Poly(lactide-co-glycolide) Microspheres for MRI-Monitored Delivery of Sorafenib Locally to Liver Tumors

Jeane Chen\textsuperscript{1,2}, Sarah B. White\textsuperscript{2,3}, Kathleen R. Harris\textsuperscript{2}, Jonathan WT. Yap\textsuperscript{4}, Robert J. Lewandowski\textsuperscript{2}, Lonnie D. Shea\textsuperscript{1}, Andrew C. Larson\textsuperscript{2,4}

\textsuperscript{1}Chemical and Biological Engineering, Northwestern University, Evanston, IL, USA
\textsuperscript{2}Radiology, Northwestern University, Chicago, IL, USA
\textsuperscript{3}Vascular Interventional Radiology, Medical College of Wisconsin, Milwaukee, WI, USA
\textsuperscript{4}Biomedical Engineering, Northwestern University, Evanston, IL, USA

Jeanechen2015@u.northwestern.edu

Abstract Summary
Sorafenib is a multikinase inhibitor chemotherapeutic effective in treating hepatocellular carcinoma (HCC). However, the oral tablet formulation is associated with severe side effects, resulting in poor patient tolerance. To address such issues, this study works to re-formulate sorafenib as part of an embolic microsphere platform to enable local delivery of sorafenib to the liver tumors via catheters placed in tumor-feeding arteries.

Introduction
Hepatocellular carcinoma (HCC) is the most common form of primary liver cancer and the third cause of cancer-related death [1, 2]. In 2007, sorafenib was approved by the FDA for the treatment of HCC, and it has quickly become the standard of care for patients with advanced stage disease [3, 4]. It is formulated as an oral tablet and thus, the systemic administration results in severe side effects. Systemic administration of chemotherapy has typically been avoided for HCC via transarterial chemoembolization (TACE), where doctors place a catheter in the femoral artery and use imaging to guide the catheter specifically into tumor-feeding arteries. Once the catheter is placed, embolic beads loaded with chemotherapy can be delivered locally to the tumor and also embolize tumor feeding arteries to starve the tumor. As sorafenib has not been formulated for such a procedure due to its hydrophobicity, the purpose of this project is to re-formulate sorafenib for TACE procedures using poly(lactide-co-glycolide) (PLG) microspheres. Additionally, we have co-encapsulated iron oxide nanoparticles into our PLG microspheres to allow for magnetic resonance imaging (MRI) monitoring of microsphere delivery to the liver tumors.

Experimental Methods
Microspheres were fabricated via a double emulsion/solvent evaporation method. Specifically, PLG was dissolved in dichloromethane and added to sorafenib in DMSO. The water phase consisted of a suspension containing iron oxide. The oil phase and water phase were combined and homogenized for 30 seconds before adding a solution containing polyvinyl alcohol. Afterwards, the solution was homogenized again for 2 min. and poured into a stirred beaker containing a 0.5% polyvinyl alcohol solution. The microspheres were stirred in the beaker at least three hours for solvent evaporation before they were collected, washed and lyophilized.

The microspheres were then characterized for size, loading, and release. Additionally, they were also characterized for MRI properties, where the microspheres were embedded in agar gels and imaged \textit{in vitro} under a 7 Tesla MRI scanner (Bruker Clinscan). After these initial characterization studies, five rabbits were implanted with VX2 tumor fragments in the liver under ultrasound guidance. PLG sorafenib iron oxide microsphere TACE was performed on these rabbits when the tumors were over 1 cm in diameter and after catheter placement in the tumor feeding arteries, 50 mg of microspheres were infused. Pre- and post-infusion 7T MRI was performed to allow evaluation of microsphere biodistribution. After the post-procedural MRI, the rabbits were sacrificed and the tumor, liver, stomach and gallbladder were harvested. Prussian blue staining for the iron oxide within the microspheres was performed on these tissue samples for purposes of biodistribution analyses to confirm what was observed under MRI imaging.

Results and Discussion
The fabricated microspheres had diameters ranging from 2.5 µm to 63 µm, with an average of 13 µm. Loading and release studies indicated that the weight percentage of sorafenib and iron oxide in the microspheres were 8.8% and 0.89%, respectively and that 21% and 28% of the loaded sorafenib and iron oxide released in 72 hours, respectively.
Figure 1: *In vitro* sorafenib and iron oxide release from the fabricated poly(lactide-co-glycolide) microspheres at 37°C.

MRI characterization studies qualitatively showed signal decreases due to the negative contrast effects of the iron oxide within the microspheres and quantitatively showed a decrease in $T_2$ values from 111.1 to 33.3 ms when increasing microsphere concentrations from 0 to 5 mg/mL. X-ray Digital Subtraction Angiography performed during the procedure indicated poor lack of perfusion within the tumor-feeding vessels after microsphere delivery, thus confirming proper vessel embolization. Comparisons of pre and post-procedural MRI allowed indicated signal loss at the tumor after microsphere delivery. This observation was confirmed by Prussian blue staining, which showed primary microsphere deposition in the tumor periphery. Prussian blue staining also showed limited non-targeted delivery to surrounding organs such as the stomach and gallbladder. No microspheres were seen to have passed through an arterial venous shunt and thus to have gone to the lungs.

Conclusion

Sorafenib can be formulated for local delivery using a PLG microsphere platform to improve patient outcomes. The developed PLG microspheres demonstrated proof of concept in a rabbit VX2 model and allowed for MRI monitoring of the delivery.

References


Acknowledgements

The authors would like to acknowledge Jodi Nicolai for her assistance with maintaining the rabbit VX2 cell line.