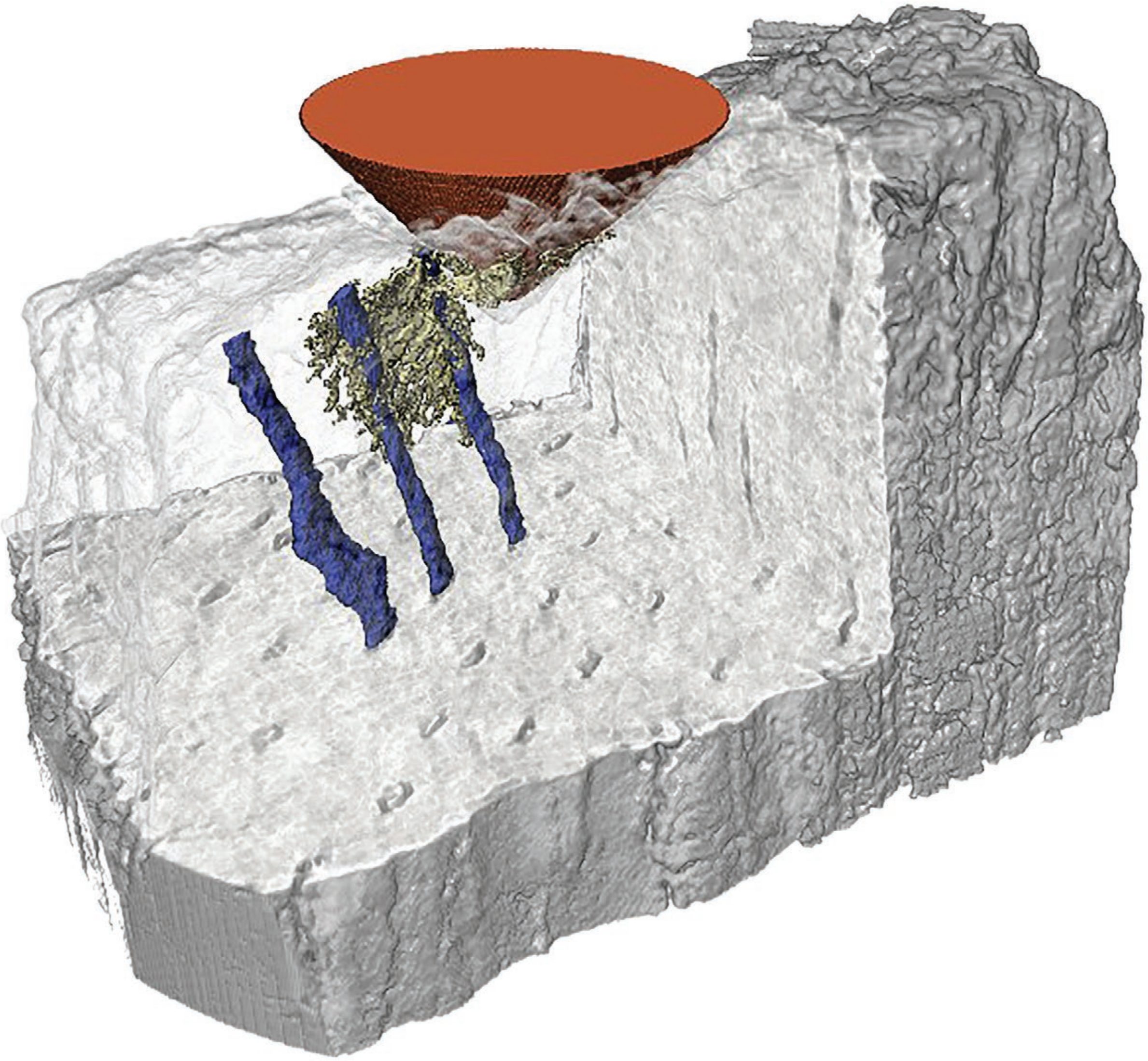

In situ 3D Imaging of Crack Growth in Dentin
at the Nanoscale

We make it visible.

