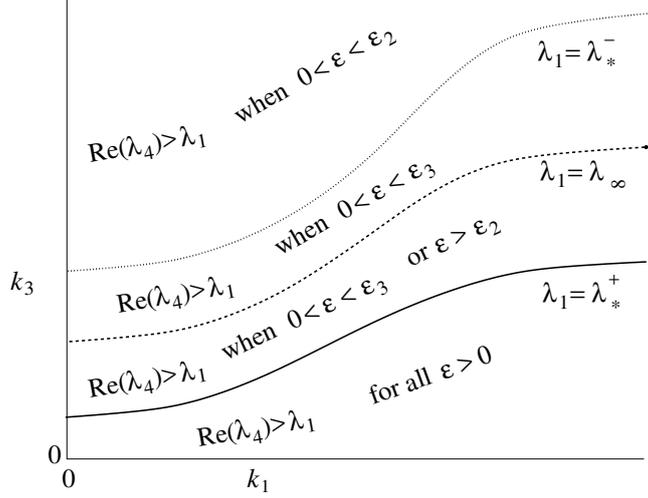
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Figure 8: The relative positioning of the eigenvalues in the k_1-k_3 plane with k_2 and k_4 fixed. Recall that $\lambda_\infty(k_3) = -k_3(1 + h_0)$ and $\lambda_*^\pm(k_3) = -k_3(1 + h_0 \pm \sqrt{h_0(1 + h_0)})$, thus the three curves are relations of the form $k_3 = \lambda_1(k_1, k_2, k_4)/f(h_0)$, with $f(h_0)$ the appropriate function.

Note that $\lambda_1 + k_3(1 + h_0) = \lambda_1 - \lambda_\infty \neq 0$ in this interval and so $\epsilon_2(\lambda_1, k_3)$ is well defined. Thus $\epsilon_2(\lambda_1, k_3) = \epsilon_1^-(k_3)$, when $\lambda_1 = \lambda_*^-$. Altogether, we conclude that in this λ_1 interval, we have that $\lambda_4 > \lambda_1$ if $0 < \epsilon < \epsilon_2(\lambda_1, k_3)$.

- $\lambda_\infty(k_3) \leq \lambda_1(k_1, k_2, k_4) \leq \lambda_*^-(k_3)$ (i.e., $\frac{-\lambda_1}{1+h_0} \leq k_3 \leq \frac{-\lambda_1}{1+h_0-\sqrt{h_0(1+h_0)}}$):

  Here λ_1 is in the interval where λ_4 takes complex values, hence for $\epsilon < \epsilon_1^-(k_3)$, λ_4 is real and will be the eigenvalue closest to zero. At $\epsilon = \epsilon_1^-(k_3)$, λ_4 and λ_3 merge and become a complex conjugate pair. For ϵ just larger than ϵ_1^-, the real part of the complex pair will be closest to zero and this will be the case until the real part of the complex eigenvalues crosses λ_1. This occurs when $\mathrm{Re}(\lambda_{3,4}) = \lambda_1$ and since $\mathrm{Re}(\lambda_{3,4}) = -(k_3 + \epsilon)/2$, this crossing occurs at $\epsilon = \epsilon_3$, where

  $$\epsilon_3(\lambda_1, k_3) := -(k_3 + 2\lambda_1)$$

  Since $\epsilon_1^\pm = -(k_3 + 2\lambda_*^\pm)$, it follows immediately that

  $$\epsilon_1^-(k_3) < \epsilon_3(\lambda_1, k_3) < \epsilon_1^+(k_3), \quad \text{for } \lambda_*^+ < \lambda_1 < \lambda_*^-. \tag{62}$$

  Note that $\epsilon_3(\lambda_1, k_3) = \epsilon_1^-(k_3) = \epsilon_2(\lambda_1, k_3)$, when $\lambda_1 = \lambda_*^-$. Thus in this interval, we have that $\mathrm{Re}(\lambda_4) > \lambda_1$ for $0 < \epsilon < \epsilon_3(\lambda_1, k_3)$.

- $\lambda_*^+(k_3) \leq \lambda_1(k_1, k_2, k_4) < \lambda_\infty(k_3)$ (i.e., $\frac{-\lambda_1}{1+h_0+\sqrt{h_0(1+h_0)}} \leq k_3 < \frac{-\lambda_1}{1+h_0}$):

  Now λ_1 is in the interval where λ_4 takes both real and complex values. If $\epsilon > \epsilon_1^+(k_3)$, then λ_4 is real and we can use the calculation in the first point to conclude that $\lambda_4 > \lambda_1$ if $\epsilon > \epsilon_2(\lambda_1, k_3)$. If $\epsilon_1^-(k_3) < \epsilon < \epsilon_1^+(k_3)$, then λ_4 is complex and the calculation above gives that $\mathrm{Re}(\lambda_4) > \lambda_1$ if $0 < \epsilon < \epsilon_3(\lambda_1, k_3)$. Note that $\epsilon_2(\lambda_1, k_3) > \epsilon_1^+(k_3) > \epsilon_3(\lambda_1, k_3)$ in the open λ_1 interval and that $\epsilon_3(\lambda_1, k_3)$ and $\epsilon_2(\lambda_1, k_3)$ collide with $\epsilon_1^+(k_3)$ when $\lambda_1 = \lambda_*^+(k_3)$. Furthermore, $\epsilon_2(\lambda_1, k_3) \to \infty$ for $\lambda_1 \to \lambda_\infty$. So altogether we have that in this interval

  $$\mathrm{Re}(\lambda_4) > \lambda_1 \quad \text{if} \ \ 0 < \epsilon < \epsilon_3(\lambda_1, k_3) \ \ \text{or} \ \ \epsilon > \epsilon_2(\lambda_1, k_3).$$

- $\lambda_1(k_1, k_2, k_4) < \lambda_*^+(k_3)$ (i.e., $k_3 < \frac{-\lambda_1}{1+h_0+\sqrt{h_0(1+h_0)}}$):

  Since $\mathrm{Re}(\lambda_4) \geq \lambda_*^+(k_3)$, we see immediately that in this interval $\mathrm{Re}(\lambda_4) > \lambda_1$ for all $\epsilon > 0$.

To visualise this information, we have sketched the λ_1 intervals in the k_1-k_3 plane in Fig. 8.

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5.3.2 Rebound in the latter stages of the evolution

Now that we have determined the positioning of the eigenvalues, we can state when rebound occurs in the latter stages of the evolution.

Theorem 5.12 *Define m as in Theorem 4.10, i.e., $m = \inf\left\{\frac{h(y)-1}{y-1} \mid 0 \leq y < 1\right\}$ and define $f_*^+(h_0) = 1 + h_0 + \sqrt{h_0(1+h_0)}$, hence $\lambda_*^+ = -k_3 f_*^+(h_0)$.*

(i) If $k_3 < \min\left(\frac{-\lambda_1}{f_*^+(h_0)}, \frac{k_4}{1+m}\right)$, then rebound occurs generically for $0 < \epsilon < \epsilon_1^+(k_3)$.

(ii) If $k_1 > k_4$ and

> *(a) $\frac{-\lambda_1}{f_*^+(h_0)} < k_3 < \min\left(\frac{-\lambda_1}{1+h_0}, \frac{k_4}{1+m}\right)$, then rebound occurs generically for $0 < \epsilon < \epsilon_2(\lambda_1, k_3)$;*
>
> *(b) $k_3 > \min\left(\frac{-\lambda_1}{1+h_0}, \frac{k_4}{1+m}\right)$, then rebound occurs generically for all $\epsilon > 0$.*

(iii) If $k_1 < k_4$ and

> *(a) $-\frac{\lambda_1}{f_*^+(h_0)} < k_3 \leq -\lambda_1$ then rebound occurs generically for $0 < \epsilon < \epsilon_3(\lambda_1, k_3)$;*
>
> *(b) $k_3 > -\lambda_1$ then rebound occurs generically for $0 < \epsilon < -\lambda_1$.*

These results are summarised in Fig. 9.
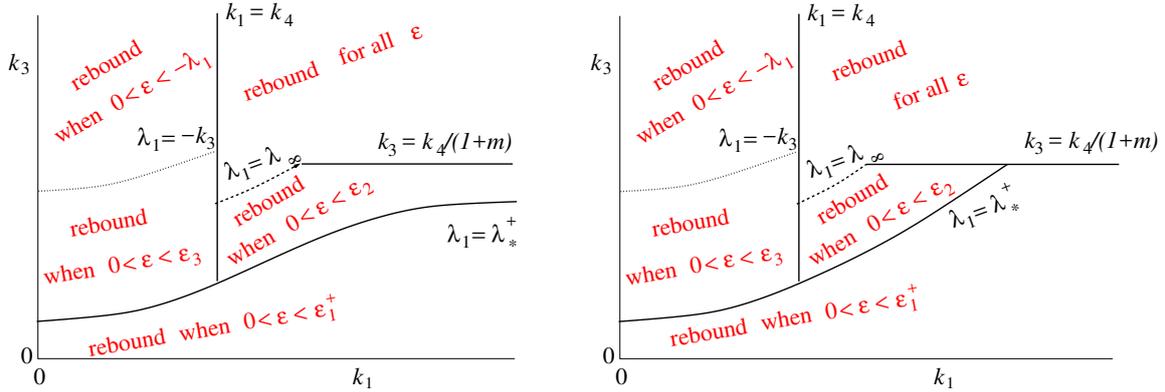

Figure 9: Rebound overview, on the left for $k_2 \leq k_4 \frac{f_*^+ - (1+m)}{1+m}$ and on the right for $k_2 > k_4 \frac{f_*^+ - (1+m)}{1+m}$. Note that $f_*^+(h_0) > 1 + h_0 \geq 1 + m$. Recall that $\lambda_*^+ = -k_3 f_*^+(h_0)$ and $\lambda_\infty = -k_3(1+h_0)$.

The proof of this theorem will be given via two lemmas which determine when the eigenvalue closest to zero can be associated with rebound. First we consider the case that the eigenvalue λ_4 (or its real part if complex) is closest to zero.

Lemma 5.13 *If the eigenvalue λ_4 (or its real part if complex) is closest to zero and*

- $0 < \epsilon < \epsilon_1^+$ *or*

- $\epsilon \geq \epsilon_1^+$, *and* $k_3 > \frac{k_4}{1+m}$,

then rebound occurs (generically only for $\epsilon_1^- < \epsilon < \epsilon_1^+$).

The proof of this lemma can be found in Appendix B.

Next we consider the case that the eigenvalue λ_1 is closest to zero.

Lemma 5.14 *If λ_1 is the eigenvalue closest to zero, then rebound occurs generically if $k_1 > k_4$ or if $\epsilon < -\lambda_1$.*

Again, the proof of this lemma can be found in Appendix B.

With the lemmas above, we are ready to prove Theorem 5.12.

Proof of Theorem 5.12 The results in this Theorem are essentially obtained by applying Lemmas 5.13 and 5.14 in each of the four regions described in Section 5.3.1. The results from this process are summarised in Table 1. The observations about the relative position of ϵ_2, ϵ_3 and ϵ^+ in the various regions follow from (61) and (62). In the regions $\lambda_*^+ < \lambda_1 < \lambda_*^-$, the case $\mathrm{Re}(\lambda_4) < \lambda_1$ leads to the range $\epsilon_3 < \epsilon < -\lambda_1$. This range is not empty only when $\epsilon_3 < -\lambda_1$. The definition of ϵ_3 shows that this condition holds if $\lambda_1 > -k_3$, which is why this condition is included in the second row. For the third row, we observe that $\lambda_\infty = -k_3(1 + h_0) < -k_3$, so the range $\epsilon_3 < \epsilon < -\lambda_1$ is always empty if $\lambda_1 < \lambda_\infty$.

