BIOGRAPHICAL SKETCH

NAME: Alessandro Fraldi

POSITION TITLE: Associate Professor of Histology, University of Naples "Federico II'
Principal Investigator, CEINGE-*Advanced Biotechnology*

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Faculty of Science, University of Naples "Federico II", Italy	BSc DEGREE (cum laude)	10/2000	MOLECULAR BIOLOGY
Department of Genetics, Faculty of Science, University of Naples "Federico II", Italy	PhD	03/2004	HUMAN GENETICS
Telethon Institute of Genetics and Medicine (TIGEM), Naples, Italy		2004-2011	HUMAN GENETICS/ MOLECULAR THERAPY
Lysosomal Disease Research Unit" Woman's and Children's Hospital, Adelaide, Australia		2005-2006	HUMAN GENETICS

A. PERSONAL STATEMENT

My laboratory mainly focuses on (I) the study of lysosomal dysfunction and protein aggregation in neurodegenerative lysosomal diseases and (II) the development of innovative therapeutic strategies for the treatment of brain pathology in mucopolysaccharidoses (MPS), a group of severe diseases belonging to the larger family of lysosomal storage diseases (LSDs).

Over the years I have accumulated experience in the study of LSDs. I have worked on both characterization of MPS phenotype, particularly the neurological aspects, and testing new therapeutic approaches for MPS treatment using mice and pigs as model systems. My lab developed novel brain directed AAV-mediated gene therapy approaches for Sanfilippo syndrome, one of the most common and severe form of MPS. One of these studies constituted the basis for designing the first gene therapy clinical trial for Sanfilippo A patients that is now under the phase II/III. I have also built a very solid expertise in cell biology and biochemistry, which I applied to study the molecular mechanisms underlying MPS neuropathogenesis using both cellular and mouse models of MPSs with the final aim to identify new druggable therapeutic targets. Ongoing projects in my lab are focused on developing therapies for the treatment of the CNS in Sanfilippo syndrome and other neuronopathic MPSs based (i) on gene/ RNA delivery and (ii) on the use of anti-amyloids drugs to counteract lysosomal-autophagy pathway dysfunction that we identified as a major factor driving neurodegeneration in these severe neuropathologies;

1. Giaccio M, Monaco A, Galiano L, Parente A, Borzacchiello L, Rubino R, Klärner F-G, Killa D, Perna C, Piccolo P, Marotta M. Pan X, Khijniak M, Siddique I, Schrader T, Pshezhetsky AV, Sorrentino NC, Bitan G and **Fraldi A***. *Anti-amyloid treatment is broadly effective in neuronopathic mucopolysaccharidoses and synergizes with gene therapy in MPS-IIIA.* **Molecular Therapy 2024**. Nov 6;32(11):4108-4121. ***corresponding author** PubMed PMID: 39342429
2. Monaco A, Maffia V, Sorrentino NC, Sambri I, Ezhova Y, Giuliano T, Cacace V, Nusco E, De Risi M, De Leonibus E, Schrader, T, Klärner FG, Bitan G, **Fraldi A*.** *The amyloid inhibitor CLR01 relieves autophagy and ameliorates neuropathology in a severe lysosomal storage disease.* **Molecular Therapy 2020** Apr 8;28(4):1167-1176 ***corresponding author** PubMed PMID: 32087148
3. Sambri I, D'Alessio R, Ezhova Y, Giuliano T, Sorrentino NC, Cacace V, De Risi M, Cataldi M, Annunziato L, De Leonibus E and **Fraldi, A***. *Lysosomal dysfunction disrupts presynaptic maintenance and restoration of presynaptic function prevents neurodegeneration in lysosomal storage diseases.* **EMBO Molecular Medicine. 2017** 9, 112-132 ***corresponding author -cover highlight-** PubMed PMID: 27881461.

4. Sorrentino NC, Cacace V, De Risi M, Maffia V, Strollo S, Tedesco N, Nusco E, Romagnoli N, Ventrella D, Huang Y, Liu N, Kalled SL, Choi VW, De Leonibus E, **Fraldi A*.** *Enhancing the therapeutic potential of sulfamidase for the treatment of Mucopolysaccharidosis IIIA* **Molecular Therapy- *Methods & Clinical Development,* 2019** Oct 28;15:333-342 ***corresponding author** PubMed PMID: 31788497.
5. Sorrentino NC, D'Orsi L, Monaco C, Nusco E, Sambri I, Spampanato C, Polishhuck EV, Saccone P, De Leonibus E, Ballabio A, **Fraldi A***. *A highly secreted sulfamidase engineered to cross the blood-brain barrier corrects the CNS pathology of mice with mucopolysaccharidoses type IIIA.* **EMBO Mol Med. 2013** May; 5(5):675-90 ***corresponding author** PubMed PMID: 23568409.

