Tube sample holder set for the SkyScan desktop microCT scanners

Method note
MCT-014

1. Introduction

The tube sample holder set (TSHS) is a solution for mounting samples for micro-CT scanning in the SkyScan 1272, 1172, 1173, 1275 and 1174 desktop scanners. It consists of a metal (aluminium) base on which tubes with four inner diameters (6, 10, 15, 20mm) can be mounted in accurate straight vertical alignment.

Note that for the SkyScan1173 and 1275 models, plastic sample tubes with a longer length are available,

The TSHS is useful for batch scanning, allowing multiple samples to be mounted in a vertical line series. It is also a solution for mounting long samples for oversize scanning.

The tube sample holder (TSH) is also useful for the centred positioning of samples in general. For best results from micro-CT scanning, the sample's midline or axis should lie along or close to the vertical axis of rotation of the sample stage. The TSH makes it easily to get well centred positioning of samples.

Note that while it is rare that your sample size will be exactly suited for secure fitting in one of the tube diameters, it is possible to employ laboratory paper tissue or gauze or similar fabrics, to "pad out" the sample, to fit inside the tube a little wider than your sample. Paper and fabrics usually have low density and will not unduly affect the overall sample x-ray opacity.

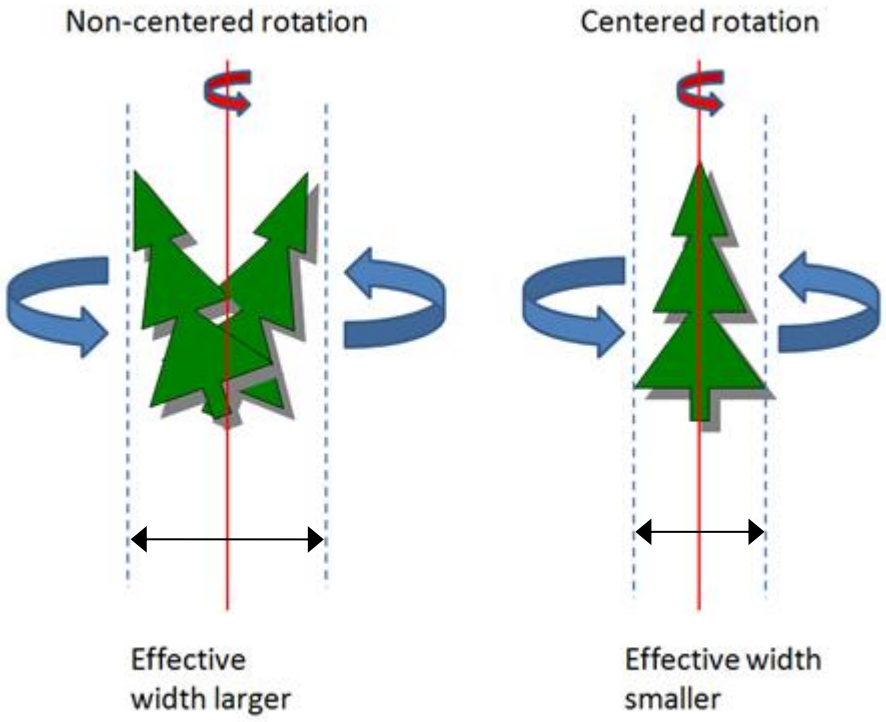


Figure 1. A well centred sample will rotate symmetrically during a scan, and have a smaller effective width, allowing higher magnification (smaller pixel size) and thus better resolution. Furthermore, post-alignment correction during reconstruction also works better with centred rotation.

2. Method

A method will be described here for loading bone samples in the vertical tube of the tube sample holder (TSH), wrapped in moist paper tissue. This method can be applied generally to biological tissue scanning.

2.1. Select vertical tube diameter

Identify the narrowest sample tube which will accommodate all the samples to be scanned. It is important that in a single experimental study, the tube used is the same for all samples. Tube inner diameters are 6, 10, 15 and 20 mm (approximately).