Region	$\mathrm{Re}(\lambda_4) > \lambda_1$	Conditions for Rebound	$\mathrm{Re}(\lambda_4) < \lambda_1$	Conditions for Rebound
$\lambda_*^- \leq \lambda_1 < 0$	$0 < \epsilon < \epsilon_2(< \epsilon_1^+)$	✓	$\epsilon > \epsilon_2$	$k_1 > k_4$ or $\epsilon_2 < \epsilon < -\lambda_1$
$\lambda_\infty \leq \lambda_1 \leq \lambda_*^-$	$0 < \epsilon < \epsilon_3(< \epsilon_1^+)$	✓	$\epsilon > \epsilon_3$	$k_1 > k_4$ or $\epsilon_3 < \epsilon < -\lambda_1$ if $\lambda_1 > -k_3$
$\lambda_*^+ \leq \lambda_1 < \lambda_\infty$	$0 < \epsilon < \epsilon_3(\leq \epsilon_1^+)$; $\epsilon > \epsilon_2(\geq \epsilon_1^+)$	✓ ; $k_3 > \frac{k_4}{1+m}$	$\epsilon_3 < \epsilon < \epsilon_2$	$k_1 > k_4$
$\lambda_1 < \lambda_*^+$	$\epsilon > 0$	$0 < \epsilon < \epsilon_1^+$ or $\epsilon \geq \epsilon_1^+$ if $k_3 > \frac{k_4}{1+m}$	–	–

Table 1: Summary of results obtained by applying Lemmas 5.13, 5.14 to the eigenvalue categories described in Section 5.3.1. The ticks indicate that rebound occurs in the given intervals.

The two columns containing conditions for rebound in Table 1 can be combined in some cases, as shown in Table 2, where we separate out two regions in the parameter plane defined by $k_1 \leq k_4$ and $k_1 > k_4$. For the third row, we have used that $\lambda_1(k_1, k_2, k_4)$ is monotonic decreasing in λ_1. Hence if $k_1 \leq k_4$, then $\lambda_1(k_1, k_2, k_4) \geq \lambda_1(k_4, k_2, k_4) = -k_4$ (see (60)). Thus if $\lambda_1 < \lambda_\infty = -k_3(1 + h_0)$ and $k_1 \leq k_4$, then $k_3 < -\frac{\lambda_1}{1+h_0} \leq \frac{k_4}{1+h_0} \leq \frac{k_4}{1+m}$.

Region	$k_1 \leq k_4$	$k_1 > k_4$
$\lambda_*^- \leq \lambda_1 < 0$	$0 < \epsilon < \epsilon_2$, $\epsilon_2 < \epsilon < -\lambda_1$	$0 < \epsilon < \epsilon_2$, $\epsilon_2 < \epsilon$
$\lambda_\infty \leq \lambda_1 \leq \lambda_*^-$	$0 < \epsilon < \epsilon_3$, $\epsilon_3 < \epsilon < -\lambda_1$ if $\lambda_1 > -k_3$	$0 < \epsilon < \epsilon_3$, $\epsilon_3 < \epsilon$
$\lambda_*^+ \leq \lambda_1 < \lambda_\infty$	$0 < \epsilon < \epsilon_3$	$0 < \epsilon < \epsilon_3$, $\epsilon_3 < \epsilon < \epsilon_2$, $\epsilon > \epsilon_2$ if $k_3 > \frac{k_4}{1+m}$
$\lambda_1 < \lambda_*^+$	$0 < \epsilon < \epsilon_1^+$, $\epsilon \geq \epsilon_1^+$ if $k_3 > \frac{k_4}{1+m}$	

Table 2: Summary of conditions for rebound derived from Table 1.

We note in Table 2 that in many cases, there are neighbouring intervals with only the boundary point between them missing. Thus, filling in these specific points (which are proved below) gives the

results shown in Table 3. The results in Theorem 5.12 are obtained by converting the eigenvalue ranges in Table 3 into conditions involving the parameters, using that $\lambda_\infty = -k_3(1+h_0)$ and $\lambda_*^+ = -k_3 f_*^+(h_0)$.

Region	$k_1 \leq k_4$	$k_1 > k_4$
$\lambda_*^- \leq \lambda_1 < 0$	$0 < \epsilon < -\lambda_1$	$\epsilon > 0$
$\lambda_\infty \leq \lambda_1 \leq \lambda_*^-$	$0 < \epsilon < \epsilon_3$ or $0 < \epsilon < -\lambda_1$ if $\lambda_1 > -k_3$	$\epsilon > 0$
$\lambda_*^+ \leq \lambda_1 < \lambda_\infty$	$0 < \epsilon < \epsilon_3$	$0 < \epsilon < \epsilon_2$ or $\epsilon > 0$ if $k_3 > \frac{k_4}{1+m}$
$\lambda_1 < \lambda_*^+$	$0 < \epsilon < \epsilon_1^+$ or $\epsilon > 0$ if $k_3 > \frac{k_4}{1+m}$	

Table 3: Summary of final conditions for rebound.

We now consider a number of special cases that fill in the gaps that allow us to go from Table 2 to Table 3.

- $\lambda_*^- < \lambda_1 < 0$, $\epsilon = \epsilon_2$.
  When $\epsilon = \epsilon_2$, then $\lambda_1 = \lambda_4$ and so there are two repeated eigenvalues that are the least negative. With some calculations, it can be seen that the eigenvectors v_1 and v_4 collide (the denominator $(k_3 + \lambda_1)(\epsilon_3 + \lambda_1) + \epsilon_2 k_3 h_0$ in the third and fourth entry vanishes, so the eigenvector has to be rescaled first to see this), hence we are in the case of an eigenvector and a generalised eigenvector. The eigenvector is $v_4 = (0, 0, k_3, k_3 + \lambda_4)$ and generically solutions will align with this eigenvector. In the proof of Lemma B.1, it is shown that $k_3 + \lambda_4 > 0$ when $\epsilon \leq \epsilon_1^-$. Now $\epsilon_2 < \epsilon_1^-$ for $\lambda_1 > \lambda_*^-$ by (61) and this implies that the third and fourth components of v_4 have the same sign. The proof in Lemma B.1 now applies to prove that generically rebound occurs in this case.

- $\lambda_1 = \lambda_*^-$, $\epsilon = \epsilon_2 = \epsilon_3 = \epsilon_1^-$.
  In this case there is a triple eigenvalue: the eigenvector $v_1 = v_4 = v_3 = (0, 0, k_3, k_3 + \lambda_4)$ together with two generalised eigenvectors. Again, generically solutions align with this eigenvector and as above, third and fourth entry have the same sign, thus rebound can be proved via a contradiction argument.

- $\lambda_*^+ < \lambda_1 < \lambda_\infty$, $\epsilon = \epsilon_2$ with $k_3 > \frac{k_4}{1+m}$.
  This is similar to the first case as the two eigenvalues λ_1 and λ_4 coincide when $\epsilon = \epsilon_2$ with an eigenvector and a generalised eigenvector in this case. Generically solutions will align asymptotically with the eigenvector. The proof of Lemma B.2 can now be applied in this case.

- $\lambda_*^+ < \lambda_1 < \lambda_*^-$, $\epsilon = \epsilon_3$ with $k_1 > k_4$ or $k_3 > -\lambda_1$.
  In this case, the real part of the complex pair $\lambda_{3,4}$ equals the eigenvalue λ_1. Approaching the steady state via the complex pair leads to rebound. The proof of Lemma 5.14 also applies with $\epsilon = \epsilon_3$ provided that $\epsilon_3 + \lambda_1 < 0$ and this occurs if $k_3 > -\lambda_1$ and so rebound occurs if the steady state is approached tangent to v_1. If the approach occurs via a linear combination of the three eigenvectors, then clearly rebound will then occur in this case also. Hence we can conclude that rebound will occur generically in this case.

The rebound results that are summarised in Table 3 are shown in Fig. 9. There are two cases shown here, depending on whether or not the curve $\lambda_1 = \lambda_*^+$ intersects the line $k_3 = \frac{k_4}{1+m}$. Since $\lim_{k_1 \to \infty} = -(k_2 + k_4)$ (see (60)), if $k_2 < k_4 \frac{f_*^+ - (1+m)}{1+m}$ then the curve $\lambda_1 = \lambda_*^+$ is always below

$k_3 = \frac{k_4}{1+m}$. Also, as $\lambda_1(k_4, k_2, k_4) = -k_4$ (see (60)), then it can be shown that the line $k_3 = \frac{k_4}{1+m}$ is above the line λ_*^+ at this point, and so the line can never go below the curve as the curve is monotonically increasing with k_1. $\quad\square$

Remarks

- Some of the rebound results that we proved in Lemmas 5.13 and 5.14 hold only generically, and some cases always give rebound. However, for the sake of simplicity, we have not distinguished between these in Theorem 5.12 but simply refer to the existence of generic rebound.

- In Theorem 5.12, we have proved the existence of rebound in a variety of cases. However, it should be noted that we have not proved or disproved the existence of rebound in other parameter regions, and so we do not have results for all possible parameter values.

- From Theorem 5.12, we can conclude that there exists $\epsilon_0 > 0$ such that rebound occurs for all ϵ satisfying $0 < \epsilon < \epsilon_0$ (see Fig. 9). Thus, for all parameter values, rebound occurs for sufficiently small $\epsilon > 0$. This might seem to contradict the result for the limit $\epsilon = 0$ (the no-feedback case) where there is a large region in the parameter plane for which there is no rebound. The explanation is that for those parameter values, the magnitude of the rebound decays to zero as ϵ goes to zero.

6 Generalisations

Thus far, we have considered the basic TMDD model with feedback dynamics which is linear in the feedback moderator F. We now extend this model in two directions. First we extend the TMDD model to allow for more compartments and secondly we consider more general types of feedback dynamics. Below we show that some of the rebound results can be extended to the more general models.

6.1 Generalised TMDD Model With Feedback via a Moderator

In this section we consider a generalisation of the basic TMDD model to more compartments. It is assumed that the receptor is still only in one compartment. In the basic TMDD model, the variable y represents the normalised receptor in this compartment. Now we will assign y to represent either the normalised free receptor (as before) or it could be the normalised total amount of receptor. The normalised ligand-product components (x and z) of the basic TMDD model are generalised to a vector $\mathbf{x}$ that represents the ligand and product in the compartments. The generalised TMDD model with feedback becomes

$$\dot{\mathbf{x}} = \mathbf{f}_1(\mathbf{x}, y) \tag{63}$$
$$\dot{y} = k_3(w - y) + f_2(\mathbf{x}, y) \tag{64}$$
$$\dot{w} = \epsilon(h(y) - w) \tag{65}$$

where $\mathbf{x} \in \mathbb{R}^n$, $n \geq 1$ and $y, w \in \mathbb{R}$. We note that if $\mathbf{f}_1$ depends only on $\mathbf{x}$ but not on y, or if f_2 depends only on y but not on $\mathbf{x}$, then the coupling between $\mathbf{x}$ and y is in one direction only. In this case, the model could not be classed as a generalised TMDD model, although our results below still hold in these cases. We assume that the assumptions (h1)–(h4) on the feedback function $h(y)$ hold and we make the following assumptions on the functions $\mathbf{f}_1$ and f_2:

- There exists an $\mathbf{x}_0 \in \mathbb{R}^n$ such that the equations (63)–(65) have the steady state solution $\mathbf{x} = \mathbf{x}_0$, $y = w = 1$. This implies that $\mathbf{f}_1(\mathbf{x}_0, 1) = \mathbf{0}$ and $f_2(\mathbf{x}_0, 1) = 0$. We also assume that this steady state is globally asymptotically stable.

