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Preparation of Radiopaque Drug-Eluting Beads for Transcatheter Chemoembolization

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Abstract

Purpose—Transarterial chemoembolization (TACE) with drug eluting microspheres improves drug delivery to tumors and reduces systemic exposure compared to conventional TACE (cTACE), but lacks imaging feedback. The *objective* of this study was to develop a simple method to produce radiopaque drug eluting microspheres that could be incorporated into the current clinical TACE workflow and evaluate their performance *in vitro* and *in vivo*.

Methods—A Lipiodol[®] and ethanol solution was added to a lyophilized 100–300 μ m bead prior to loading with doxorubicin. These radiopaque drug eluting beads (DEBs, Biocompatibles UK Ltd) were evaluated *in vitro* for X-ray attenuation, composition, size, drug loading/elution, and correlation between attenuation and doxorubicin concentration. *In vivo* conspicuity was evaluated in a VX2 tumor model.

Results—Lipiodol was loaded into lyophilized beads using two glass syringes and a 3-way stopcock. Maximum bead attenuation was achieved within 30 minutes. X-ray attenuation of radiopaque beads increased linearly (21 – 867 HU) with the amount of beads (0.4 – 12.5 vol%; R^2

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Conflicts of interest:

None of the other authors have identified a conflict of interest.

= 0.9989). Doxorubicin loading efficiency and total amount eluted was similar to DC Bead®; however, the elution rate was slower for radiopaque DEBs ($P < 0.05$). Importantly, doxorubicin concentration linearly correlated with X-ray attenuation of radiopaque DEBs ($R^2 = 0.99$). Radiopaque DEBs were seen in tumor feeding arteries following administration by fluoroscopy, CT, microCT, and their location was confirmed by histology.

Conclusion—A simple and rapid method to produce radiopaque DEBs was developed. These radiopaque DEBs provided sufficient conspicuity to be visualized with X-ray imaging techniques.

Keywords

Drug-eluting bead; Lipiodol; DEB-TACE

Introduction

Transcatheter arterial chemoembolization (TACE) is a treatment option for unresectable hypervascular hepatic tumors and is performed with either drug eluting microspheres or a Lipiodol and chemotherapy emulsion, known as conventional TACE (cTACE) [1]. TACE with drug eluting microspheres may provide improved tolerability and decreased systemic drug exposure compared to cTACE [2–4] perhaps due to a slower and more sustained drug elution from the microsphere [5, 6]. A shortcoming of TACE with drug eluting microspheres is the lack of intra- and post-procedural imaging feedback that is otherwise provided by Lipiodol in cTACE.

When performing TACE with drug eluting microspheres, soluble iodinated contrast is mixed with microspheres to form a durable suspension and to visualize changes in vascular flow in order to monitor and limit reflux during the procedure. Iodinated contrast is necessary for safety but does not indicate eventual location of microspheres or drug [7, 8]. Several investigators have developed microspherical embolics which can be visualized by X-ray (e.g., fluoroscopy and CT) [7, 9], magnetic resonance imaging [10–15], or both [16]. However, these image-able embolics have yet to be translated into clinical practice.

In order for image-able drug eluting microspheres to provide clinical utility, they should satisfy several practical requirements. First, image-able microspheres must be adequately visible by X-ray. Second, image-able microspheres location should spatially correlate with drug. Third, the additional image-ability should not adversely affect drug loading/elution profiles, handling and embolic performance of microspheres. Furthermore, preparation of image-able drug eluting microspheres should be convenient, reproducible, rapid (< 1 hr) and sufficiently standardized for incorporation into the current workflow. Using these guiding practical requirements, the *objective* of this study was to develop a simple method to produce radiopaque drug eluting microspheres and to evaluate their performance *in vitro* and *in vivo*.

