Biodistribution and Biodegradation of a Novel Peptide Amphiphile Implant in a Rat Spinal Fusion Model

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Introduction: Current methods of spinal fusion exhibit significant rates of pseudoarthrosis (failed fusion) that are associated with significant morbidity. The addition of bone morphogenic protein (rhBMP-2) enhances spine fusion rates, but there are well established side effects. To mitigate these risks, implants consisting of peptide amphiphile (PA) nanofibers containing a BMP-2 binding epitope have been developed that potentiate BMP-2 activity, reducing the dose of rhBMP-2 required to elicit successful fusion in a pre-clinical model. PAs are composed of lipids and amino acids that self-assemble into cylindrical fibers in a structure analogous to an extracellular matrix of collagen fibrils. In moving towards clinical trials, a thorough understanding of the biodegradation rate, tissue distribution, and clearance are needed. This study sought to quantify the degradation rate and evaluate distribution of the material in the pre-clinical setting of posterolateral spinal fusion in rats.

Methods: The study was approved by the Northwestern University Institutional Animal Care and Use Committee. Twenty-three female Sprague-Dawley rats, aged 12-16 weeks, underwent L4-L5 posterolateral fusion with bilateral placement of implants composed of porous collagen microparticles, gadolinium (Gd)-labeled nanofibers containing a BMP-2 binding epitope, and low dose rhBMP-2 (Figure 1A, 1B). Magnetic resonance imaging (MRI) was performed post-operatively at increasing time points out to 13 weeks, followed by tissue harvest (spine, blood, kidney, liver, lung, and spleen) for inductively coupled plasmonic mass spectroscopy (ICP-MS)-based quantification of Gd (N=3/timepoint). Three animals underwent longitudinal imaging until the 13-week study endpoint. The Gd detected in harvested tissues at each timepoint was calculated as a percent of the Gd in the original implant. Additionally, two spines were harvested at 10 weeks and one at 13 weeks for manual palpation-based fusion scoring and high-resolution microcomputed tomographic imaging (μCT) for visualization of the fusion mass.

Results: Gd signal decreased gradually in the spine fusion site, from 71% of the presurgical implant concentration at 4 hours post-operatively, to 19% by 13 weeks (Figure 1C). Among the peripheral organs, the highest accumulation of Gd was 3% of the presurgical implant concentration in the liver at 4 weeks, which declined to 1.4% at 13 weeks. Gd accumulation in the kidney peaked at 4 weeks (1.6%), declining to 0.11% at 13 weeks. For the duration of the study, Gd remained below 0.05% in the spleen, 0.03% in the lung, and 0.01% in the blood (Figure 1E). MRI corroborates these findings with a notable loss of Gd signal at 8-weeks and complete loss of signal at the 13-week timepoint (Figure 1D). The spines evaluated by manual palpation and μCT all showed bilateral fusion.

Discussion: This study aimed to understand the degradation and biodistribution of a PA implant in a rat spinal fusion model. ICP-MS showed that the Gd-tagged implant remained localized in the surgical site, with limited accumulation in peripheral organs during degradation. By 13 weeks, only 19.5% of the initial Gd was detected locally in the spine. This was corroborated by lack of signal on MRI, supporting robust clearance after degradation. We posit that the Gd nadir was 19.5% due to Gd-PA nanofiber incorporation into the bony fusion mass. The Gd concentration in many of the clearance organs (blood, spleen, and lung) remained less than 0.1% for the duration of the study. This work shows timely degradation of this amino acid-based spinal fusion implant, with limited accumulation in peripheral organs and no change in spinal fusion efficacy due to the Gd label.

Significance/Clinical Relevance: This study provides new insight regarding the degradation and subsequent biodistribution of a peptide amphiphile-based implant material designed for bone regeneration. Our findings indicate that the PA implant remains well localized after placement at the L4-L5 transverse processes, without significant peripheral accumulation.

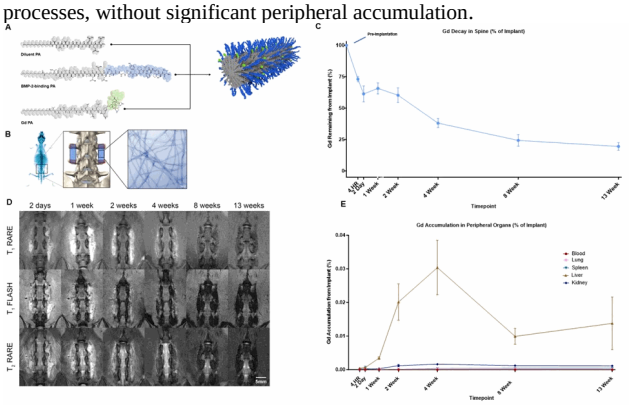

Figure 1A. Structure of the PA-based scaffold used in the study **B**. Rat spinal fusion model **C**. Gd decay over time in the spine, relative to the original concentration concentration of the scaffold pre-implantation **D.** MRI showing decreasing Gd signal from 2 days to 13 weeks **E.** Gd accumulation in peripheral organs across different timepoints.