Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Annual Report 2025

INDEX

Page 2 NICO by the numbers
Page 3 Brief historical notes
Page 6 Organisation at NICO
Page 9 Plans for internationalization
Page 9 Outreach activities
Page 10 New challenges
Page 12 ACTIVITIES OF THE SINGLE GROUPS
Page 12 PI Bonfanti- Peretto
Page 34 PI Buffo
Page 58 PI Di Cunto
Page 67 PI Bertocchi
Page 79 PI Gotti
Page 88 PI Raimondo
Page 103 PI Tamagno
Page 111 PI Tempia
Page 122 PI Vercelli
Page 153 PI Di Sapio
Page 167 Seminars

NICO 2025 by the numbers

10
Research Groups
102 Scientists

77
Peer-Reviewed
Publications

86
Collaborative Initiatives
with
International
Research Groups

51
On-going/Granted
Research Projects

16
Scientific
Conferences/workshops
organized
by NICO members

27
Invited speakers

1
Spin-off Company

1
Biobank

46
Outreach Activities
20 Invited Talks
26 Science Dissemination Initiatives

35
trained
PhD students

4845
Facebook Followers

BRIEF HISTORICAL NOTES

The Cavalieri Ottolenghi Foundation is a no-profit organisation recognized by the Piedmont Region committed to supporting research and development of structural and infrastructural Neuroscience. The Foundation comes from the legacy to the University of Turin of Annetta Cavalieri Ottolenghi in the 50s.

After decades in which the Foundation has funded scientific research projects and purchase of scientific equipment, at the end of the last century an international scientific committee of eminent personalities in Neuroscience proposed to build a centre for Neuroscience and chose Dr. Carlos Dotti, a foreign researcher, as scientific director. For some years, the research group of the Foundation was hosted at the San Luigi Hospital, Orbassano (Torino) while the construction of the building began in 2001 and was completed in 2009. Meanwhile, Dr. Dotti moved abroad with his group.

In 2009, the Board of Administrators identified in Prof. Ferdinando Rossi, University of Turin, the figure of the scientific director of the Foundation, decided to issue an announcement of selection, limited to the Piedmont Region, to select the groups to be included in the new building. Eight groups (seven of the University of Turin, one of the San Luigi Hospital) were selected by a national committee (which included Professors Bentivoglio, Bogetto, Cattaneo and Saglio, assisted by Dr. Borio, Administrative Director at the University of Turin) and in May 2010 they moved into the new building. The institute was named Neuroscience Institute Cavalieri Ottolenghi (NICO).

On January 28 and 29, 2016 a Panel of external reviewers (P. Alves, Lisbona, M. Bentivoglio, Verona, M. Celio, Fribourg) visited NICO. After evaluating activities and researcher of NICO during the first five years, they issued a report, which is available on the NICO website. On January 13 and 14, 2020, the new international Advisory Scientific Committee made a second onsite visit: their report can be found at the link https://www.nico.ottolenghi.unito.it/eng/Institute/Scientific-report

AIMS OF NICO

1) The complexity of the studies on the brain requires a multidisciplinary approach. For this, we combine complementary approaches and experiences, integrating basic research and clinical application. The birth of NICO takes full advantage of both the integration of the wealth of knowledge and the shared use of expensive equipment and laboratories formerly fragmented in university Departments.

2) Our researchers are engaged in many activities of scientific dissemination, dedicated to the public (Open days at the NICO, Stem Cell Day and Night of Researchers, public conferences) and to high school students (Neuroscience Olympics and Scientific Summer Academy). These and other initiatives are designed to bring young people to science, by sharing the commitment and passion that drives scientific research, as well as to communicate with competence and clarity a complex issue such as neuroscience.

NICO aims to perform high-level research in neuroscience geared towards the prevention, diagnosis and treatment of neurological disorders. In line with this principle, the research is focused on mechanisms that govern normal neural maturation and defects involved in mental retardation syndromes.

THE COLLABORATIVE VISION AT NICO

Since its foundation in 2010, the NICO adopted a new (relative to the Departments of origin) view of sharing all facilities, supplies and instruments by all groups. Excepted for the clinically relevant activities, which must be performed in dedicated and confined rooms to maintain privacy related to human material, all instruments are located in common facilities which are shared by all NICO members. This has initially created an organizational burden, but it has also obliged people to meet, share decisions, collaborate, and interact, also in the formation of new researchers. Internal courses on the use of instruments and facilities have been organised to improve their correct usage. Starting from the practical needs of everyday research life, this attitude has boosted collaboration and exchange of ideas among the individual researchers and ameliorated the scientific production of single researchers. To sum up, it has created a scientific environment, which, respecting the peculiarities of single researchers interacts and operates as a real community to apply

for grants, develop multidisciplinary projects, act as a whole institutional body in front of the scientific community and to the public. Finally, it represents a fundamental breakthrough to save money and to exploit the use of expensive instruments.

POSITIONING OF NICO IN THE UNIVERSITY OF TURIN

NICO is part of the University Interdepartmental centre for Neuroscience (called Neuroscience Institute of Turin – NIT), which gathers most researchers active in the field in Turin (even outside the University). NICO researchers are part of doctorate schools (Neuroscience, Bioengineering and Medical-Surgical Sciences, Molecular Biotechnology, Complex Systems for Bioquantitative Medicine and Veterinary Medicine, Sustainable Development and Cooperation) of the University of Turin and of the National doctorate in Sustainable Development and Climate Change of the IUSS (Pavia), and hosts 35 PhD students. Moreover, as lecturers at the schools of Biology, Biotechnology, Pharmacy, Medicine, Psychology and Veterinary Medicine, NICO researchers are involved in tutoring students in the preparation of theses for bachelor's and master's degrees. Moreover, NICO researchers are directly involved in the organization of the master's degree in Biotechnology for Neuroscience (of which prof. Di Cunto and Boido are respectively president and vice president).
Currently, NICO laboratories host around 50 students who are developing their Bachelor/Master thesis projects and stage.
NICO collaborates with several other research centres of the University of Turin, such as the Molecular Biotechnology centre, the IRCCS Candiolo and the Brain Imaging Centre.
NICO members belong to the Departments of Neuroscience R. Levi Montalcini, Clinical and Biological Sciences, Veterinary Sciences and Systems Biology. NICO members belonging to the Department of Neuroscience and Veterinary Sciences of UNITO have participated in the projects which were awarded by the MIUR Departments of Excellence 2017-2022. The Departments of Neuroscience and Clinical and Biological Sciences were awarded departments of excellence at the end of the 2022 for the 2023-2027 period. Therefore, NICO are involved in these five-year projects, with a significant return in terms of personnel, upgrade of instrumentation and translational science collaborations.
In particular, NICO plays a key role in the project of Excellence of the Department of Neuroscience, since it is fundamental in a) creating a C. Elegans lab for modelling neurodegenerative diseases, b) creating an IPSC and organoid platform to study neural diseases and new drugs and finally c) directing part of the research to space, in the frame of the regional and industrial plans for the "Cittadella dello Spazio" (according the Space Research Platform of UNITO).
Several groups of the NICO have projects and funding in collaboration with the Polytechnic of Turin.
Starting from 2017, the microscopy facilities at the NICO are part of the Open Access lab program of the University of Turin, and recently European project of the University of Turin RE-UNITA (workpackage 4). Within this frame, in 2020 the microscopy facilities at NICO were reorganised to create the Platform for Imaging Cavalieri Ottolenghi, PICO, https://www.nico.ottolenghi.unito.it/eng/PICO-). This facility will be soon included in the national facilities of the D34H Foundation, in order to facilitate their sustainability.

POSITIONING OF THE NICO IN ITALY AND IN THE WORLD

In addition to the one with UNITO, NICO has signed several formal agreements to host students at the Universities of Camerino, Insubria, Palermo, Parma, Siena, Trieste and Verona.
NICO researchers have several national and international collaborations, as shown by their publication record. They have also a strong rate of exchange of visits and seminars, as it can be argued from their reports in attachment. In addition, they participate to exchange programs of bachelor, graduate and doctorate degrees, and NICO is often visited and attended by foreign students. They participated in the international Young Investigators Training Program established in Italy in occasion of the 2011 World International Brain Research Organisation Meeting and of the 2014 European Neuroscience Meeting. Every second year, an international meeting (Steroids and Nervous System) is organized by Neuroendocrinology group (with the cooperation of prof. R.C. Melcangi, University of Milan): the meeting has an average 150 people attendance and more than 40 invited speakers from all over the world. The 2013, 2015, 2017, 2019, (virtual) 2021 and 2024 editions were organized with the administrative help of the Ottolenghi Foundation. The next will be held in February 2026.

In 2024, Prof. Boido and Dr. Stanga organised for the third time a biannual international workshop on motoneuron diseases which was attended by 115 people, and the next workshop is in preparation for November 2026.
NICO researchers are/have been members of committees for national and international meetings and societies and acted as referees for international peer review journals and panels of funding agencies. From January 2022 to December 2023 the NICO scientific director, A. Vercelli was acting as president of the Italian Society for Neuroscience and now serves as past president until end 2025.
The Clinical Neurobiology group organises local and national meetings on multiple sclerosis at the San Luigi Hospital.
NICO has been credited by MUR (Ministry of University and Research) in the list of the Italian Private Research Institutes for the first time in 2015 and thereafter (last time in December 2021).
Following the suggestions of the international Advisory committee to improve the international interactions of the NICO, from January 2022 NICO has hired a consultant for internationalization, Dr. Mariasilvia Ciola. In 2022 this led to informal contacts with other institutes such as the Cajal Institute of Madrid, the Department of Neuroscience of the University of Rio de Janeiro and with Israeli institutions. In 2023 a visit to several Universities in Japan (University of Tokyo, University of Osaka and Riken Institute at Kyoto) and to Brazil (Sao Paulo, Minas Gerais, Brasilia, Belo Horizonte and Rio de Janeiro) have been performed to explore the possibilities of joint projects and collaborations.
Dr. Ciola also proposed and organised the NICO participation at AIRI, the Italian Association of Industrial Research, which is the network of the main enterprises committed in R&D (e. g. pharma, bio-medical, high-tech).
Researchers at NICO participate(d) to the PNRR projects in the frame of the European "Recovery Europe" D34H (digital and biological twin of the patient), Innova, Nodes, Mnesys, Anthem and TO Move.

NICO AS A GREEN LAB

NICO and its researchers are strongly committed in the actions related to the European Green Deal.
First, its researchers are performing several projects related to the effects of pollutants on the nervous system and, on the reverse, on the beneficial effects of the green environment on brain health. Second, they participate to the new local and national doctorate schools related to Sustainable Development and Climate Change. Third, they are involved in the dissemination of these knowledges (see the organisation of the show "The mountain touch" in collaboration with the Museum of Mountains in Torino and the Italian National Institute of Health) and of the concept of One Health, strongly related to Brain Health. Fourth, together the Council of Administration, the director is preparing a plan of reduction of energy usage. Finally, a committee has been nominated in 2024 and is active to suggest policies and activities to change the attitude of the people and participate to cleaning of the environment.

NICO AS PART OF THE ALBA NETWORK

The Institute has signed the ALBA declaration on equity and inclusion in science. From the website of the ALBA network: "Members of underrepresented groups face persistent barriers to equitable representation in science, technology, engineering and mathematics (STEM), particularly at advanced stages. Although the historical basis for and manifestations of underrepresentation vary by group, discipline, and region, there are striking commonalities in the result – an apparent 'leaky pipeline' that drains the talent pool. The cost of this loss of talent is high – for individuals, for research, and for society as a whole. ALBA is a network of brain scientists committed to fostering fair & diverse scientific communities. We have drafted this document as a resource for concrete, positive, evidence-based actions that individuals and organizations at any level can take to promote equity & inclusivity. We focus specifically on two contributing factors to perpetual underrepresentation in STEM: implicit bias & workplace culture. We believe that adopting the actions below will benefit all members of the research community and the scientific enterprise itself." Prof. Boido represents the NICO in the network.

Illustration of the organizational structure and research indicating the current staff, including contractors, and their qualifications, and of the educational, scientific and instrumental activities.

ORGANIZATION OF THE NICO (Neuroscience Institute Cavalieri Ottolenghi)

Scientific Director is Prof. Alessandro Vercelli (confirmed up to June 2027). In addition to the scientific direction, he performs also the function of Administrative Director. From November 2018, prof. Annalisa Buffo was appointed vice-Director for the activities at the NICO.

Our activities are organized into ten groups:
Adult Neurogenesis (PIs Luca Bonfanti and Paolo Peretto)
Ageing and Alzheimer's disease (PI Elena Tamagno)
Brain Development and Disease (PI Alessandro Vercelli)
Clinical Neurobiology (interim PI Alessia Di Sapio)
Embryonic Neurogenesis (PI Ferdinando Di Cunto).
Nerve Regeneration (PI Stefania Raimondo – formerly S. Geuna)
Neuroendocrinology (ff PI Stefano Gotti)
Neurophysiology of Neurodegenerative Diseases (PI Filippo Tempia)
Neuropsychopharmacology (ff PI Ilaria Bertocchi)
Physiopathology of Neural Stem Cells (PI Annalisa Buffo)

STAFF

Employees directly depending on the Foundation consist of **three secretaries** (Maria Lo Grande, Susanna Monteleone and Alessandra Moretti) and **two technicians** (Sri Satuti Werdiningsih and Martir Dyrmishi).

We have professional counseling contracts with 1) **a journalist, for digital and strategic communication and disseminations activities** (i.e. press office, events, website and social media manager), Dr. Barbara Magnani, 2) a **consultant for fund raising and stakeholder management**, Dr. Alessandra Gerbo, and 3) **a consultant for internationalization**, Dr. Mariasilvia Ciola.

According to the Convention with the University of Turin and with San Luigi Hospital of Orbassano, the NICO hosts:
- **University staff**: 7 full professors, 15 associate professors, 8 university research assistants, 5 technicians, 27 post-docs/bursaries and 35 doctoral students;
- **Hospital staff**: 1 manager biologist, 2 specialists in Clinical Biochemistry, 2 post-doc fellows.

About 70 graduating students of Biology, Biotechnology, Medicine and Psychology perform experiments for their thesis at the NICO.

LABS AND EQUIPMENT

Thanks to the involvement of the Department of Neuroscience R- Levi Montalcini, which was awarded twice the seal of department of excellence, NICO could update its facilities consistently.

Molecular and cellular neurobiology, neuroanatomy

The laboratory is equipped with several excellent quality research light microscopes, in particular, two confocal microscopes (Leica SP5 and Nikon), which are being dismissed soon, and a Nikon ViCo system, an Axioscan Zeiss slide scanner and an UltraMicroscope LaVision/Miltenyi Biotec. In 2025, it has acquired a Zeiss LSM 980 Super-resolution confocal microscope. A Sartorius IncuCyte S3 microscope for automated measurements and Multielectrode Arrays are available for cell cultures. Moreover, a two-photon microscope Nikon (A1MP) is in function. A Zeiss Gemini 460 scanning electron microscope for 3D reconstructions is in function. A conventional transmission electron microscope is available in the Department of Cell Biology, San Luigi Hospital, adjacent NICO.

There are also various imaging systems with computerized microscopes and photo / digital video cameras that allow morphometric investigations, studies densitometry quantitative autoradiography, image processing and statistical analysis. Two Neurolucida systems are in the microscopy facility.
For neurohistological studies, sliding or rotational microtomes, 3 vibratomes and 4 cryostats are available.
In 2026 the facility will be implemented with a Slice and a Vesalio systems by Microbrightfield and a spinning disk microscope.
The facility is run by two experienced technicians in confocal and electron microscopy.

Animal facility

The structures devoted to the experimental animals include rooms dedicated to housing and breeding, spaces dedicated to behavioural tests and, finally, rooms equipped for surgery on rodents. The laboratory for behavioural tests is equipped with mazes and infrared cameras for the behavioural analysis of locomotor activity, anxiety, depression and memory. There is also a computerized video analysis (Ethovision XT video track system) to analyse scanned images of behavioural tests. Finally, dedicated spaces, equipped for P2 procedures are available to use viruses of the corresponding biosafety level and to inject them in animals.

Cellular And Molecular Biology

NICO has excellent facilities for research in the field of molecular and cell biology, and a dedicated and experienced staff for tissue culture experiments and molecular biology.
Tools for cell biology experiments allow cell count, freezing, plating tissue culture and cell transfection and transduction. For in vitro and ex vivo cultures (primary cultures, tissue explants, organotypic cultures, neurospheres) inverted microscopes are available and a system that allows the acquisition of images in time-lapse of viable cells. A cell culture room devoted to human pluripotent cell derived 2D and 3D models has recently been implemented.
In addition, NICO provides expertise and services related to molecular biology techniques, such as the preparation and analysis of proteins, DNA, RNA and microRNA. The instrumentation of molecular biology platform includes a semi-automatic system for the purification of nucleic acids, three machines for Real-Time PCR, an electroporator for bacteria or mammalian cells, as well as many other instruments as a standard laboratory for extraction and analysis of DNA, RNA and proteins.

Electrophysiology

The laboratory of neurophysiology provides tools for the preparation of micro-sections of nervous tissue that can be maintained in vitro for several hours. There are two experimental stations for patch clamp recordings of membrane potential or ionic current of single neurons in sections. These positions are furnished with complete tools for the electrical stimulation of the axons and for application of pharmacological substances. They can also make extracellular recordings to study synaptic plasticity.
For in vivo recordings, systems for multichannel recordings and calcium photometry are in place.

C. Elegans Facility

The C. elegans laboratory offers all the necessary expertise, instruments and strains necessary to utilize this genetically-tractable animal model for studying disorders involving altered neural development and neurodegeneration. In particular, many strains expressing fluorescent neuro-specific markers, an oocyte-injection setup for production of transgenic strains and three setups for functional assays and behavioral analysis are available. The facility is run by a dedicated technician.

Clinical Neurobiology Laboratory (Cnl)

The CNL offers diagnostic services and consulting for the interior (San Luigi Hospital) and external (10 Departments of neurology in the region) diagnosis of multiple sclerosis.
The diagnostic tests offered include cytochemical examination of cerebrospinal fluid, immunoisoelectrofocusing to search for oligoclonal bands and several essays for the detection of viral nucleic acids. In addition, the laboratory provides a diagnostic service for neuronal paraneoplastic antibodies.

Currently the CNL is one of the few laboratories in Italy capable of providing a diagnostic service for the detection of antibodies NMO-IgG and anti-AQP4.
Finally, the CNL offers various services for monitoring patients with multiple sclerosis treated with different drugs; in this regard the lab performs a service in Italy and Europe for the serological titration of antibodies against interferon-beta (using three different methods) and natalizumab (Tysabri) potentially produced by patients treated with these drugs.
The laboratory is also equipped with a service for the evaluation of the biological activity of interferon-beta through the measurement of gene expression of specific proteins induced by interferon (such as MxA).

Common Services

In addition to spaces dedicated to animal facility and laboratories, there are two rooms for the secretariat, a staff kitchen, a hall serving as cafeteria and meeting point (with automatic machines), a seminar room and a room for deep freezers.

Personnel

During 2025, University personnel stabilized in number, excepted for the arrival of new technicians and the promotion of some researchers.
Dr. A. Di Sapio, chief of the Hospital Neurology Unit, serves as PI of the group of Clinical Neurobiology.
The contract with Charles River has just been renewed by the University of Torino for the animal house.

Upcoming Projects On Instrumentation, Personnel And Facilities

To further promote the implementation of instruments, the Scientific Director applied to Regione Piemonte for an implementation of the microscopy facilities, together with Polytechnic of Turin: the project (Pastiss+) was awarded, the project is effective from December 2025 and new equipment will be acquired for the microscopy facility in 2026.

Some Considerations Regarding Research Funding

Members of the NICO have raised in the year 2024 more than 7.5 million € in grants for UNITO and 0.4 million € for FCO. Moreover, Alessandro Vercelli is local coordinator for the PNRR project D34H, for which UNITO is receiving 4.3 M€, and for the PNRR project INNOVA (Ministry of Health) and collaborate to the project TO Move. Prof. Boido and Calì participate in the PNRR project NODES: from October 2024 Prof. Boido is the leader of the Spoke 5 "Industry for Health and Silver Economy". The members of the Department of Neuroscience who work at NICO actively participate in the project of Excellence of MUR who was awarded at the end of 2022 and is effective in 2023-2027 for an overall grant of around 8 M€, in the field of Basic Neuroscience: the project was written by A. Vercelli. Also, Prof. Boido, Buffo and Vercelli received three cascade grants from PNRR projects (Mnesys and Anthem) and Prof. Di Cunto participates to a PNRR-TR1-2023 project. Many of these project have closed their activities in 2025 or will close soon, and the researchers are applying for new grants. In particular, Prof. Buffo has been awarded a Synergy grant at the end of 2025.
The current agreement between UNITO and FCO foresees a contribution from UNITO to FCO of the 50% of the running costs of NICO from UNITO (around 200.000 € in 2025). Members of UNITO working at NICO apply for the governmental funding though UNITO (which is relevant to the ranking of UNITO in Italy, and of the Departments with members affiliated to NICO among the other Departments. 12 projects from NICO researchers were financed by the PRIN plan of the Ministry of Research: all these grants terminated in 2025.
In addition, when possible, members of NICO apply directly to agencies though FCO administration. This led to a certain amount of grants directly administered by FCO: the relative amount of grants was very low in 2019 (33.000 €) and is increased significantly in the following years notwithstanding the reduction of activity of the Clinical group due to the retirement of the PI. The agreement with the San Luigi hospital (under renewal) foresees a yearly contribution of 25.000 € to FCO.
The Foundation Cavalieri Ottolenghi, has been awarded a three-year (2023-25) contribution from the Italian Ministry of Research as private Research institution.

Fund Raising

From 2024 the collaboration with fundraising consultant Dr. Alessandra Gerbo went on in 2025. The main objectives of her work were to develop further relations with philanthropic foundations and other stakeholders with a potential role in helping the Foundation's fundraising, to coordinate the NICO 5 x 1000 campaign and to ensure the development of the new website include the fundraising perspective.
In 2025, NICO, with the help of the Industrial Liaison Office of UNITO, we organized a meeting with companies in order to show our scientific and dissemination activities. After this event, it has been organized a series of seminars which companies may provide to their employees, under the frame of their welfare programs, with a donation to the NICO.
Dr. Gerbo also worked to the submission of some project requests in response to call issued by local philanthropic foundations. The most relevant development in this respect was the winning of an award of 50.000 € from Compagnia di San Paolo's Next Generation You Program. The amount has been used to develop an organisational strengthening programme lasting 18 months (from December 2024 until mid-2026) for all the members of NICO. The programme, written in collaboration with ON! Srl consultants (that also help its implementation), focuses on many aspects strictly connected with fundraising: strategic positioning and articulation of NICO's value proposition in relation to different stakeholders, boosting of management skills for key staff members, assessment of internal processes and their improvement to gain a more strategic vision and a greater efficiency in planning and decision-making.
Finally, a program for fellowships for young talents was funded following support from charities and foundations, in which every year a few salaries are offered to young people after a competitive selection, then the winners can choose a group of research in which to spend the year of work. The chosen groups must cofund the fellowship.

PLANS FOR INTERNATIONALIZATION

During 2024, the internationalization programme began to take shape and the first achievements were accomplished.
As far as the Neuroscience Institute of excellence Network – NIN is concerned, the first two agreements were signed with the Instituto Cajal in Madrid and IDOR in Rio de Janeiro. Moreover, talks are ongoing with the Neuroscience Department of the University of Osaka (Japan) and the Centre for Cognition and Sociality of the Institute for Basic Science - IBS (Republic of Korea).
Prof. Vercelli served as vice-Rector for Biomedical Research until 2024 and in this capacity, he participated in other institutional missions of University of Torino. In particular, on the occasion of the second mission to South Korea (September 2024), agreements were finalised with IBS both for the start of collaboration at PhD level and for the participation of IBS neuroscience centres in the network promoted by NICO.
New collaborations for NICO researchers were also made possible thanks to the director's participation in the institutional mission to Singapore in June 2024. Webinars with the National University of Singapore Medical School took place in the second half of the year.
In 2025 members of the IDOR and IBS visited the NICO, and together with members of the NICO participated in a symposium. Two PhDs were enrolled in the collaboration with IBS and will spend half of the doctorate in Daejeon to foster the collaboration.
In the meantime, NICO project to create an international network of Neuroscience Institutes was funded by Compagnia di San Paolo. In 2025, we started to create the network, and discussed with the Rita Levi Montalcini Foundation the possibility to name the network "Rita Levi Montalcini Neuronet". All this organizational work will serve to the organization of the Celebrations for the Rita Levi Montalcini Nobel Prize 40Th Anniversary in collaboration with UNITO, to be held in end December 2026 together with the launch of the RLM Neuronet.

OUTREACH ACTIVITIES

From the perspective of educational and scientific dissemination the aims of NICO are:
• to promote scientific culture, and in particular knowledge of neuroscience, in high schools, through multimedia tools that reduce the economic impact of training initiatives.
• to provide basic skills on the normal functioning of the brain and neurodegenerative processes;
• to explain the importance of basic research and the impact on society of tomorrow;

• to create synergies and exchange of expertise / experience in the world of university research, the school and society, represented in this case from the large network of voluntary associations active in the field of disability and dementia.

Dissemination activities in 2025

• Unistem Day (March 14); UniStem Day 2025, dedicated to Giulio Regeni;
• Olympic Games of Neuroscience Piedmont (March 15);
• Brain Awareness Week (March 11 and 12)
• 5xmille Campaign (on web and social media, from April to September)
• Pint of Science (Scientific events in pubs May 19-21)
• Festival dell'Innovazione e della Scienza Settimo T.se (October 5-12) 2 Scientific Cafès
• TrendSanità Policy and Procurement in Health Care (online and in presence Conferences)
• Web (Italian research news and news || English research news) and social media dissemination (Facebook - Instagram - LinkedIn - YouTube)

NICO is engaged in scientific activities dedicated to high school students - Olympic Games of Neuroscience and Unistem Day, national and international – as well as to general public (Researchers' Night and Brain Awareness Week).
These activities were possible thanks to a partnerships network that, starting by the University of Turin has expanded in the years throughout other universities, associations (e.g. Non-profit Associations) and institutions like Centre Agora Science (which brings together the University of Turin and East Piedmont and Polytechnic of Turin). They have allowed establishing direct contacts with teachers and high school students. NICO is organizing the regional competition of the World Olympic Games of Neuroscience: every year in the world, high school students participate in a competition to stimulate interest in the study of neuroscience. The competition begins with the sending of educational materials to schools, then a local selection in schools (in Piedmont hundreds of students), regional (at the NICO) and finally a national one in which the Italian "champion" is chosen for the world competition.

The Institute has a strong link with the Piedmont Associations of patients with disabilities (e.g. the Coordination Committee for Tetraplegic and Paraplegic patients of Piedmont - Sportello Lesioni spinali) and neurodegenerative diseases and their families (CAAP - Coordination committee of Alzheimer Associations of Piedmont -12 local associations - the Ass. Of Parkinson friends of different provinces of the region, the Association Girotondo Onlus for SMA patients in Biella, etc.).

NICO is involved in the organization of a series of dissemination lectures for the public, some of which on the Brain Awareness Week (which is held worldwide in March). The goal is to provide accurate information on scientific topics not easy to understand / disclose - such as the state of research and therapies available on neurodegenerative diseases - and often the subject of simplification and distortion.

Organization and scientific supervision of Unistem Day (yearly, international event; NICO organizes each year the Turin edition), Aula Magna of Palazzetto Aldo Moro (with 400 students at the secondary school).

SCIENTIFIC SEMINARS AT NICO

19 seminars were held on Friday afternoons. For invited speakers, see the attached list.

NEW CHALLENGES

The perceived need for change must be contextualized within a scenario of profound transformation that the Foundation and NICO are going through, produced by the convergence of three main instances. The first is represented by the perspective, in 2027, of moving the Institute and its laboratories to the new university center which is being built in the area of the so-called "Ex Scalo Vallino" in Via Nizza, with the consequent disposal of the structure in Orbassano, which currently has onerous management costs. From this perspective, further resources could therefore become available to fuel the organisation's mission. Secondly, the Reform of the Third Sector, which in some way forces the Foundation to deal with a profoundly evolving scenario

and to have to evaluate possible changes to best seize the opportunities that will arise. Thirdly, the need to adapt the operational and management structure of the Foundation to new and more complex needs triggered, for example, by digitalisation, by the increase in economic resources to manage and by competition from other non-profit entities on the market for donations from private: to this aim, we are developing a project funded by Compagnia di San Paolo in the frame of the program NGY.
Finally, following the requests of the Board of Auditors, and of the Council of Administration, the Foundation is preparing a regulation and a website for transparency in the public administration, to comply anti-corruption rules by the ANAC.
In light of the profound changes underway and with a view to enhancing the potential demonstrated by NICO, the Foundation therefore feels the need to return to focusing attention on itself, on the organization and on the processes that characterize it, in order to adapt to the new context and to be able to grasp the potential that flows through it in the design of its medium-long term development strategy.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: **Adult Neuroplasticity**

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Principal Investigator 1

Luca Bonfanti
- Degree: DVM, PhD
- Nationality: Italian
- Birthdate: 19/05/1962
- Gender: Male
- Phone: +39 011 6706606
- Email: luca.bonfanti@unito.it
- Position: Principal Investigator

Principal Investigator 2

Paolo Peretto
- Degree: PhD
- Nationality: Italian
- Birthdate: 18/09/1963
- Gender: Male
- Phone: +39 011 6706605
- Email: paolo.peretto@unito.it
- Position: Principal Investigator

Personnel

Silvia De Marchis
- Degree: PhD
- Nationality: Italian
- Birthdate: 14/09/1966
- Gender: Female
- Phone: +39 011 6706605
- Email: silvia.demarchis@unito.it
- Position: Associate Professor
- Role & Expertise: Lead researcher on postnatal neurogenesis in mouse models.

Federico Luzzati
- Degree: PhD
- Nationality: Italian
- Birthdate: 20/10/1974
- Gender: Male
- Phone: +39 011 6706615
- Email: federico.luzzati@unito.it
- Position: Associate Professor
- Role & Expertise: Lead researcher on lesion-induced neurogenesis in the striatum of mammals.

Serena Bovetti

- Degree: PhD
- Nationality: Italian
- Birthdate: 13/09/1977
- Gender: Female
- Phone: +39 011 6706613
- Email: serena.bovetti@unito.it
- Position: Associate Professor
- Role & Expertise: Lead researcher on the study of neural networks involved in sexual imprinting.

Sara Bonzano

- Degree: PhD
- Nationality: Italian
- Birthdate: 22/03/1987
- Gender: Female
- Phone: +39 011 6706632
- Email: sara.bonzano@unito.it
- Position: Assistant Professor (RTD-A)
- Role & Expertise: Cellular and molecular analyses of adult neurogenesis in the olfactory bulb and hippocampus; morphometric assessment of mitochondria ex vivo.

Stefano Zucca

- Degree: PhD
- Nationality: Italian
- Birthdate: 29/05/1988
- Gender: Male
- Phone: +39 011 6706632
- Email: stefano.zucca@unito.it
- Position: MSCA Post-doctoral Fellow
- Role & Expertise: Two-photon and light-sheet microscopy, electrophysiology.

Marco Fogli

- Degree: PhD
- Nationality: Italian
- Birthdate: 23/09/1993
- Gender: Male
- Phone: +39 011 6706632
- Email: marco.fogli@unito.it
- Position: PostDoc – Research Fellow (since February 2024)
- Role & Expertise: Cellular and molecular analyses of lesion-induced neurogenesis and analysis of human brain organoids.

Ilaria Ghia

- Degree: Biological Sciences
- Nationality: Italian
- Birthdate: 04/01/1996
- Gender: Female
- Phone: +39 011 6706632
- Email: ilaria.ghia@unito.it
- Position: PhD Student (37th Cycle)
- Role & Expertise: Two-photon and light-sheet microscopy, histology, mouse models.

Eleonora Dallorto

- Degree: Biological Sciences
- Nationality: Italian
- Birthdate: 04/07/1996
- Gender: Female
- Phone: +39 011 6706632
- Email: eleonora.dallorto@unito.it
- Position: PhD Student (37th Cycle)
- Role & Expertise: Histology, Western blot analysis, confocal and high-resolution microscopy, morphometric analysis, mouse models, human iPSC-derived neurons in vitro.

Alessia Pattaro

- Degree: Biological Sciences
- Nationality: Italian
- Birthdate: 20/11/1997
- Gender: Female
- Phone: +39 011 6706632
- Email: alessia.pattaro@unito.it
- Position: PhD Student (38th Cycle)
- Role & Expertise: Cellular and molecular analyses of immature neurons in mammals.

Flavia Maria Domizia Di Fiore

- Degree: Chemistry and Pharmaceutical Technology
- Nationality: Italian
- Birthdate: 13/03/1998
- Gender: Female
- Phone: +39 011 6706632
- Email: flaviamariadomizia.difiore@unito.it
- Position: PhD Student (40th Cycle)
- Role & Expertise: Two-photon and light-sheet microscopy, histology, mouse

Annapaola Lippolis

- Degree: Biotechnology for Neuroscience
- Nationality: Italian
- Birthdate: 03/06/2000
- Gender: Female
- Phone: +39 011 6706632
- Email: annapaola.lippolis@unito.it
- Position: PhD Student (41st Cycle)
- Role & Expertise: Histology, confocal microscopy, light-sheet microscopy, mouse models.

Demetra Avincola

- Degree: Neurobiology
- Nationality: Italian
- Birthdate: 07/11/1994
- Gender: Female
- Phone: +39 011 6706632
- Email: demetra.avincola@unito.it
- Position: PhD Student (41st Cycle)
- Role & Expertise: Optogenetics, chemogenetics, fiber photometry, RNA-seq, immunohistochemistry, genotyping, confocal microscopy, Thunder microscopy and histology.

Alessandro Zanone
- Degree: Biotechnology for Neuroscience
- Nationality: Italian
- Birthdate: 15/07/2000
- Gender: Male
- Phone: +39 011 6706632
- Email: alessandro.zanone@edu.unito.it
- Position: Research Fellow
- Role & Expertise: Cellular and molecular analyses of immature neurons in mammals.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
October 2023 - February 2026	Characterization and modulation of "immature" neurons: a potentially exploitable reservoir of non-newly generated cells involved in plasticity of the adult rodent and human cerebral cortex	Luca Bonfanti	PRIN2022	Coordinator	65,000 Euro	UNITO
November 2023-February 2026	Nr2f1-dependent regulation of Mitochondrial Function in Neural Development and Disease	Silvia De Marchis	PRIN 2022	Coordinator	71,466 Euro	UNITO
December 2023-February 2026	The role of miR-211 in neuronal aging: From Disease Mechanisms to Therapy	Silvia De Marchis	PNRR-PRIN 2022	PI	79,685 Euro	UNITO
November 2023-February 2026	Discovering the Effectors of Lifestyle-driven Memory enhancement via Inflammation	Serena Bovetti	PRIN-PNRR 2022	PI	81.000 euro	UNITO
Sept2024-Jan2027	Malattie del neurosviluppo: studio dei meccanismi	Sara Bonzano	Fondazione CRT (Bando Erogazioni	Coordinator	30.000 Euro	UNITO

	patogenetici e individuazione di nuovi potenziali target terapeutici		Ordinarie 2024 - I			
Undetermined	Neuroni alternativi	Luca Bonfanti	Donazioni	Coordinator	25.000 Euro	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Luca Bonfanti, Full professor (PI)

Supervised PhD students	Alessia Pattaro (third year)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Sebastien Couillard-Despres, University of Salzburg, Austria; Chet C. Sherwood, George Washington University, USA Juan Nacher, University of Valencia, Spain;
● Invited talks:	- *Stem cell-dependent and stem cell-independent neurogenesis in widely different mammalian brains.* Talk at Stem Cell Research Italy Meeting 2025, L'Aquila (11-13 settembre 2025) - *Comparative neuroplasticity: evolutionary and translational implications* – Neural Regeneration Research webinar (September 26th, 2025) - *Trade-offs in brain structural plasticity of widely different mammals* – Seminario nella Laurea Magistrale in Neurobiology, Pavia (26 novembre 2025) - *Stem cell-dependent and stem cell-independent neurogenesis: new insights in comparative neuroplasticity* – Seminario online per il National Center for Brain Research (CNCC) of the Romanian Academy (27 novembre 2025)
● Science communication:	- *Dagli studi sul cervello alle ricadute sull'apprendimento* Intervento di apertura al meeting ANP (Associazione Nazionale Dipendenti pubblici e Alte professionalità nella Scuola); Riccione (26-27 Febbraio 2025) - *La scienza nell'Enciclopedia italiana* – Evento organizzato da Treccani all'istituto Manara di Roma, a cura di Luca Bonfanti (9 maggio 2025) - *Neuroplasticità, i superpoteri del cervello* – Conferenza alla Fondazione Ferrero in margine *alla "Healthy Aging Week" Alba (13 novembre 2025)* - *Neuroni immaturi: la plasticità neuronale e le mille risorse del cervello* - Seminario online per AizoOn (24 novembre 2025)
● Editorial duties:	- Editor in chief *Frontiers in Neurogenesis* - Reviewing editor in *Frontiers in Mammal Science* - Joined the Editorial Board of *Regenerative Medicine Reports* (Wolters Kluwer)
● others	*Membership in the Cost action LIFT* (Lifting Farm Animal Lives – laying the foundations for positive animal welfare - CA21124) - *Membership in the Cost action AFFECT-EVO* (An Evolutionary View to Understanding Affective States across Species - CA23106) - *Membro del Comitato scientifico di I.C.S.E.M.* (International Center for Studies on Educational Methodologies)

Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Paolo Marcello Peretto, Full professor (PI)

Supervised PhD students:	Flavia Maria Domizia Di Fiore (co-tutor with S. Bovetti)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Dustin Penn (Konrad Lorenz Institute of Ethology, Veterinary Medicine University, Vienna); Paolo Giacobini (Inserm, UDSL, School of Medicine, Lille, France).
● Invited talks:	
● Science communication:	I Vertebrati: "*Un Giorno all'Università*" 2024-2025; 28 febbraio 2025
● Editorial duties:	Associate Editor Frontiers in Neuroscience - Referee for Scientific Journals
● others	70° Convegno GEI-SIBSC 2025 MODENA- Neuro I, chair "Neurodevelopment" 21st National Congress SINS 2025 PISA- Chair Symposium Th.2.F
Organizational activities and responsibilities at NICO:	Representative of the personnel for safety
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	SINS PISA 2025- Symposium Th.2.F, Navigating the Dynamic world of olfactory processing
Technology transfer achievements (patents, etc.):	

Silvia De Marchis, Associate professor, Lead researcher

Supervised PhD students:	Eleonora Dallorto - Ilaria Ghia (co-tutor with S. Bovetti) - Annapaola Lippolis (co-tutor with S. Bonzano)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Michèle Studer, INSERM U636, Nice Sophia Antipolis, France; Wojciech Krezel INSERM, IGBMC, Strasbourg, France. Christian Schaaf, Institute for Human Genetics, Heidelberg, Germany
● Invited talks:	"*Exploring Pathogenic Mechanisms of BBSOAS Neurodevelopmental Disorder*" NR2F1 Foundation - 2nd Strategic Research Meeting. Morristown, New Jersey (USA) October 16th , 2025
● Science communication:	"*BBSOAS et Plasticité Neurale une Piste pour Comprendre la Maladie*" 3ème journée de rencontre de l'association NR2F1 France entre les familles, les chercheurs et les cliniciens autour du BBSOAS - 5 Décembre 2025 - Paris - France
● Editorial duties:	Referee for Neurobiology of Disease; Cell Reports

• others	-Member of the Scientific Advisory Board of the Jérôme Lejeune Fondation - Referee for grants. -Referee for the Deutsche Forschungsgemeinschaft (DFG) German Research Foundation -Member of the PhD THESIS committee, dott. Valentina Scarpetta - June 2025 - University of Turin
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	Coordinator of the teaching committee of the PhD program in Neuroscience
Workshops, Schools or Conferences organized:	Organizer and chair of the Symposium "*Mitochondria in Brain Physiology and Pathology: Mechanistic Insights and Clinical Implications*" NeuroFrance 2025 Montpellier, France, May 14th-16th, 2025
Technology transfer achievements (patents, etc.):	

Serena Bovetti, Associate professor, Lead researcher

Supervised PhD students:	Ilaria Ghia (co-tutor with Silvia De Marchis) Flavia Maria Domizia Di Fiore (co-tutor with Paolo Peretto) Demetra Avincola (co-tutor with Seung Hee Lee)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	Paolo Giacobini, Lille, France Dustin Penn, Konrad Lorenz Institute, Vienna (Austria) Sylvain Gigan, ENS, Paris (France) Cathie Ventalon, ENS, Paris (France) Bianca Silva, IPMC – CNRS-UCA UMR7275, Sophia-Antipolis, Nice (France)
• Invited talks:	-"*Olfactory dopaminergic cells: key players in sexual odor recognition*" 21st National Congress SINS 2025 PISA -"*Olfactory dopaminergic cells: key players in sexual odor perception*" ECRO Bilbao (Spain)
• Science communication:	Relatrice all'evento LE VOCI DI FUORI Te lo dico cantando. Teatro Vittoria - 27 settembre 2025, SuperSonix Science Festival
• Editorial duties:	- Reviewing editor for *Frontiers in Neural Circuits, Molecular Neurobiology, European Journal of Neuroscience*
• others	External PhD Advisor for: Marta Ribodio; Anita Romiti, Emma Merlin
Organizational activities and responsibilities at NICO:	Responsible of the two-photon microscope Responsible of the BSL2 surgical room
Speakers invited:	- Dr. Cornelius Gross EMBL, Monterotodo, Rome) - Dr Salaman Zubedat (Spatial Perception and Memory Lab, Department of Neuroscience, The Rappaport Faculty of Medicine, Technion-Israel Institute of Technology, Israel)
Other organizational activities:	
Workshops, Schools or Conferences organized:	Organizer and Chair of the Symposium: Navigating the Dynamic World of Olfactory Processing. 21st National Congress SINS 2025 PISA Organizer and Chair of the Symposium: Plasticity and modulation in the bulbar glomerular layer. ECRO Bilbao (Spain)

Technology transfer achievements (patents, etc.):	

Federico Luzzati, Associate professor, Lead researcher

Supervised PhD students:	
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Benedikt Berninger, King's College London, UK; Philip Greulich, University of Southampton, UK Matteo Bergami, University of Cologne, Germany
● Invited talks:	-15th October 2025. Centre for Developmental Neurobiology &MRC Centre for Neurodevelopmental Disorders, **King's College London.** Title: *Brain injury reactivates a developmental program driving generation and integration of transient LGE-class interneurons* -**SINS 2025** Congress, Pisa 10th-13th/09/2025 Title: *Astrocytes Generated Lge-Class Interneurons Transiently Integrate in the Lesioned Striatum.*
● Science communication:	
● Editorial duties:	Referee for international journals
● others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	-Prof. Paolo Malatesta, University of Genoa and IRCCS Policlinico San Martino, Genoa -Prof. Benedikt Berninger, King's College London
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Sara Bonzano – Assistant professor RTD-A

Supervised PhD students:	Annapaola Lippolis (co-tutor with Silvia De Marchis)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	M. Studer (iBV, UCA, CNRS, INSERM - Nice, France); K. Wojciech, Q. Brassart (INSERM, IGBMC - Strasbourg, France)
● Invited talks:	
● Science communication:	Storie di ricerca dall'Università di Torino -Dissemination article on the Frida website - May 2025 (italian) *https://frida.unito.it/come-il-cervello-perde-la-sua-plasticita-tra-geni-e-malattie-rare*
● Editorial duties:	Referee for: Nature Methods, Clinical case reports
● others	**Poster presentation**

	SINS, Pisa - September 2025: "*Niche-specific control of Adult Neural Stem/Progenitor cell proliferation by NR2F1*" Bonzano S, Iacono F, Hamieh K, Quaresima S, Lippolis A, Dallorto E, Cacci E, Peretto P, Licursi V, Lupo G, De Marchis S **Membership of Scientific Societies:** SINS, BraYn
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	Local Organizer Committee: BraYn Conference 2025, Turin, Italy
Technology transfer achievements (patents, etc.):	

Stefano Zucca, Assegnista di Ricerca (postdoc)

Supervised PhD students:	
Outreach activities	
● International collaborations:	Dustin Penn (Konrad Lorenz Institute of Ethology, Veterinary Medicine University, Vienna); PSylvain Gigan (Laboratoire Kastler-Brossel Sorbonne Université, Paris); Paolo Giacobini (Inserm, UDSL, School of Medicine, Lille, France).
● Invited talks:	- GEI-SIBSC Conference *"Prefrontal neurons encode multimodal courtship cues"* - SINS 2025 Conference *"Multisensory integration of social cues in the ventromedial prefrontal cortex"* - Neuroclub Talk - Ospedale San Raffaele *"Multisensory integration of social cues in the ventromedial prefrontal cortex"*
Science communication:	-UNIGHT – Notte Europea delle Ricercatrici e dei Ricercatori 2025 – *"I cinque sensi dell'amore: come il cervello ci guida nella scelta del partner"* -"Science is Wonderful 2025-2026" - MSCA Action for science communication
● Editorial duties:	
● others	Poster Presentation: BraYn 2025 Conference, Turin. *"Multisensory integration of social cues in the ventromedial prefrontal cortex"*
Organizational activities and responsibilities at NICO:	Responsible for the in vivo electrophysiology setup Support and Well-being Committee
Speakers invited:	- Dr. Katja Rehinard - SISSA. *"The neuronal basis of context-dependent behaviour adaptation"*
Other organizational activities:	
Workshops, Schools or Conferences organized:	Local Organizer Committee: BraYn Conference 2025, Turin, Italy.
Technology transfer achievements (patents, etc.):	

Marco Fogli, Assegnista di Ricerca (postdoc)

Supervised PhD students:	
Honors, prizes, awards:	On January 30th 2025, I defended my PhD Thesis, thus completing my PhD in Neuroscience (University of Turin)
Outreach activities	

● International collaborations:	Philip Greulich (University of Southampton)
● Invited talks:	
● Science communication:	
● Editorial duties:	
● others	**Poster presentations:** - *"The big niche: Striatal and SVZ astrocytes share a widespread neurogenic potential and commitment towards LGE-class interneurons".* M. Fogli, G. Nato, P. Greulich, J. Pinto, M. Ribodino, G. Valsania, V. Proserpio, S. Oliviero, B. Berninger, P. Peretto, A. Buffo and F. Luzzati. "ISSCR Neural Stem Cells Symposium 2025". Athens (Greece), 3-4 April 2025. - *"Dynamic spatiotemporal activation of a pervasive neurogenic competence in striatal astrocytes supports continuous neurogenesis following injury".* M. Fogli, G. Nato, P. Greulich, J. Pinto, M. Ribodino, G. Valsania, V. Proserpio, S. Oliviero, B. Berninger, P. Peretto, A. Buffo and F. Luzzati. "Engineering Stem Cells and Stem Cell Niches", NovoNordisk Science Cluster Conference, Hillerod (Denmark), 23-27 November 2025 **Membership of Scientific Societies:** International Society of Stem Cell Research (FENS)
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Ilaria Ghia, PhD Student

Supervised PhD students:	
Honors, prizes, awards:	- Rank 11-20 Acknowledged Poster Contributor at the 21st SINS National Congress and PhD Meeting for the first-author poster entitled *"A Cre-lox technique to address olfactory dopaminergic cell functional heterogeneity"*. - 3rd Prize Award at the science communication competition "My 3-min PhD Thesis" 2025 Edition by UNITA - Universitas Montium. Talk title: *"What Does Love Smell Like? Studying Dopamine in the Mouse Olfactory Bulb to Understand How Sexual Odors Are Processed in the Brain"*.
Outreach activities	
● International collaborations:	
● Invited talks:	
● Science communication:	- U-NIGHT Notte Europea delle ricercatrici e dei ricercatori 2025. Talk: *"Che odore ha l'amore?* Il contest "My 3-Minute PhD Thesis" di comunicazione della ricerca per le Università di UNITA". - U-NIGHT Notte Europea delle ricercatrici e dei ricercatori 2025. Laboratorio della ricerca: "I cinque sensi dell'amore: come il cervello ci aiuta a scegliere". - "My 3-min PhD Thesis". UNITA - Universitas Montium - Science communication competition for PhD candidates. - Public engagement events *"Ricercatori alla Spina"* (participant and organizer).