Materials and Methods

Materials

Acrylamido Polyvinyl Alcohol–co–Acrylamido-2-methylpropane sulfonate drug eluting beads (DEBs) were lyophilized in the presence of an excipient (referred to as “dry bead” in this manuscript), by Biocompatibles UK Ltd (Farnham, UK). The excipient (Mannitol) serves as a matrix forming additive, creating an amorphous phase in the spaces vacated by the water during lyophilization. Upon hydration, the excipient rapidly dissolves as the hydrogel reforms the swollen, compressible beads (~2 mL beads/vial). The dry bead is supplied in sterile, vacuum-sealed vials and allows for incorporation of an X-ray contrast agent (Lipiodol®, Guerbet, Paris, France) and doxorubicin (Bedford Laboratories, Bedford, OH) within the bead structure. Note that standard commercial DC Bead® (available in a hydrated form) is not capable of Lipiodol loading and other commercially available dry beads were not investigated in our study. Additional materials needed to make radiopaque DEBs include ethanol, 18G or larger needles (minimum 1.5 inch length), two glass syringes (10 and 20 mL 1000 Series Gastight Syringe, Hamilton, Reno, NV), sterile water, and a metal 3-way stopcock (Merit Medical, South Jordan, UT).

Preparation of Radiopaque DEBs

A wide range of conditions (Supplemental materials, Table 1) were tested to develop a simple procedure that would yield optimal radiopaque DEBs. Overall, the procedure consisted of the following steps: 1) Load Lipiodol into dry bead to make X-ray visible (i.e. radiopaque) beads, 2) Remove excess Lipiodol solution not contained within the radiopaque beads, 3) Load doxorubicin into radiopaque beads to make radiopaque DEBs and 4) Prepare a suspension of radiopaque DEBs for embolization. A detailed description of the steps is outlined in supplemental materials and shown in Schematic 1.

In Vitro Characterization

The full description of in vitro characterization may be found in supplemental information. Briefly, bead size and appearance was evaluated with light microscopy. Iodine content was determined with elemental analysis (MEDAC Ltd, UK). Doxorubicin and Lipiodol elution was measured in a Caleva USP2 dissolution bath monitored by a Cary 50 UV-visible spectrophotometer (Varian, Australia) and elemental analysis, respectively. Attenuation of homogeneous radiopaque bead phantoms was measured with 256 Slice CT (Philips Healthcare, Cleveland, OH) to relate radiopaque bead concentration to attenuation. MicroCT imaging and analysis of phantoms was performed with a SkyScan 1172 high-resolution micro-CT (Skyscan, Konitch, BE) to evaluate the radiopacity of each individual bead, as well as, intra-bead distribution of iodine/Lipiodol.

In Vivo and Ex Vivo Evaluation of Radiopaque DEB

All animal studies were conducted under an animal use protocol approved by the Institutional Animal Care and Use Committee. Additional details may be found in supplemental information. Briefly, Vx2 tumors were propagated as described by Ranjan et al. [17]. Femoral access and catheter placement in the proper hepatic artery was conducted

with a 3F sheath (Cook, Bloomington, IN) and a 2.4 Fr microcatheter (Terumo, Somerset, NJ) under fluoroscopic guidance. Routine fluoroscopy was conducted using the 9900 Elite Digital Mobile Super C-arm (GE Healthcare/OEC Medical Systems, Inc., Waukesha, WI). Radiopaque beads (100–300 μ m) were mixed 1:20 in saline/Isovue 300 contrast and delivered under fluoroscopic monitoring at a rate of approximately 1 ml/min (including agitation time) until stasis in the main hepatic artery was determined angiographically.

Harvested liver tissue was imaged on 256 slice CT (Philips Healthcare). To resolve individual radiopaque beads within tissue an Inveon microCT (Siemens Preclinical Solutions, Knoxville, TN) was used.

Doxorubicin Concentration versus Attenuation

Ultra high performance liquid chromatography (UHPLC) was used to quantify the amount of doxorubicin (see supplemental information for details) in radiopaque DEB phantoms and correlate with the attenuation values from CT. Radiopaque DEBs (75mg doxorubicin/2 ml bead volume) containing agarose phantoms were made and imaged (percent bead volume ranging from 0–12.5% in 1 mL phantoms). Following imaging, doxorubicin extraction from the phantoms was performed by adding 10 mL of saturated KCl/ethanol (1:1 v/v) at room temperature with mixing, centrifugation, removal of the supernatant and replenishing of the KCl/ethanol solution until the supernatant was cleared. By using phantom volume measured on imaging, doxorubicin content (mg) was converted into doxorubicin concentration (mg/ml).