In situ 3D Imaging of Crack Growth in Dentin
at the Nanoscale

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Dentin is a nano-composite material that forms the bulk of teeth, and provides teeth with fracture toughness. A better understanding of fracture in dentin is important to develop a framework for failure prediction, not only for clinical understanding but also for developing biomimetic restorative materials that are able to mimic the tissue's mechanical response. This study demonstrates how a novel nanomechnical test stage for the ZEISS Xradia 810 Ultra X-ray microscope can be used to study the progressive crack growth in dentin *in situ* at nanoscale resolutions.

Introduction

In 2012, Americans' spend on dental care reached $111 billion[1], with fractured teeth being commonly observed in dental practice. In particular, dentin at exposed root surfaces often exhibit notches, which are commonly the sites of failure following the cyclic loading that teeth undergo. Treatment for cracked teeth is often expensive and time consuming. A better understanding of how teeth fracture is key to the development of better oral treatments, including biomimetic restorative materials and the establishment of a framework for failure prediction in clinical research. Furthermore, such information would prove valuable to material scientists in order to design the tougher materials of the future.

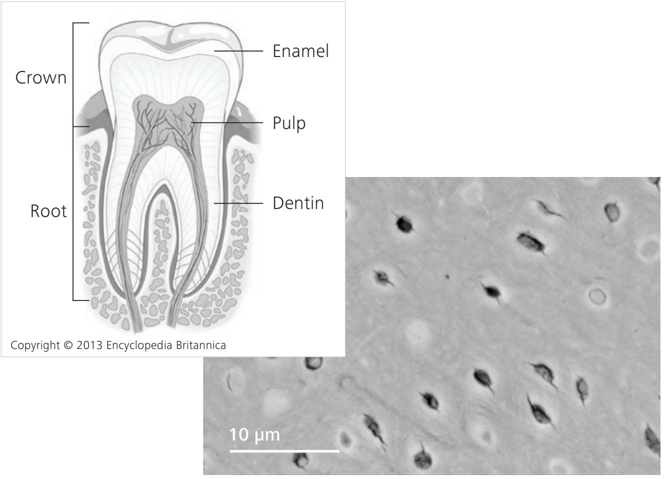

Figure 1. Left: Schematic diagram of the structure of teeth, showing the pulp, dentin and enamel. Right: An SEM image showing micro-cracking around tubules in dentin.

Dentin is a nano-composite material which forms the majority of the mineralised tissue in teeth. A key feature of dentin is the presence of tubules, which are microscopic channels followed by odontoblasts during dentinogenesis[2]. These tubules run radially from the pulp to the dentin-enamel junction in teeth, as shown in Figure 1. The enamel layer provides the tooth with hardness and strength while the underlying dentin provides toughness and hence resistance to fracture[3].

Dentin is composed of nano-crystalline hydroxyapatite (65 wt%), type I collagen fibrils (20 wt%), and fluid (15 wt%), the majority of which is found within the tubules. The dentin immediately surrounding the tubules (the peritubular dentin which is ~0.5-1 µm thick) is highly mineralised, while the remaining intertubular dentin comprises a mesh of collagen reinforced with hydroxyapatite. This random collagen scaffold is planar and oriented perpendicular to the long axes of the tubules[4]. The mineral in dentin is believed to provide strength, while the collagen provides toughness. However, the orientation of the tubules and the collagen scaffold gives rise to anisotropy in the mechanical properties such fracture toughness. Two of the principal mechanisms that have been proposed for the fracture resistance of dentin include the uncracked-ligament bridging ahead of the main crack tip and collagen fibrils bridging at the wake of the crack tip[3, 5-7].

