VISUALIZATION OF THE INTERACTION OF NANOPARTICLES WITH BIOLOGICAL CELLS BY AFM

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Abstract – In the last decade, the interest to the interaction of nanoparticles with biological cells is increased. The most promising and little studied area of nanoparticles application is medicine. Due to the unique properties nanoparticles of silver, zinc, copper and others are used against certain viruses, for the treatment of burns, destruction of tumor cells, as a part of the drugs delivering system to some organ. It is known that nanoparticles can have an impact on the functional state of the cells. One method that can be used to assess the state of the cells is an atomic force microscopy (AFM). ACM can not only visualize, but to assess the changes in the mechanical characteristics of the objects. There are many modern techniques for the visualization of biological microorganisms. Atomic force microscopy was chosen because it allows you to get a true three-dimensional surface topography and does not require complex sample preparation. Further investigation using the dynamic mode AFM allows to obtain sample microtomography. This mode allows you to automatically obtain a set of bundle AFM images of the same area at different values of the load applied to the scanning process. This study demonstrated approach for visualizing the interaction of nanoparticles with the cell and its internal structure by micromechanical destructive impact.

I. INTRODUCTION

Due to the nanomaterial's application in various fields of human activity it is necessary to study the biological effects of various nanoparticles and nanomaterials, especially their action on the human and animals organisms. The actual task is to determine the degree of nanoparticles toxicity for humans and, therefore, the potential risk of nanoparticles and medicines on their basis. For the last decade, the data about positive (therapeutic effect) and about negative (stimulation of various diseases) effects of metal nanoparticles on living organisms are accumulated. One of the most popular objects of investigation is silver nanoparticles. Mainly research works are connected with the determination of the antimicrobial activity of nanoparticles [1 – 3]. The data on the effects of silver nanoparticles (Ag) at the higher organisms are not numerous. Investigation of nanosilver toxicity, showed that cell's vitality after interaction with Ag nanoparticles depends on the cell's type, on nanoparticles size, and their concentration [4].

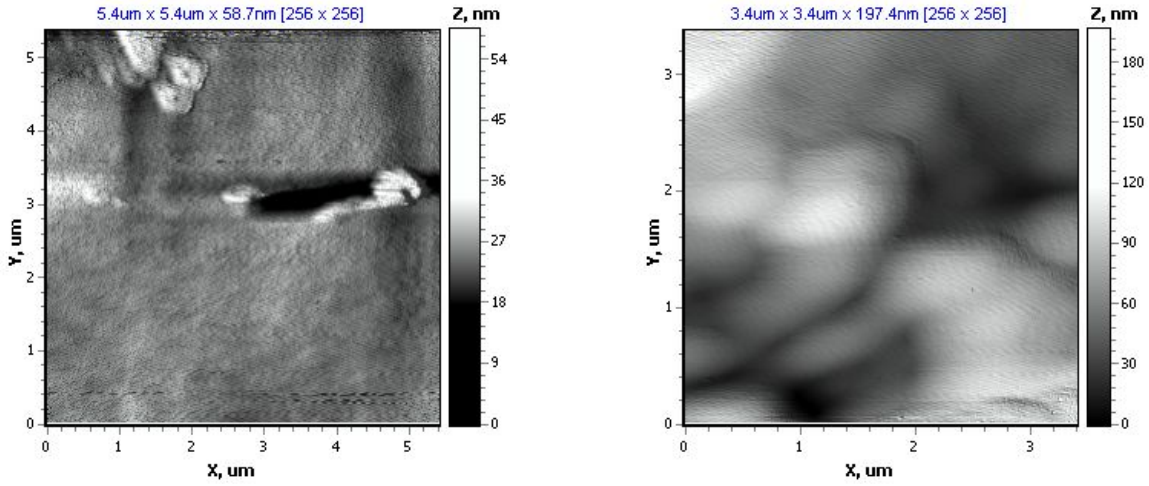
The purpose of this work was to visualize the nanoparticles on the cellular membrane surface. This task can be solved by the method of atomic force microscopy (AFM). This method allows to visualize the cell surface, and also to determine the way of interaction of nanoparticles with biological cells: by means of linkng of nanoparticles with a surface of the cellular membrane or by means of nanoparticles penetration into the cell – endocytosis.

II. MATERIALS AND METHODS

In this research we used the atomic force microscope "NT-206" ("MikroTestMashines" Belarus). The CSC 38 probe («MicroMash») with the curvature radius of 30 nm and console rigidity of 0.03 N/m was used in the static mode and the NSC 11 probe with the curvature radius of 30 nm and console rigidity of 3 N/m - in dynamic mode. The object of investigation is the culture cell line MDBK (bovine kidney cells), and silver nitrate nanoparticles ($AgNO_3$).

