CHANGES IN CELLULAR STIFFNESS RELATED TO CANCER-INDUCED CYTOSKELETON REORGANIZATION

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Introduction

Cellular mechanics undergo complex changes during cancer progression in parallel with metabolic reprogramming due to altered demands on its motility. As the cytoskeleton forms a major part of cellular stiffness, this study focuses mainly on exploring cytoskeletal changes in cancer cells and evaluating their direct impact on cellular stiffness. However, the association between cellular stiffness and tumor cell aggressiveness is not straightforward and results vary depending on tumor type. The main aim of this study was to explore the relationship between the altered inner structural arrangement and cell aggressivity in prostate cancer cells using a combination of laboratory and computational methods.

Methods

The stiffness of prostate cancer cell line 22Rv1 and more aggressive PC-3 cells cultivated in an adherent state was evaluated using Atomic Force Microscopy (AFM) [1] with a spherical tip. Mechanical testing confirmed an increase in Young's modulus for more aggressive prostate cancer cells. The amount of cytoskeletal proteins between cell types was determined using mass spectrometry-based proteomic analysis. To reflect on these changes and assess their individual impact on the cell's mechanical response we used a previously-developed hybrid Finite Element Model of a cell body that comprises all three types of cytoskeletal fibers (actin bundles, intermediate filaments and microtubules) together with nucleus, membrane and cytoplasm (see Fig. 1) [2,3] under loading conditions mimicking the series of AFM measurements (example in Fig. 2).

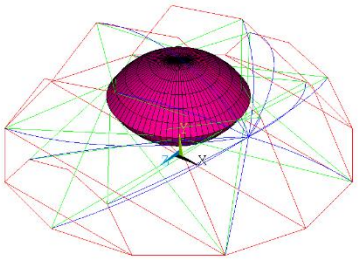

Figure 1: Inner structure of the FEM model [2]. Actin bundles (red), intermediate filaments (green), microtubules (blue) and nucleus (magenta), here shown without cytoplasm and membrane.

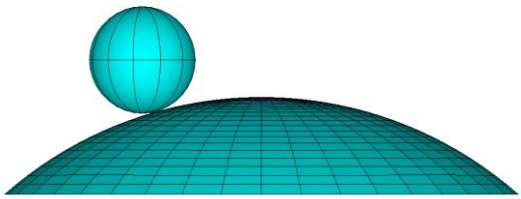

Figure 2: The setup for the simulation of AFM tip in an offset position to mimic the experimental setup.

Results

By determining the amount of tubulin, vimentin and actin in 22Rv1 and more aggressive PC-3 cell lines, we uncovered that whereas tubulin content stays almost unaltered, the stiffer PC-3 cells differ significantly in actin and vimentin content. The inner organization of the FE model is manipulated to correspond with these observations, which helps us to reveal the significance of individual cytoskeletal fibers in cancer cell mechanics. The simulations reflecting the observed changes in morphology and quantity of cytoskeletal components do not induce such stiffness differences as in the AFM experiments. This indicates that the stiffness alterations may be caused by cytoskeletal rearrangement or the existence of another mechanically relevant cell component that has not been reflected yet in the computational modeling approach.

Future prospects

Even though the cytoskeleton has been given more attention in computational cellular mechanics than other organelles, it is not entirely ruled out that there are other contributing factors such as bonds between organelles. For instance, the reorganization in the cancer cell cytoplasm also includes changes in the architecture of mitochondria, which is responsible for generating energy for the cell. Thus, the attention will be directed toward the mitochondrial organization within the cytoskeleton together with their mutual interactions and how it impacts cancer cell mechanics.

References

1. Raudenska, M. et al. Sci Rep 9, 1660 (2019).
2. Jakka VVSV and Bursa J. Comput. Biol. Med, 143(105266), 2022.
3. Jakka VVSV, Bursa J. Biomed Res. Int. 2021. Article ID 8847372

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