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Modern methods of atomic force microscopy in the biomedical research

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Modern atomic force microscopy (AFM) plays an important role in biomedical research. By selecting different scanning modes and corresponding probes, AFM allows investigation of the structure of both fixed histological sections and living biological objects, as well as very soft adhesive structures, such as natural hydrogels.

Here, we present recently introduced methods in AFM, developed specifically for the biomedical research.

PeakForce Tapping® is a relatively new mode introduced by Bruker. The mode based on the processing of force-distance curves in each point of measurement allows combination of a mild force load on the sample and a high resolution, which is especially important for imaging soft biological samples such as cells.

PeakForce QNM® (Quantitative NanoMechanics, by Bruker) is a scanning mode with the simultaneous measurement of topography and different material properties, such as Young's modulus, adhesion, deformation etc., at the nanoscale. Using PeakForce QNM®, we studied a variety of biological objects using a Multimode 8 atomic force microscope by Bruker. For example, we studied vocal fold tissues of rabbit – normal tissues, scar tissue and the scar tissues after the treatement with autologous mesenchymal stem cells (MSC). We have shown the differences between the normal and scar tissues in the packing of collagen fibrils, their thicknesses and Young's moduli. Besides, we have shown that, after the MSC treatment, both the collagen packing and Young's modulus resemble those of the normal vocal fold tissue that indicates the restoration of the original tissue's elasticity.

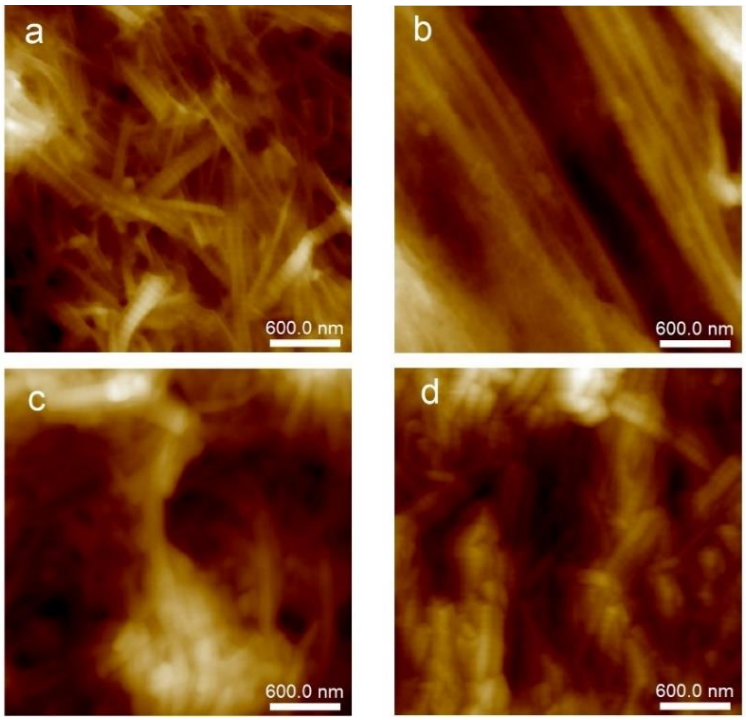

Figure 1. AFM of native matrix and the AT types: (a) native matrix of rib cartilage; (b) "classic" type of AT; (c) "fine-fibred" type of AT; (d) "intertwined" type of AT. All images have a scan size of 3x3 μm^2.

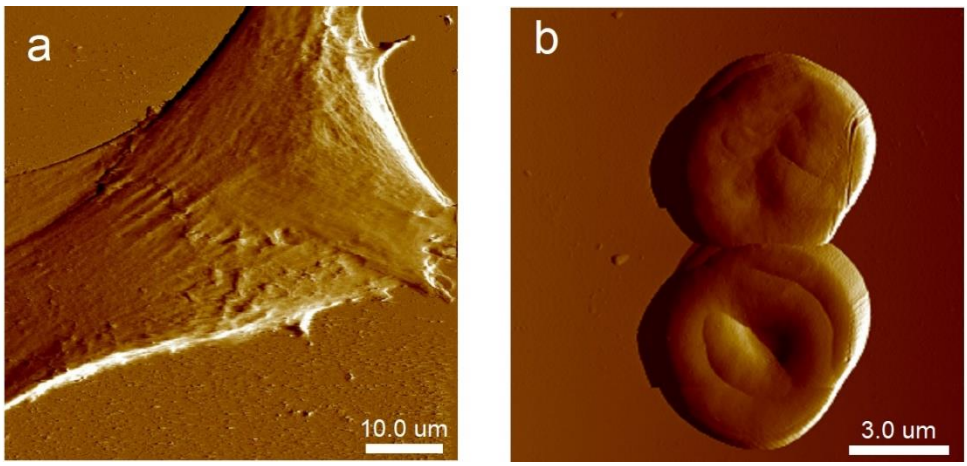

Figure 2. AFM images of live cells in the cell media. (a) human MSC, scan size 60x60 μm^2; (b) Two types of human erythrocytes: spherocyte and discocyte, scan size 14x14 μm^2.

The Fast Force Volume regime allows mapping of Young's modulus and stiffness with the preset number of points on the object's surface. It is convenient for very soft materials, such as hydrogels and certain live cells, where imaging with the Peak Force QNM® regime is not possible.

In Figure 1, we display how PeakForce Tapping® - PeakForce QNM® on air was used to study the pathology of rib cartilage in children aged 8-17 years with congenital deformations of the chest – pectus excavatum (PE) and pectus carinatum (PC). We characterized three new types of amianthoid transformation (AT) of the costal cartilage collagen fibers in children: a "classic", a "fine-fibred" and an "intertwined" type. All the AT types represent different stages of extracellular matrix transformation and have different packing and structure of collagen fibers.

In Figure 2, we demonstrate application of PeakForce Tapping® for imaging live cells in their own cell medium, using a Bioscope Resolve AFM (Bruker).