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Deformation is detrimental to cartilage matrix formation by expanded chondrocytes

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Purpose: After transplantation in-vivo, chondrocytes are subjected to deformation due to loading. Before transplantation, expansion in culture is mostly required to obtain sufficient cell numbers. The subsequent shift towards a more fibroblast-like phenotype might alter the cell’s response to deformation. We investigated the effect of expansion culture of chondrocytes on their response to deformation.

Methods and Materials: Articular chondrocytes were harvested from OA cartilage and either seeded directly or after expansion for 3 passages, on 6-wells flexible plates at a density of 300,000 cells/ well. After 5 days pre-culture, cells were stretched for 3 days using a modified Flexcell (0.5% or 3.0% uniaxial strain at 0.5 Hz) for 24 hours per day (with one hour rest period in-between) or left unstretched as control. Real-time RT-PCR was performed on aggrecan, collagen I, collagen II and proteoglycan 4 (PRG4, lubricin, superficial zone protein), SOX9, MMP3, MMP1 and MMP13. Results: In primary chondrocytes, collagen I gene expression was down-regulated in response to mechanical deformation. Expression of all other genes was not significantly altered by 0.5% or 3.0% strain. In dedifferentiated chondrocytes, strain drastically down-regulated the expression of aggrecan, collagen I and II and PRG4. SOX9, MMP3 and MMP3 were expressed higher in response to strain. Strain in combination with redifferentiation (chondrogenic) medium gave intermediate response to strain.

Conclusions: In conclusion, these results indicate that deformation of culture expanded chondrocytes is detrimental to formation of a functional extracellular matrix. As a consequence, second generation ACT procedures require external factors to protect cells from being deformed before being redifferentiated.

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Synovial fluid proteoglycans in assessment of joint degeneration severity

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Purpose: To assess cartilage tissue metabolism by virtue of synovial fluid biochemical analysis

Methods and Materials: We studied articular cartilage and synovial fluid from patients with different grades of idiopathic arthritis and OAI in 38 patients with acute injury of knee parts and ligaments without cartilage lesion. Proteoglycans of synovial fluid and of cartilage were analyzed after their fractionation into hyaluronic acid, aggregated and unaggregated proteoglycans. This allowed determining their qualitative composition of glycosaminoglycan chemical components and assessing degenerative process intensity.

Results: As degeneration progresses, the quantitative and qualitative composition of glycosaminoglycan chemical components also changes, as well as hyaluronic acid polimery and its ability to aggregate with proteoglycans. Quantity and qualitative changes of proteoglycans in synovial fluid can be used as a diagnostic test to assess degenerative process intensity.

Conclusions: In conclusion, these results indicate that deformation of cartilage matrix formation by expanded chondrocytes is detrimental to formation of a functional extracellular matrix. As a consequence, second generation ACT procedures require external factors to protect cells from being deformed before being redifferentiated.