- In the basic TMMD model, a change in the receptor level while the ligand and product are at baseline does not move the ligand or product away from baseline and we make a similar assumption for this model, i.e., $\mathbf{f}_1(\mathbf{x}_0, y) = 0$ for all $y \geq 0$ which implies that $\frac{\partial \mathbf{f}_1}{\partial y}(\mathbf{x}_0, 1) = 0$. Similarly, the function f_2 is not affected by the receptor dynamics if the ligand and product are at baseline, i.e., $f_2(\mathbf{x}_0, y) = 0$ for all $y \geq 0$ which implies that $\frac{\partial f_2}{\partial y}(\mathbf{x}_0, 1) = 0$. In fact, in both cases, all that we really require is the weaker derivative condition.

- The initial conditions are given by

$$\mathbf{x} = \mathbf{x}_1, \quad y = w = 1$$

  for some $\mathbf{x}_1 \neq \mathbf{x}_0$ with $\mathbf{x}_1$ such that $f_2(\mathbf{x}_1, 1) < 0$. Hence initially the amount of receptor will decrease.

- The Jacobian matrix $D_{\mathbf{x}}\mathbf{f}_1(\mathbf{x}_0, 1)$ has no zero or purely imaginary eigenvalues.

With these assumptions, the Jacobian matrix evaluated at the steady state is given by

$$J(\mathbf{x}_0, 1, 1) = \begin{bmatrix} D_{\mathbf{x}}\mathbf{f}_1(\mathbf{x}_0, 1) & 0 & 0 \\ D_{\mathbf{x}}f_2(\mathbf{x}_0, 1) & -k_3 & k_3 \\ 0 & -\epsilon h_0 & -\epsilon \end{bmatrix}$$

Note that the condition $\frac{\partial \mathbf{f}_1}{\partial y}(\mathbf{x}_0, 1) = 0$ leads to a block lower triangular structure in this matrix. This implies that the eigenvalues of this matrix are the eigenvalues of the Jacobian matrix $D_{\mathbf{x}}\mathbf{f}_1(\mathbf{x}_0, 1)$ and the eigenvalues of the 2×2 bottom right matrix in $J(\mathbf{x}_0, 1, 1)$. The global asymptotic stability of the steady state $(\mathbf{x}_0, 1, 1)$ implies that all eigenvalues have negative or zero real part. Note that the eigenvalues of $D_{\mathbf{x}}\mathbf{f}_1(\mathbf{x}_0, 1)$ do not depend on ϵ and that our final assumption implies that the eigenvalue closest to zero will have strictly negative real part. The 2×2 bottom right matrix is the same as in the basic TMDD model with feedback. We denote its eigenvalues by λ_n and λ_{n-1}, where the eigenvalue λ_n corresponds to λ_4 earlier and λ_{n-1} to λ_3. Thus λ_n is the closest eigenvalue to zero for sufficiently small ϵ.

We define

$$\tilde{\epsilon}_0 = \min\{\epsilon_1^-, \tilde{\epsilon}_2\}$$

where $\tilde{\epsilon}_2$ is the value of ϵ at which the eigenvalue λ_n coincides with the eigenvalue closest to zero of the matrix $D_{\mathbf{x}}f_1(\mathbf{x}_0, 1)$. Recall that ϵ_1^- is the value of ϵ for which λ_n and λ_{n-1} become a complex pair of eigenvalues. We then have the following result.

Theorem 6.1 *If the above assumptions on $\mathbf{f}_1$ and f_2 hold, then rebound occurs generically for $0 < \epsilon < \tilde{\epsilon}_0$.*

Proof The condition $f_2(\mathbf{x}_1, 1) < 0$ ensures that $\dot{y}(0) < 0$, which is required in the proof of Lemma 5.9. The w equation is unchanged in this setting and so Lemma 5.10 also holds. The proof of Lemma B.1 also holds in this setting, although the exceptional case that the trajectory approaches the steady state tangent to a different eigenvector cannot be excluded outright in this more general case, and so we only have a generic result. □

If the bottom right 2×2 matrix in $J(\mathbf{x}_0, 1, 1)$ has complex eigenvalues with real part closest to zero of all the eigenvalues, then again there will be oscillation about the steady state in y as the steady state is approached and so rebound occurs infinitely often but with exponentially decreasing amplitude.

6.2 Generalisation of the Dynamics of the Feedback

In our considerations so far, the feedback dynamics has been linear in the feedback moderator F (or w in the non-dimensional equations), see (14) or (23). In this section we will consider a more general form of feedback, and focus on the case of a small value of the parameter α (ϵ). In particular, we again assume that the receptor equation is described by (13) and describe the dynamics of F by

$$\frac{dF}{dt} = \alpha G(R, F). \tag{66}$$

Non-dimensionalising as before, we obtain a new equation for the feedback variable w ($= F$) given by

$$\dot{w} = \epsilon g(y, w), \tag{67}$$

where

$$g(y, w) = G(R_0 y, w)$$

and ϵ is defined by (25). We make a number of assumptions regarding the function g:

(h1') $g(1, 1) = 0$

(h2') $-g_1 := \frac{\partial g}{\partial y}(1, 1) < 0$

(h3') $-g_2 := \frac{\partial g}{\partial w}(1, 1) < 0$

The assumption (h1') ensures that the equations retain the steady state with $y = w = 1$. Assumption (h2') ensures that when the system is perturbed from the steady state by reducing y, which is what happens when the ligand is injected, then w will increase, resulting in greater production of the receptor. Finally, assumption (h3') ensures that the steady state $w = 1$ of (67), assuming that $y = 1$ is held fixed, is stable.

We now show that some of the results obtained previously also hold in this more general setting. We again start by considering the Jacobian matrix of equations (20)–(22), (67) evaluated at the steady state (57), which is given by

$$\begin{pmatrix} -(k_1 + 1) & \mu k_2 & 0 & 0 \\ \frac{1}{\mu} & -(k_2 + k_4) & 0 & 0 \\ -\frac{1}{\mu} & k_2 & -k_3 & k_3 \\ 0 & 0 & -\epsilon g_1 & -\epsilon g_2 \end{pmatrix} \tag{68}$$

Theorem 6.2 *If assumptions (h1'), (h2') and (h3') hold, and if $g(y, 1) > 0$ for all $y \in [0, 1)$, then there exists $\epsilon_0 > 0$ such that rebound occurs in the model given by equations (20)–(22), (67) for all ϵ satisfying $0 < \epsilon < \epsilon_0$.*

Proof We note that assumptions (h1') and (h2') imply that $g(y, 1) > 0$ for $y < 1$ sufficiently close to 1. The extra assumption requires that this holds for all $y \in [0, 1)$.

The proofs of Lemmas 5.9, 5.10 and Lemma B.1 can easily be adapted to this modified model. In particular, for Lemma 5.9, $\dot{w}(0) = 0$ as before and in this case, we have

$$\ddot{w}(0) = -\frac{\epsilon}{\mu} \frac{\partial g}{\partial y}(1, 1) = \frac{\epsilon}{\mu} g_1 > 0$$

and so Lemma 5.9 holds.

For Lemma 5.10, we note that

$$\dot{w}|_{w=1} = \epsilon g(y, 1) > 0$$

using our stated assumption. Combined with Lemma 5.9, this implies that $w(\tau)$ cannot touch or cross the line $w = 1$ when $y(\tau) < 1$. The second part of the proof of Lemma 5.10 shows by contradiction that $y(\tau) = w(\tau) = 1$ is impossible and the same proof holds in this case with h_0 replaced by g_1.

The Jacobian matrix associated with equations (49) and (67) (analogous to (51)) is given by

$$J_0 = \begin{bmatrix} -k_3 & k_3 \\ -\epsilon g_1 & -\epsilon g_2 \end{bmatrix} \tag{69}$$

which has eigenvalues $\lambda_3 = -k_3$, $\lambda_4 = 0$ when $\epsilon = 0$ as previously. These two eigenvalues collide when the discriminant of the characteristic polynomial is zero, and this first occurs when

$$\epsilon = \epsilon_1^- = \frac{k_3}{g_2^2}\left(2g_1 + g_2 - \sqrt{4g_1(g_1 + g_2)}\right).$$

With this definition of ϵ_1^-, we claim that Lemma B.1 holds also. To see this, we note that the eigenvector v_4 given by (59) is the same in this case, but with λ_4 as the eigenvalue of (69) that is closest to zero. We now have

$$k_3 + \lambda_4 = \frac{1}{2}\left(k_3 - \epsilon g_2 + \sqrt{(k_3 - \epsilon g_2)^2 - 4\epsilon k_3 g_1}\right).$$

It is easily verified that $k_3 > \epsilon_1^- g_2$ and this implies that $k_3 + \lambda_4 > 0$ for $0 < \epsilon < \epsilon_1^-$. Thus, if $0 < \epsilon < \epsilon_1^-$, then the third and fourth components of the eigenvector v_4 have the same sign and the proof of Lemma B.1 again implies that rebound occurs.

The second part of the proof of Lemma B.1 considered the non-generic cases of the trajectory approaching the steady state tangent to one of the other eigenvectors. For the eigenvector v_3 in (59), we note that it is easily verified that $k_3 + \lambda_3 > 0$ and so the same arguments apply in this case. The argument for the eigenvector v_2 is unchanged, while for v_1, the requirement for the third and fourth components of the eigenvector to be the same is $\epsilon g_2 + \lambda_1 < 0$ and it is easily verified that this holds.

If $\epsilon = \epsilon_2$ corresponds to the point at which the eigenvalues $\lambda_1 = -k_4$ and λ_2 coincide, then the conditions of Lemma B.1 hold with $\epsilon_0 = \min(\epsilon_1^-, \epsilon_2)$ and so we conclude that rebound therefore occurs for all ϵ satisfying $0 < \epsilon < \epsilon_0$. □

This result shows that for this generalised model, rebound always occurs for sufficiently small $\epsilon > 0$ even if there is no rebound without feedback ($\epsilon = 0$), as is the case for the original TMDD model with feedback moderator.

Clearly, if the bottom right 2×2 matrix in (68) has complex eigenvalues which have real part closest to zero of all the eigenvalues, then there will again be oscillation about the steady state in y and so rebound occurs infinitely often but with exponentially decreasing magnitude.

7 Applications

We now consider two applications of these results to two different models. The first is the standard TMDD model with feedback. The second is a more complicated model of the effect of efalizumab on patients with psoriasis. This model is an application of the generalizations in Section 6.

