SUMMARY: **"*Caenorhabditis elegans* as a model organism to study mitochondrial diseases associated with defects in tRNA modification"**

Post-transcriptional modification of the wobble uridine (U_{34}) of a tRNA set is an evolutionary conserved process, produced by homologous proteins from the MnmA/MTU1, MnmE/GTPBP3 and MnmG/MTO1 families. Their universal character suggests that these modifications play a crucial role in the biology of cells and organisms. In fact, mutations in the human genes *MTU1* and *GTPBP3* or *MTO1* produce acute infantile liver failure, and hypertrophic cardiomyopathy and lactic acidosis, respectively, which usually cause lethality in the first months of life. It is assumed that the primary cause of these diseases is the lack of the modifications introduced by the MTU1 protein in position 2 (a tiol group) and GTPBP3 and MTO1 proteins (a taurinomethylation group) in position 5 at U_{34} in a subgroup of mitochondrial tRNAs (mt-tRNAs). Nevertheless, the molecular mechanisms underlying these diseases (and other diseases associated with such modifications) are not clear. The reason why the typical defects of oxidative phosphorylation (due to impaired mitochondrial translation) produce such wide range of phenotypes is still unknown. Our hypothesis sustains that the mitochondria-nucleous retrograde signaling pathways triggered by the hypomodification at position 2 and 5 of U_{34} are different, and that each nuclear response is modulated by the genetic and epigenetic programs of cells and organisms.

In this work, we have used the nematode *Caenorhabditis elegans* as a model organism to study the effects of inactivating the homologue proteins to MTU1, GTPBP3 and MTO1, which we have named as MTTU-1, MTCU-1 and MTCU-2, respectively. We have proved that these nuclear encoded proteins are located in mitochondria and are involved in U_{34} modification of mt-tRNAs. The *mtcu-1* and *mtcu-2* single mutants show a reduction in fertility, while the *mttu-1* single mutant shows a reduction in fertility and a lengthening of the reproductive cycle (both phenotypes are thermosensitive). The phenotypes exhibited by the *mttu-1*, *mtcu-1* and *mtcu-2* single mutants support our hypothesis, in which the *mttu-1* single mutation, on the one hand, and the *mtcu-1* and *mtcu-2* single mutations, on the other hand, trigger different retrograde signaling pathways which produce specific nucear expression. Thus, a nuclear dependent phenotypic trait (as transcription or mt-tRNAs stability) and the expression of nuclear genes as *ucp-4*, *hsp-6*, *hsp-60* and other genes involved in mitochondrial metabolism show a differential pattern in both grup of mutants. *hsp-6* and *hsp-60* genes, which are used as mitochondrial stress response markers (UPR^{mt} or

"mitochondrial unfolded protein response") are downregulated in *mttu-1* single mutant, which could be related to fertility and reproductive cycle thermosensitivity. The three single mutants exhibit reduced expression of glycolysis and ß-oxidation genes (usually more drastic in the *mttu-1* mutant), an induction of a glutaminolysis marker, and an induction of the *ucp-4* gene, which encodes a transporter of the succinate (an intermediate of the tricarboxylic acid cycle, TCA) to the mitochondria. Due to all three single mutants display a mild OXPHOS dysfunction, we propose that the observed changes in the expression of genes involved in the mitochondrial metabolism reveal a TCA cycle reprogramming aimed to compensate the reduction of acetil-CoA (coming from glycolysis and fatty acid oxidation) though the activation of anaplerotic pathways characterized by the succinate import to mitochondria by UCP-4 and the incorporation of 2-oxoglurate from glutaminolysis. This reprogramming could be associated with the input of reduced equivalents (as $FADH_2$) to the OXPHOS system through complex II, which would compensate the putative dysfunction of complex I. In this Thesis, we also analyze the effects of the simultanous suppression of modifications at positions 2 and 5 of U_{34} in *C. elegans*. The double mutant *mtcu-2*;*mttu-1* displayed a severe OXPHOS dysfunction and a 5-fold higher AMP/ATP ratio, which was associated with embryonic lethality, developmental arrest in primary larval stages, penetrant sterility in adults and extended lifespan. This lifespan extension is modulated by signaling pathways which depend on AMPK (specifically on AAK-1 catalitic subunit) and steroid hormones, through DAF-9 and DAF-12 proteins.

In brief, this work shows for the first time in an animal model the important gene reprogramming related to mitochondrial metabolism in response to U_{34} hypomodification of mt-tRNAs, and shows new connexions between signaling patways that extend lifespan.