

JOINT EVENT

5<sup>th</sup> World Conference on **Climate Change**

&amp;

16<sup>th</sup> Annual Meeting on**Environmental Toxicology and Biological Systems**October 04-06, 2018  
London, UK

## Manuela Marcoli

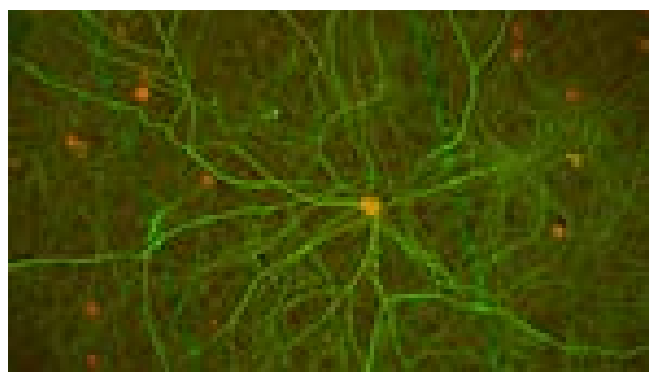
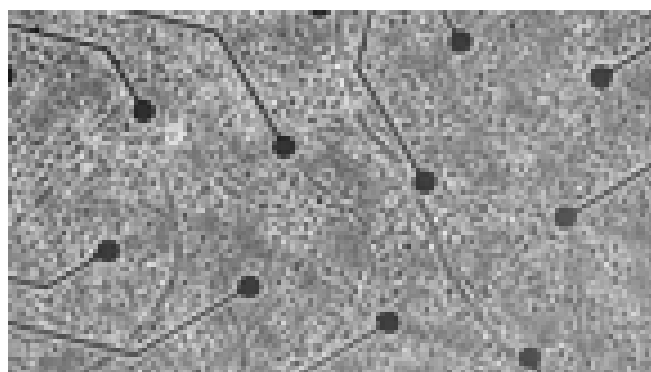
University of Genova, Italy

### Functional neuron-specific endpoints for *in vitro* neurotoxicity testing

**Statement of the Problem:** In accordance with 3Rs, alternative models are required to replace standard neurotoxicity testing. High-content, high-throughput tools are needed considering specific features of nervous system (NS) functioning to identify neurotoxic vs. cytotoxic effects. By considering intercellular communication through transmitters and transmitter sensors (receptors), and collective behavior of neuron network as relevant NS functional features, the purpose of this study is to develop tools providing neuron-specific endpoints.

**Methodology & Theoretical Orientation:** A multi-disciplinary electrophysiological, neurochemical and immunocytochemical approach, combining electrical activity recording of neuron network (on engineered micro-electrode arrays (MEAs) equipped with 60 electrodes onto which cerebrocortical neurons were cultured; data analysis through a home-made software and measurement of transmitter release was used to assess network maturation and to detect effectiveness of neuroactive/neurotoxic substances.

**Findings:** During network development, maturation of glutamatergic/GABAergic neuron networks, target for relevant neurotoxicity mechanisms (excitotoxicity) and drugs classes, was observed. In mature networks, synaptic connectivity was related to activation of glutamatergic pathways, and the system behaved as a sensitive sensor of glutamatergic transmission functioning. Activation or blockade of NMDA/AMPA receptors, or blockade of glutamate transporters, induced firing and bursting activity variations related to the effects on transmitter release. Also, the network sensed the fine transmission variations involved in synapse plasticity: the collective network behavior and glutamate release were controlled by NMDA-dependent NO-cGMP pathway, as indicated by its pharmacological manipulation (NO synthase/guanylyl cyclase inhibitors, NO donors/8Br-cGMP). By presenting examples of network activity modulation by neuroactive substances (glutamate/GABA receptor agonists/antagonists) and by known neurotoxins (e.g., domoic acid, chlorpyrifos oxon), and ineffectiveness of molecules not exhibiting acute neurotoxic effects, we report evidence that MEAs-coupled neuron networks can represent an integrated approach for neurotoxicity testing based on functional neuron-specific endpoints. They might provide an effective *in vitro* alternative tool for evaluating substance neurotoxicity, also providing a mechanistic approach.



**Figure 1:** Neuron network on MEAs; primary rat cerebrocortical neuron cultures from E19, 24 DIV. The network on microelectrode arrays is shown. Immunocytochemistry for MAP2 (green) and NeuN (red).

## **Environmental Toxicology and Biological Systems**

### **Recent Publications**

1. Frega M, Pasquale V, Tedesco M, Marcoli M, Contestabile A, et al. (2012) Cortical cultures coupled to micro-electrode arrays: a novel approach to perform in vitro excitotoxicity testing. *Neurotoxicol Teratol* 34:116–127.
2. Marcoli M, Agnati L F, Benedetti F, Genedani S, Guidolin D, et al. (2015) On the role of the extracellular space on the holistic behaviour of the brain. *Rev Neurosci* 26(5):489–506.
3. Fuxe J, Agnati L F, Marcoli M and Borroto-Escuela D (2015) Volume transmission in central dopamine and noradrenaline neurons and its astroglial target. *Neurochem Res* 40(12):2600–14.
4. Cervetto C, Vergani L, Passalacqua M, Ragazzoni M, Venturini A, et al. (2016) Astrocyte-dependent vulnerability to excitotoxicity in spermine oxidase overexpressing mouse. *Neuromolecular Med* 18:50–68.
5. Pietropaoli S, Leonetti A, Cervetto C, Venturini A, Mastrantonio R, et al. (2018) Glutamate excitotoxicity linked to spermine oxidase overexpression. *Mol Neurobiol*. 55(9):7259–7270.

### **Biography**

Manuela Marcoli has more than 20 years' experience in studying neurotransmission in the central and peripheral nervous system. She has completed her MD and PhD in Clinical Pharmacology from Pavia University, Italy. She is a Professor of Pharmacology at the University of Genova, Italy. Main research fields are glutamatergic transmission: ionotropic/metabotropic receptors and release of neuro- and gliotransmitters in physiological conditions and in pathological animal models; network activity and transmitter receptors in neuron-astrocyte networks on multi electrode arrays: effects of neuroactive substances and neurotoxicants. She has over 85 publications in peer-reviewed journals and has been serving as a Reviewer of reputed journals.

[marcoli@pharmatox.unige.it](mailto:marcoli@pharmatox.unige.it)

### **Notes:**