PhD project 2021-2024

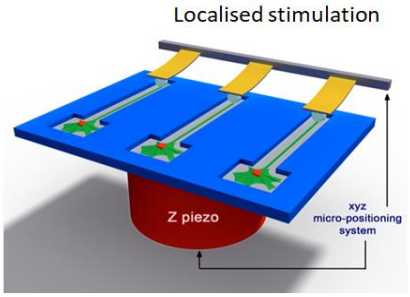
Nano-Mechanical Systems for the Understanding of cellular Mechano-Response at the Nanoscale

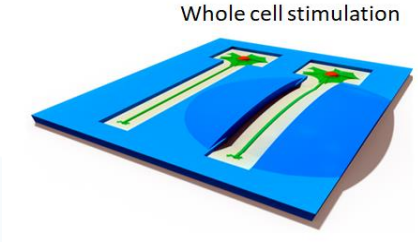
Cells are able to respond to various extracellular mechanical signals and converted them into a biological activity on the cellular scale that ranges from embryo development, cell differentiation, vasodilatation, mechanosensation [1].

Mechanoreceptor cells of the somatosensory system provides mammals with a variety of sensory response: touch, pain, itch, and proprioception. At cellular level the mechanotrasduction response has been observed to be mediated by several cellular elements: plasma-membrane [2], adhesion proteins [3], cytoskeleton [4] and mechanosensitive ion channels [5]. Recently, the mechanosensitive Piezo channels have been identified as key elements in the transduction of mechanical stimuli.

In the somatosensory system Piezo channels are located at the periphery of the Dorsal Root Ganglion (DRG) neurons, while DRG in culture lose this polarization and Piezo channels are observed evenly distributed on the cell surface: what determines their distribution during the embryo development? Can physical stimuli influence their localization in DRG? The study of the sensory elements in vivo cannot answer these questions because of their sparse distribution in the skin and of their inaccessibility to experimental mechanical stimulation. This, in turn, makes it difficult to correlate the gating mechanism of these channels with their role in physiological or pathological conditions.

In order to elucidate the origins of the mechanosensory neurons polarization and to clarify the molecular force transduction mechanism of the Piezo channels the PhD candidate will microfabricate an innovative family of active microfluidic substrates, capable of applying well defined mechanical stimulations in terms of intensity, orientation and frequency on primary DRG neurons in a controlled environment and nearly physiological condition. These devices will be used to investigate the development and polarization of mechanosensory neurons as function of precise stimuli and will be used to disentangle the cues (i.e. molecular, morphological and physical) that drive or maintain the organization and polarization of Piezo channels in mechanosensory neurons.





The functional organization of the mechanoreceptors will be investigated with live Ca++ imaging; the spatial organization will be observed with epifluorescence microscopy and super resolution microscopy with the support of the Max Planck Institute for Medical Research in Heidelberg (D). Localized mechanical stimulation using AFM in Trieste and magnetic tweezers at the University of Zurich (CH) will be used to stimulate the external cell membrane and the internal cytoplasmatic structure, respectively, in order to clarify the force intensity and the gating mechanism of Piezos at a single molecule level.

The research activity will be carried out in the 3M lab of IOM-CNR in tight collaboration with the Neurobiology group of Prof. Paul Heppenstall at SISSA (Trieste).
(https://www.iom.cnr.it/research-facilities/facilities-labs/analytical-microscopy-and-spectroscopy/3m/)

The candidate will get in touch with a rich variety of methodologies and scientific environments (e.g, neurobiology, atomic force microscopy, nanofabrication, fluorescence microscopy, super-resolution microscopy) and will work within a strong interdisciplinary environment.

Interested candidates should contact the supervisors of the project dr. Marco Lazzarino (lazzarino@iom.cnr.it) and dr.ssa Laura Andolfi (andolfi@iom.cnr.it) for further information.

References

[1] F. Martino, et al. Cellular Mechanotransduction: From Tension to Function. Frontiers in Physiology (2018) 9: 824.
[2] Y. Qi, L. Andolfi, F. Frattini, F. Mayer, M. Lazzarino, J. Hu. Membrane stiffening by STOML3 facilitates mechanosensation in sensory neurons Nat. Commun.(2015) 6:8512.
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[4] S. J Morley, Y. Qi, L. Iovino, L. Andolfi, D. Guo, N. Kalebic, L. Castaldi, C. Tischer, C. Portulano, G. Bolasco, K. Shirlekar, C. M Fusco, A. Asaro,F. Fermani, M. Sundukova, U. Matti, L. Reymond, A. De Ninno, L. Businaro, K. Johnsson, M. Lazzarino, J. Ries, Y. Schwab, J. Hu, P.A. Heppenstall. *Acetylated tubulin is essential for touch sensation in mice*. E-Life (2016) 5: e20813.
[5] JM Kefauver et al. Discoveries in structure and physiology of mechanically activated ion channels Nature (2020) 587 567-576