● Editorial duties:	na
● others	**Poster presentations:** **I. Ghia**, S. Zucca, M. Pieroni, S. Bonzano, G. Demenego, S. Lodato, P. Peretto, S. De Marchis, S.Bovetti. *A Cre-lox technique to address olfactory dopaminergic cell functional heterogeneity*. 21st SINS National Congress and PhD Meeting and ECRO 2025 and 8th BraYn 2025 **Oral presentations:** *Addressing the role of the olfactory dopaminergic population in processing ethologically relevant odors by using a multidisciplinary approach*. NICO NeuroWebinar **Member of Scientific societies**: FENS, ECRO, BraYn, FNOB
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Eleonora Dallorto, PhD student

Supervised PhD students:	
Honors, prizes, awards:	-Best Poster Presentation Award at the 21st SINS National PhD Meeting, 10th September 2025 - Travel Grant IBRO Award from the French Neuroscience Society - IBRO, which supported the attendance at NeuroFrance 2025 congress in Montpellier (France) and included the opportunity to visit two French research laboratories: Giovanni Marsicano's lab at the University of Bordeaux, and Mounia Chami's lab at IPMC-.Sophia Antipolis (Valbonne)
Outreach activities	
● International collaborations:	Michèle Studer (iBV, UCA, CNRS, INSERM - Nice, France) Christian Schaaf, Institute for Human Genetics, Heidelberg, Germany
● Invited talks:	- Selected oral presentation in the symposium *"Mitochondria in Brain Physiology and Pathology: Mechanistic Insights and Clinical Implications"* at NeuroFrance Meeting 2025 - Montpellier (France), May 14th-16th: Dallorto E., Bonzano S., Bertacchi M., Maharaux G., Studer M., De Marchis S. "*Mitochondrial regulation in the physiopathology of the neurodevelopmental disorder BBSOAS: insights from the adult mouse brain and human iPSC-derived neurons*" - w/Prof. Silvia De Marchis "*Exploring Pathogenic Mechanisms of BBSOAS Neurodevelopmental Disorder*" NR2F1 Foundation - 2nd Strategic Research Meeting. Morristown, New Jersey (USA) October 16th , 2025
● Science communication:	U-NIGHT Notte Europea delle ricercatrici e dei ricercatori 2025. Laboratorio della ricerca: *"I cinque sensi dell'amore: come il cervello ci aiuta a scegliere"*
● Editorial duties:	
● others	**Oral presentations:** iBV talks - internal seminars (CNRS, INSERM - Nice, France)

	Mitochondrial regulation in the physiopathology of the neurodevelopmental disorder BBSOAS ERA-NET NEURON strategic meeting (iBV-CNRS, INSERM - Nice, France) *Mitochondrial regulation in the physiopathology of the neurodevelopmental disorder BBSOAS* **Poster presentations:** - Dallorto E., Bonzano S., Bertacchi M., Maharaux G., Studer M., De Marchis S., 21st SINS National Meeting - Pisa (Italy), September 10th - 13th "*NR2F1 deficiency Impairs Mitochondria in Human iPSC-derived neurons*" - Dallorto E., Bonzano S., Bertacchi M.,Studer M., De Marchis S. 8th BRAYN national meeting - Torino (Italy), 29-31 October 2025 "*Loss of NR2F1 Impairs Mitochondrial Function in Mouse and Human Models of the Neurodevelopmental Disorder BBSOAS*"
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	PhD student representative of the PhD program in Neuroscience
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Alessia Pattaro, PhD student and research fellow

Supervised PhD students:	
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Chet C. Sherwood, George Washington University, USA
● Invited talks:	
● Science communication:	STEM evening at Wayside Elementary School on primate brains, Maryland (USA)
● Editorial duties:	
● others	Poster presentation: - Alessia Pattaro, Marco Ghibaudi, Chiara Corrente, Madison Hillegas, Madeline Bramel, Chet C. Sherwood, Luca Bonfanti "Cortical immature neuron density in mammals covaries with brain size and neocortical expansion: evidence from dog, horse, and macaque". 11th European Conference of Comparative Neurobiology (ECCN). May 2025, Alicante (Spain) - Alessia Pattaro, Marco Ghibaudi, Chet C. Sherwood, Luca Bonfanti "Cortical immature neuron density in macaques (Rhesus monkey) and its place in the phylogenetic variation". 8th brainstorming research assembly for young neuroscientists (BraYn). October 2025, Torino (Italy) - Alessia Pattaro, Chet C. Sherwood, Luca Bonfanti "Distribution, density, and phylogenetic variation of cortical immature neurons in macaques (Rhesus monkey)" Gruppo Italiano per lo studio della neuromorfologia (GISN). November 2025, Torino (Italy)
Organizational activities and responsibilities at NICO:	
Speakers invited:	

Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Flavia M. D.Fiore, PhD student

Supervised PhD students:	
Honors, prizes, awards:	- Best poster presentation award at 8th BraYn Congress 2025. *October 29th-31st, Turin (Italy)* - Travel Grant *by ECRO Grant Committe at ECRO Congress 2025 -September 15th-18th 2025, Bilbao (Spain).*
Outreach activities	
● International collaborations:	Dustin Penn, Sarah Zala (Konrad Lorenz Institute of Ethology, Veterinary Medicine University, Vienna
● Invited talks:	
● Science communication:	U-NIGHT Notte Europea delle ricercatrici e dei ricercatori 2025. Laboratorio della ricerca: "I cinque sensi dell'amore: come il cervello ci aiuta a scegliere"
● Editorial duties:	
● others	**Poster presentations:** Flavia M.D. Di Fiore, A. Stella, Stefano Zucca, Matteo Sartini, Sarah M. Zala, Silvia de Marchis, Dustin J. Penn, Paolo M. Peretto, Serena Bovetti. Whole-Brain encoding of opposite-sex odors in Mice.21st **SINS** National Congress and PhD Meeting. September 10th-13th, Pisa (Italy). **ECRO** 2025. September 15th-18th, Bilbao (Spain). 8th **BraYn** 2025. October 29th-31st, Turin (Italy). Zucca Stefano, Di Fiore Flavia Maria Domizia, Stella Alessandra, Mina Veronica, Peretto Paolo Marcello, Bovetti Serena. Multisensory Integration of Social Cues in the Ventromedial Prefrontal Cortex. 21st **SINS** National Congress. September 10th-13th, Pisa (Italy). **Member of Scientific societies**: SINS, ECRO, BraYn
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Annapaola Lippolis, PhD student

Supervised PhD students:	
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	
● Invited talks:	

• Science communication:	
• Editorial duties:	
• others	**Poster presentations**: **- Lippolis A.**, Belhassen I., Dallorto E., Studer M, De Marchis S., Bonzano S., "*NR2F1 Haploinsufficiency Impairs Hippocampal Oligodendrocyte lineage in BBSOAS mouse models*", 21st SINS National Congress (September 2025 - Pisa, Italy) and 8th BraYn 2025 (October 2025 - Turin, Italy) - Bonzano S., Iacono F., Hamieh K., Quaresima S., **Lippolis A**., Dallorto E., Cacci E., Peretto P., Licursi V., Lupo G., De Marchis S., "*Niche-specific control of Adult Neural Stem/Progenitor cell proliferation by NR2F1*", 21st SINS National Congress (September 2025 - Pisa, Italy) **Member of Scientific societies**: SINS, BraYn

Demetra Avincola, PdD Student

Supervised PhD students:	
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	SeungHee Lee (IBS, South Korea)
• Invited talks	
• Science communication	
• Editorial duties:	
• others	

Alessandro Zanone, Research fellllow

wSupervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
• International collaborations:	Chet C. Sherwood, George Washington University, USA
• Invited talks:	na
• Science communication:	na
• Editorial duties:	na
• others	Poster presentation: - Authors: Alessandro Zanone, Alessia Pattaro, Matteo Chincarini, Marco Ghibaudi, Michela Minero, Bruno Cozzi and Luca Bonfanti "Modulation of immature neurons in the neocortical layer II of sheep kept in different environmental conditions" Gruppo Italiano per lo studio della neuromorfologia (GISN). November 2025, Torino (Italy)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

4. Research activity in 2025

a. Summary

Different aspects of postnatal/adult brain plasticity in health and pathology were addressed:

i) the role of olfactory and acoustic cues in the shaping of neural circuits for sexual imprinting
ii) the existence of *"immature"* neurons in adult mammals, and their *trade-off* from small-brained to large-brained species
iii) the mechanisms underlying neuronal changes in the hippocampal neurogenic niche across different pathological mouse models
iv) the mechanisms and dynamics of lesion-induced acquisition of a neurogenic competence in striatal astrocytes.

b. Background and rationale

The brain's ability to adapt its organization and function is driven by environmental cues and achieved through complex cellular and molecular mechanisms that primarily occur during the postnatal critical periods. Plasticity processes continue to some extent in the mature brain involving the generation of new neurons, both in neurogenic niches and in the parenchyma, but also the persistence of neurons in an immature state. However, several questions remain open regarding the early/postnatal sensory-driven shaping of cerebral circuits, as well as the existence and role of newly generated and/or immature neurons in various mammals, including humans, under both physiological and pathological conditions.

In this complex picture, some pivotal questions are:

1) How and when the olfactory and acoustic cues shape neural brain circuits underlying reproduction? It is well accepted that early exposure to such stimuli heavily influences the reproductive behavior of females. However, no data are still available about when this process occurs during postnatal development, which circuits integrate these cues.

2) How different types of plasticity (AN versus "immature" neurons) are phylogenetically distributed among mammals? How the immature neurons behave at different ages? These questions arise by recent studies revealing conflicting results and interpretations on the existence and function of AN in the human brain, and unveiling new/alternative types of structural plasticity (i.e., neurogenesis without division).

3) How widespread is the neurogenic potential of the mature brain parenchyma, what is the fate of adult neural stem cells (NSCs) and how is regulated the identity and function of adult born neurons in physiological or pathological conditions?

The rationale of the research carried out in 2025 can be summarized as follows:

1. In many animal species, the ability to establish memories of relatives during infancy is fundamental for several vital behaviours. These memories involve different type of sensory cues, including olfactory and auditory signals whose processing is integrated into brain circuits. We previously identified several brain areas involved in this behaviour, including olfactory, hypothalamic and limbic brain areas. By focusing on some of these identified areas we performed both in vivo recording and manipulation to causally assess and test the role in uni- and multi-modal olfactory and auditory processing.

2. Based on a previous work showing that "immature" neurons are heterogeneously distributed in the mammalian brain (Piumatti et al., 2018, J Neurosci; La Rosa et al., 2020, eLife) we established a method to quantify in a comparable manner the amount of immature neurons in the subcortical regions of 10 mammals, including small-brained and large-brained species.

3.1. The neurogenic niche of the hippocampal dentate gyrus (DG) is one main site of postnatal neuroplasticity in which neurogenesis persists throughout life. Adult-born neurons play different physiological roles in learning, memory, pattern separation, and cognitive flexibility, which are altered in neuropathological conditions. Understanding the mechanisms underlying the alterations in adult

neurogenesis in animal models of neurological disorders is key in identifying the cellular/molecular targets, a prerequisite neededto develop new interventions aimed at improving cognitive functions.

3.2 We demonstrated that striatal astrocytes possess a widespread neurogenic potential and, in response to injury, actively generate neurons through spatiotemporal dynamics that closely mirror those of astroglial cells in canonical neurogenic niches across evolution. Despite this huge and highly conserved neurogenic potential, unlike in other vertebrate species the lesioned striatum do not regenerate, prompting the question as the cell fate potential of the striatal astrocytes. To resolve this issue over the last years we started an in-depth morphological, electrophysiological, transcriptional and connectivity analysis of the identity and integration capacity of these cells. Last year we completed these analyses, particularly concerning the transcriptional signature of the newly generated neurons.

c. Objectives

i) Examine the brain regions involved in the formation of sexual imprinting with specific focus on multimodal olfactory and acoustic integration.
ii) Investigate whether subcortical "immature" neurons are heterogeneous across different mammalian species and potentially more prevalent in large-brained species. Additionally, the study aimed to begin investigating cortical immature neurons in human fetal brains.
iii) Explore animal models of the neurodevelopmental disorder BBSOAS and of neuroinflammation.
iv) Determine the dynamics and mechanisms of striatal astrocytes neurogenic potential and analyze the identity and integration capacity of their neuronal progeny.

d. Results

i) *Neural network involved in sexual imprinting.* Using iDISCO tissue clearing combined with light-sheet fluorescence microscopy, we previously performed whole-brain imaging of cFos-stained neurons in female mice acutely exposed to odors, ultrasonic vocalizations (USVs), or their combination, presented from familiar or unfamiliar males. This approach allowed us to identify brain regions selectively activated under specific sensory conditions. To causally test their involvement in uni- and multimodal sensory integration, we employed in vivo imaging, along with GCaMP7 expression. Among the recruited regions, the infralimbic cortex displayed a distinct activation pattern in response to multimodal olfactory–auditory stimuli. Inhibition of this area abolished the animals' preference for multimodal sources, indicating a functional contribution to cross-modal integration. Regarding olfactory structures, we focused on the posterior dorsal olfactory bulb, where we observed selective activation of the dopaminergic population. After developing a method to specifically target this olfactory subpopulation, we are now conducting behavioral assays to assess its role in shaping opposite-sex odor preference.

ii) *Immature neurons*. In the adult mammalian brain, mainly composed of mature neurons, a limited amount of stem cell-driven neurogenesis can persist in postnatal life but is reduced in large-brained species. A population of immature, "dormant" neurons in the cortical layer II retains developmentally undifferentiated states in adulthood. We showed that in large brain mammals, in spite of well-preserved morphological and molecular features, the distribution of cortical immature neurons was highly heterogeneous, particularly abundant in the neocortex. We studied the amount of cortical immature neurons in the murine piriform cortex at different ages, showing a remarkable reduction from young to old stages, while such a decrease was far less evident in gyrencephalic species. Nevertheless, even in old rodents, the small population of immature neurons do awake and fully mature as new functional neurons. These findings suggest an evolutionary developmental mechanism for plasticity in large brains, granting a reservoir of young cells for the cerebral cortex.

iii) *Adult DG neurogenesis.* We explored the link between mitochondrial dysfunction, adult hippocampal neurogenesis and genes associated with cognitive deficits in neurodevelopmental disorders. We provided insights into how alterations in the transcriptional regulator NR2F1 may contribute to the pathophysiology of the emerging neurodevelopmental disorder Bosch-Boonstra-Schaaf optic atrophy syndrome (BBSOAS OMIM# 615722; ORPHA 401777). We investigated the impact of Nr2f1

haploinsufficiency associated with BBSOAS by constitutive heterozygous knockout mice (Nr2f1+/-). Nr2f1+/- mice exhibited impaired short-term spatial memory, reflecting a disruption of hippocampal dentate gyrus (DG)-dependent processes such as fine spatial discrimination and pattern separation. This impairment was accompanied by an increased number of activated mature DG granule cells (GCs) and reduced GABAergic inhibitory input, pointing to a dysfunctional inhibitory control onto excitatory GCs. Moreover, GABAergic terminals exhibited enhanced asynchronous release and prolonged tonic inhibition, together with an enrichment in GABAA receptors at extrasynaptic sites.

iv) *Lesion induced striatal neurogenesis*. Our results indicate that despite a transient life newly generated neurons mature and integrate into pre-existing circuits and acquire complex morphology and electrophysiological properties. Single-cell RNA-seq mapping onto an embryonic reference revealed that these cells are not related to adult striatal neuron types but instead belong to the LGE-MEIS2/PAX6 interneuron class. Notch abrogation, which mimics neurogenic activation, drives both cortical and striatal astrocytes toward this same interneuron class, revealing a shared intrinsic commitment. Surprisingly, in mice these cells are widely distributed throughout the telencephalon during embryonic and postnatal development.

e. Advancement in the field

i) *Neural network involved in sexual imprinting*: Our interdisciplinary approach, combining tissue clearing, advanced imaging techniques, and network analysis allowed to identify the brain circuits recruited by exposure to male olfactory cues in laboratory and wild mice populations.

ii) *Immature neurons:* the study of "non-newly generated, immature" neurons is revealing that these cells might represent a reservoir of "young" neurons for the (non-neurogenic) cerebral cortex of large-brained mammals.

iii) *Adult DG neurogenesis.* We demonstrate that Nr2f1 haploinsufficiency disrupts the finely tuned inhibitory control required for DG-dependent functions, highlighting inhibitory network integrity as a potential therapeutic target in BBSOAS. *Lesion induced striatal neurogenesis*: Our results indicate that unlike other vertebrates in which adult telencephalic astroglia preserve the potential to generate constitutive region-specific neurons, the homologous cells in mammals converge on the generation of a specific transient neuron class, possibly representing a reservoir for circuit plasticity in adult life.

f. Publications

- Ghibaudi M, Zanone A, Bonfanti L (2025) Brain structural plasticity in large-brained mammals: not only narrowing roads. Neural Reg. Res. 21(5):1669-1680. Review Q1
- Pattaro A. Ghibaudi, M., Corrente, C., Telitsyn, N, Graic, J.-M., Aresu, Sherwood CC, Bonfanti L. (2025) Phylogenetic variation of layer II cortical immature neurons in dog and horse confirms covariance with brain size and neocortical surface. Brain Struct. Funct. 230(6):115. Research article Q1
- Ghibaudi M, La Rosa C, Telitsyn N, Graïc J-M, Faulkes CG, Sherwood CC, Bonfanti L. (2025) Multispecies characterization of immature neurons in the mammalian amygdala reveals their expansion in primates. PLoS Biol. 23(8):e3003322. Research article Q1
- Bonato M, Montarolo F, Parolisi R, Caudana M, Abbadessa G, Cardinale V, Nato G, Pandino C, Di Cintio N, Schiavo V, Giustetto M, Luzzati F, De Francia S, Bertolotto A, Buffo A, Boda E. Acute Particulate Matter (PM10) Exposure Selectively Triggers Behavioral Alterations in the Presymptomatic Experimental Autoimmune Encephalomyelitis (EAE) Mouse Model of Multiple Sclerosis (2025). Eur J Neurosci. Nov; 62(10): e70304. doi: 10.1111/ejn.70304. Research article Q2
- Thanou I, Koutsoudaki PN, Margariti M, Luzzati F, Havaki S, Gorgoulis VG, Thomaidou D.Ectopic recruitment of neuroblasts in striatal myelin bundles and nucleus accumbens following AraC chemical

lesion.(2025). Stem Cell Reports. Sep 9;20(9):102636. doi: 10.1016/j.stemcr.2025.102636. Research article Q1

- Bovetti S, Bonzano S, Luzzati F, Dati C,De Marchis S, Peretto P. (2025). Linking Adult Olfactory Neurogenesis to Social Reproductive Stimuli: Mechanisms and Functions. Int. J. Mol. Sci. 2025, 26(1), 163
  Review Q1
- Endo, Y., Tanaka, M., Uemura, T., Tanimura, K., Desaki, Y., Ozawa, R., Bonzano, S., Maffei, M.E., Shinya, T., Galis, I. and Arimura, G.-i. (2025). Spider mite tetranins elicit different defense responses in different host habitats. Plant J, 121: e70046. https://doi.org/10.1111/tpj.70046 Research article Q1
- van Doan, C.; Bonzano, S.; Maffei, M.E. Transgenerational and Molecular Responses to Lanthanum Exposure in a Spodoptera littoralis-Brassica rapa System. Int. J. Mol. Sci. 2025, 26, 8462. https://doi.org/10.3390/ijms26178462
  Research article Q1
- Scaramuzza L, Ribodino M, Cassarino C, Morrocchi M, Gomez Gonzalez GB, Parolisi R, Sozzi E, Turrini G, Cerrato V, Conforti P, Hoxha E, Tognato R, Galeotti G, Cordiglieri C, Crosti MC, Zucca S, Lorenzati M, Bovetti S, Spaiardi P, de'Sperati C, Biella G, Ottoboni L, Parmar M, Maestri S, Besusso D, Cattaneo E, Buffo A. Transplanted human striatal progenitors exhibit functional integration and modulate host circuitry in a Huntington's disease animal model. Pharmacol Res. 2025 Sep;219:107905. doi: 10.1016/j.phrs.2025.107905. Epub 2025 Aug 10. Erratum in: Pharmacol Res. 2025 Oct;220:107934. doi: 10.1016/j.phrs.2025.107934. PMID: 40796049. Research article Q1
- Zucca S, Schulz A, Gonçalves PJ, Macke JH, Saleem AB, Solomon SG. Visual loom caused by self-movement or object-movement elicits distinct responses in mouse superior colliculus. Curr Biol. 2025 Sep 8;35(17):4241-4250.e4. doi: 10.1016/j.cub.2025.07.013. Research article Q1

5. Future directions and objectives for next years

a. Summary

Our more recent studies have been focused on exploring new and alternative angles of neural plasticity mechanisms underlying brain function/development: a new molecular regulator of mitochondrial function in adult born neurons, the neuroprotective effect of tamoxifen in vivo, the activation of parenchymal astroglia NSC potential after injury, the comparative approach to phylogenetic variation of non-newly generated “immature” neuronal populations in mammals and the role of sensory cues in shaping the organization and function of brain circuits critical for survival. In the next year, we will focus on the same topics through an in-depth analysis of the molecular/cellular mechanisms regulating adult NSC and immature neuron function in both physiological and pathological conditions and a further characterization of the immature neuron “reservoirs” in widely different mammals (including humans) and brain regions. Moreover, we will continue to investigate the organization and function of neural circuits integrating olfactory and acoustic cues responsible for sexual imprinting in female mice.

b. Background and Significance

Studies performed over the last 30 years on unravelling mechanisms driving adult brain organization and function have revealed that brain plasticity plays a key role in shaping neural circuits critical for survival. From one side it has been clearly established the brain organizational importance of several external/environmental (e.g., olfactory, visual, acoustic stimuli) and internal cues (e.g., hormones) acting during the peri- and post-natal critical periods. From the other side, the discovery of adult neurogenesis in mammals, as a further mechanism of adult neural plasticity, has opened interesting perspectives to understand brain function both in physiological and pathological conditions. Nevertheless, several unanswered questions still remain to translate these knowledges into general basic rules of neural organization/function and, possibly, to develop therapeutical strategies.

It is known that early/post-natal exposure to paternal acoustic and olfactory stimuli is critical to shape mate choice in female mice, nevertheless the neural bases of this mechanism are largely unknown. Unravelling this process will increase our knowledge about the shaping of circuits allowing multisensory integration. The research in the field of adult neurogenesis, although it has demonstrated in rodents to be involved in fundamental cognitive functions (e.g., memory and learning), it is also showing that this mechanism can be significantly heterogeneous among mammals and probably limited to the postnatal period in humans. Nevertheless, different aspects are emerging related to "new nuances" or "theme variations" of AN (e.g., the dormant neurogenic potential of the parenchyma) as well as to the discovery of other forms of plasticity related or not to AN that potentially influence, complement or compensate the role of AN in the human brain. This new vision rises new problems, opportunities and questions, such as:

A) how different types of plasticity (AN versus "immature" neurons) are phylogenetically distributed among mammals and in different brain regions?
It is now clear that different forms of brain plasticity, including AN and immature neurons, are differently present/distributed/active in different mammals. To get a picture of such heterogeneity in a high number of mammalian species and orders, including humans (and identify possible phylogenetic trends) is mandatory for correct translation of results and to identify new targets for therapeutic/preventive approaches. During the next years, the analyses will be extended to several brain sub-cortical regions and to human fetal brains
B) how, when and where salient sensory cues are integrated in the brain to sustain behaviors essential for survival (e.g., reproduction)? To this aim we will focus on the identification of neural circuits responsible for sexual imprinting in female mice). It is known that female mice use olfactory and acoustic cues from parents to learn and form memories of conspecifics and close kin, which enable them to avoid heterospecific matings as adults.
This process, called sexual imprinting, has been largely studied in different animal species but little is known about the sensory processing underlying representation of imprinted cues and how they shape brain circuits to drive mate selection.
C) how is regulated the fate of adult neural stem cells (NSCs) and the function of adult born neurons in physiological or pathological conditions? It is of paramount importance to get insight into the mechanisms regulating the neuronal vs. glial switch in different progenitor populations, conditions, and brain regions (physiological and pathological). This is particularly promising, also if/when/where a few (quiescent) progenitors are available (e.g., the adult human brain).

c. General aim and integration with mission of the Institute

Only by knowing the multifaceted roles of AN and other forms of plasticity in brain homeostasis and dysregulation we could expect to use this biological process/related forms of plasticity for translational purposes (novel therapeutic approaches for neurodegenerative diseases and preventive approaches for optimal brain function/plasticity in healthy adults and in aging; both goals ultimately in line with the NICO mission). To understand how the brain adapt to different environmental stimulations during life (from young to old individuals) is fundamental to figure out preventive strategies. In particular, to find and modulate new sources of undifferentiated/young neurons or new ways to drive quiescent (neuronal and glial) progenitors might be pivotal in translating results in large-brained species (e.g., humans) with reduced amount and/or different types of plasticity.

d. Specific objectives and strategies

- *Characterization and quantification of immature neurons in different mammals*. By using the same method employed for cortical immature neurons in 12 mammalian species, molecular, cellular, quantitative analyses will be performed in the amygdala, claustrum and external capsule, namely the subcortical regions in which these cells are expected to be present, especially in gyrencephalic mammals. *Modulation of cortical immature neurons in the sheep neocortex*: 15 brains from young sheep kept in different environmental conditions for 7 weeks (enriched environment, stress (isolation), and control group) will be analyzed for DCX+ neuron quantification, expression of markers of maturity/immaturity, and Sholl analysis. *Search for cortical immature neurons in human fetal brains* Based on an agreement with the Hospital S. Anna in Turin, we are collecting human fetal brains at different gestational stages that will be processed in order to study the

development of immature neurons in the cerebral cortex layer II and establish their total amount shortly before birth. This will give an estimation of the immature neuron reservoir in humans.

- *Neural network involved in sexual imprinting.* By using an interdisciplinary approach that combines tissue clearing, advanced imaging techniques, and network analysis we identified the brain circuits recruited by exposure to male olfactory cues in laboratory and wild mice populations (this latter part has been performed in collaboration with Prof. Dustin Penn, University of Vienna).We are now applying the Miniscope technology to target the regions previously identified, and image in vivo the neural circuits in freely-moving animals (living in the wild) during mating behavior. This part of the project will be performed in collaboration with Dr. Sylvain Gigan (Sorbonne University, Paris, France) third partner of the project and word expert in the development of advanced optical tools.

- *Hippocampal neurogenesis as a model to study BBSOAS* and the causative mechanisms and pathogenesis of intellectual disability. Stemming from our recent data on Nr2f1 function (Bonzanoet al., 2023), we aim to further characterize the mitochondrial phenotype in hippocampal newborn neurons of Nr2f1 mutant mouse models, including models carrying Nr2f1 human mutation, and human iPSCs-derived neurons *in vitro*, exploiting electron microscopy, biochemical and functional analysis as well as electrophysiology by *in vivo*/*ex vivo* approaches. On the other side, we are interested in evaluating whether the dysfunctional inhibitory control onto excitatory GCs in mouse models mimicking human BBSOAS mutations, including DNA Binding Domain-mutated Nr2f1+/R139L and Ligand Binding Domain-mutated Nr2f1+/E397* mice.

-*Mechanisms and role of astrocyte neurogenic activation.* The highly stereotyped spatial and temporal pattern of QA induced neurogenic response will be exploited to analyze at the cellular molecular level the changes in the environment and astrocytes population associated with neurogenic activation. To this aim we will use both bulk, single cell RNAseq and spatial transcriptomics to identify specific genes and cell populations associated with the activation of the neurogenic potential. Particular emphasis will be devoted to identifying pathways associated with the acquisition of primed or competent states, as observed in other stem cell systems. Selected markers of specific cell states will be further analyzed in the tissue through multiplexing immunohistochemistry and 3D reconstructions in combinations with exogenous birth dating markers such as BrdU, EdU or Ki67-CreERT2 lineage tracing. In parallel, we will perform similar analyses, in collaboration with Annalisa Buffo at NICO, in mice in which we will abrogate the expression of the transcription factor SOX2. Preliminary results indicate that mosaic deletion of this TF in astrocytes can abrogate the initiation of the neurogenic response through both cell autonomous and non-cell autonomous factors. This screening will allow us to identify factors involved in the activation of the parenchymal latent neurogenic potential. Additionally, we will verify the role of neuronal activity in the activation of striatal astrocytes neurogenic potential and in the differentiation and survival of the newly generated striatal neurons.

e. Unique features of the project research

In our research group, we address different aspects of brain structural plasticity, ranging from classic adult neurogenesis to "immature" neurons (neurogenesis without division), and including progenitor specification, hormone-linked behavior, lesion-induced repair, and the evolutionary trade-off in plasticity. We employ a combination of basic and innovative technical approaches to study different types of plasticity occurring in various brain regions of different mammalian species, from mice to humans, at the molecular, cellular, and functional levels. We believe that this comparative approach, from molecule to behavior, could broaden our understanding of brain plasticity and help us translate research data from animal models to humans accurately.
One crucial point we aim to address is the identification of mechanisms underlying the neuronal-glial switch in both canonical neurogenic sites and parenchyma to modulate endogenous progenitors. We also investigate "immature" neurons and use imaging technology in wild-living mice, which are novel topics currently addressed by only a few laboratories worldwide.
Furthermore, we are searching for a promising neuronal population that is abundantly present in large-brained mammals with reduced rates of adult neurogenesis, with particular reference to humans. We believe that this approach, in addition to providing new insights into basic neurobiology, will help overcome the current bottleneck of the "classic" adult neurogenesis vision, which is the constitutive, continuous genesis of new neurons in rodents. Instead, we explore the less-traveled roads mentioned above.

f. Methodology

Our research group is developing various innovative technologies to achieve the goals of our projects. We employ in vivo two-photon microscopy with fluorescent cell activity reporters (GCaMP) in head-restrained anesthetized and awake mice at different postnatal ages to study the functional role of neurons in diverse brain circuits (e.g., olfactory bulb and cerebral cortex) after exposure to salient sensory cues such as olfactory and acoustic stimuli. We also use two-photon imaging to investigate mitochondrial dynamics in neurogenic regions.

In collaboration with Dr. S. Gigan from the Laboratoire Kastler-Brossel Sorbonne Université in Paris and Dr. D. Penn from the Konrad Lorenz Institute of Ethology in Vienna, we aim to develop a high-throughput imaging technology based on multimodal optical fibers integrated in a wire-free head-mounted device. We already tested this approach in vivo, and, with further implementation, we aim in recording network activity from multiple brain regions with single-unit resolution, low invasiveness, and in freely-moving animals.

We have developed a customized approach to standardize and automate the production of serial section reconstructions through hierarchical imaging at the confocal microscope. This enables us to obtain 3D high-resolution reconstructions of large volumes. We use block face imaging of the specimen during sectioning as a reference for non-linear registration of the confocally acquired volumes to their original position in the intact brain. We have already used a preliminary version of this method to reconstruct the distribution of GnRH+ cells in the entire brain, the composition of neurogenic niches in the lesioned striatum, the morphology of newly generated neurons, and the distribution of their afferents. This technique may be useful in studying immature neuron populations and their modulation.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Physiopathology of neural stem cells

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Annalisa Buffo
- Degree: PhD
- Birthdate: 25/12/1967
- Phone: +39 011 6706614
- Email: annalisa.buffo@unito.it
- Position: Associate Professor of Physiology
- Role: Principal Investigator

Personnel

Enrica Boda
- Degree: PhD
- Birthdate: 08/05/1981
- Phone: +39 011 6706615
- Email: enrica.boda@unito.it
- Position: Associate Professor of Anatomy
- Role & Expertise: Lead responsible for research on oligodendroglial physiopathology.

Giulia Nato
- Degree: PhD
- Birthdate: 08/05/1986
- Phone: +39 011 6706632
- Email: giulia.nato@unito.it
- Position: Senior PostDoc
- Role & Expertise: Responsible for research on astrocyte neurogenic activation and reactivity.

Valentina Cerrato
- Degree: PhD
- Birthdate: 21/07/1988
- Phone: +39 011 6706615
- Email: valentina.cerrato@unito.it
- Position: Senior PostDoc
- Role & Expertise: Responsible for research on astrocyte heterogeneity and cerebellar development; expert in clonal, single-cell and single-nuclei analyses.

Martina Lorenzati
- Degree: PhD
- Birthdate: 30/10/1992
- Phone: +39 011 6706632
- Email: martina.lorenzati@unito.it
- Position: Junior PostDoc
- Role & Expertise: Expert in oligodendroglia biology, derivation of neural cells from hPSCs, and pathology of human glia.

Fernando Josa Prado
- Degree: PhD
- Birthdate: 17/11/1985
- Email: fernando.josaprado@unito.it
- Position: MSCA Postdoctoral Fellow
- Role & Expertise: Expertise in cellular and molecular biology. SUMOMyePath project – investigating

the role of SUMOylation in myelin biology and its implications in neurodevelopment and demyelinating pathophysiology.

Marta Ribodino
- Degree: MSc in Biotechnology
- Birthdate: 01/06/1996
- Phone: +39 011 6706632
- Email: marta.ribodino@unito.it
- Position: PhD Candidate
- Role & Expertise: Derivation of glia from hPSCs and cell therapy approaches in Huntington's disease models.

Martino Bonato
- Degree: MSc in Molecular Biotechnology
- Birthdate: 09/03/1997
- Phone: +39 011 6706632
- Email: martino.bonato@edu.unito.it
- Position: PhD Candidate
- Role & Expertise: Study of particulate matter effects in Multiple Sclerosis models.

Maryam Khastkhodaei Ardakani
- Degree: MSc in Anatomical Sciences
- Birthdate: 30/06/1991
- Phone: +39 011 6706632
- Email: maryam.khastkhodaeiardakani@unito.it
- Position: PhD Candidate
- Role & Expertise: Pharmacological and cell transplantation approaches to promote recovery in Microcephaly 17 (MCPH17) models.

Giacomo Turrini
- Degree: MSc in Biotechnology for Neuroscience
- Birthdate: 23/06/1998
- Phone: +39 3661885920
- Email: giacomo.turrini@unito.it
- Position: PhD Candidate
- Role & Expertise: Investigation of cerebellar astrocyte heterogeneity through in silico and imaging techniques.

Niccolò Di Cintio
- Degree: MSc in Neurobiology
- Birthdate: 16/11/1992
- Phone: +39 3455998634
- Email: niccolo.dicintio@unito.it
- Position: Research Fellow
- Role & Expertise: Dysmyelination and cognitive impairment.

Olga Teresa Bianciotto
- Degree: MSc in Biotechnology for Neuroscience
- Birthdate: 12/06/1999
- Phone: +39 3662845620
- Email: olgateresa.bianciotto@unito.it
- Position: PhD Candidate
- Role & Expertise: Generation of optimized cerebellar human organoids.

Arianna Contato
- Degree: MSc in Cellular and Molecular Biology
- Birthdate: 24/11/2000
- Phone: +39 3496819252
- Email: arianna.contato@edu.unito.it
- Position: Research Fellow
- Role & Expertise: Generation of integrated cellular models to study demyelination and remyelination.

Beatrice Savarese
- Degree: MSc in Medical Biotechnology
- Birthdate: 27/12/1997
- Phone: +39 3316446447
- Email: beatrice.savarese@unito.it
- Position: PhD Candidate
- Role & Expertise: Stem cell engineering strategies for cell therapy approaches in Huntington's disease models.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
2020-2025	NSC-Reconstruct Novel Strategies for Cell-based Neural Reconstruction #874758	Annalisa Buffo	H2020-SC1-BHC-2018-2020	PI of research unit, WP coordinator	680,000 €	UNITO
2023-2025	In vitro and in vivo molecular mapping of cell therapy for Huntington Disease at single-cell resolution	Annalisa Buffo	PRIN2022, MUR	PI of research unit	98.405 €	UNITO
2024-2026	INITIATE - CreazIone di una BaNca di TessutIfetAli per lo studio del Tessuto nErvoso - INITIATE	Annalisa Buffo	CRT Foundation	PI	50.000 €	UNITO
2024-2025	UNFOLD -A fully humanised integrated cellular system for scalable modeling of inflammatory demyelination	Annalisa Buffo	PNRR MUR – MNESYS_PE00000006_1.	PI	200.000 €	FCO

2025-2031	CUSTOM-MADE NEURONS FOR CELL THERAPY IN PARKINSON'S AND HUNTINGTON'S DISEASE	Annalisa Buffo	ERC Synergy	PI	2.500.000 €	UNITO
2023-2025	Targeting oligodendrocyte dysfunctions to treat cognitive defects and epilepsy in primary autosomal recessive microcephaly-17 (MCPH17) models (ID: GMR22T1066)	Enrica Boda	Telethon Foundation Multiround 21-24 – Round 1 2022 Track Basic	PI	129,500 €	Telethon
2023-2025	Role of interleukin 6 in the pathogenesis of Rett syndrome: focus on astrocyte-neuron crosstalk and its therapeutic implication (ID: P20225Z3J5)	Enrica Boda	PRIN2022-PNRR, MUR	PI of the research unit	100,000 €	UNITO
2024-2026	SUMOMyePath Unveiling the rol of SUMOylatio as novel molecula pathway in myeli biology; implication i neurodevelopmen and demyelinatin pathophysiology	Fernando Josa Prado	MSCA-Postdoctoral Fellowship Grant	Grantee – PI	188,590 €	UNITO

2026-2028	Oligodendrocyte progenitor cell based therapy in primary autosoma recessive microcephaly-17 (MCPH17) preclinical model optimization an mechanisms. (ID GMR25T2067).	Enrica Boda	Telethon Foundation Multiround 2025-2027 Round I	PI	166,000 €	UNITO

3. SCIENTIFIC ACTIVITIES IN 2025

Annalisa Buffo, PI

Supervised PhD students:	Maryam Khastkhodaei (co-supervised with E. Boda) Marta Ribodino, Giacomo Turrini (co-supervised with Valentina Cerrato), Olga Bianciotto, Beatrice Savarese
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	ERC Synergy grant Custom-Made (E Cattaneo, University of Milano, M Parmar, University of Lund; J Emneus, Danish Technical University); M Goetz, LMU, Muenchen; G Quadrato, University of Southern California; L Telley, University of Lion1. See also specific collaborations of lab members below.
• Invited talks:	--
• Science communication:	UNISTEM Day Torino 2025 – organizer https://nico.ottolenghi.unito.it/unistem-day-2025/ Featured subject in PROFESSIONAL DREAMERS, The photographer's eye. The psychologist's gaze. A photographic exhibition by T. & G. Baldizzone and R. Spagnolo, curated by I. Dhargarkar, exploring professional identity at the intersection of science, psychology, and visual storytelling (Museo Nazionale del Risorgimento Italiano, Torino, March–June 2025). Participated in associated public talks and interviews (FINECO, Torino: 25 marzo 2025; MUSEO DEL RISORGIMENTO, Torino: 9 aprile). Festival dell'Innovazione e della Scienza 2025, Settimo Torinese – Speaker – 12 ottobre 2025 Neuroni in provetta: il futuro della medicina rigenerativa Neuroscience LAB, Intesa SanPaolo Innovation Center, July 3rd 2025, On line Lecture - Speaker: Riparare il sistema nervoso: le cellule staminali e il futuro della medicina rigenerativa Key People, The Evolving Leader with Intesa San Paolo with Praxi, October 8th 2025, On line Lecture for an international

	audience - Speaker: Natural Intelligence: the Power of Brain Plasticity IV WOW Congress Italy, November 21st 2025, Torino - Speaker: Come si costruisce un progetto vincente
• Editorial duties:	Member of the Glia Editorial Board Ad hoc reviewer for the following journals in 2025: Glia, Journal of Clinical Investigations, Nature Neuroscience, Nature Communications, Advanced Science, Cerebellum.
• others	Attended meetings: -XVII European Meeting on Glial Cells in Health and Disease, Marseille, July 8–11 2025 - 75° National Congress of the Italian Society of Physiology (SIF), Turin, 17-19 September 2025. - (Brainstorming Research Assembly for Young Scientists) Brayn Conference, Turin, Oct 29-31 2025 Grant evaluator for the UK Research and Innovation, Biotechnology and Biological Sciences Research Council (BBSRC) Grant evaluator for the French National Agency, ANR, AAPG actions Grant evaluator for the European Leukodystrophy Association (ELA) International Evaluating member for tenure-eligible lecturers in Physiology, University of Barcelona, ES Membership in Scientific Societies in 2025: Federation of the European Neuroscience Societies (FENS) Italian Society of Neuroscience (SINS) Brayn association International Society for Stem Cell Research Society For Research On The Cerebellum And Ataxias (SRCA) Italian Glia Network ALBA network Co-supervisor with Dr Angela Gritti of the PhD candidate Garsia Chiara, Course in Molecular Medicine, Curriculum in Gene and Cell Therapy, Vita-Salute San Raffaele University in Milan, Italy. Advisor of the PhD candidate Mattia Battistelli, School in Molecular and Cellular Biology, University of Milan, Italy. External Examiner of the PhD Candidate Angela Genchi, PhD Programme in Molecular Medicine, Curriculum in Neuroscience and Experimental Neurology, Vita-Salute San Raffaele University, Milan, Italy.
Organizational activities and responsibilities at NICO:	- Deputy Director of NICO - Responsible of BLS2 labs at NICO
Speakers invited:	Prof. Aldo Genovesio, Univ Piemonte Orientale November 11 2025

Other organizational activities:	
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Enrica Boda, Lead responsible for research on oligodendroglial physiopathology

Supervised PhD students:	Maryam Khastkhodaei (co-supervised with A. Buffo) Martino Bonato
Honors, prizes, awards:	-
Outreach activities	
• International collaborations:	-
• Invited talks:	Oral Communication: *tDCS promotes myelin repair and plasticity in the mouse motor cortex.* 75th Congress of the Italian Society of Physiology (SIF), Turin, 18 Sept 2025 Oral Communication: *Acute particulate matter (PM_{10}) exposure selectively triggers behavioral alterations in the presymptomatic EAE mouse model of Multiple Sclerosis.* XXXIII Italian Association Neuroimmunology (AINI) Congress, Turin, 07 May 2025
• Science communication:	2 events in the frame of "A tu per tu con la scienza" organized by Diasorin Foundation and Telethon: seminars at Liceo Scientifico Valsalice, Torino and Liceo Scientifico Newton, Chivasso (TO). Participation in "Penne Amiche della Scienza" with the 2C Scuola Media di Attigliano (Terni) e 1C Scuola Media I.C. Bruno Caccia (Torino).
• Editorial duties:	In 2025, ad hoc reviewer for Glia, Disease Models & Mechanisms, European Journal of Neuroscience, Neural Regeneration Research, Frontiers in Molecular Neuroscience, Journal of Neurochemistry, Neurochemistry International, Acta Neuropathologica Communications
• others	Attended meetings: - Telethon Scientific Convention, Rimini, March 17-19 2025 - XXXIII Italian Association Neuroimmunology (AINI) Congress, Turin, 6-8 May 2025 -XVII European Meeting on Glial Cells in Health and Disease, Marseille, July 8–11 2025 - 75° National Congress of the Italian Society of Physiology (SIF), Turin, 17-19 September 2025. - Brayn (Brainstorming Research Assembly for Young Scientists) Conference, Turin, Oct 29-31 2025 Grant Reviewer for the Italian Foundation Multiple Sclerosis (FISM) Membership in Scientific Societies in 2025: Federation of the European Neuroscience Societies (FENS) Italian Society of Neuroscience (SINS) Brayn Association Italian Glia Network

	Vice-President of the BraYn **Association** Co-Founder of the Italian Glia Network (IGN) Advisor of the PhD candidate Giacomo Turrini, PhD program in Neuroscience, Turin, Italy. VIVA committee of Dr. Cecilia Zen, PhD Program in Molecular and Experimental Medicine at Humanitas University, Milan, Italy. PhD Committee member of Dr. Marco Fogli, PhD Program in Neuroscience, University of Turin, Italy.
Organizational activities and responsibilities at NICO:	-
Speakers invited:	-
Other organizational activities:	-
Workshops, Schools or Conferences organized:	Member of the Steering Committee of the BraYn conference (https://www.braynconference.com/) Lead of the Local Committee of the 8th edition of the BraYn conference 29-31 Oct 2025, Turin
Technology transfer achievements (patents, etc.):	-

Valentina Cerrato, senior PostDoc

Supervised PhD students:	Giacomo Turrini (co-supervised with Annalisa Buffo)
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	Prof. Ludovic Telley (University of Lion 1, France); Prof. Magdalena Götz (University of Munich, Germany)
• Invited talks:	"A single cell atlas to unveil the diversity of mouse cerebellar astrocytes: insights into their molecular identities, development, and functions", The 8th cerebellum days of France Cerebellum Club, Rouen, February 6-7, 2025.
• Science communication:	Co-organizer Pint of Science - Beautiful Mind - Turin, May 19th-21st 2025
• Editorial duties:	Review editor for Frontiers in Neuroscience (section Neurogenesis) Ad hoc reviewer for the following journals in 2025: Nature Communications
• others	Co-Founder of the Italian Glia Network (IGN) Elected representative of PostDocs and Residents, Dept of Neuroscience, Univ of Turin Evaluation panel member for the call: Single cell approaches for personalized medicine, Max Delbrück Center for Molecular Medicine in the Helmholtz Association – Focus area together with the Berlin Institute of Health (BIH) 2025, DE Membership in Scientific Societies in 2025: Federation of the European Neuroscience Societies (FENS) Italian Society of Neuroscience (SINS)

	Società Italiana di Fisiologia (SIF) France Cerebellum Club Attended meetings: - The 8th cerebellum days of the France Cerebellum Club, Rouen, February 6-7, 2025. - Telethon Scientific Convention, Rimini, March 17-19 2025 - Neurogenomics Conference, Human Technopole, Milan, May 19-21 2025 -XVII European Meeting on Glial Cells in Health and Disease, Marseille, July 8–11 2025 - 75° National Congress SIF, Turin, 17-19 September 2025. - Brayn Conference, Turin, Oct 29-31 2025 - Second Italian Glia Network Symposium, Nov 26 2025 (online)
Organizational activities and responsibilities at NICO:	- Group administrator for access to the Internet and to computing resources at NICO - Member of the Support and Well-being Committee at NICO
Speakers invited:	--
Other organizational activities:	Co-organizer of Pint of Science - Beautiful Mind Team - Turin, May 19th-21st 2025 Coordinator of the "In vitro and ex vivo approaches to study rodent and human neural cells" Course for the PhD in Neuroscience, Turin, May-June 2025.
Workshops, Schools or Conferences organized:	Co-organizer of the "Second Italian Glia Network Symposium", Nov 26 2025 (online)
Technology transfer achievements (patents, etc.):	--

Giulia Nato, senior PostDoc (until march 2025)

Supervised PhD students:	-
Honors, prizes, awards:	-
Outreach activities	
• International collaborations:	Benedikt Berninger (King's College; London), Prof. Magdalena Götz (University of Munich, Germany);
• Invited talks:	--
• Science communication:	--
• Editorial duties:	--
• others	Membership in Scientific Societies: Italian Society of Neuroscience (SINS) Italian Glia Network ISSCR Member Attended meetings: ISSCR 2025 Athens International Symposium 3-4April 2025 Presented poster: - Astrocyte-generated transient LGE class interneurons functionally integrate into the lesioned striatum. - The big niche: Striatal and SVZ astrocytes share a widespread neurogenic potential and commitment towards LGE-class interneurons
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--

Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Martina Lorenzati, junior PostDoc

Supervised PhD students:	Olga Bianciotto (co-supervised with Annalisa Buffo)
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	Fernando De Castro (Cajal Institute, Madrid, Spain); NSC-Reconstruct network (E Cattaneo, University of Milano); Sabah Mozafari (University of Paris, France); Juan Garcìa Leòn (University of Malaga, Spain).
• Invited talks:	"Stem Cells meet Glial Cells – Insights into the generation of in vitro models of human development and disease", 2nd Italian IPSC User Meeting – Thermofisher Scientific, February 20th, 2025 – Bologna
• Science communication:	Local co-organizer for Pint of Science - Beautiful Mind, Turin, May 19th-21st 2025
• Editorial duties:	-
• others	Membership in Scientific Societies: Italian Glia Network Italian Society of Neuroscience (SINS) Federation of European Societies (FENS) Attended meetings: - Neurogenomics Conference, Human Technopole, Milan, May 19-21 2025 - 75° National Congress SIF, Turin, 17-19 September 2025 - Second Italian Glia Network Symposium, Nov 26 2025 (online) Co-author of posters presented at: IZN/Sorbonne Neuroscience Summer school, Germany, 06-12/07/2025; MNESYS – A multiscale integrated approach to the study of the nervous system in health and disease, Spoke 3 Meeting, Naples, 11/07/2024; Brayn Conference, Turin, 29-31/10/2025. Co-author of poster presented at the XXXV congress GISN 2025, Turin, 28-29 November 2025 *'Overexpression of Anosmin-1 alters the myelination pattern and the morphology of axonal components in cerebellar purkinje cells'*
Organizational activities and responsibilities at NICO:	Co-Responsible of the Dissection Room
Speakers invited:	--
Other organizational activities:	Coordinator of the "In vitro and ex vivo approaches to study rodent and human neural cells" Course for the PhD in Neuroscience, Turin, May-June 2025
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Fernando Josa Prado, MSCA-PF

Supervised PhD students:	--
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	Prof. Dr. Arne Battefeld (Univeristy of Bordeaux, France) Dr. Fernando de Castro Soubriet (Cajal Institute, Madrid, Spain) Dr. Diego Clemente (HNP, Toledo, Spain)
• Invited talks:	--
• Science communication:	--
• Editorial duties:	*Ad hoc* reviewer for: Plos One; Journal of Cellular and Molecular Medicine
• others	• European Commission – REA: Expert - Project evaluator. Special expertise in Researchers Career Development- activated for SWAFS and COFUND calls. • EURAXESS projects advisor and collaborator (Currently Science4Refugees; Past – PIPERS, EURAXIND, TOPIV and TOPV) • Member of the Direction Board member of GADEA Science Foundation (Spain) • Member of Spanish and Italian Chapters of Marie Curie Alumni Association (MCAA) • MCAA representative at COARA's TURIN working group. • Internationalization advisor for the Madrid's Region Government through Madrid Foundation (Spain). • Board Member (and Founder) of the Association for High-Level Management of Research, Innovation and Tech-transfer Institutions (PADIIT, Spain) • Member of the Internation Committee of the Spanish Planetology and Astrobiology Network (REDESPA, Spain) • Founder and formed. President of Scientist Returned to Spain Association (CRE) • Doctoral thesis external evaluator Attended Courses in 2025 • Basics of Data Literacy - An OSCT Practical Workshop (June 2025, OSCT, Turin • Open Science and Evidence-Informed Decision Making – specialist and Instructor. Skills4EOSC (Horizon Europe, Online) Attended Meetings • Brayn Conference, Turin, 29-31/10/2025 Membership in Scientific Societies in 2025: • BraYn Association • Italian Glia network • Spanish Glia Network
Organizational activities and responsibilities at NICO:	--
Speakers invited:	• Brayn Conference, Turin, 29-31/10/2025
Other organizational activities:	•
Workshops, Schools or Conferences organized:	• Collaborator with the Doctoral School of UniTo and Euraxess UniTo (Lucia Salto): • BEYOND ACCADEMIA Seminar. UniTo Doctoral School 17/06/2025

