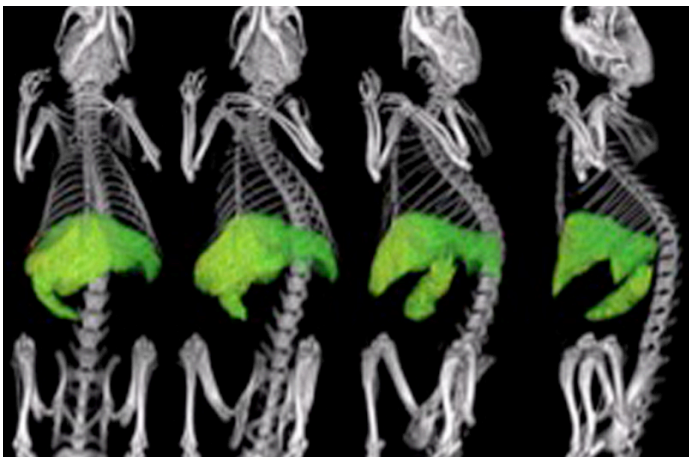

Rat Liver and Kidney Contrast-Enhanced µCT Imaging with the Albira CT

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Preclinical CT imaging is probably most commonly used in studies of bone anatomy and disease and as an anatomical reference for functional (i.e. PET and SPECT) modalities. Adipose tissue and lung tissue have innate contrast for CT imaging that can also be leveraged for preclinical studies of obesity and pulmonary disease (Sasser et al., 2012; Davison et al., 2013). Most soft tissue anatomy has limited innate CT contrast. However, some studies of soft tissue anatomy can be done using CT contrast agents (Lusic and Grinstaff, 2013).

There are a variety of commercial CT contrast agents. Exitron™ nano agents and Visipaque™ are used in small animal studies for liver and kidney contrast, respectively. Exitron™ nano 6000 and 12000 are alkaline earth metal nanoparticle agents that provide both liver and spleen CT contrast (Boll et al., 2011). Visipaque™ is an iodixanol based CT contrast agent that is approved for clinical arteriography/angiography, though in rodents Visipaque™ readily provides contrast in the kidneys and bladder (Heglund et al., 1995).

Contrast imaging, including liver contrast imaging using Exitron™ nano agents and kidney contrast imaging using Visipaque™, was demonstrated in mice and employing the Albira CT by Wathen et al. (2013). While mouse imaging techniques are established with a variety of agents (Rothe et al., 2015), there is less information available on contrast imaging for rats, which are up to 10-fold larger by weight. Here, we have identified basic methods to image kidney and liver systems in rats using two (Exitron™ and Visipaque™) commercially available CT contrast agents. These methods may be applied in future studies of liver and kidney disease using rat models.

Methods

Contrast Agents and Imaging

Rats were anesthetized using a mixture of isoflurane gas (3 % in O_2 (v/v)) at a flow rate of 2 L/min for animal preparation and imaging. For liver contrast, three rats were injected via tail vein with a bolus injection of 250 µL Exitron™ nano 12000 (Miltenyi Biotec, Auburn, CA, USA) and imaged immediately after injection and 24 and 48 hours post injection. Albira CT acquisitions were made using the Albira "Good setting" (400 projections, 125 µm voxels) at high dose (400 µA) and high voltage (45 kVp).

For kidney imaging three rats were injected via tail vein with a single bolus injection of 750 µL of Visipaque™ (GE Healthcare, USA) 270 mg/ml and imaged immediately after injection, and at intervals for three hours thereafter. Albira CT acquisitions were made using the Albira "Standard setting" (200 projections, 125 µm voxels) setting at high dose (400 µA) and high voltage (45 kVp).

