

THE OSTEOCYTE LACUNO-CANALICULAR NETWORK AT THE BONE-IMPLANT INTERPHASE IMAGED WITH FOCUSED ION BEAM – SCANNING ELECTRON MICROSCOPY

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Introduction

The biomechanical properties of the interphase between an implant and the host bone determine long-term stability of the implant. The compositional, structural and mechanical properties of the bone-implant interphase (BII) evolve during healing and vary at different length scales due to the hierarchical arrangement of bone tissue [1]. In particular, bone nano- and microstructure orientation and arrangement are important predictors of key mechanical properties such as bone strength [2]. However, these structural features at the BII are not fully understood [3], especially the spatio-temporal evolution during healing, and the impact on the resulting BII strength. This study aims at investigating the microstructure of the BII using electron microscopy techniques.

Methods

Specimen. A standardized coin-shaped implant (TiAl6V4, Ø5mm, H3mm, Fig. 1A) was osseointegrated for 7 weeks in the cortical bone of a rabbit tibia. The extracted specimen was embedded into PMMA, cut transversely into a $2 \times 5 \times 2 \text{ mm}^3$ cube and polished to expose the mineralized tissue at the BII (Fig 1B).

Imaging. A FIB-SEM (FEI Helios NanoLab 660) in back-scattered electron (BSE) mode was used to image a $20 \times 30 \times 30 \mu\text{m}^3$ region close to the implant (Fig 1B). 3D images were obtained using the “Slice and View” mode (Fig 1C) with 20nm distance between two slices. Image analysis was done using ImageJ (v.1.53f51).

Results

Ellipsoidal osteocyte lacunae ($\sim 10 \mu\text{m}$ long axis, $\sim 5 \mu\text{m}$ short axis) were observed and appeared aligned parallel with the implant surface (Fig 1D). Multiple canaliculi ($\text{Ø } 100\text{-}300 \text{ nm}$) were seen connected to the lacunae, permeating the bone matrix. Collagen fibrils were found to alternate in orientation within 2-3 μm thick lamellae, also aligned parallel with the implant surface (Fig 1D).

Discussion

The utilised implant model allows access to a bone-implant interphase where the tissue is certain to be newly formed. This enables investigation of the lacuna-canalicular network (LCN) in young bone close to an implant, and comparison to mature bone. Alternating orientation in collagen fibrils within lamellar structures was found to be consistent with previous observations in

mature bone [4] and osteocyte lacunae were observed to align along the implant as seen previously for screw-type implants [5]. Further work will focus on extending the image analysis to investigate porosity distribution and alignment of the collagen fibrils within the LCN. Structural specificities of the newly formed tissue will be compared to a mature region in the same specimen.

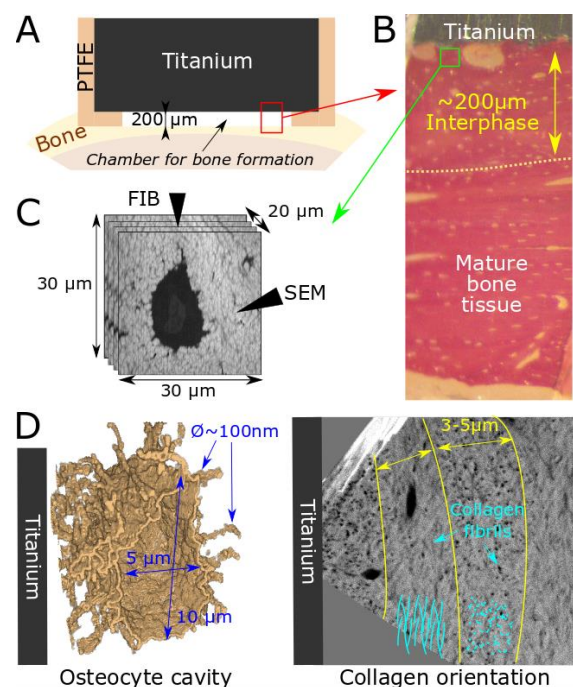


Figure 1: Standardized implant (A), specimen with bone tissue stained red with Picrofushin (B), FIB-SEM “slice and view” (C); osteocyte cavity and lacuna-canalicular network in immature bone tissue (D, left) and changes in collagen fibrils orientation (D, right).

References

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