Technology transfer achievements (patents, etc.):	-

Marta Ribodino, PhD candidate

Supervised PhD students:	na
Honors, prizes, awards:	Winner of the "Enrica Marzola" award, recognizing outstanding scientific achievements by PhD students in the Neuroscience program, University of Turin, October 2025
Outreach activities	
● International collaborations:	ERC Synergy grant Custom-Made (E Cattaneo, University of Milano, M Parmar, University of Lund; J Emneus, Danish Technical University) prof. Tomas Bjorklund (Lund University, Sweden)
● Invited talks:	*"Functional integration of human striatal progenitor-grafts in a HD animal model".* DataBlitz at the Annual Symposium of the Network for European CNS Transplantation and Restoration (NECTAR) 2025, Coimbra, Portugal, 20-22 October 2025.
● Science communication:	--
● Editorial duties:	--
● others	Membership in Scientific Societies in 2025: Italian Society of Neuroscience (SINS) ALBA Network Italian Glia network Attended meetings: Italian Society of Physiology (SIF) 2025 Conference, Turin, 17-19 September 2025. *Functional integration and circuit reconstruction by hESC-derived striatal progenitors in the QA-lesion model – poster presentation.*
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	Under-40 Symposium organizer and speaker at the meeting of the Italian Society for Neuroscience (SINS) 2025, Pisa, 10-13 September. Talk title:"*Optimizing Cell-Replacement Approaches to Reconstruct Striatal Circuitries in Huntington's Disease*".
Technology transfer achievements (patents, etc.):	

Martino Bonato, PhD candidate

Supervised PhD students:	na
Honors, prizes, awards:	--
Outreach activities	
● International collaborations:	--
● Invited talks:	"*Unraveling the role of oligodendrocyte progenitor cells in the development of the cortical inhibitory system*". Oral communication at the Italian Society for Neuroscience (SINS) PhD meeting 2025, Pisa, 10 September 2025

	"*Unraveling the role of oligodendrocyte progenitor cells in the development of the cortical inhibitory system*". Oral communication at SINS 2025, Pisa, 10-13 September 2025 "*Short-term exposure to particulate matter triggers selective molecular and behavioral alterations in a mouse model of Multiple Sclerosis*". Speed Talk at BraYn Conference 2025, Torino, 29-31 October 2025
● Science communication:	Pint of Science 2025, Turin, 21 May 2025 "Come respira il nostro cervello? Interazioni tra sclerosi multipla ed inquinamento atmosferico" – scientific communication for public engagement
● Editorial duties:	--
● others	Membership in Scientific Societies in 2025: Italian Society of Neuroscience (SINS) BraYn Association Italian Glia network (IGN) Gruppo Italiano per lo studio della Neuromorfologia (GISN) Attended meetings: BraYn Conference 2025, Turin, 29-31 October 2025 "*Short-term exposure to particulate matter triggers selective molecular and behavioral alterations in a mouse model of Multiple Sclerosis*" – poster presentation XXXV congress GISN 2025, Turin, 28-29 November 2025 "*Short-term exposure to particulate matter triggers selective molecular and behavioral alterations in a mouse model of Multiple Sclerosis*" – poster presentation
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Maryam KhastkhodaeiArdakani, PhD candidate and Research Fellow (until May 30th 2025)

Supervised PhD students:	na
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	--
• Invited talks:	--
• Science communication:	--
• Editorial duties:	--
• others	- Incucyte® Virtual Training: Base Analysis Software - NICO Webinar: Periodically at 2.00 p.m.- https://www.nico.ottolenghi.unito.it/Agenda/NICO-NeuroWebinar Membership in Scientific Societies in 2025: -Society for Neuroscience (SfN)

	- Italian Glia Network (IGN) - ALBA Network Co-author of posters presented at: IZN/Sorbonne Neuroscience Summer school, Germany, 06-12/07/2025; MNESYS – A multiscale integrated approach to the study of the nervous system in health and disease, Spoke 3 Meeting, Naples, 11/07/2024; Brayn Conference, Turin, 29-31/10/2025.
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Giacomo Turrini, PhD candidate

Supervised PhD students:	na
Honors, prizes, awards:	Best Oral Presentation at the Italian Society of Neuroscience (SINS) PhD meeting, Company of Biology Travel fellowship October 2025
Outreach activities	
● International collaborations:	Prof. Ludovic Telley (University of Lion 1, France)
● Invited talks:	*"Unraveling the Developmental Origins of Cerebellar Astrocyte Heterogeneity"* - oral presentation SINS PhD meeting 2025, Pisa, *10th September 2025*
● Science communication:	
● Editorial duties:	

• others	<u>Membership in Scientific Societies in 2025</u>: Italian Society of Neuroscience (SINS) BRAYN Assosciation Federation of European Neurosciecne Societies (FENS) Italian Glia Network (IGN) Attended Meetings • 8th Cerebellum Day, Rouen 6th, 7th February 2025 *"Molecular diversity of adult mouse cerebellar astrocytes"* – poster presentation • Neurogenomics Conference, Milan 19th, 20th, 21st May 2025 *"Molecular diversity of adult mouse cerebellar astrocytes"* – poster presentation • Glia 2025, Marseille 8th, 9th, 10th, 11th July 2025 *"Molecular diversity of adult mouse cerebellar astrocytes"* – poster presentation • SINS 2025, Pisa, *10th, 11th, 12th, 13th September 2025"Molecular diversity of adult mouse cerebellar astrocytes"* – poster presentation • Brayn 2025, Turin, 29th, 30th,31st October 2025 "*Molecular diversity of adult mouse cerebellar astrocytes"* – poster presentation
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Niccoló Di Cintio, role

Supervised PhD students:	na
Honors, prizes, awards:	--
Outreach activities	
• International collaborations:	--
• Invited talks:	--
• Science communication:	--
• Editorial duties:	--
• others	Participation in GENIE Summer School: "The Impact of sex differences on glia-neuron interactions in brain health and disease" 29th September - 2nd October 2025, held in Regensburg and supported by the Federal Ministry of Education and Research (BMBF). Poster presentations at congresses: -Di Cintio N., Bonato M., Khastkhodaei Ardakani M., Roggero O., Renna A., Montarolo F., Frasca A., Cambiaghi M., Sacchetti B., Buffo A., Boda E. "Cortical hypomyelination is associated with cognitive impairment in a mouse model of

	oligodendroglia-specific deletion of Citron Kinase". 10-14 settembre 2025, Pisa (Italia) - 21° Congresso SINS. - Di Cintio N., Bonato M., Khastkhodaei Ardakani M., Roggero O., Renna A., Montarolo F., Marchiotto F., Scalera M., Vannini E., Frasca A., Cambiaghi M., Sacchetti B., Buffo A., Boda "Oligodendroglia-specific ablation of Citron Kinase is associated with cortical hypomyelination and cognitive impairment which are mitigated by PTEN co-deletion" 29-31 Ottobre 2025, Torino (Italia) - 8° Brainstorming Research Assembly For Young Neuroscientists
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	na

Olga Teresa Bianciotto, PhD candidate

Supervised PhD students:	na
Honors, prizes, awards:	--
Outreach activities	
● International collaborations:	Prof. Giorgia Quadrato (University of Southern California, USA)
● Invited talks:	--
● Science communication:	--
● Editorial duties:	--
● others	Membership in Scientific Societies: Italian Glia Network ALBA Network BraYn Association Attended meetings: • 75° National Congress SIF, Turin, 17-19 September 2025 Poster presentation: *"Enhancing human cerebellar organoids with enrichment of region-specific glia"* • Brayn 2025, Turin , 29-31 October 2025 Poster presentation: *"Enhancing human cerebellar organoids with enrichment of region-specific glia"* • Second Italian Glia Network Symposium, Nov 26 2025 (online)
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

Arianna Contato, Research fellow (until August 9th 2025)

Supervised PhD students:	na
Honors, prizes, awards:	–
Outreach activities	
• International collaborations:	–
• Invited talks:	–
• Science communication:	–
• Editorial duties:	–
• others	Attended Meetings: - IZN/Sorbonne Neuroscience Summer school, Germany, 06-12/07/2025, poster Presentation - MNESYS – A multiscale integrated approach to the study of the nervous system in health and disease, Spoke 3 Meeting, Naples, 11/07/2024, poster presentation - Brayn Conference, Turin, 29-31/10/2025, poster presentation
Organizational activities and responsibilities at NICO:	–
Speakers invited:	–
Other organizational activities:	–
Workshops, Schools or Conferences organized:	–
Technology transfer achievements (patents, etc.):	–

Beatrice Savarese, PhD candidate from November 2025

Supervised PhD students:	na
Honors, prizes, awards:	Best short talk, myDev conference
Outreach activities	
• International collaborations:	ERC Synergy grant Custom-Made (E Cattaneo, University of Milano, M Parmar, University of Lund; J Emneus, Danish Technical University)
• Invited talks:	--
• Science communication:	--
• Editorial duties:	--
• others	Attended Meetings: MyDev (https://mydev.unimi.it/) , Milan, 13/06/2025, short talk and poster presentation
Organizational activities and responsibilities at NICO:	--
Speakers invited:	--
Other organizational activities:	--
Workshops, Schools or Conferences organized:	--
Technology transfer achievements (patents, etc.):	--

4. Research activity in 2025

a. Summary

In 2025, our group advanced cell replacement strategies for Huntington's disease and defined new principles of astrocyte diversification. We provided preclinical evidence for functional integration of human striatal progenitors and mapped astrocyte maturation trajectories and regulatory logic. Moreover, based on epidemiological data linking exposure to high levels of air pollution and Multiple Sclerosis (MS) onset and worsening, we investigated the effects of exposure to particulate matter (PM) on the disease course and biological responses in an experimental mouse model of MS.

b. Background and rationale

Our research is centers around two main areas: (i) glial cell physiopathology and (ii) the reconstruction of functional CNS circuits through cell replacement approaches using pluripotent stem cell derived human neurons.

Multiple sclerosis (MS) is a chronic disease of the central nervous system (CNS), where neuroinflammation and autoimmune response against myelin lead to functional impairments, cognitive and psychiatric symptoms. Exposure to air pollution - in particular to peaks of PM - has been associated with an increase in hospital admissions for MS onset and relapses and exacerbated neuroinflammation in MS patients. In 2025, we aimed at understanding whether individuals primed to develop autoimmunity (as it occurs in MS and in the experimental autoimmune encephalomyelitis-EAE-animal model of MS) respond differently to PM compared to healthy subjects, and at disclosing the biological substrate of such higher vulnerability.

Neurodegenerative and neurodevelopmental disorders are characterized by the progressive loss or dysfunction of specific neural cell types and by profound alterations of glial support. Our research addresses these challenges through two complementary lines of investigation: (i) reconstruction of functional neural circuits using human pluripotent stem cell–derived neurons, with a focus on Huntington's disease (HD), and (ii) dissection of astrocyte diversity, developmental trajectories and regulatory programs in the mammalian brain.

In HD, current gene-targeting approaches hold promise at early disease stages but cannot restore neuronal circuitry once degeneration is established. Cell replacement therefore represents a necessary regenerative strategy. However, its clinical translation critically depends on demonstrating long-term survival, appropriate differentiation, circuit integration and functional impact of transplanted human neurons in vivo.

In parallel, increasing evidence indicates that astrocytes are not a uniform population but consist of regionally and developmentally specialized subtypes that shape circuit maturation, plasticity and disease responses. Yet, how astrocyte diversity emerges, how lineage history constrains fate, and which transcriptional programs govern astrocyte maturation remain incompletely understood.

c. Objectives

Our objectives were to:

(i) assess the long-term maturation, circuit integration and functional impact of human striatal progenitors transplanted in HD models;
(ii-iii) define astrocyte types, subtypes and maturation trajectories in the cerebellum; identify lineage relationships and transcriptional regulators governing astrocyte diversification;
(iv) test whether exposure to PM10 influences the disease course and severity in EAE mouse model of MS;
(v) test whether individuals primed to develop autoimmunity set up a different biological response to PM compared to healthy subjects.

d. Results

Reconstruction of functional CNS circuits through cell replacement
Huntington's disease – cell replacement

We provided preclinical evidence that human embryonic stem cell–derived striatal progenitors survive long term after transplantation into a rodent HD model, undergo progressive maturation and reconstruct striatal circuitry. Using single-nucleus RNA sequencing, we resolved the molecular composition of the grafts, identifying D1- and D2-type medium spiny neurons, interneurons and regionally specified astrocytes. Anatomical tracing, electrophysiology and in vivo calcium recordings demonstrated reciprocal connectivity with host basal ganglia circuits. Importantly, chemogenetic modulation of graft activity altered striatum-dependent behaviors, establishing functional integration and causal contribution of transplanted cells (Scaramuzza, Ribodino, et al., 2025).

Astrocyte diversity and maturation

We generated a comprehensive single-cell and spatial atlas of cerebellar astrocytes, identifying canonical astrocyte types and previously unrecognized subtypes with distinct regional distributions and functional signatures. By integrating postnatal single-cell datasets, trajectory inference and clonal lineage reconstruction, we delineated multiple maturation routes originating from both multipotent and fate-restricted progenitors. These trajectories revealed divergence, convergence and lineage constraints in astrocyte development. Regulatory network analysis uncovered a hierarchical transcriptional code operating at successive levels, from regional patterning and lineage history to astrocyte type and subtype specification. Together, these results define the developmental logic underlying astrocyte heterogeneity and provide a framework applicable to other brain regions. These data are available to the scientific community (https://www.biorxiv.org/content/10.1101/2025.07.17.665323v1) and were submitted for publication to Nat Neurosci. We are currently working on the revision of the study.

Air pollution and MS

We investigated whether an acute exposure to PM10 - occurring either before immunization or during the presymptomatic phase - affects the disease course and the behavioral phenotype of the MOG35–55-induced EAE mouse model of MS. While acute PM10 exposures did not significantly accelerate the emergence of motor deficits, nor modify the disease course in EAE mice, presymptomatic EAE - but not healthy - mice selectively showed increased disinhibited, risk-taking, and novelty-seeking behaviors after being exposed to PM10 (Bonato et al., 2025a).

To assess the biological substrate of the selective response of EAE mice to PM10, we characterized plasmatic extracellular vesicles (EVs) and their miRNA cargo in healthy and presymptomatic EAE mice early after exposure to PM10, compared to unexposed healthy and EAE mice. Results showed that the response of EAE mice to PM10 did not differ in terms of EV number or source, compared to that of healthy mice.Yet, remarkable differences existed in the identity of deregulated EV-associated miRNAs, which, in EAE mice, were predicted to target several MS-relevant biological processes and nervous system-, immune- and inflammation-related pathways, possibly contributing to disease worsening.

e. Advancement in the field

Our work advances the field by demonstrating that human striatal progenitors can functionally integrate into diseased basal ganglia circuits and that the modulation of graft activity influences behavior. These data strengthen the rationale for cell replacement therapies in HD.

In parallel, we provide a unified framework linking astrocyte diversity, lineage history and regulatory logic, highlighting a lineage-intrinsic contribution to how astrocyte identities emerge in the mammalian brain.

Finally, our study shows for the first time a selective vulnerability of immunologically primed mice toward the effects of PM10, occurring before the emergence of overt motor impairment and presenting as specific behavioral alterations and a unique biological response mediated by plasmatic EVs.

f. Publications

- Bonato M, Montarolo F, Parolisi R, Caudana M, Abbadessa G, Cardinale V, Nato G, Pandino C, Di Cintio N, Schiavo V, Giustetto M, Luzzati F, De Francia S, Bertolotto A, Buffo A, Boda E. Acute Particulate Matter (PM_{10}) Exposure Selectively Triggers Behavioral Alterations in the Presymptomatic Experimental Autoimmune Encephalomyelitis (EAE) Mouse Model of Multiple Sclerosis. Eur J Neurosci. 2025 Nov;62(10):e70304.
  Q2 General Neuroscience

- Scaramuzza L, Ribodino M, Cassarino C, Morrocchi M, Gomez Gonzalez GB, Parolisi R, Sozzi E, Turrini G, Cerrato V, Conforti P, Hoxha E, Tognato R, Galeotti G, Cordiglieri C, Crosti MC, Zucca S, Lorenzati M, Bovetti S, Spaiardi P, de'Sperati C, Biella G, Ottoboni L, Parmar M, Maestri S, Besusso D, Cattaneo E, Buffo A. (2025). Transplanted human striatal progenitors exhibit functional integration and modulate host circuitry in a Huntington's disease animal model. Pharmacol Res. Sep; 219:107905.
  Research article Q1, Pharmachology
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- Iegiani G, Pallavicini G, Pezzotta A, Brix AM, Ferraro A, Gai M, Boda E, Bielas S, Pistocchi AS, Di Cunto F (2025). CITK modulates BRCA1 recruitment at DNA double strand breaks sites through HDAC6. Cell Death Dis 16, 320. doi: 10.1038/s41419-025-07655-4.

5. Future directions and objectives for next years

a. Summary

In 2026 we will address critical gaps in the restoration of complex neural networks using regenerative medicine approaches. In particular, we will investigate the role of astrocytes within human grafted cells and systematically map and profile correctly connected neurons in transplants designed to reconstruct striatal circuits.

Our efforts to understand astrocyte heterogeneity will specifically focus on the work requested by the reviewers for the resubmission of our study. We will expand our investigation to human cerebellar astrocytes and perform in silico perturbation analyses of transcriptional regulators identified in mouse cerebellar astrocytes that govern the maturation of distinct astrocytic subtypes. In parallel, we will assess the impact of adding neuroglial cells on neuronal maturation and activity in human cerebellar organoids.

In parallel, we will advance our efforts in identifying novel therapeutic approaches aimed at promoting oligodendroglial functions and (re)myelination in de-/hypo-myelinating pathologies. Specifically, we will investigate the effects of non-invasive brain stimulation in a mouse model of myelin injury. In addition, we will assess pharmacological approaches targeting oxidative stress in cases of developmental hypomyelination.

b. Background and Significance:

Reconstruction of functional CNS circuits through cell replacement

Human neurons grafted in animal models of Parkinson's disease have shown the ability to correctly wire the host brain through diffuse projections, marking significant progress towards the clinics.
However, far less is known about the reconstruction of more complex circuits, such as those affected in Huntington's disease (HD). We have shown that human striatal grafts in HD rodent models display spontaneous activity, respond to sensory stimulation, and can be chemogenetically modulated to influence striatum-dependent behaviors (Scaramuzza, Ribodino et al., 2025). Despite these advances, graft-dependent functional recovery remains limited under non-stimulated conditions.
These findings raise key unresolved questions: (i) to what extent are the direct and indirect striatal pathways anatomically and functionally reconstructed, and is precise circuit reconstruction required for therapeutic efficacy? (ii) how does graft cellular composition influence wiring, integration, and behavioral outcomes? Addressing these questions is central to our ongoing efforts to optimize cell replacement strategies for complex circuit repair.

Glial physiopathology

Glial cells are now recognized as essential regulators of brain function and major contributors to neurological disease. Our work focuses on glial specification during development and on strategies to rescue glial dysfunction in pathological contexts.

We recently generated a comprehensive single-cell and spatial transcriptomic atlas of mouse cerebellar astrocytes, revealing extensive molecular and functional heterogeneity (Cerrato et al., 2025, doi.org/10.1101/2025.07.17.665323 *preprint*). By integrating sc/snRNA-seq, spatial transcriptomics, lineage tracing, and trajectory inference, we identified known astrocyte populations and uncovered previously unrecognized subtypes with specialized functions. Reconstruction of postnatal maturation trajectories demonstrated that cerebellar astrocytes arise from multiple progenitor pools through divergent, convergent, and fate-restricted developmental paths. Importantly, we defined a hierarchical transcriptional regulatory code governing astrocyte identity, from regional lineage specification to mature subtype identity. This framework provides a general model for understanding astrocyte diversification across brain regions. However, astrocyte diversity and development in the human cerebellum remain unexplored, and the functional roles of the transcriptional regulators inferred in mouse requires experimental validation.

In demyelinating pathologies such as MS, myelin repair is inefficient or incomplete, leading to persistent axonal dysfunction and neurodegeneration. Boosting remyelination is thus now considered a key therapeutic goal in regenerative medicine. Experimental studies have provided insights into the cell-intrinsic and environmental mechanisms governing the different steps of remyelination. Although a few of these target mechanisms have already entered clinical trials, effective therapies to restore myelin remain unavailable. Notably, recent evidence suggests that modulation of neuronal activity may represent a promising non-pharmacological strategy to promote remyelination.

Moreover, oligodendrocytes are also affected in a number of developmental diseases including the MCPH17 microcephaly. Yet, the mechanistic aspects of these pathologies are largely unknown, and therapies are lacking. As regards MCPH17, in the Cit-k KO model of the disease we found prominent alterations of oligodendroglia and other glial types which are associated with oxidative stress (Boda et al., 2022, unpublished observations). We propose to investigate whether early antioxidant intervention can mitigate glial pathology and alter disease progression.

c. General aim and integration with mission of the Institute

In 2026, our research will pursue three main objectives.

First, in cell replacement studies, we will investigate the role of graft-derived astrocytes in graft maturation and function, and map reconstructed striatal circuits at single-cell resolution.

Second, in glial biology, we will adopt a comparative approach to determine whether the heterogeneity and transcriptional regulation of adult mouse cerebellar astrocytes are conserved in the human cerebellum and can be recapitulated in human cerebellar organoid models.

Third, we will develop and preclinically validate therapeutic strategies for demyelinating and neurodevelopmental disorders.

Through these activities, we contribute to the mission of NICO by: (i) studying reparative plasticity in the diseased brain; (ii) advancing fundamental knowledge of glial cell pathophysiology; (iii) exploring therapeutic approaches for CNS neurodegenerative and neurodevelopmental diseases; and (iv) developing innovative humanized experimental models and analytical tools.

d. Specific objectives and strategies

Reconstruction of functional CNS circuits through cell replacement

In 2026, the reorganization of our team and research activities driven by the objectives of the Synergy Grant CUSTOM_MADE will continue. A major focus will be to dissect the contribution of astrocytes to graft maturation and circuit reconstruction. To this end, we will leverage single-nucleus RNA-sequencing datasets from striatal grafts and host tissue in HD and PD models to resolve the identity and functional state of graft-derived astrocyte populations. These analyses will inform hypotheses on the beneficial versus detrimental roles of astrocytes within the graft.
In parallel, we will generate a modified H9 human pluripotent stem cell line in which astrocyte differentiation can be selectively ablated. Grafting experiments using this line will allow us to directly test whether the astrocytic component promotes or impairs graft maturation, integration, and functional circuit assembly.

To map reconstructed circuits, we will profile graft-derived neurons that establish correct connectivity with the globus pallidus externus or the substantia nigra pars reticulata. In parallel, we will select and optimize recombinant AAV capsids with enhanced selectivity for D1, D2, or pan-medium spiny neurons, enabling precise circuit tracing and manipulation.
Coll: Prof E Cattaneo, Univ Milano; M. Parmar, and T. Bjorklund, Univ Lund; J. Emneus, Technical University Copenhagen.

Glial physiopathology

Astrocyte heterogeneity

We will integrate proprietary and publicly available human sn/sc RNA-sequencing and spatial transcriptomics datasets to define cerebellar astrocyte subtypes and reconstruct their developmental trajectories. This will allow us to determine to what extent astrocyte heterogeneity observed in mice is conserved or expanded in the human cerebellum.
In parallel, mouse astrocyte heterogeneity and candidate transcriptional regulators will be validated by RNAscope and immunohistochemistry. LoF and GoF approaches, both in vivo and in vitro, will be employed to establish the functional role of these regulatory mechanisms.
Coll: Dr. L. Telley, Université Lyon 1; M. Götz, LMU.

In a complementary line of research, we will use engineered cerebellar organoids seeded with pre-formed glia and assembled with gliospheres to test whether glial addition enhances neuronal maturation. We will also assess whether human glia in this context acquire fetal-like or adult-like molecular and functional features.
Coll: Dr. G. Quadrato, USC.

Promoting myelin repair/plasticity and limiting damage in demyelinating or neurodevelopmental disorders

Enhancing neuronal activity has been shown to promote oligodendrogenesis and remyelination in preclinical studies. Thus, in 2026, we will investigate the suitability of a non-invasive brain stimulation technique (i.e. transcranial direct current stimulation - tDCS) as an option to boost myelin repair and plasticity in a mouse model of myelin injury.
Coll: Dr. M Cambiaghi, Univ of Verona.

Finally, we will investigate if and to what extent NAC (an FDA approved glutathione precursor) administration starting during either embryonic and postnatal development can rescue neuroanatomical and functional abnormalities in MCPH17 mouse mutants.
Coll: Prof. A. Pistocchi (Univ of Milan), Prof. M. Boido, Prof. F. Di Cunto (NICO), Prof. C. Riganti (Dept of Oncology, MBC, Univ of Turin).

e. Unique features of the project research:

Our research stands out in several ways:

- an innovative, glia-centered perspective is distinctive of our scientific approach. By focusing on glial cells as active regulators of circuit formation, plasticity, and disease, we address fundamental mechanisms that remain insufficiently explored and move beyond traditional neuron-centric paradigms;
- we extensively leverage human cell–based models and human data to increase the depth, relevance, and translational potential of our studies. The use of human pluripotent stem cell–derived neurons and glial cells, together with the development of novel human experimental platforms, allows us to investigate human-specific aspects of neural circuit organization, dysfunction and repair that cannot be fully captured in animal models alone;
- our work integrates multilevel methodological approaches with cutting-edge technologies. By combining omics analyses, advanced in vivo manipulations, behavior, human in vitro systems, and computational tools, we ensure both conceptual innovation and strong methodological rigor. This integrative strategy enables the investigation of biological processes across molecular, cellular, circuit, and functional scales.

Within this framework, we address fundamental and still unresolved research questions using original models and approaches.

A major focus is the plasticity of the diseased mature brain. We ask to what extent complex neural circuits can be reconstructed in the adult pathological brain through the addition of new, healthy neurons, and how much anatomical reconstruction is required to sustain functional circuitry. These questions are addressed using basal ganglia circuits as a model system, combined with human cells and platforms that enable the study of human neural circuits.
In parallel, we focus on glial cell physiopathology, aiming to unravel the mechanisms of glial specification and maturation and their interactions with neurons in both physiological and pathological conditions. We introduce further originality by developing and leveraging strategies promoting glia-based repair in conditions of hypo/de myelination and in neurodevelopmental disorder.

f. Methodology

Over the past years, we have invested substantially in integrating state-of-the-art technologies, emerging non-invasive therapeutic approaches, and advanced analytical pipelines to enhance both the quality and the multilevel comprehensiveness of our research. Building on this foundation, additional innovations will be developed in 2026, including:

- Optimization of human cerebellar organoid models with enriched oligodendroglial cells;
- Identification and validation of recombinant AAV capsids selectively targeting MSns

These innovations are further strengthened by our established expertise in histological and high-resolution immunohistochemical analyses, behavioral phenotyping, and mouse genomics technologies.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Embryonic neurogenesis

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Ferdinando Di Cunto
- Degree: MD, PhD
- Birthdate: 20/12/1969
- Phone: +39 011 6706616
- Email: ferdinando.dicunto@unito.it
- Position: Full Professor
- Role: Principal Investigator

Personnel

Christian Pritz
- Degree: PhD in Molecular Biology
- Birthdate: 30/08/1983
- Phone: +39 011 6706616
- Email: christian.pritz@unito.it
- Position: Tenure Track Researcher
- Role & Expertise: Generation and analysis of *C. elegans* models of brain diseases.

Alessia Ferraro
- Degree: MSc in Biotechnology
- Birthdate: 22/08/1997
- Phone: +39 011 6706616
- Email: alessia.ferraro@unito.it
- Position: PhD Student
- Role & Expertise: Molecular biology and biochemical analysis of genetically modified cellular and mouse models of microcephaly and brain tumors.

Flavia Greco
- Birthdate: 09/06/1998
- Degree: MSc in Cellular and Molecular Biology
- Phone: +39 011 6706616
- Email: flavia.greco@unito.it
- Position: PhD Student
- Role & Expertise: Generation and analysis of *C. elegans* models of brain diseases.

Alessia Ghibaudo
- Degree: MSc in Cellular and Molecular Biology
- Birthdate: 19/01/1999
- Phone: +39 011 6706616
- Email: alessia.ghibaudo@unito.it
- Position: Research Fellow
- Role & Expertise: Generation and analysis of *C. elegans* models of brain diseases.

Letizia Lazzerini
- Degree: MSc in Molecular and Applied Biology
- Birthdate: 18/06/1998
- Phone: +39 011 6706616
- Email: lazzerini.letizia08@gmail.com
- Position: Research Fellow (Fondazione Venesio)
- Role & Expertise: In vitro and in vivo analysis of neuronal cells.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
01/01/2020-30/06/2025	Development of Citron Kinase as a therapeutic target for brain tumors. IG 23341	PI	AIRC	Coordinator	855.000 Euro	UNITO
28/09/2023-28/02/2026	Dissection of common mechanisms in genetic primary microcephaly. 2022M75NN8	PI	PRIN 2022 funded by EU through Next Generation EU program.	Coordinator	72.289 Euro	UNITO
26/09/2024-25/09/2026	IDH1/2 mutation inhibition in lower grade gliomas: from preclinical models to clinical applications PNRR-TR1-2023-12378219	PI	Next Generation EU PNRR 2023	Unit leader	175.000 Euro	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Ferdinando Di Cunto (PI)

Supervised PhD students:	Direct supervisor of: Alessia Ferraro Co-supervisor of: Flavia Greco, Gianna Pavarino, Vanessa Chiappini, Sofia Dotta, Ersilia Nicorvo.
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	- Prof. Stephanie Bielas, Department of Human Genetics, University of Michigan Medical School, Ann Arbor, Michigan, USA.
• Invited talks:	
• Science communication:	- 18-03-2025: BIEVOL Association (www.bievol.org) - XVI BIOETHICS WEEK - Speech entitled: Comparison between different types of natural intelligence and artificial intelligence - 21-01-2025 Invited talk in the 'Liberi oltre STEM podcast, entitled: 'Genetics and Intelligence: Debunking Some Myths? https://www.youtube.com/watch?v=vwVRl0NntP0 (more than 8000 views). - 13-05-2025 Invited online seminar for Intesa San Paolo Innovation Center, entitled: 'The nervous system in the deep space'

• Editorial duties:	- Associated Editor of PLoS ONE - Associated Editor of Frontiers in Neurogenesis
• others	Memberships in Scientific Societies in 2025: - Federation of the European Neuroscience Societies (FENS) - American Society of Cell Biology (ASCB) - Italian Society for Neuroscience (SINS) - Italian Society for Biophysics and Molecular Biology (SIBBM) - Italian Society of Genetics (AGI)
Organizational activities and responsibilities at NICO:	- Data management - Responsible for Corruption Prevention and Transparency
Speakers invited:	Salvatore Adinolfi – University of Torino - Frataxin: a biochemistry tale from molecular to pathophysiology of neurodegeneration.
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Chrisitan Pritz, Tenure Track Researcher

Supervised PhD students:	Co-supervisor of: Flavia Greco
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	Alon Zaslaver, Department of Genetics, Silberman Institute for Life Sciences, Edmond J. Safra Campus, The Hebrew University of Jerusalem, Jerusalem, Israel.
• Invited talks:	
• Science communication:	8th Brainstorming Research Assembly for Young Neuroscientists (29-31. October 2025), Torino

Alessia Ferraro, PhD student

Supervised PhD students:	
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	
• Invited talks:	
• Science communication:	June 14-19, 2025 (Lisbon) European Association for Cancer Research (EACR) Annual Meeting October 14-16, 2025 (Bilbao) European Association for Cancer Research (EACR) Conference, Cancer-neuroscience October 29-31, 2025 (Turin) Brayn Annual Meeting

4. Research activity in 2025

a. Summary

We study the genetic and molecular mechanisms that control neuron generation, survival and differentiation during normal brain development and how the alteration of these processes may lead to neurodevelopmental

disorders, in particular primary microcephaly. To this aim, we currently use a combination of experimental and computational methods to analyze in vitro (including brain organoids) and in vivo (mouse and C. elegans) models. We also study the relevance of these mechanisms for brain cancers.

b. Background and rationale

The human brain is composed of approximately 90 billion neurons, which are generated during embryonic life starting from many different types of neural stem cells, whose proliferation is extremely well organized in space and time. If too few neurons are produced, or too many neurons die during development, the brain volume can be very compromised, a condition commonly known as microcephaly. Although a significantly reduced brain volume can be compatible with normal brain function and intelligence, microcephaly is frequently associated with strongly invalidating symptoms, such as intellectual disability, epilepsy and cerebral palsy. Microcephaly can be the result of rare genetic disorders or is produced by environmental factors, such as hypoxia, drugs and alcohol exposure or infectious agents, such as Rubella, Toxoplasmosis, Cytomegalovirus or Zyka virus. Research conducted in the last decade has shown that all these conditions may affect common molecular pathways, regulating genome stability, cell proliferation, cell survival and determination of cell identity.
The main focus of our group is to understand the molecular events activated by genetic and non-genetic conditions leading to normal neuronal numbers and neuronal differentiation. In particular, we have been studying for many years the neurological syndrome produced by CITK inactivation, characterized by microcephaly, ataxia and epilepsy. This syndrome has recently been identified in humans with the name of MCPH17. Neural progenitors of humans or mice carrying CITK mutations fail to divide and undergo programmed cell death, leading to strong reduction of the final neuron number. During the last few years we have dedicated much effort to clarify the causal relationship between these events and the other mechanisms classically associated with microcephaly, such as asymmetric cell division of neural precursors and DNA damage. In the last few years, we have underscored that most of the biological activities of CITK can be reconciled with the modifications which it produces on the dynamics of microtubule cytoskeleton. For these reasons, we are investigating how microtubule-dependent events may simultaneously affect mitotic fidelity and genomic integrity. Our latest achievement was to highlight the relevance of human brain organoids in the study of human-specific mechanisms of primary microcephaly, which cannot be adequately represented in rodent models. Using this assay, we showed that the catalytic activity of CITK is much more essential in human than in mouse development, paving the way for further studies on the relevan cell biology. Finally, considering the strong involvement of CITK in proliferation, we are addressing the hypothesis that it may be required also by brain tumor cells, in particular those that characterize the pediatric tumor medulloblastoma, with the aim of developing new specific drugs for these devastating tumors.

c. Objectives

Our constant research aims have so far been to clarify:

1. how mutations in Citron kinase lead to microcephaly;
2. what are the molecular consequences of CITK loss;
3. what is the mechanism linking CITK and microtubule dynamics;
4. CITK as a possible target for cancer therapy.
5. During the last two years we have started to approach other neurodevelopmental disorders, using the power of the genetically tractable organism C. elegans. In particular, we have established a collaboration with the group of Prof. Alfredo Brusco, who has recently bcome a Faculty member of the Department of Neuroscience, to address the biological role of mutations recently recognized as causative alterations of Autism Spectrum Disorder syndromes.

d. Results

1. We have discovered that, besides impairing cytokinesis, CITK may lead to microcephaly by two additional mechanisms. The first is through an alteration of the cell division plan, which may affect cell the rate of exit from the cell cycle and therefore reduce the number of neurons possibly generated by neural progenitors. An interaction between CITK and the prominent microcephaly protein ASPM is essential for this function. The second mechanism is by directly regulating genomic stability, independently from its role in cytokinesis.

Indeed, we found that cells lacking CITK display increased DNA damage early in the cell cycle. An alteration of DNA repair mechanisms may be the leading cause of this phenotype. Increased DNA damage leads to P53 activation, which is the main cause of apoptosis in CITK null models. Indeed, we found that the inactivation of P53 in CITK knockout animals leads to a disappearance of apoptosis, and strongly improves the overall neurological phenotype. The latter discovery could pave the way to the identification of new possible therapeutic strategies for apoptosis-related microcephaly. We have recently shown that DNA damage, apoptosis and cytokinesis failure produced by CITK loss are strongly dependent on the presence of the non-kinase domains of CITK, while the catalytic activity seems specifically necessary for polarized cytokinesis in developing human brain organoids.
2. CITK was originally identified as a protein important for remodeling the actin cytoskeleton. We have discovered that it may be even more important to regulate the stability of microtubules, and that this function is crucial both for completing cytokinesis and for spindle orientation. Moreover, we have established the microtubules play a crucial role in the CITK-dependent maintenance of genomic stability, by modulating the recruitment of BRCA1 and other components of the Homologous Recombination repair pathway to DNA double strand breaks, through the HDAC6 protein. By using the Zebrafish model, we also provided proof of principle that HDAC6 modulation could rescue, at least in part, the microcephaly produced by CITK loss.
3. Goldberg-Shprintzen disease (GOSHS) is a rare microcephaly syndrome accompanied by intellectual disability, dysmorphic facial features, peripheral neuropathy and Hirschsprung disease. It is associated with recessive mutations in the gene encoding kinesin family member 1-binding protein (KIF1BP, also known as KIFBP). The encoded protein regulates axon microtubules dynamics, kinesin attachment and mitochondrial biogenesis, but it is not clear how its loss could lead to microcephaly. We identified KIF1BP in the interactome of CITK. KIF1BP and CITK interact under physiological conditions in mitotic cells. Similar to CITK, KIF1BP is enriched at the midbody ring and is required for cytokinesis. The association between KIF1BP and CITK can be influenced by CITK activity, and the two proteins may antagonize each other for their midbody localization. KIF1BP knockdown decreases microtubule stability, increases KIF23 midbody levels and impairs midbody localization of KIF14, as well as of chromosome passenger complex. These data indicate that KIF1BP is a CITK interactor involved in midbody maturation and abscission, and may play a crucial role downstream of CITK also in microtubule remodeling and prevention of DNA double strand breaks accumulation. Recently, we have found that another important link between CITK and microtubule may be represented by the HDAC6 proteins, whose inhibition can rescue microcephaly in a zebrafish model of MCPH17.
4. We have addressed the possibility that the function of CITK may be essential for proliferation in medulloblastomas, devastating brain tumors of the infancy that urgently require the development of new therapies. To do so, we have produced a conditional model for deleting CITK in medulloblastomas arising in mutant mice. We have also addressed whether the discoveries which we have published for Medulloblastoma may apply to more prevalent brain tumors, such as glioblastomas, and may increase the radiosensitivity of both tumor types.
5. The recruitment of Dr. Christian Pritz allowed us to implement all the necessary setups to use the C. elegans model at NICO. Using this genetically tractable system we have started to address the molecular bases of two severe neurodevelopmental disorders whose causative mutations have recently been found by Prof. Brusco laboratory, as wella as of MCPH17.

e. Advancement in the field

The results which we obtained have contributed important advances in the field of microcephaly studies. In addition, we have provided evidence that genes involved in primary microcephaly are suitable targets for brain tumor therapy, in particular medulloblastoma. The understanding of mechanisms which may link microtubule dynamics to the maintenance of genome integrity by homologous recombination would be a major advancement in the field. Moreover, the modeling of neurodevelopmental disorders in C. elegans is expected to provide major contribution to the molecular and cellular understanding of disease mechanisms in a wide variety of neurodevelopmental disorders.

f. Publications

- Errico F, Russo R, Carrillo F, Nuzzo T, di Vito R, Canonico E, Pedone PV, Di Cunto F, Esposito T, Usiello A, Chambery A. (2025). Free D-aspartate modulates the expression of proteins linked to schizophrenia and autism spectrum disorder during early postnatal life. Cell Mol Life Sci. Dec 24;83(1):3. Research article (Q1).
- Di Cunto F, Catania MV, Simola N, Mingardi J, Serra M, Campanelli F. (2025). Neurobiological underpinnings of neurodegenerative and neuropsychiatric disorders: from models to therapy. Front Neurosci. Jun 9;19:1627262.
  Editorial (Q2).
- Iegiani G, Pallavicini G, Pezzotta A, Brix A, Ferraro A, Gai M, Boda E, Bielas SL, Pistocchi A, Di Cunto F. (2025). CITK modulates BRCA1 recruitment at DNA double strand breaks sites through HDAC6. Cell Death Dis. Apr 20;16(1):320.
  Research article (Q1).
- Chiappini V, Castillo MFV, Biancardi F, Di Cunto F, Magistretti PJ, Vercelli A, Agus M, Calì C. (2025). Ultrastructure of astrocytes using volume electron microscopy: A scoping review. J Physiol. Mar 3.
  Review Q1

5. Future directions and objectives for next years

a. Summary

During the next three years, we plan to continue the development of the current research lines and in particular:

1. We will continue to dissect the molecular mechanisms by which Citron kinase loss leads to microcephaly. To this regard, we plan to study the mechanisms by which CITK loss alters microtubule nucleation and stability. Moreover, we will investigate how CITK mutation or inactivation leads to DNA damage. Finally, we will address the possible causal relationships between these events and will investigate how they relate to the other genes so far involved in primary microcephaly and, possibly, in medulloblastoma progression.
2. We will continue to address the role of CITK in brain tumors and in their radiosensitivity. In particular, if the AIRC grant proposal will be funded, we plan to concentrate on the development of specific CITK inhibitors.
3. We will strengthen our efforts to increase the collaborations between NICO and clinical researchers of the Department of Neuroscience. Specifically, we are working on the implementation at NICO of the genetically tractable model C. elegans, which will be of invaluable help in addressing the biological significance of mutations identified in a clinical setting, in patients affected by neurodevelopmental and neurodegenerative disorders.

b. Background and Significance

***Neurodevelopmental disorders and intellectual disability*.**
Neurodevelopmental disorders (NDD) comprise a heterogeneous group of clinical diagnoses, including autism-spectrum disorders, intellectual disability (ID), attention deficit/hyperactivity and epilepsy. Although these syndromes are usually presented as distinct entities in the fifth edition of the Diagnostic Statistical Manual of mental disorders (DSM5) NDD have the tendency to co-occur in the context of complex clinical syndromes, often characterized by recurrence in families. NDD are frequently very invalidating and possess an enormous social impact, because they affect up to 3% of children. Modern molecular genetics technologies, based on massively parallel sequencing platforms, have allowed to identify many genetic alterations significantly associated to NDD. In few cases, the identification of the underlying mutations has rapidly allowed to identify a possible therapy. Despite these advances, the diagnostic and therapeutic approach to NDD is still very critical, especially because of their extremely heterogeneous and multifactorial origin.

Microcephaly

Congenital microcephaly (CM) is a heterogeneous group of disorders characterized by reduced head circumference at birth, to at least 3 standard deviations below the mean. CM can be the result of non-genetic conditions, such as viral infections and toxic exposure, or it can be generated by rare genetic disorders, with mostly autosomal recessive inheritance. Under More than 450 loci associated with microcephaly are known in the OMIM database. Primary hereditary microcephaly (MCPH) is the simplest form of genetic CM, in which brain size reduction is accompanied by grossly normal brain architecture and mild to moderate intellectual disability. Pure MCPH is a rare condition, since genetic CM is more often associated with syndromic features and co-morbidities, including structural brain abnormalities, seizures, palsy, ataxia, short stature, skeletal abnormalities and cancer predisposition. Although these conditions are usually classified as separate clinical entities, the elucidation of their genetic, molecular and cellular basis is revealing a high degree of overlap. Our studies are aimed at significantly extending the current knowledge on these disorders, and to identify possible therapeutic strategies. Interestingly enough, most of the identified MCPH genes play crucial roles in cell division and genomic integrity of neural progenitor cells, making them very interesting candidates as targets for drug development in brain cancers.

c. General aim and integration with mission of the Institute

The general aim of our group is to significantly advance knowledge on neurodevelopmental disorders, in particular microcephaly syndromes. In the next three years, we plan to extend our studies to selected forms of autism spectrum disorders. Since intellectual disability and behavioral abnormalities are the most important clinical consequences of these conditions, we think that our research is fully consistent with the mission of the Foundation and of the Institute.

d. Specific objectives and strategies

1. Validation of new potential CITK partners and substrates identified through proteomics.
To identify CITK physical interactors and substrates, we performed a proteomics screen. We identified many proteins capable of forming complexes with CITK independently of kinase activity, but also 34 proteins specifically co-purified by the catalytically inactive CITK bait. Importantly, the latter list contains many tubulins and tubulin-related molecules, suggesting that kinase activity is crucial for regulating CITK-microtubules interactions. To this regard, we are currently investigating the interplay between CITK, CYLD and HDAC6, since the latter proteins are connected both to the regulation of microtubule stability and to DNA repair.
2. Hypothesis-driven investigation of the molecular mechanisms through which CITK regulates microtubule dynamics.
We found that CITK controls cytokinesis and spindle orientation by altering microtubule dynamics, a scenario supported by the results of our proteomics screen. These functions involve at least in part the capability of CITK to modulate TUBB3 phosphorylation through CK2a recruitment. Moreover KIF14, whose loss leads to microcephaly in mice and humans, is a partner of CITK in regulation of midbody stability. Since kinesins play established roles in microtubule dynamics and CK2a has been involved in kinesins' regulation, we will set out to obtain more information about the interplay between all these molecules.
3. Hypothesis-driven investigation of the molecular mechanisms through which CITK prevents DSBs accumulation.
An important question raised by our studies is how CITK protects cells from accumulation of DNA damage, independently of its role in cytokinesis. Therefore, we will continue to investigate in detail the mechanisms by which CITK may affect RAD51 and BRCA1, which shows reduced recruitment to foci, dependent on the presence of CITK protein but on kinase activity. Moreover, we need to address whether other repair pathways, in particular the non-homologous end joining (NHEJ), are also compromised by CITK loss. We will also continue to address the relationships between CITK and microtubule dynamics in control of genome stability.
4. Implementation of new mouse and human MCPH17 pre-clinical models.
We aim at translating our mechanistic findings to experimental models directly relevant for the human disease. Since most MCPH17 patients carry kinase dead mutations, we have undertaken the production a new mouse model, characterized by a similar alteration. We would also like to explore the potential usefulness of neural progenitor cells derived from MCPH17 patients as a possible platform for drug screening

and validation, by transferring to these cells our discovery that the effects of CITK loss can be alleviated by P53 inactivation.

5. Identification of new genes involved in NDD.

We will work with our collaborators in the Genetics and Neuroscience departments to identify NDD patients who may carry novel genetic alterations. In particular, we will use our computational skills to analyze the copy number variation data and the exome sequencing data produced by our collaborators, to identify the variants most likely causing the disorders. We plan to validate the most interesting alterations using both transgenic and knockdown models in the genetically tractable model *C. elegans*. Moreover, we plan to use neural stem cell culture and also human forebrain organoids, derived from patient-specific induced pluripotent stem cells

e. Unique features of the project research

The most peculiar aspect of our group is our capability to combine different approaches, including computational biology, biochemistry, molecular biology and experimental analysis in cultured and in vivo models for approaching sophisticated biological questions related to brain development and brain disorders.

f. Methodology

The most innovative aspects of our research will be:

1. the use of human brain organoids, derived from induced pluripotent stem cells. These sophisticated cultures are ideally suited to reproduce in culture the fundamental cellular events that characterize the first stages of brain embryonic development, especially those that are specific of humans and cannot be therefore mimicked by mouse models. We have partially setup this system at NICO, and we plan to use it both for our studies on microcephaly and for functionally characterize the new NDD genes which we should identify with our collaborators.

2. the extensive use of computational biology/bioinformatics techniques, with the aim of directing and optimizing the experimental work.

3. the development of the genetically tractable model *C. elegans* among the main platforms of the Institute.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Neuropsychopharmacology

1. LABORATORY DESCRIPTION – PERSONNEL

Principal Investigator

Ilaria Bertocchi
- Degree: PhD
- Birthdate: 13/04/1982
- Phone: +39 011 6706611
- Email: ilaria.bertocchi@unito.it
- Position: Research Assistant (RTD-B)
- Role: Principal Investigator

Personnel

Alessandra Oberto
- Degree: PhD
- Birthdate: 24/10/1967
- Phone: +39 011 6706611
- Email: alessandra.oberto@unito.it
- Position: Research Associate (RU)
- Role & Expertise: Design, supervision and conduction of experiments with expertise in biotechnology, molecular and cellular biology, immunohistochemistry and laboratory rodents.

Lorenzo Cifarelli
- Degree: Medical Biotechnology
- Birthdate: 17/07/1997
- Phone: +39 011 6706632
- Email: lorenzo.cifarelli@unito.it
- Position: PhD Student
- Role & Expertise: Conduction of experiments with expertise in handling murine models of neuropsychiatric disorders, behavioral tests, epifluorescence microscopy, image analysis and data analysis.

Alessandra Lodi
- Degree: Neuroscience
- Birthdate: 18/01/1994
- Phone: +39 011 6706611
- Email: alessandra.lodi@unito.it
- Position: PhD Student
- Role & Expertise: Conduction of experiments with expertise in molecular and cellular biology, immunohistochemistry, laboratory rodents and flies, behavioral tests, image analysis and data analysis.

