Modeling and Analysis of the Signal Transduction Process with Delays Involving G Protein Coupled Receptors as a Drug Target

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***Abstract*—**Mathematical modeling has played a significant role in modern biology and pharmacology and has become a powerful tool for examining GPCR pathways. Modeling can be used to validate hypothesized mechanisms, and identify relevant data. More importantly, it can suggest new drug targets, designs of experiments, and new explanations for observed phenomena. G protein coupled receptors (GPCRs) constitute the largest family of cell membrane receptors which are subject to being targeted by an estimated 50% of current pharmaceuticals. Thus, better understanding of GPCRs and the signal transduction pathways they mediate will lead to new drug targets. Signal transduction is the process by which a cell recognizes and extracellular signal and converts that signal into an intracellular response. Subjected to transient stimuli, biological systems can exhibit early responses and/or late responses. In this study, we use mathematical modelling and analysis to study dynamical mechanisms of biological memory and delayed response to external stimuli. A delay model of signaling pathways involving G-proteins is analyzed to show that the model admits positive solutions and is uniformly persistent. Global stability of the system is shown to be attainable under certain conditions on the system's parametric values. It is found that the delays τ_I in response to inhibition and τ_R in G protein mediated response to external stimuli of the receptors do not appear to impact on the persistent and stability characteristics of this system.

***Keywords*—**Delay differential equations; omega limit set; persistence; signal transduction; stability.

I. Introduction

MATHEMATICAL and computer modeling can help incorporate complicated hypothesized mechanisms and parameters, such as rates and concentrations, allowing both qualitative and quantitative insights. Models can vary significantly in the level of detail used to describe molecular interactions. For example, models of GPCR signaling may include vast detail in the G-protein activation/deactivation cycle, such as in the work of Bornheimer *et al*. [1] or may have no explicit inclusion of G-proteins at all but rather lump their effect into what happens to a downstream component, as in Linderman's work [2]. We have to choose how fine- or coarse-grained we make our model. The appropriate choice depends on the question we are asking and also on the data we are able to obtain for model validation and testing. According to Linderman [2], "Modeling is meant to be an iterative process with experimentation, each one driving the other."

First, a "minimal model" is constructed and then grows in complexity by incorporating new hypotheses and new data. According to Linderman [2], "Simple is good. There is no glory to be had by constructing a complicated model when a simple one can elucidate the key features of the biology; in fact, a complicated model may obscure a simple result." Although models typically involve parameter values which are often difficult to ascertain, often the parametric values themselves are less important than the qualitative insight [2], for example how the receptor trafficking or dimerization effects the dose response curves.

As evidence that modeling is becoming an increasingly useful tool for understanding GPCR pathways, the United States Food and Drug Administration Critical Path Initiative has recently identified model-based drug development, including drug and disease modeling, as an important goal (www.fda.gov/oc/initiatives/criticalpath). New discoveries and theories generated by model construction have been appearing in many prominent biology related journals. The hypotheses on which the models are based have been the impetus for the development of new experimental techniques, such as RNA silencing of pathway molecules and novel fluorescent probes that allow for single cell kinetic data, in order to both generate data for model building and allow testing of key model findings [2].

It is well recognized that G-protein-coupled receptors (GPCRs) are the largest family of cell membrane receptors [2]. The fact that an estimated 50% of current pharmaceuticals target GPCRs [3] indicates that further increases in our

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understanding of GPCRs and the signaling pathways they mediate will without doubt lead to new drug targets. Use of mathematical and computational models has played an increasingly important role in modern biology and pharmacology [2, 4, 5] and offers a powerful tool for examining GPCR pathways. Apart from allowing us to better understand hypothesized mechanisms, they can be used to execute virtual (*in silico*) experiments, interpret data, suggest new drug targets, suggest designs of experiments to validate the model, and offer insightful explanations for observed phenomena [2].

Abnormalities of signal transduction pathways have been linked to the development of many serious disorders, such as cancer which derives from a cell that has lost the ability to respond normally to controls from outside, or inside, the cell. Signal transduction is the process by which information from an extracellular signal is transmitted from the plasma membrane into the cell and along an intracellular chain of signaling molecules to stimulate a cellular response [6].

The signal transduction pathway is a three step process; reception, transduction, and the response step. Reception is the target cell's detection of a signal transmitted from cell's surrounding environment. A chemical signal is detected when it binds to a receptor protein located at the cell's surface or inside the cell. Signal transduction converts the external stimuli into a form that can bring about a specific cellular response. In the third stage of cell signaling, the transduction process brings about a cellular response. This can be any of many different cellular activities, such as activation of a certain enzyme, rearrangement of the cytoskeleton, or activation of specific genes [7]. After a signal transduction pathway has been initiated and the information has been transduced to affect other cellular processes, the signaling processes must be terminated. Without such termination, cells lose their responsiveness to new signals. Signal processes that fail to properly terminate can lead to uncontrolled cell growth and the possibility of cancer [8]. In addition, we know of many situations where altered signaling pathways produce dramatic changes in cell survival, cell proliferation, morphology, angiogenesis, longevity, or other properties that characterize cancer cells [9]. For this reason, better understanding of the signal transduction process has been a subject of intense investigation.

