Deformation induced chondrocyte damage

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Mark van Turnhout[1], Stefan de Vries[2], René van Donkelaar[2] & Cees Oomens[1]
Eindhoven University of Technology, Department of Biomedical Engineering.
[1] Soft Tissue Biomechanics and Engineering, [2] Orthopaedic Biomechanics.

1. Introduction

As collaborators in the NIBIB/NIH project 'Predicting cell deformation from body level mechanical loads' we are investigating mechanical chondrocyte damage thresholds. Thus, we wish to know how much deformation a chondrocyte can endure, and for how long, before it damages (dies).

2. Methods

Bovine chondrocytes are cultured in 3-D agarose constructs. These constructs are subjected to nonuniform compression at a confocal microscope (figure 3). At the microscope, we can image living and dead chondrocytes with fluorescent CTG/PI staining in time and space [1, & figure 1].

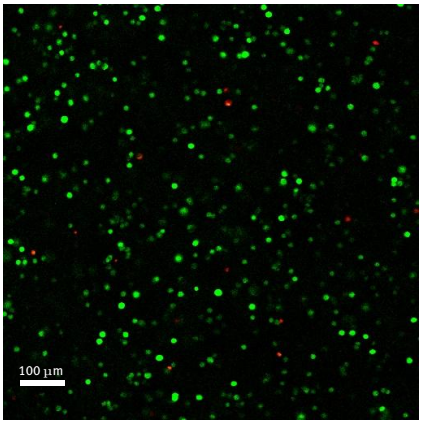

Figure 1: Example of live (CTG, green) – dead (PI, red) staining of the chondrocytes, as imaged with the CLSM.

3. Results

Experiments were performed after 1, 2, and 3 weeks of culturing. Axial compression levels at the centre of the construct were 25 % and 35 %. The fraction of dead cells was monitored before compression (t^-), immediately after compression (t^+) and after compression for 13 hours $(t+13)$ and 64 hours $(t+64)$ (figure 2).

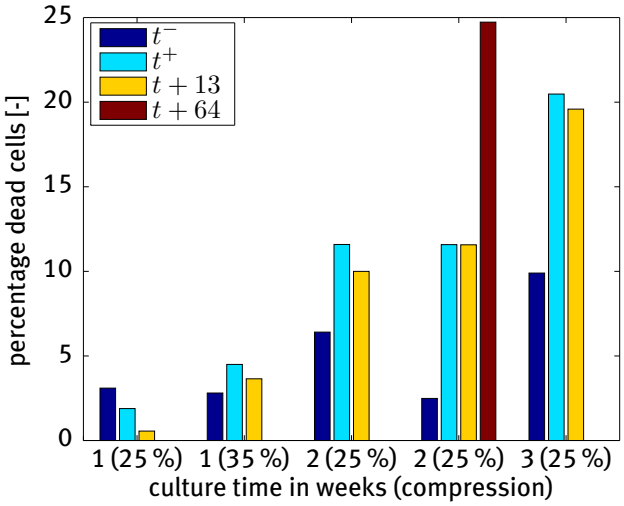

Figure 2: Percentage dead cells at the construct symmetry axis in five experiments before compression (t^-), immediately after compression (t^-), and long after compression $(t+13, t+64)$.

4. Conclusions

- either we can resurrect cells by compression, or we have to critically evaluate our methods and analysis
- longer culture times appear to increase the cells' vulnerability to deformation
- patience is a virtue if you wish to see substantial chondrocyte death in young samples at 25 % compression

References

[1] D. Gawlitta *et al.* (2004). Evaluation of a continuous quantification method of apoptosis and necrosis in tissue cultures, *Cryotechnology*, **46**:139–150.

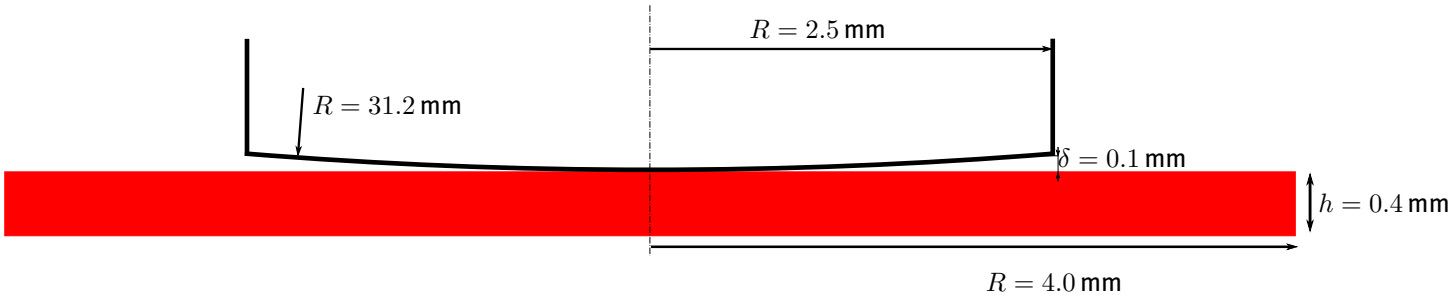

Figure 3: Chondrocyte deformation is induced by deformation of the agarose construct (red). We use an indenter with an spherical end to obtain a variety of deformations in a single experiment. Figure is to scale.