B. POSITIONS AND HONORS

Positions and Employment

2023-present	Associate Professor of Histology, Dept of Translational Medicine University of Naples "Federico II'
2022-2023	Associate Professor of Medical Genetics, Dept of Translational Medicine University of Naples "Federico II'
2021-present	Faculty and Principal Investigator, CEINGE-*Advanced Biotechnology,* Naples, Italy
2017-2022	Assistant Professor, Dept of Translational Medicine University of Naples "Federico II'
2013-present	Faculty Member of the European School of Molecular Medicine (SEMM)
2011-2020	Faculty and Investigator, Molecular Therapy Program, Telethon Institute of Genetics and Medicine (TIGEM), Naples, Italy

Other Experience and Professional Activities

-Journal Reviewer: "Ad hoc" reviewer for Science Translational Medicine, EMBO Mol. Med., Human Molecular Genetics, Autophagy, Human Gene Therapy, PLOS One, Neuroscience and Scientific Reports

-Reviewer panel member: European Research Council (ERC), Medical Research Council (MRC), Sanfilippo Children's Foundation, Agency for Health Quality and Assessment of Catalonia.

2024-present Member, Italian Histology Association
2020-present President of Scientific Advisory Board, Sanfilippo Fighters Foundation
2017-present Member, Italian Medical Genetics School
2005-present Member, American Society of Gene & Cell Therapy

2023-	Meeting Organization: Co-organizer and Scientific committee member of Mucopolysaccharidosis III Workshop, (Naples, Italy, 2017)
2017-	Meeting Organization: Co-organizer and Scientific committee member of XVII National Congress of Italian Neuroscience Society (SINS), (Ischia, Italy, 2017)

Invited lectures to internationally established conferences/advances schools

2009-	FEBS advanced course, Ortona, 2009
2011-	Sanfilippo research and treatment workshop Northampton, 2011
2011-	Brains for Brain conference Frankfort, 2011
2013-	XIX Annual Meeting German Society for Gene Therapy (DG-GT), 2013
2013-	Brains for Brain conference Frankfort, 2013
2015-	John van Geest Centre for Brain Repair, University of Cambridge, 2015
2017-	Telethon Convention, Riva del Garda, 2017
2017-	II evento nazionale sulle mucopolisaccaridosi, Milan, 2017
2017-	International Workshop on "Inhibition of Protein-Protein Interactions in Alzheimer's Disease and Related Proteinopathies" Lille University, 2017
2019-	International Workshop on "Strategies and tools for modulating pathologic protein self-assembly, Porto (Portugal) March 21-22, 2019
2019-	ESGCT Spring School 2019, Tigem, Pozzuoli, April 3-5, 2019
2019-	23rd ESN biennial meeting on molecular mechanisms of regulation of the nervous system, Milan, September 1-4, 2019
2019-	EGSLD course on Lysosomes and Lysosomal Diseases, Catalonia (Spain),10-13th October 2019
2021-	Recordati Neurometabolic course: "its all in the brain" May 26-27 2021- Virtual event
2022-	Solving Sanfilippo Symposium, Adelaide (Australia), April 1, 2022- Mixed-mode event
2022-	ADVANCE 2022 | Sanfilippo Syndrome Community Conference. Virtual Event: 7-8 July 2022
2023-	Telethon Convention, Riva del Garda, March 13-15, 2023

2023- Mucopolysaccharidosis III Workshop, Naples (Italy), May 26-27 2023- Mixed-mode event
2024- ADVANCE 2024 | Sanfilippo Syndrome Community Conference. Virtual Event: 29-30 Oct 2024

Honors

2000- B.Sc. Cum Laude, University of Naples "Federico II"
2002- International Mobility Program Award, University of Naples "Federico II"
2003- EMBO fellowship, program 2003
2004- Ph.D. Summa cum Laude, University of Naples "Federico II"
2005- EMBO fellowship, program 2005

Academic Qualifications

National Academic Qualification
Full professor: 05/E1 General Biochemistry; 05/E2 Molecular Biology, 06/A1 Medical Genetics
Associate professor: 05/H2 Histology; 06/A1 Medical Genetics

