WEBINAR:

(March 13, 2024) Overview of Preclinical Small Animal and Multimodal Imaging

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Questions and answers from the March 13, 2024 webinar titled: *"Overview of Preclinical Small Animal and Multimodal Imaging"*

This document includes questions we received and answered during the webinar, as well as those that we did not have time to address.

1. **What is the resolution of CT?**

Tonya Coulthard: That is highly dependent on the system used and the level of radiation dose that you choose to use to acquire the image. For example, with micro-CT system you could achieve sub-micron resolution, but more typically 2-5µm, however the radiation dose required for this type of image requires that the sample be *ex vivo*. For *in vivo* applications, resolution of 50-100µm is routinely achieved by most newer systems, with radiation levels below that which would affect the outcome of a longitudinal study with multiple imaging timepoints.

2. **Is there an instrument or a system that allows to combine PET with bioluminescence?**

Tonya Coulthard: I had the exact question yesterday from someone I was talking with. I'm not aware of a system on the market that directly combines them simultaneously or sequential in the same instrument. However, there are lots of different animal shuttles that can take the animal between an optical or BLI system and a PET system.

For example, we have one that works very nicely with our optical imaging system, and is able to go in every other imaging system that we have. This allows for multimodal imaging, we also work with third-party software to allow users to co-register images from different imaging modalities.

Interesting that you mentioned BLI and PET, because PET is traditionally a 3D image, and BLI is traditionally a 2D image. We have options to do, 2D PET, 2 BLI, 3D BLI, and 3D PET. And so, considering if you're doing 2D, or 3D comes into effect when you're co-registering those images as well. But again, we work with third-party software that is imaging system

agnostic. So, you can take images from really any imaging system that we have, and other manufacturers as well and co-register them within the software, either using fiducial markers or by manually translating, rotating, growing, and stretching those images.

3. **Were all the slides generated on equipment that you support?**

Tonya Coulthard: Yes, all of the images that I showed were acquired on systems that we sell and support. This wasn't meant to be Scintica specific, but the images that you saw were acquired on the systems that we sell as well, and we're happy to chat through any of those with you, just reaching out to info at Scintica.com, or visiting the website and submitting your questions there.

4. **How good is your 3D BLI, in terms of resolution and accuracy.**

Tonya Coulthard: It's a great question, and BLI is something that I've worked with through 2 companies that I worked with. I mentioned that I worked with Aspect Imaging which is the MRI company that we represent now. I worked for them directly before, and we were working with a team to develop a BLI co-registered with the MRI. And then, of course, with our optical imaging system, we have the BLI capabilities. So, the way the images are acquired is multiple; different, wavelengths are acquired. Then there's an equation that allows us to look at the intensity of those signals at the different wavelengths, and approximate where that tissue, or where that signal originated from at the depth within that tissue, the resolution is again similar to what we would have on 2D BLI. But we're able to localize where that signal came from. There are validation studies done in terms of making sure that the math and the equations work and using a known source.

5. **For fluorescence CCD, do you have a probe to target such as things as TNFa *in vivo,* for example using confocal?**

Tonya Coulthard: We do not have many consumable agents, such as antibodies to detect TNFalpha, for confocal or other applications. I do believe there are commercially available agents that could be used for this purpose available. I think the most reliable source would be from commercial antibody suppliers.

6. **If we have any phantoms for calibrations of the system**

Tonya Coulthard: We do. So, there's a group from Phantech. They're based out of Wisconsin, and they are imagers themselves, and they've developed a whole series of different phantoms that you can load with your own PET agents, and they have a mouse that they can put in your optical agents as well to help with calibrating. There are NIST standards that can be used for the bioluminescent side that allow you to know the exact number of photons that are coming up for calibration. But the PET phantoms would allow

you to calibrate radioactive dose as well as partial volume effects as well. These are available on our website.

4. Can we share the presentation?

Tonya Coulthard: Absolutely. So, in a couple of days you'll get a link to the recording the slide deck will be shared on that link. But if you want the actual slides, just reach out to us at info@scintica.com and they'll pass the email onto me, and I can share you share with you a download link to the slide deck as well.