

 <p>UniSR Università Vita-Salute San Raffaele</p>	<p>APPLICATION TO ACT AS SUPERVISOR AND RESEARCH PROJECT PROPOSAL</p>	<p>MO 20-5 ed. 01 del 21/02/2025 PO 20 Page 4 of 10</p>
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PROJECT

Supervisor: ___Dario Bonanomi_____

Title: Endothelial-Mesenchymal Interactions in Nerve Regeneration and Cancer

Curriculum: _____ Cellular and Molecular Biology

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

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

Background/gap of knowledge: Peripheral nerves regenerate after injury, unlike the nerves in the central nervous system (CNS). However, they are also vulnerable to tumorigenesis, such as in Neurofibromatosis Type 1 (NF1), a disorder affecting 1 in 3,000 births. Neurofibromas, nerve tumors associated with this genetic condition, emerge when Schwann cell (SC) precursors lose both copies of the Nf1 gene [1, 2]. Neurofibromas resemble chronic wounds, suggesting they may arise from disrupted nerve healing. Although SC precursors initiate tumorigenesis, they represent a minor fraction of the tumor mass. In contrast, fibroblast-like mesenchymal cells (MES) are abundant but remain understudied [3, 4]. Our recent findings show that deregulated angiogenesis after nerve injury leads to abnormal stromal remodeling and impaired repair, hinting that similar vascular-mesenchymal dysfunction may underlie tumor growth [5].

Rationale and hypothesis: Tumors have been considered “open wounds” [6]. Aligning with this, we propose that aberrant angiogenesis in neurofibroma drives a shift in mesenchymal cells from a reparative to a cancer-promoting phenotype, mimicking failed wound healing. This vascular-mesenchymal crosstalk may convert the regenerative environment of injured nerves into a fibrotic, tumor-permissive niche. Understanding this transition could uncover strategies to redirect tumor stroma toward repair.

Objectives and specific aims: The goal of this project is to define and modulate the vascular-mesenchymal axis in nerve tumors. The student will use a neurofibroma model based on removal of Nf1 from SCs during embryogenesis and establish whether nerve injury accelerates tumor growth [7]. Tumors will be monitored via MRI and characterized histologically, focusing on endothelial cells (ECs) and mesenchymal populations. Next, ECs and MES present in



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neurofibromas or nerve wounds will be profiled using single-cell RNA-seq. Comparing tumors, early lesions, and repaired nerves will identify signatures that distinguish tumor-promoting vs. reparative niches. Candidate regulators will be validated in mouse tissue and examined in patient neurofibroma samples with the support of clinical pathologists (collaborations in place). Finally, the tumor vasculature and stroma will be targeted with pharmacological and viral (AAV) approaches to manipulate key signaling axes identified in the molecular screen. As a proof of principle, we will test whether anti-angiogenic therapy following nerve injury redirects MES activation, as we have previously demonstrated in physiological healing [4], limiting tumor growth. Prior to in vivo studies, candidate molecular targets will be screened in iPSC-derived neurofibroma assembloids (in collaboration with Dr. Igor Adameyko, Karolinska Institute, Sweden).

Expected outcomes: This work will clarify how endothelial-stromal interactions influence neurofibroma development and establish whether the nerve microenvironment can be reprogrammed toward repair. Insights gained may open therapeutic avenues in NF1 and other fibrovascular tumors.

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

Mouse handling; design of mouse crosses; Basic surgeries in mouse models (nerve injury); Injections of cells, viral particles and drugs; tissue dissection and processing for immunohistochemistry/histology; Tissue clearing and imaging; molecular approaches including cloning, preparation of RNA probes, cDNA library preparation; FACS sorting and basic analysis; Image processing and quantitative analysis of imaging and gene expression data. Interaction with bioinformaticians will be encouraged to build skills in interpretation and analysis of computational data (programming skills are optional).

References (max. 15)

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[4] Kershner, L. J., Choi, K., Wu, J., Zhang, X., Perrino, M., Salomonis, N., Shern, J. F., & Ratner, N. (2022). Multiple Nf1 Schwann cell populations reprogram the plexiform neurofibroma tumor microenvironment. *JCI Insight*, 7(18), e154513. <https://doi.org/10.1172/jci.insight.154513>

[5] Bhat, G. P., Maurizio, A., Motta, A., Podini, P., Diprima, S., Malpighi, C., Brambilla, I., Martins, L., Badaloni, A., Boselli, D., Bianchi, F., Pellegatta, M., Genua, M., Ostuni, R., Del Carro, U., Taveggia, C., de Pretis, S., Quattrini, A., & Bonanomi, D. (2024). Structured wound angiogenesis instructs mesenchymal barrier compartments in the regenerating nerve. *Neuron*, 112(2), 209–229.e11. DOI: [10.1016/j.neuron.2023.10.025](https://doi.org/10.1016/j.neuron.2023.10.025)

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[7] Ribeiro, S., Napoli, I., White, I. J., Parrinello, S., Flanagan, A. M., Suter, U., Parada, L. F., & Lloyd, A. C. (2013). Injury Signals Cooperate with Nf1 Loss to Relieve the Tumor-Suppressive Environment of Adult Peripheral Nerve. *Cell Reports*, 5(1), 126–136. <https://doi.org/10.1016/j.celrep.2013.08.033>