Green Synthesis of Gold Quantum dots and Cadmium free Quantum dots for Cancer Cell Imaging: Haemocompatibility and cytotoxicity Evaluation.

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Statement of Purpose: Highly fluorescent quantum dots (QDs) have attracted significant interest in nanobiotechnoilogy. These nanomaterials were found to be superior over conventional organic dyes in many of their photophysical features. Largely QDs structures are based on heavy metal combinations like CdSe, CdTe and HgTe. Though they possess excellent fluorescence, their biological applications are limited due to the inherent toxicity by heavy metal ion release. Such applications demand biofriendly QDs simultaneously having the required emission properties. QDs have been extensively studied in bio imaging and targeted cancer therapy. However, investigations on their haemocompatibility, which is a mandatory in targeted therapy as the QDs has to voyage through the systemic circulation (blood) to reach the target are limited. Keeping this in mind we synthesized gold quantum dots by "green" synthetic route using bovine serum albumin (BSA), human serum albumin (HSA) and lysozyme. The generated QDs were then characterized by analytical techniques and evaluated for their haemocompatibility and cytotoxicity in comparison with the silica coated blue emitting ZnSe and glutathione (GSH) coated orange emitting CdTe QDs. Our results showed that highly fluorescent Near Infrared (NIR) emitting gold QDs can be synthesized using proteins and they are more haemocomapatible and less cytotoxic comparing to ZnSe/silica and CdTe/GSH QDs.

Methods: All the reagents were used as obtained without further purification until and unless stated. Deionised Milli Q water (DI) with a resistivity of 18.2 Ω m was used through out the experiment. $HAuCl_4$. $3H_2O$, bovine serum albumin (BSA), HSA, lysozyme, $CdCl_2$, Se, Te membrane dialysis bag (molecular weight cutoff 12kDa) were obtained from sigma-Aldrich. UV-Visible spectrophotometer (Varian Cary 50conc) was used for absorbance study. Fluorescence studies were conducted with Fluorescence spectrophotometer (Varian Cary Eclipse). The C6 glyoma cancer cells were used for cell studies. Human blood was obtained from unmedicated healthy volunteers.

Synthesis of QDs: A biofriendly method was followed for the synthesis of gold QDs. The BSA gold QDs (nanoclusters) were synthesized as reported elsewhere[1]. The other gold QDs were synthesized by the same procedure with minor modification. The synthesis of ZnSe/silica and CdTe/GSH were based on reported procedure with modifications[2-3].

Results: Through green approach, we synthesized blue emitting ZnSe/Silica QDs, protein coated NIR emitting gold QDs and orange emitting CdTe/GSH QDs.

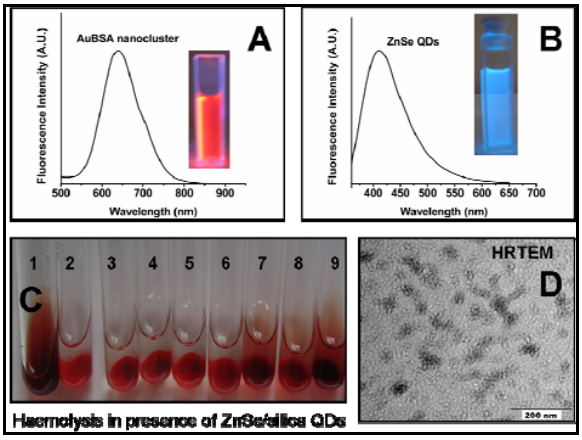

Figure 1. The fluorescence emission spectrum of BSA coated gold QDs and its image on UV excitation **(A)**. The fluorescence emission spectrum of ZnSe/silica QDs in water and its image on UV excitation (B). The haemolysis results **(C)** of sample **B** and its HRTEM characterization.

The synthesized nanostructures were characterized with instrumental techniques **(figure1).** We assessed haemocompatibility and cytotoxicity of these materials to explore their potential for targeted drug delivery and imaging applications. The haemolysis tests, platelet, RBC and WBC aggregation were performed using standard protocols. The silica and GSH coated QDs were shown more cytotoxicity to C6-glyoma cancer cells and were less haemocompatible compared to the protein generated gold nanoclusters (QDs). The haemolysis result in figure 1**(c)** shows concentration dependant haemolysis (**3-9**) compared to positive (**1**) and negative controls (**2**).

Conclusions: We synthesized highly fluorescent (NIR) gold QDs which were shown excellent biocompatibility indicating their suitability as NIR emitting QDs for in vivo applications. The *in vivo* imaging studies are in progress in our laboratory.

References:

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