

Boolean Modelling of Catecholamine synthesis and degradation pathways

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Abstract:

Catecholamines are the common name used for the hormones like epinephrine (adrenaline), norepinephrine (noradrenaline), and dopamine. Catecholamines are neurotransmitters which are involved in various regulatory processes and its involvement in various disorders like cardiovascular, neurological, psychiatric has been extensively studied. The qualitative dynamics of the catecholamine synthesis and degradation pathway is studied here using synchronous and asynchronous analysis with the help of Boolean modelling. The effect of various inhibitors on the key enzyme viz. Monoamine oxidase (MAO) of the degradation pathway was also studied and the network was validated. The results obtained gives an important insight into the state cycle and steady state of the catecholamine regulatory network.

Introduction

Catecholamines are the class of hormones and chemical neurotransmitters which are involved in multiple regulatory processes. These are monoamines which contains a catechol and a side chain amine. The key sites of catecholamine synthesis are chromaffin cells of adrenal medulla but some of these cells are also found in heart, liver, kidney, gonads, adrenergic neurons of the post ganglionic sympathetic system and central nervous system[1][2][3]. The release of these catecholamines from the adrenal medulla takes place in response to fight-or-flight or in stress conditions. . Plasma and urinary levels of catecholamines has long been known as a marker for neuroendocrine tumours like pheochromocytomas[4].

There are four major processes/pathways associated with catecholamines viz. synthesis, storage, secretion and degradation. The catecholamine biosynthesis is initiated by tyrosine which itself is produced from phenylalanine

through the activity of phenylalanine hydroxylase in the liver. Monoamine Oxidases (MAO) are the important enzymes which are involved in deamination of these catecholamines and hence plays a major role in its metabolism.

The results here shows the Boolean modelling[5] for the two important pathways i.e. synthesis and degradation of catecholamines. The complete pathway for synthesis and degradation is depicted in Figure 3[6].

Background

A systems biology approach was used to model the entire network. The already modelled cholesterol synthesis pathway was used as the basis for the present model. A similar approach involving Boolean modelling was incorporated in which activation corresponds to “ON” and inhibition corresponds to “OFF” state.

¹ This is a course project done under supervision of Dr. Karthik Raman

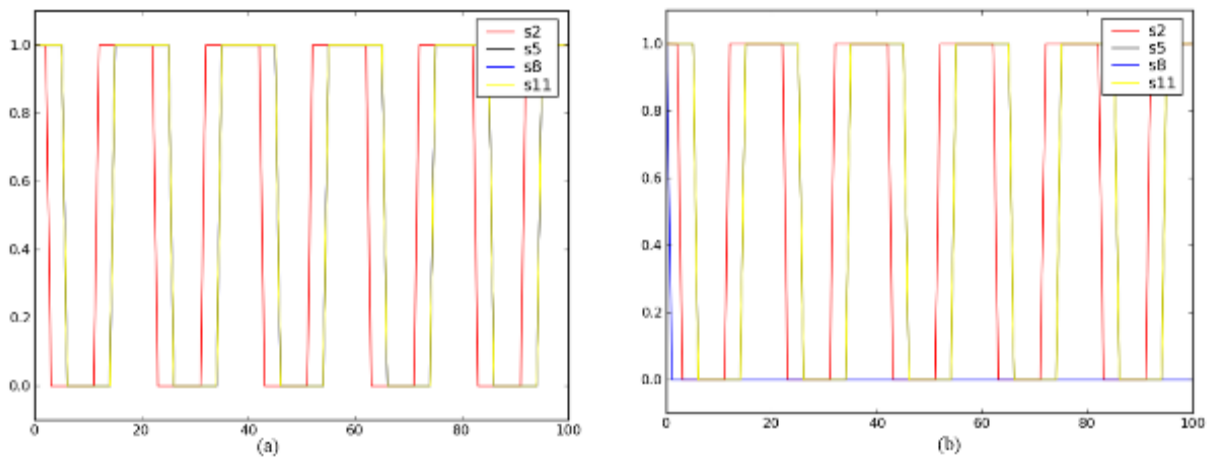


Figure 1: (a) Synchronous analysis of the cholesterol model in the absence of statins and behaviour of key enzymes. The yellow colour represents the overlap of the three enzymes represented by three different colours. (b) Synchronous analysis of the cholesterol pathway in the presence of statins and behaviour of key enzymes. The line in blue colour represents the enzyme HMG-CoA Synthase