7.1 Example 1

We consider the TMDD equations including feedback with moderator given by (11)–(14). We use parameter values for the IgE mAb omalizumab [20, 1], namely $k_{e(L)} = 0.024$ day^{-1}, $k_{e(P)} = 0.201$ day^{-1}, $k_{out} = 0.823$ day^{-1}, $R_0 = 2.688$ nM, $k_{off} = 0.900$ day^{-1}, $k_{on} = 0.592$ (nM day)$^{-1}$, $k_{in} = k_{out}R_0 = 2.212224$ nM day^{-1}, and initial drug dose $L_0 = 14.8148$ nM. These parameter values have $k_{e(L)} < k_{e(P)}$ and so no rebound will occur when there is no feedback or in the case of direct feedback for any
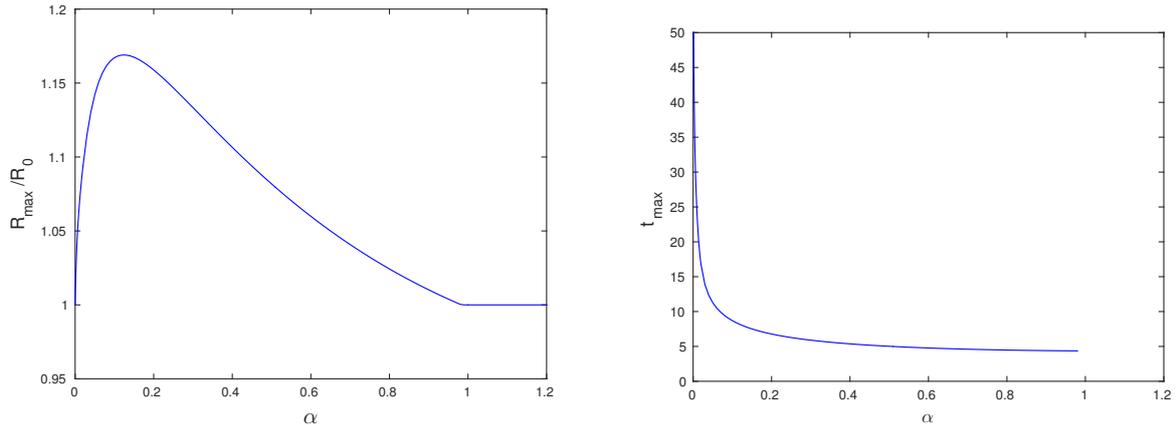
Figure 10: Left: A plot of the relative magnitude of rebound $R_{\max}/R_0$ for feedback with a moderator and the mainly linear feedback function (34) with $H_0 = 1$ for a range of values of α. Right: A plot of the time at which the maximum rebound occurs.

feedback function $H(R)$. This follows directly from Theorem 2.1, Corollary 4.4, and Theorem 4.10 since $k_{\mathrm{e(L)}} < k_{\mathrm{e(P)}}$ implies that the parameter values are in the left green region in Figs 3, 4, and 6.

However if there is feedback via a moderator then Theorem 5.12 implies that rebound occurs for all sufficiently small $\alpha > 0$, even though there is no rebound at $\alpha = 0$. The parameters for this example give

$$\lambda_1 = -0.084, \quad \lambda_4 = 0.126, \quad k_3 = 0.517.$$

Thus Theorem 5.12 gives the parameters are in the upper left region of Fig. 9 and that rebound will happen for $\alpha < 0.135$ (using $\alpha = \epsilon/0.625$). However, this is a lower bound on the region of rebound. To illustrate this, we have used the mainly linear feedback function $H(R)$ in (34) with $H_0 = 1$. The magnitude of the rebound is sufficiently small in this case that a nonlinear function $H_1(R)$ is not required. The maximum rebound is plotted in Fig. 10 for a range of values of α which does indeed show that there is rebound for all $\alpha > 0$ and sufficiently small. For this example, the rebound ends at approximately $\alpha = 0.98$. In this case, a local maximum for R persists for larger values of α, but with $R_{\max}/R_0 < 1$. The value of time $t_{\max}$ at which the maximum rebound occurs is also shown in Fig. 10. We note that as $\alpha \to 0$ then $t_{\max} \to \infty$ and so the time of maximum rebound moves to infinity as the magnitude decreases. The same mechanism does not apply when the rebound disappears around $\alpha = 1$ since a local maximum persists in this case but at values lower than baseline, as already discussed.

We have numerically sampled larger values of α and there is no indication of further rebound occurring. Of course we know that when $\alpha \to \infty$, the feedback with moderator becomes the direct feedback case, for which there is no rebound in this example.

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7.2 Example 2

In a study on the effect of efalizumab on patients with psoriasis [15], rebound was reported in some patients. The model equations proposed in [15] are

$$\frac{dX_{\text{sc}}}{dt} = -k_a X_{\text{sc}},$$

$$\frac{dX_1}{dt} = -(k_{10} + k_{12})X_1 + k_{21}X_2 - \frac{V_{\text{m}}X_1}{K_{\text{mc}}V_{\text{c}} + X_1} + F_a k_a X_{\text{sc}},$$

$$\frac{dX_2}{dt} = k_{12}X_1 - k_{21}X_2,$$

$$\frac{dX_3}{dt} = X_4 - k_{30}X_3 - \frac{V_{\text{m2}}X_3 X_1}{K_{\text{mc}}V_{\text{c}} + X_1},$$

$$\frac{dX_4}{dt} = k_{\text{off}}\left[k_{03\text{max}}\left(\frac{K_{\text{mc03}}}{K_{\text{mc03}} + X_3}\right) - X_4\right],$$

where X_{sc}, X_1 and X_2 are the amounts of efalizumab in the depot, central and peripheral compartments respectively, X_3 is the total %CD11a on the surface of each T cell and X_4 is the production rate of %CD11a to the T cell surface. In this section we will show that this model can be written as a generalised model with feedback as studied in Section 6.1. Stricly speaking, this model is not a TMDD model as the efalizumab equations decouple from the %CD11a equations. As indicated in [15], one could use $\frac{X_1 X_3}{K_{\text{mc}}V_{\text{c}} + X_1}$ instead of $\frac{X_1}{K_{\text{mc}}V_{\text{c}} + X_1}$ in the PK equations and have a more TMDD-like model. In [15], the simpler model was chosen as there was no difference in the fit of the data. If one would use the more complicated model, the parameters would need to be fitted again. So we focus in this section on the same model as in [15]. However, our analysis also applies to the more complicated model and the observed rebound would again be due to the slow feedback response.

In this model, X_3 is the total receptor, free and bound, and X_4 is the feedback moderator. The plots in Fig. 3 of [15] show rebound in the total receptor, which of course is not the same as rebound in the free receptor. However, we first show that this model fits with the generalised model that we considered in Section 6.1 and we will then discuss the issue of rebound in the free receptor. To put this model into our framework, we take $\mathbf{X} = [X_{\text{sc}}, X_1, X_2]$, $Y = X_3$ and $F = X_4/k_{03\text{max}}$. The differential equations for Y and F are then given by

$$\frac{dY}{dt} = k_{03\text{max}}F - k_{30}Y + F_2(\mathbf{X}, Y), \tag{70}$$

$$\frac{dF}{dt} = k_{\text{off}}\left(\frac{K_{\text{mc03}}}{K_{\text{mc03}} + Y} - F\right), \tag{71}$$

where

$$F_2(\mathbf{X}, Y) = -\frac{V_{\text{m2}}Y X_1}{K_{\text{mc}}V_{\text{c}} + X_1}.$$

The non-dimensional form of these equations are precisely in the form of equations (63)–(65). These equations have the unique steady state in the region of interest given by

$$X_{\text{sc}}^0 = X_1^0 = X_2^0 = 0, \quad Y^0 = \frac{k_{03\text{max}}F^0}{k_{30}},$$

where F^0 is the positive root of the quadratic equation

$$k_{03\text{max}}F^2 + k_{30}K_{\text{mc03}}F - k_{30}K_{\text{mc03}} = 0. \tag{72}$$

Substituting $F = 1 + \delta F$ into (72) gives a quadratic equation for δF which has two negative roots, and this implies that $F^0 \in (0,1)$. It is then easily verified that all the conditions stated in Section 6.1 on the functions F_1 and F_2 hold. The particular function H that is used in this model is given by

$$H(Y) = \frac{K_{\text{mc03}}}{K_{\text{mc03}} + Y}.$$

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We non-dimensionalise, considering only Y and F, by defining the new variables

$$y = \frac{Y}{Y^0}, \quad w = \frac{F}{F^0}, \quad \tau = V_{\mathrm{m}2}t.$$

The non-dimensional form of equations (70)–(71) is then

$$\begin{aligned}
\dot{y} &= k_3(w - y) + f_2(\mathbf{x}, y), \\
\dot{w} &= \epsilon(h(y) - w),
\end{aligned}$$

where dot denotes differentiation with respect to τ, $\mathbf{x}$ is the (unspecified) non-dimensional form of $\mathbf{X}$, and

$$k_3 = \frac{k_{30}}{V_{\mathrm{m}2}}, \quad \epsilon = \frac{k_{\mathrm{off}}}{V_{\mathrm{m}2}}, \quad f_2(\mathbf{x}, y) = \frac{F_2(\mathbf{X}, Y^0 y)}{V_{\mathrm{m}2}}, \quad h(y) = \frac{1}{F^0 + (1 - F^0)y}.$$

Note that we have used (72) in deriving $h(y)$. All of our assumptions (h1)–(h4) on the function $h(y)$ are now satisfied and

$$h_0 = -h'(1) = 1 - F^0 > 0$$

since $F^0 \in (0, 1)$. We tried to reproduce the numerical results shown in Fig. 3B (dashed line) of [15] but found that this was only possible by changing k_{off} from the stated value of 0.00154 day^{-1} to 0.0154 day^{-1} (the smaller value of k_{off} did show rebound, but with a smaller magnitude of about 110% of baseline). Moreover, the authors in [15] report an affinity of 0.033μg/ml, which is $(0.033/150000) \times 10^{-3}$M $= 2 \times 10^{-10}$M, or 200pM. If we use the reported k_{off} of 0.00154 day^{-1} and assume a typical k_{on} (see [3]) of 0.592 nM^{-1}/day, the ratio $k_{\mathrm{off}}/k_{\mathrm{on}} \approx 2$pM, i.e. 100-fold more potent than reported. A 10-times higher k_{off} still does not give the right affinity but at least it is closer. The values of the other parameters used, from Tables II and III of [15], are $k_{\mathrm{a}} = 0.242$ day^{-1}, $k_{10} = 0.114$ day^{-1}, $k_{12} = 0.097$ day^{-1}, $k_{21} = 0.193$ day^{-1}, $k_{30} = 0.444$ day^{-1}, $V_{\mathrm{c}} = 64.3$ mL/kg, $V_{\mathrm{m}} = 26.9$ μg/mL, $V_{\mathrm{m}2} = 2.16$ day^{-1}, $K_{\mathrm{mc}} = 0.033$ μg/mL, $F_{\mathrm{a}} = 0.564$, $k_{03\mathrm{max}} = 334$ %CD11a/day. The plot of the total receptor X_3 in this case is the solid blue line shown on the left in Fig. 11.
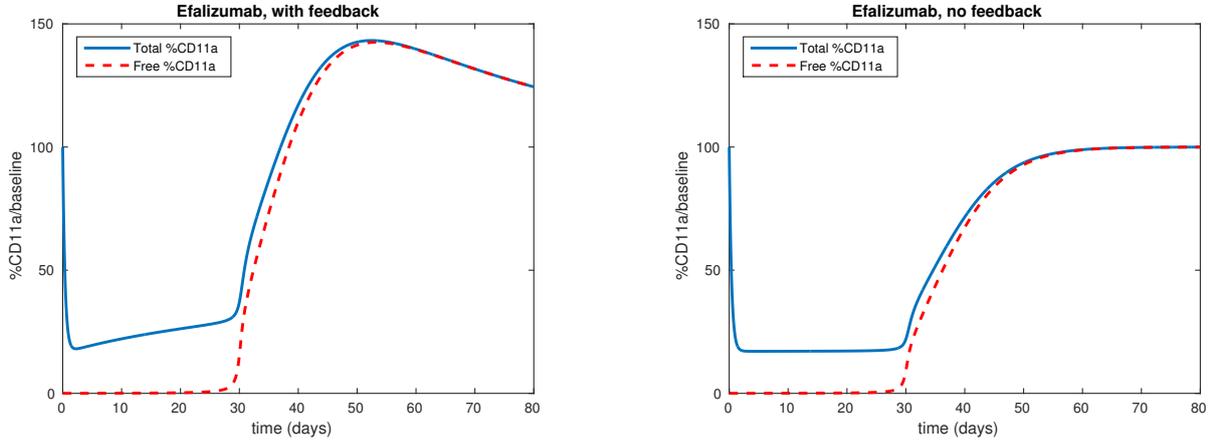

Figure 11: The total and free %CD11a relative to baseline after a single 3mg/kg intravenous dose of efalizumab. On the left is the plot with feedback turned on and rebound can be observed. On the right is the plot without feedback and no rebound occurs.

