Effect of the Addition of Chitosan Micro-beads Containing Iron Oxide Nanoparticles to the Chitosan Sponge Delivery System.

Alex P. Hoban, BS¹, Monica P. Rawson¹, Greg P. McGraw, BS¹, Warren O. Haggard, PhD², Joel D. Bumgardner, PhD², Jessica Amber Jennings, PhD¹.
¹Univ Memphis, Memphis, TN, USA, ²University of Memphis, Memphis, TN, USA.

Disclosures:

Introduction: Wound infections are becoming more difficult to treat with the introduction of increasingly antibiotic resistant bacteria. Local antibiotic delivery offers many advantages over systemic antibiotic delivery[1]. Chitosan is a biocompatible, biodegradable polymer that offers many advantages over other polymers in the drug delivery field[2, 3]. Our previous research has demonstrated that the chitosan sponge can be an effective, local antibiotic delivery system to combat and inhibit bacterial infections[4]. The development of a more patient specific stimuli responsive local delivery may prove useful as a therapeutic tool for the prevention and treatment of bacterial infections. Our goal in this study is to obtain preliminary data for the addition of iron oxide nanoparticle impregnated chitosan microbeads to the chitosan sponge for a stimuli responsive drug delivery vehicle.

Methods: The procedure from Jain et. al was followed to make 2 batches of chitosan microspheres with 40% w/v iron oxide nanoparticles. One batch was loaded with 5mg/ml amikacin and the other was not. Both batches of microspheres were then soaked separately in 5mg/ml amikacin solutions. These solutions containing the microspheres were then hydrated into chitosan sponges, frozen at -80°C, and lyophilized. 20ml of PBS was then added to each sponge and time points of 1, 3, 6, 24, 28, and 72 hours were taken. PBS was refreshed at each time point. Figure 1 shows the results of this 72 hour elution study. Chitosan sponges were made following procedure found in Noel et. al[4]. Iron oxide nanoparticles were made following procedure in Hu et. al[5]. A cytocompatibility study was also run on the iron oxide nanoparticles. This study was done using NIH 3T3 cells and varying concentrations of iron oxide nanoparticles. The DNA was measured in ng/ml at 1 day and 3 day time points.

Results: During the 72 hour elution study, there was found to be no statistical difference in the amikacin elution from the different amikacin loading procedures. All concentrations of amikacin were found to be above the MIC for Pseudomonas Aruginosa which has been reported between 2 ug/ml and 16 ug/ml[4, 6]. The sponges gave an initial burst release of 2300 ug/ml for the sponge with microbeads not preloaded with amikacin and 2023 ug/ml for the sponges with microbeads preloaded with amikacin. Over the 72 hours, this amikacin concentration steadily declined to 19.42 ug/ml and 23.00 ug/ml respectively by the 72 hour time point. The 72 hour elution figure shows the elution results for the 72 hour elution study. The results of the cytocompatibility study on varying concentrations of iron oxide nanoparticles are shown in the Cytompatibility figure. These results show that concentrations up to 0.31 mg/ml have no negative effect on NIH 3T3 cells.

Discussion: Results show that there is no statistical difference in the elution of amikacin from this chitosan sponge-microbead composite. The levels of amikacin eluted from the sponge-microbead composite were found to be above the MIC for pseudomonas aruginosa which is reported between 2ug/ml and 16ug/ml[4, 6]. A limitation of this preliminary study is that there was no external stimulus to cause a release of the loaded amikacin from the chitosan microbeads. The effect of an external stimulus will be evaluated in future studies. Further, the addition of the iron oxide nanoparticle impregnated chitosan microbeads to the chitosan sponge delivery system should be evaluated in vivo prior to any clinical application. The results from the cytocompatibility study show that iron oxide nanoparticles have little to no effect on NIH 3T3 cells when under 0.31 mg/ml concentrations.

Significance: The results of these studies are useful in determining if the addition of iron oxide impregnated chitosan microbeads to the chitosan sponge can create a viable stimuli responsive drug delivery system.

Acknowledgments: Funding for this research has been provide by the Fed Ex Institute for Technology.

References:
72hr Amikacin Elution study

Hours of elution

Concentration in ng/ml of amikacin

- Amikacin loaded after manufacturing of microspheres
- Amikacin loaded during and after manufacturing of microspheres