Evaluation of Drug Release Profile for a Combination of Antibiotics in a Demineralized Bone Matrix Carrier

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Introduction: Treatment of chronic or acute osteomyelitis is difficult, time consuming, and expensive[1]. The current standard of care involves prophylactic intravenous antibiotics, which may be inadequate if the infection site is poorly vascularized or necrotic. By directly releasing antibiotics locally to bone over time, this disadvantage is averted. Delivering antibiotics with a hydrophobic osteoinductive carrier constructed from demineralized bone matrix (DBM) may be a useful treatment for bone infection. Minocycline and rifampin are a patented antibiotic combination shown to be clinically effective against common orthopedic infections, such as S. Aureus[2,3]. The objective of this study is to evaluate release characteristics of this antibiotic combination from a lecithin-based DBM putty to determine if the release pattern is clinically relevant as a prophylactic agent against infection.

Methods: InterGro™ DBM Putty (EBI, L.P., Parsippany, NJ) was manually combined with minocycline (M) and/or rifampin (R) to form composite beads (1 cc). Group 1: DBM control, Group 2: DBM + M/R (0.625% w/w M, 0.625% w/w R), Group 3: DBM + M/R (0.125% M, 0.125% R), Group 4: DBM + M (0.125% M) and Group 5: DBM + R (0.125% R). Samples from each group (n=5) were submersed in 30 cc of phosphate buffered saline (PBS) at 25 ºC with mild agitation. PBS fluid was changed daily for eight days or until complete resorption of the lecithin carrier. Extracted solutions were measured with a UV spectrophotometer (Genesys Thermo Electron; Pittsford, NY) at 350 nm and 470 nm. Standard curves were created for both M and R to derive the cumulative % of release for each group and the results were plotted (error bars are standard error of the mean).

Results: Release profiles for M and R from Groups 1 thru 5 are presented in Figures 1 and 2, respectively. Approximately 40% of M released by Day 7. R released slightly faster, passing 40% release on Day 6. Antibiotic release up to that point progressed in a fairly constant fashion starting from Day 2 before bursting during Days 7 and 8. By Day 8, the DBM carrier had completely resorbed in all samples, thereby releasing 100% of the antibiotics. The shape of the release profiles were independent of the amount of M or R contained in Groups 2 and 3, though M and R both released faster on their own when not in the presence of the other drug.

Discussion: Prophylactic antibiotics are beneficial for preventing infection arising from accidental operative inoculation. The release profile exhibited in this study may have clinical relevance for orthopedic applications because its time scale matches the critical period of intervention for postoperative infections. Additionally, M and R are known to be extremely effective against the major bacterial species that cause osteomyelitis. The minimum inhibitory concentrations for R against S. Aureus and P. Aeruginosa, for example, as determined by agar diffusion assays, were 0.015 µg/mL and 64 µg/mL respectively[3]. M is effective against E. Coli and K. Enterobacter at 25 µg/mL and many other gram-positive and gram-negative organisms as well[4]. Both M and R are broad spectrum and are not antagonistic in combination, though they can be occasionally synergistic[5]. They are also not known to exhibit immunogenicity or allergic reactions in patients.

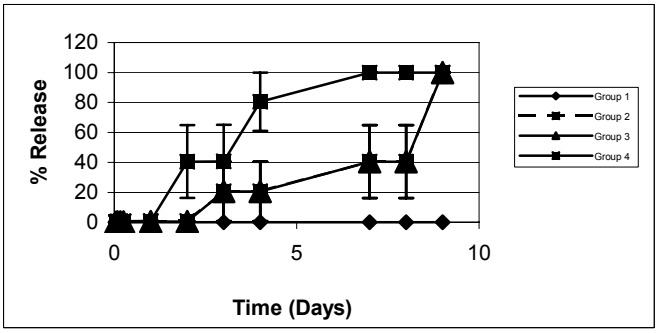

Figure 1. Minocycline release profile from DBM

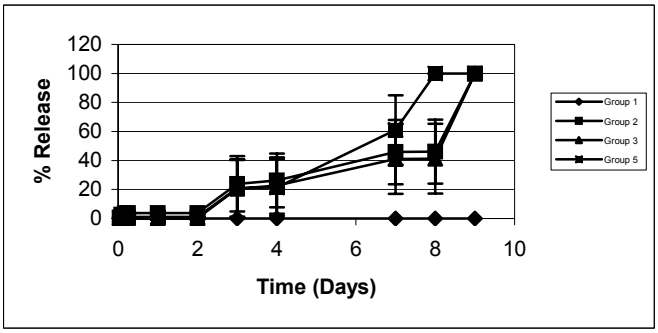

Figure 2. Rifampin release profile from DBM

Conclusions: Given the timing of release exhibited by M and R with respect to the known timescale associated with postoperative orthopedic infections and the utility of the M and R combination, it can be concluded that M/R release from DBM has clinical potential and is worthy of further evaluation. With this assembly, it may be possible to achieve optimal protection against infection without compromising bone healing and regeneration.

References:

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