The role of transcription factor Aire in development of autoreactive and regulatory $CD4^+$ T lymphocytes

AIRE transcription factor encodes tissue-specific antigens that are presented to thymocytes during their development. Perturbed expression of AIRE leads to many immunological deficiencies leading to activation of autoreactive T cells, their invasion to many organs, including liver and prostate, that induce inflammation, expression of auto-antibodies and infertility. Numerous mouse models have been developed in order to study APECED and APS1, human syndromes of AIRE deficiency, of which one was used in this dissertation.

The purpose of these studies was to address the effect of AIRE deficiency on the development and function of helper T cells, including $CD4^+Foxp3^+$ regulatory T lineage (Tregs). For this purpose, $Aire^{ko}$ mouse was crossed with TCR^{mini} mouse that expresses a limited TCR diversity, and with reporter animals, $Foxp3^{hCD2}$ (where hCD2 is expressed in the context of Foxp3 expression) and Nur77GFP (where GFP reports T cell activation in response to an antigen in a dose-dependent manner). This new mouse model $Aire^{ko}TCR^{mini}Foxp3^{hCD2}$Nur77GFP was used to study helper T cells in numerous organs, including the thymus, lymph nodes, liver, prostate, and large intestine. These studies utilized many techniques and technologies, including flow cytometry, high throughput sequencing, and the production of hybridomas from $CD4^+$ cells.

The data obtained in this dissertation confirmed a number of published data (i.e., the presence of autoreactive immunoglobulins in the serum of $Aire^{ko}$ mice, and invasion of T cells to several organs (with the liver being the most prominent one)). The studies showed that perturbations in TCR repertoire $CD4^+Foxp3^-$ and $Foxp3^+$ cells in thymus were not directed towards dominant clones. Analysis of TCR sequences form non-lymphoid organs allowed to select limited number of T cell receptors that might be responsible for tissue damage directed towards TSAs. These changes resulted in the elevated affinity of TCRs to auto-antigens, increased presence of T lymphocytes in many organs, and enhanced activation and proliferation of $CD4^+$ T cells. At the same time, reduced presentation of autoantigens to developing Tregs resulted in their decreased suppressive function and control of activated $CD4^+Foxp3^-$ autoreactive T cells, which was measured by their expression of CD73 and PD1. Elevated expression of CD44 and Nur77 and decreased expression of CD62L by effector T cells, as well as the increased frequency of $CD4^+$

hybridomas recognizing tissue-specific auto-antigens, but also presence of autoreactive clones, confirmed these data. Further experiments will be performed using mouse CNS^{mut} mouse model, where the effect of increased number of activated lymphocytes will not be overshadowed by presence of pTregs.

In conclusion, the novel mouse model $Aire^{ko}TCR^{mini}Foxp3^{hCD2}$Nur77GFP allowed deciphering the changes in helper T cells leading to the induction of autoimmune disease in mice with a mutation in AIRE transcription factor, especially manifested by inflammation in prostate and liver.