Magalì Giordano
- Degree: Medical Biotechnology
- Birthdate: 13/02/1996
- Phone: +39 011 6706632
- Email: magali.giordano@unito.it
- Position: Research Fellow
- Role & Expertise: Conduction of experiments with expertise in molecular and cellular biology, Western blot, laboratory rodents and flies, cardiovascular physiology, image analysis and data analysis.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
2023-2025 Extended till 28/02/2026	Matrix Metalloproteinase-9 and PeriNeuronal Nets: new therapeutic targets for Fragile X Syndrome	Carola Eva	PRIN: PROGETTI DI RICERCA DI RILEVANTE INTERESSE NAZIONALE	ex-PI	204.298 Euro	UNITO
28/02/2025-30/6/2027	Progetto Dieta e Salute: impatto della dieta chetogenica nel trattamento dell'epilessia farmacoresistente	Ilaria Bertocchi	Fondazione CRT	PI	38.000 Euro	UNITO

3. SCIENTIFIC ACTIVITIES IN 2025

Ilaria Bertocchi, Research assistant (RTD-B)_PI

Supervised PhD students:	Lorenzo Cifarelli and Alessandra Lodi
Honors, prizes, awards:	n/a
Outreach activities	
• International collaborations:	-Mazahir Hasan, Ikerbasque Professor and Group Leader at the Achucarro Basque Center for Neuroscience heading the Laboratory of Brain Circuit Therapeutic -José María Delgado García, full Professor of Physiology Pablo de Olavide University Division of Neurosciences, Building 21, Pablo de Olavide -Rolf Sprengel, Research Group of the Max Planck Institute for Medical Research at the Inst. for Anatomy and Cell Biology, Heidelberg University -Pierandrea Muglia, GRIN Therapeutics Inc, New York, NY, USA -Daniela Carulli, Laboratory for Neuroregeneration Netherlands Institute for Neuroscience (NIN) Meibergdreef 47 - 1105 BA Amsterdam - The Netherland
• Invited talks:	"Matrix Metalloproteinase-9 and PeriNeuronal Nets as New Therapeutic Targets for Fragile X Syndrome (FXS)" 21st Workshop on Fragile X syndrome & other Neurodevelopmental Disorders. 15-18 September 2025 Noordwijk, The Netherlands 'Exploring the effects of a ketogenic diet in mice carrying the GluN2A(N615S) mutation' 7th European GRIN Conference October 17 -19, 2025. Palace Grand Hotel Varese, Varese, Italy
• Science communication:	n/a
• Editorial duties:	Reviewer for the following journals (2024): iScience, Molecular Psychiatry, Biochemistry and Biophysics Reports

• others	Attended Congress: • 75th Congress of The Italian Society of Physiology (SIF 2025) held in Turin, Italy, from 17 to 19 September 2025. • 8th Brayn Conference 29-31 October 2025, Turin Italy Attended courses: ETICA E CONCEZIONE DEI PROGETTI, MODULI 9, 10, 11, DM 5 AGOSTO 2021 - Edizione Unica - 'The Science Bridge' advisory board member https://thesciencebridge.org/ - Comitato scientifico assmgrin2aitalia https://assmgrin2aitalia.it/comitato-scientifico/ - Comitato scientifico di associazione GRI Italia https://www.gri-italia.it/
Organizational activities and responsibilities at NICO:	In charge for behavioral labs I and II (animal facility)
Speakers invited:	n/a
Other organizational activities:	n/a
Workshops, Schools or Conferences organized:	n/a
Technology transfer achievements (patents, etc.):	n/a

Alessandra Oberto, Research associate (RU)

Supervised PhD students:	Lorenzo Cifarelli and Alessandra Lodi
Honors, prizes, awards:	n/a
Outreach activities	
• International collaborations:	-Mazahir Hasan, Ikerbasque Professor and Group Leader at the Achucarro Basque Center for Neuroscience heading the Laboratory of Brain Circuit Therapeutic -Rolf Sprengel, Research Group of the Max Planck Institute for Medical Research at the Inst. for Anatomy and Cell Biology, Heidelberg University -Pierandrea Muglia, GRIN Therapeutics Inc, New York, NY, USA -Daniela Carulli, Laboratory for Neuroregeneration Netherlands Institute for Neuroscience (NIN) Meibergdreef 47 - 1105 BA Amsterdam - The Netherland
• Invited talks:	n/a
• Science communication:	n/a
• Editorial duties:	n/a
• others	Attended Congress: • 75th Congress of The Italian Society of Physiology (SIF 2025) held in Turin, Italy, from 17 to 19 September 2025. • 7th European GRIN Conference October 17 -19, 2025. Palace Grand Hotel Varese, Varese, Italy • 8th Brayn Conference 29-31 October 2025, Turin Italy
Organizational activities and responsibilities at NICO:	In charge for behavioral labs I and II (animal facility)
Speakers invited:	n/a
Other organizational activities:	n/a
Workshops, Schools or Conferences organized:	n/a

Technology transfer achievements (patents, etc.):	n/a

Lorenzo Cifarelli, PhD student

Supervised PhD students:	n/a
Honors, prizes, awards:	FENS (*Federation of European Neuroscience Societes*) travel grant of 600€ to attend FRM (*FENS Regional Meeting*) 2025 in Oslo, Norway
Outreach activities	
• International collaborations:	Cesare Patrone, head of NeuroCardioMetabol group at Department of Clinical Science and Education, Södersjukhuset, Internal Medicine, Karolinska Institutet (Stockholm, Sweden)
• Invited talks:	21th national congress of SINS, symposium *Neurobiological Insights into Obesity and Eating Disorders*. Talk title: *High-fat-diet promotes brain insulin-resistance and alters cerebral GLUT4 and neuroplasticity*
• Science communication:	n/a
• Editorial duties:	n/a
• others	• Workshop SINS *The Neuroscience of Cognitive and Behavioural Flexibility: from Function to Dysfunction*. Poster: *Behavioral impairments and perineuronal net expression in fragile X syndrome mice differ upon age and sex* • FENS regional meeting 2025. Poster: *Ketogenic Diet in a Mouse Model of Grin2a Syndrome: Effects on Behaviour, Neuroplasticity and Neuroinflammation* • SINS PhD meeting 2025. Poster: *Neuroplasticity and behavioral changes in Fmr1KO mice across postnatal development* • 8th BraYn (*Brainstorming Research Assembly for Young Neuroscientists*) conference 2025: - Poster: *Brain-wide analysis of perineuronal nets and parvalbumin-positive interneurons in fragile X syndrome mice across postnatal development highlights region-specific alterations influenced by age and sex* - Poster: *Sex- and region-specific modulation of perineuronal nets, parvalbumin interneurons, and microglia by ketogenic diet in a Grin2a mouse model of developmental and epileptic encephalopathy* • XXVI Conference of Young SIF (*Società Italiana di Farmacologia*). Oral presentation: *High-fat-diet-induced brain insulin resistance dampens GLUT4 cerebral expression and impairs neuronal plasticity*
Organizational activities and responsibilities at NICO:	n/a
Speakers invited:	n/a
Other organizational activities:	n/a
Workshops, Schools or Conferences organized:	n/a
Technology transfer achievements (patents, etc.):	n/a

Alessandra Lodi, PhD student

Supervised PhD students:	n/a
Honors, prizes, awards:	- SIF Award for Best Oral Communication, Monothematic Congress SIF, "Social Dysfunction: Exploring Mechanisms and advancing therapeutic interventions" (Genova, 21-22/10/2025) - SIF Award for Best Oral Communication, Monothematic Congress SIF "The impact of micro- and macro-environments on immunity and inflammation: paving the way for novel immunotherapies" (Perugia, 15-16/06/2025) - PhD Scholarship 2025-2028 in the Doctoral Programme "Experimental Medicine and Therapy", University of Turin
Outreach activities	
• International collaborations:	n/a
• Invited talks:	n/a
• Science communication:	n/a
• Editorial duties:	n/a
• others	• Monothematic SIF congress (Italian Society of Pharmacology) "La moderna ricerca scientifica e le differenze: l'importanza della farmacologia e tossicologia di genere" (Foggia, 12-13/12/2025), **Oral Presentation** • 8th BraYn (Brainstorming Research Assembly for Young Neuroscientists) conference 2025 (Turin, 29-31/10/2024), **2 Posters presented** • Monothematic SIF congress (Italian Society of Pharmacology) "Social Dysfunction: Exploring Mechanisms and advancing therapeutic interventions (Genova, 21-22/10/2025), **Oral Presentation and SIF Award for Best Oral Communication** • 7th European GRIN Conference (GRIN Europe) (Varese, 17-19/10/2025), **Poster presentation** • 75th National Congress SIF (Italian Society of Physiology) (Turin, 17-19/09/2025), **Poster presentation** • 21st National SINS (Italian Society for Neuroscience) (Pisa, 10-13/09/2025), **Poster presentation** • Monothematic SIF congress (Italian Society of Pharmacology) "The impact of micro- and macro-environments on immunity and inflammation: paving the way for novel immunotherapies" (Perugia, 15-16/06/2025), **Oral presentation and SIF Award for Best Oral Communication** • Workshop SINS (Italian Society for Neuroscience): "The Neuroscience of Cognitive and Behavioral Flexibility: from Function to Dysfunction" (Milan, 27/01/2025), **Poster presentation**
Organizational activities and responsibilities at NICO:	n/a
Speakers invited:	n/a
Other organizational activities:	Contribution to the organization of the 75th National Congress SIF, Turin, 17-19/09/2025

Workshops, Schools or Conferences organized:	n/a
Technology transfer achievements (patents, etc.):	n/a

Magalì Giordano, research fellow

Supervised PhD students:	n/a
Honors, prizes, awards:	n/a
Outreach activities	
• International collaborations:	n/a
• Invited talks:	n/a
• Science communication:	n/a
• Editorial duties:	n/a
• others	• 23 – 25 October 2025 – Poster presentation XXV CONGRESSO NAZIONALE SOCIETÀ NAZIONALE DI RICERCHE CARDIOVASCOLARI. Imola, Italy. Magalì Giordano, S. Femminò, A. Brovero, R. Ciullo, C. Barale, M. Bertinaria, C. Penna, P. Pagliaro, I. Russo. "Platelet modulation in cardiac Ischemia/Reperfusion Injury: functional impact of ADP, Hyperglycemia and NLRP3 inhibition" • 5 – 7 October 2025 – Oral presentation XXIV Riunione nazionale del Gruppo di Studio delle Piastrine, Caramanico Terme (PE), Italy. Magalì Giordano, C. Barale, A. Brovero, R. Ciullo, S. Femminò, M. Bertinaria, C. Penna, P. Pagliaro, I. Russo. "ADP, iperglicemia e NLRP3: determinanti della risposta piastrinica nel danno cardiaco da ischemia/riperfusione" • 17 – 19 September 2025 – Poster presentation 75th SIF NATIONAL CONGRESS. The Italian Society of Physiology. Turin, Italy. Magalì Giordano, S. Femminò, A. Brovero, R. Ciullo, C. Barale, C. Penna, P. Pagliaro, I. Russo. "Modulatory role of platelets in ischemia/reperfusion injury: functional impact of ADP and NLRP3 inhibition"
Organizational activities and responsibilities at NICO:	n/a
Speakers invited:	n/a
Other organizational activities:	n/a
Workshops, Schools or Conferences organized:	n/a
Technology transfer achievements (patents, etc.):	n/a

4. Research activity in 2025

a. Summary

We have added new and interesting developments to the two projects we are carrying out using our mouse models of neurodevelopmental diseases: the Fragile X syndrome and the GRIN2A syndrome. This was also possible thanks to the start of new collaborations. Two of them even led us to work on external projects, which enabled us to publish in 2025, and we expect more publications in the near future.

b. Background and rationale

Mice with the knockout of the Fmr1 gene (Fmr1KO mice) well recapitulate the main symptoms of Fragile X syndrome (FXS), the most common genetic cause of intellectual disability and autism spectrum disorder (ASD). Grin2a$^{S/S}$ mice represent a unique and valuable genetic mouse model of rare syndromes known as GRIN-related developmental and epileptic encephalopathies (DEEs). Grin2a$^{S/S}$ mice carry a point mutation analogous to those found in children affected by DEE, which confers audiogenic seizures, hyperactivity, impulsivity, and dysregulated attention to the mouse (Bertocchi et al., 2021a;2023; 2024).
These two models are genetically determined; however, in addition to genetic influences, environmental factors play a critical role in shaping the individual's phenotype.
In the first model, we have focused our attention on specialized forms of extracellular matrix, the perineuronal nets (PNNs), which arise in concomitance with the closure of developmental critical periods of plasticity and control synaptic connectivity and functions in the adult brain (Bertocchi et al., 2021b). An impairment of PNN formation around GABAergic parvalbumin-positive neurons (PV+ cells) in the developing auditory cortex (AC) of Fmr1KO mice has been reported, and their pharmacological or genetic restoration significantly ameliorated FXS-associated hyperresponsivity to acoustic stimuli. Despite this evidence, the role of PNN in other areas of the FXS brain is still largely unknown. Understanding the role of PNNs in the whole wild-type brain and the functional consequences of their dysregulation in the FXS brain is crucial for identifying novel therapeutic strategies.
In the second model, we have focused our attention on sex-dependent differences in audiogenic seizure (AGS) susceptibility and in the efficacy of antiseizure medicaments (ASM) in our homozygous Grin2a$^{S/S}$ mice (Bertocchi et al., 2024). We asked if, by treating these mice with a ketogenic diet (KD), a dietary protocol documented to be highly effective for children and adults with drug-resistant epilepsy, we could obtain further information related to sex differences in this model. Furthermore, several studies suggest that the KD has positive effects on cognitive abilities and benefits from a metabolic and physiological point of view. However, only a few preclinical and clinical studies have analyzed the effects of KD, and the results have not been conclusive. Discordant data also exist regarding inflammation and the effects produced by KD at a peripheral level. This diet is therefore poorly integrated into clinical practice, mainly due to scarce knowledge on: (i) biomolecular mechanisms, (ii) specific biomarkers of efficacy/toxicity, (iii) risks deriving from prolonged use. It is essential to have new experimental data to improve the level of knowledge on KD.

c. Objectives

Thanks to our internal collaboration with Prof. Luzzati, we worked on the conclusion of our study related to PNN distribution in the whole FXS brain, and we continued to get data related to the expression of these plasticity players at different times of postnatal development in brain nuclei important for cognitive and emotional functions of Fmr1KO mice.
We also increased the number of experimental subjects to get more insights into the efficacy of KD in counteracting seizures and other deficits in our preclinical model of GRIN2A syndrome. Our new collaboration with Prof. Penna's laboratory and the acquisition of a new research fellow helped us in collecting new information related to the impact of the mutation on the cardiovascular system.
In addition, we contributed to the behavioral phenotyping of a new model of autism spectrum disorder (ASD), the SKT mouse, and we plan to further characterize it in the future, thanks to our new collaboration with Prof. Defilippi's laboratory.

d. Results

1) We finalized the statistical analysis and the production of graphs and tables for the writing of an important manuscript about the whole brain parvalbumin and perineuronal net expression analysis in Fragile X mice, by Luzzati and Cifarelli et al (in preparation). The automated analysis of PNN and PV+ cells in the entire Fmr1KO mouse brain at P60 revealed a general reduction of these plasticity players in FXS brains compared to the WT ones. The neocortex is the brain area most affected by a significant PNN reduction, whereas PV+ cell density is significantly decreased in lower limbic areas, such as the amygdala.

In parallel, we obtained the behavioral characterization of an adequate number of mice at different stages of development (P20, P40, and P60), and we are currently completing the analysis of their brains for PNN and PV+ cell distribution. In this way, we will obtain information about the differences between Fmr1KO and control C57BL/6 wild-type mice and between sexes (we used both hemizygous male and homozygous female Fmr1KO mice) in the process of PNN formation during development.
Additionally, we obtained new data about the effect of maternal care of FXS dams on pups' behaviour as adults. We documented differences in maternal behaviour of FXS and WT C57BL/ 6J dams toward their pups during the first postnatal week of life, with Fmr1KO mothers being less caring than WT controls. Interestingly, we also showed that Fmr1KO +/- mothers are less maternal and more hyperactive compared to the homozygous ones. Pups raised by Fmr1 KO +/- showed reduced sociability, short-term memory, and anxiety.
2) We validated the therapeutic efficacy of KD in counteracting seizures in our preclinical model of GRIN2A syndrome: AGS episodes were largely decreased in KD-treated Grin2a$^{S/S}$ mice compared to those subjected to a standard diet (SD). Mild effects are present relating to behavioral and cognitive functions. We also documented diet-induced peripheral inflammation after analyzing liver and blood samples. As documented with other therapeutic tools (Bertocchi et al., 2024), Grin2a$^{S/S}$ females were found to be more susceptible to AGS and AGS-induced cardiorespiratory arrest, but also more responsive to treatment. AGS susceptibility leading to death fulfills several criteria used to diagnose sudden unexpected death in epilepsy (SUDEP) in humans. SUDEP is the most common type of epilepsy-related death, and the mechanisms underlying it are still poorly understood. Considerable emphasis in SUDEP research was given to long QT syndrome and mutations that affect LQTS genes, which encode mainly for ion channels expressed throughout the body. To date, mutated NMDARs seem not to be included in this mutation list, despite evidence of their expression in peripheral tissues, including the heart, and considering the significant function of these receptors in the control of the autonomic and cardiovascular systems. Indeed, while the role of GluN2A-containing NMDARs in neuronal processes has been extensively investigated, their specific physiological function in cardiac tissues remains largely unknown. We have found that the higher excitability in females compared to males paralleled greater cardiac vulnerability, as reflected by larger infarct sizes, suggesting shared Ca^{2+}-dependent mechanisms linking synaptic brain dysfunction to circuit-level instability and peripheral tissue plasticity.
3) Prof. Defilippi's research group at MBC, University of Turin, has identified a protein, called SKT, that is predominantly expressed in the brain within dendritic spines, in the post-synaptic density (PSD). The lack of SKT in nerve cells produced threadlike, immature, and dysfunctional dendritic spines and impaired neuronal communication. We analyzed the behavior of SKT-KO mice at NICO and found motor and cognitive deficits similar to those exhibited by rodent models of autism spectrum disorder (ASD), such as Shank-3 KO mice. These results are now published (Morellato et al., 2025), but further questions remain unanswered.

e. Advancement in the field

We are honored to have contributed to the work that, for the first time, showed that the protein SKT acts as a molecular adaptor in dendritic spines, so that it can cooperate with other PSD members to allow precise neuronal communication and, in turn, ensure functional behavior. Since the behavioral phenotype of the mouse is similar to established models of autism (Shank3 KO mice), we have validated a possible new ASD model that can be used to better understand the mechanisms related to these disorders.
Staying on the line of neurodevelopmental diseases, GRIN-related DEEs and FXS are rare syndromes that currently lack effective treatments. Notably, our past work demonstrated the high predictive value of our GRIN2A mouse model, and our new data regarding the cardiovascular system in this model may shed light on new mechanisms related to SUDEP.
Such results can translate into an increase in knowledge that, over time, can generate concrete applications and positively impact the well-being of society.

f. Publications

- Morellato A, De Gregorio M, Angelini C, Torelli F, Belmonte V, Alfieri A, Bersia B, Cravero T, Gai M, Bianciotto OT, Rebecca Oddone R, Salemme V, Natalini D, Centonze G, Nigrelli F, Raspanti A, Gurgone A, D'Attanasio G, Mele P, Eva C, Lodi A, Gavello D, Carabelli V, Hidisoglu E, Chiantia G, Carbone E, Becchetti A, Giustetto M, Marcantoni A, Turco E, Bertocchi I*, Defilippi P* (2025). The

adaptor protein SKT interacts with PSD-95 and SHANK3 and affects synaptic functions. Cell Reports 44, 116206, September 23, 2025.
Research article Q1

5. Future directions and objectives for next years

a. Summary

1) We will complete our preclinical study on the therapeutic efficacy of KD in counteracting cognitive and behavioral deficits and epileptic seizures typical of early-onset epileptic and developmental encephalopathies (DEE) induced by mutations in the GRIN genes, coding for glutamate receptors.
This is fundamental for developing new drugs with an overall efficacy while avoiding side effects as much as possible. Interestingly, we revealed a sex-dependent difference not only in AGS susceptibility and in the effects of the KD, but also in the effects of the mutation in the peripheral system, particularly on cardiovascular physiology.
2) Another goal is to validate PNN and MMP-9 as new molecular targets for therapeutic intervention in FXS. We plan to preclinically assess new treatments at the most effective developmental stage, an experimental opportunity impossible to achieve with clinical studies. To do this, we have established a collaboration with NICO members and with national and international experts in the field.
We desire to apply to national and international calls for grants on rare diseases.
3) In 2025, we submitted a grant application to a foundation for a project on schizophrenia, but it was unsuccessful. We would like to secure new funding for this project, as we believe it is valuable.

b. Background and Significance

1) AGS susceptibility leading to death for cardio/respiratory arrest (RA) fulfills several criteria used to diagnose sudden unexpected death in epilepsy (SUDEP) in humans. SUDEP is the most common type of epilepsy-related death, and the mechanisms underlying it are still poorly understood. Considerable emphasis in SUDEP research was given to long QT syndrome and mutations that affect LQTS genes, which encode mainly for ion channels expressed throughout the body. To date, mutated NMDARs seem not to be included in this mutation list, despite evidence of their expression in peripheral tissues, including the heart, and considering the significant function of these receptors in the control of the autonomic and cardiovascular systems. Indeed, while the role of GluN2A-containing NMDARs in neuronal processes has been extensively investigated, their specific physiological function in cardiac tissues remains largely unknown. An in-depth study of the role of NMDARs at this level is therefore essential.
2) Perineuronal nets (PNNs) are specialized forms of extracellular matrix that arise during development and control synaptic plasticity and functions. Overexpression of matrix metalloproteinases like MMP-9 degrades PNNs around GABAergic parvalbumin-positive (PV+) neurons in the developing auditory cortex (AC) of Fmr1KO mice. Pharmacological or genetic restoration of PNNs in the AC significantly ameliorated FXS-associated hyperresponsivity to acoustic stimuli, suggesting that PNN and MMP-9 represent new FXS therapeutic targets. We conducted an automated analysis of PNN+ and PV+ cells in the entire mouse brain at P60 and found a reduction of PNNs and PV+ in Fmr1 KO brains compared to controls. Moreover, Fmr1KO mice and controls of both sexes at different stages of development were subjected to a behavioral test battery, and their brains were analyzed to study PNN formation.
3) Schizophrenia (SCZ) is considered a neurodevelopmental disorder that develops from a combination of genetic and environmental factors. Rare and common variants in the GRIN2A gene, encoding the GluN2A subunit of the NMDAR, greatly increase the risk of SCZ. These variants generally lead to NMDA receptor hypofunction. Grin2a-deficient mice ($Grin2a^{+/-}$) were recently proposed as a valuable genetic model of SCZ. These mice show EEG and behavioral changes similar to those observed in patients, together with molecular features that support the "hypo-glutamate" and "hyper-dopamine" hypotheses of SCZ. Expression of GRIN2A starts during postnatal brain development and influences the maturation of PV+ interneurons and the formation of PNNs in an activity-dependent way. Environmental stimuli during such periods can cause irreversible brain function and structure changes. Early-life maternal care, for example, can affect neural circuit formation during critical periods, impacting future cognitive and emotional functioning. Mothers with SCZ may face difficulties in parenting due to the disorder itself, exhibiting disturbances in their interactions

with their infants. Children may, in turn, exhibit unstable attachments, low self-esteem, diminished cognitive and problem-solving abilities, and reduced empathy and sociability as they grow. Postnatal stress and poor maternal care have also been associated with increased SCZ risk.
NMDA receptor function impacts maternal care: injections of an NMDAR antagonist into the medial preoptic area reduce maternal behavior in rat mothers. More importantly, mice carrying the GRIN2A S644G variant have shown impaired maternal behaviors. Thus, alterations in the GRIN2A gene can impact maternal behavior, even if the exact mechanisms behind are not fully understood and require further investigation. Despite the significance of these factors, no research has examined the complex relationship between maternal behavior and GRIN2A function in regulating brain plasticity and SCZ-like behavior in Grin2a+/- mice.

c. General aim and integration with mission of the Institute

The general aim of our research is to delve into basic nervous system mechanisms and fascinating internal and external interactions that can alter its function. Our projects are focused on the search for new therapeutic targets and treatments for various neurodevelopmental disorders and their comorbidities. Actually, our first line of research has great clinical relevance for the development of therapies for children, adolescents and adults affected not only by rare epileptic syndromes, but also by other forms of drug-resistant epilepsy and neurodisorders, in which there is an imbalance between excitatory and inhibitory neuronal transmission. For this reason, our purposes fit very well with the mission of the Institute.

d. Specific objectives and strategies

1) We hypothesize that a bidirectional interaction between brain and heart NMDAR activity may drive sex-specific changes in the effect of the GluN2A(N615S) mutation and susceptibility to SUDEP.
By using the $Grin2a^{S/S}$ mouse model, we aim to investigate whether the GluN2A(N615S) mutation contributes to oxidative stress, inflammation, or ischemic injury in the heart. This would provide new insights into new basic mechanisms related to GRINpathies. Furthermore, a second aim is to elucidate better the mechanisms underlying sex-based differences in AGS susceptibility and drug response, which could be related to differential expression of GRIN genes and a significant influence of sex hormones on glutamate transmission.
Control and mutant mice of both sexes, exposed to standard or KD for 5 weeks since weaning, and then to the AGS protocol, will be sacrificed and perfused. A whole-brain analysis of the expression of different markers for inflammation and plasticity will be conducted. The same analysis can be done for the perfused heart, with a 3D reconstruction of either hematoxylin-eosin- or immunofluorescent-stained heart sections, to examine the gross morphology, the presence of hypertrophy or necrosis, and to verify the expression of crucial proteins related to glutamate and heart functionality. In parallel, another cohort of mice will be sacrificed by cervical dislocation for the explant of the fresh heart and brain, to conduct an experimental ischemia-reperfusion protocol and to perform western blot analyses, respectively. In this way, we will collect information related to the effects of different experimental conditions (e.g, therapeutic diet, epileptic activity, and both, with their controls) on the expression of markers relative to the physiology and functioning of the brain and the heart of male and female mutant and control mice.
2) Thanks to our collaboration with the University of Rome ("La Sapienza" and Catholic University), we are screening compounds with known and putative effects on MMP-9 activity using an in vitro assay as well as human FXS and control fibroblasts; the most efficient compounds identified through the in vitro screening will then be tested in Fmr1KO mice and human FXS iPSCs-derived neurons. We also aim to analyze the selected compounds' effects on the intrinsic neuronal excitability, glutamatergic and GABAergic synaptic transmission in Fmr1KO brain slices by patch-clamp recordings. The final goal is to identify specific brain areas and corresponding time windows of intervention that could be therapeutically relevant for treating Fragile X Syndrome.
3) We hypothesized that maternal care could ameliorate or exacerbate SCZ-like symptoms and molecular signatures related to GluN2A hypofunction. Starting from the above-described research premises, we would like to complete the following aims:

Aim 1: to examine the maternal behavior of Grin2a+/- dams compared to Grin2a+/+ controls towards their pups or pups from their genotypic counterpart (cross-fostered);

Aim 2: to analyze the behavior of the littermates as adults. Eight experimental groups: males and females, cross-fostered (CF) or unfostered (UF) Grin2a+/- and Grin2a+/+ mice;
Aim 3: to perform ex-vivo analysis on brain and blood samples to examine molecular and physiological differences between experimental groups. Grin2a knock-out (KO) mice (genetic background C57Bl6) are already under breeding in our animal house, being a kind gift of our close collaborator, Prof. Sprengel.

e. Unique features of the project research

A significant feature of our project research is its high translational potential: the main objective of our projects is, in fact, to identify potential new or old therapeutic 'bullets' with a new use for conditions with unmet medical needs.
Our projects are very connected to each other and involve the collaboration of a network of experts in both basic and clinical sciences, as well as the support of the association of parents of children affected by rare syndromes (https://www.gri-italia.it).

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Neuroendocrinology

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator (Acting Group leader)

Stefano Gotti
- Degree: PhD
- Birthdate: 17/06/1971
- Phone: +39 011 6706610
- Email: stefano.gotti@unito.it
- Position: Associate Professor
- Role: Principal Investigator (Acting Group Leader)

Personnel

Marilena Marraudino
- Degree: PhD
- Birthdate: 08/06/1988
- Phone: +39 011 6706632
- Email: marilena.marraudino@unito.it
- Position: Assistant Professor (RTD-B)
- Role & Expertise: Control of reproduction and endocrine disruptors.

Chiara Ballan
- Degree: Master's Degree
- Birthdate: 20/09/1995
- Phone: +39 011 6706632
- Email: chiara.ballan@edu.unito.it
- Position: PhD Student
- Role & Expertise: Endocrine disruptors.

Elena Ricci
- Degree: Master's Degree
- Birthdate: 12/06/1998
- Phone: +39 011 6706632
- Email: elena.ricci@edu.unito.it
- Position: DNS Research Fellow
- Role & Expertise: Endocrine disruptors.

Nebai Ontiveros
- Degree: Master's Degree
- Birthdate: 20/09/1994
- Phone: +39 011 6706632
- Email: nebai.ontiveros@edu.unito.it
- Position: NICO Research Fellow
- Role & Expertise: Eating disorders models.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
2021-2024 (end 04.25)	Developmental, Reproductive and Metabolic effects of Endocrine Disruptors: the DReaM-ED study	Gotti, PI	PRIN 2020	PI of research unit	134.696 Euro	UNITO
2024-2026	Fitoestrogeni e diete ipoproteiche in madri con Anoressia Nervosa, quali le alterazioni per il neurosviluppo della prole?	Marraudino	Fondazione CRT	Coordinator	25.000 Euro	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Stefano Gotti, PI

Supervised PhD students:	Chiara Ballan
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	Cooperation with Prof. P. Collado and H. Pinos (UNED, Madrid, Spain) Cooperation with Prof. D. Grassi (Universidad Europea de Madrid, Madrid, Spain)
• Invited talks:	The effects of exposure to endocrine disruptors during pregnancy, Congresso SIAI (19/09/2025)
• Science communication:	Piattaforma Plastiche sostenibili SusPlas@Unito
• Editorial duties:	Journal Reviewer: Brain Research, Cell and Tissue Research, Physiology and Behavior, Neurological Science, Toxics, Chemosphere, Cells, Neurobiology of Disease, Molecular and Cellular Neuroscience, Journal of Neuroendocrinology, Behavioral Brain Research Guest editor of the Special Issue: Steroids and the Nervous System 2024 (Journal of Neuroendocrinology, 2025)
• others	
Organizational activities and responsibilities at NICO:	First aid and fire safety officer Responsible for Cryostat Room Responsible for Perfusion Room Responsible for -80 Room
Speakers invited:	

Other organizational activities:	
Workshops, Schools or Conferences organized:	Organizer of the 13th Meeting Steroids and Nervous System, Torino, February 21-25, 2026 Organizer of the SNS Webinar, 11/04/2025 Organizer of the SNS Webinar, 10/11/2025 Organizzatore del XXXV Congresso Nazionale Gruppo Italiano per lo Studio della Neuromorfologia (GISN)
Technology transfer achievements (patents, etc.):	

Marilena Marraudino, Assistant Professor RTD-B

Supervised PhD students:	N.A.
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	• Prof. Daniela Grassi, Neuroanatomy and Neuroendocrinology, Faculty of Basic Biomedical Science, Universidad Europea de Madrid, Madrid, Spain. • Prof. Paloma Collado, Neuro Psychoendocrinology, National Distance Education University (UNED), Department of Psychology, Madrid, Spain • Prof. Luis Miguel Garcia Segura, Neurodevelopment and Neuroinflammation, Cajal Institute, Neuroactive steroids, Madrid, Spain. • Dr. Matthieu Keller, Neuroendocrinology, INRA, Neuroendocrinologie des Interactions et Comportements Sexuels, Tours, France.
• Invited talks:	02/12/2025 - Invited by Dr. Silvia Diviccaro for a communication entitled 'Phytoestrogens as EDCs, when the term “natural” is not always synonymous with safety'. Course 'Health effects of endocrine disruptors', Dept. of Pharmacological and Biomolecular Sciences, University of Milan, Italy.
• Science communication:	20/11/2025 - Disturbatori endocrini: i pericoli nascosti nella vita quotidiana dei nostri figli. Asilo Bianca e Bernie. Incontro per educatori e genitori. 03/11/2025 - Nuove tecnologie e neurosviluppo: come la tecnologia influenza il nostro cervello. AizoOn. 10/10/2025 – Festival dell'Innovazione e della Scienza 2025. Caffè Scientifico. Distruttori endocrini e microplastiche: il loro impatto sulla salute. Settimo Torinese, Torino. 26/09/2025 - U*NIGHT - The European Researchers' Night 2025. Laboratori di ricerca. Neuroscienze presso UNITO: scopri le nostre sfide quotidiane. 19/06/2025 - Settimana della Scienza, Museo di Anatomia Umana Luigi Rolando, Torino. Ritorno al futuro: dialoghi tra anatomia storica e neuroscienze contemporanee. Il cervello dei due sessi, uno strano mosaico.

	03/02/2025. 'Il cervello nei due sessi'. Prof. Annarita Deluca, Istituto di Istruzione Superiore Santorre di Santarosa.
• Editorial duties:	• Reviewer for Hormones and Behavior, Official Journal of the Society for Behavioral Neuroendocrinology. • Topical Advisory Panel Member of section "Molecular Endocrinology and Metabolism" in the International Journal of Molecular Sciences (IJMS) of Multidisciplinary Digital Publishing Institute (MDPI).
• others	
Organizational activities and responsibilities at NICO:	Responsible for Cryostat Room Responsible for Perfusion Room
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	Organizzatore di webinar per Giovani ricercatori in qualità di membro della 'Educational Committee for International Meeting Steroids and Nervous System (SNS)' 11/04/2025 – Secondo webinar SNS per giovani ricercatori. 25/11/2025 – Terzo webinar SNS per giovani ricercatori. Organizzatore del XXXV Congresso Nazionale Gruppo Italiano per lo Studio della Neuromorfologia (GISN)
Technology transfer achievements (patents, etc.):	N.A.

4. Research activity in 2025

a. Summary

In 2025, our research examined how Endocrine Disruptors impact reproductive parameters and how, in a model of gaming disorders, several neurotransmitter dysfunctions occur.

b. Background and rationale

Steroid hormones in the nervous system mediate many biological processes. The estrogen receptors (nuclear, ERα and ERβ; membrane, GPER-1) are expressed in various brain regions. Estrogens may modulate neuronal differentiation, particularly by influencing cell migration, survival, death, and synaptic plasticity.
Thus, maintaining proper levels of gonadal hormones is crucial for the normal development and sexual differentiation of the central nervous system (CNS), as well as for reproductive behavior. Interrupting this developmental process with external estrogen treatment or gonadectomy during key prenatal and postnatal phases can cause permanent changes in CNS organization and lead to behavioral changes across various species.
Our research investigates the effects of external factors disrupting this delicate hormonal balance. We performed experiments simulating environmental changes to examine their impact on behavior and associated neural circuits.
One of our research lines investigates Endocrine Disrupting Chemicals (EDCs), substances that can alter endocrine system function and are closely linked to the development and operation of the nervous system. Examples include xenoestrogens and xenoandrogens, which can significantly affect the development and function of gonadal hormone-dependent neural circuits and associated behaviors. The impact of EDCs varies based on several factors, including the route, duration, and dose of exposure. Developmental stages are generally much more vulnerable to signal disruption than adult stages, and the effects of fetal or neonatal exposure may differ drastically from those of adult exposure.

Our lab also focuses on a rat model of gaming disorder (GD). Recognized as a mental disorder, GD exhibits notable differences between males and females, with males being more frequently affected. Additional research is necessary to understand these sex differences better, and animal models can serve as valuable tools in uncovering the neurological mechanisms behind GD. We are especially focused on how these differences influence addictive behaviors, brain activity during gaming, and possible alterations in circuits that govern the reward system.

c. Objectives

Our primary objective was to investigate EDCs, especially their effects on neural circuits and behavior. We examined how perinatal exposure to various EDCs impacts neuroendocrine circuits involved in reproduction. Additionally, we studied neurotransmitter dysfunctions in a model of gaming disorder.

d. Results

Perinatal exposure to endocrine-disrupting chemicals

Exposure to endocrine-disrupting chemicals (EDCs) is especially dangerous during critical life stages when organisms are more susceptible to hormonal fluctuations. EDCs can impact multiple aspects of life, such as maternal behavior.

Bisphenols are synthetic organic compounds widely used in plastics and are a common type of endocrine-disrupting chemicals (EDCs). These substances, whether alone or in mixtures, can disrupt hormone functions in various ways. Exposure to bisphenols, especially during critical developmental periods, can cause significant health effects. This study specifically investigates the impact of perinatal exposure to BPA and its common substitute, BPS, on anxiety-related behaviors and the serotonergic system, which plays a key role in regulating these behaviors, in adult mice.

C57BL/6J dams were orally administered 4 μg/kg body weight/day of BPA, BPS, or a control vehicle from mating until the offspring were weaned. This dose corresponds to the previous EFSA TDI for BPA. In the adult offspring, we conducted the two-bedding T-Maze test and monitored spontaneous sexual behavior (**Bonaldo et al., 2025)**. Exposure to BPA delayed the onset of puberty in females, while BPS advanced it in males, and both altered the estrous cycle in females. Sexual and sex-related behaviors were partially altered in males, especially in the BPA-exposed group.

Regarding kisspeptin immunoreactivity in the analyzed hypothalamic nuclei, in BPA- or BPS-treated females, we observed an increase in the rostral periventricular area. BPA increased immunoreactivity in the paraventricular nucleus, whereas BPS decreased it compared with control females.

Among males, there was a notable increase in the arcuate nucleus of BPA-treated males and a significant decrease in the paraventricular nucleus of BPS-treated males.

These findings suggest that perinatal exposure to low doses of BPA or BPS influences reproductive parameters, sexual behaviors, and kisspeptin hypothalamic nuclei in a sex-specific way.

Rat model of gaming disorder

Gaming Disorder (GD) is classified as a mental health disorder. It mainly impacts adolescents, who after becoming addicted, exhibit traits like social anxiety, depression, social withdrawal, and attention issues.

In a prior study (**Casile et al., published in May 2025 but available online in April 2024**), we introduced a novel GD rat model designed to explore specific aspects of the disorder. This model exhibited key GD traits such as hyperactivity and a lack of control, with the behavioral phenotype remaining stable over time. These traits were not observed in the control groups. Furthermore, analysis of c-Fos immunoreactivity across the prelimbic cortex (PrL), orbitofrontal cortex (OFC), nucleus accumbens, amygdala, and bed nucleus of the stria terminalis (BNST) showed significant differences between GD and control groups, highlighting alterations in neural activity associated with the emergence of the GD phenotype.

In a follow-up study (**Casile et al., published in January 2026 but available online in October 2025**), we used immunohistochemistry and high-performance liquid chromatography to measure ORX, DA, and 5-HT levels in rats subjected to the GD protocol. Compared to control groups, GD rats of both sexes exhibited a decrease in the density and number of ORX+, DA+, and 5-HT+ cells within the examined nuclei. Furthermore, the decrease in DA+ and 5-HT+ neurons was linked to changes in their plasma levels and modifications in the tryptophan signaling pathway, as well as a rise in inflammatory metabolites. These three systems are crucial for proper brain function, regulation of decision-making, and processing of emotional and reward-related stimuli.

Our results support the idea that neurotransmitter dysfunction is central to GD and provide a foundation for future studies on the brain and behavioral changes seen in GD patients.

e. Advancement in the field

Our results confirm that endocrine-disrupting chemicals (EDCs) can notably influence pups during the perinatal period. We detected clear changes in both males and females across multiple brain regions, along with alterations in certain behaviors. Importantly, our study also shows that BPS, a main alternative to BPA, may have detrimental effects that are potentially more severe than those of BPA.

f. Publications

- Bonaldo B, Casile A, Bettarelli M, Marraudino M, Gotti S. (2025). Perinatal exposure to bisphenol A or S alters differently sexual behavior and kisspeptin system in mice. Environ Res. Mar 15; 269. Research article (Q1).
- Casile A, Bonaldo B, Bettarelli M, Papadopoulos P, Micioni Di Bonaventura MV, Nasini S, Comai S, Sut S, Dall'Acqua S, Marraudino M, Gotti S, Cifani C. (2026). Central and peripheral monoamine changes in a rat model of Gaming Disorder. Neuropharmacology. Jan 1; 282:110703.
  Research article (Q1).
- Casile A, Marraudino M, Bonaldo B, Micioni Di Bonaventura MV, Nasini S, Cifani C, Gotti S. (2025). Novel rat model of gaming disorder: assessment of social reward and sex differences in behavior and c-Fos brain activity. Psychopharmacology (Berl). 2025 May; 242(5):1103–1122.
  Research article (Q1).
- Giatti S, Gotti S, Melcangi RC. (2025). Steroids and nervous system-2024. J Neuroendocrinol. Jul; 37(7):e70048.
  Editorial (Q1).

5. Future directions and objectives for next years

a. Summary

We will develop multiple lines of research:

a) Effects of perinatal exposure to a mixture of EDCs. Throughout life, we are exposed to multiple EDCs simultaneously, so we aim to assess the cumulative effects of these exposures on the survival of mouse pups, serotonergic circuits, and anxiety- and sex-related behaviors.
b) Effects of gonadal receptor hormone antagonists on neural circuits involved in reproduction, metabolism, and other physiological functions.
c) Effects of drug treatment in a rodent model of Gaming Disorder. After developing and validating the gaming disorder model, we plan to test various addiction-targeting drugs on it to observe their effects.

b. Background and Significance

Steroid hormones are essential for the development, growth, maturation, differentiation, and protection of both the central and peripheral nervous systems. Derived from cholesterol, they are produced in organs such as the adrenal glands, gonads, and placenta. The nervous system can also synthesize or metabolize active steroids (neurosteroids) on its own, potentially influencing nerve cell activity and survival. Gonadal steroid hormones play a key role in developing sexually dimorphic neural circuits and functions, while also regulating vital physiological processes like reproduction, metabolism, parental care, social interactions, and aggression. Understanding the mechanisms behind their actions is crucial, especially identifying which estrogen receptor types are involved in controlling these diverse circuits and behaviors.
Our research explores sex differences across various levels, the impact of stimulating different estrogen receptors, and how environmental factors influence the nervous system and behavior.
Broadly speaking, the environment can significantly influence neural circuits; in fact, numerous substances called Endocrine-Disrupting Chemicals (EDCs) can modify endocrine system functions, which are closely

linked to nervous system development and operation. Many EDCs are xenoestrogens or xenoandrogens, which—even at very low levels—can markedly influence the development and functioning of gonadal hormone-dependent neural circuits and associated behaviors. They may induce subtle effects by interfering with gene expression and other cellular mechanisms, leading to temporary changes or permanent damage. Therefore, the effects of EDCs depend on factors such as how, how long, and how much exposure occurs. Developmental stages are typically far more vulnerable to signal disruption than adult stages, and the consequences of fetal or neonatal exposure may be markedly different from those of adult exposure. What is the main problem associated with EDCs? These environmental contaminants demonstrate endocrine activity across humans, wildlife, and domestic animals. Certain "natural" EDCs, like plant phytoestrogens present in food, can substantially affect the reproductive cycles of small rodents and may have both beneficial and harmful impacts on other animals, including humans. Recently, the idea of metabolic disruptors—substances that can cause significant changes in metabolism—was introduced. Another key environmental impact is associated with parental behavior. Research has shown that a lack of maternal care can cause lasting changes in certain behaviors of pups when they reach adulthood, along with permanent modifications in neuroendocrine circuits. Often, these effects vary between males and females, likely because gonadal hormones play a role in this process.

c. General aim and integration with the mission of the Institute

Our main goal is to investigate how steroid hormones influence and regulate neural circuits that manage vital physiological processes such as reproduction, food intake, and metabolism, paying particular attention to gender differences. This objective is closely related to the neuroendocrine basis of some neurodegenerative diseases, which exhibit notable sex differences. Treatments for these diseases should always consider that some core mechanisms may differ based on sex or steroid influence. Furthermore, environmental factors also appear to play a role in disease development. Thus, endocrine-disrupting compounds (EDCs) capable of interacting with steroid hormone receptors are important candidates in this environmental impact. As a result, we intend to further our projects concentrating on the neuroendocrine system, neurodegenerative and psychiatric disorders, and their possible connections to environmental influences.

d. Specific objectives and strategies

EDCs effects.

EDCs can affect the endocrine system by interfering with hormone synthesis, bioavailability, and action, and by altering cellular proliferation and differentiation, tissue development, and various metabolic processes. Among them, BPA, BPS, and perfluoroalkyl compounds (PFAS) have spread widely in recent decades. They are manufactured through industrial processes for everyday products, are highly persistent in the environment, readily absorbed, and are slowly eliminated. Humans are exposed to them by consuming food and drinks that have come into contact with plastics. Exposure during pregnancy is associated with adverse pregnancy and birth outcomes. Gaining a clearer understanding of how these pollutants cause molecular disturbances, how these mechanisms interact, and whether they exhibit sex-specific effects is crucial for developing targeted strategies to mitigate chemical-induced risks diseases. The aim of our project is to evaluate the effects of exposure to these EDCs in dams during the critical period of pregnancy and later in pups once they become adults.

Mothers were orally treated with the single or mixture of EDCs from E0 to PND14 with very low doses (new EFSA TDI: 0.2 ng/kg body weight/day for BPA and BPS; 0.7 ng/kg body weight/day for PFOS). After the birth of the pups, the mothers' maternal and anxiety behaviors will be analyzed.

After weaning, the offspring will be monitored until postnatal day 90, at which point we will analyze their reproductive behavior. Based on previous findings, we can hypothesize that EDCs may delay the onset of puberty and disrupt the estrous cycle in females. Additionally, both bisphenols and PFOS are likely to impact sexual behavior in males. Through immunohistochemical analysis of the treated groups, we aim to reveal changes in several neuronal systems.

Effects of Estrogens on pups.

Gonadal steroids play a crucial role in influencing and maintaining hypothalamic systems that regulate metabolism and reproduction during key developmental stages. Estradiol (E2) appears to be vital in organizing these neural circuits. It interacts with two nuclear receptors, ERα and ERβ, as well as a membrane

receptor, GPR30. The serotonin (5-HT) system is vital for managing anxiety-like behaviors and is modulated by estrogen. Our goal is to identify which estrogen receptor exerts an organizational influence on anxiety-related behavior, thereby affecting the distribution of 5HT+ neurons. During the critical postnatal period, male and female CD1 mice will receive subcutaneous injections from postnatal day 5 to 12. They will be treated with E2 alone or with selective estrogen receptor antagonists—MPP, PHTPP, and G15—that inhibit ERα, ERβ, and GPR30, respectively. In adulthood, the mice will undergo behavioral assessments to measure anxiety and will be analyzed via immunohistochemistry for the 5-HT system in the Raphe Nucleus.

Rat model of gaming disorder (GD)

Although play is a crucial aspect of human behavioral development, recent studies suggest that children and adolescents who spend more time on electronic media may face intra- and interpersonal risks. Because of its strong resemblance to addictive disorders and its coexistence with social anxiety and attention deficits, the the loss of control over gaming (Gaming Disorder, GD), is classifying as a mental disorder. GD primarily affects adolescents, who, after developing an addiction, display traits such as social anxiety, depression, social isolation, and attention problems. However, recent human studies reveal several limitations, including a lack of demographic diversity, uneven age and gender representation (most studies focus on males, with few including females), differences in game types, and variations in follow-up periods. After developing and validating a new gaming disorder model, we plan to test various addiction-targeting drugs on it to observe their effects. Male and female Wistar Kyoto rats will undergo training using an innovative touchscreen platform to develop GD. At the end of the training, rats will be randomly assigned to either the vehicle group or the Fluoxetine-treated group to determine whether drug treatment decreases play-related behaviors and increases social interaction in rats of both sexes.

e. Unique features of the project research

The Neuroendocrinology team has a longstanding interest in how steroid hormones interact with nervous tissue, focusing on behavior as the main physiological measure. Multiple steroid hormone receptors are found across various brain regions, and steroid hormones are known to affect neuronal and glial cell differentiation, survival, and protection. Consequently, gaining a deeper understanding of how steroid hormones relate to the nervous system is essential. These interactions might also partly account for gender differences observed in both normal and disease states. Furthermore, in the last twenty years, understanding how environmental factors influence human and animal physiology to cause diseases has gained significance in biomedical research. It is understandable that numerous synthetic substances may bind to hormone receptors, possibly leading to endocrine imbalances and health issues. For years, the neuroendocrine effects were overlooked, and nervous tissue was not a main focus of research. Furthermore, this was not regarded as a key endpoint in developing toxicological tests for regulating EDC use. Our group's research focuses on the potential risks of EDCs, especially their impact on the developing central nervous system. Overall, our work improves understanding of gender differences in healthy brains, various neural disorders, and the complex interactions between neural circuits, behavior, and environmental contaminants.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Peripheral Nerve Regeneration Unit

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Stefania Raimondo
- Degree: PhD in Physiology; Master's Degree in Biological Sciences (University of Turin)
- Birthdate: 25/02/1977
- Phone: +39 011 6705433
- Email: stefania.raimondo@unito.it
- Position: Full Professor
- Role: Principal Investigator (Acting Group Leader succeeding Stefano Geuna, Rector of the University of Turin until 30 September 2025)

Personnel

Giulia Ronchi
- Degree: PhD in Neuroscience; Master's Degree in Neurobiology (University of Turin)
- Birthdate: 27/11/1982
- Phone: +39 011 6705433
- Email: giulia.ronchi@unito.it
- Position: Associate Professor
- Role & Expertise: In vivo models for peripheral nerve regeneration study.

Giovanna Gambarotta
- Degree: PhD in Cellular Sciences and Technologies; Master's Degree in Biological Sciences (University of Turin)
- Birthdate: 22/08/1967
- Phone: +39 011 6705436
- Email: giovanna.gambarotta@unito.it
- Position: Associate Professor
- Role & Expertise: In vitro models and biomolecular analysis for peripheral nerve regeneration study.

Federica Fregnan
- Degree: PhD in Neuroscience; Master's Degree in Biological Sciences (University of Turin)
- Birthdate: 02/07/1976
- Phone: +39 011 6705433
- Email: federica.fregnan@unito.it
- Position: Research Technician
- Role & Expertise: In vitro model for peripheral nerve regeneration study.

Luisa Muratori
- Degree: PhD in Experimental Medicine and Therapy; Master's Degree in Neurobiology (University of Turin)
- Birthdate: 02/05/1984
- Phone: +39 011 6703577
- Email: luisa.muratori@unito.it
- Position: Research Assistant
- Role & Expertise: In vitro and in vivo models for autonomic nervous system regeneration.

Marwa El Soury
- Degree: PhD in Neuroscience (University of Turin); Master's Degree in Molecular Biology and Biotechnology (Alexandria University)
- Birthdate: 22/04/1991
- Phone: +39 011 6705436
- Email: marwa.elsoury@unito.it
- Position: Post-doctoral Fellowship Recipient (concluded 30/09/2025)
- Role & Expertise: Biomolecular analysis of peripheral nerve regeneration.

Federica Zen
- Degree: Master's Degree in Industrial Biotechnologies (University of Padova)
- Birthdate: 06/04/1995
- Phone: +39 011 6705436
- Email: federica.zen@unito.it
- Position: PhD Student, PhD Program in Experimental Medicine and Therapy (concluded 31/05/2025)
- Role & Expertise: Biomolecular analysis of peripheral nerve regeneration.

Marina Garcia Bejarano
- Degree: Master's Degree in Tissue Engineering and Advanced Therapies (University of Granada)
- Birthdate: 14/10/1996
- Phone: +39 011 6705436
- Email: marina.garciabejerano@unito.it
- Position: PhD Student, PhD in Neuroscience
- Role & Expertise: Biomolecular analysis of peripheral nerve regeneration; development of gold nanoparticles for in vitro and in vivo siRNA delivery.

Miriam Metafune
- Degree: Master's Degree in Molecular Biology (University of Parma)
- Birthdate: 18/12/1994
- Phone: +39 011 6703577
- Email: miriam.metafune@unito.it
- Position: PhD Student, PhD Program in Experimental Medicine and Therapy
- Role & Expertise: Study of the capability of biomaterials to promote peripheral nerve regeneration.

