Original Article

Providing a Stochastic Petri Net Model for Interactions of the Immune System and B16-F10 Tumor Cells in order to Investigate the Effect of Myeloid-Derived Suppressor Cells (MDSC) on Behavioral States of Tumor

S. Shafiekhani[1,2], S. Rahbar[1,2], F. Akbarian[1,2], J. Hadjati[3], A.Allahverdy[4], A. H. Jafari[1,2*]

1- Department of Medical Physics and Biomedical Engineering, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran.

2- Research Center for Biomedical Technologies and Robotics (RCBTR), Tehran University of Medical Sciences, Tehran, Iran.

3- Department of Medical Immunology, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran.

4- Radiology Department, Allied Faculty, Mazandaran University of Medical Sciences, Sari, Iran.

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A B S T R A C T

Purpose- Using mathematical models for cancer treatment has had excellent results in recent years. Modeling of the tumor-immune interactions is possible by several mathematical models. Stochastic models such as Stochastic Petri Net (SPN) consider the random effects and uncertainty in the biological environments. Therefore, they are good choices for the simulation of biological systems, specially the complex dynamical network of tumor-immune interactions.

Methods- In this study, we have modeled the interactions of the B16-F10 tumor cells, Cytotoxic T Cells (CTL) and MDSC by SPN. By a systematic search on immunology resources, we identified the behaviors, characteristics, and effective interactions between these cells. We used SPN to construct the dynamics of these cells, therefore a dynamical network of tumor-immune interactions (DNTII) has been made. By considering these cells as places and all interactions as transitions of SPN, we can simulate this complex biological network. The model has some control parameters that their regulation causes DNTII to mimic different behaviors of tumor-immune system, such as tumor escape and degradation.

Results- The model can properly simulate complete dynamical network of tumor-immune interactions compared to the biological reality. This model is capable to represent different behavior of tumor-immune system such as tumor escape from immune response, overcoming the immune system on the tumor cells.

Conclusion- By using this model, we can test different immunology hypothesis in a simulation environment without spending any time and money.

1. Introduction

The main challenge in the biology is exploring the basic rules governing the structure and function of biological networks [1]. One of the most complex networks is a single cell [2]. The interactions between different cells follow complicated rules that mathematical modeling can help to identify. This complexity

*Corresponding Author:
Amir Homayoun Jafari, PhD
Department of Medical Physics & Biomedical Engineering, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran.
Tel: (+98)2166466383 / Fax: (+98)2166482654
Email: h_jafari@tums.ac.ir

increases in cancer environment. Therefore, the nonlinear Dynamical Network of Tumor-Immune Interactions may exhibit different behaviors [3]. There are some key components in DNTII that control the dynamic of network with a dominant effect. According to biological literatures, tumor cells, Cytotoxic T cells (CTL) and MDSCs are key components of DNTII [4]. Some disorders and perturbations in genetic factors cause a healthy cell to transfer to a tumor cell with non-standard functions. The Main immune response against tumor cell is expressed by CTL. In fact, in micro-environment of tumor, tumor cells release some antigens that are recognizable by CTLs, then CTLs immigrate to tumor site and encounter. The increased immune response creates an inflammatory environment, therefore MDSCs are produced and decrease the immune response. This preamble stated the interactions between key components of DNTII concisely [5].

Mathematical modeling of biological networks is a very effective tool which is widely used for the system identification and optimization of drug delivery systems [6]. A number of mathematical methods for simulating the DNTII have been proposed, for example [7-10] are Ordinary Differential Equation (ODE) models that describe the average behavior of DNTIIs without regarding stochastic effects. Because of determinism rigidity of ODE models, we prefer to use stochastic models to capture the inherent randomness of DNTII. One of the best models in the system biology and mathematical modeling of DNTII is Petri Nets (PN) [11-13]. PN is a comprehensive mathematical model which describes the dynamical network quantitatively. SPN is an extended form of PN which regards to stochastic nature of interactions. Therefore, SPN is suitable for modeling of DNTII [14].

In this study, we have constructed a SPN model to simulate the DNTII. This model has control parameters to display different behaviors according to the reality of the biological system.

2. Methods

In the following, the biological background of tumor-immune system and the concept of stochastic petri net is expressed, then the model is described in detail.