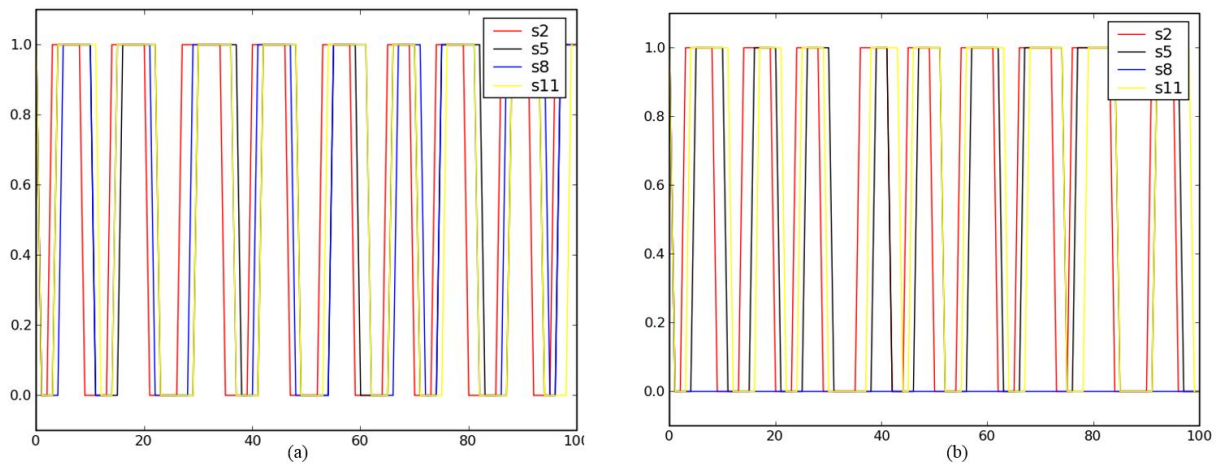


Figure 2: (a) Asynchronous analysis of the cholesterol model in the absence of statins and behaviour of key enzymes. (b) Asynchronous analysis of the cholesterol pathway in the presence of statins and behaviour of key enzymes. The line in blue colour represents the enzyme HMG-CoA Synthase

The synchronous and asynchronous analysis for the cholesterol pathway[7] was performed to check for the state cycle and steady state of the pathway. The Boolean equations were written as per the cholesterol model given by Kervizic *et al.* 2008. The effect of statin on the model along with the key metabolites and enzymes like mSREBP (s2), HMG-CoA Synthase (s5), HMG CoA-Reductase (s6) and FPP Synthase (s11) were studied. The synchronous (Figure 1) and asynchronous analysis (Figure 2) for the same is shown.

It was also observed that the system was reaching equilibrium above 1000 iterations with the state cycle length of 20. A similar

approach was used to model the system of research i.e. the catecholamine synthesis and degradation pathway.

Results and Discussions:

The catecholamine synthesis and degradation pathway is depicted in detail in Figure 3. The aim is to build a regulatory model which can be used to find the equilibrium and steady state of the system.

