The Elution Behavior of Daptomycin Loaded Calcium Sulfate Pellets

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Statement of Purpose: This study investigated an optimal method to incorporate daptomycin (Cubist Pharmaceuticals; Lexington, MA) in a degradable bone graft substitute, calcium sulfate. Daptomycin is an antibiotic that has activity against antibiotic-resistant Gram-positive organisms and could be useful in the local treatment of musculoskeletal infections.

Methods: Calcium sulfate hemihydrate ($CaSO_4$• ½ H2O) powder was mixed with a potassium sulfate solution (4% K_2SO_4 by weight in DI water, a known accelerant), which was mixed with varying weight percentages of daptomycin per powder weight (2%, 4%, and 6%). The mixing ratio was 0.25g of K_2SO_4 solution per 1g $CaSO_4$ for the 2% and 4% daptomycin percentages, and 0.31g solution per 1g $CaSO_4$ for the 6% daptomycin pellet (a higher solution amount was required to allow dissolution of daptomycin into the mixture). After mixing the $CaSO_4$ powder and K_2SO_4 solution for two minutes, lyophilized daptomycin was added at a weight to bring the final composition to 2%, 4%, or 6% daptomycin, and mixed for an additional minute. The $CaSO_4$-K_2SO_4-daptomycin mixture was cast in a silicone elastomer pellet mold (4.8 mm diameter x 3.3 mm height) and allowed to cure for 24 hours. Average pellet weight after 24 hours was approximately 102.3 mg for the 2% pellets, 110.6 mg for the 4% pellets, and 89.5 mg for the 6% pellets. Elution characteristics were determined for each group of pellets. For each group, three elution samples consisting of 8 pellets each were tested. Each sample consisted of 8 pellets in 20 ml of phosphate buffered saline (PBS) kept at 37°C for the duration of the test. Aliquots of the eluent (1 ml) were collected for each sample on days 1, 2, 5, 7, 10, 14, 21, 28, and 35. The pellets were dried at 40°C for one hour and their weights were recorded at each time interval. The entire amount of PBS was replaced at each time interval, and the pellets were returned to the elution vessels at 37°C. Aliquots were frozen at - 8°C until the elution was completed. All aliquots were tested using HPLC (Varian ProStar, C8 column, 0.45% $NH_4H_2PO_4$ mobile phase) for daptomycin concentration. Daptomycin concentrations were extrapolated from a concentration curve.

Results/Discussion: The 6% daptomycin pellets were completely dissolved by day 21, the 4% pellets at day 28, and the 2% daptomycin pellets at day 35. This is consistent with the observations that daptomycin inhibits the setting of calcium sulfate dihydrate from hemihydrate. The greater amount of potassium sulfate solution added to the 6% daptomycin pellet was necessary to produce a pellet, and may affect the elution profile. Current experiments using 4% daptomycin pellets with 0.31g K_2SO_4 solution per gram of powder are ongoing. Figure 1 presents a sample elution profile of the three groups from each set averaged. Figure 2 presents an average pellet weight loss for each of the three concentrations.

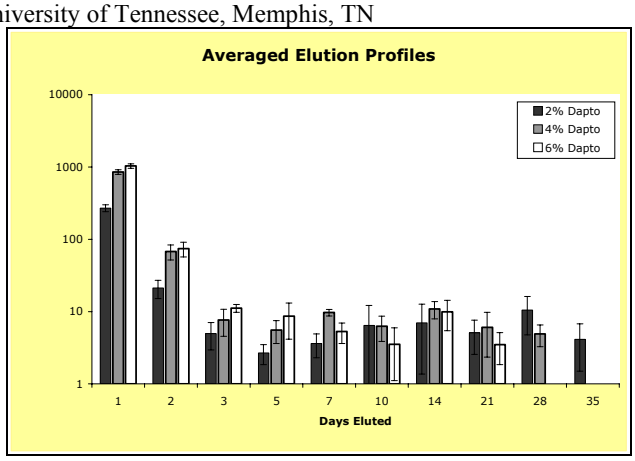

Figure 1. Elution profiles, adjusted for initial pellet weight. Note logarithmic scale on the Y axis.

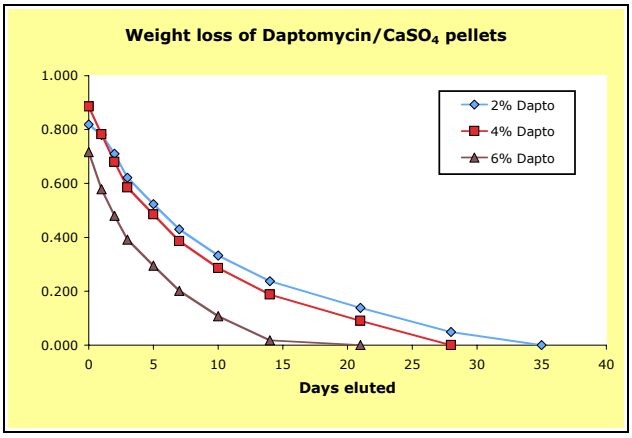

Figure 2. Weight loss of pellets during experiment

Conclusions: The greater concentrations of daptomycin appear to increase dissolution rate. Greater concentrations of daptomycin increase elution rates before day 7, however, after day 7, the elution of daptomycin from the pellet was observed to be similar for all three groups of pellets. Ongoing experiments will assess the affect of potassium sulfate solution amount on the elution profile and weight loss.

This study demonstrates calcium sulfate can be used to provide a suitable pellet for the local delivery of daptomycin with a degradable material.

References:

[1] Moseley JP, et. al., ORS Trans., 2004, p.1063
[2] Webb N, et. al., Musculoskeletal Infection Society 16th Annual Meeting, August 11-12, 2006, Abs. #23

Acknowledgments:

- Dr. Judith Steenbergen, Ph.D. of Cubist Pharmaceuticals for technical support
- Cubist Pharmaceuticals for material donations and research funding
- Wright Medical Technology for material donations.