

Synchrotron radiation-based micro computed tomography in the assessment of dentin de- and re-mineralization

Florian Kernen^{1,2,3}, Tuomas Waltimo³, Hans Deyhle¹, Felix Beckmann⁴, Wendelin Stark⁵, and Bert Müller^{1,2}



¹Institute of Materials Science, Dental School, University of Basel, Switzerland

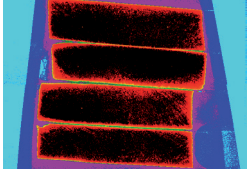
²Biomaterials Science Center, University of Basel, Switzerland

³Institute of Preventive Dentistry and Oral Microbiology, Dental School, University of Basel, Switzerland

⁴Institute of Materials Research, GKSS-Research Center Geesthacht, Germany

⁵Department of chemistry, ETH Zürich, Switzerland

INTRODUCTION



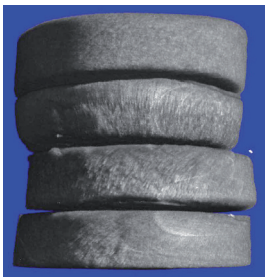
Since the introduction in the 1970s, bio-active glasses (BAG) have been applied in dentistry to augment alveolar ridges and to treat periodontal pockets. BAGs in water release their ionic compounds, resulting in a basic environment intolerable for most oral micro-organisms. In contrast to commonly used disinfectants in dentistry, BAGs induce dentin re-mineralization and the formation of calcium phosphate precipitations. Detailed data on the re-mineralization capacity, however, are rare. Synchrotron-radiation based micro computed tomography (SRμCT) allows to determine the local density distribution within a sample. The effect of BAG on artificially de-mineralized dentin specimens as examined *in vitro*.

SRμCT

Synchrotron radiation sources offer high brilliance monochromatic x-rays. SRμCT is a well established method for non-destructively determining the local x-ray opacity, and therefore the local density within a specimen with high contrast.

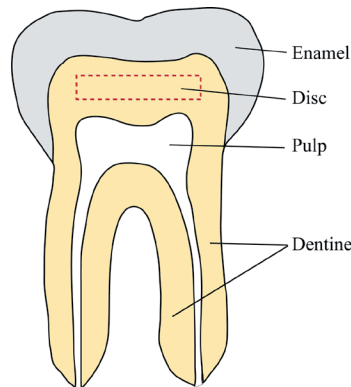
Through the histogram analysis of a dataset, density variations within a specimen can be quantified.

The dentin discs were placed in an Eppendorf tube in saline solution. The tomographic scans were recorded at the W2 beamline at HASYLAB (DESY, Hamburg, Germany) with 27 keV photon energy.



SPECIMEN PREPARATION

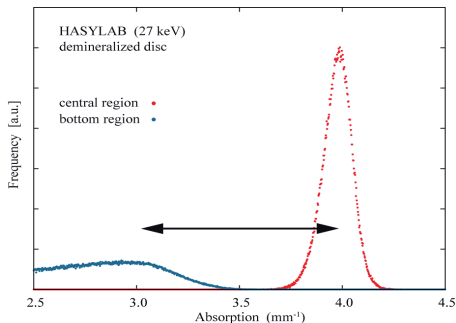
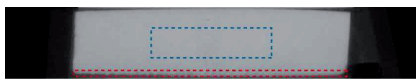
Four dentin discs were mechanically removed from freshly extracted human third molars using a trephine bur cylinder with a diameter of 4 mm. Subsequently, the discs, with a thickness of 0.8 mm, were cut from the dentin section of the cylinder using a microtome saw (Leica, Germany). The discs were autoclaved at 121 °C for 15 min before storage in sterile saline solution at a temperature of 5 °C.



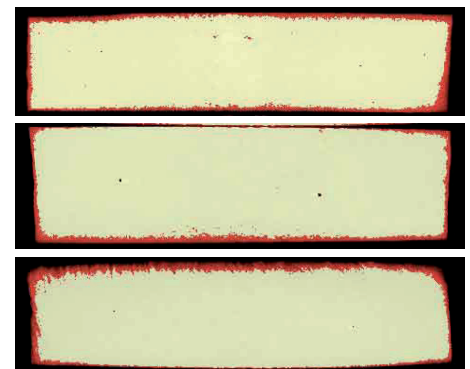
Disc 1 served as control and was left untreated. The other three discs were de-mineralized using 10% citric acid for a period of 10 min, to simulate carious lesion. Experimental flame spray-derived nanoparticulate bio-active glass SiO₂ (47.8%), P₂O₅ (4.6%), CaO (25.1%), Na₂O (22.6%) (NanoBAG, Department of Chemistry, ETH Zürich) was suspended in physiological saline (1:5 wt/vol). For the re-mineralization, discs 3 and 4 were incubated in the suspension at a temperature of 37 °C for 24 h and 7 d, respectively. Disc 2 was not incubated with BAG to allow for the quantification of de-mineralization

| Specimen | De-mineralization | Re-mineralization |
|----------|-----------------------------|------------------------------|
| Disc 1 | No treatment | No treatment |
| Disc 2 | Citric acid 37°C for 10 min | No treatment |
| Disc 3 | Citric acid 37°C for 10 min | NanoBAG 45S5 at 37°C for 24h |
| Disc 4 | Citric acid 37°C for 10 min | NanoBAG 45S5 at 37°C for 7d |

RESULTS



Only the rim region of the samples was affected by the treatment. The citric acid and BAG particles seem to penetrate the dentin only up to a depth of about 80 μm. The graph shows the difference in absorption, and thus HA-density, between the inner (blue) and outer (red) region of a demineralized sample. Note that only the peak position, not the curve height, is relevant for density determination. The pictures show a cross section through specimens 1,2 and 4. Shown in red are the rim zones, which exhibited lower hydroxyapatite density. Although such a lower density can be observed in all of the samples, it is thicker in the demineralized specimen, whereas a re-mineralization effect can be seen in the BAG-treated dentin.



CONCLUSION AND ACKNOWLEDGEMENT

SRμCT permits to locally determine spatial variations in dentin density and composition. As these properties can vary along the tooth, a sequential investigation of one specimen in a non-destructive way has to be preferred with respect to direct comparison between different discs. The treatment with citric acid and BAG only affects a rim region 80 to 100 μm deep. Therefore, surface-sensitive analysis methods such as Atomic Force Microscopy could be a valuable complement to SRμCT.

The authors acknowledge the valuable support of the beamline W2 team (HASYLAB at DESY, Hamburg, Germany) during the data acquisition.