

 <p>UniSR Università Vita-Salute San Raffaele</p>	<p>APPLICATION TO ACT AS SUPERVISOR AND RESEARCH PROJECT PROPOSAL</p>	<p>MO 20-5 ed. 02 of 16/01/2026 PO 20 Page 5 of 11</p>
---	--	---

PROJECT

Supervisor: _____Dario Bonanomi_____

Title: __Characterizing and Targeting the Blood-Nerve barrier in ALS

Curriculum: _____Neurosciences and Experimental Neurology _

Link to the personal page of the University or relevant hospital site website: __ <https://research.hsr.it/en/divisions/neuroscience/molecular-neurobiology.html> _____

Description of the Project (max 3,000 characters including spaces)

Background/gap of knowledge

The nervous system is protected by specialized vascular barriers, which are well characterized in the central nervous system (CNS), such as the blood-brain and blood-spinal cord-barriers^{1,2}. In contrast, the blood-nerve barrier (BNB), which preserves the homeostasis of peripheral nerves, remains poorly understood³. The BNB represents both a critical site of vulnerability in diseases affecting the peripheral nervous system (PNS) and a barrier to the effective delivery of therapeutics⁴.

Importantly, the BNB is structurally and molecularly distinct from the CNS vasculature⁵. Our lab has begun to define these unique features using transcriptomic approaches, mouse models, and functional assays. We have identified specific endothelial properties and cellular interactions that regulate BNB stability and are altered in disease⁶.

In amyotrophic lateral sclerosis (ALS), traditionally considered a CNS disorder, emerging evidence indicates early involvement of the PNS^{7,8}. Motor neurons extend long axons through peripheral nerves, and damage to these structures occurs early in disease progression. Our data show that BNB alterations are an early event in ALS models and patient samples, suggesting a potential role in disease initiation and propagation⁶.

Rationale and hypothesis

The early involvement of the BNB in ALS suggests that this interface is both vulnerable and potentially targetable. We hypothesize that early BNB disruption permits immune infiltration and accumulation of toxic factors, leading to destabilization of peripheral nerve homeostasis and

 <p>UniSR Università Vita-Salute San Raffaele</p>	<p>APPLICATION TO ACT AS SUPERVISOR AND RESEARCH PROJECT PROPOSAL</p>	<p>MO 20-5 ed. 02 of 16/01/2026 PO 20 Page 6 of 11</p>
---	--	---

exacerbation of neurodegeneration. Restoring BNB integrity may therefore mitigate disease progression.

Objectives and specific aims

This project aims to define the molecular and cellular mechanisms regulating BNB function and its alteration in ALS. It will leverage the lab's expertise and tools to achieve:

- 1) Molecular characterization of the BNB using single-cell RNA sequencing and integration with spatial transcriptomics datasets to identify deregulated pathways in ALS models.
- 2) Functional targeting of candidate pathways by manipulating endothelial cells in peripheral nerves using AAV-based approaches and genetic models to test causality and therapeutic potential.

The project may also extend to other PNS-related disorders, including demyelinating diseases and pain syndromes.

Expected outcomes

We expect to identify molecular signatures of BNB dysfunction, including disease-specific and shared pathways across PNS disorders. This work will define key regulators of vascular stability in peripheral nerves and establish whether targeting the BNB can preserve nerve integrity and limit disease progression.

Skills that the student should acquire (max. 600 characters including spaces):s

The student will gain expertise in neurovascular biology. They will develop skills in mouse models of neurodegeneration, including viral and drug delivery surgeries, behavioral analysis, immunohistochemistry, and imaging. They will also be trained in single-cell and spatial transcriptomics, functional vascular assays, molecular targeting approaches (AAV, genetics), data integration, and critical analysis of disease mechanisms in ALS and related disorders.

References (max. 15)

1. Lacoste, B., Prat, A., Freitas-Andrade, M., and Gu, C. (2024). The Blood-Brain Barrier: Composition, Properties, and Roles in Brain Health. *Cold Spring Harb Perspect Biol.* 10.1101/cshperspect.a041422.
2. Ruiz de Almodovar, C., Dupraz, S., and Bonanomi, D. (2025). Neurovascular dynamics in the spinal cord from development to pathophysiology. *Neuron.* 10.1016/j.neuron.2025.09.017.
3. Mizisin, A.P., and Weerasuriya, A. (2011). Homeostatic regulation of the endoneurial microenvironment during development, aging and in response to trauma, disease and toxic insult. *Acta Neuropathol* 121, 291-312. 10.1007/s00401-010-0783-x.



UniSR

Università Vita-Salute
San Raffaele

**APPLICATION TO ACT AS SUPERVISOR AND
RESEARCH PROJECT PROPOSAL**

MO 20-5

ed. 02 of 16/01/2026

PO 20

Page 7 of 11

4. Reinhold, A.K., and Rittner, H.L. (2017). Barrier function in the peripheral and central nervous system—a review. *Pflugers Arch* 469, 123–134. 10.1007/s00424-016-1920-8.
5. Malong, L., Napoli, I., Casal, G., White, I.J., Stierli, S., Vaughan, A., Cattin, A.L., Burden, J.J., Hng, K.I., Bossio, A., et al. (2023). Characterization of the structure and control of the blood-nerve barrier identifies avenues for therapeutic delivery. *Dev Cell* 58, 174–191 e178. 10.1016/j.devcel.2023.01.002.
6. Bhat GP, D.S., Beccaria CG, Podini P, Badaloni A, Motta A, Brambilla I, Martins L, Boselli D, de Pretis S, Nardo G, Bendotti C, Riva N, Iannacone M, Quattrini A, Bonanomi D (2025). Early endothelial activation at the blood-nerve barrier defines a hallmark of ALS. *bioRxiv*. <https://doi.org/10.1101/2025.08.31.673224>.
7. Ravits, J.M., and La Spada, A.R. (2009). ALS motor phenotype heterogeneity, focality, and spread: deconstructing motor neuron degeneration. *Neurology* 73, 805–811. 10.1212/WNL.0b013e3181b6bbbd.
8. Riva, N., Gentile, F., Cerri, F., Gallia, F., Podini, P., Dina, G., Falzone, Y.M., Fazio, R., Lunetta, C., Calvo, A., et al. (2022). Phosphorylated TDP-43 aggregates in peripheral motor nerves of patients with amyotrophic lateral sclerosis. *Brain* 145, 276–284. 10.1093/brain/awab285.