Green Synthesis of Silver Nanoparticles for Biological Interaction Studies

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Statement of Purpose: Silver occupies a unique niche in the field of nanotechnology due to its time tested effectiveness as an antimicrobial agent. Recent results have shown that silver nanoparticles exhibit size dependent interactions with the HIV virus that result in virucidal action [1]. There have also been some studies that indicate that silver nanoparticles may possess unique regenerative capabilities for tissue engineering due to proposed cellular level effects. The goals of this study are to 1.) Investigate the mechanism of interaction between bovine serum albumin and silver ions in the formation of colloidally stable silver nanoparticles, 2.) Control the chemical and physical properties of the nanoscale particles for assessing their modes of biological interaction.

Methods: Silver nanoparticles were synthesized in aqueous solvents using a green chemistry approach. Aqueous solutions of silver ion (0.01-0.1 M) were mixed with bovine serum albumin (BSA). The silver – BSA solutions were subsequently mixed with two reducing agents (sodium borohydride, ascorbic acid) and allowed to react under mild conditions at room temperature. The silver nanoparticles were characterized via dynamic light scattering, X-ray diffraction, Nitrogen gas adsorption, thermal gravimetric analysis, and Fourier transform infra-red spectroscopy to study the interaction between BSA and silver ions in solution and BSA and the silver nanoparticles after the reduction reaction. The chemically synthesized silver nanoparticles were exposed to model bacteria (E. coli) and mammalian cells in culture at controlled levels to investigate toxicity effects.

Results/Discussion: Silver nanoparticles were formed from aqueous solutions containing BSA and the reducing agents sodium borohydride and ascorbic acid. BSA seems to play a dual role in the reaction sequence for synthesizing silver nanoparticles. Silver is known to form protein complexes. The addition of silver salt solution to a BSA solution resulted in the formation of a white silver-protein complex. The BSA silver complex immediately changed color upon the addition of the reducing agents. In the case of sodium borohydride, the nanoparticle suspension turned dark brown due to the reduction of the complexed silver ion. The addition of ascorbic acid to the BSA-silver complex resulted in a very dark green/black colloidal suspension. Size analysis results from DLS indicates that both reducing agents resulted in silver nanoparticles, but the sodium borohydride system yielded smaller particles (< 20 nm vs. < 50 nm). BSA worked effectively for stabilizing the silver nanoparticles at lower silver concentration. The degree of particle agglomeration increased with increasing silver ion concentration. The biological activity of the silver nanoparticles during exposure to E. coli confirmed the antimicrobial action of silver nanoparticles. Ag nanoparticles were also toxic to mouse preosteoblast cells at the 1000, 500 and 100 ppm level.

Conclusions: Colloidally stable Silver nanoparticles can be synthesized using BSA and the reducing agents sodium borohydride and ascorbic acid. BSA influences the reaction mechanism by forming a protein complex with silver ions in solution and adsorbing on the silver nanoparticle surface to prevent particle aggregation. The silver nanoparticles were toxic to E. coli bacteria and MC3T3-E1 cells at levels ranging from 100-1000 ppm. Future studies will investigate the concentration dependent toxicity of silver nanoparticles to see if there are levels that lead to beneficial effects.

References: [1] (Elechiguerra JL et al.J Nanobiotech. 2005;3:6.)