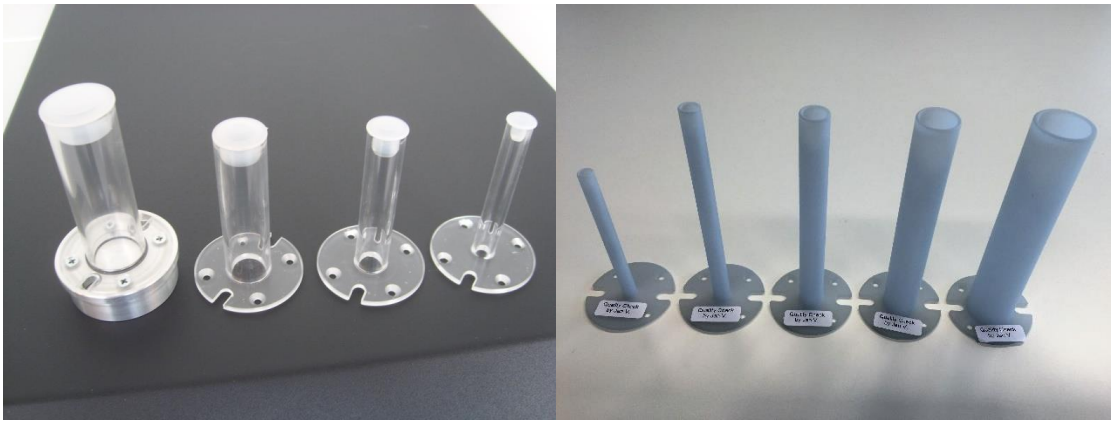


Figure 2. The set of sample tubes with disc platforms with inner diameters of (approximately) 6, 10, 15 and 20mm. The standard tube set on the left has tubes with length 8cm. The tubes on the right are elongated versions – 8.5/12 cm long – for use in the SkyScan1275 and 1273.

2.2. Take care to maintain sample identity during batch scanning

When removing multiple samples from storage for scanning, be careful to maintain sample identity.

Figure 3. Four rat distal femurs prepared for scanning, with label cards to maintain identity.

Roll up samples in a tissue paper strip

Cut a strip of paper tissue, and roll the sample up in this strip. Adjust the length of this strip so that the rolled up sample fits into the tube and slides along inside it, while in tight enough contact with the tube sides so as to maintain its position securely without the possibility of slipping during scanning.

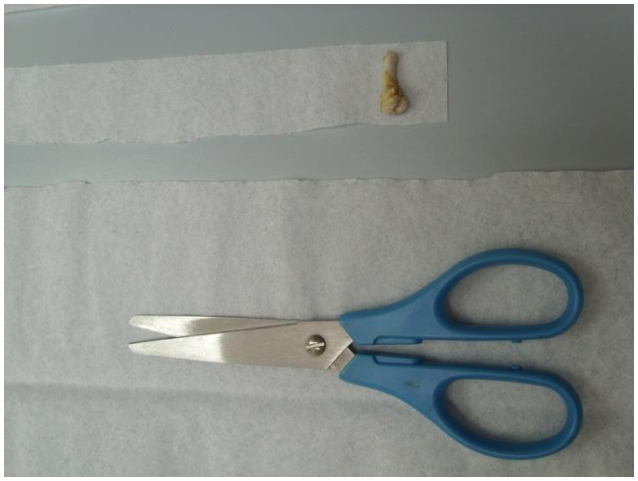


Figure 3. Preparing a paper tissue strip for wrapping biological samples.

2.4. Place the tissue-wrapped samples into the plastic tube, then wet the tissue paper

Place the wrapped sample in the tube so that it grips the side of the tube – for secure sample positioning – but can be moved easily enough (without stress to the sample). Then add several drops of water so that all the tissue surrounding each sample becomes moist. Only add the minimum water to moisten the paper tissue – free standing water around the sample is not necessary.

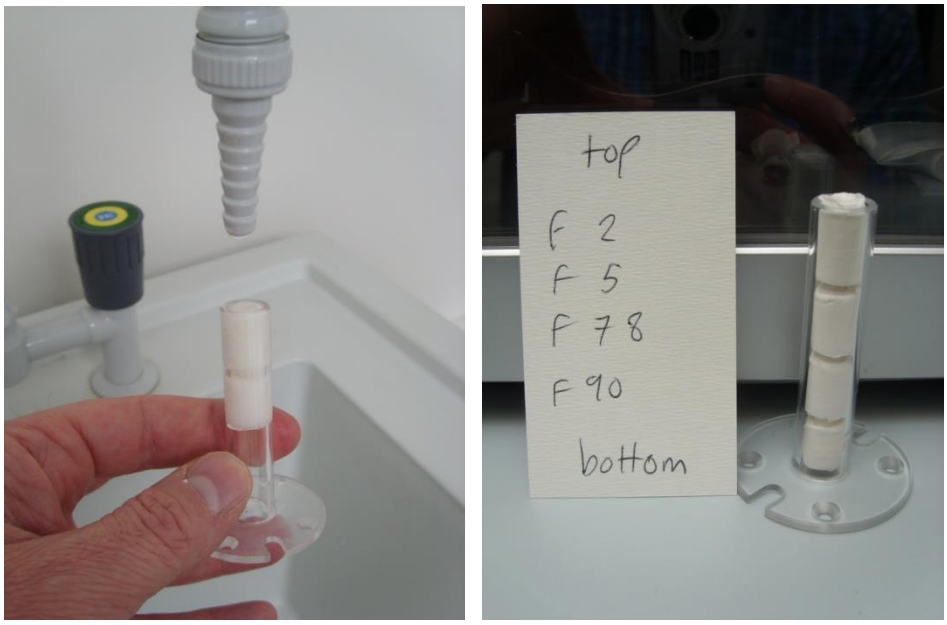


Figure 4. With the wrapped sample in the tube, add some drops of water to make the tissue paper moist. A card with sample identities listed can help maintain sample identity.

