Reviewer's report

Title: Bovine Explant Model Of Degeneration Of The Intervertebral Disc

Version: 1 Date: 11 November 2007

Reviewer: Stephen Ferguson

Reviewer's report:

General

The authors have presented a study of chemonucleolysis in an in vitro organ culture model. Following injection of trypsin or papain into bovine coccygeal disc explants, alterations to the disc were observed following three weeks of incubation. Nuclear disruption and cavity formation was evaluated by injection of a radioopaque marker into the disc and subsequent radiographic imaging. Disc water and proteoglycan content were measured. Histomorphological evaluation of disc quality, structure and cellularity was made. The authors concluded that they have established an in vitro bovine disc explant model of disc degeneration.

For me, the most interesting finding of this study is the description of the cleft formation in the nucleus, which provides an in vitro analogue for the internal disruption found during natural disc degeneration. As such, the paper is a well-written technical note which provides support for one potential method of inducing degeneration in an in vitro organ culture model.

The analysis of the thus-created "degenerated" disc is rather simplistic, relying on biochemical measurements of GAG content and hydration and visual analysis of overall disc structural competence and health. There are no quantitative measures of cell viability, therefore it is difficult to determine whether or not the model is truly useful for long-term studies, or at least to compare to the many other organ culture models described in the literature. The analysis considers only one aspect of disc degeneration, the end-point structural and biochemical changes (and also does not consider any changes to collagen). Also important, and worth at least discussing, is the relevance of alterations to the expression of anabolic and catabolic genes or the production of catabolic enzymes and enzyme inhibitors, which have been studied in other in vitro and in vivo models of disc degeneration.

Nevertheless, this is an interesting, self-contained and well-written study which provides justification for the use of enzymatic digestion to produce a compromised disc in an in vitro environment. Future studies should build on this first work to fully establish the validity of the model. The manuscript stands on its own as a technical note, although the authors should qualify the title message by highlighting that the model reproduces only one aspect of disc degeneration. The manuscript could be improved by extending the discussion, as suggested below.
Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

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Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

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Discretionary Revisions (which the author can choose to ignore)

The authors should comment on the limitations of the histological determination of disc cell health ("the disc cells themselves appeared normal") as an evaluation of the validity of an organ culture model.

The importance of other outcome measures (e.g. cell viability, total cellularity, immunohistochemistry, qRT-PCR) for an in vitro model of disc degeneration should be discussed and compared to their chosen evaluation methods.

I do not entirely agree that the use of an intact disc absolutely precludes evaluation of injectable therapies in vitro. The authors have been able to inject a certain amount of enzyme (the absolute volume was not reported, just the concentration). For GF therapy, small injection volumes would be adequate. I agree that larger volumes would be required for injection of cells & carriers, or synthetic augmentation materials. Perhaps this could be further discussed in the manuscript.

Enzymatic digestion of the nucleus is only one method for inducing a degenerative state in vitro. Other possibilities which have been reported include mechanical overload, impact loading, osmotic loading, hydrostatic over-pressure and diminished nutrient supply. The proposed model should be compared and contrasted with the other models.

What next?: Accept after discretionary revisions

Level of interest: An article whose findings are important to those with closely related research interests

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

I declare that I have no competing interests