# IMMUNOLOGICAL MARKERS OF CELL ACTIVATION IN SARCOIDOSIS

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Sarcoidosis is a systemic disease of unknown cause, which is characterized by the presence of noncaseating granulomas in one or multiple organs. Most clinical manifestations of sarcoidosis are secondary to the direct effect of the accumulation of activated immune cells in involved tissues, notably T cells and macrophages. The purpose of this thesis was to evaluate the usefulness of markers of immune cell activation in pulmonary sarcoidosis. Furthermore, genes encoding proteins related to cell activation were investigated for their influence on disease susceptibility or disease course. Sarcoidosis is likely to be a genetically complex disease that involves a combination of genetic loci conferring disease predisposition or phenotypic variation of disease manifestation.

The results in this thesis describe two novel susceptibility genes for sarcoidosis, the ITGAE gene and the IL7R gene. ITGAE encodes the alpha chain for the integrin alphaEbeta7, a protein that accounts for the interaction between T cells and epithelial cells. A specific genotype of a promoter single nucleotide polymorphism (SNP) showed significant correlation with higher percentages of CD103+CD4+ lymphocytes in bronchoalveolar lavage fluid (BALF) of sarcoidosis patients and in peripheral blood mononuclear cells that were cultured in vitro. Furthermore, this genotype was associated with fibrosis formation on chest x-ray after minimal 4 years follow-up. These chronic activated effector T cells may be important as first line of defense but might also be harmful to an already compromised epithelium.

The other novel and susceptible gene for sarcoidosis described in this thesis is IL7Ralpha. IL7Ralpha is a functional key marker of the early heterogeneity observed in effector T cells. A functional SNP in the IL7R gene was associated with the development of sarcoidosis. Altered IL-7 signaling may contribute to reduced initial immune activation, leading to persistence of the antigen and subsequent development of sarcoid granuloma. The results described in this thesis support the use of cell activation markers as sarcoidosis disease parameters, in addition to existing parameters including disease marker levels in serum, evolution of chest x-ray, and lung function data.

Furthermore, this thesis shows that peripheral blood CD69+VLA-1+ monocytes and CD8+CD28null lymphocytes may also be promising new biomarkers for respectively disease activity and disease severity. In addition, CD103 may play a role. The results of this study showed increased percentages of CD103 positive cells at time of diagnosis in BALF of sarcoidosis patients. These elevated percentages appeared to be of prognostic value for development of parenchymal infiltrates at pulmonary disease outcome independent of chest radiography at presentation.

Finally, the BALF CD103+CD4+/CD4+ ratio, combined with a relative BAL/PB CD4+/CD8+ ratio, discriminated pulmonary sarcoidosis from other interstitial lung diseases. Due to the increased possibilities to characterize the cellular and protein composition of BALF, the normal values of these novel parameters are mostly lacking for healthy controls. Therefore we determined the normal values for cellular subsets and a broad range of protein biomarkers in a group of healthy subjects who voluntarily underwent bronchoalveolar lavage. This enabled us to define unique blood and corresponding lung control values of the experimental parameters described in the different chapters of this thesis.