Abstract

Primary immunodeficiencies (PIDs) are a heterogeneous group of over 480 genetically inherited disorders of the immune system that result from a block in leukocyte development or functional defects in the immune response. PIDs have a wide range of symptoms including increased susceptibility to infections but also autoimmunity or malignancy. Detailed knowledge of PIDs is essential not only in the clinical/diagnostic field, but also in basic research, as they can serve as model systems for the study of the immune system.

The first part of the thesis focuses on the use of the SCID-RTE cytometric test, which we developed within the EuroFlow consortium, as a diagnostic tool for detection of patients suspected of severe PID. Based on the parameters studied (levels of naive $CD4^+$ and $CD8^+$ T cells, recent thymic emigrants and the activation status), we have demonstrated that the SCID-RTE tube is a sensitive test suitable for patients clinically suspected of PID, identified within the broader EuroFlow diagnostic algorithm, or the national newborn screening program. Next, within EuroFlow, we provided a sensitive immunophenotypic evaluation of B cell subset alterations in patients with predominantly antibody deficiencies (PADs), including the expression of distinct immunoglobulin subclasses on memory B cells and plasma cells. Subset dissection identified distinct immune profiles that correlated with diagnostic subtype and clinical presentation. In order to create a tool for detailed understanding to B cell immunodeficiency, we developed a mass cytometry protocol and a novel computational framework capable of detailed interrogation of human B cell developmental pathways, with the potential to reveal alternative pathways possibly present in PIDs, to suggest improved diagnostic tests or therapeutic targets.

In the second part of the thesis we investigated previously unreported role of non-apoptotic Fas signaling and its function in B cell differentiation using autoimmune lymphoproliferative syndrome (ALPS) as a model disease. Specifically, it contributes to the extrafollicular versus germinal center fate decision by modulating the activity of mTOR. Next, we identified and functionally characterized a novel mutation in the *TLR8* gene that caused an autoimmune and autoinflammatory disease in monozygotic twins. The resulting partial loss of TLR8 protein led to dysregulation between the TLR8 and TLR7 responses, based on cross-reactivity of the TLR8 receptor to TLR7 ligands and enhanced signaling of the TLR7 receptor.