Histological Evaluation

Histological assessment of the tumor tissue was conducted to confirm the radiopaque DEB location relative to the tumor as seen on radiologic imaging. Hematoxylin and eosin staining (5 μ m sections) was performed on 10 equidistantly spaced regions throughout a tumor volume. Imaging was performed on a ScanScope CS (Aperio, Vista, CA).

Statistics

All analyses were performed using GraphPad Prism 5.0 (GraphPad Software Inc.). Groups were compared for differences using a Student's T-test or one-way ANOVA with a Newman Keuls post hoc test. R^2 values are reported from linear regression (following a logarithmic transform of both the dependent and independent variables) of attenuation versus radiopaque bead concentration and doxorubicin amount versus change in attenuation of radiopaque DEBs (background attenuation of phantom material was subtracted). All p-values were two-sided, and a p-value less than 0.05 indicated statistical significance. Values are reported as mean $\pm$ SEM unless otherwise indicated.

Results

Lipiodol Loading, Washing and Attenuation of Radiopaque Beads

Dry beads (100–300 μ m) were made radiopaque by incubating them with a Lipiodol and ethanol mixture and then washing to remove excess Lipiodol. The optimal procedure was selected by evaluating the beads' radiopacity and lack of free Lipiodol within the

suspension. The conditions (Table 1, supplemental materials) tested included Lipiodol to bead ratio, Lipiodol to ethanol ratio, loading time, washing method, and the number of washing steps. A ratio of 0.4 mL of Lipiodol per 1 mL of beads was needed to achieve maximal attenuation; this attenuation did not substantially increase with a further increase in Lipiodol to bead ratio. An ethanol and Lipiodol mixture of 15:1 was used because it was found to facilitate removal of excess, unloaded Lipiodol, as compared to beads loaded with Lipiodol alone. Sequential washes with water proved to remove the excess Lipiodol mixture efficiently. This methodology was optimized using the 100–300 μm bead size but was also successful with the smaller 70–150 μm bead size.

Radiopaque bead attenuation as a function of Lipiodol loading time was evaluated up to 24hr; Figure 1A shows the results for up to 2hr of loading. These results demonstrated that 30min of incubation with a Lipiodol and ethanol mixture yielded the highest attenuation (229 ± 7.96 HU in radiopaque bead phantoms, 3 vol%) and no increased attenuation was observed with incubation periods beyond 30min (p-value > 0.05, ANOVA). Residual free Lipiodol in 5 successive washes was assessed as shown in Figure 1B. Each successive wash (10 mL sterile water) decreased the attenuation of the supernatant, and after 5 washes, the attenuation was not statistically different from deionized water ($P > 0.05$). Also, washing radiopaque beads did not remove substantial amounts of Lipiodol from the radiopaque beads (Lipiodol level in wet beads was 47–49 wt% throughout all washes). This washing method also removed excess ethanol resulting in 15 ppm (17,000ppm in the first wash). A clinical CT scanner was used to determine attenuation as a function of percent bead volume (vol%) in bead containing agarose gel phantoms, as shown in Figure 1C&D. Attenuation ($21 - 867$ HU) increased linearly with bead concentration ($0.4 - 12.5\text{vol\%}$; $R^2 = 0.9989$, p-value < 0.05).