The Boolean equations were derived based on Figure 3. Two additional compounds (inhibitors) were added to the model to study the role of widely known enzyme Monoamine Oxidase (MAO) on the steady state of the system. The enzyme is involved

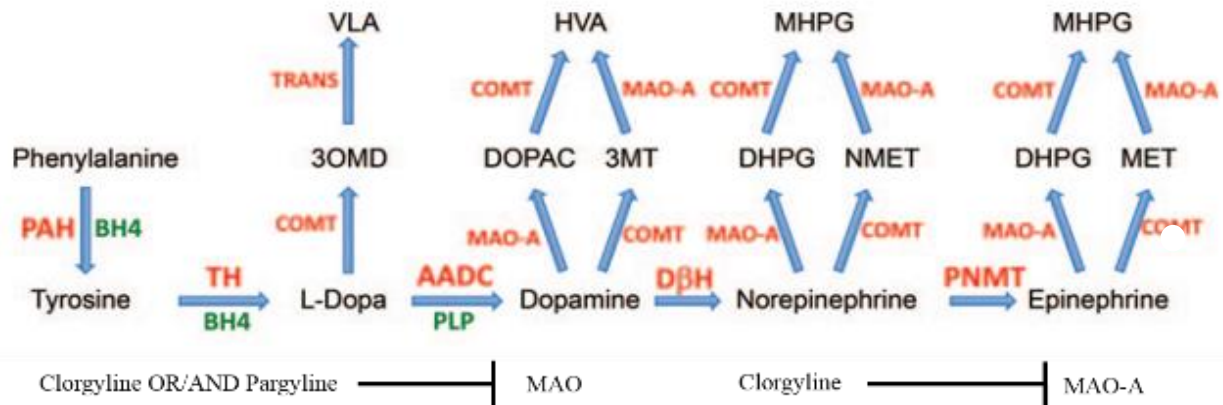


Figure 3: The detailed catecholamine synthesis and degradation pathway. Clorgyline and Pargyline are specific inhibitors of MAO-A and MAO-B respectively.

Table 1 : Boolean Equations for Catecholamine metabolism pathway

1.	$TH(t+1) = TH(t)$
2.	$L\text{-Dopa}(t+1) = \text{Tyrosine}(t) \text{ and } TH(t) \text{ and } BH4(t)$
3.	$Dopamine(t+1) = L\text{-Dopa}(t) \text{ and } AADC(t) \text{ and } PLP(t)$
4.	$DBH(t+1) = DBH(t)$
5.	$Dopamine(t+1) = Dopamine(t)$
6.	$Norepinephrine(t+1) = DBH(t) \text{ and } Dopamine(t)$
7.	$PNMT(t+1) = PNMT(t)$
8.	$Epinephrine(t+1) = PNMT(t) \text{ and } Norepinephrine(t)$
9.	$COMT(t+1) = COMT(t)$
10.	$3OMD(t+1) = COMT(t) \text{ and } L\text{-DOPA}(t)$
11.	$TRANS(t+1) = TRANS(t)$
12.	$VLA(t+1) = 3OMD(t) \text{ and } TRANS(t)$
13.	$MAO\text{-}A(t+1) = \text{not } Clorgyline(t)$
14.	$DOPAC(t+1) = MAO(t) \text{ and } Dopamine(t)$
15.	$3MT(t+1) = COMT(t) \text{ and } Dopamine(t)$
16.	$HVA(t+1) = (COMT(t) \text{ and } DPAC(t)) \text{ or } (MAO(t) \text{ and } 3MT(t))$
17.	$DHPG(t+1) = (MAO\text{-}A(t) \text{ and } Norepinephrine) \text{ or } (MAO\text{-}A(t) \text{ and } Epinephrine(t))$
18.	$NMET(t+1) = Norepinephrine(t) \text{ and } COMT(t)$
19.	$MET(t+1) = COMT(t) \text{ and } Epinephrine(t)$
20.	$MHPG(t+1) = (DHPG(t) \text{ and } COMT(t)) \text{ or } (NMET(t) \text{ and } MAO\text{-}A(t)) \text{ or } (MET(t) \text{ and } MAO\text{-}A(t))$
21.	$BH4(t+1) = BH4(t)$
22.	$PLP(t+1) = PLP(t)$
23.	$Clorgyline(t+1) = Clorgyline(t)$
24.	$Pargyline(t+1) = Pargyline(t)$
25.	$MAO(t+1) = \text{not } (Clorgyline(t) \text{ or } Pargyline(t))$