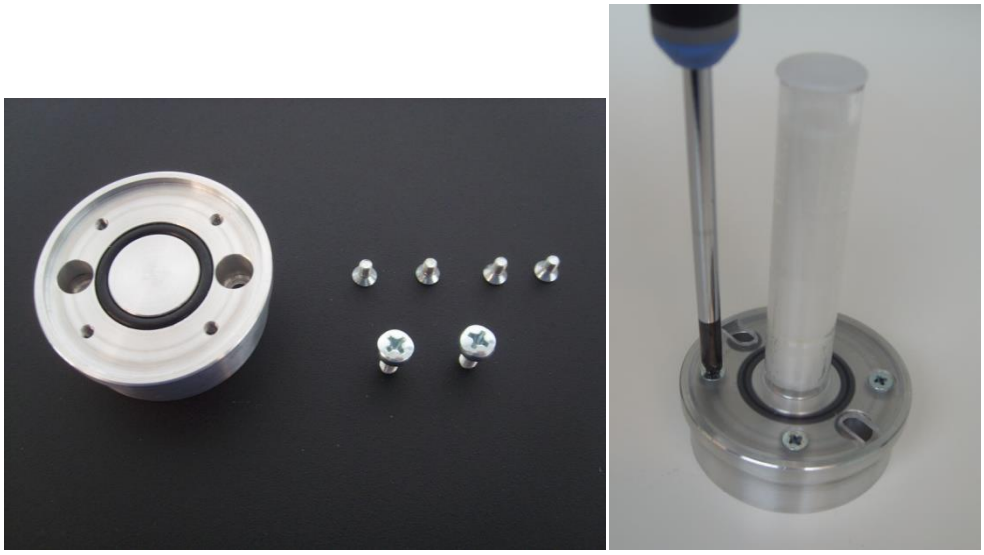
2.5. Attaching the sample holder tube to its base and into the scanner

Four small Phillips type screws attach each tube holder by its disc base to the metal stage of the TSHS.

Two larger Phillips type screws attach the whole TSH assembly (metal stage with tube attached) into the SkyScan 1172 or 1174 sample chamber, using the bolt attachments on either side of the brass stage nut.

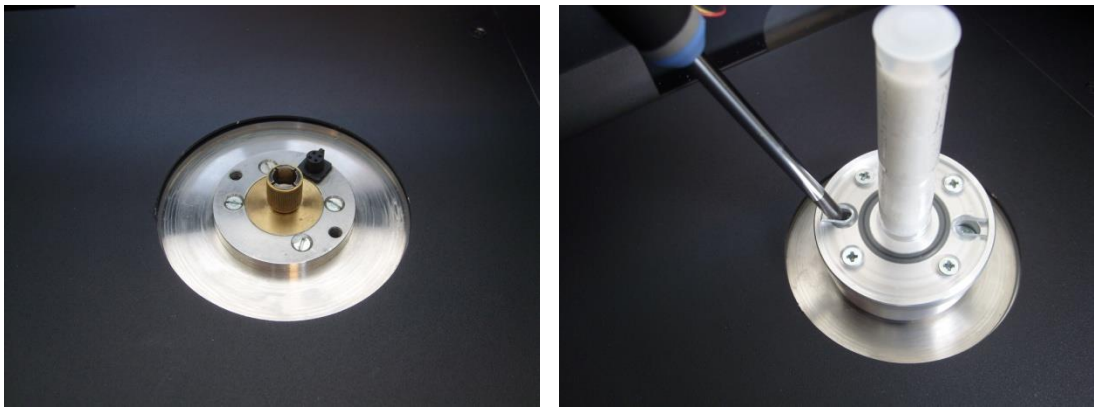
Please note – you should remove the brass stage nut before attaching the tube sample holder (TSH).

Please note – do not over-tighten any of the screws.



(a) (b)

Figure 5. The metal stage of the tube sample holder contains a rubber o-ring to keep the fitted tube water-tight. Four small Phillips-type screws attach each tube by its disc base to the metal stage (a). Two larger screws attach the assembly of stage plus tube into the scanner (b).