Doxorubicin Loaded Radiopaque DEBs

In order to make radiopaque DEBs, radiopaque beads were loaded with doxorubicin using a procedure similar to the one used to load DC Bead. Two hours were sufficient to load >99% doxorubicin (37.5mg/ml bead), as shown in Figure 2A. Doxorubicin elution over time is shown in Figure 2B and the half-life was significantly slower from radiopaque DEBs than standard DEBs (see Table 2, supplemental information; 36.2 ± 1.4 versus 12.5 ± 0.4 min; p-value < 0.05, Tukey's). Dry DEBs had a similar doxorubicin elution as DC bead by the similarity and difference test (f_1 and f_2), while radiopaque DEB's elution was significantly different from DC bead. Importantly, a similar total amount of drug is eluted after 6 hr. Bead appearance following Lipiodol or doxorubicin loading is shown in supplemental information (Supplemental Figure 1A, B and C). Hydrated beads appear spherical and blue. Radiopaque beads have a white/opaque appearance after Lipiodol loading and doxorubicin imparts a red color that is slightly less intense than doxorubicin loaded DC Bead. The radiopaque beads often appeared with a marbled internal structure, indicating inhomogeneity of Lipiodol in the bead, with coexistence of Lipiodol-rich and water-rich phases.

Attenuation of Radiopaque Beads and Radiopaque DEBs

The inhomogeneous internal structure of the radiopaque beads noted with light microscopy was also clearly observed in the micro-CT images (Figure 3). Following doxorubicin loading, the marbled appearance was reduced and the packed bead volume decreased. These

changes may be due to displacement of water from the radiopaque DEB following doxorubicin loading, leading to a more compacted bead structure as demonstrated in Figure 3. The Lipiodol content in wet radiopaque beads (before doxorubicin loading) was 42.3 ± 2.9 wt% and wet radiopaque DEBs (after doxorubicin loading) was 58.1 ± 0.6 wt%. Consistent with this increased Lipiodol content, individual radiopaque DEBs ($17,921 \pm 909.38$ HU) had a greater attenuation than radiopaque beads ($13,092 \pm 1411.0$ HU) (p -value < 0.001 , t -test). Similar to pure Lipiodol (38 wt%), the iodine content was 37.2 ± 0.1 wt% in radiopaque beads and 36.2 ± 0.1 wt% in radiopaque DEBs when the beads were dehydrated.

The doxorubicin loading efficiency into radiopaque beads was 99.98% when using 75mg of doxorubicin/2ml beads. The attenuation of radiopaque DEBs may be used to predict the concentration of doxorubicin in a phantom as shown in Figure 4. The doxorubicin concentration was linearly correlated with attenuation ($R^2 = 0.99$, $p < 0.0001$).

Bead Size Analysis and Handle-ability

The bead size changed after Lipiodol and doxorubicin loading, as shown in Table 2, supplemental information. DC Bead size (160 ± 54 μ m) decreased after doxorubicin loading (117 ± 43 μ m). Radiopaque bead size (164 ± 41 μ m) was slightly smaller than dry bead (192 ± 58 μ m). Similarly, radiopaque beads decreased in size with doxorubicin loading (radiopaque DEB = 149 ± 47 μ m).

Bead handle-ability was assessed by monitoring the durability of bead suspension and assessing transcatheter delivery. Radiopaque DEBs suspended in sterile water or saline tended to form small clumps and fall out of suspension rapidly (3.2 ± 0.45 sec), requiring frequent mixing during catheter delivery. A uniform and durable suspension of radiopaque DEBs without bead clumping was obtained by using a contrast to water ratio of 2:1. This ratio yielded a durable suspension that only required mixing every 20–30 sec (remained in suspension 21 ± 2.3 sec in a 20 cc syringe with a bead dilution of 1:20, $n = 4-5$) and was practical for clinical use. This suspension was easily delivered through a microcatheter (2.4 Fr) with performance similar to doxorubicin loaded DC Bead.

In Vivo and Ex Vivo Characterization of Radiopaque Beads

The conspicuity of radiopaque beads was studied during transcatheter embolization in rabbit Vx2 intrahepatic tumors as shown in Figure 5. Individual radiopaque beads were not visible leaving the catheter tip with real-time fluoroscopy but their accumulation in hepatic arterial branches was visualized with fluoroscopic images post embolization. Following euthanasia and liver harvest, clinical CT demonstrated filling of the large feeding arteries surrounding the Vx2 tumor leading back to the proper hepatic artery, consistent with the fluoroscopy images. MicroCT is more sensitive than clinical CT with superior resolution (31 μ m versus 1 mm) capable of visualizing individual radiopaque beads. Similar to clinical CT, microCT images demonstrated radiopaque beads mainly surrounding the tumor periphery but also indicated the presence of infrequent beads within the tumor interior.

