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Research Paper

Intrafibrillar plasticity through mineral/collagen sliding is the dominant mechanism for the extreme toughness of antler bone

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ABSTRACT

The inelastic deformability of the mineralised matrix in bones is critical to their high toughness, but the nanoscale mechanisms are incompletely understood. Antler is a tough bone type, with a nanostructure composed of mineralised collagen fibrils ~100 nm diameter. We track the fibrillar deformation of antler tissue during cyclic loading using *in situ* synchrotron small-angle X-ray diffraction (SAXD), finding that residual strain remains in the fibrils after the load was removed. During repeated unloading/reloading cycles, the fibril strain shows minimal hysteresis when plotted as a function of tissue strain, indicating that permanent plastic strain accumulates inside the fibril. We model the tensile response of the mineralised collagen fibril by a two – level staggered model – including both elastic – and inelastic regimes – with debonding between mineral and collagen within fibrils triggering macroscopic inelasticity. In the model, the subsequent frictional sliding at intrafibrillar mineral/collagen interfaces accounts for subsequent inelastic deformation of the tissue in tension. The model is compared to experimental measurements of fibrillar and mineral platelet strain during tensile deformation, measured by *in situ* synchrotron SAXD and wide-angle X-ray diffraction (WAXD) respectively, as well as macroscopic tissue stress and strain. By fitting the model predictions to experimentally observed parameters like the yield point, elastic modulus and post-yield slope, extremely good agreement is found between the model and experimental data at both the macro- and at the nanoscale. Our results provide strong evidence that intrafibrillar sliding between mineral and collagen leads to permanent plastic strain at both the fibril and the tissue level, and that the energy thus dissipated is a significant factor behind the high toughness of antler bone.

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1. Introduction

The fracture criteria and inelasticity in ceramics, metals and other synthetic materials are driven at the molecular level by well-defined mechanisms such as dislocation dynamics (Kocks et al., 1975) or grain boundary slip. In contrast, the corresponding situation in structural biological fibre composites is more complex, and is poorly understood, particularly at the supramolecular level of 1–100 nm. Biological structural materials like bone are often hierarchical fibre-composite systems built up by cell-guided self-assembly of structural units from the molecular to the macroscopic length scale (Fratzl and Weinkamer, 2007). Bone can be considered, at the nanoscale, as an anisotropic composite of thin (1–5 nm thick × 30–50 nm wide) platelets interleaved with an organic scaffold of Type I tropocollagen protein molecules (~1.1 nm wide × 300 nm long (Weiner and Wagner, 1998)). The intrafibrillar mineral and collagen together form mineralised fibrils (~50–200 nm in diameter) which, together with extrafibrillar mineral, aggregate into fibril arrays with a small fraction of amorphous (noncollagenous) proteins (Fantner et al., 2005). Fibril arrays, in turn, form lamellae ~5 μm in width (Weiner and Wagner, 1998), which can aggregate into various structures, forming the organ bone.

When bone is subjected to external mechanical forces – as encountered during physiological loading, fatigue, impact and fracture – differential slip, bond breakage and associated complex fracture and crack patterns can occur at each of the above structural levels. For low strains the mechanical response is elastic, but for macroscopic (tissue-level) strains >0.5%, a clear transition to inelastic behaviour is observed, with a reduced tangent modulus (~$1/10^{th}$ the elastic modulus), and residual strain upon removing the external stress (Currey, 2002). Failure of weak interfaces at many length scales can contribute to this inelasticity, but it is not clear which interfaces are the mechanically the most critical, nor which structural interfaces trigger the transition to inelasticity. At larger scales (~1–10 μm lamellae), microdamage (Zioupos et al., 2008) and both crack bridging and deflection (Nalla et al., 2003) are well established as toughening mechanisms, but the corresponding mechanisms (if any) for the fibrillar (<1 μm) length scale is far less understood, and the reason for the transition point of ~0.5% is not clear.

Experimentally, the determination of the deformation mechanisms is complicated by the concurrent structural deformation occurring at several levels in the hierarchy. *In situ* electron microscopy and tomography have shown that the increased length of the cracks, and partial bridging of opened cracks by ligaments at the microscale act as an extrinsic (material-independent) toughening mechanism (Koester et al., 2008). At the nanoscale, fewer experimental data on the precise mechanisms are available, although scanning probe microscopy has supported the role of weak, re-formable bonds in a noncollagenous protein "glue" between fibrils during bone fracture (Fantner et al., 2005). Understanding the mechanism is essential because a large part of the interest these structural biomaterials like bone and mollusc shells have attracted recently is due to their extreme toughness through nano- and microscale structural design (Meyers et al., 2008). Their high work to fracture has inspired the design of synthetic man-made bioinspired composites to replicate this exceptional mechanical performance (Bonderer et al., 2008; Tang et al., 2003). However, as the inelasticity mechanism is still not conclusively known, the emulation designs remain partly empirical.

Table 1 – Comparison of work to fracture and elastic moduli for antler versus bone, in wet and dry conditions (data taken from Currey et al. (2009).

Tissue	Type	Elastic modulus (GPa)	Work to fracture (kJ/m^2)
Antler:	Dry	17.5	23.4
	Wet	7.3	31
Bone:	Dry	21.3	–
	Wet	19.8	9.6

Antler is an exceptionally tough bony tissue, whose mineral content is the lowest (volume fraction Φ~0.30 (Currey, 2002)) of common bone types. It is an annually regenerated and rapidly growing tissue, consisting at the tissue level of a cortical shell and a spongy inner core (Krauss et al., 2011). It is found, except in reindeer, only in the males of deer, and antlers are used in male dominance fights for access to females in the rutting season. Its toughness and strain to failure are much higher (by a factor of 2–3 (Currey, 2002; Currey et al., 2009)) in comparison with more common bone types (Table 1), and fracture mechanics tests have shown (Launey et al., 2010) crack propagation in antler to arise largely from intrinsic (material-dependent) mechanisms, instead of the extrinsic (material-independent) mechanisms such as crack bridging typical in more highly mineralised bone (Koester et al., 2008). We have previously proposed that, in tension, one component of these intrinsic material mechanisms is an interfibrillar debonding (at the scale of ~100 nm) that starts during inelastic deformation (Krauss et al., 2009). However, several major questions remain unanswered about the inelasticity mechanisms of antler tissue.

In this paper we report direct experimental quantification of the fibrillar and subfibrillar deformation during macroscopic inelastic deformation in the deer antler. We use the observed results to develop a nanoscale structural model of the plastic deformation of the mineralised collagen fibril during macroscopic inelasticity, which is used to predict the elastic modulus, yield point, and full macroscopic stress/strain behaviour and can be extended to more highly mineralised bone types. Specifically, we were aiming at building a model for the macroscopic strain behaviour to answer several open questions as summarised below:

- We previously inferred a nanoscale toughening mechanism of *inter*fibrillar debonding at the fibril array level (Krauss et al., 2009), based on the observation of an inhomogeneous deformation at the fibrillar length scale by X-ray diffraction, but the concurrent *intra*fibrillar processes were not investigated.
- The fibrillar level response to (a) cyclic loading and reloading, and (b) the extent of plastic fibril deformation (if any) has not been reported experimentally.