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Evaluation of the penetrating ability of a perspective copper-containing drugs into cells using an electrochemical nanocapillary-based sensor

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Evaluation of the effectiveness of perspective drugs, as well as the study of their pharmacokinetics, has been one of the priority tasks at the stage of pre-clinical drug trials (Vaneev et al., 2020). In recent years, many copper-containing drugs have been developed since they have a high antimicrobial, anti-tuberculosis, anti-malarial, antifungal and anti-inflammatory activity. However, in most cases various biological activity tests are carried out by expensive, high invasive (AAS, ICP-MS etc.), or difficult to synthesize and often low-precise (fluorescent labels) analysis methods. Thus, it difficult to understand the effect of drugs on living biological systems.

Recently, we presented an analytical review detailing all the quantitative copper detection methods for the analysis of living single cells (Timoshenko et al., 2020). According to the results of a comparative analysis, the electrochemical determination of copper is one of the most promising for use in biological systems.

We used nanoscale sensors based on nanocapillaries, which were fabricated using a CO2 laser puller. Then we functionalized the nanocapillaries with pyrolytic carbon. Further modification consists of a carbon pre-etching and gold electrochemical deposition into the formed nanocavities. The fabricated electrochemical sensors are fixed on a special micromanipulator near the optical microscope, which allows to penetrate into single cells with high precision and receive an electrochemical response (Figure 1).

The developed sensor was used to study the distribution of copper in MCF7 cells under the effect of copper chloride (Figure 2A). Thus, our sensors can be used to study hereditary neurodegenerative diseases associated with dysregulation of copper (Wilson, Menkes and Alzheimer diseases).

We are currently investigating a new copper-containing cytotoxic agent based on 2-thioxoimidazolone (Krasnovskaya et al., 2020). The complexes at various concentrations were characterized by cyclic voltammetry in physiological solutions (Figure 2B).

The next step will be to study the accumulation of testing copper compounds in the nucleus and cytoplasm of living cells after treatment.

Our method is one of the applications of scanning ion conductance microscopy (SICM). The use of nanocapillaries is an unlimited opportunity for the developing of low invasive electrochemical sensors for evaluation of perspective drugs. Firstly, in conjunction with SICM, it is possible to simultaneously map the surface with high resolution and determine analytes at a local point of analyzed space. Secondly, the use of easily modifiable metals such as gold will simplify the selection ligands for all types of analytical tasks.

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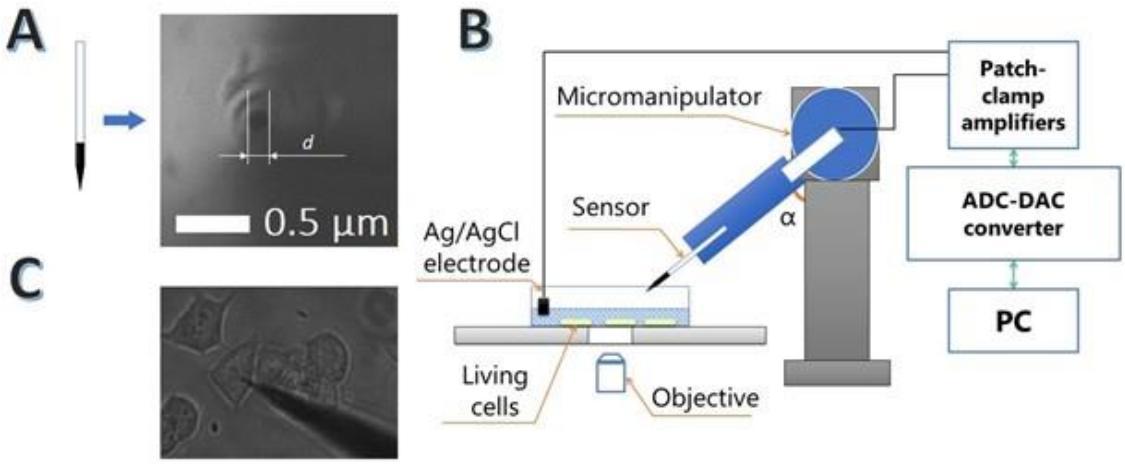

Figure 1. Electrochemical analysis of single cells. A) SEM image of nanocapillary-based sensor B) Setup scheme C) Microphotograph demonstrating a single cell measurement

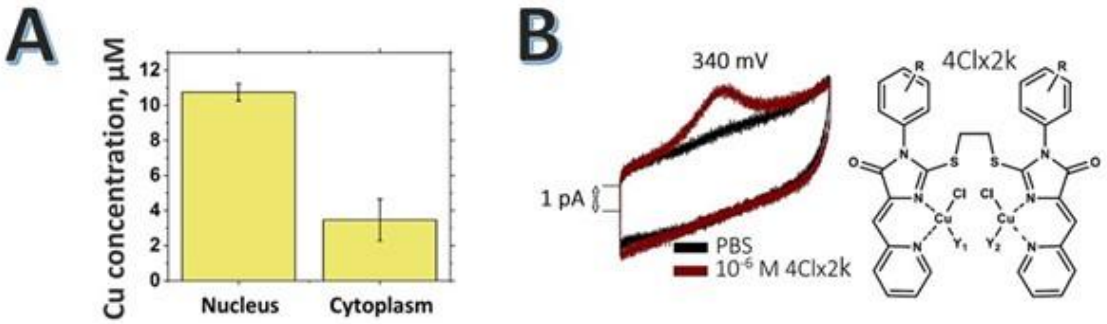

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Figure 2. A) Demonstration of imbalance in copper distribution in single cells B) Cyclic voltammetry obtained in the presence and absence of a copper compound (scan rate 400 mV/s) and structure of copper-containing compound

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