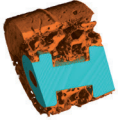


Quantification of Bone Around Surface Modified Titanium Implants

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INTRODUCTION



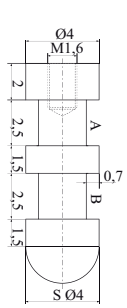
The influence of surface properties on the bone growth around titanium dental implants in relation to the healing time is of high interest for the clinical use. A common way to characterise differences of bone contact around implant surfaces is the histomorphometry. Despite the excellent lateral picture resolution of this technique, a better spatial resolution (~10µm) and easier sample preparation is required. Both aspects appear to be important for a statistical analysis of differences in bone formation around surface modified implants. Microtomography with synchrotron radiation (SRµCT) can determine the bone growth around the implants non-destructively with a high spatial resolution and with minimal sample preparation. Using the digital information from the CT-measurements, the aim is to find appropriate methods for a spatial quantification for newly formed bone.

MATERIALS

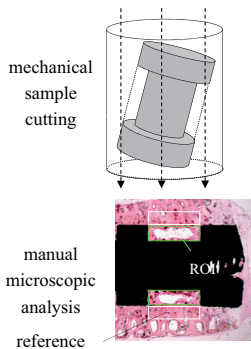
Titanium implants coated with collagen type I and III, a peptide, and without coating were placed in the femur condyle of goats for 4 and 12 weeks. After extraction and embedding in PMMA, samples with titanium implants and surrounding hard tissue (diameter = 8mm, length = 15mm) were investigated by the SRµCT experiment at the beamline BW5 of HASYLAB at DESY. Monochromatic X-Ray with a photon energy of 70 keV were used to investigate the samples under 720 different angles with a resolution of ~10µm. In a second step they are histologically prepared.

ANALYSIS

A) implant design



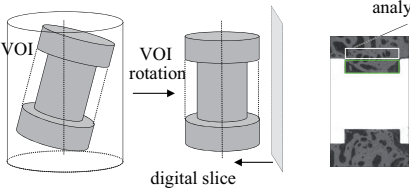
B) histological analysis



A) Schematic view of the cylindrical implant with two cavities.
B) histological procedure for the cylindrical implants.

C) SRµCT analysis

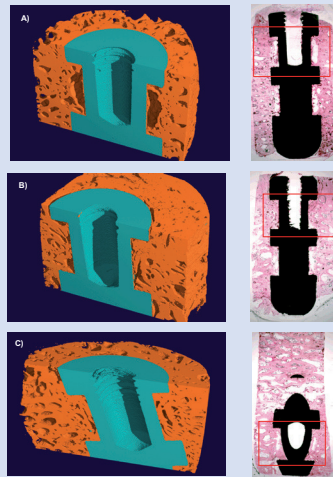
interactive ROI and reference analysis



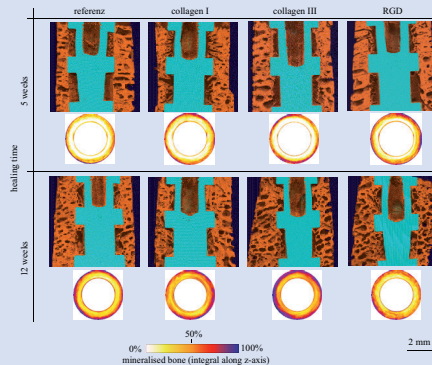
C) SRµCT-analysis procedure for the cylindrical implants. The volume of interest (VOI) is set to the cavities. The bone amount in a reference area above the VOI is calculated to take the surrounding bone quality into account.

The reconstructions obtained from the SRµCT-measurements show a clear contrast between the absorption values of mineralized bone and the titanium implant. Image artefacts usually found near the high absorbing titanium, are not visible.

VISUALIZATION



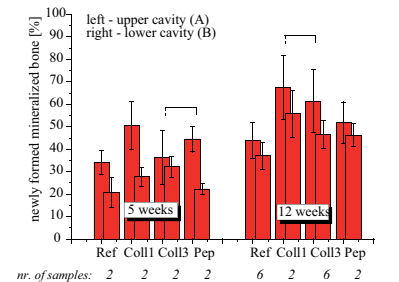
SRµCT visualization of mineralized bone (orange) and titanium implant (cyan) after a healing time of 12 weeks for A) an uncoated implant, B) implant coated with collagen type I and C) an implant coated with collagen type III. The virtual slices in the SRµCT-volume were correlated to histological micrography at nearly the same sample position (right side, red box) and demonstrate the good morphological agreement.



3D-Visualization of the mineralized bone inside the cavities of the biofunctionalized surfaces after a healing time of 5 and 12 weeks. The image below each 3D-visualization shows the integral amount of mineralized bone for all slices along the z-axis inside the cavities.

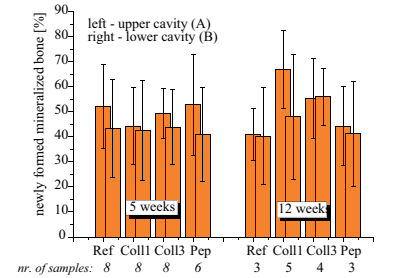
RESULTS

Bone volume - SRµCT (200 slices/sample)



Analysis of bone values inside the cavities from histological micrographs. No significant differences between the surface states could be found.

Bone volume - histology (3 slices/sample)



Analysis of bone volume from SRµCT data. For a given implantation time, all coatings differed significantly ($p < 0.01$) except those connected. No significant differences were found between materials at different healing times.

The analysis provides adequate information on the bone-implant interface and also suggest development of 3D image analysis modules to quantify the spatial bone amount around the implants.

With the integration of thresholded values for mineralized bone along the implant z-axis of the SRµCT-volume preferred zones of bone formation could be visualized. The newly formed bone is mainly visible at the inner sides of the implant cavities. The SRµCT-analysis of the cavities in cylindrical implants shows differences in bone formation for the healing time and the surface modification.

Inside a given implantation time significant differences ($p < 0.01$) in bone formation between the surface modifications were found. In contrast, the statistical testing of the histological data revealed that no significant differences in bone formation between the implantation time and surface state existed ($p < 0.01$).

CONCLUSION AND ACKNOWLEDGEMENT

SRµCT allows a clear visualization of fully mineralized bone around the implants, as a very high number of slices can be generated per sample. As a consequence, the subsequent quantification and semi-automatic bone detection lead to a high precision of the results for each sample. The advantage of histological imaging is still the superb lateral resolution and the visualization of biochemical tissue properties. On the other hand, in terms of statistical relevance, visualization and quantification is more practicable with SRµCT. Consequently, a combination of classical histology with SRµCT can be a powerful tool for an improved understanding of biological reactions around biofunctionalised implants. This study was supported by the Bundesministerium für Bildung und Forschung (BMBF) and the Deutsche Forschungsgemeinschaft (DFG). The authors thank Prof. Dr. Rainer Koch, Institut für Medizinische Informatik und Biometrie, Medizinische Fakultät Carl Gustav Carus, TU Dresden for valuable assistance in statistical analysis. The authors also thank Jacky den Bakker and Jan-Paul van der Waerden, Department of Biomaterials, UMC Nijmegen for their extensive assistance in the histological sectioning and evaluation.