Davide Pellegrino
- Degree: Master's Degree in Cellular and Molecular Biology (University of Turin)
- Birthdate: 28/02/1998
- Phone: +39 011 6705436
- Email: da.pellegrino@unito.it
- Position: PhD Student, PhD Program in Experimental Medicine and Therapy
- Role & Expertise: In vitro and in vivo models for studying the role of gut microbiota in peripheral nerve regeneration.

Debora Molinaro
- Degree: Master's Degree in Cellular and Molecular Biology (University of Turin)
- Birthdate: 12/11/1991
- Phone: +39 011 6705436
- Email: debora.molinaro@edu.unito.it; debora.molinaro.to@gmail.com
- Position: Fellowship Recipient (concluded 28/02/2025)
- Role & Expertise: In vitro and in vivo models for peripheral nerve regeneration.

Francesca Bertone
- Degree: Master's Degree in Medical Biotechnology (University of Turin)
- Birthdate: 16/09/1997
- Phone: +39 011 6703577
- Email: francesca.bertone@unito.it
- Position: PhD Student, PhD Program in Experimental Medicine and Therapy
- Role & Expertise: In vitro models for nervous system regeneration, biochemical analysis of nervous tissue and behavioural tests on murine models.

Simona Rando
- Degree: Master's Degree in Molecular Biotechnology (University of Turin)
- Birthdate: 02/03/1999
- Phone: +39 011 6705433
- Email: simona.rando@unito.it
- Position: PhD Student, PhD Program in Experimental Medicine and Therapy
- Role & Expertise: In vitro hiPSC models for studying diabetic neuropathy.

Giuliana Paravizzini
- Degree: Master's Degree in Biomedical Engineering (University of Turin)
- Birthdate: 09/09/1998
- Phone: +39 011 6705433
- Email: giuliana.paravizzini@unito.it
- Position: Fellowship Recipient
- Role & Expertise: Application of bioengineering principles for the development of biomaterials for peripheral nerve regeneration.

Sajjad Farzin
- Degree: Master's Degree in Cellular and Molecular Biology (University of Turin)
- Birthdate: 27/10/1997
- Phone: +39 011 6705433
- Email: sajjad.farzin@edu.unito.it
- Position: Fellowship Recipient
- Role & Expertise: In vitro and in vivo models to study nerve regeneration under conditions of gut dysbiosis.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
12/2022 - 11/2026	D34Health	Stefania Raimondo	PNRR MUR/MH	Team component	325.895 Euro	UNITO
10/2023 - 09/2025	Development of nano/micro-engineered devices for applications in peripheral nervous system pathological models	Stefania Raimondo	PRIN-MUR	PI of research Unit	59.238 Euro	UNITO
10/2023 - 09/2025	Gut and NeuroMuscular	Giulia Ronchi	PRIN-MUR	Coordinator	106.345 Euro	UNITO

	system: investigating the impact of microbiota on nerve regeneration and muscle reinnervation after peripheral nerve injury (Gut-NeuroMuscle) 20227YB93W					
12/2023 - 11/2025	New insights into myelin maintenance and peripheral nerve regeneration: the role of Beclin1 (INNER-BECN1) P2022Y2A3L	Giulia Ronchi	PRIN-PNRR	PI of research Unit	107.130 Euro	UNITO
11/12/2025 11/12/2026	Studio dell'efficacia del condotto SilkBridge® nella riparazione di lesioni nervose con grave perdita di sostanza	Luisa Muratori	KLISBio S.r.l,	PI of research	50.000 Euro	UNITO

3. SCIENTIFIC ACTIVITIES IN 2025

Stefania Raimondo, (PI)

Supervised PhD students:	Federica Zen Miriam Metafune Simona Rando Alessia Nuzzi
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	- University of Granada, Spain, prof. Victor Sebastian Carriel - University of Hannover, Germany, prof Kirsten Haastert-Talini - University of Zaragoza, Spain, prof Jesus Ciriza - University of Porto, Portugal, prof. Ana Colette Maurício - Universidade de Lisboa, Portugal, Dr. André Luís Bombeiro -Kyushu Institute of Technology, Fukuoka, Japan, prof. Yuki Shirosaki - Acibadem University, Turkey, Prof. Mustafa Aktekin

• Invited talks:	
• Science communication:	- Apr–Jun 2025 – *Doctor for a Day*, medical orientation program – School of Medicine, Simulation Center, A.O.U. San Luigi Gonzaga.
• Editorial duties:	- Editorial Board Member of Frontiers in Neuroanatomy - Editorial Board Member of Biomedicines
• others	- President of ESPNR (The European Society for the Study of Peripheral Nerve Repair and Regeneration) - Member of "NANBIOSIS Scientific Advisory Board", Spain
Organizational activities and responsibilities at NICO:	
Speakers invited:	- António J. Salgado, Life and Health Sciences Research Institute (ICVS), School of Medicine, University of Minho, Braga, Portugal; ICVS/3Bs PT Associated Lab, Braga/Guimarães, Portugal, "Stem Cells Secretome in Central Nervous System Regenerative Medicine: Insights from Pre-Clinical Models of Injury and degeneration" 9/5/2025. - Paul Weiringa, Maastricht University and MERLN Institute for Technology-Inspired Regenerative Medicine, "The Peripheral Nervous System in Tissue Engineering: from Scaffold Design to Functional In Vitro Models" 13/6/2025.
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Giulia Ronchi, Associate Professor

Supervised PhD students:	Davide Pellegrino (PhD) Francesca Bertone (PhD)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	University of Hannover, Germany, prof Kirsten Haastert-Talini Faculdade de Medicina da Universidade de Lisboa, Dr. Andrè Bombeiro University of Porto, Portugal, prof. Ana Colette Maurício and Prof. Rui Damásio Alvites Karolinska Institutet, Solna, Sweden, Dr. Harald Lund
• Invited talks:	23rd May 2025 - 14th Workshop of Advanced Microscopy Techniques for Biomedical Applications– "*Transmission Electron Microscopy: A Powerful Lens into the Peripheral Nervous System*" 4th June 2025, DFG-Priority Programme "Codeχ"- SPP 2416 Chitin, chitosan and chito-oligosaccharides and their interaction with proteins of the extracellular matrix and cellular signalling. "*Chitosan-Based Scaffolds as a Promising Approach for Peripheral Nerve Regeneration*"
• Science communication:	Penne Amiche Della Scienza
• Editorial duties:	Youth Editorial Board Member di Neural Regeneration Research (NRR) Referee for Journal of Clinical Investigation, Antioxidants, International Journal of Molecular Sciences, Heliyoin
• others	Reviewer of a grant proposal for DGF - German Research Foundation

Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Giovanna Gambarotta, Associate Professor

Supervised PhD students:	Marina Garcia Bejarano (PhD in Neuroscience) in cotutelle with University of Zaragoza (prof Jesus M. de la Fuente) Federica Zen (PhD) (co-supervised with Stefania Raimondo)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	University of Zaragoza, Spain, prof Jesus M. de la Fuente University of Granada, Spain, prof Victor Sebastian Carriel University of Hannover, Germany, prof Kirsten Haastert-Talini
• Invited talks:	
• Science communication:	
• Editorial duties:	Editorial Board Member of Brain Sciences Associate Editor for Cellular Neuropathology (section Frontiers in Cellular Neuroscience) Referee for several journals.
• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	Italian patent for Industrial Invention number 102015000071499 entitled "*Conjugate of Neuregulin 1 for the treatment of peripheral nerve lesions*", release date: 18/04/2018, expiry: 11/11/2035. Inventors: Stefano Geuna, Abraham Shahar, Ofra Ziv-Polat, Giovanna Gambarotta, Federica Fregnan.

Federica Fregnan, Research technician

Supervised PhD students:	Miriam Metafune (PhD)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	- Kyushu Institute of Technology, Fukuoka, Japan, prof. Yuki Shirosaki - University of Hannover, Germany, prof Kirsten Haastert-Talini
• Invited talks:	
• Science communication:	- Apr–Jun 2025 – *Doctor for a Day*, medical orientation program – School of Medicine, Simulation Center, A.O.U. San Luigi Gonzaga. - Penne Amiche Della Scienza
• Editorial duties:	Reviewer for Scientific International Journal, MDPI

• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	Italian patent for Industrial Invention number 102015000071499 entitled "Conjugate of Neuregulin 1 for the treatment of peripheral nerve lesions", release date: 18/04/2018, expiry: 11/11/2035. Inventors: Stefano Geuna, Abraham Shahar, Ofra Ziv-Polat, Giovanna Gambarotta, Federica Fregnan.

Luisa Muratori, Research Assistant

Supervised PhD students:	Miriam Metafune (PhD) Simona Rando (PhD)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	University of Zaragoza, Spain, prof Jesus Ciriza
• Invited talks:	9th May 2025-7th Brunelli Hand Surgery Club, Faculty of Medicine and Surgery, University of Brescia, Italy. Talk entitled: "Tissue engineering in the reconstruction of the nerve gap".
• Science communication:	
• Editorial duties:	
• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	Member of the organizing committee of the XXXV CONGRESSO GISN 2025, Torino (28-29 Novembre 2025)
Technology transfer achievements (patents, etc.):	

4. Research activity in 2025

a. Summary

Research activities focus on elucidating biomolecular and biological mechanisms underlying peripheral nerve repair and regeneration, aiming to identify novel therapeutic strategies for severe somatic and autonomic nerve injuries. Recent efforts investigate the role of the microbiota in nerve regeneration, as well as extracellular matrix remodelling after nerve perfusion. In parallel, novel biomaterials have been tested in vitro and in vivo to evaluate their potential to promote nerve repair.

b. Background and rationale

Peripheral nerve injuries represent a significant clinical challenge, commonly resulting from traumatic events or iatrogenic interventions. Such injuries often result in partial or complete loss of motor and sensory functions, with consequences for patients' life quality. Although the peripheral nervous system (PNS) retains a remarkable intrinsic ability to regenerate throughout adulthood, spontaneous recovery following nerve injury is often incomplete. This limitation is particularly evident in cases of severe nerve damage and long-

gap injuries, where functional recovery is often unsatisfactory. The increasing incidence of peripheral nerve injuries, together with the growing number of patients undergoing nerve-related surgical interventions, has highlighted the necessity for the development of advanced therapeutic strategies. Consequently, peripheral nerve regeneration has become a major focus of biomedical and translational research, with the overarching aim of improving structural repair and, more importantly, functional recovery. In mild to moderate injuries, direct end-to-end nerve repair may be feasible; however, in severe cases, direct suturing is not possible. Under these conditions, nerve grafting is required to bridge the gap between the proximal and distal nerve stumps, providing a permissive pathway for axonal regeneration and eventual reinnervation of the original peripheral targets. Autologous nerve grafts, typically harvested from sensory nerves, are currently considered the gold standard due to their high efficacy and biocompatibility. Nevertheless, their clinical application is limited by several drawbacks, including the need for additional surgical procedures, donor site morbidity, and permanent sensory deficits at the harvest site. These limitations have driven the development of alternative grafting solutions, encompassing both biological and synthetic substitutes, some of which are already in clinical use. Despite these advances, the outcomes of nerve reconstruction procedures remain highly variable, and complete functional recovery is achieved only in a minority of patients. The success of peripheral nerve repair is influenced by multiple interrelated factors. These include the anatomical location and extension of the injury, the delay between injury and surgical intervention, and the capacity of denervated muscles to accept reinnervation and recover from atrophy. Additional critical determinants are the progressive decline in axonal regenerative potential following prolonged axotomy, the loss of Schwann cell support over time, patient age, and the presence of concomitant pathological conditions. Together, these factors contribute to the complexity of nerve regeneration and underscore the need for a multifaceted research approach. Research in this field is inherently interdisciplinary, integrating principles from neuroscience, cell and molecular biology, bioengineering, and clinical sciences. The primary objective is not only to elucidate the biological and biomolecular events that occur following nerve injury, but also to identify and optimize therapeutic strategies capable of enhancing axonal regeneration, supporting target reinnervation, and ultimately restoring motor and sensory function. Advancing knowledge in peripheral nerve regeneration is therefore essential to improve clinical outcomes and to develop more effective, patient-centred treatments for nerve injuries.

c. Objectives

The group aimed to elucidate biomolecular and biological mechanisms underlying peripheral nerve regeneration and to develop strategies to enhance functional recovery after peripheral nerve injury. These objectives were addressed through multiple complementary research activities, including: i) the analysis of the role of Beclin1 into myelin maintenance and peripheral nerve regeneration; ii) the investigation of the role of gut microbiota during nerve repair; iii) the evaluation of biomaterials as potential devices for nerve repair; iv) the assessment of allograft efficacy in the repair of nerve lesions associated with tissue loss; v) the exploration of novel therapeutic strategies to improve outcomes following injury to periprostatic nerve bundles; vi) the development of gold nanoparticle-conjugated siRNAs to investigate the role of specific molecular factors in nerve regeneration; vii) the establishment of an in vitro model based on human induced pluripotent stem cells (hiPSCs); viii) the comparison between delayed and immediate nerve repair.

d. Results

<u>i) Role of Beclin1 in myelin maintenance and peripheral nerve regeneration</u>

This study investigated the role of Beclin1, a key autophagy regulator, in adult peripheral nerve myelin maintenance. Peripheral nerves from tamoxifen-treated P0-creERT2:Becn1fl/fl mice were analysed at 4, 8, and 12 weeks post-induction. At 12 weeks, mutant mice showed significant alterations in axon and fibre diameters and reduced myelin thickness compared with controls. From week 8, Schwann cells displayed enlarged cytoplasm filled with vesicular structures. These findings demonstrate a critical role for Beclin1 in maintaining myelin integrity in the adult peripheral nervous system.

<u>ii) Role of gut microbiota during nerve repair</u>

To assess the impact of gut dysbiosis on nerve regeneration, mice received an antibiotic cocktail before and/or after nerve injury. The experimental groups were analyzed at 2, 7, and 28 days post-injury. Functional testing revealed delayed motor recovery at 10 days in mice treated with antibiotics before injury, while no

differences were observed at 28 days. Morphoquantitative analyses confirmed comparable regeneration among groups at later stages. RNA sequencing identified differentially expressed genes in antibiotic-treated mice, and further analyses are ongoing

iii) Studying biomaterials as nerve repair device
Innovative materials were evaluated for their ability to promote nerve regeneration. In particular in vitro analysis using glial and neuronal cell models were used to assess biocompatibility, glial cell proliferation, migration, adhesion and axonal elongation of sensory and motor neurons. In particular, Zein-derived scaffolds (to be used as conduit fillers or membranes), Chitosan70-CMC30-RGO electroconductive devices, PVDF piezoelectric membranes showed promising results for supporting peripheral nerve regeneration.

iv) Acellular allograft for nerve lesions with tissue loss
Porcine xenografts were used as a translational model to assess host immune responses following implantation in a rat model. Porcine-derived grafts represent a relevant platform for evaluating immunological compatibility and host–graft interactions, enabling the assessment of innate and adaptive immune responses and providing insights into immunogenicity to support the optimization of xenogeneic biomaterials for clinical use.

v) Studying new strategies to improve outcome after periprostatic nerve bundles after injury
Radical prostatectomy for prostate cancer resections results in erectile dysfunction due to damage of the periprostatic nerve bundles. Engineered nano/micro-structured active membranes in piezoelectric materials devices were applied *in vivo* to repair cavernous nerve injury on adult rats. The post recovery time was 60 days to evaluate the ability of the devices to allow autonomic axonal regeneration and functional recovery.

vi) Gold nanoparticle-conjugated siRNAs
Gold nanoparticle–conjugated siRNAs were developed to investigate the role of specific molecular factors involved in peripheral nerve regeneration.

vii) Development of an *in vitro* model using human induced pluripotent stem cells (hiPSC)
To develop a “human” peripheral nerve *in vitro* model, human induced pluripotent stem cells (hiPSC) were grown and stimulated to differentiate towards a motor and sensory neuron phenotype and towards Schwann cells. Currently, we successfully obtained hiPSC-derived motor and sensory neurons and hiPSC-derived Schwann cell, which we are analysing morphologically and for the expression of differentiation markers through quantitative Real time.

viii) Delayed versus immediate nerve repair
Transcriptomic and morphological analyses were performed on nerves regenerated within a conduit to characterize the different stages of regeneration and to assess the impact of delayed nerve repair on peripheral nerve regeneration.

e. Advancement in the field

Research activity significantly advanced the field of peripheral nerve regeneration by integrating innovative biomaterials, translational in vivo models, and human-relevant in vitro systems:

- Novel biomaterials supported Schwann cell proliferation, migration, and neurite outgrowth, confirming their potential as next-generation nerve repair devices.
- Piezoelectric membranes tested in vivo for the first time to promote autonomic nerve regeneration.
- Decellularized porcine nerve allografts enabled the analysis of host–graft immune interactions, supporting the optimization of xenogeneic biomaterials for clinical nerve reconstruction.
- hiPSC-derived neurons and glial cells provided predictive human-relevant models for studying nerve regeneration, neuropathies, and biomaterial-cell interactions.
- Beclin1 identified as a key regulator of myelin maintenance in adult Schwann cells, essential for long-term peripheral nerve integrity.
- Gut microbiota perturbation modulates functional recovery after peripheral nerve injury, revealing a novel gut–peripheral nervous system axis.

- First transcriptomics analysis of nerve regeneration within a conduit

f. Publications

- Fregnan F, Muratori L, Crosio A, Zen F, Fiori C, Tonazzini I, Gambarotta G, Martinelli A, Meziere J, Raimondo S, Manfredi M, Scaccini L, Geuna S, Porpiglia F. (2025). A novel rat model for studying bilateral cavernous nerve damage in vivo: exploring the potential of chitosan-blended membranes for nerve regeneration post-radical prostatectomy. Minerva Urol Nephrol. Feb77;(1)
  Research article Q1
- Feyles E, Felicetti T, Arduino I, Rittà M, Civra A, Muratori L, Raimondo S, Lembo D, Manfroni G, Donalisio M. (2025). Broad-Spectrum Antiviral Activity of Pyridobenzothiazolone Analogues Against Respiratory Viruses. Viruses. Jun 24; (7)
  Research article Q1
- Metafune M, Muratori L, Fregnan F, Ronchi G, Raimondo S. (2025). The extracellular matrix in peripheral nerve repair and regeneration: a narrative review of its role and therapeutic potential. Front Neuroanat. Oct 21;(19)
  Review Q1
- Mahmoudian M, Trotta F, Raimondo S, Bussolino F, Arese M. (2025). Cell Membrane-Integrated Neuroligin-1 Regulates the Anti-Inflammatory Effects of CRC Cell-Derived Exosomes. Int J Mol Sci. Jan 9; (2)
  Research article Q1
- Scaccini L, Escarda-Castro E, Seijas-Gamardo A, Wu H, Coimbra TS, Raimondo S, Fregnan F, Muratori L, Cecchini M, Moroni L, Wieringa P, Tonazzini I. (2025). Soft Scaffolds for Nerve Repair: Investigating Glycerol-Plasticized Chitosan Microstructures With In Vitro Complex Models. J Biomed Mater Res B Appl Biomater. Nov;113(11)
  Research article Q2
- Önger ME, Geuna S, Elhaj AE, Raimondo S, Kaplan S. (2025). Regeneration responses of Ankaferd Blood Stopper, platelet-rich-plasma, and *Momordica charantia*on sciatic nerve injury in the obese rats. Biomed Mater. May 23;(4)
  Research article Q2
- Alvites R, Lopes B, Coelho A, Sousa AC, Sousa P, Moreira A, Rêma A, Simões M, Mendonça C, Atayde L, Prada J, Pires I, Silva B, João F, Metafune M, Bertone F, Raimondo S, Alves N, Luís AL, Maurício AC. (2025). Combined use of olfactory mucosal mesenchymal stem cells conditioned medium and neural guide conduits promotes nerve regeneration in an ovine model. Front Cell Dev Biol. May 2;(13)
  Research article Q1
- Ronchi G, Pellegrino D, El Soury M, Amato O, Gaia F, Farzin S, Nuzzi R, Basic M, Bolsega S, Geuna S, Cescon M, Haastert-Talini K, Gambarotta G. (2025). Gut microbiota regulates optic nerve fiber myelination. Front Cell Dev Biol. Feb 20;(13)
  Research article Q1
- Lombardo-Hernandez J, Mansilla-Guardiola J, Aucello R, Botta C, García-Esteban MT, Murciano-Cespedosa A, Muñoz-Rodríguez D, Quarta E, Mateos González Á, Juan-Llamas C, Rantsiou K, Geuna S, Cocolin L, Herrera-Rincon C. (2025). An in vitro neurobacterial interface reveals direct modulation of neuronal function by gut bacteria. Sci Rep. Jul 15;(1)
  Research article Q1
- Quarta E, Bourqqia-Ramzi M, Muñoz-Rodriguez D, García-Esteban MT, Murciano-Cespedosa A, Mateos González Á, Conejero-Meca FJ, Lombardo-Hernandez J, Mansilla-Guardiola J, Baroni S, Geninatti Crich S, Geuna S, Munaron L, Chiabrando D, Herrera-Rincon C (2025). Impact of Iron Deficiency on the Growth and Bioelectrical Profile of Different Gut Bacteria. Microbiologyopen. Jun14;(3)
  Research article Q2
- Lopes B, Sousa P, de Sousa Moreira A, Sousa AC, Rêma A, Atayde L, Salgado AJ, Geuna S, Alvites R, Maurício AC. (2025). Neuronal Differentiation and Exosome Profiling of Dental Pulp Stem Cells: Unveiling Their Potential for Nerve Repair. Int J Mol Sci. Oct 6;(19)

Research article Q1

- Sousa P, Lopes B, Sousa AC, de Sousa Moreira A, Rêma A, Alvites R, Geuna S, Alves N, Maurício AC. (2025). Rat Hair Follicle Stem Cell-Derived Exosomes: Isolation, Characterization and Comparative Analysis of Their In Vitro Wound Healing Potential. Int J Mol Sci. May 25;(11):5081
  Research article Q1
- García-Bejarano M, Aucello R, Zen F, El Soury M, Cordero F, de la Fuente JM, Perroteau I, Ronchi G, Gambarotta G. (2025). Dynamic regulation of *Rgs16* and its correlation with Neuregulin1 expression in acute and chronic nerve injury. Front Cell Dev Biol. Mar 27;(13)
  Research article Q1
- Gambarotto L, Russo L, Bresolin S, Persano L, D'Amore R, Ronchi G, Zen F, Muratori L, Cani A, Negro S, Megighian A, Calabrò S, Braghetta P, Bizzotto D, Cescon M. (2025). Schwann Cell-Specific Ablation of Beclin 1 Impairs Myelination and Leads to Motor and Sensory Neuropathy in Mice. Adv Sci (Weinh). Feb;12(5)
  Research article Q1
- Ronchi G, Cescon M, Gambarotta G, Haastert-Talini K. (2025). Peripheral nervous system and gut microbiota: Emerging evidence on increased mechanistic understanding to reveal innovative strategies for peripheral nerve regeneration. Neural Regen Res. Apr 1;(4)
  Perspective article Q1
- Aucello R, Pernice S, Tortarolo D, Calogero RA, Herrera-Rincon C, Ronchi G, Geuna S, Cordero F, Lió P, Beccuti M. (2025). UnifiedGreatMod: a new holistic modelling paradigm for studying biological systems on a complete and harmonious scale. Bioinformatics. Mar 4;(3)
  Research article Q1
- Ronchi G, Ackva C, Fregnan F, Zen F, Muratori L, Crosio A, Metzen J, Haastert-Talini K. (2025). Chitosan-Based Materials for Peripheral Nerve Repair-New Pre-Clinical Data on Degradation Behavior at the Nerve Repair Site and Critical Opinion on Their Translational Impact. Int J Mol Sci. Jan 30;(3)
  Research article Q1
- Bartesaghi L, Giangreco B, Chiappini V, Veloz Castillo MF, Monaco M, Médard JJ, Gambarotta G, Agus M, Calì C. (2025). Tight Spaces, Tighter Signals: Spatial Constraints as Drivers of Peripheral Myelination. Cells. Jun 18;(12)
  Research article Q1
- Zucca G, Perucchini M, Vigani B, Valentino C, Patterlini V, Climani G, D'Angelo D, Sonvico F, Muratori L, Civra A, Lembo D, Ruggeri M, Sandri G, Rossi S. (2025). Development of a nasal spray based on cyclodextrin/hydrophobically-modified hydroxypropyl-methyl cellulose for the prevention of viral respiratory infections. Int J Biol Macromol. Apr; 299
  Research article Q1

5. Future directions and objectives for next years

a. Summary

The research group plans to expand its focus on emerging topics that advance fundamental and translational discoveries in peripheral nerve regeneration, with the goal of developing innovative therapies to improve patient outcomes following somatic and autonomic peripheral nerve injuries. Specifically, among the most innovative objectives, sildenafil-conjugated nanomicelles will be tested in vitro on glial cells and neurons to evaluate drug release profiles and their biological effects on cellular responses relevant to nerve regeneration. The analysis of innovative biomaterials will continue with further in vitro experiments and in vivo assessment for the most promising. In particular, we will work on the functionalization of the promising scaffolds with topographical cues or with molecules that can stimulate the nerve regeneration (such as PDEi). Ongoing studies will further investigate the impact of dysbiosis on peripheral nerve regeneration following nerve injury, with the aim of elucidating how microbiota imbalance influences regenerative processes and functional recovery. At the same time, neurons and glial cells will be used to develop an *in vitro* model of diabetic neuropathy and to investigate the mechanisms underlying the onset of diabetic neuropathy. Schwann cells, motor neurons and/or sensitive neurons derived from human induced pluripotent stem cells (hiPSC)

will be also used with this aim. Lastly, the biological and molecular mechanisms underlying nerve regeneration will be further investigated to expand current knowledge in the field.

b. Background and Significance

Development of single-component nanomicelles and binary nanomicelles for the controlled release of neuroprotective and neuroregenerative drugs

In order to improve the outcome of nerve regeneration, this project aims to develop and validate an innovative biodegradable polymeric scaffold integrated with single-component nanomicelles (NM) and binary nanomicelles (BNM) for the controlled release of neuroprotective and neuroregenerative molecule

The scaffold will act as a temporary extracellular matrix, providing structural support for axonal regrowth, and creating a favorable microenvironment for Schwann cell migration and axonal elongation while the integrated nanomicelles will enable the sustained delivery of bioactive factors essential for peripheral nerve repair. The approach relies on biocompatible and biodegradable materials to ensure safety and seamless integration with host tissue. The ultimate goal is to deliver an effective, scalable therapeutic platform that overcomes the limitations of traditional approaches and significantly improves clinical outcomes.

Studying biomaterials as possible nerve repair device

Innovative materials have been assessed *in vitro* for their ability to promote nerve regeneration: Zein-derived scaffolds (to be used as conduit fillers or membranes), Chitosan70-CMC30-RGO electroconductive devices, PVDF piezoelectric membranes. Preliminary experiments demonstrated their potential use for repairing somatic and autonomic lesions. Therefore, future experiments will be focused on further improve their efficacy testing them both in advanced in vitro models and in vivo, on somatic and autonomic nervous system.

The Role of Dysbiosis in Peripheral Nerve Regeneration

Future studies on the role of dysbiosis in peripheral nerve regeneration will be essential to elucidate the mechanisms through which the gut microbiota influences nerve repair. In particular, it will be important to identify specific microbiota-derived metabolites that impact peripheral nervous system function and regeneration, and to define how dysbiosis-driven modulation of the immune system affects the inflammatory and regenerative responses following nerve injury. A deeper understanding of these gut–immune–nerve interactions may reveal novel therapeutic targets to enhance peripheral nerve repair and functional recovery.

Role of Beclin 1 into myelin maintenance and peripheral nerve regeneration

Given its essential role in adult myelin maintenance, future work will address the function of Beclin1 during peripheral nerve injury and regeneration. In particular, we will investigate how Beclin1 regulates Schwann cell plasticity, myelin remodeling, and axon–glia interactions following nerve damage, as well as its impact on functional recovery. Defining the contribution of Beclin1-dependent pathways to nerve repair will provide new mechanistic insight into peripheral nerve regeneration and may identify novel therapeutic targets to enhance myelin repair and regenerative outcomes.

Development of an in vitro model using human induced pluripotent stem cells (hiPSC)

Schwann cells, motor neurons and/or sensitive neurons derived from human induced pluripotent stem cells (hiPSC) will be used for developing an in vitro model for studying nerve regeneration and neuropathies (such as diabetic neuropathies). Primary cultures of neurons and glial cells will be used as model for evaluating culture conditions that reproduce the diabetic neuropathy (hyperglycemia and hypoxia).

c. General aim and integration with mission of the Institute

The group’s primary objective is to develop innovative strategies to enhance functional recovery following traumatic and iatrogenic nerve injuries. Nerve damage constitutes a major cause of neurological disability, with significant consequences for patients’ quality of life, including psychosocial and relational aspects. Addressing this clinical challenge requires a coordinated, interdisciplinary effort integrating expertise in both central and peripheral nervous system research, in accordance with the mission of NICO.

d. Specific objectives and strategies

Nanomicelle-Based Strategy for In Vivo Nerve Regeneration
Based on in vitro findings, future studies will focus on in vivo validation of the developed system. Nanomicelles (NM and BNM) will be incorporated into a functionalized membrane and tested in a rat model of cavernous nerve injury. The membrane will be applied at the lesion site to provide structural support and enable the sustained release of sildenafil, a neuroprotective and pro-regenerative agent.

Studying the relationship between microbiota and peripheral nerve regeneration
Future research will be focused on elucidating the specific roles of the gut microbiota composition and metabolites, as well as the signaling pathways through which they may influence nerve degeneration and regeneration.

Role of Beclin1 into myelin maintenance and peripheral nerve regeneration
Currently, we are investigating the *in vivo* impact of Beclin1-regulated processes on myelin maintenance, nerve regeneration, and skeletal muscle innervation using an inducible Schwann cell-specific Beclin1 knockout mouse model, where the gene is deleted in response to tamoxifen.

Studying biomaterials as possible nerve repair device
The analysis of the efficacy of the innovative materials will continue with further advanced *in vitro* experiments and *in vivo* for the most promising. In particular, we will work on the functionalization of the promising scaffolds with topographical cues or with molecules that can stimulate the nerve regeneration (such as PDEi). The in vitro analysis will be performed using neural and glial cell models, co-cultures, and organotypic cultures, in vivo analysis will be carried out on both somatic and autonomic nervous system.

Development of an *in vitro* model using human induced pluripotent stem cells (hiPSC)
An in vitro model of diabetic neuropathy will be developed to investigate the molecular and cellular mechanisms underlying the onset and progression of the disease, as well as to evaluate potential therapeutic intervention strategies. Primary cultures of rat neurons and glial cells will be employed to establish culture conditions that closely mimic diabetic pathology. These conditions will subsequently be applied to differentiated human induced pluripotent stem cells (hiPSCs) in order to generate a human-relevant in vitro model of diabetic neuropathy.

e. Unique features of the project research

The project research is internationally recognized for its innovative contributions to peripheral nerve repair and regeneration. The research activity addresses key biological and translational challenges associated with peripheral nervous system regeneration through the systematic integration of *in vitro* and *in vivo* experimental models. This combined approach enables the investigation of molecular, cellular, and functional mechanisms of nerve injury and repair, while supporting the validation of experimental findings in clinically relevant contexts.
The research project is characterized by a strong interdisciplinary framework that integrates complementary expertise in neurobiology, biomaterials science, bioengineering, immunology, and translational neuroscience. This multidisciplinary organization allows for a comprehensive analysis of peripheral nerve regeneration processes and supports the development of innovative experimental strategies. The project further benefits from established collaborations with multiple European and international research groups, facilitating scientific exchange, methodological standardization, and access to specialized infrastructures and technologies.
A defining feature of the project research is its strong translational focus. Research outcomes are evaluated for their applicability to the development of novel therapeutic strategies aimed at improving functional recovery following peripheral nerve injuries. By bridging basic research and preclinical validation, the project contributes to the generation of robust and reproducible evidence with clear potential for successful clinical translation, in alignment with current priorities in regenerative medicine and peripheral nerve repair.

f. Methodology

The most innovative aspects of our research will be:

1. The use of hiPSC as *in vitro* model for the study of peripheral nerve
2. The extensive use of computational biology/bioinformatics techniques
3. Thanks to the acquisition of a fluorescence microscope equipped for time-lapse imaging, together with the availability of expression vectors encoding fluorescent proteins that label autophagosomes and lysosomes, it will be possible to monitor auto-phagolysosome formation under different experimental conditions, in particular in the presence or absence of Beclin1.
4. Thanks to the establishment of a metabolomics facility within the department of clinical and biological sciences, adjacent to the NICO, it will be possible to analyze the different metabolites present in the serum of animals with a normal microbiota or with dysbiosis.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Ageing and Alzheimer's disease

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Elena Tamagno
- Degree: PhD
- Birthdate: 14 July 1967
- Nationality: Italian
- Gender: Female
- Phone +39116707764
- E-mail: elena.tamagno@unito.it
- Position: Full Professor
- Role & expertise: Pathogenesis of Alzheimer's disease

Personnel

Michela Guglielmotto
- Degree: PhD
- Birthdate: 28/02/1977
- Nationality: Italian
- Gender: Female
- Phone: +390116707704
- Email: michela.guglielmotto@unito.it
- Position: Associate Professor
- Role & expertise: Pathogenesis of Alzheimer's disease

Giulia Morellato
- Degree: PhD student
- Birthdate: 05/12/1998
- Nationality: Italian
- Gender: Female
- Phone: +390116707732
- Email: g.morello@unito.it
- Position: PhD student
- Role & expertise: Pathogenesis of Alzheimer's disease

Elisa Porchietto
- Degree: PhD
- Birthdate: 26/08/1997
- Nationality: Italian
- Gender: Female
- Phone: +390116707732
- Email: elisa.porchietto@unito.it
- Position: Post doctoral researcher
- Role & expertise: Pathogenesis of Alzheimer's disease and molecular biology

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Giulia Cicilese
- Degree: PhD student
- Birthdate: 18/02/1998
- Nationality: Italian
- Gender: Female
- Phone: +390116707732
- Email: giulia.cicilese@unito.it
- Position: Biotechnologist, CRT Foundation research fellow
- Role & expertise: Pathogenesis of Alzheimer's disease and molecular biology

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
18/10/23 17/10/25	Extracellular vesicles derived from preconditioned Mesenchymal stem cell: a new challenge for AD therapy? Prot. 202229X8HW	Elena Tamagno	PRIN 2022	PI	67.911	UNITO
22/12/2023 30/06/2026	106831 - (2023.1759) - 'Studio di marcatori plasmatici precoci della malattia di Alzheimer: ruolo del miRNA218	Michela Guglielmotto	CRT Bando Richieste Ordinarie 2023	PI	30.000	UNITO

3. SCIENTIFIC ACTIVITIES IN 2025

Elena Tamagno, Role (PI)

Supervised PhD students:	Giulia Morello
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	Thomas Fenlz, Associate Professor, Clinic for Anesthesiology and Intensive Care TUM School of Medicine and Health Technical University of Munich; Gerhard Rammes, Associate Professor, Clinic for Anesthesiology and Intensive Care TUM School of Medicine and Health Technical University of Munich; George Tetz, MD, PhD, Department of Systems Biology Human Microbiology Institute, NY.
• Invited talks:	

• Science communication:	
• Editorial duties:	
• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Supervised PhD students:	Gulia Cicilese (November 2025)
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	Thomas Fenlz, Associate Professor, Clinic for Anesthesiology and Intensive Care TUM School of Medicine and Health Technical University of Munich; Gerhard Rammes, Associate Professor, Clinic for Anesthesiology and Intensive Care TUM School of Medicine and Health Technical University of Munich; George Tetz, MD, PhD, Department of Systems Biology Human Microbiology Institute, NY.
• Invited talks:	
• Science communication:	
• Editorial duties:	Associate Editor in Molecular Diagnostics and Therapeutics of Frontiers in Molecular Biosciences
• others	
Organizational activities and responsibilities at NICO:	Responsible for the Cell Culture room (floor 0) Responsible for the dissection room (floor -1) Responsible for Western Blot Lab (floor 0) Responsible for Western Blot /Bacteria Lab (floor -1)
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

4. Research activity in 2025

a. Summary

Alzheimer's disease (AD) is a progressive neurodegenerative disorder affecting millions of people worldwide, characterised by the accumulation of Aβ, hyperphosphorylation of the tau protein, neuroinflammation and synaptic loss. Given the lack of effective treatments, new approaches are needed. Extracellular vesicles derived from mesenchymal stem cells exhibit immunomodulatory and neuroprotective potential, similar to GHRH and its agonist MR-409. Furthermore, scientific evidence has been obtained to support the use of miRNA-218 as a molecular biomarker.

b. Background and rationale

Alzheimer's disease (AD) is the most common form of neurodegenerative dementia, accounting for approximately 70–80% of dementia cases worldwide. It is characterized by a progressive decline in cognitive and executive functions and begins years before the appearance of clinical symptoms. Although disease-modifying treatments, including monoclonal antibodies targeting amyloid accumulation, represent an important advance, their efficacy remains limited, highlighting the need for innovative therapeutic strategies targeting multiple pathogenic mechanisms.

Mesenchymal stem cells (MSCs) and their extracellular vesicles (MSC-EVs) have emerged as promising tools against inflammation and neuronal degeneration. Increasing evidence suggests that many of the protective effects of MSCs are mainly mediated by secreted soluble factors, cytokines and, particularly, EVs carrying immunomodulatory and neuroprotective molecules. Several preclinical studies, including our own in AD models, have demonstrated the therapeutic potential of MSC-EVs. Moreover, MSC preconditioning with physical, chemical, biological or genetic stimuli can enhance the therapeutic properties of both MSCs and their EVs. However, a systematic characterization of EV cargo following different preconditioning protocols is still lacking.

Recently, research into microRNAs (miRNAs) has taken on particular significance in relation to their possible role in the pathogenesis of numerous diseases. These small non-coding RNAs regulate gene expression and are involved in cellular stress responses, synaptic plasticity, neuronal differentiation and neurodegeneration. Several miRNAs are differentially expressed in AD and may represent minimally invasive biomarkers capable of detecting disease-related changes before symptom onset. Particular attention will be given to miR-218, a CNS-enriched miRNA implicated in neurodegenerative disorders and potentially involved in tau phosphorylation through estrogen receptor-dependent mechanisms. ERα may increase miR-218 expression, reducing PTPα and promoting GSK3β activation and PP2A inhibition, thereby favoring tau hyperphosphorylation, whereas ERβ may exert opposite effects.

To assess its clinical relevance, miR-218 levels were measured by Real-Time RT-PCR in CSF and plasma from AD and MCI patients and controls, considering sex and disease stage. miR-218 was increased in MCI and AD patients of both genders. These findings support further investigation of miR-218 as a potential biomarker and therapeutic target.

A complementary therapeutic approach involves growth hormone-releasing hormone (GHRH), a hypothalamic peptide with extra-pituitary anti-inflammatory and neuroprotective effects mediated by GHRH receptors. GHRH and its agonist MR-409 have shown protective effects in neurological disease models. Our preliminary in vitro data demonstrate that GHRH(1-44)NH_2 attenuates Aβ(1-42)-induced toxicity in neural stem cells while promoting neurogenesis and neuronal differentiation. Together, these findings support the investigation of MSC-EVs, miR-218 and GHRH/MR-409 as complementary strategies to target key mechanisms of AD pathogenesis.

c. Objectives

The objectives for each project are:

- to characterize the protective action of preconditioned MSC-EVs relatively to amyloid precursor protein (APP) processing and amyloid β (Aβ) formation, microtubule associated protein tau (MAPT) phosphorylation and aggregation, inflammation and activation of glial cells, the crucial hallmarks of AD;
- to establish a correlation between miRNA-218 expression and the pathogenesis of AD, and to identify a new biomarker already present in the plasma of MCI patients, with a view to developing a potential therapeutic approach to slow the progression of the disease;
- to investigate the potential protective effect of MR-409 in the mouse model of Alzheimer's disease, the 5xFAD transgenic mouse.

d. Results

The results obtained support the therapeutic potential of extracellular vesicles (EVs) derived from mesenchymal stem cells (MSC-EVs) and of growth hormone-releasing hormone (GHRH) signaling in experimental Alzheimer's disease (AD), while providing new evidence on the role of miRNA-218 as a potential biomarker of disease progression.
Two types of MSC-EVs, obtained using different preconditioning protocols, were administered intranasally to 3-month-old male 5xFAD mice. This route was selected to bypass the blood-brain barrier and facilitate EV delivery to target brain regions. Both EV preparations reduced astrogliosis not only in anterior brain areas but also in the hippocampus and cortex, regions critically involved in AD pathology. A concomitant reduction in amyloid plaque density was observed in the same areas, with a more pronounced effect following treatment with EVs derived from oxygen and glucose deprivation (OGD)-preconditioned MSCs. These findings indicate that MSC-EVs can reach relevant brain regions and modulate important pathological features of AD, although the possibility that different EV preparations exert region- or target-specific effects requires further investigation. Despite these histological effects, treated 5xFAD mice did not show significant behavioural improvement. This result may be explained by the severe phenotype of this model, in which extensive amyloid deposition begins as early as two months of age and may induce irreversible alterations in cognitive and behavioural functions before treatment. Conversely, wild-type mice receiving the same EV treatments displayed reduced anxiety and improved object discrimination, indicating that EVs can reach brain regions involved in memory and positively influence behavioural performance. Similar results were obtained in hTau mice, which showed higher discrimination index scores following EV treatment compared with PBS-treated controls.
Furthermore, the characterization of the newly generated 5xTg-AD/hTauTg mouse model provided important information on the temporal relationship between amyloid and tau pathology. Both major histological markers of AD became detectable from approximately five months of age, whereas at three months animals exhibited mainly extracellular β-amyloid deposits. This temporal characterization identified an appropriate experimental window for investigating molecular changes associated with disease progression and allowed the relationship between endogenous amyloid production and pathological tau alterations to be investigated. Attention was devoted to miRNA-218. Plasma levels were analyzed in female 5xTg-AD/hTauTg mice aged 2, 5, and 8 months under comparable estrous conditions. miRNA-218 levels progressively increased with age and were significantly higher in 8-month-old compared with 2-month-old mice. In contrast, no significant age-dependent changes were detected in female wild-type or 5xFAD mice, and previous analyses in hTau mice at two months showed no significant alterations in brain miRNA-218 levels following Aβ1-42 exposure. These findings suggest that miRNA-218 modulation is associated not only with estrogen-dependent mechanisms but also with disease progression and the accumulation of pathological features, including senile plaques and neurofibrillary tangles. Thus, miRNA-218 may represent a promising biomarker and potential therapeutic target in AD.
The study demonstrated neurogenic and neuroprotective effects of GHRH and its agonist MR-409. In vitro, GHRH(1-44)NH_2 promoted survival, proliferation, and neuronal differentiation of rat neural stem cells and human neuroblastoma cells. In neuron-like cells exposed to Aβ, GHRH preserved synaptic integrity and cholinergic function by preventing reductions in synaptic markers and BDNF and attenuating the Aβ-induced increase in acetylcholinesterase activity. In vivo, MR-409 treatment of 5xFAD mice reduced amyloid-β accumulation, tau phosphorylation, neuroinflammation, neuronal and synaptic loss, and improved cognitive performance. Overall, these findings indicate that GHRH signaling acts on multiple pathological mechanisms underlying AD and may therefore represent a promising therapeutic approach.
The main limitations include the use of the 5xFAD model, which predominantly reflects amyloid-driven pathology and therefore has limited direct applicability to sporadic AD and primary tauopathies, and the exclusive use of male mice in the EV and MR-409 studies, which may have prevented the identification of sex-specific differences. Future studies including both sexes and complementary AD models will be essential to validate these findings and clarify their translational relevance.

e. Advancement in the field

Despite extensive research and substantial economic investment, Alzheimer's disease (AD) remains incurable, partly due to the complexity of its pathological mechanisms and the high failure rate of drug development. The limitations of the traditional "one drug–one target" approach highlight the need for multifunctional strategies capable of simultaneously modulating different disease pathways. Our research

addresses this challenge by investigating mechanisms underlying AD pathology, identifying predictive plasma biomarkers for early diagnosis, and exploring environmental and behavioural factors contributing to disease onset and progression.

5. Future directions and objectives for next years

a. Summary

Professional collaboration with Gabriella Testa, Associate Professor of General Pathology at the University of Turin and a winner of the Airalzh Grant for Young Researcher 2024.
Title: A study of the ability of hydroxytyrosol in olive oil to prevent the hyperphosphorylation of tau protein in Alzheimer's disease
The proposed research project aims to demonstrate that the administration of extra virgin olive oil (EVOO) enriched with hydroxytyrosol (HXT) to mice expressing human tau (hTau) can prevent the accumulation of hyperphosphorylated tau and counteracting the formation of neurofibrillary tangles (NFTs) in Alzheimer's disease (AD).

b. Background and Significance

Alzheimer's disease (AD) is characterised by the intracellular accumulation of neurofibrillary tangles (NFTs) consisting of hyperphosphorylated tau protein and by the deposition of extracellular beta-amyloid (Aβ) plaques. Experimental evidence points to the very early onset of tauopathy and its key role in mediating Aβ toxicity in AD. Although tau's involvement in the pathogenesis of AD is well established, the mechanisms leading to its accumulation are not yet fully understood. A dysfunction in the mechanisms responsible for tau degradation, such as the ubiquitin-proteasome system (UPS) and the lysosomal autophagic pathway (ALP), could lead to the accumulation of insoluble NFTs at the neuronal level [3–4], which, by impairing axonal transport and neuronal function, result in neurodegeneration in AD. The use of a nutraceutical approach, based on the use of bioactive compounds found in food, is proving increasingly promising for the treatment of neurodegenerative diseases. In particular, numerous studies report the beneficial effects of dietary polyphenols in the treatment of AD, highlighting their potential application in disease prevention. A growing body of experimental evidence demonstrates that the Mediterranean diet reduces the risk of developing AD.

c. General aim and integration with mission of the Institute

Hydroxytyrosol (HXT), one of the most potent polyphenols found in extra virgin olive oil (EVOO), has recently been identified as a bioactive molecule with beneficial effects in AD: it is capable of improving cognitive function , counteracting Aβ-induced toxicity and preventing tau aggregation in vitro. However, to date, there is no evidence available regarding HXT's ability to counteract the accumulation of hyperphosphorylated tau protein.

d. Specific objectives and strategies

Following oral administration of HXT via gavage, a pharmacokinetic analysis will be carried out and the main biochemical parameters in the blood, as well as the cognitive abilities of the mice, will be assessed (Objective 1). The biodistribution of HXT in the brain and its ability to prevent the intraneuronal accumulation of hyperphosphorylated tau protein in hTau mice, in which tau phosphorylation has been induced by okadaic acid (OKA) via ICV, will be analysed (Objective 2). The molecular mechanisms involved in the protective effect of HXT will be investigated in depth, in particular the mechanisms of tau degradation via the ubiquitin-proteasome system (UPS) and the lysosomal autophagy pathway (ALP), both of which are regulated by the neuroprotective enzyme sirtuin 1, which we have already shown to be induced by HXT in vitro (Objective 3).

e. Unique features of the project research

Thanks to its ability to penetrate brain tissue and its non-cytotoxic nature, hydroxytyrosol (HXT) is considered a potential nutraceutical for the prevention of AD. It has also been observed that the use of extra virgin olive oil as a carrier promotes the absorption of HXT in the gut and its bioavailability in various tissues of the body, including the brain. Despite the recognised neuroprotective action of HXT against Aβ-induced toxicity, no studies are available on its effect in counteracting tauopathy. We have demonstrated, in neuronal cells, that HXT is a potent activator of the neuroprotective deacetylase sirtuin 1 (SIRT-1) and that the activation of SIRT-1 by the cerebrosterol 24-hydroxycholesterol (24-OHC) promotes the degradation of tau via proteasomal activation. Recent studies have also revealed that HXT, through the induction of SIRT-1, is capable of promoting autophagy. However, to date, the effect of HXT on tau degradation via the UPS and/or ALP remains unexplored. The study of how HXT, delivered via extra virgin olive oil (EVOO), activates tau degradation systems shows great promise for the development of a preventive strategy to avoid the accumulation of hyperphosphorylated tau and counteract the formation of GNF, which characterise AD.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Neurophysiology of neurodegenerative diseases

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Filippo Tempia
- Degree: MD, PhD
- Birthdate: 20/08/1960
- Phone: +39 011 6706609
- Email: filippo.tempia@unito.it
- Position: Full Professor
- Role: Principal Investigator

Personnel

Eriola Hoxha
- Degree: PhD
- Birthdate: 26/01/1981
- Phone: +39 011 6706609
- Email: eriola.hoxha@unito.it
- Position: Associate Professor
- Role & Expertise: Supervision of research activities; patch-clamp electrophysiology, molecular biology and neuroinflammation.

Francesca Montarolo
- Degree: PhD
- Birthdate: 14/05/1983
- Phone: +39 011 6706609
- Email: francesca.montarolo@unito.it
- Position: Researcher Tenure Track (RTT) in Physiology
- Role & Expertise: Behavioral experiments, histology and molecular biology.

Anita Maria Rominto
- Degree: MS
- Birthdate: 19/07/1996
- Phone: +39 011 6706609
- Email: anitamaria.rominto@unito.it
- Position: PhD Student
- Role & Expertise: Behavioral experiments, histology, molecular biology and patch-clamp

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Luna Berrino
- Degree: MS
- Birthdate: 19/09/1997
- Phone: +39 011 6706609
- Email: luna.berrino@unito.it
- Position: Research Fellow
- Role & Expertise: Behavioral experiments, histology, molecular biology and patch-clamp

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Matilde Loddo
- Degree: MS
- Birthdate: 19/01/2002
- Phone: +39 011 6706609
- Email: matildeloddo7@gmail.com
- Position: Research Fellow (from 03/10/2025)
- Role & Expertise: Behavioral experiments, histology, molecular biology and patch-clamp.

