

SLG Functionalized MEA for Enhanced Detection of Neural Network Development

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Planar Micro-Electrode Arrays (MEAs) are a valuable tool for the recording of spontaneous activity of primary neuronal network, thus providing the possibility to monitor the electrophysiological activity of neurons from single spikes to network bursts [1]. Recently, in order to improve performances of MEAs in terms of signal to noise ratio and enhancement of cell-electrode coupling, different strategies have been proposed including employment of carbon based materials, that have gained popularity [2], among which graphene is. Since it has been demonstrated that the viability of neuronal cells and average neurite length were significantly enhanced on graphene substrate [3], in the current study we have employed single layer graphene (SLG) to functionalize the surface of MEAs.

First, we successfully transferred SLG via wet etching onto commercial planar 60 electrode devices; then we recorded the electrophysiological development of neuronal cultures at the whole network level up to completion of network maturation, and we compared it to conventional (i.e., not functionalized) MEAs. We recorded the spontaneous activity for 90 minutes at seven different developmental phases; i.e. days in vitro

(DIV), starting from DIV 7 up to DIV 25. Confocal microscopy images of immunolabeled samples demonstrated morphology of healthy cells on both SLG and control substrates. Neural specific MAP2 immunostaining demonstrated the higher number of neurons on SLG with respect to the control. The most striking results were the observation of spikes and bursts activity at an earlier developmental phase and of strongly synchronized neuronal networks on SLG-MEA versus control. These observations are in well agreement with our previous study of single neuron synaptogenesis by patch clamp, where earlier synaptogenesis on SLG compared to the control was detected. The results have been corroborated with the mean firing rate and mean bursting rate results showing higher and statistically significant values on SLG-MEA.

References

- [1] P. Charlesworth et al., Neural development, 10 (2015) 1
- [2] S. Ostrovsky et al, Rsc Advances, 6 (2016) 41714-41723
- [3] D. Convertino et al, Front. Neurosci., 12 (2018)
- [4] S. Keshavan et al, Acta biomaterialia, 65 (2018) 363-375

Figures

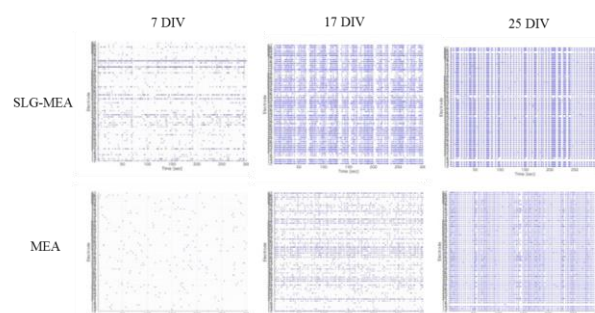


Figure 1: 5-s raster plots of spontaneous activity recorded at three different developmental phases: 7 DIV, 17 DIV and 25 DIV