

Conjugated Nanoparticle Induced Apoptosis in Cancer Cells

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Abstract:

The activity of cells is highly dependent on their extracellular environments composed of soluble factors, surrounding cells and extracellular matrices (ECMs). These extracellular stimuli activate intracellular machinery called signal transduction, which directs the cells to a specific phenotype. The purpose of this project was to develop new materials to chemically and mechanically engineer cellular environments and look at their impact on cellular activities using a nanobiointerface and nanomaterial synthesis and characterization.

Summary of Research:

X polymer was conjugated with a Y nanoparticle in an effort to induce apoptosis in cancer cells of Z cancer type. Preparation of the conjugated particle included steps to wash the particle and obtain an adequate size before inserting the conjugated particle in the cancer cell. Obtaining an adequate size involved putting the functionalized nanoparticle through several cycles of ultra-centrifuge utilizing an adequate solvent that mimicked the natural body environment that Z cancer cells are found in. The ultra-centrifuge process allowed for an effective particle size, and sufficient collection of functionalized nanoparticle concentration.

Particle concentration was confirmed utilizing a spectrometer. A comparison sample's concentration utilizing the manufacture's information was utilized to determine if the concentration collected was enough to perform cellular experiments. Sufficient concentration allowed for the testing protocol to determine if the polymer in question was binding to the correct site of the Z cancer cell.

Additionally, Western Blotting was followed to analyze the phosphorylation levels of the Z cancer cells after insertion of the functionalized nanoparticle.

The beforehand procedures allowed for fruitful results for further cellular experiments to include apoptosis.

In preparation for the apoptosis experiment, the protocol for Annexin V was followed. In addition of apoptosis testing, necrosis was also tested for utilizing PI. A control group and a group with the soluble polymer was also prepared for comparison purposes with our sample.

Results and Conclusions:

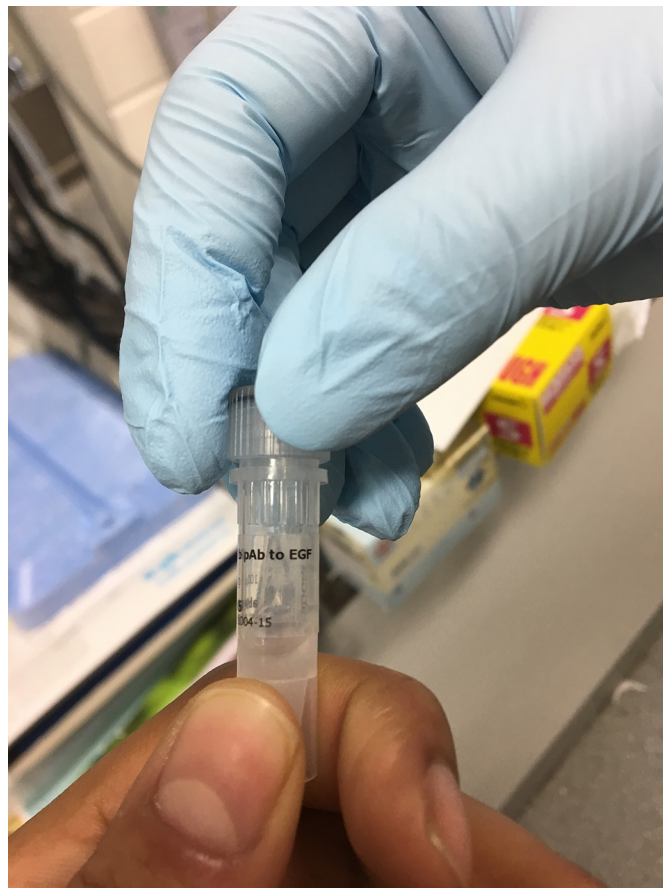
Putting the conjugated nanoparticles through an ultra-centrifuge procedure proved to be an effective method to separate the functionalized particles from the non-functionalized particles. The spectrometer revealed a sufficient enough concentration to performed the cellular experiments. Thus, confirming that the set ultra-centrifuge parameters were adequate and efficient. The phosphorylation levels obtained contradicted prior results and expectations, therefore, further analysis of the polymer concentration on the nanoparticle needs to be performed. The Annexin V protein group was successful in the determination of apoptosis. The PI group allowed for the analysis of necrosis in the Z cancer cells. The apoptosis and necrosis levels observed were promising thus fortifying the need to continue studies to understand the mechanisms in play during the apoptotic and necrotic stage.

Future Work:

Further work needs to be performed to determine if the material we used is the most efficient and durable. Additionally, the size of the functionalized nanoparticle has been revised to explore the possibility of other sizes being more efficient at inducing apoptosis or necrosis. Lastly, further work is needed to understand the biological and physical mechanism from the moment of insertion to the apoptotic stage as many unknowns remain.

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Isabel works on her project in the lab at NIMS.



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