## Multifunctional NGF-Au-SPIO Nanoparticles: Magnetically Directing Neurite Extension and Orientation

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Multilayered nanoparticles are promising carriers to deliver therapeutic biomolecules across the blood-brain barrier (BBB) for neurodegenerative diseases and traumatic brain injuries. Capable of tissue imaging, drug delivery, and photothermal therapy, multifunctional nanoparticles can also generate mechanical tensile forces when combined with an external magnetic field as a novel means to enhance nerve repair. In this study, superparamagnetic iron oxide (SPIO) cores coated with gold nanoshells were synthesized by the co-precipitation and iterative hydroxylamine seeding methods, respectively. TEM imaging, UV-Vis spectroscopy, and zeta potential tests characterized the magnetoplasmonic nanospheres with a mean diameter of  $21.2 \pm 4.4$  nm as stable and monodisperse. Nerve growth factor (NGF) functionalized to Au-SPIO nanoparticles (NGF-Au-SPIONs) were then cultured with PC12 phochromocytoma cells, leading to prolonged cell survival and proliferation at 72h. An innovative 3D-printed Halbach configuration was designed to induce a strong 0.5 T radially outward magnetic force to accelerate nerve regeneration. NGF-Au-SPIONs (10 µg/ml) induced neuronal differentiation of PC12 cells (>31% under magnetic effect), along with significantly increased neurite length and preferential alignment toward the magnetic force (p<0.05). Data supports the novel NGF-Au-SPIONs across an in vitro BBB model and stem cell tracking via magnetic resonance imaging (MRI). Ultimately, multifunctional nanoparticles can be accurately guided by an inexpensive magnetic device, serving as one of the first models to treat both neurological disorders and cancers in the brain.

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