Fibrillar-level mechanics of the healthy and aged bone-cartilage unit

Himadri Shikhar Gupta*[1], Waqas Badar[1], Martin M. Knight[1], Sheetal R. Inamdar[1], Peter Fratzl[2], Tim Snow[3], and Nicholas J Terrill[3]

[1]School of Engineering and Materials Science, Queen Mary University of London – United Kingdom
[2]Max Planck Institute of Colloids and Interfaces – Germany
[3]Diamond Light Source, Harwell Science Campus, Harwell – United Kingdom

Abstract

The bone-cartilage interface at the ends of long bones is essential for distributing physiological loads across articulating joints, and facilitating smooth, pain-free joint movement (1). Structural and mechanical degradation of this bone-cartilage unit (BCU) is a key factor in developing osteoarthritis, affecting over 500 million individuals globally. Despite its biomedical and mechanobiological importance, the *in situ* structural deformation of the collagen-based extracellular matrix (ECM) within the BCU has been difficult to quantify due to the small length-scale of the deforming units (mainly Type II collagen fibrils $<$ 100 nm in diameter and proteoglycans along with water) and the graded, hierarchical structure of the ECM across the BCU, from the superficial (surface) layer to the underlying subchondral bone.
This study introduces a 3D synchrotron-based small-angle X-ray modelling and spatially correlative mechano-imaging approach to measure nanoscale fibrillar deformation across the intact BCU under biomechanical loading. Synchrotron small-angle X-ray scattering can detect the fibrillar-level strains, reorientation and intrafibrillar molecular organisation of the collagen fibrillar network, and when combined with compressive loading and high-brilliance synchrotron radiation, enables microscale maps of the fibrillar-level scattering to be determined. However, the anisotropic, graded structure of the scattering fibrils makes interpretation of the scattering signal complex. Here, we developed 3D fibre-symmetric models of the scattering from the fibrils to link the measured scattering to intrinsic fibrillar structural parameters, like the axial tropocollagen stagger (D-period, linked to pre-strain), fibril orientation, diameter and axial strain heterogeneity. Using this approach, we identified previously hidden deformation mechanisms within the semicrystalline collagen fibril network (2,3), which are spatially graded across the BCU. These include shifts in fibril crystallinity, pre-strain, lateral compression, axial contraction, and molecular disordering (2). In the superficial cartilage layer, collagen fibrils exhibit heightened molecular disorder. Progressing deeper toward the bone interface, fibrils transition from reorientation-driven strain at the microscale to molecular kinking strain. Simultaneously, fibrils undergo greater lateral compression than axial contraction, likely due to intrafibrillar water expulsion under load. The mineralized cartilage and bone closest to the interface with articular cartilage exhibits compressive deformation, but on going deeper into the subchondral bone, the fibril strain reduces. Further, the trabecular bone under the BCU shows a much lower pre-strain than the articular cartilage and the mineralized interface.

*Speaker

Age-related cross-linking of the BCU, simulated using *in vitro* glycation using ribose, further alters these mechanics by reducing fibril pre-strain, increasing fibril diameter, and diminishing the nanofibrous network's resilience by lowering fibril deformability. These findings significantly enhance our understanding of the nanoscale mechanobiology of the bone-cartilage interface and open pathways for targeted biomarkers and therapeutic interventions.

References: (1) D.M. Findlay et al., *Bone Res.* (2016); (2) W. Badar et al., *Adv. Sci.* (2024). (3) W. Badar et al., *PLoS One* (2022).