Histological examination of tumor sections confirmed presence of radiopaque beads in large arteries (> 100 μ m) in liver parenchyma near the tumor and some radiopaque beads within

the tumor (Figure 6). The presence of radiopaque beads (e.g., tumor interior or tumor periphery) found with clinical CT and microCT was confirmed by histology.

Discussion

The lack of imaging feedback during TACE with drug eluting microspheres remains a technical challenge. Several groups have reported on development of microspheres that can be imaged with fluoroscopy, CT and/or MRI [7–16] but these have not yet been translated to clinical practice. While availability of MR image-able [10–15] or multimodality image-able beads [16] are attractive options for research questions, we chose to focus on X-ray image-able microspheres because of the wide availability of fluoroscopy and cone beam CT in current clinical practice. In addition, while advanced MRI techniques such as four-dimensional transcatheter intra-arterial perfusion MRI are capable of providing real-time imaging feedback during TACE [18, 19], these imaging techniques are not widely available. A relatively simple and clinically viable method is provided herein for efficiently loading Lipiodol into a lyophilized version of DC Bead, yielding radiopaque DEBs.

As a requisite to clinical adoption, radiopaque drug eluting microspheres must perform similarly to currently available drug eluting microspheres and provide imaging feedback, without adversely affecting workflow. Since the method to produce radiopaque DEBs was rapid and used tools readily available to interventional radiologists or pharmacists, it has potential for incorporation into the current clinical workflow. In the future, it may be ideal to have a have pre-made, ready-to-use radiopaque drug eluting microspheres.

Overall, radiopaque DEBs perform similarly to doxorubicin loaded DC Bead. One difference with radiopaque DEBs is their increased density, compared to DC Bead, due to Lipiodol loading that resulted in more rapid settling (with a density similar to Lipiodol). Rapid settling is not a clinical concern when sufficient soluble contrast is used in the bead suspension. Doxorubicin loading and elution were relatively similar between radiopaque DEBs and DC Bead. The slightly slower drug elution rate of radiopaque DEBs may be due to slower counter ion migration through the Lipiodol phase into the DEB and/or slower migration of doxorubicin through the Lipiodol phase out of the DEB. Furthermore, that larger diameter of radiopaque DEBs compared to DC Bead may reduce the elution rate. Slower drug elution from a DEB is of unknown clinical significance but theoretically may increase the exposure duration of cancer cells to doxorubicin. The radiopaque beads reported herein are more radiopaque than other attempts modify PVA-based beads with covalent attachment of Iodine [20], highlighting the utility of Lipiodol as a contrast agent.

Imaging feedback provided by radiopaque drug eluting microspheres could be useful for two reasons. First, direct visualization of microspheres could help detect and/or limit non-target embolization. For example, during the Vx2 intrahepatic tumor embolization with radiopaque beads, non-target embolization of the gastric arteries was seen with fluoroscopy and confirmed at necropsy. In clinical practice, imaging feedback will depend on the interplay between sensitivity of the imaging modality used to detect the radiopaque DEBs (i.e., cone beam CT is more sensitive than fluoroscopy) and conspicuity of the radiopaque DEBs. Second, since DEB location reliably predicts drug location due to the controlled elution,

visualization of embolized radiopaque DEBs could serve as an imaging surrogate of drug delivery in embolized tissues [8, 21, 22]. This is supported by the fact that doxorubicin concentration is linearly correlated with attenuation of radiopaque DEBs *in vitro* ($R^2 = 0.99$, $p\text{-value} < 0.0001$). This is an important distinction from cTACE where Lipiodol attenuation did not correlate well with drug location *in vivo* ($R^2 = 0.0099$) [23]. Further *in vivo* studies are needed to demonstrate this relationship between radiopaque DEBs and bioavailable drug concentration in tissues.