2.1. Biological Background of Tumor-Immune System

Tumors are generally the result of uncontrolled growth of host tissue cells. In normal tissues, growth rate of cells is controlled by several mechanisms. Many factors such as genetic mutations due to carcinogens may cause failure of these mechanisms and disrupt the cellular metabolic processes. Tumor cells are mainly able to proliferate in a shorter time than normal cells. In our model, it is assumed that the tumor cells can proliferate autonomously with a constant rate and are not responsive to apoptosis signals from microenvironment. Alternatively, it is assumed that the only agent which eliminates tumor cells from the microenvironment is the effector cells of immune system [15].

Effector cells are the relatively short-lived activated cells that respond to adaptive immune stimulus and referred to the collection of plasma cells which secrete antibodies, and activated T cells which include cytotoxic T cells and helper T cells and carry out cell-mediated responses. Cytotoxic T cells begin to exist at a high frequency during an adaptive immune response in the tumor microenvironment following a recognition of tumor antigens by antigen presenting cells. Effector cells are steadily recruited to the tumor microenvironment to sustain an effective response to tumor growth. In agreement with this function of adaptive immune system, we have considered a recruitment of effector cells due to the presence of tumor. The recruitment rate of the effectors is dependent on the tumor population. Present effector cells in microenvironment are able to encounter tumor cells and may eliminate them completely. Like many other cells of immune system, effector cells may undergo apoptosis after a finite number of encounters or by aging. To model this attribute, we have assumed a half-life of effector cells.

Similar to chronic infections, tumors can cause altered haematopoiesis which in long term leads to an expansion of immunosuppressive activities of immune system. One of these immunosuppressive mechanisms is the differentiation of myeloid cells formed in the bone marrow toward MDSCs. MDSCs differentiate to dendritic cells, macrophages and neutrophils in healthy individuals. MDSCs

suppressor effect lies in their ability to inhibit T cell proliferation and activation. They also accelerate tumor progression and metastasis. Growing tumors secret a variety of chemokines and molecules which are necessary for myeloid cells differentiation to MDSCs. In order to simulate this effect in the model, we have considered MDSC generation as a result of tumor growth [16, 17].

2.2. Concept of Stochastic Petri Net (SPN)

The SPN model is implemented in Matlab environment. A simple PN is composed of two groups of nodes that are places and transitions. Also there are some arcs with specified weight that connect places to transitions and transitions to places. Places and transitions describe the cells (or proteins) and interaction between the places respectively. Places and transitions express the static part of PN and the dynamic of PN is modeled by flowing of tokens between places through transitions. In fact, tokens describe the amount of each place, for example the concentration of a specific protein or the population of a cell [18].

Each transition has some input places and output places. When tokens of input places of transition t_i be more than the weight of related arcs between that input places and transition, t_i , this transition is enabled. In PN, enabled transition is fired immediately, and in SPN is fired with a stochastic delay with an exponential probability distribution. After firing of a transition, t_i, tokens flow from input places of this transition to output places. The amount of transitioned tokens between input places and output places correspond to the weight of related arcs between places and transitions. For example, in PN of Figure 1, if transition T1 is fired, the amount of w_{11} tokens and w_{21} tokens from places A and B are reduced respectively and the amount of w'_{13} tokens are added to place C [18].

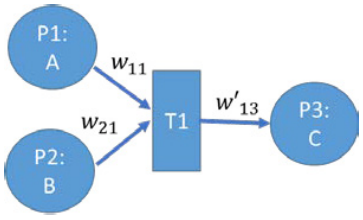

Figure 1. A PN with three places, one transition and three arcs relate places and transition. Circles and square depict places and transition respectively. The values of w_{11},w_{12} and w'_{13} are the weights of preplaces and post places of transition T1.

As it was mentioned, in SPN, enabled transitions are fired with a delay ($t \in T$) which is stochastic variable ($X_t \in [0,1)$) with the following probability distribution:

$$f_{X_t}(\tau) = \gamma_t(m).\exp(-\gamma_t(m).\tau) \qquad \tau \geq 0 \qquad (1)$$

$$\text{Mass action(k)} = \gamma \qquad (2)$$

In Equation (1), γ is a function of marking of SPN which is calculated with the mass action kinetic law. The mass action kinetic law for reaction A+B →C is as follow:

$$\frac{d}{dt}[C] = k\,[A]\,[B] = \gamma \qquad (3)$$

2.3. SPN for Modeling of DNTII

We have considered MDSC, CTL and Tumor cell as places of SPN and 7 transitions for modeling of their interactions and execution of the cells as depicted in Figure 2. First, we have assigned the initial values to the cells, then run SPN model to achieve the dynamics of the system. The dynamics of the model is achieved by firing of only one transition from 7 transitions at any time step. Each transition has a rate which is computed by the mass action law according to Equations (2) and (3). Then, these rates are normalized to their summation. These normalized rates describe the probability of firing of each enabled transition. Therefore, with subsequent firing of enabled transitions during time, the dynamic of model is completed.