In the work of Rattanakul *et al.* [10], a model was proposed for the signal transduction pathway which involves G protein coupled receptors (GCPRs) consisting of a system of two differential equations governing the interaction between the inhibitor protein and the ligand-receptor complexes. Signal transduction across the plasma membrane is mediated by membrane receptor bound proteins which connect the genetically controlled biochemical reactions in the cytosol to the production of the second messenger, leading to desired intracellular responses.

In order for a life form to be able to function properly, its cellular constituents need to have the ability to efficiently communicate with each other. This requires that cells have a mechanism to detect and respond specifically to external signals [11]. In [12], Lodish *et al.* described one of the more complex strategies for cell communications which involves a three-stage G protein coupled enzyme cascade.

In the first stage, the reception stage, a specialized membrane receptor protein interacts with a particular ligand, or absorbs a photon of light of a particular wavelength, and thus becomes activated. In the second stage, the transduction stage, the activated receptor, the density of which will be denoted R, triggers a heterotrimeric G protein to exchange GDP (guanosine diphosphate) for the nucleotide guanosine triphosphate (GTP). The α-subunit and the β-γ complex of the G protein then dissociate, after which the GTP-bound α-subunit then diffuses along the membrane and binds to an effector, activating it and leading to an appearance of GTPase activity resulting in the conversion of active α-GTP to inactive α-GDP and thereby inhibiting the activation of AC by G proteins.

In this mechanism we consider the GTP and GDP ligands to play the roles of activating agent (A) and inhibiting agent (I), respectively. We let G be the amount of α-subunits of G proteins in the resting state, and G^* be that in the active state.

Then, as in [10] we will be able to write the following governing equations for A, I, and G^*.

$$\frac{dA}{dt} = -k_{-a}A + k_a R \tag{1}$$

$$\frac{dI}{dt} = -k_{-i}I + k_i R \tag{2}$$

$$\frac{dG^*}{dt} = -k_{-r}IG^* + k_r AG \tag{3}$$

Letting C be the concentration of the second messenger, such as cAMP, which represents the output signal of the transduction process, then the cAMP in turns acts as a second messenger and amplifies the initial signal [13]. Thus, following previous works by Levchenko and Iglesias [14] and Iglesias [15] the rate equation for the ligand-receptor complex density on the cellular membrane surface at time t should read as follows.

$$\frac{dR}{dt} = -\tilde{a}_3 R - \frac{\tilde{b}_1 R}{\tilde{b}_2 + R} + k_R C \tag{4}$$

where the first term on the right is the removal rate, the second accounts for its transport through the cell membrane which saturates as R increases, the third accounts for the signal amplification arising from the synthesis of cAMP.

As in [14-15], we now assume that the activated regulators G^*, A, and C equilibrate relatively quickly so that we shall be able to arrive at the following:

$$C = \frac{\tilde{b}_4 R^2}{\left(\tilde{b}_3 R + I\right)^2} + K_C \tag{5}$$

at equilibrium.

Substituting (5) into (4), one arrives at the following system:

$$\frac{dR}{dt} = -\tilde{a}_3 R - \frac{\tilde{b}_1 R}{\tilde{b}_2 + R} + \frac{k_R \tilde{b}_4 R^2}{\left(\tilde{b}_3 R + I\right)^2} + k_R K_C \qquad (6)$$

$$\frac{dI}{dt} = -k_{-i} I + k_i R \qquad (7)$$

We refer the readers to [10, 14-15] for further details, while we have assumed here that homogeneous distribution of the ligand-receptor complexes and the inhibitor protein on the cell membrane has been attained.

According to Han *et al.* [16], "memory is a ubiquitous phenomenon in biological systems, yet its impacts, and how to manipulate it at the sub-cellular level, remain poorly understood. Subjected to transient stimuli, biological systems can exhibit short early responses and/or prolonged late responses". Although experimental evidence has provided some intuitive explanation at the basic molecular level, it does little to clarify the important dynamics that could lead us to discover possible therapeutic strategies in the setting of human diseases. In such attempts, mathematical modelling can go a long way in illuminating the underlying dynamic intricacies which may not be attained in experimental executions alone.

In this work, two significant time delays have been incorporated in the system. One is the delay τ_I in the response of the ligand-receptor complexes (R) to the action of the inhibitor protein, while the other is the delay τ_R in the response of the inhibitor protein (I) to the changes in the density of R.