in catecholamine degradation pathway. Here, the effects of the two types of Monoamine Oxidases (MAO-A and MAO-B) were investigated. All the three catecholamines i.e. Dopamine, Epinephrine and Norepinephrine can be degraded by MAO-A while only Dopamine is degraded

by MAO-B. Thus, to make our analysis simpler, MAO is used instead of both MAO-A or MAO-B for the degradation pathway of Dopamine to HVA. Clorgyline is a widely known inhibitor which is specific to MAO-A while Pargyline is a specific inhibitor to MAO-B. Since, for this analysis MAO is

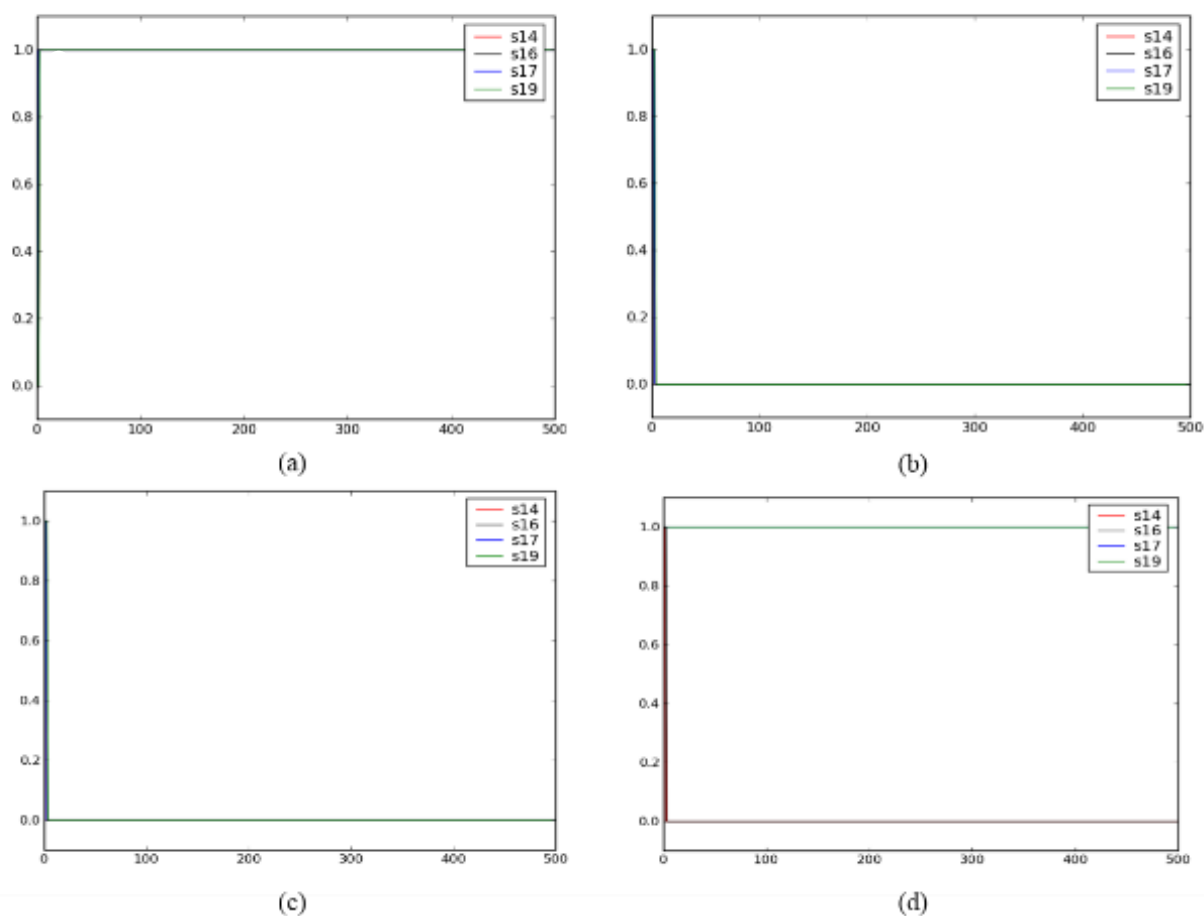


Figure 4: Modelling of catecholamine pathway. The four substrates are DOPAC (s14), HVA(s16), DHPG(s17), MHPG(s19) are shown. (a) Graph obtained in absence of both the inhibitors. (b) Graph representing presence of both the inhibitors. (c) Graph in presence of Clorgyline inhibitor. (d) Graph obtained in presence of Pargyline inhibitor.

considered as either of MAO-A or MAO-B. Thus, MAO can be inhibited by both Clorgyline and Pargyline. The Boolean equations are developed keeping the above stated assumption in mind. The detailed Boolean equations are shown in Table 1. The effect of other metabolites and enzymes on this pathway is not considered here and it is assumed that the metabolites/enzymes are self-renewed after time t , if it is shown as not getting affected by any other enzymes in the system considered. It is also assumed that all other small metabolites like ions are in excess and doesn't affect the model in any respect. Based on the above listed Boolean equations, simulation was done to obtain the working model.

Synchronous analysis approach was used to model the pathway. In order to confirm the

correctness of the model, the values of the MAO-A dependent metabolites were monitored in the presence or absence of both the inhibitors. There are four cases possible which is clearly shown in Figure 4. It was easily noticed that in the presence of Clorgyline inhibitor, production of all the substrates were inhibited while in the presence of Pargyline only HVA and DOPAC were affected. Hence, the results suggests that the Boolean modelling of the network was perfectly achieved.

It was also observed that the system was reaching steady state at random input values and at 500 iterations. Asynchronous analysis was also tried but could not obtain any convincing results. In future, it would be interesting to perform asynchronous analysis and stochastic update on the model

