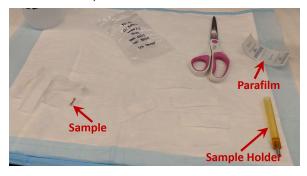
Video Demo - How to set up a scan on μCT35

By PCMD MicroCT Imaging Core

Youtube link: https://www.youtube.com/watch?v=QUt0QqIYJ80
All our video tutorials are listed on our website: https://www.med.upenn.edu/orl/uct/user-tutorials.html

Step 1: Wrap & put the sample into the sample holder

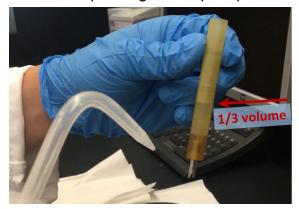
Prepare your sample using these materials: sample holder, parafilm, scissor, tweezer, gauze. In this video, mouse femurs were used to demo the setup process.



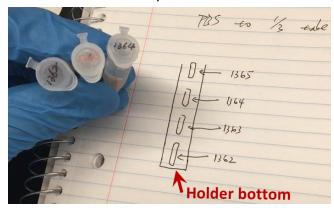
1. Cut the gauze into several strips. (Note: The size and quantity depend on your samples.)



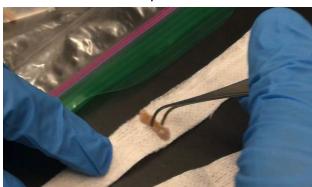
- 2. Suppose you want to scan samples at $6\mu m$ resolution on the $\mu CT35$ scanner. You should use 11.5mm (diameter) sample holders. Fill $\frac{1}{3}$ of the sample holder with PBS.
- Note 1: if you are unsure to use which sample holder, please consult with the manager.
- Note 2: Besides PBS, you can also fill the sample holder with ethanol, water, 10% formalin, or other medium, depending on the post-processing of your sample.

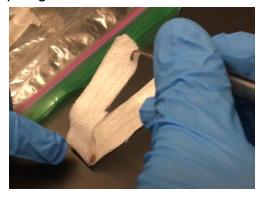


3. If you have multiple samples to load, please remember to record the locations of the samples within the sample holder.



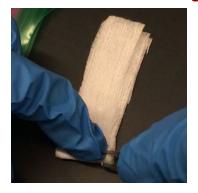
4. Place the sample in the middle of the strip of gauze, and then fold it.





5. Use a tweezer to grab and roll the sample in gauze while pressing FIRMLY. **This step is IMPORTANT to avoid motion artifacts during scanning.**





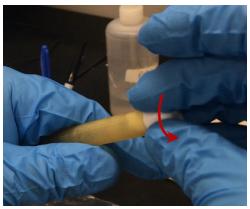
6. (Optional: If you want to fit as many samples as possible into a sample holder) Use your hand to feel the sample location inside the gauze, and then trim extra gauze accordingly.
Attention: Avoid cutting samples while trimming the gauze!



7. After the 1st round of gauze wrapping, if the sample holder is still too large to fit the sample securely, you should continue wrapping in gauze. Use your hand to roll the samples over gauze.



8. Twist and push the sample into the sample holder. (Make sure the sample is secure inside the gauze!)

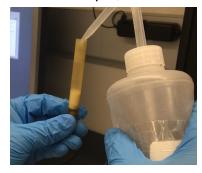


9. Once the sample is inside the sample holder, use a tweezer to push the sample toward the bottom.





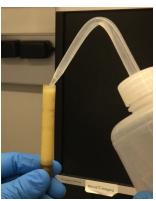
10. Fill more PBS into the sample holder. This is to ensure the sample is immersed in media. For example, in this demo, the 11.5mm sample holder can fit 4 mouse femurs.





- 11. (Optional for scanning multiple samples) Repeat steps #4 to #9
- **12.** After all samples are loaded into the sample holder, add more PBS to ensure all of the samples wrapped in gauze are immersed in medium.



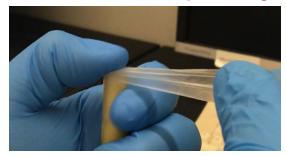


13. Cut a piece of parafilm, peel and then stretch it out entirely.



14. Wrap the top of the sample holder with parafilm. After each round of wrapping, use your finger to push on the top to ensure a tight seal.

IMPORTANT to avoid liquid leakage from the sample holder, which can damage the scanner!!!



