

---

## BIOGRAPHICAL SKETCH

---

NAME: Fioriti, Luana

eRA COMMONS USER NAME (credential, e.g., agency login): FIOLUN

POSITION TITLE: Head of Laboratory, Istituto di Ricerche Farmacologiche Mario Negri, IRCCS

EDUCATION/TRAINING

| INSTITUTION AND LOCATION           | DEGREE  | Completion Date | FIELD OF STUDY |
|------------------------------------|---------|-----------------|----------------|
| University of Milan, Milan (Italy) | B.Sc.   | 02/2001         | Biology        |
| University of Milan, Italy         | M.S.    | 07/2004         | Biotechnology  |
| Open University, London, UK        | Ph.D.   | 02/2006         | Pharmacology   |
| Columbia University                | Postdoc | 10/2010         | Neuroscience   |

### A. Personal Statement

The goal of my research is to design novel therapeutic approaches to prevent or delay the onset of neurodegenerative disease. Over the past two decades, my research has focused on how protein conformation governs neuronal resilience, memory persistence, and vulnerability to neurodegeneration. I have repeatedly demonstrated that prion-like protein assembly can be either adaptive and neuroprotective or pathogenic and neurotoxic, depending on how cellular proteostasis mechanisms engage these conformational states. My group has contributed several of the defining conceptual advances establishing prion-like protein function as a regulated, physiological process, and not solely a mechanism of disease.

We have identified the small ubiquitin-like modifier SUMO2 as a master regulatory node that stabilizes proteostasis networks and preserves synaptic and metabolic function. Our work demonstrates that enhancing SUMO2 conjugation prevents and reverses cognitive impairment in preclinical Alzheimer's models, even after pathology is established. Like ubiquitin, SUMO can be covalently attached to target proteins and affect several aspects of the protein function. My interest in SUMOylation started during my postdoctoral training at Columbia University, in the lab of Nobel Laureate prof. Eric Kandel, a well-known expert in the field of memory, when I was studying the role of the prion-like protein CPEB3 in learning and memory. The observation that SUMO2 conjugation is an inhibitor constraint of the aggregation of the prion-like protein CPEB3, inspired my recent studies on regulation of tau aggregation and toxicity by SUMO in Alzheimer's disease (AD) and frontotemporal dementia (FTD), and PSP. In particular we are testing molecules able to modulate pharmacologically SUMO2 conjugation in vitro and in vivo, which could be further developed into drugs for patients. Our preclinical data suggest that modulation of SUMO2 reduces Tau aggregation, improves mitochondrial and synaptic function, and reduces neuroinflammation. I will ensure that the project is conducted in an efficient manner and with the highest scientific standards. In sum, I have demonstrated a record of successful and productive research projects in the neurobiology and neurodegenerative fields and I have the necessary expertise to achieve the goals of this proposal.

Ongoing and recently completed projects that I would like to highlight include:

1R01NS134902 (Quinzii/Fioriti – multiple PIs) 09/01/2023 – 08/31/2028

*Role of SUMOylation in mitochondrial/synaptic axis dysfunction induced by abnormal tau in FTD*

AARG-17-505136 (Fioriti – PI) 03/01/2017 – 05/31/2022

*Role of Sumoylation in the aggregation and toxicity of tau*

Three citations derived from work in my laboratory, not included in Section C of this biosketch, and directly relevant to this application are listed below:

**Fioriti L**, Wijesekara N, Argyrousi EK, Matsuzaki S, Takamura H, Satoh K, Han K, Yamauchi H, Staniszewski A, Acquarone E, Orsini F, Martucci A, Katayama T, Arancio O, Fraser PE. Genetic and pharmacologic enhancement of SUMO2 conjugation prevents and reverses cognitive impairment and synaptotoxicity in a preclinical model of Alzheimer's disease. *Alzheimers Dement*. 2025 Mar;21(3):e70606. doi: 10.1002/alz.70606. PMID: 40047257; PMCID: PMC11883658.

Orsini F, Pascente R, Martucci A, Palacino S, Fraser P, Arancio O, **Fioriti L**. SUMO2 rescues neuronal and glial cells from the toxicity of P301L Tau mutant. *Front Cell Neurosci*. 2024 Dec 12;18:1437995. doi: 10.3389/fncel.2024.1437995. PMID: 39726633; PMCID: PMC11669524.