Knowledge of drug delivery in embolized tissue could be used to determine completeness or adequacy of treatment and provide a sound rationale for additional treatment. For example, a region of tumor that does not contain radiopaque DEBs may be defined as an under-dosed region that requires further treatment; either with embolization of additional tumor feeding arteries or with thermal ablation (e.g., radiofrequency ablation). In this regard, knowledge of local drug delivery may provide complementary information for determining optimal embolization endpoints that would augment the current clinical angiographic techniques and potentially improve therapeutic outcomes. It has been shown that the location of residual soluble iodinated contrast seen on immediate post procedure CT following TACE with drug eluting microspheres may indicate the region(s) that were embolized [24] and this information may indicate when treatment may have been incomplete. Furthermore, marginal contrast retention [25] as well as tumor contrast retention, vascular contrast retention, and defects in contrast retention [25, 26] predict for imaging response. Although this is useful information confirming that embolization was performed from a tumor feeding artery, location of the iodinated contrast does not always indicate the eventual DEB or drug [7, 8] location or show regions of nontarget embolization. The persistent opacification of embolized vessels with radiopaque beads following soluble contrast washout, may provide additional value beyond soluble contrast retention.

This study has several limitations. The Vx2 animal model allows for the investigation of many interventional procedures but does not represent the complexity and size of human liver cancer. A correlation between doxorubicin concentration and attenuation was only demonstrated *in vitro* and should be confirmed by future *in vivo* studies. A portable C-arm, which is inferior to most imaging systems found in a clinical interventional suite, was used for the embolization procedure. The use of high-end clinical imaging systems including cone beam CT may characterize the true utility of radiopaque DEBs.

As TACE with drug eluting microspheres continues to evolve, improvements may be based on scientific and clinical evidence demonstrating the relative contribution of drug-related cytotoxic effects and embolic-related ischemic effects. In this regard, clinical knowledge of drug location and concentration in embolized tissues, using clinical tools such as navigation and cone beam CT, may help to educate this debate and provide a rational basis for optimal TACE and additional or combination therapies.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Lewandowski RJ, Geschwind JF, Liapi E, Salem R. Transcatheter intraarterial therapies: rationale and overview. *Radiology*. 2011; 259:641–57. [PubMed: 21602502]
2. Lammer J, Malagari K, Vogl T, et al. Prospective randomized study of doxorubicin-eluting-bead embolization in the treatment of hepatocellular carcinoma: results of the PRECISION V study. *Cardiovascular and interventional radiology*. 2010; 33:41–52. [PubMed: 19908093]
3. Poon RTP, Tso WK, Pang RWC, et al. A Phase I/II Trial of Chemoembolization for Hepatocellular Carcinoma Using a Novel Intra-Arterial Drug-Eluting Bead. *Clinical Gastroenterology and Hepatology*. 2007; 5:1100–8. [PubMed: 17627902]
4. Varela M, Real MI, Burrell M, et al. Chemoembolization of hepatocellular carcinoma with drug eluting beads: efficacy and doxorubicin pharmacokinetics. *J Hepatol*. 2007; 46:474–81. [PubMed: 17239480]
5. Lewis AL, Gonzalez MV, Lloyd AW, et al. DC bead: in vitro characterization of a drug-delivery device for transarterial chemoembolization. *Journal of vascular and interventional radiology: JVIR*. 2006; 17:335–42. [PubMed: 16517780]
6. Johnson PJ, Kalayci C, Dobbs N, et al. Pharmacokinetics and toxicity of intraarterial adriamycin for hepatocellular carcinoma: effect of coadministration of lipiodol. *Journal of hepatology*. 1991; 13:120–7. [PubMed: 1655867]
7. Sharma KV, Dreher MR, Tang Y, et al. Development of “imageable” beads for transcatheter embolotherapy. *Journal of vascular and interventional radiology: JVIR*. 2010; 21:865–76. [PubMed: 20494290]
8. Dreher MR, Sharma KV, Woods DL, et al. Radiopaque drug-eluting beads for transcatheter embolotherapy: experimental study of drug penetration and coverage in swine. *Journal of vascular and interventional radiology: JVIR*. 2012; 23:257–64 e4. [PubMed: 22178039]
9. Saralidze K, van Hooy-Corstjens CS, Koole LH, Knetsch ML. New acrylic microspheres for arterial embolization: combining radiopacity for precise localization with immobilized thrombin to trigger local blood coagulation. *Biomaterials*. 2007; 28:2457–64. [PubMed: 17257667]
10. Gupta T, Virmani S, Neidt TM, et al. MR tracking of iron-labeled glass radioembolization microspheres during transcatheter delivery to rabbit VX2 liver tumors: feasibility study. *Radiology*. 2008; 249:845–54. [PubMed: 18840788]
11. Lee KH, Liapi E, Vossen JA, et al. Distribution of iron oxide-containing Embosphere particles after transcatheter arterial embolization in an animal model of liver cancer: Evaluation with MR Imaging and implication for therapy. *Journal of vascular and interventional radiology: JVIR*. 2008; 19:1490–6. [PubMed: 18755602]
12. Namur J, Chapot R, Pelage JP, et al. MR imaging detection of superparamagnetic iron oxide loaded tris-acryl embolization microspheres. *Journal of vascular and interventional radiology: JVIR*. 2007; 18:1287–95. [PubMed: 17911520]