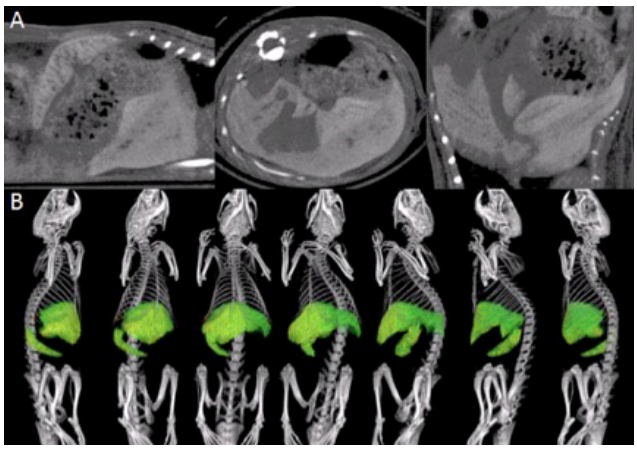
Image Presentation

All images were reconstructed with the Albira Reconstructor Software Advanced option at high resolution with a FBP algorithm. Images were processed in the PMOD (PMOD Technologies Ltd., Zurich, Switzerland) software and VolView (Kitware Inc.) software. 3D image presentation was performed according to the methods detailed by Wathen et al. (2012).

Results/Discussion

Preliminary studies (not shown) were made to identify a general working volume range of Exitron™ nano 12000 and Visipaque™ for contrast imaging in rats. Not surprisingly, a greater working volume is required for rat imaging than for mouse imaging. At 250 µL Exitron™ nano 12000 provided clear contrast for the liver and spleen for at least 48 hours after the initial bolus injection (Figure 1). This protocol provides a foundation for applied liver contrast imaging in rats. Previously Exitron™ agents have been used in µCT imaging mouse liver metastasis models (Boll et al., 2013), and the protocol described here could feasibly be translatable to imaging of rat liver tumor models.

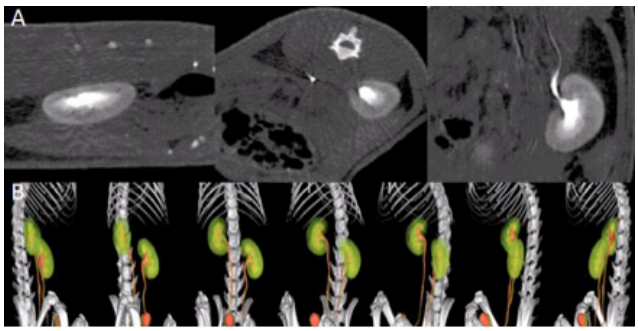
Figure 1



Rat liver and spleen contrast enhanced µCT imaging using Exitron™ nano 12000. A) Liver and spleen are clearly visible in rat injected with Exitron™ nano 12000 and imaged by µCT, here showing sagittal, transverse, and coronal views. B) 3D view of rat liver and spleen contrast enhanced µCT imaging.

A working volume of 750 µL of Visipaque™ was found to provide good contrast for kidney and bladder contrast in rats (Figure 2). The contrast agent cleared from the kidneys completely within 3 hours post injection, though the bladder contrast was visible for the 3 hour duration of imaging studies. The results provide a basis for applied kidney imaging using the Albira CT system. One potential application for this protocol would be for enhanced anatomical CT reference for multimodal PET and/or SPECT imaging. This could be particularly relevant in disease models where fine anatomical localization related to the kidney or bladder is required. Additionally, a method for evaluating kidney clearance rates in mice using Visipaque™ and X-ray imaging was reported (Orton et al., 2010). Similar analysis for kidney clearance in rats would likely be possible through adoption of the methods reported here, and using the Albira CT 2D dynamic scan setting.

Figure 2



Rat kidney contrast enhanced µCT imaging using Visipaque™. A) Kidneys are clearly visible in rats injected with Visipaque™ and imaged by µCT, here showing sagittal, transverse, and coronal views. In these view, the renal cortex and renal pelvis can be easily contrasted. B) 3D view of rat kidney contrast enhanced microCT imaging. In this view of the lower anatomy the ureter (orange) and bladder are clearly visible.

Conclusion

Rodent liver and kidney tissue has an innately low contrast with µCT imaging. Exitron™ and Visipaque™ contrast agents have been used in µCT studies of mouse liver and kidney disease. Workable volumes and useable timing for both agents to perform µCT contrast imaging in rats and employing the Albira CT could be identified. These methods should be applicable for studies in relevant rat liver, kidney, and bladder disease models.

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