Evaluation of Gold / Gold-Sulfide nanoparticles for use in photothermal ablation cancer therapy.

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Introduction:

Metal nanoshells, are a class of optically tunable nanoparticle that consist of a dielectric core and a metal shell [1, 2].We have previously used gold nanoshells with a silica core for photothermal tumor ablation [3, 4]. Silica gold nanoshells used in previous studies were on the order of 130-160 nm in diameter; this size allowed sufficient accumulation of nanoshells in the tumors through the enhanced permeability and retention effect (EPR), to allow successful photothermal ablation of the tumors. However we seek to improve the technology further by developing smaller more strongly absorbing gold nanoparticles for therapeutic purposes. Gold coated Gold-Sulfide (Au/Au_2S) nanoshells have been previously demonstrated [5]. The particles are formed through a self assembly process and the resulting suspension consists of solid gold colloidal particles as a contaminant as well as Au/Au_2S nanoshells. In this study we remove the majority of the colloidal gold contaminants to allow the remaining NIR absorbing Au/Au_2S nanoshells to be concentrated in sufficient quantities for use in heating assessment studies and in *in vitro* ablation of cultured cells using an 808nm laser.

Methods and Materials:

Nanoshell Fabrication:

Gold coated Gold-Sulfide (Au/Au_2S) nanoshells are made by self assembly. 2.3 mM chloroauric acid, $HAuCl_4$, (Aldrich, Milwaukee) and 3.1 mM sodium sulfide, Na_2S (Aldrich, Milwaukee) are mixed and aged for 24-48 hours. Ratios ranging from 1:1.5 to 1:2.5 (Na_2S:$HAuCl_4$) and reacted to completion to select for the best NIR absorbing nanoparticles. NIR absorption characteristics of the nanoshells were determined using a UV-Vis spectrophotometer (Carey 50 Varian, Walnut Creek, CA). After production, the suspension was centrifuged in multiple steps to remove the smaller colloidal gold particles from the supernatant thus concentrating the fraction of NIR absorbing nanoparticles in the pellet.

Heating Assessment of Au/Au_2S nanoshells.

1 ml samples of Au/Au_2S nanoshells diluted at varying total extinction were used in cuvettes with the 808 laser (Coherent, Santa Clara, CA) at varying power outputs from 2-7 watts. A K type thermocouple was inserted into the cuvette out of the path of the laser to measure temperature changes. Sample of Silica-Gold nanoshells were diluted to equivalent extinctions and measured similarly for comparison. Temperature measurements were performed on water as negative controls.

In vitro cell ablation.

PC-3, prostate cancer cells were cultured in well plates. Cells were incubated with nanoshells and media for 6 hours. Cells were rinsed with PBS and kept on PBS during exposure to laser. Cells were exposed to laser spot of 1.2 mm at 60W/cm^2 for 3 minutes.

Results/Discussion:

We have observed higher temperature increases and faster heating rates for Au/Au_2S nanoshells compared to silica / gold nanoshells at equivalent extinctions due to theoretical higher absorption efficiency.

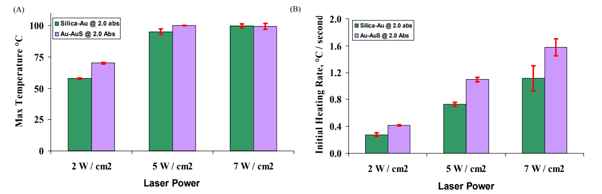

Figure 1: Au/Au_2S & Silica/Gold nanoshells (A) Final temperature after heating for 3 minutes of laser irradiation. (B) Initial temperature rate measured in the first 30 seconds of heating experiment.

After incubation with nanoshells and subsequent exposure to laser irradiation we performed calcein AM / ethidium staining and observed an area of cell death that corresponds to the laser spot size, this is similar to previous experiments and controls using silica gold nanoshells. We see no damage when cells are exposed to the laser only.

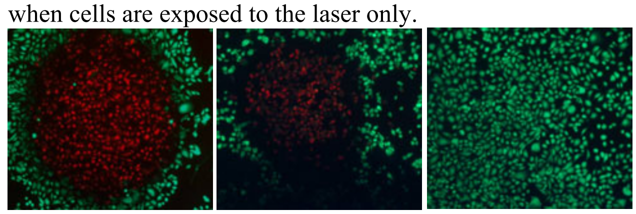

Figure 2: Demonstration of kill zone where cells were exposed to Laser + Au/Au_2S nanoshells (A) Laser + Silica/Gold nanoshells (B) or Laser Only (C).

Conclusions & Future Plans:

We have demonstrated that the Au/Au_2S nanoshells can heat faster and to a higher final temperature than silica gold nanoshells at equivalent extinctions and have demonstrated the ability to kills cells with this combination of nanoshells + laser. We believe that this will further our development of nanoshell based cancer therapies. We will optimize concentration of nanoshells and light intensity for cell ablation, investigate toxicity with these new particles and prepare for *in vivo* therapy.

References

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