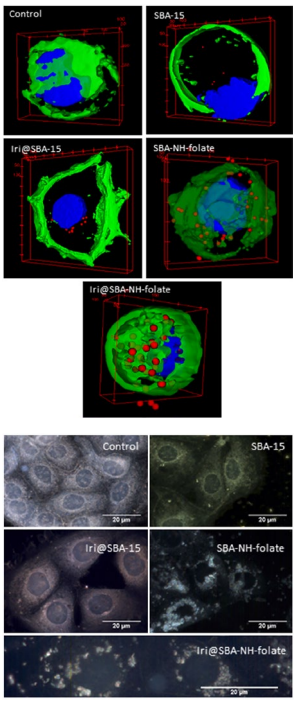
Nano-Biomaterials' Interactions with Cells: Challenges and Pportunities

Mihaela G. MOISESCU
Biophysics and Cellular Biotechnology Department, Excellence Center for Research in Biophysics and Cellular Biotechnology, Faculty of Medicine, Carol Davila University of Medicine and Pharmacy, 8 Eroii Sanitari Blvd., 050474 Bucharest, Romania
mihaela.moisescu@umfcd.ro

Development and engineering of nanomaterials (NMs) for efficient therapy of various pathologies (*e.g.*, cancer) know an extensive progress over the last two decades with special attention given to modulating the material properties through synthesis processes (*e.g.*, chemical structure, size, area, pore volume, chemical groups attached to the surface, *etc*) aiming to improve targeting and drug delivery. Another key point is a deeper knowledge of materials effects on living cells by NMs accurate tracking from membrane penetration to NMs final intracellular destination. Various NMs physico-chemical parameters as well as the type and biological features of the cell influence the intracellular fate of the nanomaterial. Thereupon several cell-based, including label-free technologies got an increasing attention in the preclinical NMs-based drug delivery studies.
During the talk, it will be presented our group's experience in evaluating the effects of several categories of NMs carrying cytotoxic molecules (*e.g.*, irinotecan) and under various functionalization *(e.g.*, folate, fucoidan) on cells in culture (*e.g.*, murine fibroblasts, human colonic adenocarcinoma cells). Results obtained using end-point tests (like formazan-based colorimetric tests, comet assay for DNA) as well as real-time label-free evaluation of cell growth (like impedance-based cell technique) will be discussed for different nanoparticles in terms of chemical structure (like mesoporous silica, zinc oxide) or molecules modifying the cellular processes involved in the NMs incorporation (like inhibitors of endocytosis).



Caco-2 cells incubated 24h with SBA-15 mesoporous silica nanoparticles functionalized with folate and loaded with irinotecan [1].
(up) 3D reconstructions of cells (cytoplasm green, nuclear DNA blue and nanoparticles red). Green zone appears discontinuous for visualization purposes.
(down) Hyperspectral images of cells where the intracellular accumulations of nanoparticles can be seen as bright spots.

A combination of visualization techniques implying fluorescence, hyperspectral and enhanced dark field microscopy was used to quantitatively evaluate the penetration and localization of NMs within a cell.
A methodology for processing serial Z-stacks of fluorescent and dark field images was developed to obtain 3D cell images [2]. Then, the nanomaterials concentrations (zinc oxide or SBA-15 nanoparticles) were calculated in 3D reconstructions of cellular compartments like nucleus, cytoplasm, shells adjacent to nucleus or membrane.
Moreover, by hyperspectral microscopy not-labelled NMs were intracellularly found, counted, and tracked in correlation with their spectral signature
A preferential accumulation of NMs in the close vicinity of specific cell structures will be presented and the advantages and limitations of these combined techniques will be discussed. It will be shown that the cytotoxic effects of some drugs were enhanced when delivered by NMs compared to administration in homogeneous solution.

References

[1] L.C. Miclea, M. Mihailescu, N. Tarba, et al., *Nanoscale*, **2022**, *14*, 12744.
[2] M. Mihailescu, L.C. Miclea, A.M. Pleava, et al., *Biomed. Opt. Express*, **2023**, *14*, 2796–2810.