Rhabdomyosarcoma (RMS) is the most common type of soft tissue sarcoma in children and adolescents. The reasons for the development of RMS are not fully understood, but it seems that its development is linked to biological errors in stem cells. The prognosis for patients with advanced RMS is poor, especially if metastases occur. Patients with Li-Fraumeni syndrome (LFS) are particularly susceptible to RMS due to mutation in p53 gene.

The epithelial-mesenchymal (EMT) transition process can be defined as series of morphogenetic lesions that increase motility, invasiveness and the ability to metastasize. Transcription factors (TFs) playing a key role in EMT are the family of SNAIL proteins and the helix-loop-helix proteins family.

We have shown recently, that TFs associated with EMT are found in RMS cells. In addition, we have shown that the expression of SNAIL1 has a positive effect on the growth of RMS *in vivo*. These data allow us to make an interesting hypothesis that TFs associated with EMT directly or through an interaction with myogenic TFs can play a key role in the development of RMS.

In our project, we will use human pluripotent cell lines from patients with LFS and human pluripotent cell lines without p53 gene expression created by genetic modification. Cells will be differentiated into myogenic and mesenchymal lines to determine, which cell type is responsible for RMS development. In addition, we will develop human pluripotent cell lines with decreased or increased expression of TFs associated with EMT and myogenesis to determine interactions between these transcriptional factors during rhabodomyogenesis and myogenesis, proliferation, migration and angiogenesis. In order to investigate the role of EMT related TFs *in vivo*, human myogenic and mesenchymal stem cells obtained by differentiation of pluripotent cells will be injected subcutaneously and intravenously into immunocompromised mice to investigate their ability to form tumors and metastases.

By defining the role of EMT related and myogenic TFs in rhabdomyogenesis and myogenesis, we have a chance to better understand the complexity of interactions and linkages between TFs related to EMT and myogenesis in the development and progression of RMS. This will help us to learn more about the mechanisms underlying the RMS development. In the future, our research may open up opportunities to develop therapeutic tools that can be used in clinical trials.

Our recently published results confirm that the development of RMS is, at least in part, due to a defect in early stem cell differentiation. We have shown that SNAIL1 and MET receptor, are responsible for keeping RMS cells in an undifferentiated state and that their reduction may force differentiation of RMS cells. In the proposed project, we will study interactions between EMT related TFs and myogenic TFs in relation to rhabdomyogenesis and myogenesis. Such a comprehensive analysis of the interaction between TFs EMT-related and myogenic TFs in RMS development has not been published before. We hope that the results obtained during the project will increase the knowledge about RMS biology and help in developing new, more effective RMS therapies.