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Ex Vivo observation of human nucleus pulposus chondrocytes isolated from degenerated intervertebral discs

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Study Design: We performed an *ex vivo* study to observe cell morphology and viability of human nucleus pulposus (NP) chondrocytes isolated from degenerated intervertebral discs (IVD).

Purpose: To better understand the biological behavior of NP chondrocytes in monolayer cultures.

Overview of Literature: Biological repair of IVDs by cell-based therapy has been shown to be feasible in clinical trials. As one of the most promising transplanting seeds, how the isolated NP chondrocytes behavior *ex vivo* has not been fully understood.

Methods: Human NP chondrocytes were harvested from 20 degenerated IVDs and cultured in monolayers. Histological and immunochemistry staining was used to detect cell morphology change. Cell viability was studied by analyzing cell cycle distribution and apoptotic rate in the primary and subcultured cells.

Results: The round or polygonal primary NP chondrocytes had an average adherence time of 7 days and took nearly 31 days to reach 95% confluence. The spindle-shaped P1 NP chondrocytes increased growth kinetics and took about 12 hours to adhere and 6.6 days to get 95% confluent (Figure 1). Immunochemistry staining of collagen II was positive in the cell cytoplasm. Nearly 90% of the confluent NP chondrocytes stayed in G1 phase while 16% underwent apoptosis (Table 1). No significant difference of the collagen II expression, cell cycle distribution or the apoptosis indices were detected between the primary and subcultured NP chondrocytes.

Conclusions: Human NP chondrocytes undergo significant morphological change in monolayer cultures. Cell cycle distribution pattern and apoptosis index of the cultured NP chondrocytes potentially influence their clinical transplantation or laboratory use.

Feng Wang has completed his MS degree at the age of 27 years from Southeast University and is doing doctoral studies from Southeast University School of Medicine. He has published 6 papers about the pathogenesis of intervertebral disc degeneration. His laboratory is investigating the phenotype changes of senescent disc cells and how cellular senescence influences the degenerating process of intervertebral disc.