Based on earlier investigations and modeling efforts of Giang *et al.* [17], and Palumbo *et al.* [18], we study a mathematical model for the signal transduction process consisting of delay-differential equations modified from the model studied by Rattanakul *et al.* [10] discussed above. The model is then analyzed by using the ω–limit set of a positive solution and constructing a full time solution. We first show that the model is persistent under certain conditions, in which case the levels of the inhibitors and ligand-receptor complex are bounded above and below by positive constants. Moreover, under certain conditions, oscillation about the respective basal levels, which is of clinical interest for control purposes, may be observed or else the system converges to a positive steady state.

Finally, the global stability of the model system will be investigated. Conditions on the system parameters are given which ensure the global stability of the steady state of the system at its basal levels.

II. The Reference Model

To incorporate delay mechanism, based on the model discussed in the previous section, we consider the system of two delay differential equations which governs the interaction between the ligand-receptor complexes $R(t)$ and the inhibitor protein $I(t)$ as follows:

$$\frac{dR}{dt} = -a_1 R - \frac{b_1 R}{b_2 + R} + \frac{b_3 R^2}{(b_4 R + I(t-\tau_I))^2} + a_4 \qquad (8)$$

$$\frac{dI}{dt} = -a_2 I + a_3 R(t-\tau_R) \qquad (9)$$

where the first term on the right of (8) and (9) are the removal rates of the corresponding state variables, the second term on the right of (8) is the rate that R is internalized through the cell membrane, the third term accounts for the amplification effect on the production of R due to the secretion of the secondary hormone or signal with a delay τ_I, and a_4 is the zero order production rate of R. The second term on the right of (9), on the other hand, is the production rate of inhibiting protein I in response of the increase in R at the time τ_R sec earlier. We first show that the model system (8)–(9) has a positive solution.

Theorem 1 System (8)–(9) admits positive solution provided that $R(t) > 0$ on the initial interval $[-\tau_R, 0]$ and $I(t) > 0$ on the initial interval $[-\tau_I, 0]$.

Proof Let $R(t) > 0$ over an initial interval $[-\tau_R, 0]$. According to the continuity of the solution of a differential equation, $R(t)$ would become non-positive if there existed a $t_0 > 0$ such that

$$R(t_0) = 0$$

and $R(t) > 0$ for any $t, 0 \le t < t_0$.

Then, necessarily, $\left.\frac{dR}{dt}\right|_{t=t_0} \le 0$, which is a contradiction because

$$\left.\frac{dR}{dt}\right|_{t=t_0} = -a_1 R(t_0) - \frac{b_1 R(t_0)}{b_2 + R(t_0)} + \frac{b_3 R^2(t_0)}{(b_4 R + I(t_0 - \tau_I))^2} + a_4 = a_4 > 0. \qquad (10)$$

This proves that, if $R(t) > 0$ over $[-\tau_R, 0]$ then $R(t)$ never vanishes and is positive for all $t \ge -\tau_R$. Similarly, it can be proven that, if $I(t) > 0$ over an initial interval $[-\tau_I, 0]$, also $I(t)$ never vanishes and is positive for all later time. If there existed a $t_0 > 0$ such that

$$I(t_0) = 0$$

and $I(t) > 0$ for any $t, 0 \le t < t_0$.

Then, necessarily, $\left.\frac{dI}{dt}\right|_{t=t_0} \le 0$, which is a contradiction because

$$\left.\frac{dI}{dt}\right|_{t=t_0} = -a_2 I(t_0) + a_3 R(t_0 - \tau_R) \quad (11)$$
$$= a_3 R(t_0 - \tau_R) > 0. \quad \blacksquare$$

III. The Uniform Persistence

This section investigates some properties involving the solution of (8)–(9) and the equilibrium point (R_b, I_b) which, by definition, satisfies the following system:

$$a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b}{(b_4 R_b + I_b)^2} = a_4 \quad (12)$$

$$I_b = \frac{a_3}{a_2} R_b \quad (13)$$

In what follows, we let

$$R_m = \liminf_{t \to +\infty} R(t), \qquad R_M = \limsup_{t \to +\infty} R(t),$$
$$I_m = \liminf_{t \to +\infty} I(t), \qquad I_M = \limsup_{t \to +\infty} I(t).$$

Theorem 2 Under the assumptions of Theorem 1, system (8)–(9) is persistent.

Proof Recall that a model is persistent if there exists a pair of positive real numbers (m, M) such that there exists a $\bar{t}$ such that

$$0 < m < X_i(t) < M < +\infty, \quad \text{for all } t \geq \bar{t}$$

for each component X_i of the state vector.

