In Vitro Immunotoxicity of Superhydrophilic Superparamagnetic Iron Oxide Nanoparticles

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1. Introduction

During the last decade, superparamagnetic iron oxide nanoparticles (SPIONs) have attracted the scientific community's interest due to the multitude of applications in many fields of biomedicine namely as contrast media in diagnosis or as carriers for targeted drug delivery.

2. Purpose

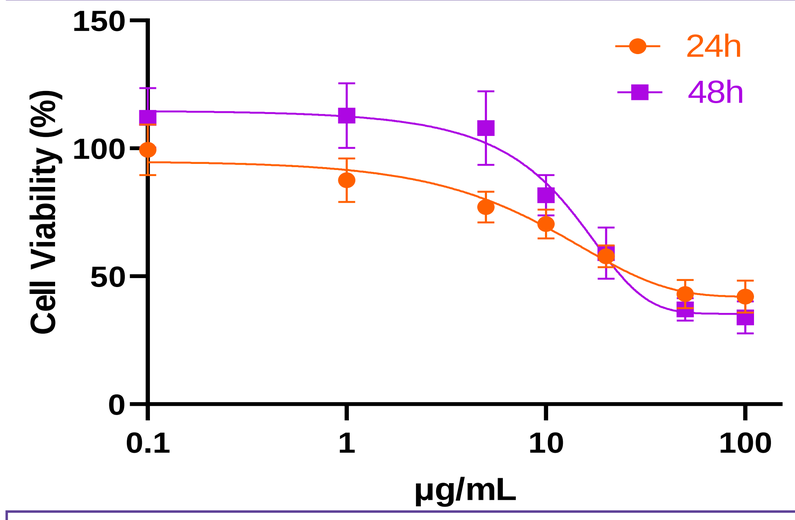
We investigated the immunotoxicity of hydrophilic and surface functionalized SPIONs *in vitro* against THP-1-derived macrophages.

3. Methodology

a. Estimation of cell viability of THP-1 cells (acute monocytic leukemia cells) by colorimetric MTT assay.
b. Quantification of intracellular production of Reactive Oxygen Species (ROS) by flow cytometer under the same conditions and after staining with DCF-DA.
c. Induction of Apoptosis/Necrosis by Annexin V-FITC and Propidium Iodide (PI) double staining after 24 and 48 hours of exposure to different concentrations of SPIONs and flow cytometer analysis.
d. Estimation of the percentages of cell population in different phases of the cell cycle (PI staining).

4. Results

4a. Cell Viability

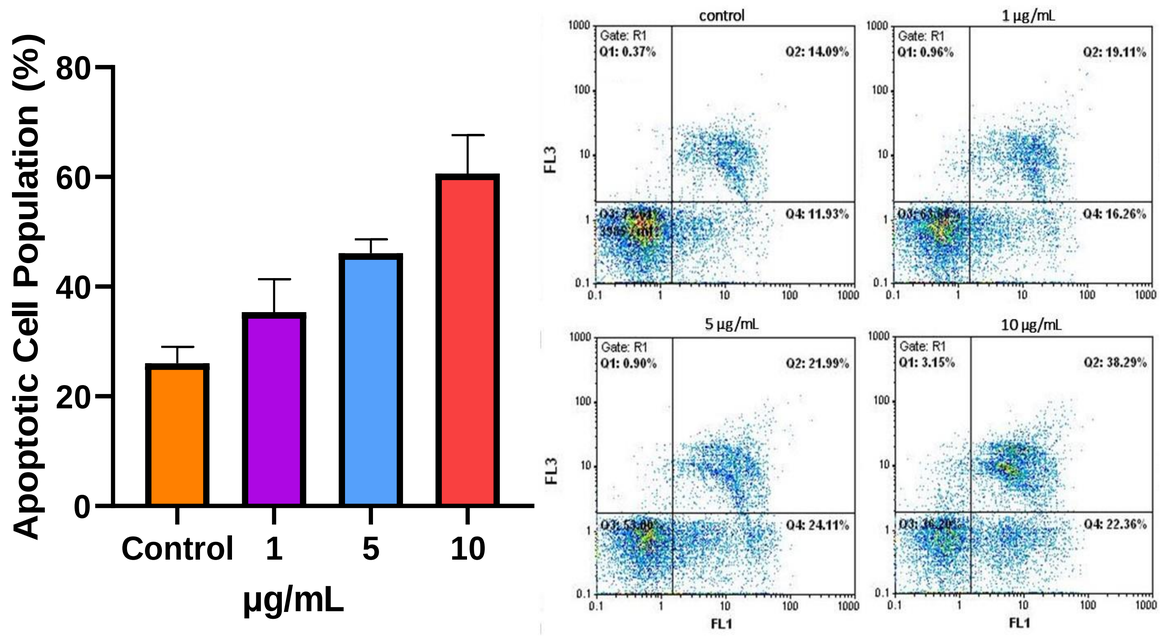


A dose-dependent reduction in cell viability was recorded after treatment for 24 and 48 hours, which was more evident at doses higher than 5 µg/mL.

4b. ROS Production

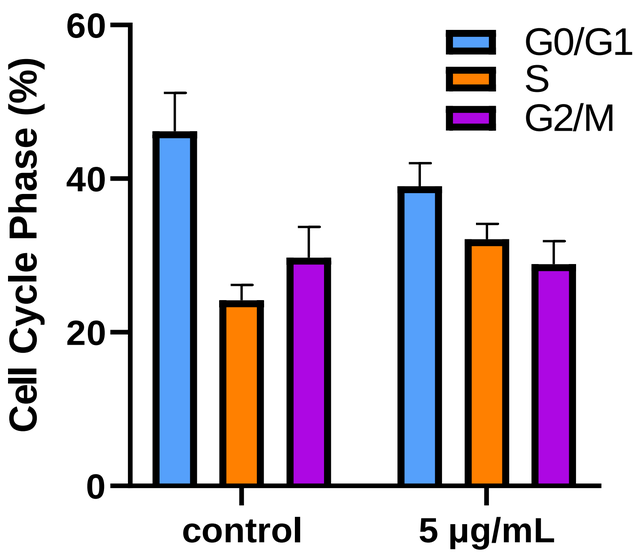
A mild increase in ROS formation was noted at doses lower than 2.5 µg/mL. However, at higher doses, ROS formation by SPIONs was more intense.

4c. Induction of Apoptosis



SPIONs also induced a dose-dependent increase in the apoptotic cell population. Specifically, apoptosis was increased 1.4x, 1.8x and 2.4x fold after treatment with 1, 5 and 10 µg/mL for 24 hours.

4d. Cell Cycle Analysis



Cell cycle analysis revealed that SPIONs (5 µg/mL) arrested THP-1-derived macrophages at S-phase (increased by 8%).

	Control	5 µg/mL
G0/G1	46.2±4.0%	39.0±3.3%
S	24.2±2.8%	32.1±2.5%
G2/M	29.7±3.9%	28.9±3.4%

5. Conclusions

Our results showed that the ultra-small SPIONs (diameter ~4 nm) induced immunotoxic effects causing the death of THP-1-derived macrophages. Nonetheless, this novel synthetic approach allows proceeding to further modifications to produce improved SPIONs that can deliver their beneficial promises to the biomedical field.

6. Bibliography

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Demigod Project

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