Mansour G, Seminara, Mercurio, Bianchi, Porta, Dembech C, Perez S, Polito L, Durall C, Orsini F, **Fioriti L**, Comolli D, De Paola M, De Simoni MG, Gobbi M and Fumagalli S. Glycan-coated nanoparticles mimicking the ischemic glycocalyx scavenge the complement system conferring protection after ischemic stroke in mice. *Science advances* Manuscript Number: aed8928, under review

## **B. Positions and Honors**

### **Positions**

|                 |                                                                                                                                                                             |
|-----------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 11/2024-Pres.   | Adjunct Associate Professor, Mt. Sinai Icahn School of Medicine, New York                                                                                                   |
| 01/2021-Pres.   | Associate Member, Neurobiology Lab at UC Santa Barbara (directed by Kenneth Kosik)                                                                                          |
| 04/2019-Pres.   | Adjunct Associate Research Scientist, Department of Pathology and Cell biology, Taub Institute for the Aging brain, Columbia University                                     |
| 01/2019-12/2021 | Consultant, Plico Biotech, New York                                                                                                                                         |
| 01/2017-Pres.   | Head of Laboratory, Department of Neuroscience, Mario Negri Institute, Milan, Italy                                                                                         |
| 07/2016-12/2017 | Consultant, Rottapharm Biotech, Monza, Italy                                                                                                                                |
| 10/2010-05/2016 | Associate Research Scientist, Department of Neuroscience, Columbia University                                                                                               |
| 06/2007-10/2010 | Postdoctoral fellow, Howard Hughes Medical Institute                                                                                                                        |
| 10/2005-05/2007 | Postdoctoral Research Scientist, Department of Neuroscience, Columbia University                                                                                            |
| 04/2002-12/2005 | PhD Student, Open University, Prion Unit (directed by Dr. Roberto Chiesa), Dulbecco Telethon Institute and Mario Negri Institute for Pharmacological Research, Milan, Italy |

### **Honors**

|           |                                                                                                  |
|-----------|--------------------------------------------------------------------------------------------------|
| 2015      | Telethon Career Award                                                                            |
| 2009      | Mentorship Award, Columbia Mentoring Initiative                                                  |
| 2006-2007 | Fellowship, Italian Academy for Advanced Studies at Columbia University                          |
| 2004-2005 | Fellowship, "Fondazione Monzino" for PhD students                                                |
| 2002-2004 | Fellowship, the University of Milan to attend post-graduate School in Biotechnology Applications |
| 2002      | Scholarship grant to present at the World Alzheimer Congress, Stockholm                          |
| 2001-2002 | Fellowship, Mario Negri Institute for Pharmacological Research                                   |

**Professional Societies:** Society for Neuroscience (USA), New York Academy of Science, Molecular and Cellular Cognition Society, Italian Society for neuroscience, American Association for the Advancement of Science, **Reviewer:** Journal of Alzheimer Disease, Brain Sciences, Frontiers, Acta Neuropathologica, EMBO Journal, JBC, Alzheimer's Association, **Editorial Boards:** Neural Plasticity, Journal of Neuroscience Research

**Advisory Boards:** Yeshiva University Biotech Program, Metropolitan Montessori School Health Advisory

### C. Contributions to Science

The long-term **goal of my research is to develop therapeutic anti-aggregation tools**, which eventually can be used in *in vivo* situations. The overall objective I have is to study the cellular and molecular biology and pathology of prion-like infections and to use gained understanding for **delineating novel targets** for such intervention, in particular in the context of Alzheimer's Disease, Frontotemporal Dementia, and Huntington's Disease. Specifically, my most significant contributions to science are:

