Quantifying intervertebral disc inter-lamellar and inter-bundle mechanics

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Introduction: Intervertebral disc is the key element of spine flexibility; the annulus can resist high and diverse mechanical loadings, but can be severely affected by small defects such as needle puncture. Previous studies concentrated their efforts on evaluating lamellar mechanics, but there is still limited understanding of the interlamellar interactions. Cells are present in the interlamellar space and therefore their viability can be strongly affected by this mechanical environment.

Aim: To quantify the intra- and inter-lamellar strain of intervertebral disc outer annulus under tension.

Materials and Methods: Twenty-one cow tail intervertebral discs, obtained from a local abattoir, were measured in this study. Discs were carefully dissected in order to expose the outer annulus (OA) and detach both endplates; the nucleus and part of the inner annulus were also excised, leaving a ring of OA. Samples were mounted on a micro-testing device equipped with two flat hooks to load the OA in circumferential tensile strain. A tension test was performed to 5% strain in 1% strain steps. The sample was imaged during this test with a confocal microscope fitted with a 10x/0.4NA air objective (Olympus UPlanS Apo) and an 800-nm mode-locked femto-second Ti:Sapphire laser. This excites second harmonic generation in the sample, thus allowing visualization of collagen fibres in regions of interest (ROI) of approximately 0.5 x 0.4 mm. The displacement field was measured in the ROI, and it was used to measure linear and shear strain within a lamella, between lamellae and between bundles.

Results: Out of 21 samples, seven showed two adjacent lamellae and nine showed well-separated fibre bundles with the same fibre alignment; the remaining five appeared homogeneous. Fibres formed two groups with average inclinations of 23° ± 13° and -19° ± 11° relative to the disc transverse plane. Maximal local circumferential strain (1.7 %) was much smaller than the imposed 5 % strain. Strain was concentrated between lamellae and between bundles, rather than intra lamellar: final intra-lamellar linear strain (at 5% macroscopic strain) was 1.2% (range [0.3% – 3.4%]) while intra-lamellar shear was 2.2° [0.3° - 6.6°]. Final linear strain between lamellae was 2.1% [0.2% – 7.6%] while shear was 0.7° [0.2° - 1.6°]. Final linear strain between bundles was 3.7% [0.4% - 9.4%] while shear was 2.9° [0.8° – 10.5°].

Conclusion: Strain between lamellae and between bundles was between 1.4 and 3.2 times higher than within the lamella, apart from inter-lamellar shear strain which was lower than intra-lamella. Lower intra lamellar strains could be expected, because lamellae are strong fibre reinforced structures. However, to our knowledge, this is the first time that this behaviour has been quantified by direct observation of the collagen network during loading. These high strain concentrations, with peaks of 10.5° shearing and 7.6% straining between lamellae, suggest that the catastrophic effects of local injury may originate in the inter-lamellar and inter-bundle spaces.