To find out how feedback influences the model, we ran a simulation of the model when the feedback was turned off, i.e., we took X_4 to be constant at its baseline value. As can be seen on the right in Fig. 11, without feedback there is no rebound. So clearly the rebound in this example is caused by the feedback.

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Next we will interpret these results using our analysis. Using the parameter values in [15], but the modified value of k_{off}, we find that

$$\epsilon = 7.13 \times 10^{-3},$$

and since this is so small, we expect that the eigenvalue closest to zero of the Jacobian matrix evaluated at the steady state will come from the lower 2×2 diagonal block. This is indeed the case since the eigenvalue closest to zero of this block is -1.37×10^{-2} while the eigenvalue closest to zero from the remaining 3×3 block $D_{\mathbf{x}}\mathbf{f}_1(0,1)$ is -8.87×10^{-2}. Thus, in the notation of Theorem 6.1, we have $0 < \epsilon < \epsilon_0$ and so this Theorem says that we generically have rebound in the total receptor X_3, as confirmed in Fig. 11 and by the numerical results in [15].

The two eigenvalues of the lower 2×2 diagonal block collide at $\lambda = -0.122$ when $\epsilon = 0.0390$, but since there are eigenvalues of $D_{\mathbf{x}}\mathbf{f}_1(0,1)$ that are closer to zero than this, we would not expect to see oscillatory convergence to the steady state in this model by simply increasing ϵ (k_{off}), as described in Lemma B.3.

Finally, we recall that the variable X_3 in the above model is the total of the free and bound receptor. The free receptor on T cells is given by $K_{\mathrm{mc}}V_{\mathrm{c}}X_3/(K_{\mathrm{mc}}V_{\mathrm{c}} + X_1)$ [15] and we note that if $X_1 = 0$, then the free receptor is the same as the total receptor. Now X_1 is the amount of efalizumab in the central compartment and while this is present, the total receptor X_3 is kept at a low level. However, once X_1 has been almost depleted, then X_3 starts to rise again and rebound occurs. Thus, X_1 is very low in the phase of the dynamics in which rebound in X_3 occurs, and this implies that the free receptor is very similar to X_3 and so will also rebound. The free receptor for the above example is also shown in Fig. 11 as the red dashed line and it can clearly be seen that this also has rebound which is very similar to that for the total receptor as anticipated.

8 Conclusions and Discussion

In this paper we have extended our previous work on rebound in the basic TMDD model by including feedback. The feedback is negative, thus the rate of receptor production increases when the concentration drops below baseline (and vice versa). We modelled the feedback with an additional dynamic equation for the feedback moderator. If the feedback responds very fast, a quasi equilibrium approximation can be used and the feedback can be incorporated into the synthesis term itself.

We have shown that rebound is more likely to occur if feedback is present and that the likelihood of feedback occurring depends on the response speed of the feedback. If the feedback responds very slowly, there will always be rebound. In the psoriasis example presented in Section 7.2, it can be seen that the rebound can be significant, an increase of over 40% of baseline is observed. The TMDD example in Section 7.1 shows that this is not always the case. In general, it is a challenge to describe the main parameters that influence the magnitude of the rebound. One important parameter is obviously the response speed of the feedback. If the response of the feedback goes to zero, then we converge to the case without feedback. Thus for most values of the elimination parameters the magnitude of the rebound will diminish if the response of the feedback slows down to zero and in the limit the rebound will have gone. The exceptions are of course those elimination parameter values for which rebound is observed if no feedback is present (the elimination rate of the antibody-protein complex is less than both the elimination rate of the antibody and the elimination rate of the protein $(k_{\mathrm{e(P)}} < k_{\mathrm{e(L)}}$ and $k_{\mathrm{e(P)}} < k_{\mathrm{out}}))$.

At first sight, it might be surprising that there is always rebound for a very slow feedback response rate $(0 < \alpha \ll 1)$, while there are large regions without rebound when the feedback moderator does not change and there is no feedback $(\alpha = 0)$. However, it can be seen that, in the absence of feedback dynamics $(\alpha = \epsilon = 0)$, a larger initial condition for the moderator w will lead to a steady state with $y = w$ which is above the baseline. Furthermore, Lemma 5.9 shows that when the moderator starts at baseline, ligand is added and feedback is turned on $(\alpha, \epsilon > 0)$, then the moderator w initially increases to above its baseline value, albeit on a slow time scale since α is small. However, this implies that

w will also only decrease back to its steady state value of 1 on a slow time scale. While $w > 1$, y converges towards w on a faster time scale and will therefore itself exceed 1, resulting in rebound, followed by a slow relaxation of y and w back to their final steady state.

For moderately slow feedback response rates, the rebound is expected to be maximal, while for very fast feedback responses ($\alpha \gg 1$), the rebound will be approximated by the direct feedback limit. The results for the direct feedback model are qualitatively very similar to the results without feedback: the existence or non-existence of rebound again depends only on the three elimination rates $k_{e(L)}$, k_{out} and $k_{e(P)}$. An extra feature now is that the characteristics of the feedback function $H(R)$ play a role as well if $k_{e(L)} > k_{e(P)}$. In particular, in both the direct feedback and no feedback models there is no rebound if the elimination rate of the ligand is less than the elimination rate of the antibody-protein complex ($k_{e(L)} < k_{e(P)}$). Furthermore, in both models there will be rebound if the elimination rate of the antibody-protein complex is less than both the elimination rate of the antibody and the elimination rate of the protein ($k_{e(P)} < k_{e(L)}$ and $k_{e(P)} < k_{out}$). However, if $k_{out} < k_{e(P)} < k_{e(L)}$, then the presence of feedback will increase the region in which rebound occurs. The details depend on the characteristics of the feedback function $H(R)$ and can be found in Theorem 4.10. Again, in both models, the association and dissociation rate parameters k_{on} and k_{off} do not influence the occurrence of rebound, though they will play a role in the magnitude of the rebound if it occurs. There are some preliminary observations that a large k_{on} will dampen rebound, but this needs further work.

In the general feedback model, we have focused on proving the existence of rebound. In general, it is challenging to prove the non-existence of rebound as it has to be shown that the receptor is bounded by its baseline for all time. We expect that no rebound will occur for $k_{e(L)} < k_{e(P)}$ or small k_{out} for a sufficiently fast response rate (α large), but we have not yet obtained analytical results about this. The fast response is a singular limit and it can be shown that the dynamics has to be close to the limiting case. Thus if any rebound did happen, it would have to be very small.

Furthermore, we have also shown in this paper that most of the results obtained on rebound in the model with slow feedback can be extended to more general systems of TMDD equations such as equations with more compartments. We illustrated this with the example of psoriasis, where rebound is predicted by the model and underpins the observations in patients (see Section 7).

It must be acknowledged that during target discovery and validation, knowledge of the quantitative relationship between the target neutralisation and the nature of the negative feedback is often not known. However, using prior knowledge on the target of interest and similar targets, the model can be used to evaluate the possibility of a feedback and its potential magnitude, and thus confirm the suitability of the target.

A   Proofs of Invariance and Stability Results

Proof of Lemma 4.1 The phase space is bounded by the three planes $x = 0$, $y = 0$ and $z = 0$. If the vector field given by the equations (16)–(18) on these planes is directed inside or is tangent to the octant, then the given region is invariant [18].

Equations (16) and (17) are the same as in the simple TMDD model, and so the direction of the vector field on the planes $x = 0$ and $z = 0$ was established in the proof of Lemma 2.1 of [2] (see also [17]).

The only difference in this case is in the derivative normal to the plane $y = 0$, which is now

$$\dot{y}|_{y=0} = k_3 h(0) + k_2 z.$$

From assumption (h3), we see that $h(0) > 1$ and so $\dot{y}|_{y=0} > 0$ when $z \geq 0$ as before. The rest of the proof is unchanged.   □

Proof of Lemma 4.2 Setting the derivatives to zero in (16) and (17) and solving for x and z, the only valid solution possible is $x = z = 0$. Substituting these into (18) with the derivative set to zero,

we obtain
$$k_3(h(y) - y) = 0.$$

By assumptions (h2) and (h3), $y = 1$ is the unique non-negative solution of this equation. Thus the unique steady state in the positive octant of equations (16)–(18) is given by $x = z = 0$ and $y = 1$.

To show the global stability, we note that $u = x + \mu z$ again decreases monotonically to zero using the same approach as in Lemma 2.3 of [2] and since x and z must be non-negative, we conclude that $\lim_{\tau \to \infty} x(\tau) = \lim_{\tau \to \infty} z(\tau) = 0$, as for the original TMDD model. Similar to the original TMDD model, the y-axis, i.e, the manifold $x = 0$, $z = 0$, forms an invariant manifold as $\dot{x} \mid_{x=0,\,z=0} = 0$ and $\dot{z} \mid_{x=0,\,z=0} = 0$. On this invariant manifold, the y-dynamics is given by

$$\dot{y} = k_3(h(y) - y).$$

From the assumptions on the function h, it follows immediately that for $0 < y < 1$, $h(y) - y > 1 - y > 0$; for $y > 1$, $h(y) - y < 1 - y < 0$; and for $y = 1$, we have $h(y) - y = 1 - 1 = 0$. Thus $y = 1$ is an attractor for the positive y-axis. Intuitively, these two observations together give that the steady state $(x, y, z) = (0, 1, 0)$ is a global attractor for the positive octant. To make this formal, we consider the y-dynamics in a neighbourhood of the y-axis.