III. RESULTS

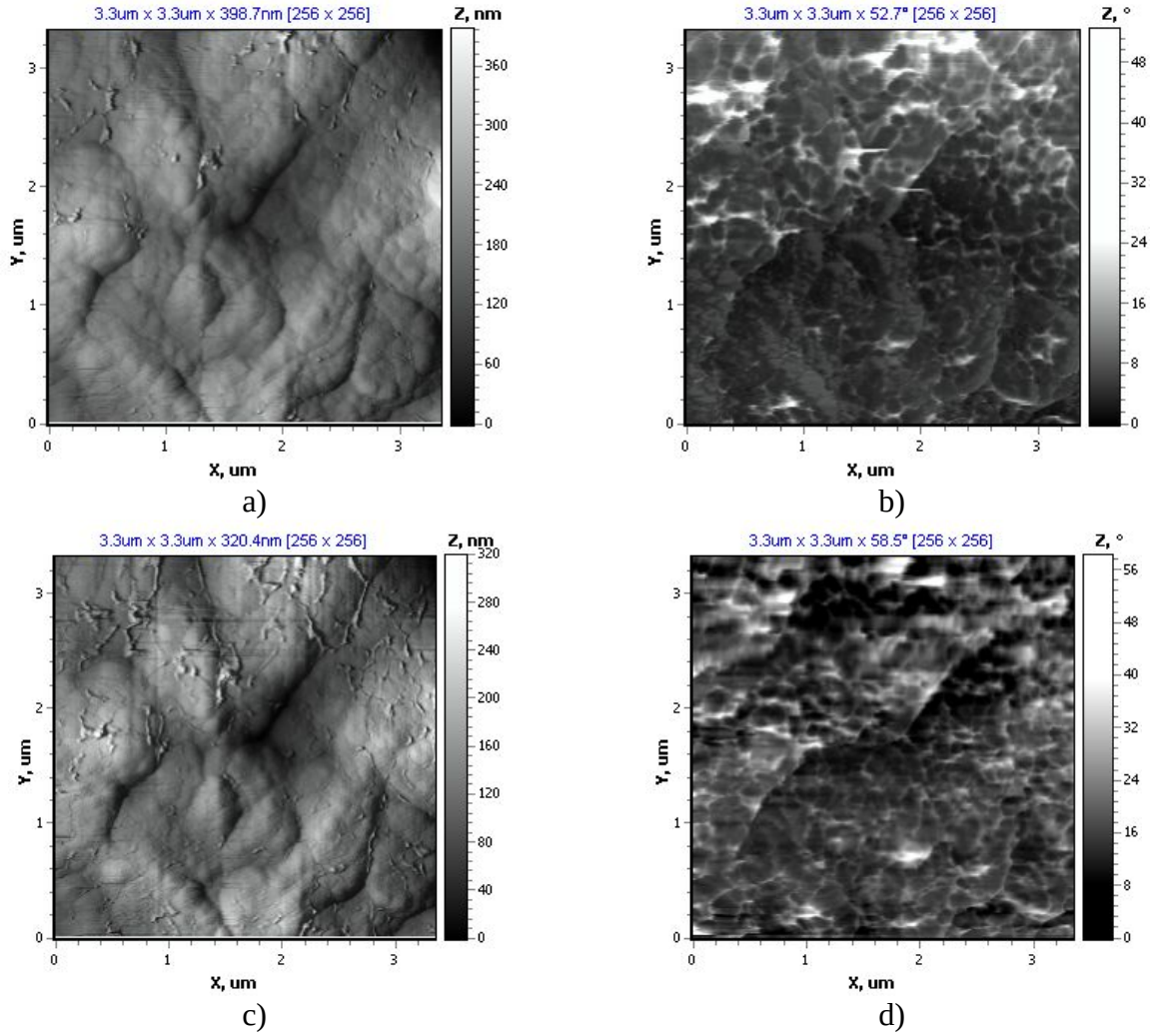
Static mode of AFM allows to visualize the surface of cell membranes and can be used as a control method of binding of the nanoparticles with the cell surface. The dynamic mode allows to reveal the objects in case of their penetration into the cell. Figure shows the cell surface with nanoparticles (Fig. 1a) scanning in contact mode and cell's surface after the cells incubation with nanoparticles (Fig. 1b).



a) b)

Figure 1 – AFM images of the surface area of the MDBK cell:
a – nanoparticles on cell's membrane – scanning area 5,4×5,4 µm;
b – cell's membrane after the cells incubation with nanoparticles – scanning area 3,4×3,4 µm

The investigations in dynamic mode were made also at different load (from 30 till 90 %). The task was to visualize the internal structure of the cell. It is established that when the load increases to 70 % of the microstructure of the cell membrane is visible more distinctly (Fig. 2 c, d).



a) b)

c) d)

Figure 2 – AFM images of the cell's surface: a, b – at 40% load; c, d – at 70% load;
a, c – topography of the cell surface, c, d – phase contrast image; scanning area 3,3x3,3 µm

IV. Conclusions

Thus, it is established that it is possible to define a way of interaction of nanoparticles with biological cells by atomic force microscope. Changing the load applied during the scanning proses it is possible to obtain the images of the internal structure of the cell.

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