CHITOSAN COATED NANOPARTICLES FOR INFLUENZA TREATMENT

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ABSTRACT
Influenza A virus infects 5-20% of the US population, resulting in over 200,000 hospitalizations and ~40,000 deaths annually. Much of influenza’s pathophysiology results from an over-exuberant immune response. Volatile anesthetics, such as isoflurane, are effective in suppressing the immune system. We hypothesize that a nanomedicine of Intralipid® nanoparticles loaded with isoflurane can modulate the immune system response and aid those infected with the influenza virus. When injected in its original formulation, the nanomedicine induces narcosis. To prevent narcosis, positively-charged chitosan is electrostatically bound to the negatively-charged Intralipid® nanoparticles. The coating allows for a controlled, localized release of isoflurane to the macrophages in the target organ (i.e., spleen). Through dynamic light scattering measurements, the size of the particles and the surface charge are measured to confirm the presence of the chitosan coating. Current testing indicates attachment of chitosan with aggregation of nanoparticles. Syringe filters are being used to remove large aggregates.

BACKGROUND
Volatile anesthetic preconditioning protection for ischemia/reperfusion (I/R) is ‘well’ studied in:
- Multiple species (human/rabbit/dog/rat)
- Multiple organ I/R injuries (heart, lung, liver, brain kidney)
Volatile anesthetic have off target properties (non anesthetic):
- Immune system modulation-which alters the response of Natural Killer cells caused by exposure to isoflurane
- Multiple organ I/R injuries

MATERIALS AND METHODS
- Intralipid®-20% soybean oil emulsion (Baxter Healthcare)
- Isoflurane-hydrophobic liquid at room temperature. BP 48.5 C
- Chitosan- MW 5000Da, polysaccharide. Does not elicit shelfish allergies
- In a 5 mL centrifuge tube, add 8.0% v/v isoflurane to 1 mL of Intralipid®. Cap and vortex for 15 seconds. Add 3 mg/mL chitosan to isoflurane-Intralipid® emulsion. Add isoflurane saturated water to fill. Cap and vortex for 15 seconds. Caprifuge isoflurane-Intralipid® emulsions at 18500 RCP for 15 minutes. Decant.
- Size and zeta potential determined by DLS (ZetaPals90, Brookhaven Instruments)

RESULTS
- Intralipid® emulsion droplets are 270 nm and -36mV.
- Chitosan concentration and mw influences chitosan iso-intralipid® emulsion droplet size
- Chitosan iso-intralipid® emulsion droplets are 400-1000 nm and +30 mV
- Adding either salt solution or glycol does not reduce aggregation of particles (data not shown)

RESULTS CONTINUED
- Addition of a chitosan coating to the Intralipid® emulsion increases the size of the particles and induces aggregation
- The large aggregates can be removed using filters but a loss of product is sustained

FUTURE DIRECTIONS
- Calculate the amount of isoflurane present in the coated nanoparticles using Raman and/or fluorescence spectroscopy
- Inject chitosan coated emulsion into mice to observe presence or absence of narcosis

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REFERENCES