The proof is achieved by proving the following four statements:

$$1) R_M < +\infty, \quad 2) I_M < +\infty, \quad 3) R_m > 0, \quad 4) I_m > 0.$$

Step 1. In order to show the boundedness of the evolution of the ligand-receptor complex, assume that $R_M = +\infty$, which means, due to continuity, that there is a time sequence $\{t_n\} \subset [0, +\infty)$ such that

$$\lim_{n \to \infty} t_n = +\infty,$$
$$\lim_{n \to \infty} R(t_n) = +\infty,$$

with

$$\left.\frac{dR}{dt}\right|_{t=t_n} \geq 0.$$

However,

$$\left.\frac{dR}{dt}\right|_{t=t_n} = -a_1 R(t_n) - \frac{b_1 R(t_n)}{b_2 + R(t_n)} + \frac{b_3 R^2(t_n)}{(b_4 R(t_n) + I(t_n - \tau_I))^2} + a_4 \to -\infty, \quad (18)$$

which is a contradiction, and therefore $R_M < +\infty$.

Step 2. In order to show the boundedness of the evolution of the inhibitor, assume that $I_M = +\infty$, which means, due to continuity, that there is a time sequence $\{t_n\} \subset [0, +\infty)$ such that

$$\lim_{n \to \infty} t_n = +\infty,$$
$$\lim_{n \to \infty} I(t_n) = +\infty,$$

with

$$\left.\frac{dI}{dt}\right|_{t=t_n} \geq 0.$$

However,

$$\left.\frac{dI}{dt}\right|_{t=t_n} = -a_2 I(t_n) + a_3 R(t_n - \tau_R) \to -\infty, \quad (14)$$

which is a contradiction, so that $I_M < +\infty$.

Step 3. Suppose $R_m < +\infty$ (otherwise $R_m > 0$ is trivially verified). Due to continuity, there exists a time sequence $\{t_n\} \subset [0, +\infty)$ such that

$$\lim_{n \to \infty} t_n = +\infty,$$
$$\lim_{n \to \infty} R(t_n) = R_m,$$

with

$$\left.\frac{dR}{dt}\right|_{t=t_n} = 0.$$

This means that:

$$0 = \lim_{n \to \infty} \left(-a_1 R(t_n) - \frac{b_1 R(t_n)}{b_2 + R(t_n)} + \frac{b_3 R^2(t_n)}{(b_4 R(t_n) + I(t_n - \tau_I))^2} + a_4 \right)$$
$$= -a_1 R_m - \frac{b_1 R_m}{b_2 + R_m} + \frac{b_3 R_m^2}{(b_4 R_m + I(t_n - \tau_I))^2} + a_4$$
$$\geq -a_1 R_m - \frac{b_1 R_m}{b_2 + R_m} + \frac{b_3 R_m^2}{(b_4 R_m + I_M)^2} + a_4. \quad (15)$$

Therefore, we have

$$0 < a_4 \le a_1 R_m + \frac{b_1 R_m}{b_2 + R_m} - \frac{b_3 R_m^2}{(b_4 R_m + I_M)^2}. \qquad (16)$$

According to this inequality, we must have $R_m > 0$, or otherwise we would have $a_4 \le 0$.

Step 4. From Step 2, it follows that $I_m \le I_M < +\infty$. Due to continuity, there exists a time sequence $\{t_n\} \subset [0, +\infty)$ such that

$$\lim_{n \to \infty} t_n = +\infty,$$
$$\lim_{n \to \infty} I(t_n) = I_m,$$

with

$$\left.\frac{dI}{dt}\right|_{t = t_n} = 0.$$

This means that:

$$\begin{aligned} 0 &= \lim_{n \to \infty} \left(-a_2 I(t_n) + a_3 R(t_n - \tau_R) \right) \\ &= -a_2 I_m + a_3 R(t_n - \tau_R) \\ &\ge -a_2 I_m + a_3 R_m. \end{aligned} \qquad (17)$$

Then, we have $a_3 R_m \le a_2 I_m$.

Therefore, $I_m > 0$, or otherwise we would have $a_3 \le 0$. ■

Remark 3 As a consequence of Theorems 1 and 2, system (8)–(9) admits positive bounded solutions for any positive initial condition.

Remark 4 Under the assumptions of Theorem 2, uniform persistence of the system (8)–(9) physically represents the fact that perpetual response to external stimuli, such as drug treatments, and inhibiting agents will be at work in a healthy subject.

Theorem 5 Under the assumptions of Theorem 1, let (R, I) be a bounded positive solution of (8)–(9). Then,

$$I_m \le I_b \le I_M, \qquad (19)$$
$$R_m \le R_b \le R_M. \qquad (20)$$

provided $\frac{b_1}{b_3} \gg 1$.