and get relevant information from the system about its steady state and equilibrium state. The lack of quantitative information like binding constants, kinetic parameters etc. for any pathway makes Boolean model a very useful tool to predict the behaviour of a particular system. We can very well rely on the qualitative description of the type “A activates (inhibits) B” in this type of model[8].

On comparing Boolean modelling with other methods like Flux Balance Analysis (FBA) suggests that FBA alone is not a very good method to model a regulatory network. FBA is a very well-known method for determining the metabolic fluxes at steady state. FBA doesn't account for various regulatory effects like gene expression or activation of enzymes by signal transduction. However, there are few exceptions which are basically the modified version of FBA that captures the full dynamic of the cell like Integrated Hybrid Models[9] incorporating Petri nets, metabolic and signalling networks are modelled using Petri nets[10] and Boolean networks which has been widely used to model regulatory networks along with the protein signalling networks[11][12].

In short, in order to get the best results, one can use a combination of FBA and Boolean networks to model this type of complex regulatory networks.

Conclusions:

Catecholamine metabolism pathway was modelled for the first time using Boolean networks. Synchronous and asynchronous analysis was studied in great detail using cholesterol and catecholamine regulatory network. Further improvements can be made by incorporating a hybrid technique to model the system.

Acknowledgements:

The author would like to acknowledge Dr. Karthik Raman for helping him with the project whenever required. The author would also like to acknowledge Mr. Vinayak Gupta, Cardiovascular Genetics Lab from IIT Madras for helping in clarifying the concepts involved in the project. I would also like to thank my friends Miss Nandita Damaraju (4th year undergraduate student from Department of Biotechnology, IIT Madras), Mr. Eddy Hudson (4th year undergraduate student from Department of Biotechnology, IIT Madras), and Mr. Aditya Sapate (4th year undergraduate student from Department of Computer Science & Engineering, IIT Madras) for their useful inputs in the project.

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