Let $\epsilon > 0$. Since $\lim_{\tau \to \infty} x(\tau) = \lim_{\tau \to \infty} z(\tau) = 0$, then there is some $T > 0$ such that for $\tau > T$, we have

$$0 \le x(\tau) \le \epsilon \mu k_3 \quad \text{and} \quad 0 \le z(\tau) \le \epsilon k_3 / k_2.$$

Thus for $\tau > T$, the y-dynamics (18) can be estimated by

$$k_3 \left[h(y) - (1 + \epsilon)y \right] \le \dot{y} \le k_3 \left[h(y) + \epsilon - y \right].$$

If $y(T) > 1$, then $y(\tau) \ge 1$ on some τ-interval beyond T. In this interval we have $h(y(\tau)) \le 1$, thus

$$\dot{y} \le k_3 \left[1 + \epsilon - y \right] \quad \text{or} \quad \frac{d}{d\tau} \left[e^{k_3 \tau} (y - (1 + \epsilon)) \right] \le 0.$$

Thus

$$y(\tau) \le (1 + \epsilon) + e^{-k_3(\tau - T)} (y(T) - (1 + \epsilon)). \tag{73}$$

Similarly, if $y(T) < 1$, then $y(\tau) \le 1$ on some τ-interval beyond T. In this interval we have $h(y(\tau)) \ge 1$, thus

$$\dot{y} \ge k_3 \left[1 - (1 + \epsilon)y \right] \quad \text{or} \quad \frac{d}{d\tau} \left[e^{k_3(1+\epsilon)\tau} \left(y - \frac{1}{1 + \epsilon} \right) \right] \ge 0$$

Thus

$$y(\tau) \ge 1 - \frac{\epsilon}{1 + \epsilon} + e^{-k_3(1+\epsilon)(\tau - T)} \left(y(T) - \frac{1}{1 + \epsilon} \right). \tag{74}$$

If $y(\tau) > 1$ for all $\tau \ge T$, then the estimate (73) is valid for all $\tau > T$ and we can conclude that there is some $T_1 > 0$ such that $1 \le y(\tau) \le 1 + 2\epsilon$ for $\tau > T_1$. Similarly, if $y(\tau) < 1$ for all $\tau \ge T$, then the estimate (74) is valid for all $\tau > T$ and we can again conclude that there is some $T_1 > 0$ such that $1 \ge y(\tau) \ge 1 - 2\epsilon$ for $\tau > T_1$

However, it is also possible that there is some $\widehat{T} \ge T$ such that $y(\widehat{T}) = 1$. In this case, (73) gives that if $y(\tau) \ge 1$ in some interval $[\widehat{T}, \widehat{T}_1]$, then we can estimate for τ in this interval

$$1 \le y(\tau) \le (1 + \epsilon) + e^{-k_3(\tau - T)} \left(y(\widehat{T}) - (1 + \epsilon) \right) = (1 + \epsilon) + e^{-k_3(\tau - T)} (1 - (1 + \epsilon)) \le (1 + \epsilon)$$

And similarly, if $y(\tau) \le 1$ in some interval $[\widehat{T}, \widehat{T}_1]$, then (74) gives that for τ in this interval, we have

$$1 \ge y(\tau) \ge 1 - \frac{\epsilon}{1 + \epsilon} + e^{-k_3(1+\epsilon)(\tau - T)} \left(y(\widehat{T}) - \frac{1}{1 + \epsilon} \right) = 1 - \frac{\epsilon}{1 + \epsilon} + e^{-k_3(1+\epsilon)(\tau - T)} \left(1 - \frac{1}{1 + \epsilon} \right) \ge 1 - \epsilon$$

Thus if there is some $\widehat{T} \geq T$ such that $y(\widehat{T}) = 1$, then the interval $[\widehat{T}, \infty)$ can be divided in intervals $[a, b)$ such that $y(a) = 1 = y(b)$ and $y(\tau) - 1$ is either negative or positive for $\tau \in (a, b)$. On each of those intervals, one of the two estimates above can be used and hence we get that $|y(\tau) - 1| < \epsilon < 2\epsilon$ for all $\tau \geq \widehat{T}$.

Combining the three cases, we can conclude that for all $\epsilon > 0$, there is some $\widetilde{T} > 0$ such that $|y(\tau) - 1| < \epsilon < 2\epsilon$ for all $\tau > \widetilde{T}$. Thus $\lim_{\tau \to \infty} y(\tau) = 0$ and we can conclude that all trajectories must converge to the steady state $(x, y, z) = (0, 1, 0)$ as $t \to \infty$. $\quad\square$

Proof of Lemma 5.2 From (53) we note that

$$
\begin{aligned}
\lambda_4 &= \frac{1}{2}(k_3 + \epsilon)\left(-1 + \sqrt{1 - \frac{4\epsilon k_3(1 + h_0)}{(k_3 + \epsilon)^2}}\right) = \frac{1}{2}(k_3 + \epsilon)\left(\frac{\frac{4\epsilon k_3(1 + h_0)}{(k_3 + \epsilon)^2}}{-1 - \sqrt{1 - \frac{4\epsilon k_3(1 + h_0)}{(k_3 + \epsilon)^2}}}\right) \\
&= \frac{\frac{2\epsilon k_3(1 + h_0)}{(k_3 + \epsilon)}}{-1 - \sqrt{1 - \frac{4\epsilon k_3(1 + h_0)}{(k_3 + \epsilon)^2}}}
\end{aligned}
$$

Taking the limit in this expression as $\epsilon \to \infty$ gives the stated result. $\quad\square$

Proof of Lemma 5.3 We define the Lyapunov function

$$
V(y, w) = \frac{1}{k_3}\int_1^y (1 - h(y'))\, dy' + \frac{1}{2\epsilon}(w - 1)^2.
$$

For the steady state to be globally asymptotically stable, we have to prove that V is positive definite and continuous and that $\dot{V}$ is negative definite and continuous relative to the steady state $y = w = 1$ on the whole of the phase space [9].

By hypothesis (h4), V and $\dot{V}$ are continuous. To show that V is positive definite, we note that $V(1, 1) = 0$ and that hypothesis (h3) implies that

$$
1 - h(y) \begin{cases} < 0 & \text{if } y < 1 \\ = 0 & \text{if } y = 1 \\ > 0 & \text{if } y > 1 \end{cases} \tag{75}
$$

Thus,

$$
\int_1^y (1 - h(y'))\, dy' \begin{cases} > 0 & \text{if } y \neq 1 \\ = 0 & \text{if } y = 1 \end{cases}
$$

and so $V(y, w) > 0$ if $(y, w) \neq (1, 1)$.

Differentiating V with respect to τ gives

$$
\begin{aligned}
\dot{V} &= \frac{1}{k_3}(1 - h(y))\dot{y} + \frac{1}{\epsilon}(w - 1)\dot{w} \\
&= (1 - h(y))(w - y) + (w - 1)(h(y) - w) \\
&= -(w - 1)^2 - (1 - h(y))(y - 1).
\end{aligned}
$$

We note that (75) implies that $(1 - h(y))(y - 1) > 0$ if $y \neq 1$ and so $\dot{V} < 0$ if $(y, w) \neq (1, 1)$. Also, $\dot{V}(1, 1) = 0$. Thus, $\dot{V}$ is negative definite.

As all the above conditions are satisfied, we conclude that the steady state $(y, w) = (1, 1)$ is globally asymptotically stable. $\quad\square$
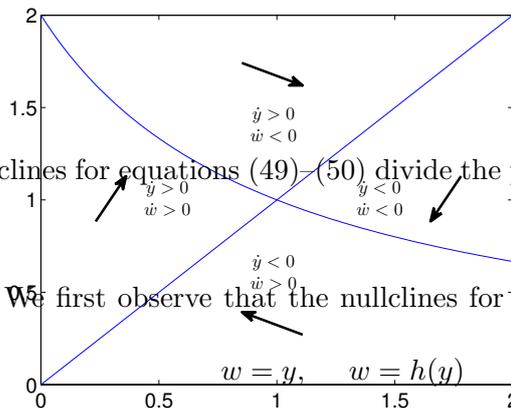
Figure 12: The two nullclines for equations (49)–(50) divide the phase plane into four regions in which $\dot{y}$ and $\dot{w}$ have one sign.

Proof of Lemma 5.4 We first observe that the nullclines for equations (49)–(50) are given by the two curves

$$w = y, \qquad w = h(y)$$

which intersect at the steady state $y = w = 1$. These two curves divide the phase plane into four regions in which the signs of $\dot{y}$ and $\dot{w}$ do not change, as illustrated in Fig. 12.

We note from (55) that

$$
\begin{aligned}
k_3 - \epsilon_1^{\pm} &= k_3 \left(-2h_0 \mp \sqrt{4h_0(1 + h_0)} \right) \\
&= -2k_3 h_0 \left(1 \pm \sqrt{1 + \frac{1}{h_0}} \right)
\end{aligned}
$$

and it follows from this that

$$\epsilon_1^- < k_3 < \epsilon_1^+. \tag{76}$$

We next observe from (52) and (53) that

$$\lambda_i + k_3 = \frac{1}{2} \left(k_3 - \epsilon \pm \sqrt{(k_3 - \epsilon)^2 - 4\epsilon k_3 h_0} \right), \quad i = 3, 4$$

and it is easily verified from this that

$$\lambda_i + k_3 \begin{cases} < 0 & \text{if } \epsilon > k_3, \\ > 0 & \text{if } \epsilon < k_3, \end{cases} \quad i = 3, 4. \tag{77}$$

We now consider the two eigenvectors associated with the eigenvalues λ_3 and λ_4, which are given by (54). The slopes of the linearised manifolds $E_{3,4}$ associated with the eigenvectors $v_{3,4}$ in the (y, w) plane are $(k_3 + \lambda_i)/k_3$, $i = 3, 4$. We now consider separately the two ranges in ϵ for which the eigenvalues are real.

If $0 < \epsilon < \epsilon_1^-$, then from (76), we also have that $\epsilon < k_3$. From (77), this implies that $\lambda_i + k_3 > 0$, $i = 3, 4$ and so the slopes of E_3 and E_4 are both positive and as $\lambda_{3,4} < 0$, the slopes are also both less than one. Since $\lambda_3 < \lambda_4$, the manifold associated with λ_4 has the steepest slope. This situation is shown in Fig. 13(a).

Since λ_4 is the eigenvalue closest to zero, a generic trajectory will approach the steady state tangent to the linearised manifold E_4. Now the global one-dimensional stable manifolds which are tangent to E_3 and E_4 at the steady state are invariant, and hence no trajectory can cross them. Thus, all initial
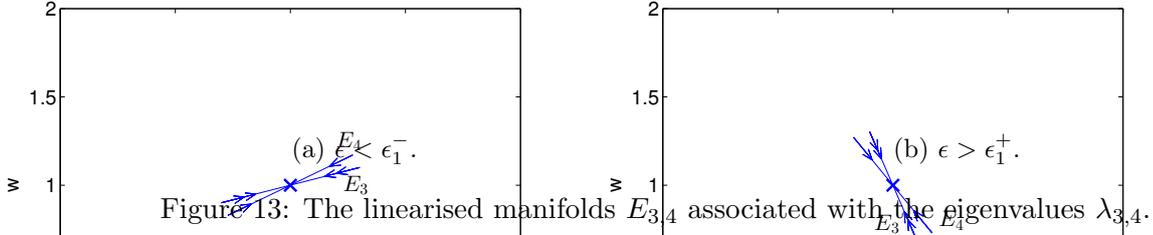
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(a) $\epsilon < \epsilon_1^-$.

(b) $\epsilon > \epsilon_1^+$.

Figure 13: The linearised manifolds $E_{3,4}$ associated with the eigenvalues $\lambda_{3,4}$.

conditions with $y(0) < 1$ but above the global stable manifold tangent to E_3 will rotate round the fixed point in a clockwise direction, as indicated in Fig. 12, and will approach the steady state tangent to E_4 from the right of the steady state for which $y > 1$. Now this global manifold must itself spiral in to the steady state in a clockwise direction as shown in Fig. 12 and so cannot cross the line $w = 1$, $0 \le y \le 1$, at least not unless it circles the steady state first. Thus, all initial conditions satisfying $w(0) > 1$, $y(0) < 1$ will give rise to trajectories with rebound as they approach the steady state, as will initial conditions with $w(0) < 1$, $y(0) < 1$ that lie above the global manifold that is tangent to E_3 at the steady state.

When $\epsilon > \epsilon_1^+$, an analogous process can be followed to show that E_3 and E_4 both have negative slopes which are less than $h'(1)$ and this is shown in Fig. 13(b). Again, any initial conditions to the right of the global manifold tangent to E_3 will rotate around the steady state in a clockwise direction and will approach the steady state with $y > 1$, giving rebound. As $\epsilon \to \infty$, we have already seen that $\lambda_3 \to -\infty$, and in this case, E_3 becomes vertical and so the region of initial conditions that gives rise to rebound, near to the steady state at least, becomes smaller as ϵ increases. Thus, for all $\epsilon \in (\epsilon_1^+, \infty)$, there are always some initial conditions with $w(0) > 1$, $y(0) < 1$ which will give rise to rebound, as claimed.