Proof By using the ω–limit set of the persistent solution (R, I), we can construct a full time solution $(\mathcal{R}, \mathcal{I})$ such that

$$I_M = \mathcal{I}(0) = \max_{t \in \mathbb{R}} I(t),$$
$$I_m \le \min_{t \in \mathbb{R}} \mathcal{I}(t),$$
$$R_m \le \mathcal{R}(t) \le R_M, \qquad \text{for all } t \in \mathbb{R}.$$

The readers are referred to the works of Giang *et al.* [15], and Palumbo *et al.* [16], for more detail on full time solutions and their applications. It follows that $\dot{\mathcal{I}}(0) = 0$, and consequently,

$$I_M = \mathcal{I}(0) = \frac{a_3}{a_2} \mathcal{R}(-\tau_R). \qquad (21)$$

First we will show that $I_M \ge I_b$. This means that we need to prove that $\mathcal{R}(-\tau_R) \ge R_b$ by showing that the assumption of $\mathcal{R}(-\tau_R) < R_b$ leads to a contradiction. Assuming that $\mathcal{R}(-\tau_R) < R_b$, by (13) we have $I_M < I_b$. Since $R_m \le \mathcal{R}(-\tau_R)$, it follows that $R_m < R_b$.

Again by using the ω-limit set of the persistent solution (R, I), we can construct a full time solution $(\mathbf{R}, \mathbf{I})$ such that

$$R_m = \mathbf{R}(0) = \min_{t \in \mathbb{R}} \mathbf{R}(t),$$
$$R_M \ge \max_{t \in \mathbb{R}} \mathbf{R}(t)$$
$$I_m \le \mathbf{I}(t) \le I_M, \qquad \text{for all } t \in \mathbb{R}$$

It follows that $\dot{\mathbf{R}}(0) = 0$, and consequently,

$$a_1 R_m + \frac{b_1 R_m}{b_2 + R_m} - \frac{b_3 R_m^2}{(b_4 R_m + \mathbf{I}(-\tau_I))^2} = a_4 \qquad (22)$$

Since we already have $I_M < I_b$ and $R_m < R_b$, with $\frac{b_1}{b_3}$ sufficiently bigger than 1 we have

$$\begin{aligned} a_4 &= a_1 R_m + \frac{b_1 R_m}{b_2 + R_m} - \frac{b_3 R_m}{(b_4 R_m + \mathbf{I}(-\tau_I))^2} \\ &< a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b}{(b_4 R_b + \mathbf{I}(-\tau_I))^2} \\ &< a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b}{(b_4 R_b + I_b)^2} = a_4 \end{aligned} \qquad (23)$$

which is a contradiction, so that $\mathcal{R}(-\tau_R) \ge R_b$. We obtain

$$I_M = \frac{a_3}{a_2} \mathcal{R}(-\tau_R) \ge \frac{a_3}{a_2} R_b = I_b. \qquad (24)$$

In addition, it follows that $R_M \ge \mathcal{R}(-\tau_R) \ge R_b$. Similarly, we may prove that $R_m \le R_b$ and $I_m \le I_b$. ■

Remark 6 It is physically meaningful that the equilibrium is bounded in the range of all bounded positive solutions, and the densities of ligand-receptor complexes and the inhibiting protein in the transduction process should eventually adjust to some levels and remain steady when we are healthy.

Corollary 6 Theorem 5 yields the following inequalities:

$$a_3 R_m \le a_2 I_m \le a_2 I_M \le a_3 R_M, \qquad (25)$$

$$a_1R_M+\frac{b_1R_M}{b_2+R_M}-\frac{b_3R_M^2}{(b_4R_M+I_m)^2}$$

$$\le a_4 \le a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m^2}{(b_4R_m+I_M)^2} \quad (26)$$

Proof We initially verify (25) by constructing a full time solution $(\mathcal{R},\mathcal{I})$ by using the ω-limit set of the persistent solution (R,I) such that

$$I_m=\mathcal{I}(0)=\min_{t\in\mathbb{R}}\mathcal{I}(t),$$
$$I_M\ge\max_{t\in\mathbb{R}}\mathcal{I}(t)$$
$$R_m\le\mathcal{R}(t)\le R_M,\qquad \text{for all } t\in\mathbb{R}.$$

It follows that $\dot{\mathcal{I}}(0)=0$, and consequently,

$$0=-a_2I_m+a_3\mathcal{R}(-\tau_R). \quad (27)$$

By the definition of $\mathcal{R}(t)$,

$$R_m\le\mathcal{R}(-\tau_R),$$

we have

$$-a_2I_m+a_3\mathcal{R}(-\tau_R)\ge-a_2I_m+a_3R_m \quad (28)$$

Therefore,

$$a_3R_m\le a_2I_m \quad (29)$$

From the proof of Theorem 5,

$$\mathcal{R}(-\tau_R)\le R_M,$$

which implies that

$$a_2I_M=a_3\mathcal{R}(-\tau_R)\le a_3R_M. \quad (30)$$

Then, it is clear that

$$a_3R_m\le a_2I_m\le a_2I_M\le a_3R_M.$$

In order to verify (26), we again construct a full time solution $(\boldsymbol{R},\boldsymbol{I})$ by using the ω-limit set of the persistent solution (R,I) such that

$$R_M=\boldsymbol{R}(0)=\max_{t\in\mathbb{R}}\boldsymbol{R}(t),$$
$$R_m\le\min_{t\in\mathbb{R}}\boldsymbol{R}(t)$$
$$I_m\le\boldsymbol{I}(t)\le I_M,\qquad \text{for all } t\in\mathbb{R}$$