C. CONTRIBUTIONS TO SCIENCE

Selected peer-reviewed publications

1. As a postdoctoral fellow I studied the mechanisms of action of SUMF1, a master regulator of the activities of sulfatases, a group of lysosomal enzymes whose deficiency causes different forms of LSDs. Moreover, during my postdoc at the Lysosomal Disease Unit in Adelaide (Australia) I contributed to characterize the first mouse model of Sanfilippo A (MPS-IIIA) and provided the first proof-of-principle demonstrating the therapeutic efficacy of AAV-mediated gene therapy approach in Sanfilippo A using the characterized mouse model;

a. **Fraldi A**, Biffi A, Lombardi A, Visigalli I, Pepe S, Settembre C, Nusco E, Auricchio A, Naldini L, Ballabio A, Cosma MP. *SUMF1 enhances sulfatase activities in vivo in five sulfatase deficiencies*. **Biochem J 2007**;403:305-12. PubMed PMID: 17206939; PubMed Central PMCID: PMC1874239.
b. **Fraldi A***, Hemsley K, Crawley A, Lombardi A, Lau A, Sutherland L, Auricchio A, Ballabio A, Hopwood J. *Functional correction of CNS lesions in an MPS-IIIA mouse model by intracerebral AAV-mediated delivery of sulfamidase and SUMF1 genes*. **Hum Mol Genet 2007**;16:2693-702. ***corresponding author.** PubMed PMID: 17725987
c. **Fraldi A**, Zito E, Annunziata F, Lombardi A, Cozzolino M, Monti M, Spampanato C, Ballabio A, Pucci P, Sitia R, Cosma MP. *Multistep, sequential control of the trafficking and function of the multiple sulfatase deficiency gene product, SUMF1 by PDI, ERGIC-53 and ERp44*. **Hum Mol Genet 2008**;17:2610-21. PubMed PMID: 18508857
d. Tardieu M., Zerah M., Husson B., de Bournonville S., Deiva K., Adamsbaum C., Vincent F., Hocquemiller M., Broissand C., Furlan V., Ballabio A., **Fraldi A**., Crystal R., Baugnon T., Roujeau T., Heard, J.M. and Danos O. *Intracerebral administration of AAV rh.10 carrying human SGSH and SUMF1 cDNAs in children with MPSIIIA disease: results of a phase I/II trial*. **Human Gene Therapy. 2014**; June 25(6):506-16. PubMed PMID: 24524415.

2. I contributed to the elucidation of mechanisms underlying lysosomal dysfunction in MPSs. We have provided the first demonstration of a block of autophagy in severe forms of MPSs. Our HMG 2007 paper describing these results is considered a landmark in the field (cited over 400 times). Many subsequent papers, inspired by our work showed similar results in several types of MPSs and other LSDs;

a. Settembre C, **Fraldi A***, Jahreiss L, Spampanato C, Venturi C, Medina D, de Pablo R, Tacchetti C, Rubinsztein DC, Ballabio A. *A block of autophagy in lysosomal storage disorders*. **Hum Mol Genet 2008**;17:119-29. ***co-first author.** PubMed PMID: 17913701
b. **Fraldi A**, Annunziata F, Lombardi A, Kaiser HJ, Medina DL, Spampanato C, Fedele AO, Polishchuk R, Sorrentino NC, Simons K, Ballabio A. *Lysosomal fusion and SNARE function are impaired by cholesterol accumulation in lysosomal storage disorders*. **EMBO J 2010**; Nov 29:3607-20. PubMed PMID: 20871593; PubMed Central PMCID: PMC2982760.
c. Medina DL, **Fraldi A***, Bouchè V, Annunziata F, Mansueto G, Spampanato C, Puri C, Pignata A, Martina JA, Sardiello M, Polischuk R, Puertollano R and Ballabio A. *Transcriptional activation of lysosomal exocytosis promotes cellular clearance.* **Dev. Cell. 2011;** 21(3):421-430. ***co-first author.** PubMed PMID: 21889421; PubMed Central PMCID: PMC3173716
d. **Fraldi A*,** Klein AD, Medina DL, Settembre C. *Brain Disorders Due to Lysosomal Dysfunction*. **Annu Rev Neurosci. 2016** Jul 8;39:277-95. ***corresponding author** PubMed PMID: 27090953

3. A research line in my lab is focused on developing new gene therapy approaches to treat the CNS in MPSs (particularly in the Sanfilippo syndrome) based on the use of AAV as viral vectors to transfer the therapeutic gene to the CNS. We have pioneered the use of engineered variants of therapeutic genes to improve the therapeutic potential of these approaches;