Crack growth in dentin has typically been studied in 2D by SEM, with a limited amount of research looking in 3D crack growth development using microtomography[8,9]. In this study, a novel *in situ* nanomechanical test stage for the Xradia Ultra nanoscale X-ray microscope was used to initiate and propagate cracks in elephant dentin (tusk) during tomography. This enabled the progressive crack growth to be studied in 3D and *in situ* (under load) at 150 nm resolution for the first time. The results can provide new insights into anisotropic fracture behaviour and crack shielding mechanisms.

High resolution time-lapse study of *in situ* crack growth in dentin

ZEISS Xradia Ultra XRM is the only laboratory-based 3D imaging system available with resolution down to 50 nm. Xradia Ultra Load Stage, a novel nanomechanical test stage, has been designed specifically for integration in Xradia Ultra XRM. It enables a range of mechanical tests to be performed whilst imaging in 3D at ultra high resolutions. The stage features a high precision piezo actuator and an integrated load cell, enabling the load-displacement curve to be measured and related to the evolving microstructure observed in the corresponding 3D tomographic reconstructions.

For this study, indentation with a diamond cone indenter was selected to initiate and propagate cracks in elephant dentin. A suitable dentin sample was prepared using mechanical polishing and mounted on the sample pin as shown in Figure 3. The test stage features fine control of the position of the indenter tip relative to the sample's upper surface. The 3D rendering of the sample in Figure 4, prior to indentation, shows the ability of Xradia 810 Ultra to capture the arrangement of the tubules with fine detail.

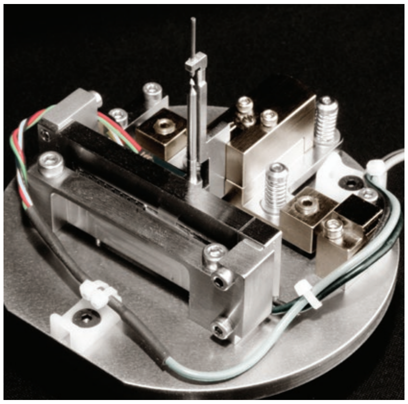

Figure 2: Xradia Ultra Load Stage

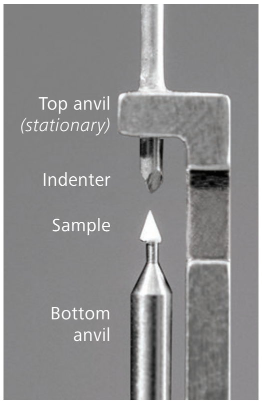

Figure 3. Elephant dentin sample mounted in Xradia Ultra Load Stage. The cone indenter diamond tip is positioned above.

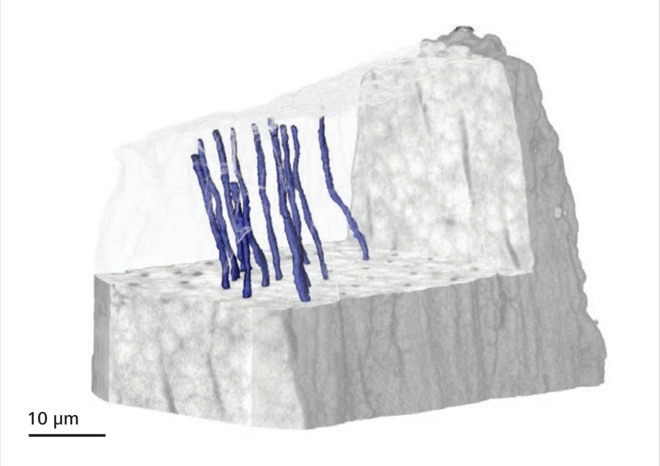

Figure 4. 3D rendering of the arrangement of selected tubules in elephant dentin captured non-destructively by Xradia 810 Ultra X-ray microscope.

Indentation was carried out in a series of stages, and at each stage the sample was held at constant displacement relative to the indenter while a tomography scan was taken. Figure 5 shows the progression of crack growth in the same virtual slice in relation to the measured load-displacement curve.