By the definition of $\boldsymbol{I}(t)$, $I_m\le\boldsymbol{I}(-\tau_I)$ and from (8), we obtain that

$$a_4=a_1R_M+\frac{b_1R_M}{b_2+R_M}-\frac{b_3R_M^2}{(b_4R_M+\mathbf{I}(-\tau_I))^2}$$

$$\ge a_1R_M+\frac{b_1R_M}{b_2+R_M}-\frac{b_3R_M^2}{(b_4R_M+I_m)^2} \quad (31)$$

Since $\boldsymbol{I}(-\tau_I)\le I_M$ and from (22), we have

$$a_4=a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m}{(b_4R_m+\mathbf{I}(-\tau_I))^2}$$

$$\le a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m}{(b_4R_m+I_M)^2} \quad (32)$$

According to (31), we therefore obtain

$$a_1R_M+\frac{b_1R_M}{b_2+R_M}-\frac{b_3R_M^2}{(b_4R_M+I_m)^2}$$

$$\le a_4\le a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m^2}{(b_4R_m+I_M)^2}. \quad \blacksquare$$

Lemma 7 By the assumption in Theorem 5, the following conditions are equivalent:

$$i)I_M=I_b,\ ii)R_m=R_b,\ iii)I_m=I_b,\ iv)R_M=R_b$$

Proof We first prove that $i)$ implies $ii)$ by supposing that $I_M=I_b$. Then, from the second inequality in (26),

$$a_4\le a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m}{(b_4R_m+I_b)^2}. \quad (33)$$

Since the function

$$a_1R+\frac{b_1R}{b_2+R}-\frac{b_3R^2}{(b_4R+I)^2}$$

is increasing in R when $\frac{b_1}{b_3}$ is sufficiently large, we have from (33) that

$$a_1R_m+\frac{b_1R_m}{b_2+R_m}-\frac{b_3R_m}{(b_4R_m+I_b)^2}$$

$$\le a_1R_b+\frac{b_1R_b}{b_2+R_b}-\frac{b_3R_b}{(b_4R_b+I_b)^2}=a_4$$

Consequently, by (33), $R_m=R_b$.

To show that $ii)$ implies $iii)$, assume that $R_m=R_b$. We construct a full time solution $(\mathcal{R},\mathcal{I})$ by using the ω-limit set of the persistent solution (R,I) such that

$$I_m=\mathcal{I}(0)=\min_{t\in\mathbb{R}}\mathcal{I}(t),$$
$$I_M\ge\max_{t\in\mathbb{R}}\mathcal{I}(t)$$
$$R_m\le\mathcal{R}(t)\le R_M,\qquad \text{for all } t\in\mathbb{R}.$$

It follows from (9) and (29) that

$$I_b = \frac{a_3}{a_2} R_b = \frac{a_3}{a_2} R_m \le I_m . \tag{34}$$

However, we have $I_m \le I_b$. Hence, we must have $I_m = I_b$.

To show that *iii*) implies *iv*), we suppose $I_m = I_b$. We consider the first inequality in (26),

$$a_4 \ge a_1 R_M + \frac{b_1 R_M}{b_2 + R_M} - \frac{b_3 R_M^2}{(b_4 R_M + I_b)^2} \tag{35}$$

Since the function $a_1 R + \frac{b_1 R}{b_2 + R} - \frac{b_3 R^2}{(b_4 R + I)^2}$ is increasing in R, it follows that

$$\begin{aligned} a_1 R_M + \frac{b_1 R_M}{b_2 + R_M} - \frac{b_3 R_M^2}{(b_4 R_M + I_b)^2} \\ \ge a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b}{(b_4 R_b + I_b)^2} = a_4 \end{aligned} \tag{36}$$

Hence, we have $R_M = R_b$.

Now, to show that *iv*) implies *i*), assume that $R_m = R_b$. From (9) and (25), we have

$$I_b = \frac{a_3}{a_2} R_b = \frac{a_3}{a_2} R_M \le I_M \tag{37}$$

Again, since $I_b \le I_M$, we have $I_M = I_b$. ■

From Lemma 7, we have that if $R \le R_b$, or $R \ge R_b$, or $I \le I_b$, or $I \ge I_b$ for all *t*, then we will have $\lim_{t \to \infty} I(t) = I_b$ and $\lim_{t \to \infty} R(t) = R_b$. This means that if the system solution does not oscillate about the equilibrium point (R_b, I_b) then it must tend eventually toward the steady state. In other words, all solutions must oscillate about the steady state levels or else they converge to (R_b, I_b) as time passes.

