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STUDY OF THE NEURONAL CELLS ON THE MICROELECTRODE ARRAY MEA60PT BY THE ATOMIC FORCE MICROSCOPY (AFM)

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The research on the nervous system is potentially important in order to understand the origin of neurological diseases. The use of the atomic force microscopy (AFM) in such complex biological system consisted from network of neuronal cells, offers a powerful tool to study cell morphology and mechanisms of the intercellular communication.

Here, we present work on development of methodology for visualization and characterization of the neural network cell system, consisted from granular neurons of cerebellum (primary culture), based on the use of the AFM (Nanoscope III, Multimode). The primary cultures of neuronal cells were obtained from cerebellum of the Wistar rats. Furthermore, cells were placed to grow on a microelectrode array MEA60Pt, previously coated with poly-L-lysine/DMEM. This particular methodology turns to be very efficient for the cell growth and the AFM imaging [1-2].

The AFM visualization was carried *in-situ* (physiological medium) and after the cell immobilization treatment, by washing the microelectrode by the ethanol-water solution (70, 80 and 100 % ethanol). A successful visualization, so far, was achieved after immobilization (dehydration) process. The high-resolution AFM images clearly show distribution and position (accommodation) of the neural cells on the MEA60Pt array (figure 1b), with nanometric details of the cell morphology, especially in the region of the cell cytoplasmic prolongation, previously not reported in the literature. The rate of the cell dehydration did not influence the imaging process and the cell morphology characteristics (figure 1a-1d).

Among different properties, we also study ability of the neural cells to conduct electrical signal between electrodes of the MEA60Pt, under external potential field. Our results shows that AFM could successfully register and identified part of the MEA60Pt, involved in the electronic circuit, however participation of the neural cells could not be clarified, so far. The further measurements are in the progress [2-3].

The methodology and results developed in our study, show that AFM could offer new inside into mechanism of the biologically complex systems and could be of a tool of the great potential for further development of nanomedicine and the molecular biology.

References:

[1] Freshney Ian R. (1994). Culture of Animal Cells: a Manual of Basic Technique.4th Wiley-Liss. Glasgow, Scotland.

[2] Acosta G. Ma.Cristina. (2005). Estudio de células del sistema nervioso por microscopía de fuerza atómica. Ms.c. Thesis with Dr. Nikola Batina, (UAM-I) and Dr. Eva Ramón Gallegos, (ENCB-IPN). México, D.F. MEXICO.

[3] Morin, F.O., Takamura, Y. and Tamiya, E., Jan 2005. Investigating neuronal activity with planar microelectrode arrays: achievements and new perspectives Journal of Bioscience and Bioengineering, Vol. 100, Issue 2.

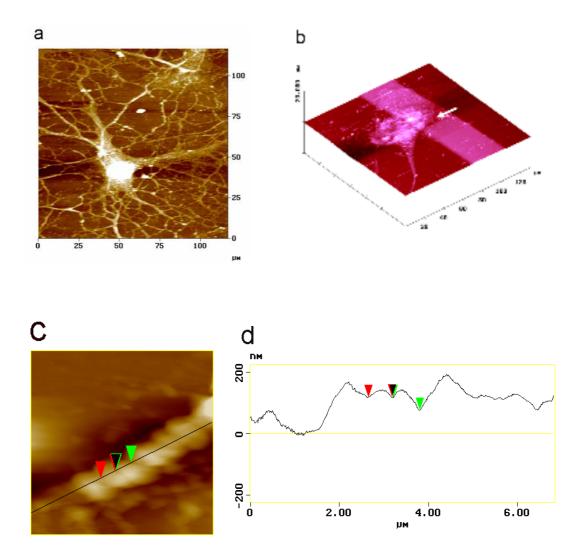


Figure 1. AFM Images of the neural cell on gold (a) and the neural cell on the MEA60Pt array (b). High resolution AFM image show the fine structural detail of the neural cell cytoplasmic prolongation (c), mainly consisted from the disc like features with diameter of 558.96 nm, in average (c and d).