1. **Abnormal forms of the prion protein in the pathogenesis of prion diseases: the extracellular pathway.** In my doctoral thesis, I investigated two alternative pathways by which PrP might exert a toxic effect towards neuronal cells: 1) the **extracellular pathway**, consisting of neurodegeneration induced by PrP aggregates deposition in the brain, simulated by the employment of synthetic PrP-derived peptides on primary neuronal cultures. My work demonstrated that the effect of the peptides was not mediated by abnormal PrP species and suggested that PrP peptides could interfere with a physiological function of PrP (Fioriti et al., 2007) (Fioriti et al., 2005a), (Biasini et al., 2006), (Dossena et al., 2008).
  - a. **Fioriti L**, Quaglio E, Massignan T, Stewart RS, Salmona M, Harris DA, Forloni G, Chiesa R. The neurotoxicity of prion protein (PrP) peptide 106-126 is independent of the expression level of PrP and is not mediated by abnormal PrP species. *Mol Cell Neurosci.* 2005; 28(1):165-76. PMID: 15607951
  - b. **Fioriti L**, Angeretti N, Colombo L, DeLuigi A, Tagliavini F, Salmona M, Chiesa R, Forloni G. Neurotoxic and gliotrophic activity of a synthetic peptide homologous to Gerstmann-Straussler-Scheinker disease amyloid protein. *J Neurosci.* 2007; 27(7):1576-83. PMCID: PMC6673725
  - c. Biasini E, Massignan T, **Fioriti L**, Rossi V, Dossena S, Salmona M, Forloni G, Bonetto V, Chiesa R. Proteomic analysis of a transgenic mouse model of inherited prion disease reveals preclinical alteration of calcineurin activity. *Proteomics.* 2006; 6(9):2823-34. PMID: 16572473
  - d. Dossena S, Imeri L, Mangieri M, Garofoli A, Ferrari L, Senatore A, Restelli E, Balducci C, Fiordaliso F, Salio M, Bianchi S, **Fioriti L**, Morbin M, Pincherle A, Marcon G, Villani F, Carli M, Tagliavini F, Forloni G, Chiesa R. Mutant prion protein expression causes motor and memory deficits and abnormal sleep patterns in a transgenic mouse model. *Neuron.* 2008; 60(4):598-609. PMID: 19038218
2. **Abnormal forms of the prion protein in the pathogenesis of prion diseases: the intracellular pathway.** The second line of research related to the toxicity of the prion protein is about the **intracellular pathway**, consisting of the production of aberrant and mislocalised PrP species, like transmembrane or cytosolic PrP, induced *in vitro* by the administration of drugs interfering with maturation and PrP metabolism. As for the intracellular pathway, no evidence of cytosolic PrP was found in neurons expressing wild-type or mutant PrPs, thus arguing against the hypothesis that perturbation of PrP metabolism through the proteasomal pathway plays a pathogenic role in prion diseases. Instead, my data suggested that PrP may exert a protective, anti-apoptotic function (**Fioriti et al., 2005b**, Orsi et al., 2006; Drisaldi et al., 2003; Restelli et al., 2010).
  - a. Restelli E, **Fioriti L**, Mantovani S, Airaghi S, Forloni G, Chiesa R. Cell Type-specific Neuroprotective Activity of Untranslocated Prion Protein. *PLoS One.* 2010; 5(10):e13725. PMCID: PMC2965675
  - b. Orsi A, **Fioriti L**, Chiesa R, Siti R. Conditions of ER stress favour the accumulation of cytosolic PrP. *J Biol Chem.* 2006; 281(41):30431-8. PMID: 16908519
  - c. **Fioriti L**, Dossena S, Stewart LR, Harris DA, Forloni G, Chiesa R. Cytosolic prion protein (PrP) is not toxic in N2a cells and primary neurons expressing pathogenic PrP mutations. *J Biol Chem.* 2005; 280(12):11320-8. PMID: 15632159
  - d. Drisaldi B, Stewart RS, Adles C, Stewart LR, Quaglio E, Biasini E, **Fioriti L**, Chiesa R, Harris DA. Mutant PrP is delayed in its exit from the endoplasmic reticulum, but neither wild-type nor mutant PrP undergoes retrotranslocation prior to proteasomal degradation. *J Biol Chem.* 2003; 278(24):21732-43. PMID:12663673

**3. Structural analysis of CPEB and other polyQ and Q/N rich prions.** After my PhD, I became intrigued by the possibility that a prion-like molecular switch might operate under more physiological conditions and mediate the normal function of a protein. While Prions were initially identified as protein infectious agents causing neurological diseases by misfolding from their native conformation to a self-propagating, aggregation-prone  $\beta$  sheet-rich secondary structure, subsequent studies delineated a group of prions in yeast, which are not pathogenic. The structural transitions of functional prions are not easily conceivable as an uncontrolled  $\beta$  sheet misfolding process. Indeed, through bioinformatics and mutagenesis studies, I found the presence of Coiled Coil (CC) domains in CPEB, Ure2, and Huntingtin protein (Fiumara et al. 2010). Using structure-guided mutagenesis, we found that CC domains regulate the aggregation, insolubility, and activity of CPEB, Ure2 and huntingtin proteins. Based on these findings, we propose a model in which CCs have a critical role in the structural dynamics of functional prions and polyQ and Q/N rich proteins. These studies have continued with deletion analysis of the CPEB3 prion domain that uncovered a tripartite organization: two aggregation-promoting domains surround a regulatory module that affects interaction with the actin cytoskeleton, thus providing direct evidence that CPEB3 is a functional prion in the mammalian brain involved in synaptic plasticity and long-term memory (Stephan et al. 2015).

- a. Fiumara F, **Fioriti L**, Kandel ER, Hendrickson WA. Essential Role of Coiled Coils for Aggregation and Activity of Q/N-Rich Prions and PolyQ Proteins. *Cell*. 2010; 143:1121-35. PMID: PMC3472970
- b. Stephan J, **Fioriti L**, Lamba N, Colnaghi L, Karl K, Derkatch I, Kandel ER. CPEB3 is a functional prion that interacts with the actin cytoskeleton. *Cell Rep*. 2015; 11(11):1772-85. PMID: 26074072
- c. Ford L, Ling E, Kandel ER, **Fioriti L**. CPEB3 inhibits translation of mRNA targets by localizing them to P bodies. *Proc Natl Acad Sci U S A*. 2019 Sep 3;116(36):18078-18087. PMID: 31416913 PMID: PMC6731686
- d. Ford LK, **Fioriti L**. Coiled-Coil Motifs of RNA-Binding Proteins: Dynamicity in RNA Regulation. *Front Cell Dev Biol*. 2020; 8:607947. PMID: PMC7710910

