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Hierarchical structure of the spiralled narwhal tusk studied by X-ray imaging

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Narwhals are fascinating animals whose tusk has caught the attention of humankind for centuries and has been associated with the unicorn myth. Narwhals have a vestigial tooth, a tusk, with a left-handed spiral structure that grows during the animal's life span principally in males. This twisted tusk has highly anisotropic mechanical properties and an extraordinarily high impact resistance [3]

We hypothesize that the orientation of the biological nanostructure reflects the spiral macro-structure, which defines the mechanical properties. Using combined 2D and 3D imaging techniques using X-rays and polarised visible light we studied its hierarchical structure at multiple length scales and with different contrasts. Small-angle X-ray scattering and diffraction imaging [3], X-ray fluorescence, and birefringence microscopy were used to investigate the anisotropy of the tusk building blocks (i.e., mineralised collagen fibrils) from the nano to the macro scale. Narwhal tusks have a central pulp chamber surrounded by primary dentine and a layer of cementum. Unlike human teeth, no enamel is present [2]. The building blocks of the narwhal cementum and dentine are identical to other mineralised tissues; however, their degree of anisotropy and spatial distribution differ from, e.g., bone [3] SAXS tensor tomography revealed a strong anisotropy in the dentine with mineralised collagen aligned in the longitudinal direction. A twist in the orientation of the longitudinal fibres was also found with radially increasing angles, creating a helical pattern from the pulp chamber to the cementum layer. The outer cementum layer that covers the entire tusk had a less anisotropic structure with radially oriented domains embedded in a longitudinally oriented matrix. At the nanoscale, the periodicity of the collagen fibrils was slightly higher in cementum than in dentine. The shape and size of the mineral particles were estimated fitting the SAXS and WAXS patterns, where the particle thickness and length increased from the inner to the outer dentine. These results not only reveal that a strong anisotropy is present in the tusk but also suggest that they follow a three-dimensional spiral structure in the nanoscale. The combination of 2D and 3D imaging techniques achieved an optimal balance between field of view, measurement time, and resolution.

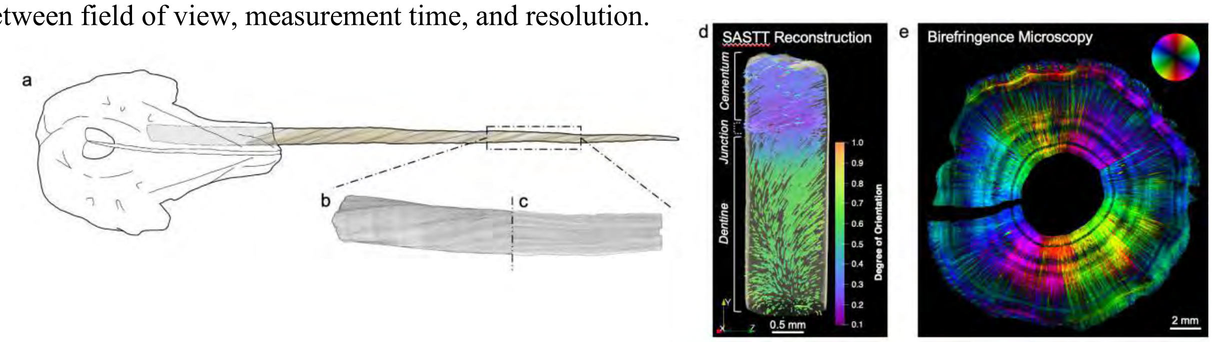

Figure 1. Schematic representation of a narwhal skull with tusk (**a**) and 3D rendered computed-tomography scan of the tusk showing its macroscopic left-handed spiral (**b,c**). 3D representation of the mineralised collagen fibrils' orientation in the narwhal tusk as the streamlines of the orientation of their reconstructed reciprocal space map (SAXS tensor tomography) (**d**). The morphology of the sample was reconstructed using the X-ray transmission signal measured during the experiment. Birefringence microscopy of a transversal slice of the narwhal tusk. The orientation of the mineralised collagen fibres follows the colour wheel where the angle is represented by the hue and the retardance is represented by the value in each pixel (**e**).

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