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The Effects of Cross Linking on Collagen Type I Nanostructure and Nanostructural Response to Uniaxial Tension

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Abstract

Collagen type I, is a fibrillar protein with a complex hierarchical structure, forming the extracellular matrices of an extensive range of organs and tissues. Applications for treated collagen materials vary vastly from commercial uses to the medical field for bioprosthesis and tissue grafts. Glycosaminoglycan (GAG), cross links naturally bridge fibrils, whilst glutaraldehyde is widely used as a synthetic linking agent in medical and other industries. No consensus has been reached regarding what contribution, if any, such cross links have on collagen structure and mechanical responses to applied stresses. This research investigated the role of GAG and glutaraldehyde cross links on the nanostructure and nanostructural response of type I collagen fibrils under uniaxial strain. Bovine pericardium was decellularised, producing native samples, or further treated with glutaraldehyde or chondroitinase ABC to produce glutaraldehyde cross linked or GAG-depleted collagen samples respectively. Synchrotron small angle X-ray scattering (SAXS), and atomic force and polarised light microscopy provided quantitative and qualitative information on collagen nanostructure. Uniaxial tensile experiments in conjunction with SAXS were performed to monitor structural changes with applied strain. Glutaraldehyde cross links constrained fibrils into more networked isotropic structures and demonstrated a mechanical function, recruiting 45% of fibrils into stretching which experienced strains of up to 6.4%. Comparison of native with chondroitinase ABC-treated samples showed GAGs do not constrain fibrils into alignment and have potential fibril lubricating effects; 12% of fibrils in native tissue experienced strains up to 4.1%, and 36% of fibrils experienced strains up to 4.6% in the GAG-depleted tissue. A higher degree of fibril sliding occurs in native tissue. Interestingly, whilst adult pericardia are more cross linked and fibrils of neonatal pericardia are more aligned, both tissues share similar propensities to form more isotropic structures with glutaraldehyde treatment. These findings build a comprehensive picture as to the function cross linking has in collagen structure and mechanical response at the nano-level, where such knowledge may prove useful for the preparation of collagen materials for specific applications.

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