**4. CPEB3 aggregation, regulation, and activity.** During my post-doc, I tested the idea that local protein synthesis, essential for memory storage, might be regulated by a CPEB functional prion. Indeed, the conditional knockout of CPEB3s that I generated has deficits in L-LTP (late phase of long term potentiation) and in the maintenance of long-term memory (**Fioriti et al.**, 2015). My findings provide the first evidence for a **functional prion in the mammalian brain** and illustrate that the CPEB-mediated protein synthesis is important in the maintenance of explicit memory. Since functional prions are potentially explosive, we were interested in knowing how they are regulated. I found that CPEB3 is dormant in the basal state and represses translation (**Fioriti et al.**, 2015), while it is activated by synaptic activity via ubiquitination mediated by the E3 ubiquitin ligase Neuralized1 (Pavlopoulos et al., 2011). On the contrary, SUMOylation appears to mediate the translational repression operated by CPEB3, by promoting its solubilization and competing with ubiquitination (Drisaldi et al., 2015). We recently obtained additional structural insights into the regulation of CPEB3 amyloid formation (Flores et al., 2025).

- a. **Fioriti L**, Myers C, Huang YY, Li X, Stephan J, Colnaghi L, Trifilieff P, Drisaldi B, Pavlopoulos E, Kandel E. The Persistence of Hippocampal-based Memory Requires CPEB3-mediated Protein Synthesis. *Neuron*. 2015; 86(6):1433-48. PMID: 26074003
- b. Drisaldi B, Colnaghi L, Levine A, Huang Y, Snyder AM, Metzger DJ, Theis M, Kandel DB, Kandel ER, **Fioriti L**. Cytoplasmic Polyadenylation Element Binding Proteins CPEB1 and CPEB3 Regulate the Translation of FosB and Are Required for Maintaining Addiction-Like Behaviors Induced by Cocaine. *Front Cell Neurosci*. 2020;14:207. PMID: PMC7365288
- c. Drisaldi B, Colnaghi L, **Fioriti L\***, Meyers C, Tarasoff J, Fraser PE, Kandel ER. SUMOylation is an inhibitory constraint that regulates the prion-like aggregation and activity of CPEB3. *Cell Rep*. 2015; 11(11):1694-702. PMID: PMC5477225 \*Co-first author.

- d. Flores MD, Sawaya MR, Boyer DR, Zink S, Tovmasyan S, Saucedo A, Richards LS, Zee CT, Cardenas J, **Fioriti L\***, Rodriguez JA. Structural insights into functional regulation of the human CPEB3 prion by an amyloid-forming segment. *Structure*. 2025 Aug 7;33(8):1314-1324.e5. doi: 10.1016/j.str.2025.05.007. Epub 2025 Jun 5. PMID: 40480223. \* co-corresponding

**5. Development of novel therapeutic approaches for AD and other neurodegenerative diseases.**

More recently I started to investigate the role of post-translational modifications in the modulation of aggregation, toxicity, and release of aggregation-prone proteins, such as Htt, Tau, and  $\alpha$ -synuclein, with the goal of designing therapeutic approaches aimed at boosting the protective SUMOylation pathway.

- a. Orsini F, Bosica M, Martucci A, De Paola M, Comolli D, Pascente R, Forloni G, Fraser PE, Arancio O, **Fioriti L**. SARS-CoV-2 Nucleocapsid Protein Induces Tau Pathological Changes That Can Be Counteracted by SUMO2. *Int J Mol Sci*. 2024 Jun 28;25(13):7169. doi: 10.3390/ijms25137169. PMID: 39000276; PMCID: PMC11241313.
- b. Ford L, **Fioriti L**, Kandel ER. Ubiquitination and SUMOylation of Amyloid and Amyloid-like Proteins in Health and Disease. *Curr Issues Mol Biol*. 2019 Aug 18;35:195-230.
- c. Zanier ER, Barzago MM, Vegliante G, Romeo M, Restelli E, Bertani I, Natale C, Colnaghi L, Colombo L, Russo L, Micotti E, **Fioriti L**, Chiesa R, Diomedea L. *C. elegans detects toxicity of traumatic brain injury generated tau*. *Neurobiol Dis*. 2021 Jun;153:105330. doi: 10.1016/j.nbd.2021.105330. Epub 2021 Mar 10. PMID: 33711491; PMCID: PMC8039186.

**Complete List of Published Work in MyBibliography:** <https://www.ncbi.nlm.nih.gov/pubmed/?term=Fioriti+Luana>