15. Make sure every part of the parafilm is tightly attached to the holder. Trim extra parafilm accordingly. (**Everything must be within the** Field of View during the scan!)



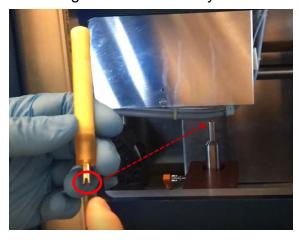


Step 2: Place the sample holder onto the rotation stage in µCT35 scanner

1. If both the X-ray tube and Door Locker indicator are OFF, you may open the door on μCT35.



2. Pay attention to the notch on the metal base of the sample holder. This notch should be aligned to successfully load the sample holder onto the rotation stage.



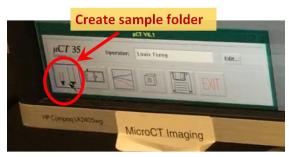




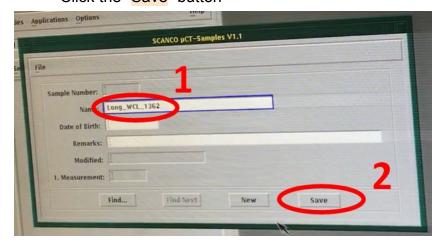
3. Close the door on µCT35. (You may push a little bit to confirm it is secure.)

Step 3: Create sample folder in the μCT35 scanner

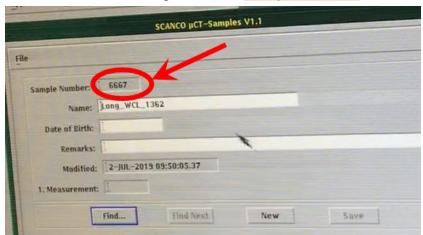
1. Click the first left button in the Control box (at the left bottom of the screen)



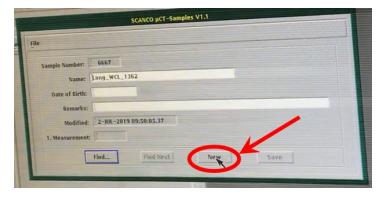
2. In the "µCT-Samples" window, enter your sample name:
Pl's initial_your Pennkey_Sample info
Click the "Save" button

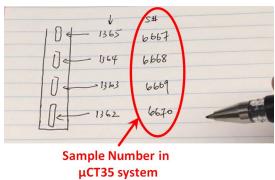


3. Write down the generated "Sample Number".



4. (Optional if you need to create multiple sample folders) Click the "New" button to create additional sample folders. Remember to record your "Sample Number" information as it will be required for subsequent steps.





5. Close the "µCT-Samples" window

Step 4: Perform a scout view in the µCT35 scanner

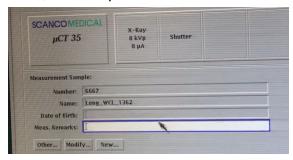
1. Click the second left button in the Control box to open the scan software



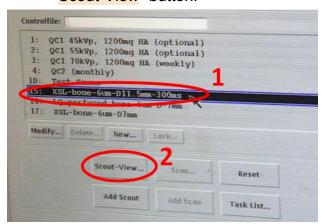
2. Enter Sample Number (generated in Step 3. It's "6667" in the video demo), and then click the "OK" button.



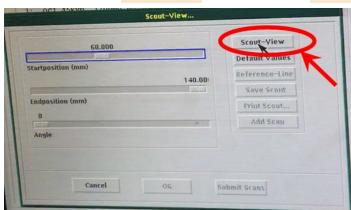
Then, the sample info will be shown in the scan software.



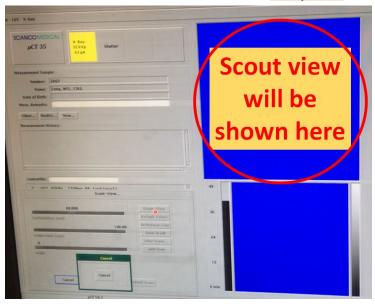
3. Select an appropriate controlfile (consult with the manager if unsure), and click the "Scout-View" button.



