Modeling of Peripheral T Cell Differentiation

Natasa Miskov-Zivanov[1], John A. P. Sekar[1], Michael S. Turner[2], Lawrence P. Kane[2], Penelope A. Morel[2] and James R. Faeder[1]

***Short Abstract* — Peripheral naïve T-cells can differentiate into several types of effector cells and the relative numbers produced of each cell type are critical for many immune-related pathologies. To study this system, we have constructed a logical model in which each molecule type is treated as a discrete variable. The model reproduced several important experimental observations and its construction helped to clarify the logical relationships among molecular inputs at several key control points in the process. Model simulations led to the design of new experiments to probe the behavior of key elements such as PTEN and IL-2 under different initial conditions.**

I. Introduction

MECHANISMS involved in DC-mediated expansion of regulatory T (Treg) cells, as opposed to helper T (Th) cells, are still not well understood, although experimental data suggest that there is a T cell-intrinsic mechanism such that low T-cell receptor (TCR) signaling levels favor Treg induction [7]. Recent studies have also emphasized the role of the Akt/mTOR signaling pathway in inhibiting the induction of Treg cells [5]. Therefore, it is essential to understand the pathways leading to DC-mediated differentiation of naïve T-cells. Preventing Treg cell induction at the level of DC-T cell interactions might be one way to eliminate antigen-specific Treg cells and thus decrease or even reverse immune suppression in cancer.

In this work we used a logical approach and, in particular, Boolean networks, to model T cell differentiation. Modeling regulatory networks by means of Boolean networks has been suggested in late sixties [3], but it received more attention in recent years [2][4]. In order to create a Boolean network for studying T–cell differentiation, we have identified a set of elements that are found to have important roles in the process of differentiation [5][7] and determined logical representation of connections between the model elements.

II. Results

The logical model of T cell differentiation was based on an extensive literature survey. Simulations were carried out using BooleanNet tool [1] with random asynchronous updates of model variables. We simulated and analyzed four scenarios, described in more detail below. For each scenario, 300 independent simulations were conducted. Stable expression of Foxp3 was used as a marker for the Treg phenotype.

Experimental results from [7] show that high antigen dose produces mostly Th cells and low antigen dose generates a high fraction of Foxp3-expressing Treg cells. Simulation results show that high antigen dose trajectories all end with Foxp3 expressed, whereas low antigen dose trajectories all end with Foxp3 absent. The high antigen dose simulations also match the experimental observation of transient Foxp3 expression. Simulations agree qualitatively with the experimental finding that a substantial fraction of the resulting cell population expresses Foxp3 when antigen is removed after 18 hours. Furthermore, simulation results also agree with the experimental finding that a substantial Foxp3 positive population is generated as a consequence of blocking Akt or mTORC1 activation with inhibitors that are added 18 hours after strong TCR stimulation. Inhibition is modeled by fixing the value of either Akt or mTORC1 at '0' after four time steps. Finally, both experiments [7] and logical model simulations indicate an inverse correlation between pS6 levels at early time points (18 hours) and Foxp3 expression later on (7 days).

III. Discussion

The logical model that we have constructed for T cell differentiation reproduces many of the important experimental results and has also initiated further experimentation. Analysis of transient Foxp3 expression for the high-antigen dose case indicates that the timing of initial Foxp3 expression relative to mTORC1 activation determines whether transient expression occurs. In our simulations, early STAT5 activation of the Foxp3 promoter occurs in all trajectories with transient Foxp3 expression, which points to the need for further experiments to determine whether interfering with STAT5 activation is sufficient to eliminate transient expression or other factors are involved that should be considered in the model. Simulation results also emphasize the need for better experimental characterization of PTEN, particularly its differential activity in Treg vs. Th. Logical simulation has also suggested the rationale behind oscillations in the Foxp3 and IL-2 values, after logical relationships are changed from 'and' to 'or' or vice versa. These oscillations either point to the representation closest to the realistic behavior, or they indicate the need for representing some element states with more than two levels.

Thus, by using the logical model that we constructed, we have identified the elements that may play a critical role in the system. We are also conducting new experiments to obtain further measurements of these key elements (*e.g.*, PTEN, SMAD3, IL-2), as well as to gain more insight into element relationships (*e.g.*, pS6 kinetics with respect to the presence of IL-2). We anticipate that these measurements will further advance our understanding of the determinants of the peripheral T cell fate, eventually leading to new strategies for reversing immune suppression in cancer in order to increase the effectiveness of vaccination-based therapies.

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[1]Department of Computational and Systems Biology, School of Medicine, University of Pittsburgh, E-mail: {nam66,jas237,faeder}@pitt.edu

[2]Department of Immunology. School of Medicine, University of Pittsburgh. E-mail: {mstst22,morel,lkane}@pitt.edu.

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