a. Sorrentino NC, D'Orsi L, Monaco C, Nusco E, Sambri I, Spampanato C, Polishhuck EV, Saccone P, De Leonibus E, Ballabio A, **Fraldi A***. *A highly secreted sulfamidase engineered to cross the blood-brain barrier corrects the CNS pathology of mice with mucopolysaccharidoses type IIIA.* **EMBO Mol Med. 2013** May; 5(5):675-90 ***corresponding author** PubMed PMID: 23568409; PubMed Central PMCID: PMC3662312
b. Sorrentino NC, Maffia V, Strollo S, Cacace V, Romagnoli N, Manfredi A, Ventrella D, Dondi F, Barone F, Giunti M, Graham AR, Huang Y, Kalled S, Auricchio A, Bacci ML, Surace EM and **Fraldi A*.** *A comprehensive map of CNS transduction by eight recombinant adeno-associated virus serotypes upon cerebrospinal fluid administration in pigs* **Molecular Therapy. 2015** Feb;24(2):276-86 ***corresponding author** PubMed PMID: 26639405; PubMed Central PMCID: PMC4817820
c. Sorrentino NC, Cacace V, De Risi M, Maffia V, Strollo S, Tedesco N, Nusco E, Romagnoli N, Ventrella D, Huang Y, Liu N, Kalled SL, Choi VW, De Leonibus E, **Fraldi A*.** *Enhancing the therapeutic potential of sulfamidase for the treatment of Mucopolysaccharidosis IIIA* **Molecular Therapy- *Methods & Clinical Development,* 2019** Oct 28;15:333-342 ***corresponding author** PubMed PMID: 31788497; PubMed Central PMCID: PMC6881609
d. **Fraldi A***, Serafini M, Sorrentino NC, Gentner B, Aiuti A, Bernardo ME. *Gene therapy for mucopolysaccharidoses: in vivo and ex vivo approaches.* **. IJ of Pediatr. 2018; 44 (Suppl 2): 130. *co-corresponding author** PubMed PMID: 30442177; PubMed Central PMCID: PMC6238250

4. More recently, I discovered a new mechanism driving neurodegeneration in Sanfilippo syndrome. By studying mouse models of neuronopathic MPS, we demonstrated that neuronal cell bodies provide a major site for progressive deposition of multiple amyloid proteins including α-synuclein, PrP, Tau and Aß-protein. Importantly, we provided evidence that preventing amyloid deposition by using CLR01, a potent broad-spectrum inhibitor of amyloid self-assembly effectively protects against neurodegeneration by relieving autophagy-lysosomal dysfunction. Our data show for the first time that targeting amyloid aggregation is an effective strategy to treat the CNS pathology in neuronopathic MPS:

a. Sambri I, D'Alessio R, Ezhova Y, Giuliano T, Sorrentino NC, Cacace V, De Risi M, Cataldi M, Annunziato L, De Leonibus E and **Fraldi, A***. *Lysosomal dysfunction disrupts presynaptic maintenance and restoration of presynaptic function prevents neurodegeneration in lysosomal storage diseases.* **EMBO Molecular Medicine. 2017** 9, 112-132 ***corresponding author -cover highlight-** PubMed PMID: 27881461; PubMed Central PMCID: PMC5210158
b. Monaco A, Maffia V, Sorrentino NC, Sambri I, Ezhova Y, Giuliano T, Cacace V, Nusco E, De Risi M, De Leonibus E, Schrader, T, Klärner FG, Bitan G, **Fraldi A*.** *The amyloid inhibitor CLR01 relieves autophagy and ameliorates neuropathology in a severe lysosomal storage disease.* **Molecular Therapy 2020** *Apr 8;28(4):1167-1176.* ***corresponding author** PubMed PMID: 32087148; PubMed Central PMCID: PMC7132627
c. Monaco A and **Fraldi A**. *Protein aggregation and autophagy dysfunction: new lessons from mucopolysaccharidoses.* **Autophagy 2021**. Aug 18;:1-2. doi: 10.1080/15548627.2021.1961076 Pubmed PMID: 34407725
d. Giaccio M, Monaco A, Galiano L, Parente A, Borzacchiello L, Rubino R, Klärner F-G, Killa D, Perna C, Piccolo P, Marotta M. Pan X, Khijniak M, Siddique I, Schrader T, Pshezhetsky AV, Sorrentino NC, Bitan G and **Fraldi A***. *Anti-amyloid treatment is broadly effective in neuronopathic mucopolysaccharidoses and synergizes with gene therapy in MPS-IIIA.* **Molecular Therapy 2024**. doi: 10.1016/j.ymthe.2024.09.030 ***corresponding author**

Complete List of Published Work in My Bibliography (H-index = 27)
https://www.ncbi.nlm.nih.gov/myncbi/1v5s3Wlkb5zMLi/bibliography/public/

D. ADDITIONAL INFORMATION:

Patents
Granted
WO 2012085622 A1 "Modification of a sulfamidase enzyme to treat CNS pathology in MPS type III".

WO2020023094 A2 "Treatment of lysosomal storage disorders”
WO/2020/036656 A2 “Inhibition of lipofuscin aggregation by molecular tweezers”