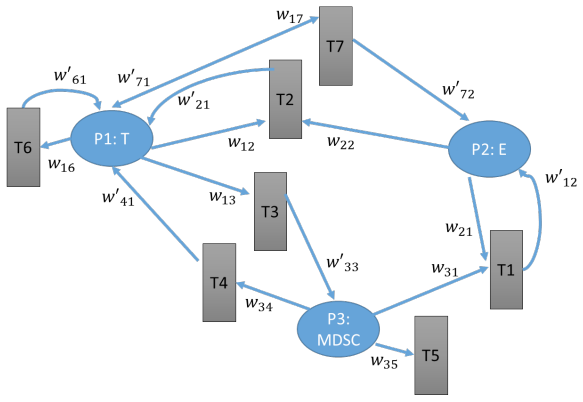

Figure 2. SPN of DNTII. Places P1, P2 and P3 (circles) describe tumor cell, effector cell and MDSC respectively and squares T1 to T7 describe different interactions between cells according to the immunology knowledge.

As mentioned before, each of these transitions describe an interaction between places. In other words, these places apply their features and behaviors through transitions. All transitions and their biological concept is described in Table 1. The model has two outcome strategies: firstly, the tumor escape from the immune response and secondly, the tumor elimination by the immune system. Changing control parameters generates different scenarios according to biological reality in a tumor microenvironment. The weights of arcs (w_{ij}, w'_{ij}, i, j=1,2,…,7) and constant rates of transitions (k_i, i=1,2,…,7) are the control parameters of the model.

Table 1. Description of reactants and reactions of DNTII.

Reactants	Transition	Description of Transition	Weight of the arcs associated with the corresponding transition for first strategy (tumor escape)	Weight of the arcs associated with the corresponding transition for second strategy (tumor degradation)
MDSC, Effector	T1	Inhibition of effector cells by MDSC in tumor microenvironment.	$w_{21}=2, w_{31}=1,$ $w'_{12}=1$ $k_1=10^{-6}$	$w_{21}=2, w_{31}=1,$ $w'_{12}=1$ $k_1=10^{-6}$
Tumor, Effector	T2	Killing tumor cells by effector cells.	$w_{12}=2, w_{22}=1$ $w'_{21}=1$ $k_2=10^{-4}$	$w_{12}=2, w_{22}=1$ $w'_{21}=1$ $k_2=10^{-4}$
Tumor, MDSC	T3	Expression of MDSC by tumor cells.	$w_{13}=1, w'_{33}=1$ $k_3=8*10^{-7}$	$w_{13}=1, w'_{33}=1$ $k_3=0.8$
MDSC, Tumor	T4	Expression of tumor cells by MDSC.	$w_{34}=1, w'_{41}=2$ $k_4=0.8$	$w_{34}=1, w'_{41}=1$ $k_4=0.8$
MDSC	T5	Random degradation of MDSC.	$w_{35}=1$ $k_5=1$	$w_{35}=1$ $k_5=1$
Tumor	T6	Self-reproduction of tumor cells.	$w_{16}=1, w'_{61}=4$ $k_6=0.7$	$w_{16}=1, w'_{61}=2$ $k_6=1$
Tumor, Effector	T7	Expression of effector cells due to the presence of tumor cells.	$w_{17}=1, w'_{71}=1$ $k_7=1$	$w_{17}=1, w'_{71}=1$ $k_7=1$

According to the literature, half-life of MDSC and effector cells is approximated to be 34 and 10 days respectively [19, 20]. Therefore, in addition to these transitions, MDSC and effector cells undergo apoptosis due to half-life.

3. Results

To assess the dynamics of tumor-immune system and predict the behavior of different cells in tumor microenvironment, we have designed a computational model which can model the behavior of system with regarding randomness in interactions. We executed the model for two strategies that are tumor escape from immune response and tumor elimination by the immune system. The dynamic of tumor cells, effector cells and MDSC for these strategies is depicted in Figure 3 and 5. As presented in these figures, SPN models the behavior of tumor cells, effector cells and MDSC in each of the two strategies of tumor escape and tumor elimination correctly. According to Figure 5, in the early time steps, tumor cells recruit MDSCs in order to immune cells response suppression. Consequently, effector cells population decreases with the increment of

the MDSCs population. However, effector cells overcome the MDSCs in the next times and the population of effector cells converge to a nonzero value and eliminate tumor cells.