We note that as the global stable manifolds may spiral in to the steady state in a clockwise direction, we cannot say whether initial conditions satisfying $w(0) < 1$, $y(0) < 1$ are above or below this global manifold, and so we cannot comment on the existence or non-existence of rebound in this case.

When $\epsilon = \epsilon_1^\pm$, the eigenvalues λ_3 and λ_4 collide as well as their eigenvectors. Instead of two eigenvectors, there is now one eigenvector $v_3 = v_4$ and a generalised eigenvector. All solutions will approach the steady state via the eigenvector, see [9]. The analysis for the approach to the steady state for the case $0 < \epsilon < \epsilon_1^-$ can be extended to the case $\epsilon = \epsilon_1^-$ and similarly, the case $\epsilon > \epsilon_1^+$ can be extended to $\epsilon = \epsilon_1^+$.

The last case to consider are the complex eigenvalues when $\epsilon_1^- < \epsilon < \epsilon_1^+$. When ϵ is in this range, the eigenvalues $\lambda_{3,4}$ are a complex conjugate pair. Thus close to the steady state, trajectories will spiral around it. Each time the trajectory loops round the steady state, there will be a part of the trajectory with $y > 1$ and so rebound occurs infinitely often, but with exponentially decreasing magnitude, which is determined by $\text{Re}(\lambda_{3,4}) = -(k_3 + \epsilon)/2$. □

Proof of Lemma 5.5 The phase space is bounded by the planes $x = 0$, $y = 0$, $z = 0$ and $w = 0$. As in the proof of Lemma 4.1, we have to show that the derivative normal to each of these planes is

non-negative [18].

The proof that the derivative normal to the planes $x = 0$ and $z = 0$ is non-negative is the same as in the proof of Lemma 4.1. On the plane $y = 0$, we find from (22) that

$$\dot{y}|_{y=0} = k_3 w + k_2 z$$

and so $\dot{y}|_{y=0} \geq 0$ when $w, z \geq 0$. Similarly on the plane $w = 0$, from (23), we obtain

$$\dot{w}|_{w=0} = \epsilon h(y)$$

and so $\dot{w}|_{w=0} > 0$ for all $y \geq 0$ using assumption (h1).

Thus, we conclude that any trajectory with $x(0)$, $y(0)$, $z(0)$, $w(0) \geq 0$ must satisfy $x(\tau)$, $y(\tau)$, $z(\tau)$, $w(\tau) \geq 0$ for all $\tau > 0$. □

Proof of Lemma 5.6 Solving the equations $\dot{x} = \dot{y} = \dot{z} = 0$ for x, y and z again gives two solutions, but one of these has y negative, so is not physically relevant and is not in the positive part of the phase plane considered in this lemma. The other solution is given by

$$x = z = 0, \quad y = w.$$

Substituting for y into the equation $\dot{w} = 0$ gives

$$h(w) - w = 0. \tag{78}$$

As in the proof of Lemma 4.2, we see that $w = 1$ is a solution of this equation using (h2) and assumption (h3) ensures that this solution is unique. □

Proof of Lemma 5.7 We showed in [2] that λ_1 and λ_2 are always real and negative. Furthermore, Lemma 5.2 gives that λ_3 and λ_4 are always negative and real or complex with negative real part. So the steady state is linearly stable. □

Proof of Lemma 5.8 The linear stability proved in Lemma 5.7 implies local stability, i.e., there is some $\delta > 0$ such that all solutions in a δ neighbourhood of the fixed point $(0, 0, 1, 1)$ will converge to this fixed point as $\tau \to \infty$. Or formally, if at some τ_0 we have $||(x(\tau_0), z(\tau_0), y(\tau_0), w(\tau_0)) - (0, 0, 1, 1)|| < \delta$, then $\lim_{\tau \to \infty}(x(\tau), z(\tau), y(\tau), w(\tau)) = (0, 0, 1, 1)$. For later use, we define the following quantities

$$b_1 = \min\left\{h(y) - 1 \mid 0 \leq y \leq 1 - \frac{\delta}{2}\right\}, \quad b_2 = \min\left\{1 - h(y) \mid y \geq 1 + \frac{\delta}{2}\right\}, \quad b = \min(b_1, b_2);$$

and

$$B = \sup\{|h(y) - 1| \mid y \geq 0\}.$$

The assumptions (h1)–(h4) ensure that $b > 0$ and $|h(y) - 1|$ is bounded, hence B is well defined.

Let $(x(\tau), z(\tau), y(\tau), w(\tau))$ be a solution of the TMDD system with moderator feedback (20)–(23). Following the same arguments as in Lemma 2.3 of [2], it follows that the normalised total amount of drug $u = x + \mu z$ decreases monotonically to zero. Since x and z must be non-negative, this implies that $\lim_{\tau \to \infty} x(\tau) = \lim_{\tau \to \infty} z(\tau) = 0$, as for the original TMDD model. Thus there is some $T > 0$ such that for all $\tau > T$, we have

$$|x(\tau)| < \min\left(\frac{b\delta}{8}, \frac{\delta^2}{16}\right) \min\left(\frac{\mu k_3}{B}, 1\right) \text{ and } |z(\tau)| < \min\left(\frac{b\delta}{8}, \frac{\delta^2}{16}\right) \min\left(\frac{k_3}{k_2 B}, 1\right). \tag{79}$$

Next we consider the function $V(y, w)$ as defined in the proof of Lemma 5.3, i.e.,

$$V(y, w) = \frac{1}{k_3} \int_1^y (1 - h(y')) \, dy' + \frac{1}{2\epsilon}(w - 1)^2.$$

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It is shown in that proof that V is non-negative for $y \geq 0$ and $w \geq 0$. We will show here that $V(y(\tau), w(\tau))$ is decreasing when $\tau > T$ and $|y(\tau) - 1| \geq \frac{\delta}{2}$ or $|w(\tau) - 1| \geq \frac{\delta}{2}$ and that this implies that any solution has to enter the local δ neighbourhood in which solutions get attracted to the fixed point.

Let $\tau > T$, i.e, $x(\tau)$ and $z(\tau)$ satisfy (79). Then we have

$$
\begin{aligned}
\dot{V}(y, w) &= \frac{1}{k_3}(1 - h(y))\dot{y} + \frac{(w-1)\dot{w}}{\epsilon} \\
&= \frac{1}{k_3}(1 - h(y))\left(k_3(w-y) - \frac{xy}{\mu} + k_2 z\right) + (w-1)(h(y) - w) \\
&= -(w-1)^2 - (1 - h(y))(y-1) - \frac{(1-h(y))}{k_3}\left(\frac{xy}{\mu} - k_2 z\right).
\end{aligned}
$$

Next we consider each term. First we note that for every $w \geq 0$ and $y \geq 0$, we have that $-(w-1)^2 \leq 0$ and $-(1-h(y))(y-1) \leq 0$. Furthermore, if $|w-1| \geq \frac{\delta}{2}$, then $-(w-1)^2 \leq -\frac{\delta^2}{4}$. Also, if $|y-1| \geq \frac{\delta}{2}$ then the fact that $(1-y)(h(y)-1) > 0$ and the definition of b give that $-(1-y)(h(y)-1) \leq -\frac{b\delta}{2}$. The definition of B gives that $\frac{(1-h(y))k_2 z}{k_3} \leq \frac{Bk_2|z|}{k_3} < \min\left(\frac{b\delta}{8}, \frac{\delta^2}{16}\right)$. Finally, if $y > 1$, then $1 - h(y) > 0$, thus $-\frac{(1-h(y))}{k_3}\frac{xy}{\mu} \leq 0$. If $0 \leq y \leq 1$, then the definition of B gives $-\frac{(1-h(y))}{k_3}\frac{xy}{\mu} \leq \frac{B}{k_3}\frac{x}{\mu}$. So together we get that $-\frac{(1-h(y))}{k_3}\frac{xy}{\mu} < \min\left(\frac{b\delta}{8}, \frac{\delta^2}{16}\right)$. With these four estimates we get that if $|w-1| \geq \frac{\delta}{2}$ or $|y-1| \geq \frac{\delta}{2}$, then

$$
\dot{V}(y(\tau), w(\tau)) < -\min\left(\frac{b\delta}{2}, \frac{\delta^2}{4}\right) + 2\min\left(\frac{b\delta}{8}, \frac{\delta^2}{16}\right) = -\min\left(\frac{b\delta}{4}, \frac{\delta^2}{8}\right).
$$

If we assume that for all $\tau > T$, either $|w(\tau) - 1| \geq \frac{\delta}{2}$ or $|y(\tau) - 1| \geq \frac{\delta}{2}$ (or both), then integration of the estimate above gives that

$$
V(\tau) < V(T) - \min\left(\frac{b\delta}{4}, \frac{\delta^2}{8}\right)(\tau - T) < 0
$$

for τ sufficiently large. However this contradicts the fact that V is non-negative, hence our assumption is incorrect and we can conclude that there is some $\tau_0 > T$ such that $|w(\tau_0) - 1| < \frac{\delta}{2}$ and $|y(\tau_0) - 1| < \frac{\delta}{2}$. Since $\tau_0 > T$, we also have $|x(\tau_0)| < \frac{\delta^2}{16}$ and $|z(\tau_0)| < \frac{\delta^2}{16}$. Thus $||(x(\tau_0), z(\tau_0), y(\tau_0), w(\tau_0)) - (0, 0, 1, 1)|| < \delta$ and the local stability implies that the solution will converge to the steady state as $\tau \to \infty$.

B Proof of Rebound Lemmas

Lemma 5.13 is proved by assuming that λ_4 is closest to zero and then considering the three ϵ intervals for which the value of λ_4 is qualitatively different.

Lemma B.1 *If $0 < \epsilon \leq \epsilon_1^-$ and the eigenvalue λ_4 is closest to zero then rebound occurs.*

Proof First we consider the case $0 < \epsilon < \epsilon_1^-$. Generically the trajectory must approach the steady state tangent to the eigenvector v_4 given in (59) since λ_4 is real and is the eigenvalue closest to zero. We note that

$$
k_3 + \lambda_4 = \frac{1}{2}\left(k_3 - \epsilon + \sqrt{(k_3 - \epsilon)^2 - 4\epsilon k_3 h_0}\right).
$$

We see from (53) that the term under the square root is positive if λ_4 is real (i.e., $\epsilon \notin (\epsilon_1^-, \epsilon_1^+)$), and thus the expression above immediately implies that $k_3 + \lambda_4 > 0$ if $\epsilon < \epsilon_1^- < k_3$ and $k_3 + \lambda_4 < 0$ if $\epsilon > \epsilon_1^+ > k_3$. Therefore, if $0 < \epsilon < \epsilon_1^-$, then the third and fourth components of the eigenvector v_4

have the same sign. We therefore have two cases to consider. In the first case, y and w both approach the steady state from above, and clearly rebound occurs in this case. Alternatively, y and w both approach the steady state from below and Corollary 5.11 implies that there is rebound in this case also.

As in the proof of Theorem 3.5 in [2], we must again consider the possibility that for particular parameter values, the generic situation considered above does not occur, and so the trajectory approaches the steady state tangent to one of the other eigenvectors.

