## Characterization of the osteocyte lacunocanalicular network in subchondral bone of healthy mice

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When bone and cartilage are joined together, both tissues present specific adaptation strategies to solve the biomechanical and biological dissimilarities between them. The bone that articulates in the joint is called subchondral bone (SB) and is separated from the articular cartilage by a region of mineralized cartilage. At the cellular level, mineralized cartilage is populated by chondrocytes whereas SB contains osteocytes. The latter are highly interconnected through the osteocyte lacunocanalicular network (LCN). Osteocytes are believed to regulate the biological processes of bone remodeling and mineralization [1]. Changes in SB are involved in joint aging and pathologies, such as osteoarthritis (OA). Common modifications observed in OA are subchondral thickening and increased subchondral remodeling [2]. The specific architecture of the LCN in the SB remains poorly characterized, limiting our understanding of its role in subchondral modifications. This study focuses on the LCN of SB and on the possible connections between osteocytes in SB and chondrocytes in mineralized cartilage. We analyzed a 10-week-old male C57Bl/6J mouse tibia and LCN was stained with rhodamine and imaged using confocal laser scanning microscopy (Stellaris 8, Leica) at a lateral resolution of 0.18 μm over a depth of 40 μm. Network analysis was performed with the dedicated open-source software TINA [3]. Canalicular density (Ca.Dn) exhibited high spatial heterogeneity, with lower Ca.Dn observed near the bone-cartilage and the bone-marrow interface (Fig. 1). Numerous stained chondrocyte lacunae (Fig. 2) were found over the full width of the articular cartilage, irrespective of their proximity to SB. Our initial results provide reference data for future work comparing healthy and post-traumatic OA scenarios.

## Acknowledgement

This project has received funding from the FWO and F.R.S.-FNRS under the Excellence of Science (EOS) programme (EOS No. 40007553)

## References

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- [2] Madry et al., Knee Surgery, Sports Traumatology, Arthroscopy, 2010. 18(4): p. 419-433.
- [3] https://bitbucket.org/refelix/tina/src/master/

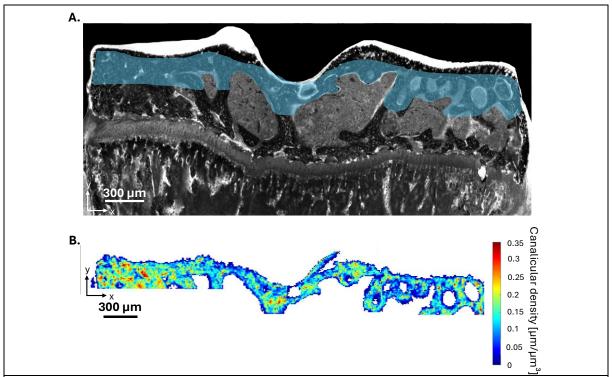


Figure 1 (A) Tibia cross-section showing the region measured with CLSM (highlighted in light blue). (B) Canalicular density map computed in subvolumes of  $400 \ \mu m^3$ .

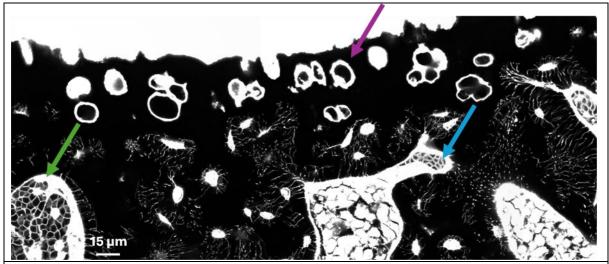


Figure 2. Close up at the bone-cartilage interface highlighting the OLCN and additional features stained with rhodamine: chondrocyte lacunae (purple arrow), bone marrow (green arrow) and blood vessel (blue arrow).