Zernike phase contrast mode in the Xradia Ultra reveals the crack evolution with high contrast. It is found that the crack propagates largely perpendicular to the tubule axis, as shown in Figure 6.

Conclusions

In situ nanomechanical testing in a nanoscale X-ray microscope provides researchers with a key tool to link the nanoscale 3D structure of a material to measured performance. Materials can be studied in a range of modes, including compression, tension and indentation. The non-destructive nature of X-rays enables tomography scans to be carried out at progressive loading stages, yielding a time-lapse evolution of the microstructure. This ‘4D’ information can be directly related to mechanical behaviour via the measured load-displacement curve. In this study, the indentation mode enabled crack growth in dentin to be studied *in situ* and at resolutions down to 150 nm. The phase contrast mode of Xradia Ultra provided a crucial advantage in enabling the cracks to be revealed with high contrast. Crack growth in the lateral direction appears to follow the mechanism postulated in Ref. 3, which involves micro-cracking at the tubules linking up with the main crack. Obvious crack bridging is observed along the crack path. Future studies will investigate the anisotropic behaviour by using a faceted indenter to generate cracks in specific orientations with respect to the tubules.

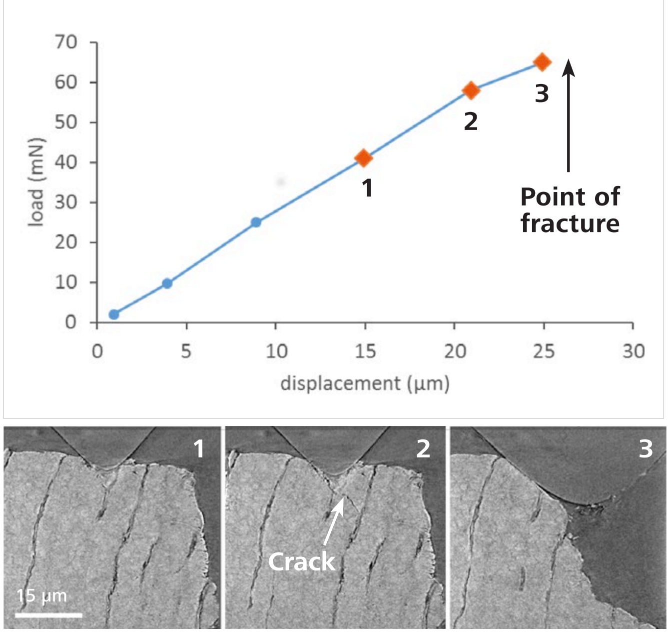

Figure 5. Progressive crack growth in elephant dentin in relation to the measured load-displacement curve. The three slices shown correspond to the last three loading stages, with complete fracture occurring after applying the highest load.

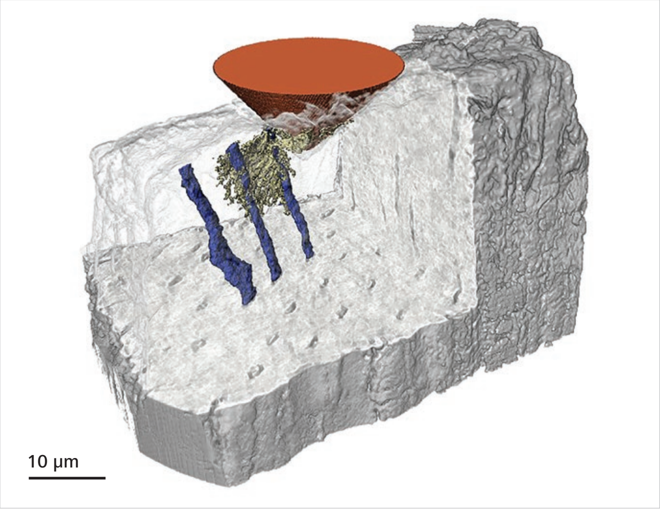

Figure 6. A rendering showing the 3D morphology of a selected crack in relation to neighbouring tubules and the indenter tip.

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