REPORT – 3rd Ph.D. Year

Virginia Bazzurro (XXXIII Cycle)

Supervisors: Prof. Alberto Diaspro, Prof. Mauro Robello

RESEARCH ACTIVITY

My research activity concerns the study of the action of pharmacological molecules, especially the Antisecretory Factor, on $GABA_A$ receptors, activated by the endogenous inhibitory neurotransmitter γ-aminobutyric acid (GABA), in rat cerebellar neurons (cerebellar granule cells) in culture.

The project aims to study the activation and the modulation of these receptors in a specific cell region for monitoring individual neuronal responses with a high spatial resolution thanks to the uncaging of a "caged GABA", RuBi-GABA, a photoactivated molecule whose activity can be controlled by a pulse of light.

I have experimentally investigated the caged molecule RuBi-GABA thanks to the electrophysiological technique of the patch-clamp in whole-cell configuration coupled with the confocal and two-photon microscopy for regulating the release of the neurotransmitter in a well-defined instant of time and space.

In particular, I deeply characterized the uncaging method. I explored how the variations of several critical physical parameters (i.e., laser power, exposure time, distance from the uncaging point from the cell of interest along the X, Y, Z spatial coordinates) influence the electrophysiological measurements and how they affect the modality and efficacy of GABA release and, consequently, the $GABA_A$ response, using photoactivated-GABA.

Furthermore, I studied the action of a pharmacological molecule, the Antisecretory Factor, on the $GABA_A$ receptor using the non-linear photoactivation of RuBi-GABA.

The Antisecretory Factor is an endogenous protein that acts *in vivo* by inhibiting intestinal hypersecretion and various forms of inflammation and is expressed in many mammalian tissues and plasma, but its biological role is mostly unknown.

The purpose was to examine the effect on a defined $GABA_A$ population, uncaging RuBi-GABA on specific areas of the neuron (i.e., soma, cone, and neurites) for understanding the mechanism of action on different receptor subtypes.

This technique allows useful studies to localize and map the receptor distribution on the plasma membrane of the neurons.

The future goal is to study the modulation of $GABA_A$ receptors by other selective pharmacological treatments using the uncaging of RuBi-GABA and the labeling with fluorescence probes suitable to trace ions for identifying the intracellular mechanism of action of such drugs.

PUBLICATIONS

Bazzurro V., Gatta E., Cupello A., Lange S., Robello M. (2018). Antisecretory Factor Modulates $GABA_A$ Receptor Activity in Neurons. J Mol Neurosci; 64(2): 312 – 320.
doi: https://doi.org/10.1007/s12031-017-1024-8

Cozzolino M., **Bazzurro V.**, Gatta E., Bianchini P., Angeli E., Robello M., Diaspro A. (2020). Precise 3D modulation of electro-optical parameters during neurotransmitter uncaging experiments with neurons in vitro. Sci Rep.; 10(1):13380.
doi: https://doi.org/10.1038/s41598-020-70217-5

CONFERENCES

- February 15th – 19th 2020, 64th Annual Meeting of the Biophysical Society, San Diego.
  Bazzurro V., Gatta E., Angeli E., Robello M., Diaspro A. (2020). *Study of the modulation of $GABA_A$ receptor by the Antisecretory Factor using RuBi-GABA uncaging with non-linear photoactivation in rat cerebellar granule cells in vitro.*

- September 14th – 18th 2020, 106° Congresso Nazionale Società Italiana di Fisica.
  Invited speaker
  Bazzurro V., Gatta E., Angeli E., Cupello A., Lange S., Robello M., Diaspro A. (2020). *Study of the Antisecretory Factor effects on $GABA_A$ receptor by using RuBi-GABA uncaging with non-linear photoactivation in rat cerebellar granule cells in vitro*.