Bioinformatics and systems biology for studying the immune system against prostate cancer

Huiming Peng, Guangxu Jin, Tao Cui, King Li, Purnima Dubey, and Xiaobo Zhou

Wake Forest School of Medicine, Medical Center Boulevard, Winston Salem, NC 27157, USA

Background: The prostate cancer (PCa) is the most commonly diagnosed malignancy and the second leading cause of cancer-related deaths in American men. Androgen ablation is a first-line therapy (immune-mediated) for PCa, but a significant number of patients eventually developed castration-resistant PCa, for which there is no cure. Thus, it is urgently needed for us to increase our understanding of the castration-related host immune system which may play critical roles in the development of effective immunotherapy for PCa.

Methods: To cope with the complexity (multi-scale issue) of the immune system we combined the interdisciplinary arts of bioinformatics and systems biology in our study. On the one hand, to initially determine the inhibitory mechanisms responsible for the transient increase in effector function following surgical castration (CX) previously observed in our lab based on prostate-specific Pten-/- mice model, we proposed a systems biology approach based on animal data, which combines coherent experimental protocols with a predictive multi-scale multi-compartment ODEs model of immune system (called M5I system) to dissect the dynamics of the host immune responses to CX in mice model. The proposed M5I system was well established by integrating four types of datasets under three types of experimental conditions and being trained with a modified genetic algorithm (MGA) which takes advantage of the HPC in TACC. Specifically designed model perturbations were then performed to predict the difference of outcomes between different therapeutic protocols. On the other hand, to test whether or not the intercellular immune mechanisms derived from the above animal studies also happen in human cases and to further extend the study on intracellular immune mechanisms, we proposed a novel bioinformatics approach (called Cox-based Immune Network) based on human data MSKCC dataset (PMID: 20579941 and GEO: GSE21032) which includes both gene expression and paired survival profiles for total 140 prostate cancer patients. First, genes' expressions were merged into immunity interactions' expressions or immune signaling interactions with own definition which combines immunity interactions information from two databases InnateDB and Reactome and the statistical p-values of Cox proportional-hazards regression between gene expression vectors and survival times. Then, the immune signaling interactions were clustered into functional networks using dynamic cut tree algorithm with the followed evaluation on grouped branches by their performance in Cox regression model. Finally, the selected functional networks were integrated into a simplified multi-scale structural map using IPA and cell-specific immune database (PMID: 15789058 and GEO: GSE22886).

Results: On the one hand, the proposed M5I system based on animal model resulted in a potential and simplified immune mechanism, which demonstrated that 1) the immune microenvironment after castration involves innate (TANs) and adaptive (CTLs and Tregs) immune cells and two key cytokines (IL2 and CCL20) produced by these cells; 2) Tregs and TANs regulate each other, thereby creating a reciprocal inhibitory loop that adversely affect CTL function. The model perturbations based on M5I system provided us with several effective therapeutic strategies to block the immune inhibitory brake and improve the immune function by depleting Tregs, TANs or CCL20 in certain ways, some of which were validated in experiments, especially worth noting that for CCL20 depletion the treatment effect increases as the treatment is extended which may be associated with the observed decrease of CCL20's peak value. On the other hand, the proposed Cox-based Immune Network based on human data resulted in a simplified multi-scale structural map consisting of intercellular interactions and intracellular

signaling pathways, in which the identified genes have been separated by positive and negative immune functions. The resulted map successfully identified out the major members and their relationships appearing in animal M5I system, which demonstrates to some extent the effectiveness of animal-derived results shifting towards human case. Most importantly, the resulted map demonstrated that CCL20-related inhibitory immune mechanism may closely related with part of the identified negative genes including two TAN-specific genes TLR9 and MYD88 and two Treg-specific genes ELK1 and SOCS1 and one TAN-Treg common gene TRAF6, which may have potential to be immunotherapeutic targets besides CCL20 for PCa treatment.

Conclusion: This study provides a framework of combined bioinformatics and systems biology approaches to studyin