Bioresorbable microsphere tracking in vivo via encapsulated iron oxide nanoparticles

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Purpose: For decades, injectable microspheres have provided controlled, sustained drug delivery over time to a local environment. Several studies have measured the in vivo effects of released drugs on the tissue surrounding the injection site, but there are few studies illustrating the location of microspheres in these tissues. Confirming the location of the delivery platform and its longevity in vivo would give insight into the vehicle biodistribution and provide insights into the length of time before the particle is cleared by the tissue. Visual imaging or tracking of these injected particles may strengthen results reported by many investigators in the field as to the efficacy of local drug delivery, because the presence of the vehicle over time could be confirmed.

Introduction: Superparamagnetic Iron Oxide (SPIO) particles are used as a gold standard for magnetic resonance imaging (MRI) contrast. SPIO particles are composed of iron oxide crystals that contain magnetic domains in random orientations that become oriented with an applied external magnetic field and are detectable via MRI. Encapsulated SPIO particles in nanospheres and other synthetic polymer constructs, have shown promise to target drug delivery and image target tissue. The goal of our study was to exploit the MRI contrast of aqueous SPIO particles by encapsulating them into the core of bioresorbable polymer microspheres to track microsphere location upon injection into tissue.

Methods: Poly(D,L-lactide-co-glycolide) 85:15 Sigma-Aldrich (St. Louis, MO) was fabricated into microspheres as described previously via water-in-oil-in-water (W/O/W) double emulsion technique with some modifications. The organic phase consisted of 5% (w/v) PLG in ethyl acetate, while the aqueous phase consisted of 0.1 mL of FeREX (Worcester, MA). The aqueous and organic phases were mixed and sonicated briefly. The resulting first emulsion was added immediately into a 1% poly(vinyl alcohol) in ethyl acetate solution and mechanically vortexed to form the second emulsion. The resulting solution was added to a hardening solution and stirred for 4 h to allow for organic solvent evaporation. The resulting microspheres were collected, washed, resuspended, and purified via coarse filtration.

Results: Scanning Electron Microscopy prior to injection (Figure 1a) demonstrated that FeREX-encapsulated microspheres had a similar morphology to typical PLG microspheres used in drug delivery. To determine the amount of SPIO particles incorporated per injection, the total iron concentration (0.058±0.013ug/mg of PLG microspheres) was measured with a Ferrozine-based colorimetric assay after dissociation of microspheres via hydrolysis with 0.1M NaOH. To determine if our microspheres were visible on MRI, 2.5mg and 5mg samples of dry microspheres were suspended in saline and injected into an explanted muscle (Figure 1b). Both injection sites were visible upon same day MRI. 10mg ethylene oxide gas sterilized microspheres injected into the myocardium of an anesthetized pig heart were also detectable via MRI at two injection locations minutes after administration (Figure 1c).

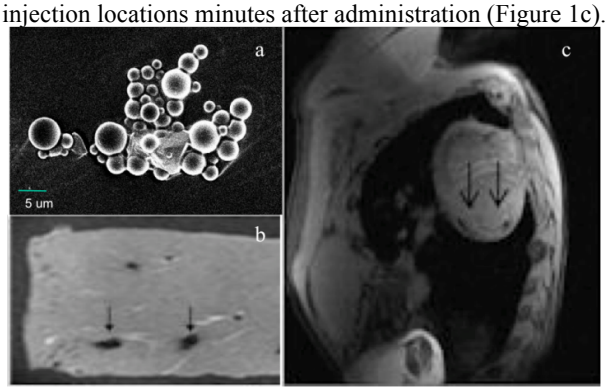

Figure 1: (a) PLG SPIO encapsulated microspheres imaged with Scanning Electron Microscopy, (b) In vitro injections of SPIO encapsulated PLG microspheres imaged with MRI, (c) In vivo myocardial detection of injected SPIO encapsulated PLG microspheres

Conclusions: Encapsulation of SPIO in biodegradable polymer microspheres allows for imaging of microsphere location in vivo. Because the particles are encapsulated in the same bioresorbable delivery vehicle used for drug administration, monitoring SPIO presence may be useful to demonstrate degradation kinetics of polymer microspheres in vivo. This tracking system may also be useful to detect whether locally injected delivery vehicles remain at the injection site or travel throughout the tissue. Future work includes temporal imaging of SPIO injected tissues to determine microsphere resorption rate, and determining the optimal microsphere load necessary to maintain a detectable MRI signal.