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
01/08/2024-31/07/2026 (extended to 24months)	Collaboration related to the link between the role of NURR1 in physiological and pathological conditions and the effects of various Immunic compounds	Francesca Montarolo	Immunic Therapeutics	PI	250.000 Euro	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Filippo Tempia, PI

Supervised PhD students:	1: Anita Maria Rominto
Outreach activities	
• International collaborations:	Prof. Fernanda Laezza, University of Texas Medical Branch, USA Guomei Tang, Columbia University Irving Medical Center, New York, USA
• Invited talks:	na
• Science communication:	na
• Editorial duties:	Associate Editor of Frontiers in Aging Neuroscience, Frontiers in Synaptic Neuroscience, Frontiers in Dementia, Journal of Neuroscience and Rehabilitation, International Journal of Brain Science, The American Journal of Alzheimer's Disease
• Others	
Organizational activities and responsibilities at NICO:	Group Leader of Neurophysiology of Neurodegenerative Diseases; Director of the NICO Animal Facility
Speakers invited:	na
Other organizational activities:[7]	na
Workshops, Schools or Conferences organized:	Chair of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025.
Technology transfer achievements (patents, etc.):	na

Eriola Hoxha, Supervisor and Researcher

Supervised PhD students:	1: Anita Maria Rominto
Outreach activities	
• International collaborations:	-Prof. Shan Zha, (Dept. of Pathology and Cell Biology), Columbia University, New York, USA. -Prof. Jeanne Nerbonne, (Dept. of Developmental Biology) Washington University School of Medicine, St. Louis, MO, USA.

	-Prof. Dorota Skowronska-Krawczyk, PhD (Dept of Physiology and Biophysics Dept of Ophthalmology, Center for Translational Vision Research School of Medicine UC Irvine, USA. -Prof. Maximiliano Jose Nigro, (Dept. of Language and Literature), Kavli Institute for Systems Neuroscience, NTNU, Trondheim, NO -Prof. Magdalena Dziembowska (Faculty of Biology) University of Warsaw, Poland.
• Invited talks:	*Early cerebellar circuitry alterations in a murine model of Ataxia-Telangiectasia*. 21st SINS National Congress, September 10th-13th 2025, Pisa, Italy in the frame of the symposium: "Models and approaches to tackle neurodevelopmental disorders heterogeneity".
• Science communication:	Invited speaker at the Istituto di Istruzione superiore "Balbo-Palli", Casale Monferrato, Italy, November 11th, 2025. Seminars covering the fundamentals of biomineralization, brain mineralogy, and lipid-associated neurodegeneration.
• Editorial duties:	Editor (Handling Editor), *Frontiers in Aging Neuroscience.* Editorial Board Member, *Frontiers in Cellular and Molecular Mechanisms of Brain Aging.* Associate Editor, *Behavioral and Brain Functions.*
• others	Referee for the grant agency National Science Center Poland (NCN) (PRELUDIUM 24). Referee for research grant proposals submitted to the *Fondation Maladies Rares* (France), within the "AnDDI-Rares 2025" call. The foundation supports research on rare diseases in France. Member of the Italian Society of Neuroscience (SINS), of the Italian Society of Physiology (SIF), and of the European Brain and Behaviour Society (EBBS)
Organizational activities and responsibilities at NICO:	Responsible for the water ultrapurification systems at NICO
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Chair of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025.
Technology transfer achievements (patents, etc.):	na

Francesca Montarolo, Researcher Tenure Track (RTT)

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
• International collaborations:	na
• Invited talks:	na
• Science communication:	Invited speaker at the Liceo Scientifico Statale "Alessandro Volta" (classe 4Cs, Liceo delle Scienze Applicate, disciplina Scienze

	naturali – PTOF, referente Prof. Silvia Gillo), Turin, Italy, 8 may 2025.
• Editorial duties:	na
• Others[6]	Poster presenting author at the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025. Title: Neurodegeneration and white matter defects in a neuron-specific Atm kinase-dead murine model of Ataxia-Telangiectasia Referee for the FISM (Federazione Italiana Sclerosi Multipla) grant. Member of the Italian Society of Neuroscience (SINS) and of the Italian Society of Physiology (SIF)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	Local Organizer of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025.
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Anita Rominto, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
• International collaborations:	Research stay of 3 months at the Kavli Institute for Systems Neuroscience in Trondheim, Norway, in the lab of professor Nigro
• Invited talks:	Selected for oral communication at the PhD Meeting of the Italian Society for Neuroscience (SINS), Pisa, Italy, 10 September 2025. Title: Chronic Social Defeat Stress reduces prefrontal cortex excitability in mice. Poster presenting author at the 21st National Congress of the Italian Society for Neuroscience (SINS), Pisa, Italy, 10-13 September 2025. Title: Chronic Social Defeat Stress reduces prefrontal cortex excitability in mice. Poster presenting author at the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025. Title: Depression in mice causes decreased neuronal excitability and enhanced frequency adaptation in medial prefrontal cortex pyramidal neurons. Poster presenting author at the 8th BraYn meeting, 29-31 October 2025. Title: Decreased neuronal excitability and enhanced frequency adaptation in medial prefrontal cortex pyramidal neurons of depressed mice.

• Science communication:	na
• Editorial duties:	na
• Others	Operational Staff- Local Organizing Team of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Luna Berrino, research fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
• International collaborations:	na
• Invited talks:	na
• Science communication:	na
• Editorial duties:	na
• Others	na
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	Operational Staff- Local Organizing Team of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025.
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Matilde Loddo, research fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
• International collaborations:	na
• Invited talks:	na
• Science communication:	na
• Editorial duties:	na
• Others	na
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:[7]	Operational Staff- Local Organizing Team of the 75th SIF National Congress - The Italian Society of Physiology and Joint Meeting of

	Italian-Slovenian Physiological Societies, Turin, Italy, 17-19 September 2025.
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

4. Research activity in 2025

a. Summary

In an animal model of AT, we found a deficit in action potential conduction. We found that the enzyme Elovl5 is necessary for proper action potential conduction in the axon. In an animal model of depression, in the medial prefrontal cortex we found a deficit in action potential generation, due to a more pronounced frequency adaptation. We are studying the effect of the NURR1 modulation in the control of neuroinflammation.

b. Background and rationale

1. Ataxia-teleangiectasia (AT) is an autosomal recessive disorder caused by loss of function of the ATM kinase and is characterized by an early onset, progressive cerebellar ataxia, oculocutaneous telangiectasia, immunodeficiency, pulmonary disease and increased risk of developing cancer. Histological studies from autoptic brain material revealed an important degeneration of Purkinje cells (PCs) with a compromised cerebellar structure. The Atm protein modulates the correct presynaptic vesicle release at glutamatergic synapses and controls GABAergic tone during development. Transcriptomics on AT cerebellum in early asymptomatic phase showed a possible compromised cerebellar glutamatergic signaling paralleled by a deranged calcium homeostasis, which might be involved in the mechanism of PCs death and ataxia.

2. Action potential conduction in the axon must take place at high velocity. This is particularly important in the cerebellum, in which timing of motor control is crucial. Elovl5 deletion in mice or mutation in humans causes cerebellar motor deficits. Our hypothesis is that Elovl5-dependent PUFAs are essential to preserve the correct velocity of action potential conduction along the axons of Purkinje cells.

3. The molecular mechanisms of depression are not clearly understood, and antidepressant drugs have a low rate of efficacy. The development of more effective drugs is hindered by the lack of knowledge of the neuronal alteration present in depression. The medial prefrontal cortex (mPFC) is a main hub controlling the circuit involved in stress and depression. Moreover, functional alterations of neuronal electrical signaling are associated with dysregulation of regulatory pathways. The kinase GSK3 is hypothesized to be a pivotal regulator of neuronal excitability and synaptic transmission. The discovery of the role of GSK3 in depression would open a new avenue to the study of the molecular basis of this disease, which is the leading cause of lifelong disability due to its high prevalence in the population.

4. Multiple Sclerosis (MS) is an autoimmune disease of CNS characterized by neuro-inflammation demyelinatio, and neurodegeneration without definitive cure. Our previous studies demonstrated a reduced level of NURR1 gene expression in the blood of patients with MS compared to healthy controls. NURR1 (NUclear Receptor-Related Protein 1) is a transcription factor that exerts anti-inflammatory role in the peripheral blood and central nervous system (CNS). A class of disease-modifying treatments (DMTs) recently discovered by Immunic Therapeutics and proposed to manage MS also act through a not-well described modulation of NURR.

c. Objectives

Aim 1. Functional alterations in cerebellar Purkinje cells in an animal model of AT. The aim was to find differences in action potential activity and/or in synaptic signaling.

Aim 2. Role of Elovl5 in action potential conduction. The aim was to detect an impairment in the conduction velocity of action potentials. The final objective was to understand the mechanisms that control action potential velocity in the axons of cerebellar Purkinje cells.

Aim 3. Neuronal mechanisms of depression. The aim of the research was to investigate, in pyramidal neurons of the mPFC, the alterations in action potential firing induced by the development of a depressive-like behavior following the chronic social defeat stress paradigm.

Aim 4. Role of NURR1 in neuro-inflammation. The aim was to dissect the role of NURR1 by means of *in-vitro* and *in-vivo* systems of inflammation using molecules able to modulate its function.

d. Results

Aim 1. Functional alterations in cerebellar Purkinje cells in an animal model of AT. Action potentials were evoked in cerebellar Purkinje cells by direct depolarization via the patch pipette. We found that such evoked firing showed aberrantly higher frequency compared to control animals. This might be a mechanism of the early signs of ataxia in AT. While the parallel fiber-Purkinje cell synapse was intact, we found that the climbing fiber-Purkinje cell synapse showed a more pronounced paired pulse depression, which is a form of synaptic plasticity in which the calcium-dependent release of neurotransmitter is involved.

Aim 2. Role of Elovl5 in action potential conduction. In mice with deletion of Elovl5, the conduction of action potentials showed a slowed velocity. However, conduction failures were not present. Action potentials in Elovl5 deficient mice normally triggered neurotransmitter release. These results suggest a specific role of Elovl5 in preserving high velocity conduction of electrical signals along the Purkinje cell axon. Such deficit might, at least in part, be responsible for motor symptom of Elovl5 deficient mice and of patients with ELOVL5 mutations.

Aim 3. Neuronal mechanisms of depression. For the induction of a depressed-like behavior in mice we used the chronic social defeat stress (CSDS) paradigm. Pyramidal neurons of the mPFC were recorded by patch-clamp in acutely prepared tissue slices. In contrast with previous findings with different induction protocols, which used indirect techniques and gave contrasting results, we found a deficit in action potential generation, consisting in a pronounced frequency adaptation, increase of action potential threshold and enhanced afterhyperpolarization.

Aim 4. Our results show that the disease-modifying treatments recently discovered by Immunic Therapeutics are able to modulate the expression of the NURR1-target genes in primary murine cell cultures and to alter the neuropathological features characteristic of MS in *ex-vivo* neuronal slice cultures. Also, the clinical course of a murine model of MS (experimental eutoimmune encephalomyelitis), is altered by the administration of the proposed disease-modifying treatment.

e. Advancement in the field

The finding of deficits in action potential firing and of synaptic transmission in the animal model of AT are completely novel, and they are an indispensable knowledge to design a therapy.

Our results on the importance of Elovl5 in the conduction velocity of action potentials indicate a primary role of Elovl5-dependent PUFAs for axonal functional properties.

Our results revealing action potential deficit in the animal model of depression provide the first insight into the neural mechanisms responsible for this psychiatric disorder. These results represent a starting point to investigate the role of Gsk3 and Fgf14, which might become novel targets of a new generation of antidepressant drugs.

Our findings on NURR1 and neuroinflammation will open up future therapeutic strategies to treat MS by acting directly on the CNS and not just the immune system, as is generally the case with most disease-modifying treatments.

f. Publications

- Bonato M*, Montarolo F*, Parolisi R, Caudana M, Abbadessa G, Cardinale V, Nato G, Pandino C, Di Cintio N, Schiavo V, Giustetto M, Luzzati F, De Francia S, Bertolotto A, Buffo A, Boda E. (2025). Acute Particulate Matter (PM10) Exposure Selectively Triggers Behavioral Alterations in the Presymptomatic Experimental Autoimmune Encephalomyelitis (EAE) Mouse Model of Multiple Sclerosis. Eur J Neurosci. Nov;62(10):e70304. *equally contributed to the work
  Research article Q2
- Rominto AM, Montarolo F, Berrino L, Loddo M, Bertolotto A, Tempia F, Hoxha E. (2025). Depression in mice causes decreased neuronal excitability and enhanced frequency adaptation in medial prefrontal cortex pyramidal neurons. Sci Rep. Nov 3;15(1):38402
  Research article Q1
- Montarolo F, Gao F, Loddo M, Milek J, Zawadzka M, Miniaci MC, Brusco A, Balbo I, Dziembowska M, Tempia F, Skowronska-Krawczyk D, Hoxha E. (2025). Impact of Elovl5 deficiency on cerebellar excitatory synaptic transmission in mice. J Neurosci. Aug 11:e1529242025
  Research article Q1
- Bonato M, Cerrato V, Dioni L, Montarolo F, Parolisi R, Bertolotto A, Bollati V, Ferrari L and Boda E. (2025). Short-term exposure to particulate matter triggers a selective alteration of plasma extracellular vesicle-packaged miRNAs in a mouse model of multiple sclerosis. Front. Immunol. 16:1596935.
  Research article Q1
- Montarolo F, Rominto AM, Berrino L, Bertolotto A, Laezza F, Tempia F, Hoxha E. (2025). Deletion of Fgf14 confers resilience to basal and stress-induced depressive-like behavior and reduces anxiety in mice. Transl Psychiatry. 2025 Apr 9;15(1):136.
  Research article Q1
- Montarolo F, Thielens S, Bove M, Bertolotto A, Tempia F, Hoxha E. (2025). NURR1 deficiency is associated to altered microglial phenotype in male mice. Mol Neurobiol. 2025 Mar 8.
  Research article Q1
- Lado W, Wu X, Choi S, Dong Y, Yang G, Arancio O, Tempia F, Miniaci MC, Sulzer D, Mosharov E, Tang G. (2025). Synaptic plasticity deficits in a mouse model of Timothy syndrome: LTP saturation and its pharmacological rescue by nifedipine. Biomed Pharmacother. 184:117896.
  Research article Q1

5. Future directions and objectives for next years

a. Summary

In the animal model of AT, in which we found disfunction in action potentials and synaptic transmission, we plan to search for alterations in calcium signaling. In another line of research, we plan to study the role of Elovl5 in the conduction of action potentials in the central nervous system. Starting from our results on neuronal firing in animal model of depression, we plan to investigate the role of Gsk3 and Fgf14 in such functional alterations. The role of NURR1 in neuroinflammation will be further investigated.

b. Background and Significance

1. A-T is an autosomal recessive disorder caused by loss of function of the ATM kinase and is characterized by an early onset, progressive cerebellar ataxia, oculocutaneous telangiectasia, immunodeficiency, pulmonary disease and increased risk of developing cancer. Histological studies from autoptic brain material revealed an important degeneration of PCs with an unavoidable compromised cerebellar structure. The Atm protein is involved in several cellular responses to damage, including ionizing radiations, oxidative stress, hypoxic ischemia. Despite such knowledge on the role of Atm in response to cell stress, the mechanisms that cause PC death are still unknown. Recent indirect evidence suggests a role of excitotoxicity and dysregulation

of calcium signaling. Our aim is to exploit a novel animal model of A-T to uncover the mechanisms causing PC lesion. In the last year the research was focused on the early alterations in PC physiology, including action potential firing and postsynaptic events. However, indirect evidence suggests an important involvement of deficits in calcium signaling.
2.. We previously ì showed that Elovl5-dependent PUFAs are important for proper action potential conduction in peripheral nerves and that the Purkinje cells axon requires Elovl5 dependent PUFA. High velocity of action potential conduction in the axon is necessary to preserve the timing of electrical signaling. This is particularly important in the cerebellum, in which timing of motor control is crucial. Elovl5 deletion in mice or mutation in humans causes cerebellar motor deficits. Our hypothesis is that Elovl5-dependent PUFAs are essential to preserve the correct velocity of action potential conduction along the axons of Purkinje cells.
3. Currently available therapies for mood disorders, including Major Depressive Disorder (MDD) and Bipolar Disorder (BD), require long-term treatment and have limited efficacy. The discovery of the cellular and molecular mechanisms of depression is required for the development of therapies with higher efficacy. The involvement of GSK3 in mood disorders is supported by genetic studies and investigations on the mechanisms of action of lithium, mood stabilizers and antidepressant drugs. GSK3 controls neuronal excitability and synaptic transmission. It is negatively regulated by phosphorylation at serine residues, while tyrosine phosphorylation promotes its activity. Lower GSK3 level are specific of patients with BD relative to MDD. Mutant mice with a constitutive GSK3 hyperactivity have increased susceptibility to depression, but the molecular and electrophysiological mechanisms are not known. Moreover, the role of GSK3 in the brain regions involved in depression is still unknown. Fgf14 is a cytoplasmic protein that regulated the function of voltage-dependent sodium channels. Our findings on the resilience of Fgf14 knockout mice to depression will be a starting point to investigate the involvement of Fgf14 in the pathogenesis of depression.
4. Multiple Sclerosis (MS) is an autoimmune disease of the central nervous system characterized by neuro-inflammation, demyelination, and neurodegeneration, without definitive cure. Our previous studies demonstrated a reduced level of *NURR1* gene expression in the blood of patients with MS compared to healthy controls. A class of disease-modifying treatments recently discovered by Immunic Therapeutics and proposed to manage MS also act through a not-well described modulation of NURR1.

c. General aim and integration with mission of the Institute

Our projects regard the neuronal bases of several neurologic and psychiatric disorders. Regarding neurologic diseases, we plan to focus on the neurophysiological basis of A-T and MS. A general aim about psychiatric disorders is to identify new signaling pathways, including GSK3and FGF14, involved in mood disorders. The mission of the Institute is exactly the same as ours, namely, to advance scientific knowledge regarding brain disorders, including psychiatric diseases such as depression and neurologic diseases like cerebellar ataxias and basic mechanisms, which are altered in neurological disorders.

d. Specific objectives and strategies

Aim 1: The main aim of this phase of research on the neuronal mechanisms of AT is to find dysregulated calcium signals exploiting our animal model. The alteration that we found in the short-term plasticity of the climbing fiber-Purkinje cell synapse suggests a problem with calcium signaling. We plan to start with an evaluation of the basal calcium level in Purkinje cells. Calcium signals evoked in Purkinje cells by depolarization or by synaptic activation will be studied in vitro in cerebellar slices. Spontaneous calcium signals produced by climbing fiber activity will be studied in vivo by two photon microscopy.

Aim 2: The aim of this research is to determine the effects of Elovl5 deficiency on action potential conduction along axons in the central nervous system. In slices of cerebellum we plan to stimulate the Purkinje cell axon and simultaneously record backpropagated action potentials with an electrode in the soma.

Aim 3: Role of Gsk3 and Fgf14 in the control of action potentials following induction of depression. The aim of the project is to identify changes in neuronal function induced by Gsk3 dysregulated activity in the mPFC of murine models of depression. For the induction of a depressed-like behavior in mice we will use the chronic social defeat stress (CSDS) paradigm. Our previous results showed that Gsk3β becomes hyperactive in mice susceptible to CSDS. Thus, after induction of a depressed-like behavior by CSDS, a

thorough evaluation of neuronal activity in the mPFC will be performed. In the next year we plan to start with an evaluation of synaptic transmission following CSDS. In addition to the experiments in normal mice after CSDS, in order to identify which changes in neuronal function are caused by Gsk3 hyperactivity we will study mPFC neuronal activity in knock-in mice (abbreviated Gsk3-KI) in which regulatory serines of Gsk3α and Gsk3β have been mutated into alanines, so that both isoforms cannot be inhibited and are constitutively hyperactive (McManus et al., 2005). Gsk3-KI mice under basal conditions (not subjected to a stress protocol) do not display spontaneous anxiety or depressed-like behavior. However, they are highly susceptible to depression in response to stress protocols. An electrophysiological analysis of action potentials will be conducted in CSDS and Gsk3-KI mice. Depression in patients and mice is associated with decreased neuronal activity in the mPFC. Our goal is to detect action potential firing alterations caused by changes of Gsk3 activity following CSDS. In parallel, neuronal activity in the PFC will be studied in $Fgf14^{-/-}$ mice, which we showed to be resilient to depression.

Aim 4: We would like to extend the project about the role of NURR1 during neuro-inflammation, enlarging the in-vitro, ex-vivo and in-vivo experimental setting to better dissect the molecular aspect related to the NURR1 modulation during neuroinflammation.

e. Unique features of the project research

1. In ataxia-teleangiectasia, calcium dysregulation might be a major mechanism of Purkinje cell dysfunction and ultimately cell degeneration. The identification of such mechanisms would open the way to design new treatments to rescue Purkinje cells and prevent ataxia in patients with AT.

2. A deficit in action potential conduction, especially in cerebellar neurons, could disrupt the precise timing of signals necessary for motor control. Moreover, the results might provide new knowledge about the role of the lipidic composition of myelin for the optimization of action potential conduction at high velocity.

3. By the experiments of Aim3 we expect to identify Gsk3 and Fgf14 alterations involved in mood disorders. A possible clinical impact is the possibility to utilize a Gsk3 dosage assay to guide and refine the diagnosis. We expect to characterize the Gsk3-modulation profile of different mood disorders and the effects of therapy. The study of functional neuronal alterations of pyramidal neurons of the prefrontal cortex would be a first result in a new line of research aimed at discovering the neuronal mechanisms of mood disorders. We expect to determine the role of the Gsk3 pathway in the induction of the alterations of action potentials and synaptic transmission. This would open the way to the development of new drugs with a better efficacy relative to current therapies.

4. In MS, NURR1 could influence neuro-inflammatory aspects of the disease. The dissection of this mechanism could open up future therapeutic strategies to treat MS by acting directly on the central nervous system.

f. Methodology

We plan to perform some of the measures of neuronal activity by in vivo two photon imaging.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Brain development and disease

1. LABORATORY DESCRIPTION – PERSONNEL

Principal Investigator

Vercelli, Alessandro
- Degree: MD, PhD
- Birthdate: 09/07/1961
- Phone: +39 011 670 6617
- Email: alessandro.vercelli@unito.it
- Position: Full Professor
- Role: Principal Investigator

Personnel

Boido, Marina
- Degree: PhD
- Birthdate: 06/09/1980
- Phone: +39 011 670 6613
- Email: marina.boido@unito.it
- Position: Full Professor (since October 2025)
- Role: Spinal cord injury, motor neuron diseases (ALS and SMA), aging, drug repositioning, stem cells, exposome

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Calì, Corrado
- Degree: PhD
- Birthdate: 27/11/1982
- Phone: +39 011 670 3447
- Email: corrado.cali@unito.it
- Position: Associate Professor
- Role: Glia, astrocytes, 3D electron microscopy, 3D modeling and analysis, VR (Virtual Reality), AR (Augmented Reality)

Stanga, Serena
- Degree: PhD
- Birthdate: 03/06/1983
- Phone: +39 011 670 6632
- Email: serena.stanga@unito.it
- Position: Associate Professor
- Role: Ageing and Alzheimer's disease (AD), motor neuron diseases (ALS and SMA), molecular neuroscience, primary neuronal cultures, mitochondrial dysfunctions

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Marvaldi, Letizia
- Degree: PhD
- Birthdate: 23/01/1983
- Phone: +39 011 670 6632
- Email: letizia.marvaldi@unito.it
- Position: Associate Professor
- Role: Pain and neurite outgrowth signalling, neurogenetics of pain and neuronal regeneration and survival, importins, age and gender differences in pain perception and axonal outgrowth

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Schellino, Roberta
- Degree: PhD
- Birthdate: 11/02/1985
- Phone: +39 011 670 6632
- Email: roberta.schellino@unito.it
- Position: Associate Professor (since November 2025)
- Role: Neurogenesis, motor neuron diseases (SMA and ALS), neuromuscular junctions, aging, green therapy, histology, confocal imaging, cell tracing, behavior

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Pacca, Paolo
- Degree: MD, PhD
- Birthdate: 23/06/1984
- Phone: +39 011 670 6632
- Email: paolo.pacca@unito.it
- Position: Assistant Professor RTD-A
- Role: Glymphatic system, motor neuron diseases, spinal cord injury, cell culture, muscular atrophy, aging, robotic technology for elderly care

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Menduti, Giovanna
- Degree: PhD
- Birthdate: 14/04/1991
- Phone: +39 011 670 6632
- Email: giovanna.menduti@unito.it
- Position: II Level Technologist
- Role: Cellular and molecular neurobiology, spinal muscular atrophy, drug repositioning

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Bartesaghi, Luca
- Degree: PhD
- Birthdate: 28/12/1980
- Phone: +39 011 670 6632
- Email: luca.bartesaghi@unito.it
- Position: Post-doc Research Fellow (since September 2024)
- Role: Myelinating glia, developmental neuroscience, light microscopy, 3D modelling, molecular biology, cell culture, animal experimentation

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La Rocca, Federica
- Degree: PhD
- Birthdate: 18/07/1994
- Phone: +39 011 670 6632
- Email: federica.larocca@unito.it
- Position: Post-doc Fellow (since December 2024)
- Role: Spinal muscular atrophy, neuromuscular diseases

Mattioni, Anna
- Degree: PhD
- Birthdate: 02/12/1986
- Phone: +39 011 670 6632
- Email: anna.mattioni@unito.it
- Position: Post-doc Fellow (since October 2024)
- Role: Ageing and Alzheimer’s disease, cell culture, organoids, iPSCs, molecular and cellular biology, correlative microscopy

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Mezzanotte, Mariarosa
- Degree: PhD
- Birthdate: 19/08/1985
- Phone: +39 011 670 6632
- Email: mariarosa.mezzanotte@unito.it
- Position: Post-doc Fellow
- Role: Ageing and Alzheimer's disease, brain iron metabolism, histological and molecular analysis

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Chasseur, Silvia
- Degree: Master's Degree in Biotechnology for Neuroscience
- Birthdate: 01/11/1999
- Phone: +39 345 953 5393
- Email: silvia.chasseur@unito.it
- Position: Research Fellow
- Role: Spinal cord injury, bioprinting, cell culture, cellular and molecular analysis, pollution

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Chicote, Javier
- Degree: Bachelor's Degree
- Birthdate: 18/01/1987
- Phone: +39 011 670 6632
- Email: javier.chicotegonzalez@unito.it
- Position: PhD Student (since November 2025)
- Role: Ageing and Alzheimer's disease, brain iron metabolism, histological and molecular analysis

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Iezzi, Giulia
- Degree: Master's Degree in Medical Biotechnology
- Birthdate: 05/10/1999
- Phone: +39 011 670 6632
- Email: giulia.iezzi@unito.it
- Position: Research Fellow (since July 2024)
- Role: Chemotherapy-induced cognitive impairment, spinal muscular atrophy, histological analysis

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Musso, Giada
- Degree: MSc in Cellular and Molecular Biology
- Birthdate: 27/03/1999
- Phone: +39 011 670 6632
- Email: gi.musso@unito.it
- Position: Research Fellow
- Role: Pain research and injury signals, embryonic and adult DRG culture, neurite outgrowth assay, molecular biology and imaging

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Tuninetti, Alessandra
- Degree: Master's Degree in Animal Biotechnology
- Birthdate: 13/06/1998
- Phone: +39 011 670 6632
- Email: alessandra.tuninetti@unito.it
- Position: Research Fellow
- Role: Glymphatic system in spinal cord injury and motor neuron diseases, biomarkers

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Caretto, Anna
- Degree: Master's Degree in CTF
- Birthdate: 08/07/1995
- Phone: +39 011 670 6632
- Email: anna.caretto@unito.it
- Position: PhD Student
- Role: Spinal muscular atrophy, neuromuscular diseases

Chiappini, Vanessa
- Degree: Master's Degree in Biomedical Engineering
- Birthdate: 23/07/1997
- Phone: +39 011 670 6632
- Email: vanessa.chiappini@unito.it
- Position: PhD Student
- Role: Spinal cord injury, bioprinting, cell culture

Dallere, Sveva
- Degree: Master's Degree in Medical Biotechnology
- Birthdate: 23/07/1997
- Phone: +39 011 670 6632
- Email: sveva.dallere@edu.unito.it
- Position: PhD Student
- Role: Cell culture, muscular atrophy, Alzheimer disease, iPSCs, terpenes, electron microscopy, correlative microscopy

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Dotta, Sofia
- Degree: Master's Degree in Biotechnology for Neuroscience
- Birthdate: 21/09/1999
- Phone: +39 011 670 6632
- Email: sofia.dotta@unito.it
- Position: PhD Student
- Role: Pain research during aging, embryonic and adult DRG culture, neurite outgrowth assay, molecular biology and imaging

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Ferrero, Clelia
- Degree: Master's Degree in Neurobiology
- Birthdate: 30/09/1997
- Phone: +39 011 670 6632
- Email: clelia.ferrero@unito.it
- Position: PhD Student
- Role: Amyotrophic lateral sclerosis, Frontotemporal Dementia, stem cells, iPSCs, organoids

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Nicorvo, Ersilia
- Degree: Master's Degree in Neuroscience
- Birthdate: 31/05/1995
- Phone: +39 011 670 6632
- Email: ersilia.nicorvo@unito.it
- Position: PhD Student
- Role: Spinal muscular atrophy, drug reposition, cell culture, iPSCs, glioblastoma, neuron-tumor crosstalk

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Pavarino, Gianna
- Degree: Master's Degree in Molecular Biotechnology
- Birthdate: 02/05/1997
- Phone: +39 011 670 6632
- Email: gianna.pavarino@unito.it
- Position: PhD Student
- Role: Spinal muscular atrophy, depression, molecular biology, iPSCs, terpenes

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Rasà, Daniela Maria
- Degree: Master's Degree in Biology
- Birthdate: 11/09/1990
- Phone: +39 011 670 6632
- Email: danielamaria.rasa@unito.it
- Position: PhD Student
- Role: Cell culture, motor neuron diseases (SMA and ALS), stress, drug repositioning, molecular and cellular biology

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Scimia, Noemi
- Degree: Master's Degree in Biotechnology for Neuroscience
- Birthdate: 25/02/1999
- Phone: +39 011 670 6632
- Email: noemi.scimia@unito.it
- Position: PhD Student
- Role: Aging and Alzheimer's disease, motor neuron diseases (ALS and SMA), brain iron metabolism, histological and molecular analysis

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Veloz-Castillo, Maria Fernanda
- Degree: Master's Degree in BioScience
- Birthdate: 03/01/1995
- Phone: +39 351 922 6321
- Email: mariafernanda.velozcastillo@unito.it
- Position: PhD Student
- Role: Brain energy metabolism, behavior, image analysis

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
2025	SMA	Vercelli A./Boido M	Girotondo Onlus	Coordinator	15,000 €	FCO
2023-2025	Eloquent areas in the CC of patients undergoing glioma surgery	Vercelli A./Garbossa D.	Reply	Coordinator	100,000	FCO
2021-2025	The involvement of the small heat shock protein HSPB8 in amyotrophic lateral sclerosis	Vercelli A.	AFM Telethon	Coordinator	36,000 €	UNITO

2022-2026	D34H	Vercelli A.	PNRR MUR/MH	Coordinator UNITO	4,3 M€	UNITO
2025-2026	Daisy&Ron	Vercelli A.	PNRR/Anthem Foundation	Coordinator	300,00€	UNITO
2023-2027	Department of Excellence	Vercelli A.	MUR	Coordinator DNS - UNITO	7.75 M€	UNITO
2023-2025	Understanding and targeting Chemotherapy-related neurotoxicity	Vercelli A.	PRIN 2022	P.I. of research unit	85,000 €	UNITO
2023-2026	Innova	Vercelli A	PNRR – Ministry of Health	Coordinator UNITO	279,850	UNITO
2025-2027	Pastiss+	Vercelli A	Regione Piemonte	Coordinator UNITO	3 M€	UNITO
2023-2026	A combinatorial pharmacotherapeutic approach to counteract Spinal Muscular Atrophy	Boido M.	AFM Telethon	P.I. of research unit	90,000 €	UNITO
2023-2025	Uncovering the mechanism of action and synergistic potential of a SMA repositioned therapy	Boido M.	SMA Europe	Coordinator	60,000 €	FCO
2022-2025	NODES - INNDIANA	Boido M.	PNRR	P.I. of the research unit	60,000 €	UNITO
2023-2025	A 3D Bioprinted spinal cord Model to reach functional meaningful and clinically translatable regeneration	Boido M.	PRIN 2022	Coordinator	61,711 €	UNITO
2024-2025	A multiscale imaging approach to study early alterations in an Alzheimer cerebral organoid (MULTIMALZ)	Boido M.	PNRR MUR	Coordinator (Co-PI Roberta Schellino)	280,960 €	FCO
2024-2027	REACT: Rebalancing the Epitranscriptional landscape as an AdvanCed Therapy in SMA	Boido M.	CURE SMA 2024	P.I. of the research unit	12,420 $	FCO
2024-2027	InibiRE l'infiAmmazione per Contrastare l'aTrofIa muscOlare nell'aNziano (REACTION)	Boido M.	Fondazione CRT (progetti interdipartimentali)	P.I. of the research unit	14,950 €	UNITO
2021-2024	NODES - TINCARE	Calì C.	PNRR	P.I. of the research unit	60,000 €	UNITO
2022-2025	Cross-talk between intrinsic and extrinsic mechanism in importin alpha 3 knock-out mice.	Marvaldi L.	Programma per Giovani Ricercatori "Rita Levi Montalcini"	Coordinator	251,581 €	UNITO

	Investigating the neurite outgrowth in embryonic neurons and the control gene regulation in importin alpha3 knock-out mice.					
2023-2025	Targeting mAconitase and KIF5A to rescue mitochondrial mobility and function for the treatment of motor neuron diseases	Stanga S.	PRIN 2022	P.I. of the research unit	102,625€	UNITO
2023-2025	Evaluation of the effect of leriglitazone on brain iron deposits in a murine model of Alzheimer's disease	Stanga S.	Minoryx therapeutics	Coordinator and P.I. of the research unit	60,000€	FCO
2024-2026	Evaluation of the effect of leriglitazone on cognitive behaviour in a murine model of Alzheimer's disease	Stanga S.	Minoryx therapeutics	Coordinator and P.I. of the research unit	34,800 €	FCO
2025-2028	Nuova classe di farmaci per la prevenzione della neuroinfiammazione nella cura della Malattia di Alzheimer	Stanga S.	Fondazione CRT	Coordinator	40,000€	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Alessandro Vercelli, PI

Supervised PhD students:	A. Caretto (co-tutorship with M. Boido), S. Dallere (co-tutorship with M. Boido), G. Pavarino (co-tutorship with M. Boido), S. Dotta (co-tutorship with L. Marvaldi), C. Ferrero (co-tutorship with M. Boido)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	G. Aumayr (Austria), M. Summers (Australia), M. Sato (Osaka University, Japan), J. De Felipe (Cajal Institute), Serenella Tolomeo (Singapore), Helena Vieira (Lisboa, Portugal), Pharmafox Therapeutics
● Invited talks:	21 March 2025 L'anatomia del glinfatico. Accademia medicina 16 May 2025 Pyramidal neurons in brain development and disease Osaka University Japan 15 October 2025 World anatomy day 17 October 2025 Tempo di vivere tempo di morire, Università di Sassari

	4 December 2025 Cortical alterations in SMA. University of Oxford
● Science communication:	Malattie neurodegenerative e prevenzione. CRC My AHA (my active and healthy ageing) e Daisy&Ron: prevenire la fragilità nell'anziano – Brain@gym 2025 My AHA (my active and healthy ageing) e Daisy&Ron: prevenire la fragilità nell'anziano con la tecnologia – Rotary Torino Palazzo Reale 2025 Webinar "Il cervello al centro del nostro benessere" for Aizoon Weelbeing program, Torino, Italy (06/10/2025) Podcast Vois (https://www.youtube.com/watch?v=aUI7BYwsjIs)
● Editorial duties:	Associate Editor Frontiers Ageing Neuroscience
● others	
Organizational activities and responsibilities at NICO:	Scientific Director
Speakers iynvited:	Javier DeFelipe (Cajal Institute, Madrid, Spain); Serenella Tolomeo (Institute of High Performance Computing, A*STAR, Singapore); Makoto Sato (University of Osaka)
Other organizational activities:	Honorary President of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Workshops, Schools or Conferences organized:	NICO-IBS workshop
Technology transfer achievements (patents, etc.):	President Spin-off committee, UNITO; UNITO representative in European Institute of Innovation & Technology Health

Marina Boido, Full Professor

Supervised PhD students:	A. Caretto (co-tutorship with A. Vercelli), C. Ferrero (co-tutorship with A. Vercelli), DM Rasà, AA Flores da Silva (co-tutorship with A.C. Cristovao, Univ. Beira Interior), S. Dallere (co-tutorship with A. Vercelli), G. Pavarino (co-tutorship with A. Vercelli)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Prof. Artero (Univ. Valencia), Prof. Soler (University of Lleida, Spain), Pharmafox Therapeutics AG (Switzerland), Dr. Martinat (I-STEM, Corbeil-Essonnes, France), P. Smeriglio (Institut de Myologie, Paris, France), A. Prochiantz (Collège de France, Paris, France), Prof. Cristovao (Univ. Beira Interior, Portugal), Dr Rico Schieweck (University of Luxembourg)
● Invited talks:	Invited talks at meetings: "The green therapy effects on depression and Alzheimer' disease", 97° SIBS meeting, Palermo, 12/04/2025 "Beyond motor neurons: neuromuscular junction and skeletal muscle contribution to spinal muscular atrophy pathogenesis". University of Osaka, Japan, 16/05/2025 "Organoidi cerebrali: un nuovo modello 3D per decifrare la Demenza Frontotemporale", D34H webinar, 25/06/2025 "Advances in spinal cord injury treatment: from cell therapy to biomaterials and tissue engineering", Regenerative Medicine Academy, Novara, 15/10/2025 "Sustainability in Biology: Plastics & Human Health", Specializing Master in Sustainability in Health, online, 05/12/2025 "Neurodegenerative diseases: from pathogenetic mechanisms to treatments" IBS-UniTO Neuroscience Workshop (14/01/2025)

	Invitation upon selection at meetings: "New Repurposed Drugs for the Treatment of Spinal Muscular Atrophy", 21° SINS meeting, Pisa, 10-13/09/2025
● Science communication:	Regional coordinator (Piedmont) of Olimpiadi delle Neuroscienze", Regional stage, Turin (15/03/2025) Conference "Ricostruire il sistema nervoso grazie ad una stampante 3D", Festival dell'Innovazione e della Scienza, Settimo T.se (TO) (06/10/2025). Webinar "I benefici della natura sulla salute del cervello", for Aizoon Weelbeing program, Torino, Italy (15/12/2025)
● Editorial duties:	Associate Editor for Frontiers in Medical Engineering Review editor for the journal Frontiers in Aging Neuroscience
● others	General Secretary in the SIBS directive board; Coordinator of CU6/CU Alpha in the national PhD "Sustainable Development and Climate Change"; Representative of the Italian Society for Neuroscience to the Representative of the Italian Society for Neuroscience and NICO to the ALBA Network; Councilor of the SINS Governing board.
Organizational activities and responsibilities at NICO:	Responsible for the infrastructure in open access "In vivo and behavioral studies" Organization of the NICO NeuroWebinars
Speakers invited:	Daniela Ferrari (Dept. Biotechnology and Biosciences, UNIMIB)
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Conference Chair of the Local Organizing and Scientific Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Corrado Calì, Associate Professor

Supervised PhD students:	M. F. Veloz Castillo, V. Chiappini
Honors, prizes, awards:	Best paper honorable mention at VBCM Conference 2025, October 3rd, Delft, Netherlands
Outreach activities	
● International collaborations:	Pierre Magistretti (KAUST, Saudi Arabia), Marco Agus (Hamad Bin Khalifa University, Qatar), Markus Hadwiger (KAUST, Saudi Arabia), Leena Ibrahim (KAUST, Saudi Arabia)
● Invited talks:	Invited talks at meetings: "3D Ultrastructural Analysis of Astrocytic Glycogen-Derived Lactate in Synaptic Plasticity and Brain Energy Metabolism", Japan Society for Neuroscience National Meeting, Niigata, Japan, 24/07/2025 "Structural reshaping of potentiated synapses requires glycogen", Plenary lecture, International Neuronal Energy Dynamics Congress, Norom, Ankara, Turkye, 2/11/2025
● Science communication:	Rotary night, 14/05/2025; UNIGHT presenter, 26/09/2025
● Editorial duties:	Associate editor of Advanced Technology in Neuroscience
● others	Member of the review panel of EIC Pathfinder program; Member of the review panel of the Brayn Starting Grant; Responsible for Erasmus program of Biotechnology for Neuroscience course

Organizational activities and responsibilities at NICO:	Responsible for the Gemini 460 SEM Volutome system
Speakers invited:	Meltem Bahcelioglu (NOROM, Ankara, Turkye)
Other organizational activities:	President of Intravides SRL; Scientific Officer at Cllinatec Endowment Fund (Grenoble, France)
Workshops, Schools or Conferences organized:	Member of the Local Organizing and Scientific Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025); Member of the Scientific Committee of the International Neuronal Energy Dynamics Congress, NOROM, Ankara, Turkye; Member of the Organizing Committee of the Disaster Victim Identification Course, University of Turin, June 2025; Organizer of the 1st MITIC-DNS course "Changes in Future Medicine", 23/09/2025
Technology transfer achievements (patents, etc.):	na

Letizia Marvaldi, Associate Professor

Supervised PhD students:	S. Dotta (co-tutorship with A.Vercelli)
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Dr. Franziska Rother (MDC Berlin; Germany), Prof. Michaela Kress (Medical University Innsbruck, Austria), Prof. Mike Fainzilber and Dr. Ida Rishal (Weizmann Institute of Science, Rehovot, Israel), Prof. Marc Suter (University of Lausanne, Switzerland).
● Invited talks:	Invitation upon selection at meetings: "Paw posture is a robust indicator for injury, pain, and age", EMBO Workshop on Cell Biology of the Nervous System: Cellular Mechanisms of Communication, Heraklion, Greece (05-08/05/2025)
● Science communication:	na
● Editorial duties:	na
● others	Posters at meetings: Pritz CO, Dotta S, Freund P, Musso G, Rother F, Bader M, Okladnikov N, Marvaldi L. Paw posture is a robust indicator for injury, pain and age. 8th BRAYN Conference, Turin, Italy (30/10/2025) Dotta S, Musso G, Muraglia L, Parmar A, Fusco S, Rother F, Bader M, Hartmann E, Pritz CO, Marvaldi L. The link between neuronal branching and pain. Congresso Gruppo Italiano per lo studio della Neuromorfologia (GISN), Torino, Italy (28/11/2025)
Organizational activities and responsibilities at NICO:	Coordination for BioRender license
Speakers invited:	Dr. Michael Tsoory (Weizmann Institute, Israel)
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Roberta Schellino, Associate Professor

Supervised PhD students:	na

Honors, prizes, awards:	
Outreach activities	
● International collaborations:	J. Willem Vrijbloed (Pharmafox Therapeutics AG); R. Fariello (Pharmafox Therapeutics AG); Prof.. Silvia Pozzi (ULaval, CERVO research institute, Quebec, Canada)
● Invited talks:	Invitation upon selection at meetings: Panelist of the symposium "Motor Neuron Disease: advances into therapeutic strategies and biomarker discovery", 21st national congress of the Italian Society of Neuroscience (SINS), Pisa, Italy. (10-13/09/2025). Talk: "Time-dependent Cortical Projection Neuron alterations in SMA model". Schellino R, Iezzi G, Caretto A, La Rocca F, Boido M, Vercelli A. Webinar: "Time-dependent characterization of cortical projection neuron's alterations in a murine model of Spinal Muscular Atrophy" IBS-UniTO Neuroscience Workshop. (14/01/2025)
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025) Outreach meetings for fourth- and fifth-year students of "I.I.S. Alberto Castigliano" (Asti) on Rita Levi-Montalcini's life and discoveries and scientific research at the NICO laboratories. (25/03/25 and 05/04/25)
● Editorial duties:	Review board for the journals: Frontiers in Neuroscience (ISSN 1662-453X), Frontiers group; Brain Communications (ISSN 2632-1297), Oxford Academic.
● others	Poster at meeting: Schellino R, Iezzi G, Caretto A, La Rocca F, Boido M, Vercelli A. Beyond the motor neuron: exploring the dynamics of cortical projection neurons in SMA disease. 97th Congress of the Italian Society of Experimental Biology (SIBS), Palermo, Italy (10-13/04/2025) Schellino R, Stanga S, Boido M et al. An emerging era in Italy: The impact of the PALS-led volunteers from Associazione conSLAncio Onlus; Associazione conSLAncio Onlus. ENCALS, Torino, Italy (3-6/06/2025) La Rocca F*, Schellino R*, Iezzi G, Caretto A, Boido M, Vercelli A "A new perspective on SMA disease beyond spinal motor neurons: what role do cortical projection neurons play?". 8th Brainstorming Research Assembly for Young Neuroscientists (BRAYN), Torino, Italy (29-31/10/2025) La Rocca F*, Schellino R*, Iezzi G, Caretto A, Boido M, Vercelli A. Beyond the spinal cord: morphological alterations of cortical projection neuron in Spinal Muscular Atrophy. XXXV GISN (Gruppo Italiano per lo Studio della Neuromorfologia) Conference. Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	Responsible for E800 Nikon Eclipse fluorescence microscope
Speakers invited:	Dr. Roberta Piovesana (Université de Montréal, Canada); Prof. Silvia Pozzi (Universitè Laval and CERVO research center, Quebec, Canada)
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Member of the Local Organizing and Scientific Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Serena Stanga, Associate Professor

Supervised PhD students:	Noemi Scimia (co-tutorship with A. Vercelli); Javier Chicote (co-tutorship with C. Guiot)
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Prof. P. Kienlen-Campard (UCLouvain, Bruxelles Belgium); Prof. F. Piguet (Inserm, Paris, France); Prof. A. Prochiantz and Dr. A. Di Nardo (College de France, Paris, France); Prof. C. Svendsen, Dr. D. Lall and Dr. F. Diaz (Cedars-Sinai, Los Angeles, USA); Minoryx Therapeutics (Spain, Belgium)
● Invited talks:	Invited talks: "Mitocondri: un denominatore comune tra le malattie neurodegenerative" from DGM Farmaceutici at Circolo dei Lettori, Torino, Italy 15.02.25 and Garden Ristò Roma, Italy 22.03.25
● Science communication:	Member of the UNISTEM Day organization of Attività 1 – Il Corpo Digitale at anatomage, Torino, Italy 14/03/2025 Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025) Organizer of "Pint of Science" Beautiful Mind Torino, Italy (19-21/05/2025) Article for Frida - Storie di ricerca/Scienza e tecnologia: "Braccio di ferro con l'Alzheimer: comprendere i meccanismi del cervello per una cura precoce" online Frida 12/06/2025 Organizer and speaker of "Ritorno al futuro: dialoghi tra anatomia storica e neuroscienze contemporanee" in collaboration with Museo di Anatomia Umana Luigi Rolando, Torino, Italy (19 and 27/06/2025) Webinar "Invecchiamento attivo e prevenzione delle malattie neurodegenerative" for Aizoon Weelbeing program, Torino, Italy (10/11/2025)
● Editorial duties:	Project evaluation for the French association for ALS research; Review editor for Aging and Disease
● others	Posters at meetings: Stanga S, Lall D, Diaz F, Di Lullo E, Mezzanotte M, Chicote J, Scimia N, Hernandez S, Sansa A, Prochiantz A, Svendsen Cn. A Role For Dysfunctional Mitochondria In ALS. 97° Congresso Nazionale Società Italiana di Biologia Sperimentale (SIBS), Palermo, Italy (10-13/04/2025) Stanga S, Schellino R, Boido M et al. Improving online caregiver training for ALS patients in Italy: a four-year update. Associazione conSLAncio Onlus, ENCALS Torino, Italy (3-6/06/2025) Mezzanotte M, Chicote J, Scimia N, Riganti C, Porporato P, Stanga S. Iron Dyshomeostasis And Mitochondrial Features' Alteration In Early Alzheimer's Disease. SINS meeting, Pisa, Italy (10-13/09/2025) Mezzanotte M, Chicote J, Scimia N, Riganti C, Porporato P, Stanga S. Iron Dyshomeostasis And Mitochondrial Features' Alteration In Early Alzheimer's Disease. New Horizons in Neurodegeneration, Leuven, Belgium (9-10/10/2025)
Organizational activities and responsibilities at NICO:	Responsible for the Cell Culture room (floor 0) Responsible for the dissection room (floor -1)

	Member of the Green Policy Committee NICO NEWS online monthly
Speakers invited:	na
Other organizational activities:	Organization of the Lab meetings 2025 of the group "Brain Development & Disease"
Workshops, Schools or Conferences organized:	Member of the Local Organizing and Scientific Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Paolo Pacca, Assistant Professor RTD-A

Supervised PhD students:	na
Honors, prizes, awards:	National Scientific qualification as Associate Professor, Academic Recruitment Field 05/H - Human anatomy, according to the Italian higher education system, in the call 2023/2025 (Ministerial Decree n. 1796/2023) for the disciplinary field of 05/H1 - Human anatomy
Outreach activities	
● International collaborations:	Helena Vieira (Lisboa, Portugal); J. Willem Vrijbloed (Pharmafox Therapeutics AG); R. Fariello (Pharmafox Therapeutics AG)
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Poster at meeting: Pacca P, Tuninetti A, Vercelli A, Boido M. AQP4 Overexpression and Polarization in ALS and SMA Mouse models. 21st National Congress of the Italian Society for Neuroscience (SINS 2025), Pisa, Italy, (10-13/09/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Member of the Local Organizing and Scientific Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Giovanna Menduti, II level technologist