(a) (b)

Figure 6. The brass stage nut (a) should be removed entirely before fitting the tube sample holder. Then attach the tube sample holder assembly by the two larger screws through the two holes on either side of the metal stage (b).

Once the tube sample holder containing wetted tissue-wrapped bone samples is fitted to the scanner, place the plastic lid on the top of the tube.

Then proceed to carry out the scan of the four samples, using the batch scanning function in both scanners. Refer to the manual of the 1172 and 1174 scanners, and the "SkyScan 1172 method notes" document.

2.6. Scanning samples in water or other liquid medium

The metal base securing the plastic tube holders contains a rubber O-ring inside a circular recess (figure 5a). The plastic tubes have a flat basal disc which is held by screws onto the metal base (figure 5b). The pressure of the tube basal disc held against the metal base and O-ring maintains a water-tight seal. The tubes are also provided with plastic lids. Therefore the tube is water-tight and can be filled with water or other (non-corrosive) liquid medium such as water, saline or dilute formalin (e.g. 4% and phosphate buffered) for biological samples.

NOTE ABOUT ALCOHOL

Early models of the tube holder set were made of **polished acrylic** plastic. These look shiny and highly transparent. However polished acrylic becomes damaged by cracking (or fine crazing on the surface) by exposure to ethanol and probably other alcohols. So if your tubes are acrylic, do not put alcohol in them, and alcohol preserved samples should be wrapped in parafilm or wetted paper tissue. However the later and current model of tube holder set employs tubes made of **polycarbonate**. This looks a little darker and less transparent – and less shiny. However polycarbonate is unaffected by alcohol and is generally a chemically resistant plastic.

When holding samples in liquid, always attach the plastic lid to the top of the tube.

Please note: Care should be exercised if using the TSH filled with fluid. The tube should be prepared and filled with fluid away from the scanner, not inside the scanner sample chamber. Obviously – remove the whole assembly with the two large screws, from the

scanner after scanning: don't remove the tube from the base with the four small screws, or the fluid will leak out from the bottom of the tube! Exercise common sense.

2.7. Fitting the tube sample holder to the SkyScan 1174 scanner

The attachment of the tube sample holder assembly to the 1174 is the same as for the 1172 – the same two bolt holes on either side of the brass stage nut are provided. The nut should be removed prior to fitting the TSH in both scanners.

However one special step is required for the SkyScan 1174: the stage elevation needs to be lowered to the lowest level, to make space for fitting the TSH and for the screw-driver to fit inside the scanner sample chamber. The stage is more robust to handling in the lowest, bottom position.

In the z-position control in the 1174 scanner, enter the value of 100mm to move the stage to the lowest position. The actual value of the bottom z position is variable but much less than 100 (higher values correspond to a lower physical position).

Please note – "normally" the stage controls of pixel size, rotation and stage elevation are inaccessible when the scanner door is open. To over-ride this, when the door is open the control button can be pressed on the keyboard, then the stage controls will be available. Open the stage elevation control in this way if necessary to lower the stage to the lowest position (z-position 100mm) to allow the TSH to be fitted as shown in figure 7.

(a) (b) (c)

Figure 7. The highest (a) and lowest (b) stage elevation in the SkyScan 1174 scanner chamber. Lower the stage to the lowest elevation to allow space for fitting the tube stage holder (c) with a screwdriver.

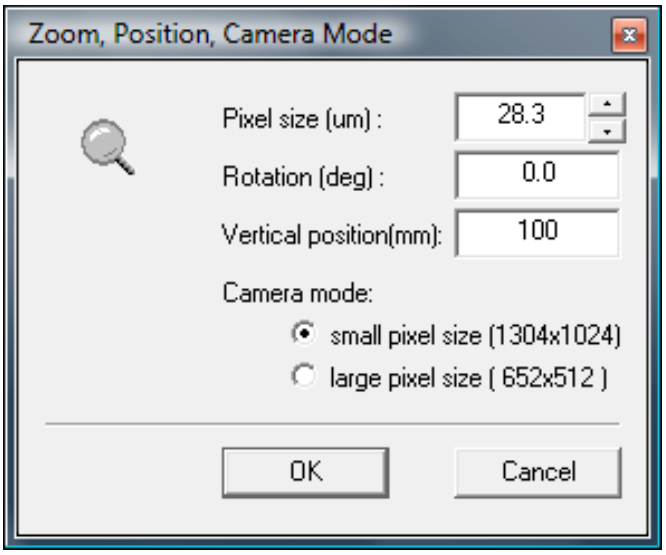


Figure 8. Set stage elevation to the bottom position by entering the value of 100mm, in order to allow installation and removal of the tube sample holder.