Remark 8 Every non-constant periodic solution of (8) and (9) must oscillate around the basal level (R_b, I_b); otherwise, the inequality (25) or (26) forces all strictly bounded positive solutions to converge to (R_b, I_b).

IV. Global Stability of the Basal Level

We next give conditions which ensure the global stability of the model system.

Theorem 9 Suppose

$$a_1 > \max(M_1 I_b, M_2 I_b), \tag{38}$$

$$L_1 = \frac{a_3}{a_2}, \tag{39}$$

$$L_2 = \frac{M_1 R_b}{a_1 - M_1 R_b}, \tag{40}$$

$$L_3 = \frac{M_2 R_b}{a_1 - M_2 R_b}, \tag{41}$$

where

$$M_1 = \frac{b_3}{(b_4 R_b + I_b)(b_4 R_m + I_M)} \left[\frac{R_b}{b_4 R_b + I_b} + \frac{R_m}{b_4 R_m + I_M} \right],$$

$$M_2 = \frac{b_3}{(b_4 R_b + I_b)(b_4 R_M + I_M)} \left[\frac{R_b}{b_4 R_b + I_b} + \frac{R_M}{b_4 R_M + I_m} \right].$$

Then,

$$I_M - I_b \le L_1 \left(R_M - R_b \right), \tag{42}$$

$$I_b - I_m \le L_1 \left(R_b - R_m \right), \tag{43}$$

$$R_b - R_m \le L_2 \left(I_M - I_b \right), \tag{44}$$

$$R_M - R_b \le L_3 \left(I_b - I_M \right). \tag{45}$$

If $L_1^2 L_2 L_3 < 1$, then every positive solution of the system (8)–(9) converges to the positive equilibrium, or equivalently, the basal levels are globally attractive.

Proof Let (R, I) be a solution of system (8)–(9). We now construct a full time solution $(\mathcal{R}, \mathcal{I})$ such that

$$I_M = \mathcal{I}(0) = \max_{t \in \mathbb{R}} \mathcal{I}(t),$$

$$I_m \le \min_{t \in \mathbb{R}} \mathcal{I}(t)$$

$$R_m \le \mathcal{R}(t) \le R_M, \qquad \text{for all } t \in \mathbb{R}.$$

As before, from (13) and (21), we have

$$I_M - I_b = \frac{a_3}{a_2} \left(\mathcal{R}(-\tau_R) - R_b \right) \le L_1 \left(R_M - R_b \right),$$

with L_1 as in (39). In the same way, we again construct a full time solution $(\mathcal{R}, \mathcal{I})$ such that

$$I_m = \mathcal{I}(0) = \min_{t \in \mathbb{R}} \mathcal{I}(t),$$

$$I_M \ge \max_{t \in \mathbb{R}} \mathcal{I}(t)$$

$$R_m \le \mathcal{R}(t) \le R_M, \qquad \text{for all } t \in \mathbb{R}.$$

It follows from (13) and (25) that

$$I_b - I_m = \frac{a_3}{a_2}\left(R_b - \mathcal{R}(-\tau_R)\right) \le L_1\left(R_b - R_m\right),$$

with L_1 as in (39).

On the other hand, as before, we can construct a full time solution $(\mathbf{R},\mathbf{I})$ such that

$$R_m = \mathbf{R}(0) = \min_{t\in\mathbb{R}} \mathbf{R}(t),$$
$$R_M \ge \max_{t\in\mathbb{R}} \mathbf{R}(t)$$
$$I_m \le \mathbf{I}(t) \le I_M, \qquad \text{for all } t \in \mathbb{R}$$

Then $\dot{\mathbf{R}}(0) = 0$ and (12) holds, and consequently,

$$a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2} = a_4$$
$$= a_1 R_m + \frac{b_1 R_m}{b_2 + R_m} - \frac{b_3 R_m^2}{(b_4 R_m + \mathbf{I}(-\tau_I))^2}.$$

By the definition of $\mathbf{I}(t)$, $\mathbf{I}(-\tau_I) \le I_M$, we have

$$a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2}$$
$$\le a_1 R_m + \frac{b_1 R_m}{b_2 + R_m} - \frac{b_3 R_m^2}{(b_4 R_m + I_M)^2}.$$

Then,

$$a_1\left(R_b - R_m\right) \le -\left(\frac{b_1 R_b}{b_2 + R_b} - \frac{b_1 R_m}{b_2 + R_m}\right) + \left(\frac{b_3 R_b^2}{(b_4 R_b + I_b)^2} - \frac{b_3 R_m^2}{(b_4 R_m + I_M)^2}\right)$$
$$\le \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2} - \frac{b_3 R_m^2}{(b_4 R_m + I_M)^2} = M_1(R_b I_M - R_m I_b)$$

That is,

$$a_1\left(R_b - R_m\right) \le M_1(R_b I_M - R_m I_b)$$
$$\le M_1(R_b I_M - R_b I_b + R_b I_b - R_m I_b)$$
$$= M_1(R_b(I_M - I_b) + I_b(R_b - R_m)),$$

or

$$\left(a_1 - M_1 I_b\right)\left(R_b - R_m\right) \le M_1 R_b (I_M - I_b).$$

Therefore,

$$R_b - R_m \le L_2\left(I_M - I_b\right)$$

provided (38) holds.

