P12-42
Comparative study of the cytotoxicity of hydroxyapatite, tricalcium phosphate and calcium phosphate nanomaterials on Panc-1 and HEK293 Cell Line

S. Cesmeli, C. Oksel Karakus
Izmir Institute of Technology, Department of Bioengineering, Izmir, Turkey

Calcium phosphate-based bioceramic nanoparticles have been actively used in a range of therapeutic applications. Although they are mostly considered as biocompatible materials, the circulation of nanoparticles in the bloodstream raise further questions as to what degree of cellular damage they are capable of causing once carried out to vital organs such as kidney and pancreas. Therefore, there is a clear need to explore potential cellular damage induced by commercially used bioceramic nanoparticles such as hydroxyapatite (HAp), tricalcium phosphate (TCP) and calcium phosphate (CaP).

Cytotoxicity can be determined based on different parameters in cells, such as membrane integrity and mitochondrial damage. In particular, mitochondrial damage is quantified by the increase in the activity of mitochondrial dehydrogenases, resulting from an expansion in the number of viable cells. Mitochondrial dehydrogenases forms formazan dye after the reaction with water-soluble tetrazolium salt (WST-1) reagent. The disruption of the cell membrane results in the translocation of cellular components (i.e., phospholipids, proteins, and enzymes) from inside the cell to the outside. Lactate dehydrogenase (LDH) is a cytosolic enzyme which is released upon cell lysis and can be measured by a reaction in which tetrazolium salt is converted into a red formazan product. The formation of reactive oxygen species (ROS) is another important mechanism which is known to cause damage in the basic building blocks of the cell including DNA, protein and lipids.

In this study, two different cell lines, pancreas cancer (Panc-1) and human embryonic kidney (HEK293), were exposed to varying concentrations of HAp, TCP and CaP nanoparticles. The cytotoxic effects were assessed by WST-1, LDH and dichlorofluorescein diacetate (DCFDA) assays. Prior to toxicity assessments, proliferation experiments for both cell lines were conducted to assess possible interferences with assay components and to determine the optimal number of cells. No interference was observed between bioceramic nanopowders and LDH assay components, indicating that it can be used to evaluate cytotoxicity of nanoparticles. As expected, cell line- and nanoparticle-specific differences were observed in the cytotoxicity of tested materials. In particular, TCP showed relatively higher toxicity compared to Hap and CaP on Panc-1 cell line. None of the tested nanoparticles induced cytotoxicity on HEK293 cell line. Results of ROS assay were in line with cell viability assessments. Taken together, these findings suggest that bioceramic nanoparticles do not induce any significant cytotoxic effect and can be safely used in biomedical applications.

References
