DOTTORATO DI RICERCA IN BIOLOGIA CELLULARE E DELLO SVILUPPO

Proposta di progetto per Dottorato

Titolo della ricerca Spastin elevating approaches: new therapeutic perspectives in Hereditary Spastic Paraplegia Type 4 (SPG4-HSP)

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Struttura(e) ospitante(i): IBPM-CNR c/o Sapienza- dept BBCD

Summary

SPG4-HSP is the most common motorneuron hereditary disease, in which low extremity weakness and spasticity are the predominant symptoms (Burger et al., 2000). Current therapies are symptomatic, and usually provide inadequate relief of symptoms. Dominant mutations of SPG4, encoding the microtubule (MT) severing enzyme, spastin, lead to pathogenesis mainly by haplo-insufficiency (Solowska et al., 2010). A gene-dosage rescue of neurite defects in SPG4-HSP patients' neurons showed that restoring physiological spastin levels halt pathological phenotypes (Havlicek et al., 2014). The future for innovative therapy design is encouraging, since the knowledge about the underlying molecular mechanisms of this disease is progressively growing. However, SPG4-HSP therapeutics urgently require a systematic analysis of quantifiable clinical, molecular and digital parameters to develop rigorous biomarkers and clinical outcome assessment measures.

Recently, we found that inhibition of spastin degradation leads to an increase of spastin protein levels and rescues neurite defects in spastin-deficient models, providing the proof of principle that manipulating spastin degradation pathway is a strategy to develop spastin-elevating therapies (Sardina et al., 2020). In particular, we identified a promising drug that we are now validating in opportune animal models. During these studies, we also observed peculiar MT network defects in SPG4-HSP patient-derived lymphoblastoid cells (LCLs) that are rescued by pharmacologically elevating spastin.

Thus, we plan i) to dissect the spastin degradation pathway by identifying the molecular players and exploring the contribution of neddylation ii) to characterize MT network architecture in LCLs from SPG4-HSP patient families and iii) to evaluate their response to spastin elevating approaches.

Achieving the goals of this project will improve the knowledge on a novel spastin druggable pathway and will reveal prognostic and predictive biomarkers for SPG4-HSP patients.

Pertinent Publications of the proponent (last 5 years)

1) Sardina F, Pisciottani A, Ferrara M, Valente D, Casella M, Crescenzi M, Peschiaroli A, Casali C, Soddu S, Grierson AJ, **Rinaldo C** Spastin recovery in hereditary spastic paraplegia by preventing neddylation-dependent degradation. Life Sci Alliance. 2020 Oct 26;3(12):e202000799. doi: 10.26508/lsa.202000799.
2) Sardina F, Monteonofrio L, Ferrara M, Magi F, Soddu S, **Rinaldo C.**HIPK2 Is Required for Midbody Remnant Removal Through Autophagy-Mediated Degradation. Front Cell Dev Biol. 2020 Sep 15;8:572094.
3) Gatti V, Ferrara M, Virdia I, Matteoni S, Monteonofrio L, di Martino S, Diodoro MG, Di Rocco G, **Rinaldo C***, Soddu S. An Alternative Splice Variant of HIPK2 with Intron Retention Contributes to Cytokinesis. Cells. 2020 Feb 20;9(2):484. *co- last author.
4) Monteonofrio L, Valente D, **Rinaldo** C, Soddu S. Extrachromosomal Histone H2B Contributes to the Formation of the Abscission Site for Cell Division. Cells. 2019 Nov 5;8(11). pii: E1391.

5) Contadini C, Monteonofrio L, Virdia I, Prodosmo A, Valente D, Chessa L, Musio A, Fava LL, **Rinaldo C**, Di Rocco G, Soddu S. p53 mitotic centrosome localization preserves centrosome integrity and works as sensor for the mitotic surveillance pathway. Cell Death Dis. 2019 Nov 7;10(11):850.
6) Willan J, Cleasby AJ, Flores-Rodriguez N, Stefani F, **Rinaldo C**, Pisciottani A, Grant E, Woodman P, Bryant HE, Ciani B ESCRT-III is necessary for the integrity of the nuclear envelope in micronuclei but is aberrant at ruptured micronuclear envelopes generating damage. Oncogenesis. 2019 Apr 15;8(5):29.
7) A Pisciottani, L Biancolillo, M Ferrara, D Valente, F Sardina, L Monteonofrio, S Camerini, M Crescenzi, S Soddu, **C Rinaldo** HIPK2 phosphorylates the microtubule-severing enzyme spastin at S268 for abscission. Cells, 2019 8(7), 684.
8) Scaglione A, Monteonofrio L, Parisi G, Cecchetti C, Siepi F, **Rinaldo C**, Giorgi A, Verzili D, Zamparelli C, Savino C, Soddu S, Vallone B, Montemiglio LC. Effects of Y361-auto-phosphorylation on structural plasticity of the HIPK2 kinase domain. Protein Sci. 2018 Mar;27(3):725-737
9) Monteonofrio L, Valente D, Ferrara M, Camerini S, Miscione R, Crescenzi M, **Rinaldo C***, Soddu S. HIPK2 and extrachromosomal histone H2B are separately recruited by Aurora-B for cytokinesis. Oncogene. 2018 Mar22 *co- last author.
10) Barbiero I, Valente D, Chandola C, Magi F, Bergo A, Monteonofrio L, Tramarin M, Fazzari M, Soddu S, Landsberger N, **Rinaldo C***, Kilstrup-Nielsen C. CDKL5 localizes at the centrosome and midbody and is required for faithful cell division. Sci Rep. 2017 Jul 24;7(1):6228 *co- last author
11) Pierantoni GM, Conte A, **Rinaldo C**, Tornincasa M, Gerlini R, Valente D, Izzo A, Fusco A. Hmga1null mouse embryonic fibroblasts display downregulation of spindle assembly checkpoint gene expression associated to nuclear and karyotypic abnormalities. Cell Cycle. 2016 Feb 18:0.

References (other citations, if appropriate)

- Bürger J, Fonknechten N, Hoeltzenbein M, Neumann L, Bratanoff E, Hazan J, Reis A. (2000) Hereditary spastic paraplegia caused by mutations in the SPG4 gene. Eur J Hum Genet 10:771-6. doi:10.1038/sj.ejhg.5200528

- Solowska JM, Garbern JY, Baas PW. (2010) Evaluation of loss of function as an explanation for SPG4-based hereditary spastic paraplegia. Hum Mol Genet.14:2767-79. doi: 10.1093/hmg/ddq177

- Havlicek S, Kohl Z, Mishra HK, Prots I, Eberhardt E, Denguir N, Wend H, Plötz S, Boyer L, Marchetto MC, Aigner S, Sticht H, Groemer TW, Hehr U, Lampert A, Schlötzer-Schrehardt U, Winkler J, Gage FH, Winner B. Gene dosage-dependent rescue of HSP neurite defects in SPG4 patients' neurons Hum Mol Genet. 2014 May 15;23(10):2527-41. doi: 10.1093/hmg/ddt644

- Sardina F, Pisciottani A, Ferrara M, Valente D, Casella M, Crescenzi M, Peschiaroli A, Casali C, Soddu S, Grierson AJ, Rinaldo C Spastin recovery in hereditary spastic paraplegia by preventing neddylation-dependent degradation. Life Sci Alliance. 2020 Oct 26;3(12):e202000799. doi: 10.26508/lsa.202000799.