We again construct a full time solution $(\boldsymbol{R},\boldsymbol{I})$ such that

$$R_M = \boldsymbol{R}(0) = \max_{t\in\mathbb{R}} \boldsymbol{R}(t),$$
$$R_m \le \min_{t\in\mathbb{R}} \boldsymbol{R}(t)$$
$$I_m \le \boldsymbol{I}(t) \le I_M, \qquad \text{for all } t \in \mathbb{R}$$

Then $\dot{\mathbf{R}}(0) = 0$ and (12) holds, and consequently,

$$a_1 R_M + \frac{b_1 R_M}{b_2 + R_M} - \frac{b_3 R_M^2}{(b_4 R_M + \mathbf{I}(t-\tau_I))^2} = a_4$$
$$= a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2}$$

By the definition of $\boldsymbol{I}(t)$, $I_m \le \boldsymbol{I}(-\tau_I)$ and thus,

$$a_1 R_M + \frac{b_1 R_M}{b_2 + R_M} - \frac{b_3 R_M^2}{(b_4 R_M + \mathbf{I}(-\tau_I))^2} \le a_1 R_b + \frac{b_1 R_b}{b_2 + R_b} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2}$$

Then,

$$a_1\left(R_M - R_b\right) \le -\left(\frac{b_1 R_M}{b_2 + R_M} - \frac{b_1 R_b}{b_2 + R_b}\right) + \left(\frac{b_3 R_M^2}{(b_4 R_M + I_m)^2} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2}\right)$$
$$\le \frac{b_3 R_M^2}{(b_4 R_M + I_m)^2} - \frac{b_3 R_b^2}{(b_4 R_b + I_b)^2} = M_2(R_M I_b - R_b I_M)$$

That is,

$$a_1\left(R_M - R_b\right) \le M_2(R_M I_b - R_b I_M)$$
$$\le M_2(R_M I_b - R_b I_b + R_b I_b - R_b I_M)$$
$$\le M_2(I_b(R_M - R_b) + R_b(I_b - I_m)),$$

or

$$\left(a_1 - M_2 I_b\right)\left(R_M - R_b\right) \le M_1 R_b (I_b - I_M).$$

Therefore,

$$R_M - R_b \le L_2\left(I_b - I_M\right)$$

provided (39) holds.

We can conclude that

$$I_M - I_b \le L_1^2 L_2 L_3\left(I_M - I_b\right),$$

and therefore, if $L_1^2 L_2 L_3 < 1$, it happens that $I_M = I_b$ from which the theorem is proven. ■

V. Conclusion

It has been shown that the model of the signal transduction pathway consisting of two delay differential equations (8)–(9) is uniformly persistent.

Moreover, it appears that the delays τ_I and τ_R do not appear to explicitly impact on the persistent characteristics of this system. At least, no restrictions on the delays are needed for persistence or stability of the system. However, if we consider the proofs of our results, we see that the values of *R* and *I*, the densities of activated GPCR and inhibitor protein, $R(-\tau_I)$, $I(-\tau_I)$, $R(-\tau_R)$, and $I(-\tau_R)$, are related to the quantities R_m, R_M, I_m, and I_M. These in turns bound the values of the basal levels R_b, and I_b, while R_b, and I_b appear in the expressions of the quantities L_1 to L_3 in Theorem 9 that delineates the parametric regions where the equilibrium of the system is globally stable. Thus, the delays τ_I and τ_R in fact implicitly effect the stability behaviour of the signaling pathway.

Our model analysis therefore indicates that the process outcome can be potentially engineered by controlling the levels of the densities of activated G-protein coupled receptors and inhibitors during the initial time intervals $(-\tau_I, 0)$ and $(\tau_R, 0)$ so that the global stability may be preserved or possibly destroyed, whatever is the desired outcome. In fact, we found that a solution of the model system must oscillate about the steady state level (R_b, I_b) unless it converges to the equilibrium point as time passes. When such oscillatory behavior may occurs and how the delays τ_I and τ_R effect the behavior of these oscillating solutions are subjects for future investigations.

The conclusions reached in this study are expected to bear important implications for experimental investigations to identify the mechanisms for biological memory and for the development of therapeutic strategies to modulate signaling network responses in the setting of human diseases.

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