Synchrotron X-Ray CT of Wet Cortical Bone’s Nanostructure: Technique and Results

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Several Levels of Hierarchical Structures

Fibril bundles and Patterns of fibrils (~100 nm)

Visualizing bone’s 3D structures at sub-micron scales

[Access Science website]
Synchrotron X-ray computed tomography (CT) offers nano resolution (~ tens of nm) as compared to traditional CT.

**Experimental challenges arise**

- Sample Size; X-ray transparency at nano resolution limits the sample size to 10 – 15 micron
- Wet environment required for reliable bone structure imaging
Experimental challenges – Sample Preparation

Objectives:

• Extract ~ 10 micron needle-like sample cut in long. / trans.
• Free standing and rotate 180 deg, no vibration
• Create no side damage to substrate due to cutting
• Handle and ship to Chicago
• Remain wet at all time

Solution:
Microscopic ultra fast laser ablation
1. Slice cut, long./trans. ~ 5 x 30 mm polished to 100 micron thickness
2. Femtosecond Laser cut to the size shown below

PBS Solution wets the bone surface from opposite side of laser-matter interaction while cutting
3. Rotate 90 deg to laser thinning to ~ 10 micron
4. Rotate another 90 deg to cut needle
5. Pulse laser to put a neck ~ 10 micron
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Cylindrical chamber made of 20 µm thick, X-ray transparent Kapton film

The specimens were partly immersed in PBS solution, not to interfere with the X-ray path.

The chamber provided a moist environment for the specimens during the 3 hours tomography scans.
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Some Results

Fibrolamellar bone packets
Osteocyte Lacunae
Lamellar region
Blood channel

Canaliculi

X-Ray
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Conclusion and Remarks:

• A methodology for Nano-tomography of wet bone has been summarized.

• Ultra fast-laser ablation has been employed for extracting micron-size needle sample from bone in wet condition and with no side effect to sample.

• A simple but very effective technique has been used to take hard X-ray projections in wet condition during long-running CT.

• A wet sample cut in bone’s lamellar region between Osteocyte Lacunae and Blood channels.

• A 3D net work of a fibrils bundles as well as sub-micron Canaliculi have been visualized for wet bone.
Thank you