UNIVERSITÀ DEGLI STUDI DI MILANO

DiSFeB for Excellence 2018-2022

The Department of Pharmacological and Biomolecular Sciences (DiSFeB)

presents

Wednesday, September 18th 2019

Via Balzaretti 9, Milan

Deepening the characterization and behaviour of mutants of the BCL-2 Associated Athanogene 3 (BAG3)

Tedesco B.*[1], Adriaenssens E.*[3], Mediani L.*[1], Crippa V.[1], Carra S.[3], Timmerman V.[2], Poletti A.[1]

[1]Dipartimento di Scienze Farmacologiche e Biomolecolari, Centro di Eccellenza sulle Malattie Neurodegenerative, Università degli Studi di Milano, Milano, Italy; [2]Peripheral Neuropathy Research Group, Department of Biomedical Sciences, Institute Born Bunge, University of Antwerp, Antwerpen, Belgium; [3]Department of Biomedical, Metabolic and Neural Sciences, University of Modena and Reggio Emilia, and Center for Neuroscience and Neurotechnology, Modena, Italy; *Co-Authors

barbara.tedesco@unimi.it

Keywords: BAG3, Chaperone-assisted selective autophagy, neuropathy, myopathy

ABSTRACT

The chaperone-assisted selective autophagy (CASA) is a pathway of the protein quality control (PQC) system, that plays a fundamental role in the maintenance of the quality of the proteome in neurons, particularly motoneurons, and muscle cells. CASA has been firstly described in muscle cells, where it assures the disposal of damaged structural proteins. In neurons, it has been demonstrated that CASA acts in the removal of misfolded and aggregation prone proteins. CASA relies on the formation of a multimeric complex, namely CASA complex. The Bcl-2-Associated Athanogene 3 (BAG3) is the scaffold protein for CASA complex assembly: it interacts with the Heat Shock Proteins HSPB8 and HSC70/HSP70 and the dynein-machinery. While the chaperones recognize misfolded substrates, dynein routes substrates to the microtubule-organizing center (MTOC), where aggresomes form. Mutations in CASA complex members are associated to diseases that affect neurons and muscle cells. Regarding BAG3, three mutations at position P209 in one of the two IPV domains, which enable the interaction with HSPB8, cause myopathies (P209Q/L) or neuropathy (P209S). Instead, a mutation in the BAG domain (E455K) is associated to cardiomyopathy. I already showed that BAG3 P209 mutants form insoluble species and aggregates, while the E455K mutant is characterized by a higher solubility in respect to BAG3 wild type (WT). Moreover, these P209 mutants are characterized by a detrimental function in the removal of a well-known substrate of the CASA complex (the Amyotrophic Lateral Sclerosis-related SOD1-G93A). Here, I show my new findings on BAG3 mutants behaviour. I confirm the aggregation prone behaviour of BAG3 P209 mutants by flow cytometry. I show that these P209 mutants aggregates sticks to the perinuclear region, in correspondence of the aggresome. I will then show that inhibition of the dynein-mediated retrograde transport is not sufficient to favour the clearance or solubilization of preformed P209 aggregates, but that genetic silencing of a dynein subunit interferes with the formation of P209 aberrant aggresomes. Finally, I will show that trehalose, a natural compound that enhances autophagy and favours the clearance of misfolded and aggregate prone proteins, has the same beneficial activity on the removal of P209 BAG3 aggregates, an effect that was already observed in models of motoneuron diseases and other neurodegenerative conditions. In summary, these results expand the knowledge on the role of the interactions that orchestrate CASA pathway and open a window on a possible therapeutic approach.