6 Summary

Human LRRK1 (leucine-rich repeat kinase 1) is a multi domain protein of over 200 kDa, which contains several essential domains for signal transduction. This thesis describes the protein-protein interaction mediating Ankyrin, leucine-rich and WD-40 repeats of LRRK1 and is focusing on its protein kinase and the GTPase-like domain (Roc domain; Ras of complex proteins). The Roc domain, which is always connected with a 300-400 bp spanning COR domain (C-terminal of Roc), classifies LRRK1 to the group of ROCO proteins, a distinct subfamily of Ras-like GTPases of unknown function. Through characterizing the biochemistry of LRRK1 here, for the first time, the mechanism of regulation of a ROCO protein is described.

In-depth bioinformatic analyses of the architecture of the LRRK1 protein domains led to the discovery and completion of the protein repeats known so far. A complete transcript, fragments and mutated versions of LRRK1 mRNA were generated and, via plasmid transfections, expressed in HeLa and HEK293FT cells. Through analyzing the exclusively cytosolic LRRK1, it could be proven that LRRK1 is an active protein kinase which specifically binds GTP or GDP to its Roc domain. Moreover, the rate of autophosphorylation indicated that LRRK1 kinase activity is selectively stimulated by GTP and not GDP. The analysis of several LRRK1 mutants led to the understanding of the intramolecular mechanism of the autophosphorylation. Based upon the signal transduction of small monomer GTPases a GTP-dependent activation model of LRRK1 was postulated.

LRRK1 represents the first known GTP regulated protein kinase where regulatory and enzymatic functions are joined in a single protein. Furthermore, LRRK1 meets distinct criteria for representing a molecular switch that can be regulated in many ways. Because of the described sequence homology the presented activation model constitutes a future potential in the understanding of the function of other ROCO proteins. Comprehending the mutations of LRRK2, the closest relative of LRRK1, which contribute to the pathology in Parkinson’s disease is of uttermost interest. If LRRK1, whose gene is situated in a gene locus suggested to trigger Alzheimer’s disease, participates in the pathology of neurodegenerative disorders, it will certainly be subject to further studies.