Mutations in mitofusin 2 (MFN2) gene are one of the underlying causes of the most common inherited neuropathy, Charcot-Marie-Tooth (CMT). A novel MFN2 mutation (??539) was detected as the cause for an autosomal recessive inherited fetalonset neuroaxonal dystrophy (FNAD) in a dog model. FNAD pathology manifests in the nervous system, causing secondary effects on muscle functions. MFN2 is a nuclear encoded, mitochondrial outer membrane protein involved in maintaining mitochondrial shape, integrity, and dynamics through protein interaction networks. The aim of this thesis was to investigate the effects of the novel MFN2 mutation at the molecular and cellular levels. Mutant MFN2 mRNA transcript analysis revealed active transcription of the mutant MFN2 allele in tissues derived from both homozygous and heterozygous dogs. Immunoblotting, and immunocytochemistry revealed a decreased amount of mutant MFN2 protein in tissues of diseased dogs. Polarography performed on mitochondrial extracts derived from affected pups did not detect any mitochondrial respiratory dysfunctions. Mitochondrial structure and distribution assayed through live-cell imaging of in-vitro cultured primary canine fibroblasts in combination with mitochondria specific dyes showed maintained mitochondrial morphology and networks in cells derived from affected pups. Analysis of mitochondrial membrane potential, an indicator of
mitochondrial integrity and health, suggested a reduced membrane potential in affected pup mitochondria. Mitochondrial autophagy and recycling stress was not detected in vitro under normal culture conditions. The mitochondrial fusion function of mutant MFN2 is lost as revealed by specific knock down of its fusion complementing cellular paralogue MFN1 in affected pup primary fibroblast cells. In conclusion, the ??539 mutation in MFN2 results in loss of protein expression which is proposed to affect an MFN2 function relevant to nervous system derived tissues. Mitochondrial structure and assayed functions are maintained in affected dog cultured primary fibroblast cells in spite of possible membrane potential declines.