
SELECTED ABSTRACTS DELIVERED AT THE 9TH ANNUAL AOSPINE NORTH AMERICA FELLOWS FORUM

Consistent with EBSJ's commitment to fostering quality research, we are pleased to feature some of the most highly rated abstracts from the 9th Annual AOSpine North America Fellows Forum in Banff, Canada. Enhancing the quality of evidence in spine care means acknowledging and supporting the efforts of young researchers within our AOSpine North America network. We look forward to seeing more from these promising researchers in the future.

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Maintenance of cell viability in axially loaded intervertebral disc organ culture

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INTRODUCTION

Whole disc organ culture is needed for preclinical testing of biological repair of the degenerate intervertebral disc. Such organ culture is hampered by two major limitations: first, obtaining adequate nutrition through the calcified cartilage end plates and adjacent vertebral bone; second, by loss of tissue integrity if the end plates are removed from the discs. In this work we use a bioreactor and a recently described technique for whole disc isolation that overcomes these problems.

METHODS

Six bovine caudal discs were isolated by parallel bone cuts close to the cartilage end plates, and the bone and the adjacent calcified part of the cartilaginous end plate was removed (CEP discs). The explants were maintained in specific culture media which were changed twice weekly. The specimens were initially cultured for 7 days without applied external load. The discs were then dynamically loaded for 19 consecutive days using our bioreactor. It consists of a culture chamber in which the disc can be dynamically loaded in a uniform manner. The culture chamber can accommodate disc up to 60 mm in diameter. The discs are loaded in the culture chamber by upper and lower platens, which conform to the shape of the remaining cartilaginous end plate and permit fluid flow across its surface. Load displacement behavior was monitored throughout the culturing period for five discs because of exclusion of one contaminated disc. Cell viability was evaluated on termination of the experiment in a 1 mm slice through the center of the disc, visualized using a live/dead fluorescence assay (Live/Dead[®], Invitrogen) and confocal microscopy.

No conflict of interest.

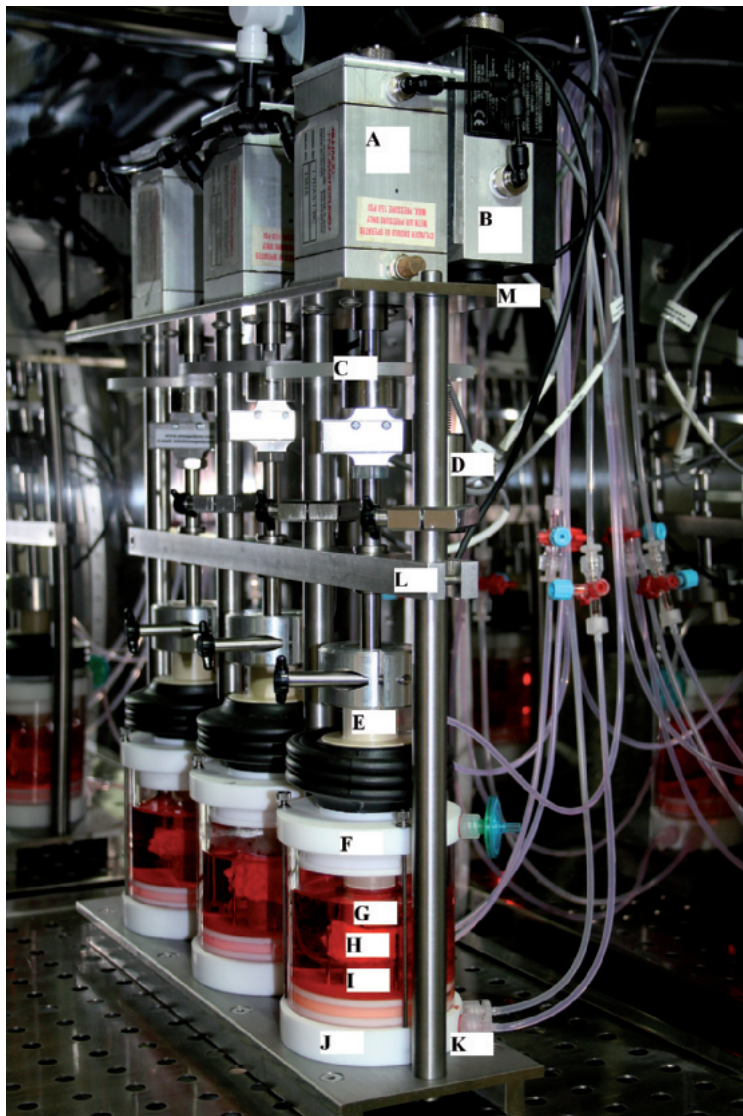
RESULTS

We have to date cultured bovine discs for 4 weeks under cyclic physiological loading, with maintained cell viability. The morphological changes for the discs are the following: on average the discs decreased 7.7% in height, increased 5.9% in diameter, and increased 2.5% in weight compared with the time of isolation. The proportion of live-to-dead cells was analyzed in the center of NP and AF. A viability of $88.4\% \pm 3.9$ was observed in the NP and $80.8\% \pm 8.9$ in the AF after a total of 4 weeks in culture.

CONCLUSIONS

Using a novel disc preparation and isolation method along with an axially loading bioreactor, we were able to culture bovine caudal discs for a total of 28 days. After this time the discs exhibited excellent cell viability for NP and AF. The bioreactor culture system is ideally situated for studying intervertebral discs in an in situ environment. The culture system also provides an experimental platform where numerous parameters can be evaluated, eg, O_2 tension, glucose levels, and the effect of growth factors. As such, it is equally useful for studying the role of load in inducing disc degeneration or the role of biological stimuli in restoring disc function.

Fig 1 The three bioreactor units that form the bioreactor system; A–E, loading unit; F–K, culture chamber unit.



- A Rolling diaphragm linear actuator
- B Proportional pressure control valve
- C LVDT touch plate
- D Linear variable displacement transducer (LVDT)
- E Plunger specimen contact system
- F Top plate with vent
- G Upper platen end plate
- H IVD specimen location
- I Lower platen end plate
- J Base plate
- K Flow adaptor
- L Anti-shear bracket
- M Bioreactor unit frame