13. Wilson MW, Fidelman N, Weber OM, et al. Experimental renal artery embolization in a combined MR imaging/angiographic unit. *Journal of vascular and interventional radiology: JVIR*. 2003; 14:1169–75. [PubMed: 14514809]
14. Cilliers R, Song Y, Kohlmeir EK, Larson AC, Omary RA, Meade TJ. Modification of embolic-PVA particles with MR contrast agents. *Magnetic resonance in medicine: official journal of the Society of Magnetic Resonance in Medicine/Society o*. 2008; 59:898–902.
15. Seevinck PR, Seppenwoolde JH, Zwanenburg JJ, Nijssen JF, Bakker CJ. FID sampling superior to spin-echo sampling for T2*-based quantification of holmium-loaded microspheres: theory and experiment. *Magnetic resonance in medicine: official journal of the Society of Magnetic Resonance in Medicine/Society o*. 2008; 60:1466–76.
16. Bartling SH, Budjan J, Aviv H, et al. First multimodal embolization particles visible on x-ray/computed tomography and magnetic resonance imaging. *Invest Radiol*. 2011; 46:178–86. [PubMed: 21263332]
17. Ranjan A, Jacobs GC, Woods DL, et al. Image-guided drug delivery with magnetic resonance guided high intensity focused ultrasound and temperature sensitive liposomes in a rabbit Vx2 tumor model. *Journal of controlled release: official journal of the Controlled Release Society*. 2012; 158:487–94. [PubMed: 22210162]
18. Sato KT, Wang D, Lewandowski RJ, et al. Four-dimensional transcatheter intraarterial perfusion MRI monitoring of radiofrequency ablation of rabbit VX2 liver tumors. *Journal of magnetic resonance imaging: JMRI*. 2011; 34:563–9. [PubMed: 21761464]
19. Gaba RC, Wang D, Lewandowski RJ, et al. Four-dimensional transcatheter intraarterial perfusion MR imaging for monitoring chemoembolization of hepatocellular carcinoma: preliminary results. *Journal of vascular and interventional radiology: JVIR*. 2008; 19:1589–95. [PubMed: 18818097]
20. Negussie AH, Dreher MR, Johnson CG, et al. Synthesis and characterization of image-able polyvinyl alcohol microspheres for image-guided chemoembolization. *Journal of materials science Materials in medicine*. 2015; 26:198. [PubMed: 26105830]
21. Namur J, Citron SJ, Sellers MT, et al. Embolization of hepatocellular carcinoma with drug-eluting beads: doxorubicin tissue concentration and distribution in patient liver explants. *J Hepatol*. 2011; 55:1332–8. [PubMed: 21703190]
22. Namur J, Wassef M, Millot JM, Lewis AL, Manfait M, Laurent A. Drug-eluting beads for liver embolization: concentration of doxorubicin in tissue and in beads in a pig model. *Journal of vascular and interventional radiology: JVIR*. 2010; 21:259–67. [PubMed: 20123210]
23. Gaba RC, Baumgarten S, Omene BO, et al. Ethiodized Oil Uptake Does Not Predict Doxorubicin Drug Delivery after Chemoembolization in VX2 Liver Tumors. *Journal of vascular and interventional radiology: JVIR*. 2011
24. Golowa YS, Cynamon J, Reinus JF, et al. Value of noncontrast CT immediately after transarterial chemoembolization of hepatocellular carcinoma with drug-eluting beads. *Journal of vascular and interventional radiology: JVIR*. 2012; 23:1031–5. [PubMed: 22739645]
25. Suk Oh J, Jong Chun H, Gil Choi B, Gyu Lee H. Transarterial chemoembolization with drug-eluting beads in hepatocellular carcinoma: usefulness of contrast saturation features on cone-beam computed tomography imaging for predicting short-term tumor response. *Journal of vascular and interventional radiology: JVIR*. 2013; 24:483–9. [PubMed: 23452553]
26. Wang X, Erinjeri JP, Jia X, et al. Pattern of retained contrast on immediate postprocedure computed tomography (CT) after particle embolization of liver tumors predicts subsequent treatment response. *Cardiovascular and interventional radiology*. 2013; 36:1030–8. [PubMed: 23152036]