We first consider the eigenvector v_3 (see (59)). For this vector, we note that $k_3 + \lambda_3 > 0$, by a similar argument to the one above for $k_3 + \lambda_4$, and so the third and fourth components of the eigenvector v_3 have the same sign. Thus, the above proof by contradiction also holds if the trajectory approaches the steady state tangent to v_3.

For the eigenvector v_2, we note, as in the proof of Theorem 3.5 of [2], that this eigenvector points out of the phase space, since the first and second components have opposite sign, and so no trajectory can approach the steady state tangent to this eigenvector.

Finally we consider the eigenvector v_1. We note that the third and fourth components of this eigenvector will have the same sign if $\epsilon + \lambda_1 < 0$. Since λ_4 is real and closest to zero, the definition of λ_4 (53) gives

$$\epsilon + \lambda_1 < \epsilon + \lambda_4 = \frac{1}{2}\left(\epsilon - k_3 + \sqrt{(k_3 - \epsilon)^2 - 4\epsilon k_3 h_0}\right) < 0$$

as $\epsilon < \epsilon_1^- < k_3$. So we can conclude that the third and fourth entries in the eigenvector v_1 have the same sign, and so the above proof by contradiction also holds if the trajectory approaches the steady state tangent to v_1.

This covers all possible cases for $\epsilon < \epsilon_1^-$, and so we conclude that rebound must occur for those ϵ values.

Finally we consider the case $\epsilon = \epsilon_1^-$, which is when the Jacobian matrix has two repeated real eigenvalues $\lambda_3 = \lambda_4(= \lambda_*^-)$ that are the least negative. In this case, the two corresponding eigenvectors satisfy $v_3 = v_4$ and there is a generalised eigenvector v_g which is given by

$$v_g = (0, 0, 0, 1)$$

The solutions on the two-dimensional linearised manifold spanned by v_3 and v_g are linear combinations of $e^{\lambda_3 t}(v_g + t v_3)$ and $e^{\lambda_3 t} v_3$ [9] and asymptotically will align with the eigenvector v_3. It is easily verified that $k_3 + \lambda_*^- > 0$, and so the third and fourth components of v_3 have the same sign. The same arguments as above show that rebound must then occur in this case, and that the trajectory cannot approach the steady state tangent to v_1 or v_2. □

Next we consider the case that $\epsilon > \epsilon_1^+(k_3)$ and λ_4 is real and the eigenvalue closest to zero. This can only happen if $\lambda_1 < \lambda_\infty$. In the case of the direct feedback, we analysed this region by using that the total amount of receptor $(v = y + z)$ stays on one side of the plane $v = 1$, see Lemma 4.7. As observed earlier, this property does not hold any more for the feedback with a moderator, but we can define a related quantity and show a partial result.

We write $h(y)$ as in (35), i.e., $h(y) = 1 + (1 - y)\tilde{h}(y)$ and define the bounds m and M as in (41). Next we define

$$\tilde{v} = y + z + \frac{k_3 w}{\epsilon}.$$

At baseline, we have $\tilde{v} = 1 + \frac{k_3}{\epsilon}$ and the initial condition is also in the plane $\tilde{v} = 1 + \frac{k_3}{\epsilon}$. By using this plane, we will now prove that there is rebound if k_3 is sufficiently large compared to k_4.

Lemma B.2 *If $\epsilon \geq \epsilon_1^+$, the eigenvalue λ_4 is closest to zero, and $k_3 > \frac{k_4}{1+m}$, then rebound occurs.*

Proof Using the definition of $\tilde{v}$, we get that

$$
\begin{aligned}
\dot{\tilde{v}} &= -k_4 z + k_3(h(y) - y) \\
&= (1 - y)(k_3(1 + \tilde{h}(y)) - k_4) + k_4\left(1 + \frac{k_3}{\epsilon} - \tilde{v}\right) + \frac{k_3 k_4}{\epsilon}(w - 1). \quad (80)
\end{aligned}
$$

Evaluating this expression at $\tau = 0$, gives $\dot{\tilde{v}}(0) = 0$. Differentiating once more shows that

$$
\ddot{\tilde{v}}(0) = \frac{1}{\mu}\left(k_3(1 + h_0) - k_4\right) > 0,
$$

as $k_4 < k_3(1 + m) < k_3(1 + h_0)$. Thus $\tilde{v}(\tau) > 1 + \frac{k_3}{\epsilon}$ for sufficiently small $\tau > 0$.

We will prove the occurrence of rebound with a contradiction argument and so we assume that there is no rebound, i.e., $y(\tau) < 1$ for all $\tau > 0$. With Lemma 5.10, this implies that $w(\tau) > 1$ for all $\tau > 0$. Hence on the plane $\tilde{v} = 1 + \frac{k_3}{\epsilon}$, equation (80) gives

$$
\dot{\tilde{v}}|_{\tilde{v}=1+\frac{k_3}{\epsilon}} = (1 - y)(k_3(1 + \tilde{h}(y)) - k_4) + \frac{k_3 k_4}{\epsilon}(w - 1) > (1 - y)(k_3(1 + m) - k_4).
$$

Since $k_3(1 + m) > k_4$, this implies that $\dot{\tilde{v}}|_{\tilde{v}=1+\frac{k_3}{\epsilon}} > 0$, hence $\tilde{v}(\tau) > 1 + \frac{k_3}{\epsilon}$ for all $\tau > 0$.

Since λ_4 is the eigenvalue closest to zero, generically the solution will approach the steady state along the line spanned by the eigenvector v_4. Furthermore, the outward normal to the plane $\tilde{v} = 1 + \frac{k_3}{\epsilon}$ is $\nabla \tilde{v} = (0, 1, 1, k_3/\epsilon)$. Thus with the expression for v_4 in (59) and $\epsilon > \epsilon_1^+$, it follows that its fourth component is

$$
k_3 + \lambda_4 < k_3 + \lambda_\infty = k_3 - k_3(1 + h_0) = -k_3 h_0 < 0.
$$

As the solution has $y < 1$ and $w > 1$, the solution comes in via $-v_4$. But this vector is below the plane $\tilde{v} = 1 + \frac{k_3}{\epsilon}$ as

$$
(\nabla \tilde{v}, -v_4) = -\frac{k_3}{\epsilon}(\epsilon + k_3 + \lambda_4) < 0,
$$

as, by the definition of λ_4, we have $\epsilon + k_3 + \lambda_4 = \frac{1}{2}(k_3 + \epsilon + \sqrt{D}) > 0$. This contradicts the fact that the solution is always above the plane, hence our assumption that there is no rebound is false.

Finally, we consider the case $\epsilon = \epsilon_1^+$. This case is similar to that when $\epsilon = \epsilon_1^-$ in the proof of Lemma B.1 in that there is a repeated eigenvalue $\lambda_3 = \lambda_4$ with one eigenvector $v_3 = v_4$ and a generalised eigenvector v_g. The solution will again asymptotically align with the eigenvector v_4 which has fourth component $k_3 + \lambda_*^+ < 0$ and so the above proof by contradiction works in this case also. $\qquad \square$

Next we look at the case that the eigenvalue λ_4 is complex (i.e, $\epsilon_1^- < \epsilon < \epsilon_1^+$) and its real part is closest to zero.

Lemma B.3 *If the eigenvalue λ_4 is complex and its real part is larger than λ_1, hence closer to zero, then y generically converges to its steady state value $y = 1$ in an oscillatory fashion, and so rebound occurs infinitely many times, but with exponentially decreasing magnitude.*

Proof If the complex conjugate pair of eigenvalues $\lambda_{3,4}$ have real part that is closer to zero than λ_1, then the Hartman-Grobman Theorem [7] gives that generically trajectories will converge to the steady state in the phase space tangent to the (y, w) plane and will spiral around the steady state $(y, w) = (1, 1)$. Each time the trajectory loops round the steady state, there will be a part of the trajectory with $y > 1$ and so rebound occurs infinitely often, but with exponentially decreasing magnitude, which is determined by $\mathrm{Re}(\lambda_{3,4}) = -(k_3 + \epsilon)/2$.

As in the previous case, we also need to consider the possibility that for particular parameter values, the trajectory might approach tangent to a different eigenvector. We again note that the eigenvector v_2 points out of the phase space, and so a trajectory cannot approach the steady state tangent to

this eigenvector. However, the eigenvector v_1 points into the phase space, since the first and second components of the eigenvector are both positive. Thus, we are unable to exclude the possibility that an orbit could approach the steady state tangent to this eigenvector for particular parameter values. However, this is very unlikely to occur in practice and is not a generic case. $\quad\square$

Proof of Lemma 5.13 This follows immediately by combining Lemmas B.1-B.3 and using that $\epsilon_1^- < \epsilon < \epsilon_1^+$ implies that λ_4 is complex.

Next we will consider the case that λ_1 is the eigenvalue closest to zero. The eigenvector v_1 corresponding to the eigenvalue λ_1 is given in (59). The first two entries in this eigenvector are the same as we had previously for the case with no feedback, and since both of these quantities are positive (see Lemma 2.7 of [2]), we must take a positive multiple of v_1 to ensure that x and z are positive in the direction of this eigenvector. We now consider the signs of the third and fourth components of this vector, although we note that since the steady state values for y and w are non-zero, there is no requirement that these entries be positive. First we determine the sign of the denominator in the expressions for the third and fourth components of v_1.

Lemma B.4 *If λ_1 is the least negative eigenvalue of the matrix (58) then*

$$(k_3 + \lambda_1)(\epsilon + \lambda_1) + \epsilon k_3 h_0 > 0.$$

Proof The eigenvalues λ_3 and λ_4 are found from the bottom right 2×2 matrix in (58) and hence are roots of the characteristic polynomial

$$p(\lambda) = (k_3 + \lambda)(\epsilon + \lambda) + \epsilon k_3 h_0.$$

We note that the quantity of interest is given by $p(\lambda_1)$.

Now $p(0) = \epsilon k_3(1 + h_0) > 0$. If the eigenvalues λ_3 and λ_4 are real and negative (with $\lambda_3 < \lambda_4$) then $p(\lambda) > 0$ for $\lambda \in (\lambda_4, 0]$. Since λ_1 is the eigenvalue closest to zero, it must lie in this range and so in this case $p(\lambda_1) > 0$.

Alternatively, if the eigenvalues λ_3 and λ_4 are complex, then $p(\lambda)$ has no real roots and so $p(\lambda) > 0$ for all λ, and in particular $p(\lambda_1) > 0$.

Thus, in both cases, $p(\lambda_1) > 0$ as required. $\quad\square$

Proof of Lemma 5.14 The ratio of the third and fourth components of v_1 (which we denote by $v_{1,3}$ and $v_{1,4}$ respectively) is given by

$$\frac{v_{1,3}}{v_{1,4}} = -\frac{\epsilon + \lambda_1}{\epsilon h_0}.$$

Thus, if $\epsilon + \lambda_1 < 0$, then $v_{1,3}$ and $v_{1,4}$ have the same sign and the proof of rebound given in Lemma B.1 holds in this case also.

If $k_1 > k_4$, then we note from Lemma 2.8 of [2] that $k_4 + \lambda_1 < 0$ and so, using the result of Lemma B.4, we then see that $v_{1,4} < 0$ in this case. Since we are taking a positive multiple of the eigenvector, this implies that w must approach its steady state value from below and so, by Corollary 5.11, rebound must occur in this case also.

As in previous cases, we should again consider the possibility that a trajectory could approach the steady state tangent to a different eigenvector. As previously, the eigenvector v_2 again points out of the invariant region since the first two entries have opposite sign. However, it is not possible to determine the global invariant manifold that approaches the steady state tangent to the eigenvectors v_3 or v_4 and so we can only state a generic result. $\quad\square$

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