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Prof. Artero (Univ. Valencia), Dr. Martinat (I-STEM, Corbeil-Essonnes, France)
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Invitation upon selection at meetings: "A novel therapeutic candidate for Spinal Muscular Atrophy identified through drug repositioning: evidence from a mouse

	model and patient-derived cells", 8th Brainstorming Research Assembly for Young Neuroscientists (BRaYN), Turin, Italy (29–31/10/2025). Posters at meetings: Menduti G, Ferrini F, Caretto A, Hassan A, Di Vito R, Beltrando G, Marnetto D, Usiello A, Di Cunto F, Boido M, Vercelli A. Cortical dysregulations of GABA signaling and metabolism in Spinal Muscular Atrophy: results from SMAΔ7 mice. 8th Brainstorming Research Assembly for Young Neuroscientists (BRaYN), Turin, Italy (29–31/10/2025) Menduti G, Ferrini F, Caretto A, Hassan A, Di Vito R, Beltrando G, Marnetto D, Usiello A, Di Cunto F, Boido M, Vercelli A. Morphological and synaptic changes of the cortical GABAergic inhibitory system in Spinal Muscular Atrophy. XXXV GISN 2025, Turin, Italy (28-29/11/ 2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Federica La Rocca, Post-doc fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Dr. Rico Schieweck (University of Luxembourg)
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Posters at meetings: La Rocca F*, Schellino R*, Iezzi G, Caretto A, Boido M, Vercelli A. A new perspective on SMA disease beyond spinal motor neurons: what role do cortical projection neurons play?. 8th Brainstorming Research Assembly for Young Neuroscientists, BraYn Conference 2025, Turin, Italy (29-31/10/2025) La Rocca F*, Schellino R*, Iezzi G, Caretto A, Boido M, Vercelli A. Beyond the spinal cord: morphological alterations of cortical projection neuron in spinal muscular atrophy. XXXV Congresso Nazionale del Gruppo Italiano per lo Studio della Neuromorfologia, GISN 2025, Turin, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Anna Mattioni, Post-doc fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	na
● Invited talks:	Invitation upon selection at meetings: "Multimalz: a multiscale imaging approach to study early alteration in an Alzheimer cerebral organoid", Second MNESYS Spoke 2 Meeting Neural Plasticity & Connectivity, Naples, Italy (24-25/09/2025)
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Posters at meetings: Mattioni A, Ferrero C, Parolisi R, Dallere S, Chiappini V, Signorino E, Schellino R, Boido M. Multimalz: a multiscale imaging approach to study early alteration in an Alzheimer cerebral organoid. MNESYS III annual meeting, Genoa, Italy, (27-28/02/2025) Mattioni A, Ferrero C, Parolisi R, Dallere S, Chiappini V, Signorino E, Schellino R, Boido M. Multimalz: a multiscale imaging approach to study early alteration in an Alzheimer cerebral organoid. Second MNESYS Spoke 2 Meeting Neural Plasticity & Connectivity, Naples, Italy (24-25/09/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Mariarosa Mezzanotte, Post-doc fellow

Supervised PhD students:	na
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Minoryx Therapeutics (Spain, Belgium); Prof. Clive Svendsen, Cedars-Sinai Medical Center (USA)
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025) Staff for science communication festival "Pint of Science" Beautiful Mind, Torino, Italy (19/05/25) Talks at science communication nights "Ricercatori alla Spina" Torino, Italy (21/05/2025)
● Editorial duties:	na
● others	Posters at meetings:

	Mezzanotte M, Chicote J, Scimia N, Riganti C, Porporato Paolo Ettore, Stanga S. Iron dyshomeostasis and mitochondrial features' alteration in the early phase of Alzheimer's disease in 5xFAD mouse model. 97° Congresso Nazionale Società Italiana di Biologia Sperimentale (SIBS), Palermo, Italy (10-13/04/2025) Mezzanotte M, Chicote J, Scimia N, Riganti C, Porporato Paolo Ettore, Stanga S. 5xFAD mouse model: investigating iron imbalance and mitochondrial features' alteration. XXXV Congresso Gruppo Italiano per lo studio della Neuromorfologia (GISN), Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Silvia Chasseur, Research fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	na
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Poster at meetings: Chasseur S, Traldi C, Chiappini V, Tonda-Turo C, Boido M. A 3D-bioprinted platform for modeling spinal cord architecture and neural activity in vitro. SINS 2025 (21st National Congress of the Italian Society for Neuroscience), Pisa, Italy (10-13/09/2025) Chasseur S, Traldi C, Chiappini V, Tonda-Turo C, Boido M. Engineering the spinal cord in vitro: a multi-scaffold 3D system for modeling cytoarchitecture and neural activity. BraYn Conference 2025 (8th Brainstorming Research Assembly for Young Neuroscientists), Torino, Italy (29-31/10/2025) Chasseur S, Traldi C, Chiappini V, Tonda-Turo C, Boido M. Multi Scaffold 3D bioprinted model of spinal cord architecture in vitro. GISN 2025 (35th Congress Gruppo Italiano per lo Studio della Neuromorfologia), Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Giulia Iezzi, Research fellow

Supervised PhD students:	na

Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	na
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● Others	Posters at meetings: Iezzi G, Marmiroli P, Vercelli A, Cavaletti G. Understanding Chemo-Fog: Cortical Structural Changes in Mice Treated with Oxaliplatin. 21st National Congress of the Italian Society for Neuroscience (SINS), Pisa, Italy (10-13/09/2025) Iezzi G, Marmiroli P, Vercelli A, Cavaletti G. Cortical Dendritic Remodeling in a Murine Model of Oxaliplatin-Induced Neurotoxicity. 8th Brainstorming Research Assembly for Young Neuroscientists (BRAYN), Turin, Italy (29-31/10/2025) Iezzi G, Marmiroli P, Vercelli A, Cavaletti G. Chemotherapy-induced remodeling of cortical pyramidal neurons: morphological effects of oxaliplatin and paclitaxel. XXXV convegno nazionale del gruppo italiano per lo studio della neuromorfologia (GISN), Turin, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Giada Musso, Research Fellow

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Prof. Michaela Kress (Medical University Innsbruck)
● Invited talks:	na
● Science communication:	na
● Editorial duties:	na
● others	Posters at meeting: Musso G, Dotta S, Testa L, Mollo A, De Carlo L, Muraglia L, Hartmann E, Bader M, Buffo A, Rother F, Marvaldi L. The interplay between importin a3 and CBX2 in axonal regeneration and pain. 8th Brainstorming Research Assembly for Young Neuroscientists conference, Turin, Italy (29-30-31/10/2025) Musso G, Dotta S, Testa L, Mollo A, De Carlo L, Muraglia L, Hartmann E, Bader M, Buffo A, Rother F, Marvaldi L. The interplay between importin a3 and CBX2 in axonal regeneration and pain. EMBO workshop "Cell Biology of the Nervous System: Cellular Mechanisms of Communication", Heraklion, Crete, Greece (05-08/05/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na

Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Alessandra Tuninetti, Research fellow

Supervised PhD students:	
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	na
● Invited talks:	na
● Science communication:	na
● Editorial duties:	na
● Others	Posters at meetings: Tuninetti A, Pacca P, Boido M, Vercelli A. AQP4 overexpression and polarization in ALS and SMA mouse models. 8th Brainstorming Research Assembly for Young Neuroscientists (BRAYN), Turin, Italy (29-31/10/2025) Tuninetti A, Boido M, Vercelli A, Pacca P. Change in AQP4 expression and localization during amyotrophic lateral sclerosis and spinal muscular atrophy progression. XXXV convegno nazionale del gruppo italiano per lo studio della neuromorfologia (GISN), Turin, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	na

Vanessa Chiappini, PhD student

Supervised PhD students:	
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	na
● Invited talks:	GRS on Volume Electron Microscopy (Capturing Subcellular Architecture in Cells, Tissues and Organisms) “Insights into Lafora Disease: Distribution of Corpora Amylacea and Their Ultrastructural Relationship with Glial Processes” GRC on Volume Electron Microscopy (Capturing Subcellular Architecture in Cells, Tissues and Organisms), “Mapping Glycogen Aggregates and Identifying Ultrastructural Changes in a Lafora Disease Mouse Model”
● Science communication:	na
● Editorial duties:	na
● Others	Posters at meetings: Mapping glycogen aggregates and Identifying ultrastructural changes in a Lafora disease mouse model. GRS and GRC on

	Volume Electron Microscopy (Capturing Subcellular Architecture in Cells, Tissues and Organisms) Mapping Glycogen Aggregates and Identifying Ultrastructural Changes in a Lafora Disease Mouse Model. Neuroscience 2025, San Diego, USA
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Javier Chicote, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	
Outreach activities	
● International collaborations:	Minoryx Therapeutics (Spain, Belgium)
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Posters at meetings: Chicote J, Rosano V, Mezzanotte M, Bertinaria M, Marini E, Vercelli V, Stanga S. A novel brain-penetrant inflammasome inhibitor as a treatment for Alzheimer's disease. XXI Italian Society for Neuroscience (SINS) Congress, Pisa, Italy (10-13/09/2025) Chicote J, Rosano V, Mezzanotte M, Bertinaria M, Marini E, Vercelli V, Stanga S. A novel drug targeting the inflammasome ameliorates AD pathology in the murine model 5xFAD. XXXV Congresso Nazionale Gruppo italiano per lo studio della neuromorfologia (GISN), Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Sveva Dallere, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Javier DeFelipe (Laboratorio Cajal de Circuitos Corticales, Madrid, Spain); J. Willem Vrijbloed (Pharmafox Therapeutics AG); Ruggero Fariello (Pharmafox Therapeutics AG); Makoto Sato and Yuichiro Oka (Osaka University, Japan)
● Invited talks:	na
● Science communication:	na

● Editorial duties:	na
● others	Posters at meetings: Dallere S, Chasseur S, Pavarino M, Sgorbini B, Boido M, Vercelli A. Terpenes and Alzheimer's Disease: in Vitro Neuroprotective Effects. SINS National Congress, Pisa, Italy (10-13/09/2025) Turégano-López M, Dallere S, Blázquez Llorca L, Merchán Pérez A, DeFelipe J. Application of Volume Electron Microscopy to the Study of Alzheimer's Disease. CTB day 2025, Madrid, Spain (19/06/2025)
Organizational activities and responsibilities at NICO:	Member of the Green Policy Committee
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Sofia Dotta, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Prof. Michaela Kress (Medical University Innsbruck), Dr. Franziska Rother (MBC, Berlin), Prof. Marc Suter (University of Lausanne, Switzerland)
● Invited talks:	na
● Science communication:	na
● Editorial duties:	na
● others	Poster at meeting: Dotta S, Muraglia L, Musso G, Testa L, Mollo A, Luzzati F, Bader M, Rother F, Buffo A, Vercelli A, Marvaldi L. Importin alpha3 controls age-dependent axonal branching via a ribosomal protein, 8th BRAYN Conference, Turin, Italy (30/10/2025) Dotta S, Musso G, Testa L, Muraglia L, Luzzati F, Parmar A, Bader M, Rother F, Kern G, Buffo A, Kress M, Vercelli A, Marvaldi L. Importin alpha3 controls age-dependent axonal branching via a ribosomal protein, EMBO workshop – Cell Biology of the Nervous System: Cellular Mechanisms of Communication, Heraklion, Greece (05/05/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

Clelia Ferrero, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	

● International collaborations:	na
● Invited talks:	na
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	na
● others	Posters at meetings: Ferrero C, Pacca P, Vercelli A, Boido M. Cerebral organoids as a model for Frontotemporal Dementia (FTD). BraYn Conference, Turin, Italy, (30-31/10/2025) Ferrero C, Pacca P, Vercelli A, Boido M. Human ipsc-derived cerebral organoids as a model for c9orf72-linked frontotemporal dementia: insights into neuronal and glial differentiation dynamics. GISN Congress, Turin, Italy (28-29/11/2025) Ferrero C, Pacca P, Vercelli A, Boido M. Modeling and Characterization of Frontotemporal Dementia via Cerebral Organoids. SINS congress Italy (10-13/09/2025) & SINS National meeting of PhD students in Neuroscience, Pisa, Italy (09/09/2025) Ferrero C, Vercelli A, Boido M. Cerebral organoids as a model for Frontotemporal Dementia: Investigating the role of C9orf72 mutation and Golgi dysfunction. Brain Organoid Summer School #BOSS, Bilbao, Spain (10-13/06/2025) Ferrero C, Vercelli A, Boido M. Cerebral organoids as a 3D model for Frontotemporal Dementia (FTD): insights into disease mechanisms. SIBS Congress , Palermo, Italy (10-13/04/2025) Ferrero C, Pacca P, Vercelli A, Boido M. Exploring the potential of cerebral organoids as a 3D model for understanding the pathological mechanisms of Frontotemporal Dementia (FTD). Medicina di precisione e personalizzata, Artificial Intelligence e biological twin, Politecnico of Turin, Turin, Italy, (19-20/03/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Member of the Young Researchers Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Ersilia Nicorvo, PhD Student

Supervised PhD students:	na
Honors, prizes, awards:	"Premio Speciale Camillo Golgi", SIBS Congress, April 2025, Palermo (Italy)
Outreach activities	
● International collaborations:	Piera Smeriglio (Paris, France)
● Invited talks:	"Neuron-tumor crosstalk in vitro: investigating co-culture systems and morphological characterization", Event "Celebrazione della costituzione della Società Italiana di Biologia Sperimentale", Pavia, Italy (27/05/2025)
● Science communication:	Member of the local organizing committee at "Olimpiadi delle Neuroscienze", Turin, Italy (15/03/2025)
● Editorial duties:	
● Others	Posters at meetings:

	Nicorvo E, Ferraro A, Boido M, Di Cunto F, Vercelli A. Neuron-tumor crosstalk in vitro: investigating co-culture systems and morphological characterization. SIBS Congress, Palermo, Italy (10-13/04/2025) Nicorvo E, Ferraro A, Boido M, Di Cunto F, Vercelli A. In Vitro Modeling of Neuron-Glioma Interactions Using Co-Culture Systems. SINS Congress, Pisa, Italy, (10-13/09/2025) Nicorvo E, Ferraro A, Pacca P, Boido M, Di Cunto F, Vercelli A. Modeling Neuron–Glioma Interactions In Vitro Using Co-Culture Systems. Brayn Congress, Torino, Italy (29-31/10/2025) Nicorvo E, Ferraro A, Pacca P, Boido M, Di Cunto F, Vercelli A. Exploring Neuron–Glioma Signaling Through In Vitro Co-Cultures. GISN Congress, Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Member of the Young Researchers Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	

Noemi Scimia, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	Prof. Clive Svendsen, Cedars-Sinai Medical Center (USA)
● Invited talks:	
● Science communication:	Participation at the event “UNIGHT - La Notte Europea delle Ricercatrici e dei Ricercatori 2025. Le neuroscienze di base in Neuroendocrinologia e Neurodegenerazione al microscopio. Gira la ruota e Gioca con noi”. Torino, Italy 26/09/2025
● Editorial duties:	na
● others	Posters at meetings: Scimia N, Carletti RM, Rasà MD, Ferrari D, Valsecchi V, Boido M, Stanga S. “Role Of Mitochondrial Dysfunction In Motor Neuron Diseases” New Horizons in Neurodegeneration, Leuven, Belgium (9-10/10/2025) Scimia N, Carletti RM, Rasà MD, Ferrari D, Boido M, Stanga S. “Mitochondrial dysfunction as a key factor of motor neuron degeneration: insights from ALS cellular model”. XXXV Congresso Gruppo Italiano per lo studio della Neuromorfologia (GISN), Torino, Italy (28-29/11/2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	na
Workshops, Schools or Conferences organized:	Member of the Young Researchers Committee of the 35th National Conference of the Italian Group for the Study of Neuromorphology (GISN), Torino, Italy (28-29/11/2025)
Technology transfer achievements (patents, etc.):	na

Maria Fernanda Veloz-Castillo, PhD student

Supervised PhD students:	na
Honors, prizes, awards:	na
Outreach activities	
● International collaborations:	BioElectron Microscopy Facility at the University of Lausanne, Switzerland
● Invited talks:	The Role of Astrocytic Glycogen Metabolism in Learning and Long- term memory (CHUV, Lausanne, Switzerland, October 2025)
● Science communication:	Organization of a global campaign to upload photographs of women in STEM into Wikimedia Commons. Organization of Wikipedia edit-a-thons to improve the biographical content of women in science.
● Editorial duties:	na
● others	Posters at meetings: Gordon Research Seminar and Gordon Research Conference on Volume Electron Microscopy, Barcelona, Spain (April 2025) Neuroscience 2025, San Diego, USA (November 2025)
Organizational activities and responsibilities at NICO:	na
Speakers invited:	na
Other organizational activities:	Collaboration with UniTo by hosting metaverse lecture across continents
Workshops, Schools or Conferences organized:	na
Technology transfer achievements (patents, etc.):	na

4. Research activity in 2025

a. Summary

We study CNS/PNS development and the neurobiological mechanisms and molecular pathways leading to normal development, neurodegeneration and pain. We are interested in neuronal cell death pathways (in development and neurodegeneration) and in the fine-tuning of brain-energy metabolism, a complex paradigm involving a strong astrocyte-neuron interplay. We are also studying drug repositioning approaches in preclinical models. We are also interested in pain signaling and neuronal regeneration.

b. Background and rationale

The study of the CNS represents a great challenge of the 21st century, and neurodevelopmental and neurodegenerative disorders provide major insights in the understanding of its anatomy, physiology and pathology and the design of new therapies. Many cellular events and mechanisms occurring during development may have profound influences on the adult nervous system, and healthy aging may be considered as the last phase of neural development.
Only a multidisciplinary and holistic approach, from molecules to brain areas, from development to disease, can provide new insights on brain function, disease and repair. Understanding the CNS development and how neurons establish synaptic connections and create networks is key to the comprehension of brain function and disease, and to design new therapeutic strategies. To this regard, astrocytes participate in tuning neuronal networks at various levels (structural, metabolic, chemical), to be fully understood. To address these questions, we compare normal brains with TG models, and we employ a range of cellular and animal models of neurodegenerative diseases developed over the years to study underlying molecular mechanisms and to

test different therapies and targeted inhibitors aimed at preventing disease and promoting brain repair at cellular, network, and behavioral levels. Within the framework of our participation in Italian PNRR projects, we are involved in the development of an iPSC and organoid based-platform for modeling neural development and diseases (AD and FTD), to identify early pathological and ultrastructural alterations (using innovative strategies such as 3D-EM) and to test novel drugs in vitro. Finally, we maintain a close connection with clinicians in order to both foster a translation from bench to bedside, and get a continuous feedback on the clinical needs. We are also involved in the search for biomarkers of disease, both to predict and to study the progression. The advancement of science does not only consist of new ideas, concepts and mechanisms to be understood but also of new tools which allow to investigate the nervous system from new points of view. Indeed, we are spending part of our time and economical efforts to cross-breed neuroscience with other disciplines, with a focus on technological improvement, since only the contamination among different forms of knowledge may provide breakthrough innovation in the field. In particular, we are improving our technology to study the microscale with 3D EM and exploit correlative microscopy approaches, in strict collaboration with Zeiss. The collection of increasing amount of data with Internet of Things and big data pose new challenges to Neuroscience and we would like to participate in this new era.

c. Objectives

We aim to understand the structural/functional building blocks of the cerebral cortex and spinal cord, and their circuitry, as substrate for CNS activities and entities, which may be disrupted in several congenital and degenerative diseases. We also aim to clarify the astrocyte-neuron metabolic interplay, by analyzing the overall morphology of individual astrocytes, through machine learning, high-throughput 3D imaging, 3D models, VR tools. We also intend to understand the communication between intrinsic and extrinsic factors in sensory neurons. Moreover, we study the neuronal death mechanisms and neuroinflammation during development and disease, and how to prevent/modulate them.
In collaboration with clinicians, we are also interested in a) the effect of (green) environment on mental health and neurodegenerative diseases, b) the glymphatic system in ageing and disease and c) the analysis of data in big international datasets such as UK biobank.

d. Results

Astrocytes-neurons structural crosstalk
CC and VC acquired a new dataset from microwave-fixed mice, analyzed using a custom-made DL network developed by MFVC, capable of automatically segment glycogen and mitochondria. The increase in glycogen positively correlates with learning, but we are increasing the n. Similarly, we are acquiring and analyzing a dataset from hippocampal dentate gyrus of Lafora Disease mice, to correlate the early presence of Lafora bodies (a dysfunctional form of glycogen) to the progression of the neurodegeneration.

Altered cellular/molecular mechanisms in SMA and therapeutic approaches
AV, MB & RS (with GM and FLR) are studying the cerebral cortex of SMA mice at different postnatal ages: compared to WT, we observed i) selective vulnerability of specific cortical projection neuron populations to SMN deficiency, associated with numerical and morphological alterations, ii) an association between SMN deficiency and neuronal dysregulation of GABAergic signaling and metabolism. In coll. with A. Usiello, we are investigating alterations in amino acid metabolism in SMA, with a focus on neurometabolic mechanisms contributing to synaptic dysfunction and disease progression (Hassan et al., 2025).
MB is also testing different therapeutic approaches. We are evaluating the efficacy of repositioned SMN-dependent (e.g Haloperidol, in coll. with Univ. Valencia/Inserm; with GM) and -independent drugs (in coll. with CNR, Naples; with DMR, EN, SC, SS), demonstrating their efficacy in counteracting neurodegeneration, neuroinflammation, muscular atrophy and mechanism of action. In 2025, we demonstrated the effective repurposing of 10-Phenothiazine (Rasà et al., 2025b).

ALS pathophysiology and therapeutic strategies
PP, AV, MB (with AT) are exploring the potential role of the glymphatic system in motor neuron diseases. MB and AV (in coll. with D. Ferrari; with DMR and CF) optimized a non-clinical protocol of ICV human neural stem cell transplantation in $SOD1^{G93A}$ mice and demonstrated the safety and efficacy (Lombardi et al.,

2025). SS & NS (in coll, with D. Ferrari) are studying how $SOD1^{G93A}$ mutation impacts on mitochondria trafficking along motor neuron axons in in vitro models by advanced live imaging.

Spinal cord injury
MB (in coll. with C. Tonda-Turo, with SC) is developing a 3D in vitro spinal cord model by bioprinting technique: we have been able to mimic the neural microenvironment, which can support neural stem cell differentiation and the formation of functional neuronal networks (Traldi et al., 2025). Further experiments are ongoing using hiPSCs and reproducing injury.

Lifestyle & Active and Healthy Ageing
AV, MB, RS (with GP) are investigating the effects of exposome on CNS (Dallere et al., 2025). Exposure to urban green environments in depressed patients induced early improvement of depressive symptoms and associated biohumoral and epigenetic changes in inflammation- and stress-related markers (Pavarino et al., 2025). With SDa, we also observed positive effects of green exposure treating *in vitro* both murine and hiPSC-derived AD neurons.
MB&DMR demonstrated that stress worsens ALS pathology and increases motor neuron vulnerability: PI3K/Akt and focal adhesion pathways are key mediators of this response (Rasà et al., 2025a).
SS, AV & MM recorded beneficial effects of long-term spontaneous exercise in mice cognition, and are now looking if this is concomitantly clearing from neuroinflammation and brain iron overload.
In coll. with M. Marraudino, MB&SC are also studying in vitro the effects of micro/nanoplastics.

Aging and Alzheimer's disease (AD)
SS, MM & JC are evaluating the iron levels available for intracellular metabolic reactions in the brain, their implications in determining cytotoxic effects, the decline in cognitive/motor skills and how mitochondria, iron and amyloid deposits interact in the brain.
In collaboration with Minoryx Therapeutics they are testing a novel molecule targeting mitochondrial dysfunctions to identify new potential therapies for AD.
MB, AV&RS (with SDa, CF, AM), are investigating early ultrastructural alterations using correlative microscopy and 3D electron microscopy in an iPSC-derived AD organoid model. Early cortical alterations are under investigation in a FTD organoid models.

Glioblastoma (GBM)
AV, MB & EN are investigating neuron–tumor interactions in GBM using in vitro co-culture models. Tumor-derived soluble factors significantly impair neuronal viability, reduce neurite outgrowth, and increase cytotoxicity. Molecular analyses reveal marked downregulation of cytoskeletal and synaptic proteins, indicating disruption of neuronal structure and function.

Investigation of Importin alpha3 (IMPA3) mutant mice in neuronal regeneration, sex and ageing
In coll. with F. Rother, LM (with SDo) is studying the role of IMPA3 in sensory and motor behavioral during age sex-based differences and after sciatic crush lesion. We performed behavioral tests and acquired paw posture pictures to monitor pain and recovery response. We collected tissues for molecular and histological analyses. We measured neurite outgrowth from embryos to adult DRG sensory neurons, studying the cross-talk between intrinsic and extrinsic mechanisms. In coll. with M. Fainzilber and I. Rishal, LM is: i) studying the role of KLF6 TF during development, ii) studying the paw posture, motor function and pain (with C.O. Pritz and SDo). In coll. with M. Suter (with SDo) we study neuropathic pain inflammation in IMPA3 mutant mice upon injury response.

e. Advancement in the field

Our group works on several hot topics in Neuroscience, such as axonal development/growth in brain physiology and pathology, study of cell complexity and interplay through 3D models, cell death and therapy. It is also involved in the study of anatomical/functional connectivity of the human brain, and how it is altered in disease. In 2025, we have obtained significant results in the field of SMA and MN diseases, by identifying

new therapeutic approaches, which extend the lifespan of the animal models of disease, improve motor performance and delay MN neurodegeneration.
In 2025, we obtained significant results in the field of exposome-related studies, including exposure to green environments, demonstrating their potential as integrative approaches for the treatment of mental health–related disorders.
The ongoing projects on digital and biological twin of the patient in the D34H will allow to change the paradigms in precision and predictive medicine. We are advancing microscopy technologies to investigate the micro- and meso-connectome, integrating confocal, light-sheet and 3D-EM within a correlative microscopy framework.

f. Publications

- Ahmad Z, Alzubaidi M, Al-Thelaya K, Calì C, Boughorbel S, Schneider J, Shah U, Househ M. Advancing open-source visual analytics in digital pathology: A systematic review of tools, trends, and clinical applications. J Pathol Inform. 2025 May;18:100454.
  Review (Q1).
- Bartesaghi L, Giangreco B, Chiappini V, Veloz Castillo MF, Monaco M, Médard JJ, Gambarotta G, Agus M, Calì C. Tight Spaces, Tighter Signals: Spatial Constraints as Drivers of Peripheral Myelination. Cells. 2025;14(12):926.
  Research article (Q1).
- Calì C, Wang X. Advances in volume electron microscopy for brain imaging: methods, applications, and affordability. Front Neurosci. 2025;19:1561852.
  Editorial (Q2).
- Cangelosi A, Riberi G, Titolo P, Salvi M, Molinari F, Ulrich L, Vezzetti E, Agus M, Calì C. Augmented reality simulation framework for minimally invasive orthopedic surgery. Comput Biol Med. 2025;189:109943.
  Research article (Q1).
- Chiappini V, Veloz Castillo MF, Biancardi F, Di Cunto F, Magistretti PJ, Vercelli A, Agus M, Calì C. Ultrastructure of astrocytes using volume electron microscopy: A scoping review. J Physiol. Epub ahead of print 2025.
  Review (Q1).
- Dallere S, Rasà DM, Pavarino G, Schellino R, Vercelli A, Boido M. The exposome from neurodevelopment to neurodegeneration: A narrative review. Neurosci Biobehav Rev. 2025 Sep;176:106247.
  Review (Q1).
- Diederich NJ, Brüne M, Allen JS, Bender N, Bruner E, Changeux JP, Calì C, et al. Buried Treasure? Overlooked and Newly Discovered Evolutionary Contributions to Human Brain Diseases. Ann Neurol. 2025 Dec;98(6):1178-1195.
  Review (Q1).
- Hassan A, di Vito R, Caretto A, Nuzzo T, D'Amico A, Panicucci C, Bruno C, Bertini E, Vercelli A, Boido M, Errico F, Pellizzoni L, Usiello A. Nusinersen corrects L-arginine deficiency in the cerebrospinal fluid of patients with severe spinal muscular atrophy. Neurobiol Dis. 2025 Oct 1;214:107046.
  Research article (Q1).
- Lombardi I, Ferrero C, Vulcano E, Rasà DM, Gelati M, Pastor D, Carletti RM, de la Morena S, Profico DC, Longobardi S, Lazzarino E, Perciballi E, Rosati JD, Martinez S, Vercelli A, Vescovi AL, Boido M, Ferrari D. Safety and efficacy evaluation of intracerebroventricular human neural stem cell transplantation in SOD1 mice as a novel approach for ALS. J Transl Med. 2025 May 9;23(1):529.
  Research article (Q1).
- Madeira R, Esteves D, Pinto N, Vercelli A, Vaz Patto M. Acceptance Factors and Barriers to the Implementation of a Digital Intervention With Older Adults With Dementia or Caregivers: Protocol for an Umbrella Review. JMIR Res Protoc. 2025 Feb 24;14:e56584.

- Pavarino G, Brasso C, Boido M, Carluccio A, Cirulli F, Mengozzi G, Schellino R, Vercelli A, Rocca P. An observational feasibility study on the impact of green exposure on major depressive episode symptomatology and inflammatory biomarkers. Front Psychiatry. 2025 Nov 10;16:1631393.
  Research article (Q1).
- Rasà DM, Stoppa I, Bérenger-Currias N, Pasho E, Ciura S, Kabashi E, Martinat C, Boido M. Stress exposure affects amyotrophic lateral sclerosis pathogenesis via PI3K/Akt and focal adhesion pathways: evidence from three experimental models. Sci Rep. 2025a May 21;15(1):17583.
  Research article (Q1).
- Rasà DM*, Stanga S*, Santonicola P, Mazzarella N, Camera A, Matino I, Zampi G, Boido M, Di Schiavi E, Vercelli A. 10H-phenothiazine exerts beneficial effects in spinal muscular atrophy in vitro and in vivo models. Sci Rep. 2025b Dec 16;15(1):45757.
  Research article (Q1).
- Riberi G, Cangelosi A, Titolo P, Dutto E, Salvi M, Molinari F, Ulrich L, Agus M, Calì C. Validation Study on Iatrogenic Nerve Damage Reduction Using Augmented Reality on Elbow Phantom. Mayo Clin Proc Digit Health. 2025;3(2):100221.
  Research article (Q2).
- Rother F, Parmar AR, Bodenhagen JS, Marvaldi L, Hartmann E, Bader M. Deficiency in KPNA4, but Not in KPNA3, Causes Attention Deficit/Hyperactivity Disorder like Symptoms in Mice. Genes. 2025 Jun 6;16(6):690.
  Research article (Q2).
- Shah U, Agus M, Boges D, Aldous H, Chiappini V, Alzubaidi M, Hadwiger M, Magistretti PJ, Househ M, Calì C. AI-guided immersive exploration of brain ultrastructure for collaborative analysis and education. Comput Graph. 2025;129:104239.
  Research article (Q2).
- Shah U, Alzubaidi M, Agus M, Calì C, Magistretti PJ, Househ M. Deep learning for brain electron microscopy segmentation: Advances, challenges, and future directions in connectomics and ultrastructure analysis. Comput Graph. 2025;121:104391.
  Research article (Q2).
- Traldi C, Chiappini V, Chasseur S, Aiello F, Boido M, Tonda-Turo C. 3D in vitro modeling of neural microenvironment through a multi-scaffold assembly approach. Mater Today Bio. 2025 Jul 14;33:102086.
  Research article (Q1).

5. Future directions and objectives for next years

a. Summary

We will exploit our previous research on i) axonal growth in the CNS and PNS, ii) the astrocytic morphology and their interplay with neurons, iii) mechanisms of neuronal death in neurodegenerative and age-associated diseases, iv) multiscale network analysis v) in vitro models of neurodegenerative diseases (iPSCs and organoids). We will continue investigating how urban green environmental spaces and natural compound inhalation affect mental health and can counteract neurodegeneration. We aim at identifying some new therapeutic targets for neurodegenerative diseases (as SMA, ALS and AD). By investigating the role of mitochondrial-iron metabolism and physical exercise in healthy aging and AD, we also aim to understand why elderly people present systemic anemia but accumulate iron in the CNS, a feature common to many neurodegenerative diseases: we plan to study the mechanisms responsible for age-dependent brain iron increase and its potential involvement in the neurodegenerative processes in AD. We also intend to investigate the PNS, by looking at pain signaling and the neurogenetics of pain: we have established DRG culture in embryos, adult and aged animals to monitor neuronal growth assay and neuronal survival upon neurotrophin stimulations in importin alpha 3 ko mice.
AV is coordinating the UNITO units involved in the PNRR project D34H, which aims to build a digital and biological twin of the patient, by analyzing clinical datasets to predict the evolution of a disease (AD, FTD, GBM), and another PNRR project, INNOVA, aimed at identifying biomarkers of disease (ALS). In the PNRR Mnesys project, we are also creating biological models in vitro from iPSCs and organoids from

patients, to identify new and early markers of disease (AD). These projects are related to personalized and precision medicine.
We are exploiting new techniques for in vitro analysis of brain development and disease modeling, such as light sheet microscopy and innovative clearing protocols, bioprinting, correlative microscopy, 3D EM, and semi-automated tool for 3D analysis in VR.

b. Background and Significance

There is a growing interest in studying the development and disease of the CNS in terms of networks: genes, miRNAs and molecular networks at a ultramicroscopic level of magnitude, synaptic networks at the microscale, and anatomical and functional networks at the meso- and macroscale. Perturbances in the networks at the different scale levels may result in developmental or neurodegenerative disorders. Considering the enormous economic and social impacts, finding a cure for neurodegenerative disorders remains a priority in science. Researchers are focused on identifying the common pathogenic processes shared among these diseases, in order to design new treatments and/or drug combinations and repurposing.
To this extent, one may refer to "damage networks": is it possible that some brain areas or specific cell types are more vulnerable than others to damage, or maybe more relevant than others for the onset of disease and of functional disorders? Such perturbances may be responsible for developmental disorders, such as schizophrenia, autism, epilepsy where there is an altered connectivity in terms of synapses and axonal connections, and of excitability. Also, neural networks may underlie the spread of neurodegenerative diseases in the CNS, such as for the Braak hypothesis of the molecular and cellular damage. Understanding the mechanisms of the onset and establishment of neural disorders at different scale levels is dramatically relevant to design neuroprotective and repair strategies to prevent and modify disease progression, and understanding the co-occurrence and overall interactions among these diseases is the first step for drug development. These strategies therefore may be at a genetic, molecular, cellular and behavioral level, and must be considered in a holistic strategy. The majority of neurodegenerative disorders have significant genetic components, with genetic heritability such as for AD, ALS and SMA that we largely study within our group.
To this aim we will collaborate with F. Di Cunto, F. Pizzagalli and F. Cauda (fMRI) to investigate the existence of damage networks in some neurodegenerative and psychiatric diseases. We are strongly connected to clinicians working within the field of neurodegenerative diseases, such as I. Rainero (Turin, AD), P. Rocca (Turin, Schizophrenia and other psychiatric disorders) and T. Mongini (Turin, SMA): we intend to continue and implement this kind of collaborations in order to have a continuous exchange of ideas, data and therapeutic strategies to favor a back and forth flow of information and bidirectional translation to find innovative therapeutic solutions.
Moreover, the fine-tuning of synaptic signaling and brain-energy metabolism is another key process and hot topic in the CNS study. The fact that neurons express the machinery allowing them to self-sustain their basic functions is counterintuitive with respect to the assumption that astrocytes undergo a plethora of supporting roles for neurons, importantly metabolic support and fine tuning of synaptic transmission via gliotransmission, two faces of the same coin. The high spatial compartmentalization of astrocytes might be the key to solve such complex interplay between the two.
AV has been awarded a grant to apply Robotics in elderly care. The project (Daisy&Ron) is coordinated by him and is in collaboration with Teoresis and Intravides companies.

c. General aim and integration with mission of the Institute

Our research aims to understand some basic mechanisms of neural development, whose alterations may be involved in the onset of neuropsychiatric diseases and of cell death in neurodegenerative diseases. We are interested in investigating the micro-, meso- and macro-scale of the CNS as the fundamental principles of brain function and disease. We are exploring the astrocyte-neuron crosstalk, to decipher whether this activity could be mediated by gliotransmission or metabolic support, and whether these two are spatially co-localized: understanding physiological processes can help to treat pathological states of the brain. Our findings are finally aimed to promote healthy aging and to develop new therapeutic strategies to prevent neurodegenerative diseases and to support brain repair and pain state. Therefore, our research is perfectly fitted to study "the interdependence between physico-chemical state of the human body and the expression of the psyche", and fully integrated with the Institute mission.

d. Specific objectives and strategies

Astrocyte-neuron interplay
Thanks to the new vEM Zeiss Volutome, we are finalizing the characterization of the Laforin-KO model (in collaboration with R. Alkhateer). Moreover, newly acquired datasets on microwave-fixed brains allowed to correlate learning and memory with glycogen turnover. We plan to further improve the field of view thanks to a strategic collaboration with the synchrotron facility in Grenoble (Lucas Benoit), to identify hotspots where to focus the analysis.

Cortical development
In collaboration with Prof. Makoto and Oka (Osaka, Japan), AV, together with SD, is studying pyramidal neuron development. Moreover, together with Prof. Pizzagalli and PP, he continues to study the development of sulci and gyri in cerebral cortex, particularly that of Rolandic Scissure, in large human datasets, correlated with neurodevelopmental diseases.

Altered mechanisms in motor neuron diseases, and therapeutic approaches
SMA: MB, AV, RS (with GI and FLR) are investigating the selective vulnerability of cortical projection neurons to SMN deficiency, aiming to identify alterations that may contribute to cortical morpho-functional changes involved in disease progression. With GM, MB will extend the studies on SMN influence on neurotransmitter metabolism and release among neurons and astrocytes in cortex and spinal cord.
With FLR (in coll. with Dr Schieweck and Viero), we continue studying the SMN role in regulating methylated RNAs in neurons, to elucidate its impact on the epitranscriptome and translatome.
Moreover, MB (with GM, EN, SC, SS) will further test SMN-independent approaches for SMA, testing in vivo repurposed drugs, also in combination with the available SMN-dep therapies.
ALS: MB and RS are collaborating with M. Rigoni (UniPD), M. Milanese (UniGE) and R. Granata (UniTO) to test novel molecules as potential therapeutic agents in ALS murine models.
SMA/ALS: SS (with MM, JCG and NS) will study mitochondrial mobility and function by targeting mitochondrial enzymes and kinases in hiPSC-MNs, in collab. with V. Valsecchi, C. Svendsen and A. Prochiantz. Moreover, PP, AV, MB (with AT) are exploring the potential role of the glymphatic system in ALS and SMA: in particular we study the contribution of AQP4 to neuronal homeostasis, particularly in the context of motor neuron degeneration.

Aging, AD and sarcopenia
SS (with MM, JCG and NS) by using cellular and animal models of healthy aging and AD will evaluate new possible mechanisms implicated in the cognitive and motor decline in AD and the possible mechanism behind the beneficial effect of long-term regular exercise during healthy aging.
In coll with F. Spyrakis, MB is setting-up an in vitro model of inflammation-induced muscle atrophy, in order to test novel anti-inflammatory compounds to counteract age-related muscle atrophy, targeting NLRP3 inflammasome pathways.

iPSCs and organoids
AV and MB (with EN, CF, SC, SDa, RS, PP) are exploiting iPSCs and organoids to identify early cellular and ultrastructural changes/alterations, as possible predictive biomarkers: to this aim, we are performing a multimodal correlative imaging on different spatial (tissue, cellular, ultrastructural) and temporal scales. With EN, we are also exploring *in vitro* the neuron-tumor interactions. AV, LM and SDo in coll. with A. Buffo and G. Gambarotta will implement iPSCs sensory neurons to identify therapeutic target markers. Human niPSCs

Exposome effects on CNS
In coll. with Prof. Rocca (UniTo) and F. Cirulli (ISS), AV, MB, RS (and AT, SD) will continue studying the effects of living in the green on depression, schizophrenia and AD, from a clinical, behavioral and biochemical marker point of view. These studies will be useful for the new Green deal program of Horizon Europe.
In coll. with M. Marraudino, we will extend our studies on the effects of micro/nanoplastics in vitro and in vivo.

Neuropathic pain
AV, LM, SDo are interested in how neuropathic pain is modulated by gender, aging, social interaction and rare disease in the peripheral nervous system. Research into this interesting interaction will unlock novel approaches to personalized pain therapy.

e. Unique features of the project research

Some of our i) research topics, ii) methodologies employed and iii) external collaborations with top institutes, scientists and biotech companies, allow us to be involved in hot topics of research. Our studies on axonal thickness and its plasticity depending on the neuronal pattern of origin/projection, and on activity represent a new field which may have very important significance not only for normal development but also for disease. Our experience on some molecular pathways related to neuronal death, such as JNK and those related to autophagy, is a specific competence which allowed us to design and test new therapeutic drugs. Moreover, the current collaborations will give us the opportunity to patent some of the tested treatments (as recently occurred for Haloperidol use in SMA).
The unique feature and ultimate goal of studying the mechanisms in age-associated neurodegeneration is to identify systemic biomarkers, prognostic of the cerebral iron status that may be predictive of cognitive impairment. An analysis of these potential markers will be conducted at the clinical level on elderly populations characterized by cognitive impairment.
The emergence of new concepts in brain function and disease in terms of networks and damage networks may be fundamental for investigating the onset of disease and eventually prevent its full development. Moreover, our group is one of the major groups working with stem cell therapy at a preclinical level in Italy and Europe.
Finally, our unique approach combining 3D models and VR has previously put our research in evidence, and we currently collaborate with a network of top-ranked scientists in the Visual Computing community, including Harvard (USA; Hanspeter Pfister), KAUST (Saudi Arabia; Pierre Magistretti, Markus Hadwiger) and Hamad Bin Khalifa University (Qatar; Marco Agus). Recent microscopes are now acquiring bigger and bigger datasets (in the range of the Tera, if not Petabytes), and to this aim we are exploring new analytical strategies using newly developed quantum computing techniques, made available on cloud (e.g., IONQ).

f. Methodology

The collaboration with the Polytechnic and INRIM (Istituto Nazionale di Ricerca Metrologica) will allow to design biosensors and lab-on-chip for the detection of biomarkers. AV&MB will further develop in vitro models of pathologies of the nervous system and tumors based on human iPSCs and organoids ("biological twin").
We will implement and further advance a multimodal correlative imaging approach to study these 3D models, integrating microscopes across scales from mm-scale live-cell light microscopy to nm-scale volume electron microscopy. These analyses will be complemented by functional electrophysiological recordings using newly acquired MEA platforms (3Brain).
SS (and MM, JCG) will combine advanced in vitro and ex vivo techniques to study mitochondrial dynamics during aging and dementia: with live imaging, we will trace and reconstruct mitochondrial networks, and by 2PM on organotypic cultures of brain slices of mice models we will investigate mitochondrial dysfunctions related to the amyloid pathology.
We are using neuronal tracers, light sheet microscopy and 3D EM to generate tridimensional volume visualizations of brains, showing the alterations in cortical architecture and in neuronal projections of SMA animals at different postnatal ages (AV, MB, RS together with GM, GI and FLR), as well as structural astrocytes/neurons interplay (CC). Indeed, the new Zeiss Volutome is now available and fully functional, after the installation and setup phase: this system is the state of the art for volume Electron Microscopy, and is able to automatically acquire thousands of serial section images, unsupervised. All these techniques will also require development of novel visualization and analysis techniques that will be developed using the aid of VR.
The collaboration with prof. Cauda (Psychology Dept.) will allow using voxel-based morphometry, fMRI and tractography in ageing subjects. A collaboration is also under discussion to develop a neuroinformatic approach in studies of neurodegenerative diseases with F. Di Cunto, F. Pizzagalli and P. Provero.

Fondazione Cavalieri Ottolenghi

Neuroscience Institute Cavalieri Ottolenghi

Internal Annual Report 2025

Laboratory name: Clinical Neurobiology

1. LABORATORY DESCRIPTION – PERSONNEL:

Principal Investigator

Di Sapio, Alessia
- Degree: MD
- Birthdate: 22/10/1966
- Phone: +39 011 9026 697
- Email: a.disapio@sanluigi.piemonte.it
- Position: Head of the Regional Reference Center for Multiple Sclerosis (CRESM)
- Role: Principal Investigator

Personnel

Sala, Arianna
- Degree: MSc, Board Certification
- Birthdate: 22/05/1972
- Phone: +39 011 670 6601
- Email: sala.arianna72@gmail.com
- Position: Resident Medical Biologist at the SCDO Neurology Unit, AOU San Luigi Gonzaga, Orbassano
- Role: CSF analysis, diagnostic and prognostic tests for Multiple Sclerosis (MS) and Neuromyelitis Optica (NMO), drug immunogenicity

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Valentino, Paola (since 17/02/2025)
- Degree: MSc, Board Certification
- Birthdate: 11/08/1981
- Phone: +39 011 670 5576
- Email: p.valentino@sanluigi.piemonte.it
- Position: Medical Biotechnologist
- Role: Management of the CRESM Biobank, translational research in fluid biomarkers for Multiple Sclerosis, diagnostic tests for MS

Bava, Cecilia Irene (until 31/05/2025)
- Degree: MSc
- Birthdate: 25/11/1996
- Phone: +39 011 670 6635
- Email: cecilia.bava@edu.unito.it
- Position: Molecular Biotechnologist
- Role: Sample processing and storage in the CRESM Biobank, diagnostic tests for MS patients

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Giorgi, Lucia
- Degree: MSc
- Birthdate: 12/06/1995
- Phone: +39 011 670 6635
- Email: lucy.giorgi@gmail.com
- Position: Cellular and Molecular Biologist
- Role: Diagnostic tests for MS patients, sample processing and storage in the CRESM Biobank

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Aghaei Kouhi, Komeil
- Degree: MSc
- Birthdate: 03/09/1994
- Phone: +39 011 670 6635
- Email: komeil.aghaeikouhi@unito.it
- Position: Cellular and Molecular Biologist

- Role: Sample processing and storage in the CRESM Biobank, research activities, and diagnostic tests for MS patients

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Bertolotto, Antonio
- Degree: MD
- Birthdate: 12/02/1952
- Phone: +39 011 670 6600
- Email: antonio.bertolotto@gmail.com
- Position: Voluntary Visitor (Expert in Multiple Sclerosis)
- Role: Head of the Regional Reference Center for Multiple Sclerosis (CRESM) until 01/04/2021

2. CURRENT GRANTS

Starting-end date	Project Title and ID	Beneficiary	Funding Program/Agency	Role of the unit	Overall Amount Funded	Managed by FCO/UNITO
2023 -2025	Ruolo dei NFL nella diagnosi e monitoraggio dei pazienti con Sclerosi Multipla: il progetto pilota del CRESM	Alessia Di Sapio	Roche	PI	100,000 Euro	FCO
2025-2026	Studio dei Neurofilamenti sierici nei pazienti con Sclerosi Multipla e NMOSD nella pratica clinica corrente	Alessia Di Sapio	Roche Diagnostics	PI	40.000 Euro	FCO

3. SCIENTIFIC ACTIVITIES IN 2025

Alessia Di Sapio (acting PI)

Supervised PhD students:	NA
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	
• Invited talks:	Speaker at the Conference: “L’utilizzo precoce degli HET nella sclerosi Multipla, Cherasco 15/3/2025 Speaker at the Conference “Ravulution: l’evoluzione del trattamento NMOSD”, Venezia 26/3/2025 Speaker at the Conference “Space of care: Le opportunità di Cladribina. Focus sugli stili di vita del paziente con SM” Torino, 20/5/25 Speaker at the conference “Brain Matters”, Venezia 6/11/2025

	Speaker at the webinar “Mavenews 4th edition”, 25/11/2025
• Science communication:	Outreach event for MS patient “Alimentazione e attività fisica: strategie di benessere nella sclerosi multipla”, Orbassano 13 giugno 2025
• Editorial duties:	
• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	Scientific Director and speaker of the Preceptorship “Biomarcatori Fluidi in SM: il Laboratorio a fianco del Neurologo nella personalizzazione della Cura”, Orbassano 16-17/01/2025 Scientific Director and speaker of the Preceptorship “Biomarcatori Fluidi in sclerosi multipla e malattie neurologiche immunomediate”, Orbassano 13-14/11 2025 Chair of the Scientific Committee of the Conference “Nord-Ovest Network Meeting: La Neuroinfiammazione: misurarla e combatterla nella Sclerosi Multipla e in altre sue manifestazioni”. Torino, 31/1/2025 Scientific Director at the webinar: “Sclerosi Multipla, Terapia ad alta efficacia e PML: la gestione del rischio nella pratica clinica”. 17/12/2025
Technology transfer achievements (patents, etc.):	

Arianna Sala, Resident Biologist

Supervised PhD students:	NA
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	
• Invited talks:	- Biomarcatore: dalla ricerca alla validazione di una metodica e sua applicazione clinica. Orbassano 16-17/01/2025 -I biomarcatori nella NMOSD: tecniche di rilevazione a confronto. Orbassano 16-17/01/2025 -MS Dimension: evoluzione ed esperienze in Sclerosi Multipla. Relazione dal titolo:Immunowell-Dati RWE-. Napoli 13-14 Maggio 2025 - Relatore unico all’evento “Training su test ELISA e confront IW e stratify” Milano 17/09/2025 - I biomarcatori nella NMOSD: tecniche di rilevazione a confronto. Orbassano 13-14 Novembre 2025 -Il rebus degli ab anti JCV: confronto Immunowell e stratify. Orbassano 13-14 Novembre 2025 - Il test per gli ab anti AQP4: intervento nel meeting “La rete piemontese per la NMOSD: opinioni a confronto” Torino 19/11/2025

	- speaker at the webinar: “Sclerosi Multipla, Terapia ad alta efficacia e PML: la gestione del rischio nella pratica clinica”. 17/12/2025
• Science communication:	
• Editorial duties:	Report scientifico sul tema: Farmaci anti CD-20 nella SM. 41th ECTRIMS Meeting, Barcellona, September 24-26.
• others	Presenter of a poster titled: "Longitudinal dinamics of serum NFL and GFAP in AQP4 NMOSD patients: a real-world results of a 3 to 10 years follow up" at 41th ECTRIMS Meeting, Barcellona, September 24-26
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Paola Valentino, Resident Biologist, Medical Biotechnologist

Supervised PhD students:	- Cecilia Irene Bava, Postgraduate Medical Specialization (Residency) in Clinical Pathology and Clinical Biochemistry - Gada Mohamed Abdel Azim, Postgraduate Medical Specialization (Residency) in Clinical Pathology and Clinical Biochemistry
Honors, prizes, awards:	
Outreach activities	
• International collaborations:	
• Invited talks:	- Biomarcatori fluidi in Sclerosi Multipla e Malattie Neurologiche Immunomediate: “La Banca Biologica: engagement del paziente e Standard di qualità dei materiali e del dato clinico”; “NFLs: dalla ricerca alla validazione di un test e alla sua applicazione clinica; tecniche di rilevazione a confronto” (AOU San Luigi, 14/11/2025)
• Science communication:	- Relazione “La Biobanca del CRESM a servizio della ricerca sulla Sclerosi Multipla”, webinar “Importanza delle Reti e del Networking nel Biobanking” (Biotech week 2025) - Poster “Il tesoro delle Biobanche” (INTOtheBRAIN Le Neuroscienze di domani, NICO 3/04/2025)
• Editorial duties:	
• others	- First and presenting author of Poster “The Treasure of Biobanks: How CRESM Biobank Serves Multiple Sclerosis Research” (41st ECTRIMS Congress, 24-26/09/2025) - Presenting author of Poster “Establishment and validation of normative values for serum glial fibrillary acid protein (sGFAP) in a healthy Italian cohort” (41st ECTRIMS Congress, 24-26/09/2025) - Participation in Advisory Board “Potenziale applicazione clinica del test Elecsys NfL nella gestione della Sclerosi Multipla” (Roche, 13/10/2025)

	- Participation in the ELSI Working Group "Biobancaggio/Biobanche di ricerca e interazione con il garante" - Participation in the "Biobanking" Regional Working Group of DAIRI (Dipartimento Attività integrata Innovazione e Ricerca, Azienza Ospedaliera Alessandria)
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	Quality Management of CRESM biobank
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

Cecilia Bava, Molecular Biotechnologist

Supervised PhD students:	NA
Honors, prizes, awards:	Winner of Best Poster Award at FISM Annual Congress 2025 (Poster "Implementation of GFAP, in addition to NFL, in progressive multiple sclerosis patients: a potential tool for objective measuring of clinical and patient reported outcomes"), sharing results from FISM Fellowship 2023 project
Outreach activities	
• International collaborations:	NA
• Invited talks:	Presentation at XXXIII AINI Congress 2025 titled "Longitudinal serum NFL and GFAP dynamics in AQP4 positive and negative NMOSD patients treated with Rituximab and Tocilizumab: a real-world experience", 5-8 May 2025
• Science communication:	NA
• Editorial duties:	"NFL: focus sul test journey" (editorial project with Novartis)
• others	Presenter of a poster titled: "Longitudinal serum NFL and GFAP dynamics in AQP4 positive and negative NMOSD patients treated with rituximab and tocilizumab: a real-world experience" at Congress Disability Progression in MS, NMO and MOGAD: From Bench to bedside, Verona, 20-21 March 2025 Presenter of a poster titled: Comparison between technologies and biological approaches for CSF and serum NFL quantification: a further step towards implementation in clinical practice", XXXIII AINI Congress 2025, 5-8 May 2025 Presenter of a poster titled: " BB-CRESM: a structured institutional biobank for quality research in Multiple Sclerosis" at Europe Biobank Week Congress 2025 (EBW 2025), 13-16 May 2025 All-year-long participation to DAIRI Biobank Group activities to coordinate Biobanks in Piedmont.
Organizational activities and responsibilities at NICO:	NA
Speakers invited:	NA

Other organizational activities:	NA
Workshops, Schools or Conferences organized:	NA
Technology transfer achievements (patents, etc.):	NA

Lucia Giorgi, Cellular and Molecular Biologist

Supervised PhD students:	NA
Honors, prizes, awards:	NA
Outreach activities	
• International collaborations:	NA
• Invited talks:	NA
• Science communication:	NA
• Editorial duties:	NA
• others	Presenting author for poster titled *"sNFL as additional biomarker for clinical management of multiple sclerosis*", XXXIII AINI Congress 2025, 5-8 May 2025 Presenting author for poster titled *"BB-CRESM: a structured institutional biobank for quality research in Multiple Sclerosis*", XXXIII AINI Congress 2025, 5-8 May 2025 Participation in the "Biobanking" Regional Working Group of DAIRI (Dipartimento Attività integrata Innovazione e Ricerca, Azienza Ospedaliera Alessandria)
Organizational activities and responsibilities at NICO:	NA
Speakers invited:	NA
Other organizational activities:	NA
Workshops, Schools or Conferences organized:	NA
Technology transfer achievements (patents, etc.):	NA

Komeil Aghaei, Cellular and Molecular Biologist

Supervised PhD students:	NA
Honors, prizes, awards:	NA
Outreach activities	
• International collaborations:	NA
• Invited talks:	NA
• Science communication:	NA
• Editorial duties:	NA
• others	NA
Organizational activities and responsibilities at NICO:	NA
Speakers invited:	NA
Other organizational activities:	NA
Workshops, Schools or Conferences organized:	NA
Technology transfer achievements (patents, etc.):	NA

Antonio Bertolotto, MD

Supervised PhD students:	NA
Honors, prizes, awards:	NA
Outreach activities	
• International collaborations:	NA
• Invited talks:	Neurowebinar 7 aprile 2025 Curems.net: il trapianto autologo di cellule staminali emopoietiche come potenziale strategia per guarire la Sclerosi Multipla.
• Science communication:	Creator and manager of curems.net, a website dedicated to aHSCT (autologous Hematopoietic Stem Cell Transplantation) in MS (Multiple Sclerosis)
• Editorial duties:	European Editor.in Chief of Neurology and Therapy, impact factor 4.8, Quartile Q1
• others	
Organizational activities and responsibilities at NICO:	
Speakers invited:	
Other organizational activities:	
Workshops, Schools or Conferences organized:	
Technology transfer achievements (patents, etc.):	

4. Research activity in 2025

a. Summary

CReSM Laboratory has developed two complementary research streams:

- translational biomarker studies for monitoring Multiple Sclerosis (MS) and other neurological autoimmune disesases, as NMOSD, mainly focusing on serum neurofilament light (sNFL) and glial fibrillary acidic protein (GFAP). Normative values and longitudinal validation enhance a more personalized clinical management
- CReSM biobank (BB-CRESM) activity, supporting research at CReSM and across national and international centres in MS and autoimmune neurological diseases.

b. Background and rationale

Neurological Biomarkers

Since 2019, the CReSM Laboratory has focused on the study of sNFL using ultrasensitive Simoa technology. This enabled progressive implementation of sNFL testing in clinical practice, supported by extensive experience from pilot studies and several publications

More recently, biomarker analysis has expanded to glial fibrillary acidic protein (GFAP), increasingly recognized as a potential prognostic marker for early disease progression and clinical severity in MS. Establishing normative values allows the translation of GFAP testing into clinical practice, currently in an exploratory context.