As mentioned before, SPN applies random effects in interactions. Therefore, multiple executions of SPN can create different dynamics (with similar patterns). Figures 4 and 6 depict the region of uncertainty which is derived by 20 times of implementation of SPN for two strategies of tumor escape and tumor elimination respectively.

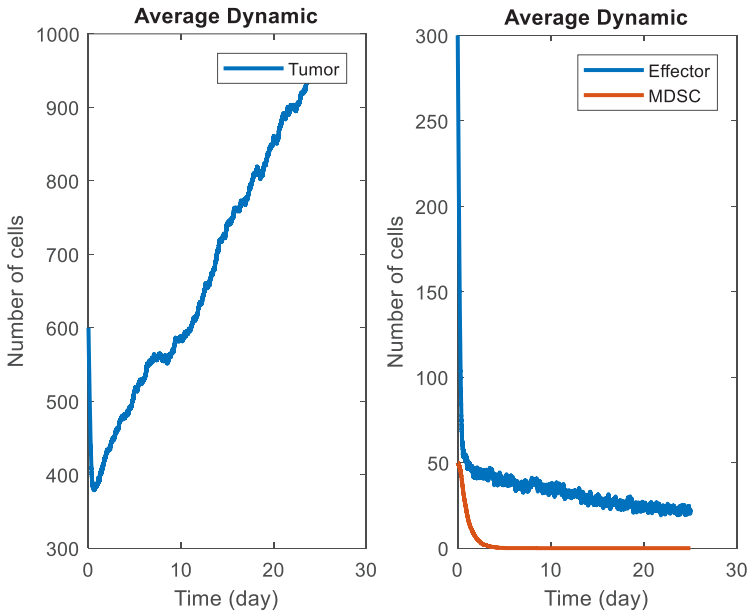

Figure 3. Average dynamic of cells in DNTII. Dynamic of tumor cells, effector cells and MDSC is generated by SPN. SPN is executed for 20 times and the average of dynamics of cells is computed.

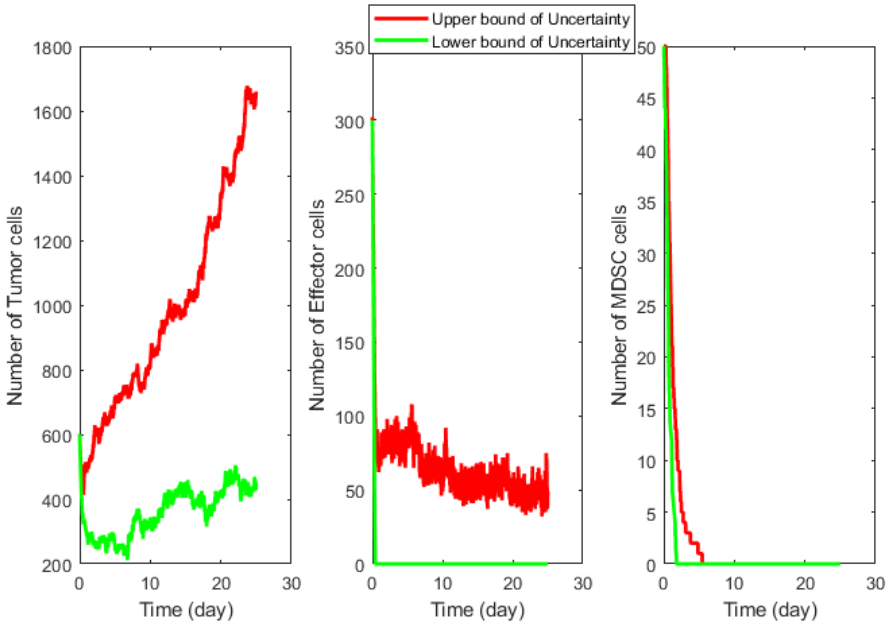

Figure 4. Uncertainty region for tumor escape phase. Region of uncertainty for tumor cells, effector cells and MDSC is generated by 20 execution of SPN.

