MICROMECHANICAL EFFECTS OF NEEDLE INJURY TO ANNULUS FIBROSUS

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Introduction

Diagnostic discography has become a controversial imaging technique to assess the intervertebral disc; on the one hand, it gives access to disc morphology and damage status, but on the other hand the needle injury necessary to inject contrast material is thought to cause or accelerate disc degeneration [1-2]. While discography might be abandoned, delivery of repair substances into the disc (hydrogels, cells, growth factors, etc.) is becoming increasingly investigated and relies equally on needle insertion. A better understanding on the effects of needle injury is therefore required to develop procedures to minimize the damage.
In this work, the effect of needle injury on the annulus fibrosus *in vitro* was quantified in cow tail disc, and compared to a control group that did not undergo puncture.

Methods

Twenty-five strips of outer *annulus fibrosus* (average size: 13mm x 5.8mm x 2.5mm thickness) were cut from thirteen cow tail intervertebral discs obtained from an abattoir. Samples were mounted in a custom-made mechanical testing rig and loaded in 1% strain steps at 1mm/min up to 5% strain. Samples were unloaded, punctured with a needle (gauges between 19 and 23) and loaded again with the same protocol. Samples were randomly assigned to two groups: fifteen samples were punctured while ten underwent the same protocol without puncture (control group). Load was measured with a 20 N load cell; elastic modulus was calculated at each strain step from the stress-strain curves.
The rig was mounted on a microscopy set up for second harmonic generation (SHG) [3], which revealed collagen structure with sub-micron resolution. Intra-bundle and inter-bundle strains were evaluated by analysis of a series images at increasing strains. Differences were analyzed with Mann-Whitney non-parametric test (α – 0.05).

Results

Figure 1 shows a typical example of puncture site; bundles of fibers can be seen bending around the hole.
The elastic modulus increased with strain. The modulus during the second loading was lower at low strain than during the first loading in both groups ($p < 0.05$), but it was similar higher strains. At 5 % applied strain, modulus of the puncture group was only reduced by 4.4 % (compared to the first loading), while it was increased by 7.3% in the control group. This difference was significant ($p = 0.007$). Inter- and intra-bundle strains, as well as inter- and intra-bundle shear strains did not significantly differ between loadings nor between groups.

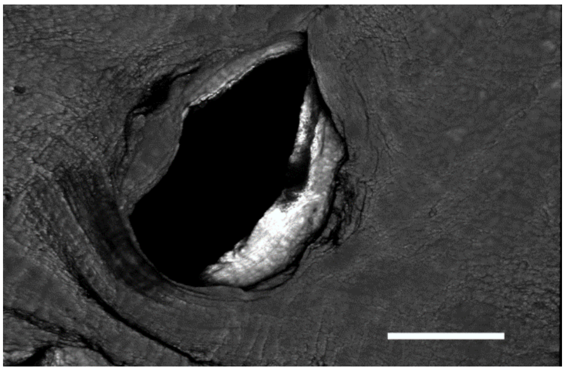

Figure 1: puncture site (scale bar is 100 μm).

Discussion

Needle puncture had a modest impact on the macro- and micromechanics of annulus fibrosus. This is surprising, especially considering that the needle diameters (0.6-1.1 mm) were relatively large compared to the width of the annulus strips (5.8 mm).
It was previously reported that the puncture has a significant mechanical effect in its immediate vicinity [4]. Indeed, SHG imaging showed that the damage was limited to the puncture (Figure 1).
In the present work, imaging of microstrain was performed at a distance of about 3-4 mm from the puncture site. The modest effects observed at this medium distance suggest that the disc degeneration induced by needle puncture might not be initiated by a disc-wide mechanical effect, but is secondary from local tissue damage, for instance a biochemical effect of local cellular death. Future work should include the analysis of the mechanical effects of puncture on the nucleus pulposus as well.

References

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