4. Click the "Scout-View" button in the "Scout-View..." window.

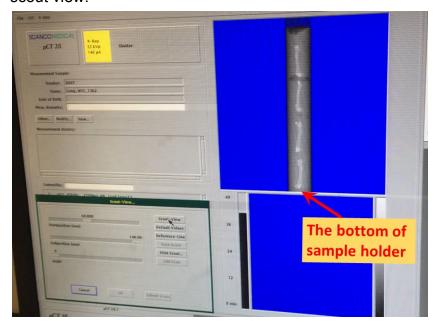


5. Wait 3~5 min. You will see the X-ray tube and Door lock indicator light turn ON.

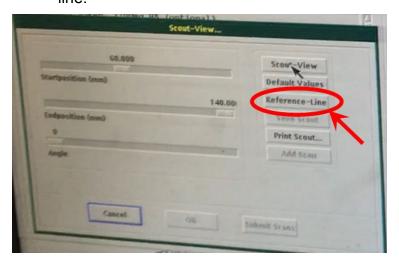


Step 5: Select the region of interests and start scan

- 1. After the scout view is finished, you will see your sample(s) in the software interface.
- Tip 1: Click and hold the **Middle** button of the mouse: enlarging the scout view.
- Tip 2: Click and hold the **Right** button of the mouse: moving the scout view.
- Tip 3: Click and hold both the **Middle** button of the mouse and Shift key: adjusting the contrast of the scout view.



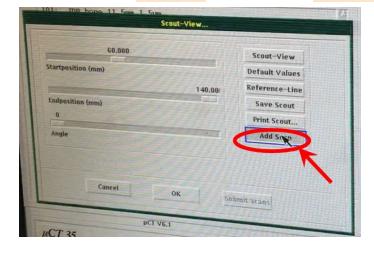
2. Click the "Reference-Line" in the "Scout-View..." window, hover the mouse to your desired region. If necessary, press and hold the Shift key to adjust the scan slice number. Once you have identified your region of interest, click the Left button of the mouse to set your reference line.





Note: If you want to change the position of the desired region, click the "Reference-Line" in the "Scout-View..." window again to cancel the previous selection.

- 3. Once your scan region is selected, the estimated time will be shown. The actual time is longer than that, usually at a factor of 1.2. As the estimated time is 41.0min in the video, the actual scan time will be $50 = 41 \times 1.2 = 1.2$
- **4.** Click the "Add Scan" button in the "Scout-View..." window.



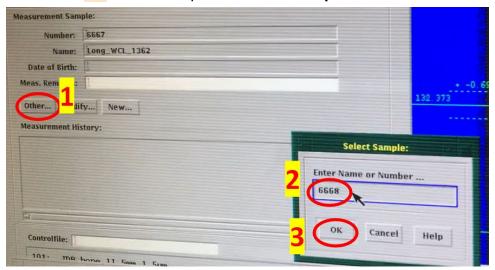
5. Click the "OK" button in the pop window "Information".



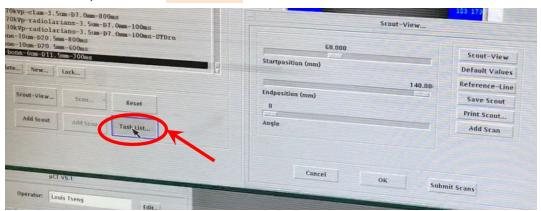
Optional: If you want to select the midshaft region, press and hold the "Alt" key, and the central region will be selected based on the length of the sample.



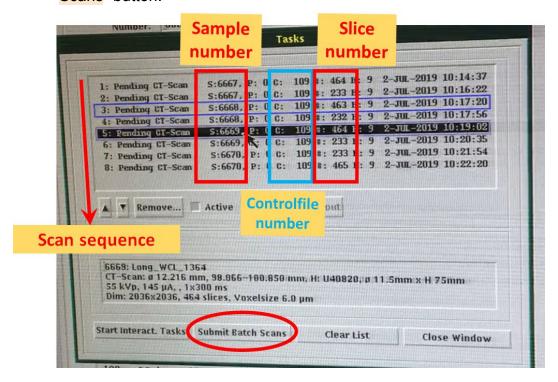
6. Optional: If you have added all regions of interest for this sample number, you can change to another sample number. Click the "Other..." button, enter your next sample number, and click the "OK" button. Repeat the above **steps 2~5**.



7. Once all sample regions are added to the scan tasklist, move the "Scout-View..." window aside, click the "Task List..." button.



8. Double check if all info in the "Tasks" window are correct. If correct, click the "Submit Batch Scans" button.



- 9. Wait for 1~2 min. The scan software will disappear.
- **10.** To confirm your scan is successfully initiated, type either "show queue" or "que" in the command window, and press Enter. You will see the "MEASUREMENT" followed by the sample number. Done!