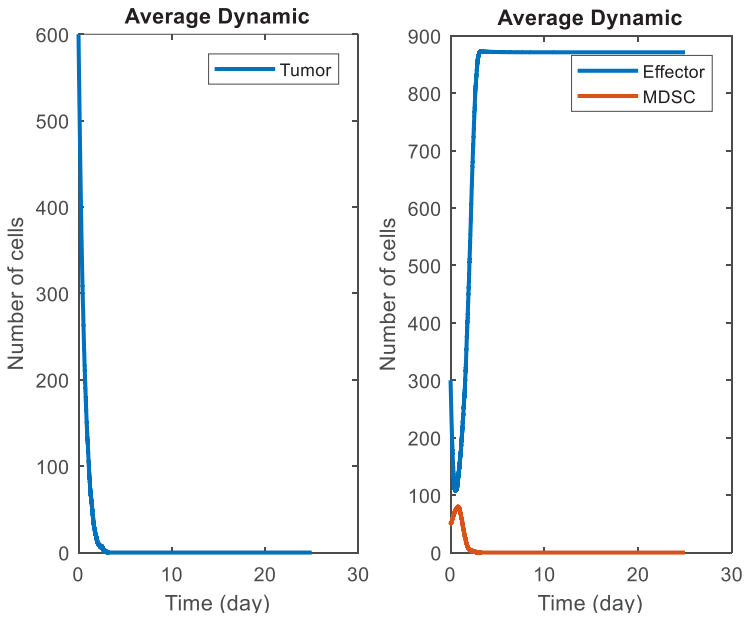

Figure 5. Average dynamic of cells in DNTII. Dynamic of tumor cells, effector cells and MDSC is generated by SPN. SPN is executed for 20 times and the average of dynamics of cells is computed.

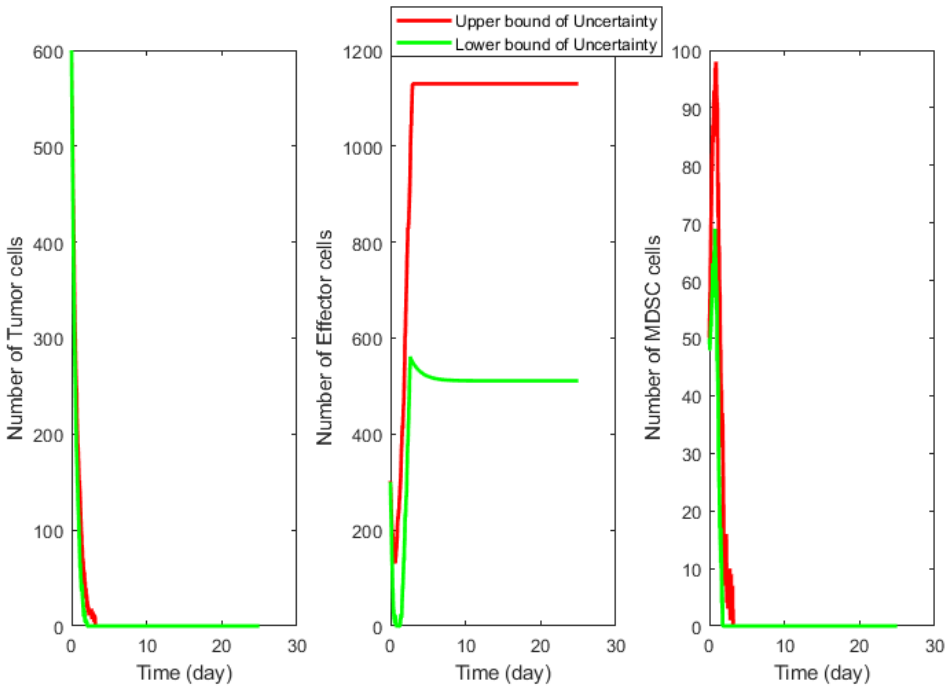

Figure 6. Uncertainty region for tumor elimination phase. Region of uncertainty for tumor cells, effector cells and MDSC is generated by 20 execution of SPN.

4. Discussion

In this study, we constructed a computational model to perform a time course study on the tumor-immune system. The non-linear Dynamical Network of Tumor-Immune Interactions (DNTII) is a very complex network and we have created a computational model to predict its dynamical behaviors. We performed a literature review for key components that are responsive in determining the dynamic of the tumor-immune system. Therefore, MDSC, effector and tumor cells are selected and a model was constructed based on these factors, their behaviors and interactions.

The computational model of this study is stochastic petri net which can exhibit inherent features of tumor-immune system like stochasticity and complexity. This model accurately predicts the dynamical behavior of tumor-immune system in the tumor escape and tumor elimination phases.

We believe that the major contribution of our survey, in addition to the offer of the use of stochastic petri net in cancer modeling, is that a deeper understanding and comprehensive analysis of the biological network can be achieved.

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