Figure 1.

Overview of Lipiodol loading, washing and attenuation of radiopaque beads. A) Attenuation of 3 vol% agarose bead phantoms made from radiopaque DEBs with variable Lipiodol incubation time periods. B) Attenuation of wash supernatant following Lipiodol loading. C) Representative CT images of radiopaque agarose bead phantoms at different bead concentration (vol%). D) Attenuation of the bead phantoms at different radiopaque bead concentration as shown in C fit with linear regression. Mean $\pm$ SEM; n=3–5.

Figure 2. Doxorubicin loading, doxorubicin elution and bead morphology. A) Doxorubicin loading over time (mean $\pm$ SEM; n=3). B) Doxorubicin elution over time (mean $\pm$ SEM; n=3). Solid line indicates a fit to first order release kinetics.

Figure 3. MicroCT images and bead composition analysis. Representative microCT images of individual beads showing that radiopaque DEBs have higher attenuation compared to radiopaque beads. The pie charts display composition of wet beads.

Figure 4.

Doxorubicin concentration versus attenuation. Radiopaque DEB (75mg doxorubicin/2 ml beads) containing agarose phantoms with various concentrations (0–12.5%) were imaged on a clinical CT scanner. Doxorubicin was extracted from these phantoms and quantified to yield doxorubicin concentration in the original phantom volume. Delta attenuation is used to indicate the change in attenuation since the attenuation of the phantom material was subtracted. These data were fit with linear regression.

Figure 5.

Rabbit Vx2 intrahepatic tumor embolized with radiopaque beads in contrast. A pre-embolization digital subtraction angiographic (DSA) image with injection from the proper hepatic artery clearly identified a tumor blush. A post-embolization native image demonstrates radiopaque beads and/or contrast within the hepatic arteries (arrow). A surface shaded display CT image of the embolized liver and a MIP microCT image of the same area (*approximate center of Vx2 tumor location) show radiopaque beads in tumor feeding vessels. Radiopaque beads were suspended in a 1:1 contrast to saline solution prior to delivery. The CT and micro CT are approximately aligned to aid in comparison.

Figure 6.

Hematoxylin and eosin images of a rabbit Vx2 tumor surrounded by normal liver embolized with radiopaque beads. A) Radiopaque beads (arrows) are observed in vessels in and around the tumor in this representative section. B) A radiopaque bead located within an artery surrounded by tumor. Note the proximity of this artery to a bile duct, suggesting that a previously existing portal triad was encased by the tumor. C) Radiopaque beads within an artery at the tumor periphery. Note small adjacent satellite metastases within normal liver tissue.

Schematic 1.

Summary of procedural steps for generating radiopaque DEBs.