The laboratory established internal normative ranges, benchmarked them against international cohorts, and validated them on internal patient samples, a necessary step for individualized data interpretation. Internal guidelines now define periodic biomarker monitoring for patient subgroups based on treatment type and/or clinical phenotype. Additionally, biomarker measurements are performed on-demand by neurologists in specific clinical scenarios.

This strategy, integrated into comprehensive multidimensional clinical assessments, provides added value for individual patient management, often guiding timely clinical decisions. Furthermore, the project enabled CReSM to extend its expertise in sNFL and GFAP measurement across the Piedmont MS Network.

Further, interesting data are related to pregnancy. Elevated sNFL levels have been observed postpartum in women with pre-eclampsia or MS; however, definitive CNS injury has not been established. Data on sNFL levels in healthy women with physiological pregnancies are currently lacking, highlighting an important area for investigation aimed to differentiate pathological from physiological sNFL increase in MS women after delivery

Biobank activities.

Research biobanks are legally established, non-profit entities that collect, store, and share human biological samples and associated data under high-quality standards, respecting participants' rights and informed consent. Sharing samples and data is central to fostering collaborative scientific research.

In 2020, the Regional Reference Center for Multiple Sclerosis (CReSM) at San Luigi Gonzaga University Hospital officially established BB-CReSM,with support from the Italian Multiple Sclerosis Foundation (FISM). BB-CReSM is the first Italian biobank specifically focused on MS and other autoimmune neurological diseases, aiming to advance research in these areas.

The biobank operates under structured governance and documented procedures, including participant information and consent forms, standard operating procedures, privacy assessments, and MDTA agreements for sample and data transfer, all approved by hospital management, the ethics committee, and the data protection officer. BB-CReSM is integrated into national and international biobanking infrastructures (BBMRI.it and BBMRI-ERIC), being the first biobank in Piedmont to join these networks.

c. Objectives

The main objectives divided into the previously described research streams were:

Neurological biomarkers

- Implement longitudinal biomarker monitoring for individualized clinical management in MS and NMOSD.
- Evaluate biomarkers' ability to detect disease activity, therapy response, and transition to progressive MS.
- Investigate the dynamics of postpartum sNFL increase in healthy women with physiological pregnancies and assess associated CNS involvement by measuring GFAP, Tau, and UCHL1 in parallel.

Biobank activities supporting research

- Maintain and implement quality standard-compliant procedures for sample and data management.
- Ensure quality control, traceability, and ethical access to biological materials.
- Provide samples and data to support national and international research, fostering translational studies and collaborative networks.

d. Results

The main results divided into the previously described research streams were:

Neurological Biomarkers

- Normative values: Normative ranges for sNFL and GFAP were established from 139 healthy controls (sGFAP at 50th, 75th, and 90th percentiles).
- Clinical validation
    - In progressive patients, GFAP, alone or in combination with NFL, captured cognitive function worsening over time andpredicted cognitive worsening

- In NMOSD patients, GFAP levels >75th percentile were more frequently associated to active disease attacks
- Longitudinal analysis of 15 NMOSD patients demonstrated correlations between biomarker levels, AQP4 status, clinical activity, radiological outcomes, and therapy response.

- Clinical implementation: The data generated from clinical validation studies described above have enabled the implementation of a patient-centred biological monitoring strategy, tailored to disease type, treatment regimen, and disease stage. This approach integrates biomarker measurements with multidimensional clinical assessments, supporting individualized decision-making and facilitating early detection of disease activity or progression.
- Pregnancy studies: Longitudinal analysis of 61 physiological pregnancies revealed substantial increases in sNFL and Ubiquitin C-terminal Hydrolase L1 (sUCHL1) after delivery. Median sNFL levels doubled on postpartum day 1 (19.84 pg/mL; range 5.02–66.19) compared to pre-delivery (8.86 pg/mL; 1.88–34.62), and further increased at days 6–10 (28.18; 7.61–48.83), before normalizing within 40 days. The type of delivery (vaginal vs. C-section) had no significant impact. sNFL levels decreased significantly with increasing parity a pattern also observed for sUCHL1, GFAP, and Tau. sUCHL1 showed a sharp increase from a median of 24.51 pre-delivery to 153.69 on postpartum day 1, returning to baseline by days 6–10. These findings reflect a phenomenon, that exhibits the following characteristics: a) Elevated post-partum sNFL, suggesting neuronal involvement. B) Elevated sUCHL1, suggesting involvement of neurons and/or lactotroph cells of the anterior pituitary; c) Triggered by parturition; d) Parity-dependent attenuation, as the intensity of the increase diminishes with higher parity; e) Self-limiting course, as it resolves within approximately 40 days f). It does not result in any evident adverse health outcomes

Biobank activities

- Quality compliance and infrastructure: BB-CReSM implemented quality-compliant procedures, including EasyTrack 2D® software for traceability, consent management, updated documentation, MDTA agreements, and recruitment of dedicated staff.
- Sample and data holdings: The biobank stores >165,000 aliquots from >3,450 participants (≈70% MS, 10% NMOSD/MOGAD, 18% other neurological conditions, 2% healthy controls), including serum, plasma, whole blood, RNA, blood cells, and CSF. Samples are collected at diagnosis, during therapy, and at relapse, ensuring longitudinal coverage.
- Quality control and accessibility: BB-CReSM maintains rigorous quality assessments for samples, data, and equipment.
- Biobank sample sharing for open Science. Over 200 samples have been distributed in years 2024-25 for external national and international research projects, including collaborations with University of Basel, Federico II Naples, and Sapienza University of Rome. In addition CRESM Biobank supported all research studies performed at CRESM, as described in the previous paragraph
- Impact: BB-CReSM supports translational research, enabling studies on disease mechanisms, biomarker discovery, and treatment response, reinforcing the centre's role as a hub for collaborative and multicentre neurological research.

e. Advancement in the field

CReSM Laboratory advanced the clinical integration of sNFL and GFAP as minimally invasive biomarkers for MS and NMOSD. Data generated at CReSM enabled a well-defined patient-centred biological monitoring strategy, tailored to disease type, treatment regimen, and disease stage, which is currently applied in clinical practice to support personalized management, therapy evaluation, and early detection of progression. Additionally, longitudinal studies in healthy pregnancies provide the first detailed characterization of sNFL, sUCHL1, GFAP, and Tau dynamics, informing interpretation of postpartum biomarker changes in women with CNS conditions such as MS.

BB-CReSM ensures quality-compliant, high-quality storage and traceability of biological samples while respecting participants' rights. Active distribution of samples and data promotes national and international collaborations, enabling translational studies, biomarker discovery, and deeper understanding of autoimmune neurological diseases. Recognized as a model of excellence and integrated into major scientific networks, BB-CReSM fosters transparency, collaboration, and the integration of research findings into clinical practice.

f. Publications

- Meloni C, Marnetto F, Fagnani C, Benincasa L, Galano D, Valentino P, Martire S, Di Sapio A, Bertolotto A, Repice A, Ballerini C, Mancosu C, Lorefice L, Frau J, Cocco E, Veroni C. EBV dysregulation is associated with immune imbalance in multiple sclerosis: evidence from integrated viral and host analyses. Neurology® Neuroimmunology & Neuroinflammation. NXI-2025-200864R1. Research article (Q1).
- Bellisario V, Squillacioti G, Valentino P, Marozio L, Bertolotto A, Bono R. Oxidative stress and multiple sclerosis: trajectories and possible predictive activities of pro and antioxidant biomolecules. Free Radic Biol Med. 2025 Dec 16; 241:582-586. doi: 10.1016/j.freeradbiomed.2025.09.052. Epub 2025 Oct 3. PMID: 41046943
  Research article (Q1
- Novi G, Bovis F, Sciolla C, Frau J, Minetti M, Napoli F, Vianello M, Greco G, Sechi E, Bellomo A, Garelli P, Spiezia AL, Fantozzi R, Schirò G, Ghezzi L, Zecca C, Signoriello E, Brambilla L, Lucchini M, Bonavita S, Schiavetti I, Malentacchi M, Cocco E, Franceschini A, Mataluni G, Gastaldi M, Rolando A, Solla P, Cellerino M, Abbadessa G, Lanzillo R, Di Sapio A, Uccelli A, Sormani MP. Long-Term Effectiveness and Safety of Rituximab in Neuromyelitis Optica Spectrum Disorder: A 5-Year Observational Study. CNS Drugs. 10.1007/s40263-025-01191-7. Advance online publication. https://doi.org/10.1007/s40263-025-01191-7
  Research article (Q1).
- Malucchi S, Perini P, Rinaldi F, Radaelli M, Malentacchi M, Bertolotto A, Di Sapio A. (2025). Disease Course in Patients Switched from Natalizumab to Alemtuzumab: An Italian Multicenter, Prospective, Observational Study. *Neurology and therapy*, 10.1007/s40120-025-00754-6. Advance online publication. https://doi.org/10.1007/s40120-025-00754-6
  Research article (Q1).
- Centonze D, Di Sapio A, Brescia Morra V, Colombo E, Inglese M, Paolicelli D, Salvetti M, Furlan R. (2025). Steps toward the implementation of neurofilaments in multiple sclerosis: patient profiles to be prioritized in clinical practice. *Frontiers in neurology*, *16*, 1571605. https://doi.org/10.3389/fneur.2025.1571605
  Research article (Q1
- Mechelli R, Umeton R, Bellucci G, Bigi R, Rinaldi V, Angelini DF, Guerrera G, Pignalosa FC, Ilari S, Patrone M, Srinivasan S, Cerono G, Romano S, Buscarinu MC, Martire S, Malucchi S, Landi D, Lorefice L, Pizzolato Umeton R, Anastasiadou E, Trivedi P, Fornasiero A, Ferraldeschi M, IMSGC WTCCC2, Di Sapio A, Marfia G, Cocco E, Centonze D, Uccelli A, Di Silvestre D, Mauri P, de Candia P, D'Alfonso S, Battistini L, Farina C, Magliozzi R, Reynolds R, Baranzini SE, Matarese G, Salvetti M, Ristori G. (2025).). A disease-specific convergence of host and Epstein-Barr virus genetics in multiple sclerosis. *Proceedings of the National Academy of Sciences of the United States of America*, *122*(14), e2418783122. https://doi.org/10.1073/pnas.2418783122
  Research article (Q1).

5. Future directions and objectives for next years

a. Summary

Over the past year, NICO has consolidated the integration of fluid biomarkers into the clinical practice of CReSM neurologists, with a particular focus on sNFL for monitoring neurodegeneration in multiple sclerosis (MS). Future directions aim to extend the use of GFAP measurements, exploring their relationship with sNFL

levels and clinical disease progression. The ultimate goal is to jointly assess neuroinflammation and neurodegeneration over time to support monitoring of "smoldering disease."
In parallel, we plan to actively promote the use of fluid biomarkers across the North-West Network through training and collaborative initiatives, harmonizing patient management. The potential utility of new microglial activation biomarkers, including CHI3L1, will also be explored to better characterize compartmentalized neuroinflammation.

Regarding CRESM Biobank, future activities include strengthening storage infrastructures, expanding the collection of biological samples, fostering collaborations with other centers, and implementing quality systems in line with BBMRI-ERIC standards and ISO 20387 accreditation. These efforts will support translational research and the monitoring of innovative therapies, particularly in progressive MS, NMOSD, and MOGAD patients.

b. Background and Significance

Management of neuroimmunological diseases, particularly multiple sclerosis (MS), is evolving toward increasingly personalized approaches based on sensitive diagnostic and predictive tools. Fluid biomarkers, such as sNFL and GFAP, provide complementary insights into neurodegenerative and neuroinflammatory processes, allowing dynamic assessment of disease activity over time. The integration of sNFL in CReSM clinical practice has already enabled more precise monitoring of disease progression and treatment response.

However, compartmentalized neuroinflammation, a key driver of "smoldering disease," requires additional specific biological indicators. Microglial activation biomarkers, such as CHI3L1, can identify subclinical inflammatory activity, offering the opportunity to detect disease processes not captured by conventional assessments.

CRESM Biobank is a strategic resource for translational research, providing high-quality biological samples for clinical and preclinical studies. Standardized collection, storage, and traceability are essential to ensure reproducibility, facilitate multi-centre collaborations, and strengthen the Institute's role as a research hub. Implementing BBMRI-ERIC quality guidelines and achieving ISO 20387 accreditation will further enhance operational standards and the value of the Biobank.

Current scientific evidence emphasizes the need to integrate fluid biomarkers, clinical data, and biological samples into a multidimensional approach. Such integration supports personalized therapeutic decisions, improves patient management, and fosters innovative neuroimmunological research. NICO's future strategies align with this vision, combining clinical practice, translational research, and high-quality biological data to reinforce the Institute's position as a centre of excellence in neuroscience.

c. General aim and integration with mission of the Institute

The general aim is to consolidate and expand the integration of fluid biomarkers and biological data into clinical practice and translational research to support increasingly personalized patient management. This aligns with NICO's mission by promoting scientific and clinical excellence, advancing innovation in diagnosis and disease monitoring, and strengthening high-quality research infrastructures in neuroscience.

d. Specific objectives and strategies

I. Expansion of fluid biomarker use in clinical practice and comparison of different analytical platforms

- Implement GFAP measurements and analyse their integration with sNFL and clinical data to monitor neurodegeneration and neuroinflammation over time.
- Perform a systematic comparison of different analytical platforms and methodologies for the measurement of neurological fluid biomarkers (including sNfL, GFAP, Tau, and related markers).
- Assess analytical performance, sensitivity, reproducibility, and inter-method agreement across assays and sample matrices.

- Validate selected methods in real-world clinical settings to support standardization, reliability, and feasibility of biomarker implementation in routine neurological practice.
- Facilitate the translation of ultra-sensitive biomarker technologies from research to clinical decision-making, promoting harmonized use across centres.

II. Promotion and harmonization of patient management across the North-West Network

- Organize training sessions and conferences to disseminate knowledge on fluid biomarkers, share clinical guidelines, and harmonize practices among centres.
- Establish a clinical network for monitoring and sharing multicentre data.

III. Investigation of new microglial activation biomarkers

- Study CHI3L1 and other markers to better characterize compartmentalized neuroinflammation.
- Integrate these findings into clinical-biological models to optimize therapeutic strategies and support clinical trials.

IV. Integration of clinical, translational, and innovative research

- Develop studies combining clinical data, fluid biomarkers, and biological samples to identify new indicators of disease progression.
- Use multidimensional approaches to improve prediction of treatment response and support personalized clinical trials.

V. Implementation of pregnancy-related biomarker studies

- Characterize serum dynamics of sNfL, UCHL1, GFAP, and Tau in women with physiological pregnancies across trimesters, the day before delivery, and early postpartum.
- Determine whether significant changes occur in these biomarkers during the peripartum period in healthy women, focusing on potential postpartum sNfL elevations previously observed in pathological contexts.
- Establish reference intervals for sNfL, UCHL1, GFAP, and Tau levels in physiological pregnancies throughout gestation and the early postpartum period.
- Compare biomarker levels between women with physiological pregnancies and women with MS during pregnancy, following the same time points.
- Explore sNfL, UCHL1, GFAP, and Tau in cerebrospinal fluid samples obtained during cesarean deliveries to provide preliminary data on CNS biomarker levels in this context.

  Overall, this study aims to deepen the understanding of neurological stress and damage biomarkers in the peripartum period in healthy women and compare them with changes observed in women with MS, helping to distinguish physiological pregnancy-related variations from potential neurological pathology.

VI. Biobank enhancement and infrastructure development

- Expand biological sample collection, including patients with progressive MS, NMOSD, and MOGAD, to support innovative research.
- Implement quality systems in line with BBMRI-ERIC Audit Programme and ISO 20387, updating SOPs, PIA, and operational documentation.
- Acquire backup systems to ensure optimal sample preservation.
- Establish collaborations with national and international centres to increase sample availability, promote translational research, and enhance NICO's scientific visibility.

e. Unique features of the project research

I. Integration of longitudinal fluid biomarkers with clinical monitoring

- The project uniquely combines serial measurements of sNfL, GFAP, and novel microglial activation markers (e.g., CHI3L1) with detailed clinical data over time.
- This approach allows simultaneous monitoring of neurodegeneration and neuroinflammation in patients with MS and related disorders, providing a multidimensional view of "smoldering disease" not captured by conventional assessments.

II. Peripartum biomarker study in healthy women and MS patients

- A distinctive component of the project is the systematic evaluation of serum and cerebrospinal fluid biomarkers during physiological pregnancy and early postpartum, with high temporal resolution (multiple time points across gestation and postpartum).
- Comparative analysis with pregnant women with MS provides a unique framework to distinguish physiological pregnancy-related changes from disease-related signals, a question rarely addressed in current research.

III. Comprehensive biobanking and high-quality data integration
- The project strengthens the Biobank infrastructure and implements advanced quality standards (BBMRI-ERIC and ISO 20387), ensuring high-quality, traceable biological samples.
- The combination of biobanking, standardized sample collection, and integrated clinical-biological datasets supports robust translational research and facilitates multi-centre collaborations.

IV. Focus on translational and personalized medicine
- By linking biomarker dynamics to clinical outcomes and treatment response, the project enables predictive modelling and a personalized approach to therapy in MS and other neuroimmunological diseases.
- The integration of novel microglial markers, peripartum studies, and longitudinal biomarker profiling positions the project at the forefront of personalized neuroimmunology research.

f. Methodology

The Clinical Neurobiology Laboratory and CReSM are equipped with state-of-the-art technologies for ultra-sensitive biomarker detection, including the SR-X Ultra-Sensitive Biomarker Detection System (Quanterix), acquired in 2018. This benchtop instrument uses Simoa bead technology, which isolates individual immunocomplexes on paramagnetic beads and traps single molecules in femtoliter-sized wells, enabling a "digital" readout. This provides up to 1000-fold greater sensitivity than conventional immunoassays, with CVs below 10%, allowing detection of biomarkers at sub-femtomolar concentrations (<1 pg/mL).

The SR-X supports multiple sample types, including serum, plasma, cerebrospinal fluid (CSF), and cell lysates, and allows multiplexed detection of up to four biomarkers per sample with minimal volumes, maximizing throughput and preserving precious patient material. It is applied across neurology, oncology, cardiology, inflammation, and infectious diseases.

In neurology, the SR-X is widely used to quantify neurofilament light chain (NFL) and other axonal damage proteins in CSF and blood, including physiological NFL levels in healthy individuals. Additional biomarkers measurable include GFAP, Tau, Aβ42, Aβ40, and alpha-synuclein, with the possibility to develop custom assays when specific antibodies are available.

These advanced methodologies are fully integrated with the CRESM Biobank, which provides high-quality, well-characterized biological samples. The combination of ultra-sensitive detection and a robust biobanking platform enables comprehensive, longitudinal studies of neurological biomarkers, supporting translational research and personalized patient management.

NeuroWebinar & Seminar 2025

1 appointment per week, on Friday at 2.00 pm
**Hybrid seminar*: both in presence and on webex*

Friday 17/1/2025 h. 2.00 pm - Hybrid seminar

Salvatore Adinolfi , Dipartimento di Scienza e Tecnologia del Farmaco, Università di Torino

Frataxin: a biochemistry tale from molecular to pathophysiology of neurodegeneration

Frataxin, a highly conserved protein found in prokaryotes and eukaryotes, is required for efficient regulation of cellular iron homeostasis. Humans with a frataxin deficiency have the cardio- and neurodegenerative disorder Friedreich's ataxia, commonly resulting from a GAA trinucleotide repeat expansion in the frataxin gene.

I will revisit the most significant milestones obtained with biochemical and biophysical approaches that have led us to our current understanding of frataxin and its functions. While frataxin function remains a point of controversy, a clearer picture emerges suggesting that frataxin is an essential element of the crucial Iron-sulfur cluster biogenesis pathway.

Host: Ferdinando Di Cunto

Thursday 30/1/2025 h. 2.00 pm - Hybrid seminar

SEMINAR CYCLE of the PhD in Neuroscience of Turin

Paolo Malatesta, University of Genoa and IRCCS Policlinico San Martino, Genoa

Tracing Clonal Evolution and Immune Evasion in Glioblastoma Progression

Human Glioblastoma remains a significant challenge in oncology research, with its early progression remaining notably elusive. In a recent study, we traced the clonal dynamics of glioblastoma evolution by co-introducing PDGFB and genetic barcodes into mouse brains, observing a sustained loss of clones during the transition to a malignant state—an effect tied to shifts in c-Myc expression levels and their downstream targets. We then extended our investigation of glioblastoma evolution by transplanting multiclonal, early- stage glioma cells into multiple immunodeficient NOD-SCID mice.

This approach allowed us to follow clonal behavior across serial transplants, revealing the

acquisition of immune-evasive capabilities by early-stage glioma clones, which later proved able to initiate tertiary tumors in immunocompetent hosts.

By combining barcode sequencing and single-cell RNA sequencing of early-stage gliomas with bulk RNA sequencing of secondary and tertiary tumors, we examined both the clonal and transcriptomic makeup of these lesions.
Among the numerous clones present in the primary tumors, only a fraction persisted through subsequent passages. Moreover, the clonal configuration of secondary tumors originating from the same primary glioma demonstrated partial overlap, hinting at a degree of predetermination in the acquisition of immune-evasive features. Our intra- and inter-clonal transcriptomic analyses across different tumor stages illuminate how novel functional traits may arise in gliomas.
Ongoing exploration of these data will reveal whether such traits stem from the expansion of clones already harboring them, or if they emerge through functional shifts within the clones themselves. Overall, our findings reinforce the significance of clonal competition, underscoring the central role of immune-system interactions in shaping these competitive dynamics.
Host: Federico Luzzati

Friday 31/1/2025 h. 2.00 pm - Hybrid seminar

SEMINAR CYCLE of the PhD in Neuroscience of Turin

Benedikt Berninger , King's College London, UK and University Medical Center Mainz, Germany

Engineering neurogenesis in the postnatal cerebral cortex by lineage reprogramming

Lineage reprogramming of glia into neurons emerges as an experimental strategy to regenerate neurons lost to disease. We explore the possibility of using proneural transcription factors and mutant variants thereof to convert cortical glia (astrocytes and oligodendrocyte progenitor cells) into induced neurons with subtype specific properties. For example, we could show that forced expression of a phospho-deficient form of achaete-scute complex like 1 (Ascl1), referred to as Ascl1SA6, together with the cell death regulator Bcl2 can promote the conversion of early postnatal astrocytes into induced neurons that feature hallmarks of fast-spiking parvalbumin-expression interneurons (Marichal et al., 2024).
Here, I will discuss how much we have learned about the transcriptional programmes underlying the conversion process, how much induced neurons resemble/differ from their endogenous counterparts, how we aim at closing the gap between induced and endogenous neurons, and to which induced neurons succeed to integrate into functional cortical circuits.
Host: Federico Luzzati

Friday 7/2/2025 h. 3.00 pm - Webinar

Roberta Piovesana, Department of Neurosciences, Université de Montréal - Groupe de Recherche sur la Signalisation Neurale et la Circuiterie, Université de Montréal, Canada

Novel role of endocannabinoid in neuromuscular junction structure and function

The peripheral nervous system has a remarkable ability to regenerate. Following nerve injury, different events happen at the axonal and neuromuscular junction (NMJ) level with the goal to generate an innervation-permissive environment. Several injury signals emerging from the injured axons and axon terminals have been identified to trigger the injury responses of Perisynaptic Schwann cells (PSCs), glial cells at the NMJ, essential for NMJ maintenance and repair. Although these NMJ-forming processes can be repeated, severe NMJ diseases can occur if a single step of NMJ formation is compromised. Thus, a better understanding of NMJ formation and maturation will help to understand both the regenerative processes that occur after nerve injury and how altered maturation leads to major defects.

Cannabinoids are frequently used in the treatment of neuropathic pain. However, despite evidence for their roles in the regulation of axonal guidance and synapse formation, their possible contribution in response to peripheral nerve injury remains unclear. In this webinar, we will explore the latest discovery on the glial Cannabinoid type 1 receptors at the NMJsfollowing nerve injury. Our results highlight a novel role of endocannabinoids in motor recovery, opening a possible therapeutic strategy for facilitating nerve repair or to address inadequate NMJ maintenance observed in motor neuron-related neurodegenerative diseases.
Host: Roberta Schellino

Friday 21/2/2025 h. 2.00 pm - Hybrid seminar

Katja Reinhard, Flexibility in Circuits & Behaviour Lab, SISSA - Trieste, Italy

The neural basis of context-dependent behaviour adaptation

Avoiding danger is one of the most essential and conserved sets of behaviours, observed in most species from crabs to primates. To optimize an animal's survival, avoidance responses need to be fast and reliable, but also flexible and adaptable to the current context. However, how this flexibility in behavioural output is implemented in the brain is largely unknown. Furthermore, it is unclear how similar the precise circuits and mechanisms underlying a conserved behaviour are across contexts and species.
The goal of my lab is to identify how information about the environment and state can adapt behavioural decision making. We approach this by using a highly standardized behaviour assay where we compare innate reactions and neural circuit activity while changing selected contextual elements. By probing the same behaviours and circuits under different circumstances, we aim to identify principles of behavioural flexibility that are conserved across contexts and species as well as to reveal elements that are most likely to change. During this seminar, I will focus on the brain circuit architecture that allows for contextual information to shape innate behaviours and talk about how habitat differences have led to permanent changes in innate behaviours and specific underlying circuit nodes.
Host: Stefano Zucca

Friday 28/3/2025 h. 2.00 pm - Webinar

Maria Ludovica Sforza, Laboratory of Neural Epigenomics, Institute for Medical Physics and Micro-tissue Engineering, Friedrich-Alexander-Universität Erlangen-Nürnberg (FAU), Erlangen, Germany; Laboratory of Nuclear Architecture in Neural Plasticity and Aging, German Center for Neurodegenerative Diseases (DZNE), Dresden, Germany

Investigating the role of Nup153 in neuronal responsiveness and its link to cognition

Neurons respond dynamically to environmental stimuli through the expression of the so-called immediate early genes (IEGs). Accurate spatio-temporal activation and repression of IEGs is vital for encoding specificity of incoming information and critical for proper neuronal plasticity, impacting on learning and memory. Nuclear pore complex proteins (Nups) play critical roles in cell type-specific gene regulation by orchestrating lineage specification and maintenance of cellular state. However, their roles in the adult brain remain largely elusive, especially in post-mitotic neurons. We hypothesize that Nups play key roles in neuronal plasticity and cognition through gene regulation. To address the importance of Nups for cognition and memory, we conducted behaviour, histological and electrophysiological assays on Nup153 heterozygous mice, one of key Nups in gene regulation.
Host: Francesca Montarolo

Tuesday 1/4/2025 h. 4.30 pm - Hybrid seminar

Javier DeFelipe, Laboratorio Cajal de Circuitos Corticales (CTB), Universidad Politécnica de Madrid and Instituto Cajal (CSIC), Madrid

Brain connectomics: exploring the connectome and synaptome

The principal goal in neuroanatomy is to define the detailed structural design of the nervous system. This challenge is one of the first steps towards understanding how neural circuits contribute to the functional organization of the nervous system, both in health and disease. The main difficulties involve unraveling the extraordinary complexity of the nervous system and to define how information flows through this finely organized synaptic network. Over the years, neuroanatomy has evolved considerably thanks to the use of classical techniques and the introduction of new procedures. The term "connectome" has recently been proposed to refer to the highly organized connection matrix of the human brain, in analogy to the human genome. However, defining how information flows through such a complex system represents so difficult a task that it would seem unlikely it could be achieved in the near future, or, for the most pessimistic, perhaps never. Circuit diagrams of the nervous system can be considered at different levels, although they are surely impossible to complete at the synaptic level. Even for a small mammal like the mouse it is impossible to fully reconstruct the brain at this level (we would need over 1.4×10^{9} sections to fully reconstruct just one mm^{3} of tissue). Therefore, complete reconstructions of a small region of the mammalian brain are feasible, while structures like the cerebral cortex cannot be fully reconstructed. Despite the technical difficulties, by adopting appropriate strategies with the tools now available coupled with the development of huge international projects, it should be possible to make spectacular advances in unraveling brain organization, even in humans. Indeed, advances in our capacity to marry macro- and microscopic data may help establish a realistic statistical model that could describe connectivity at the ultrastructural level, the "synaptome", giving us cause for optimism.

Host: Alessandro Vercelli

Tuesday 6/5/2025 h. 11.00 am - Hybrid seminar

Department of Neuroscience Rita Levi Montalcini, Unito
Main Hall (Aula Magna Neurologia), 1 piano, via Cherasco 15, Torino
Serenella Tolomeo , Institute of High Performance Computing, A*STAR

Neuroimaging in Psychiatry

Dr Tolomeo will present her research in the field of neuroimaging, cognitive neuroscience and psychiatry. The implications for prevention, intervention and future directions.

Host: Alessandro Vercelli

Friday 9/5/2025 h. 2.00 pm - Webinar

António J. Salgado , Life and Health Sciences Research Institute (ICVS), School of Medicine, University of Minho, Braga, Portugal; ICVS/3Bs PT Associated Lab, Braga/Guimarães, Portugal

Stem Cells Secretome in Central Nervous System Regenerative Medicine: Insights from Pre-Clinical Models of Injury and de generation

The low regeneration potential of the central nervous system (CNS) represents a challenge for the development of new therapeutic strategies. Mesenchymal stem cells (MSCs) have been proposed as a possible therapeutic tool for CNS disorders, namely due to the beneficial actions of their secretome. Indeed, the latter possesses a broad range of neuroregulatory factors that promote an increase in neurogenesis, inhibition of apoptosis/glial scar, immunomodulation, angiogenesis, neuronal and glial cell survival, as well as relevant neuroprotective actions into different pathophysiological contexts. Considering their protective action in lesioned sites, MSCs, and their

secretome, might also improve the integration of local progenitor cells in neuroregeneration processes. In this sense their use could represent an important vehicle for the establishment of future CNS regenerative therapies. Previously we have shown that the administration of MSCs secretome in pre-clinical models of spinal cord injury (SCI) and Parkinson's Disease (PD) led to important motor and histological improvements. In order to further improve this, in the present talk we will discuss and present the outcomes of the combinatory use of MSCs secretome and biodegrable biomaterials, particularly self-assembling peptides, lypossomes and hydrogels in in vitro and in vivo models of SCI and PD.
Host: Stefania Raimondo

Friday 13/6/2025 h. 2.00 pm - Webinar

Paul Weiringa, Maastricht University, MERLN - Institute for Technology-Inspired Regenerative Medicine

The Peripheral Nervous System in Tissue Engineering: from Scaffold Design to Functional In Vitro Models

The peripheral nervous system (PNS) plays a critical role in tissue function, homeostasis and repair, making it a compelling yet underappreciated target for tissue engineering and regenerative medicine. This lecture outlines the fundamentals of peripheral nerve biology and its broader relevance beyond nerve repair, including its influence on the regeneration and pathology of other tissues. Emphasis is placed on biofabrication strategies for engineering PNS-relevant environments, including electrospinning and melt electrowriting (MEW) technologies for producing fibrous scaffolds that mimic the extracellular matrix. Challenges and advancements in generating controlled architectures, such as microchannel networks that replicate native tissue organization, will also be discussed. Highlighting work from our lab, the lecture will showcase the development of complex 3D in vitro models aimed at recreating functional tissue-nerve interfaces in different contexts. These integrated approaches aim to advance understanding of tissue innervation in health and disease while advancing the boundaries of biofabrication.
Host: Stefania Raimondo

Wednesday 18/6/2025 h. 11.00 am - Hybrid seminar

SEMINAR CYCLE of the PhD in Neuroscience of Turin

Cornelius Gross , Epigenetics & Neurobiology Unit, European Molecular Biology Laboratory, EMBL Rome

How does context and experience shape social fear circuits?

Exposure to predators or predator-like stimuli elicit powerful negative emotions and uncontrollable escape responses across animal species. Over the last decade we have dissected the brain circuits that mediate such innate threat responses in mice in order to learn more about human fear. We have identified the brain pathways that mediate innate responses to predators and shown that these are independent from those that mediate responses to social threats. We have recorded the responses of individual neurons while animals initiate escape from threat and have identified a local microcircuit that supports the escape decision.
More recently we have turned to studying the neural circuits supporting avoidance of social threats.
We have found that social hierarchy and social context can modulate the threshold for escape from social threats, showing that innate emotional behavior responses can be reshaped by experience. We are currently exploring how territoriality impacts social fear and aggression, and how our neurocircuit findings in the mouse may be relevant for human social behavior. My talk will be preceded by a short overview of EMBL.
Host: Serena Bovetti

Friday 20/6/2025 h. 2.00 pm - Hybrid seminar

Daniela Ferrari , University of Milano-Bicocca - Department of Biotechnology and Biosciences

Investigational cell-based treatments using neural stem cells for Amyotrophic Lateral Sclerosis

Cell-based strategies continue to be among the most promising avenues for developing effective experimental treatments for disorders of the central nervous system. Due to their remarkable functional adaptability, stem cells offer a wide range of therapeutic possibilities, including cell replacement, immune system modulation, anti-inflammatory actions, trophic support and toxicitybluntingeffects.

In this context, I will provide an overview of our work and highlight key findings from both preclinical and clinical studies that have explored the therapeutic potential of neural stem cells, with a particular focus on their application in treating Amyotrophic Lateral Sclerosis (ALS).

Host: Marina Boido

Friday 4/7/2025 h. 2.00 pm - Hybrid seminar

PROGRESS REPORT Stefano Zucca (Adult Neurogenesis Group)

Multisensory Integration of Social Cues in the ventromedial Prefrontal Cortex

The ability to attract and reproduce with a suitable mate is a key driver of evolution. Mate selection depends on social interaction, and animals have evolved diverse behavioural strategies that rely on multimodal communication. Despite the importance of multisensory cues in courtship, the neural circuits responsible for integrating these signals remain poorly understood. In rodents, males use a combination of olfactory (e.g., pheromones) and acoustic (ultrasonic vocalizations, USVs) cues to court females. Recent studies show that female mice prefer USVs from unfamiliar males only when paired with sexual odors, suggesting cross-modal integration. However, where and how this sensory information converges in the brain remains unclear. We first confirmed through behavioural preference tests that female mice favor multimodal cues, but only within a social context. To explore the brain's response to unimodal and multimodal stimuli, we conducted whole-brain analysis of immediate-early gene (IEG) expression, using tissue clearing and light-sheet microscopy. Our results revealed that sexual odors predominantly drive brain activation, with the ventromedial prefrontal cortex (vmPFC) specifically responding to multisensory social cues. To further investigate vmPFC involvement, we used miniscope recording to perform one-photon functional imaging in awake animals. We found that vmPFC neurons respond to both unimodal and multimodal cues, with a distinct subset selectively activated by combined sensory inputs. By performing chemogenetic manipulation of vmPFC neurons, we are now investigating their causal role in shaping female preference for multisensory social stimuli. Together, these findings highlight the vmPFC as a key hub for integrating social cues from different modalities, suggesting its potential role in shaping mate preference.

Friday 11/7/2025 h. 2.00 pm - Hybrid seminar

Dr. Michael Tsoory, Weizmann Institute of Science, Israel

Detecting Sciatic nerve injury (SNI) induced motor impairments and recovery using the a home-cage monitoring system; a core facility unit user's perspective

The talk focuses on experiments conducted using the DVC® system in our facility. It describes the validation processes and the development of a "data pipeline". It will detail experiments aimed to assess the DVC® system's capacity to detect changes in home cage activity that stem from induced motor impairments and recovering from them.

Laboratory rodent models of nerve injury rely heavily on repeated assessments of motor functions away from the home–cage, where the laboratory animals are often forced to walk to allow

assessments of gait and stride, for example. Such assessments lead to substantial disturbance of the animals' routine and cause them some discomfort that might mask the experimental manipulation effects. In addition, these evaluations are labor-intensive and require time-consuming post-hoc analyses.
Therefore, the described study sought a home-cage-based alternative and in a series of experiments assessed the DVC® system's capacity to detect sciatic nerve injury-induced motor impairments and recovery dynamics as reflected by changes in voluntary, spontaneous, activity in the home cage.
The home – cage activity DVC® indices data indicate dynamics of recovery processes, reflected by "return to baseline levels", that correspond to those indicated by stride and gait analyses. Additional discussion will address the advantages and challenges of monitoring home cage activity from a core facility user's perspective. The lecture discusses in a critical manner issues of validity, reliability, scientific rigor and animal well fare.
Host: Letizia Marvaldi

Friday 18/7/2025 h. 2.00 pm - Webinar

Salman Zubedat , Spatial Perception and Memory Lab, Department of Neuroscience,
The Rappaport Faculty of Medicine, Technion-Israel Institute of Technology, Israel.

Large increase in dentate gyrus activity accompanies aggression in a resident-intruder task

Territoriality entails behaviors in which an individual claims and defends a spatially confined area from conspecifics. Our goal was to explore its neural foundations. We hypothesized that territoriality is supported by neural circuits linking memory and spatial representation, with the hippocampus playing a central role. To investigate this, we conducted calcium imaging of the dentate gyrus in the dorsal hippocampus during a resident-intruder paradigm. In this setup, a male ICR mouse expressing calcium indicators in the dentate gyrus was housed with a sterile female for 21 days to establish territoriality. Starting on day 21, the resident mouse was exposed to an intruder for 10-minute sessions over several days.
Initially, no notable changes were observed in behavior or neural activity. However, after 3-5 days, the number of active cells in the dentate gyrus increased by 3 to over 10 times the original level, coinciding with a shift from friendly to aggressive behavior. Interestingly, the aggressive behavior persisted even after the activity of the dentate gyrus subsided.These findings suggest that significant plastic changes occur in the dentate gyrus following the introduction of an intruder, highlighting its capacity to dramatically alter its activity in response to pivotal experiences.
Host: Serena Bovetti

Friday 24/10/2025 h. 2.00 pm - Hybrid seminar

PROGRESS REPORT

Valentina Cerrato (Research Group Physiopathology of Neural Stem Cells)

A single-cell transcriptomic atlas maps cerebellar astrocyte diversity and uncovers the transcriptional code underlying their maturation trajectories

Astrocytes are increasingly recognized as key regulators of neural circuit development and function. However, these cells are not all the same: they display remarkable diversity across and within brain regions. Yet, the full extent of this diversity and its developmental mechanisms remain poorly understood.

Using the cerebellum as a model, we combined multi-omic approaches, including single-cell RNA sequencing and spatial transcriptomics, and revealed an unprecedented degree of transcriptional diversity within cerebellar astrocytes, identifying known astrocyte types and uncovering new subtypes with functional specialization. Among them, a small subpopulation showed enriched expression of genes involved in glutamatergic vesicular release, highlighting that astrocytic heterogeneity extends beyond molecular identity and translates into functional specialization with potential direct effects on neuronal signaling.
By integrating transcriptomic data across developmental stages with lineage tracing and gene regulatory network analyses, we reconstructed the maturation trajectories of cerebellar astrocytes and the underlying molecular mechanisms. Our results traced their origins to multiple embryonic niches and perinatal progenitor sources and revealed unexpected lineage relationships between astrocytes and neurons, suggesting that some astrocyte subtypes retain lineage-imprinted traits that will eventually influence their functions and interactions with the surrounding neuronal circuits.
Our work provides a high-resolution map of cerebellar astrocyte diversity and its ontogeny, establishing a framework to explore astrocyte heterogeneity in other brain regions and in disease contexts. Our team has developed consolidated expertise in single-cell and spatial transcriptomic analyses applicable to a wide range of neurobiological questions aimed at understanding brain physiology and pathophysiology.

Friday 14/11/2025 h. 2.00 pm - Hybrid seminar
Sala Seminari, Polo Didattico, Ospedale San Luigi
Aldo Genovesio, Università del Piemonte Orientale
Istituto di Biochimica e Biologia Cellulare, CNR, Monterotondo
What can we learn by shifting the focus from individual neurons to neuronal assemblies as the fundamental units of information processing?
Changing the perspective and considering the assembly of neurons as the unit of information processing, rather than the sum of individual neurons, can reveal how a single neuron can play multiple roles in information coding, much like a chess simul master playing on multiple chessboards. A neuron may have no role in information coding when considered alone, but it can contribute to the representation of multiple types of information when its activity is coordinated with that of others.
This approach can also enhance our understanding of the formats of information representation in memory, highlighting the difference between dynamic and static coding schemes. When applied to the study of connectivity between brain areas, this perspective can uncover new modalities of coordination between neurons, such as the formation or re-entry of information loops and their role as hubs of coordination between areas.
Host: **Annalisa Buffo**

Friday 21/11/2025 h. 2.00 pm - Hybrid seminar
SEMINAR CYCLE of the PhD in Neuroscience of Turin **Makoto Sato, Department of Anatomy and Neuroscience, Graduate School of Medicine, Osaka University, Japan; Division of Child Development, United Graduate - School of Child Development (UGSCD), Osaka University, Japan**
Beyond the cytoskeletons and cortical development:Looking for the cellular mechanisms for intelligence and underlying neuronal circuit regulation
Dr Makoto Sato is currently a specially appointed professor at the United Graduate School of Child Development (UGSCD) of The University of Osaka (Osaka University) and a professor emeritus at Osaka University (2013~2025) and the University of Fukui (1998~2013), Japan. He was a full professor of the Department of Anatomy and Neuroscience, Graduate School of

Medicine, and the dean of UGSCD at Osaka Univ. He obtained a BEng (Mathema cal Informa on Engineering) from the Univ. of Tokyo, an MD and a PhD from Osaka Univ. in 1991. He was a postdoc at the Salk Ins tute, USA, before he joined Osaka City University (Osaka Metropolitan University) as an associate professor. He was made a full professor at the University of Fukui in 1998. His research interest is developmental neuroscience. He has taught neuroanatomy, human anatomy, histology, and human embryology to more than 4000 medical students. He was a board of directors of The Japanese Associa on of Anatomists and a board officer of The Japanese Society for Neurochemistry. He acted as the president of the 127 th Annual Mee ng of Anatomists in Japan (March 2022). He has won numerous research awards, including the Prizes for Science and Technology (The Commenda on for Science and Technology by the Minister of MEXT, Japan).
Host: **Alessandro Vercelli**

Friday 5/12/2025 h. 2.00 pm - Hybrid seminar
PROGRESS REPORT

Marta Ribodino (Research Group Physiopathology of Neural Stem Cells)
Toward circuit reconstruction in Huntington's Disease: functional integration of human striatal progenitors and development of tools for selective targeting of MSNs

Friday 12/12/2025 h. 2.00 pm - Hybrid seminar
PROGRESS REPORT

Ilaria Ghia (Research Group Adult Neuroplasticity)
Addressing the role of the olfactory dopaminergic population in processing ethologically relevant odors by using a multidisciplinary approach

Friday 19/12/2025 h. 11.00 am - Hybrid seminar

Silvia Pozzi, Assistant Professor, Université Laval, Quebec, Canada and CERVO Brain Research Centre
Molecular Regulators of Extracellular Matrix remodelling in ALS
Amyotrophic Lateral Sclerosis (ALS) is a devastating neurodegenerative disease that leads to progressive muscle atrophy and paralysis due to motor neuron degeneration. Despite extensive research over the past decades, ALS remains incurable. Recent advances have redefined ALS as a multicellular disorder, highlighting the critical role of cellular communication between non-neuronal cells and the degenerating motor neurons. Understanding this altered communication could reveal novel pathogenic mechanisms and therapeutic targets.
In this presentation, Dr. Pozzi will explore a key molecular pathway under investigation in her laboratory, which lies at the crossroads of neurodegeneration and neuroinflammation in ALS. Activation of this pathway accelerates extracellular matrix degradation, contributing to motor neuron death and possibly denervation. Additionally, it triggers the release of pro-inflammatory factors, exacerbating neuroinflammation. An ongoing preclinical trial demonstrates the therapeutic potential of targeting this pathway to mitigate motor neuron death in a mouse model of ALS.