Al collegio docenti del Dottorato in Medicina Molecolare

Relazione dell'attività scientifica svolta nel 1° anno di Dottorato, Anno Accademico 2018/2019, Ciclo XXXIV, nel laboratorio di Medicina Molecolare presso l'IRCSS Fondazione Stella Maris di Calambrone, Pisa

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"Study of the molecular characteristic of spastic paraplegia type 11, its impact on oxidative, autophagic and gangliosides metabolism"

BACKGROUND

Spastic paraplegia type 11 (SPG11) is one of the most prevalent autosomal recessive complicated neurodegenerative hereditary spastic paraplegia (HSP) subtypes and accounts for about 10% of all patients with HSP. AR-SPG11 is associated with mutations in the *SPG11* gene encoding spatacsin whose function is largely unclear but it seems to be associated with autophagosome recycling, impaired ganglioside metabolism, neurite plasticity.

AIM

The aim of this project is to better characterize the role of spatacsin in HSP and to perform attempts to treatments able to recover the expression level of the protein and to reduce the accumulation of gangliosides in the lysosomes. We will characterize cultured skin fibroblasts in SPG11 patients in terms of oxidative stress, autophagy, spatacsin expression and its interacting partners (SPG15, SPG48) and cellular amount of gangliosides. We will also treat cells with two FDA and EMA approved drugs, namely Ataluren and Miglustat, potentially able to rescue spatacsin readthrough transcription and function. Finally, we will validate data in *in vivo* models of SPG11.

METHODS

We will characterize in SPG11 primary cells the proteins spatacsin, spastizin, SPG48 through western blot. Oxidative stress will be evaluated through western blot of NRF2 protein, Seahorse XF Analyzers and MitoSox assay to assess free radical production. Disrupted autophagy will be studied growing cells in basal and starved conditions and measuring lysosome antibody markers such as LAMP2, lysotracker, p62 and LC3-I/LC3-II protein levels; while gangliosides amount will be analyzed thanks to a lipidomic approach.

RESULTS and EXPECTED RESULTS

In SPG11' skin fibroblasts spatacsin is almost absent in all cases, whereas SPG15, SPG48 and NRF2 protein levels are variable. We could test in parallel protein levels in blood cells. In these cell lines, we will measure oxidative metabolism, deregulated autophagy pathway and an accumulation of gangliosides. Having received the pure compound from PTC pharmaceutics, we will set up soon treatments with Ataluren to recover in part the expression of SPG11. We will also test if Miglustat will be able to lessen the production of redox species and gangliosides accumulation.

ABSTRACTS AND CONGRESSES

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EDUCATIONAL ACTIVITIES

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