Human Osteoarthritic Clones Represent a Proliferative Population of Mesenchymal Progenitor Cells

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Introduction: Osteoarthritis affects nearly 27 million people in the United States alone. Current treatment options for OA range from pain management to total joint replacement surgery. There are currently no drugs on the market that are able to slow the progression, or reverse the effects of osteoarthritis. This necessitates a better understanding of the biology of osteoarthritis, which will lead to the development of better treatment options for OA.

Advanced human OA is characterized by loss of extracellular matrix, fibrillated cartilage, and clusters of chondrocytes called “clones”. There is variability among the types of chondrocytes that make up these clones. Some are characterized by an increase in proteoglycan staining and matrix synthesis while others are characterized by decreased proteoglycan staining and phagocytosis of matrix components. Some researchers describe clones consisting of large, rounded chondrocytes with a euchromatic nucleus and other describe clones consisting of secretory cells and degenerating chondrocytes. There is also some debate as to whether chondrocytes are able to migrate and regroup into these clones, or if they are a result of chondrocyte proliferation.

In this study, we aim to further characterize the properties of the chondrocytes that compose osteoarthritic clones. We provide evidence that these clones represent a population of mesenchymal progenitor cells that have proliferative potential. We also show that the transcription factor Runx1 is present in these chondrocyte clones.

Materials and Methods: Knee joints were obtained from osteoarthritic patients with varus malalignments undergoing total knee replacement surgery (TKR) at the time of surgery in accordance with the policy on discarded samples at the University of Massachusetts Medical School. Full thickness articular cartilage samples were removed from the tibial plateau and both the medial and lateral condyles of the femur. Samples for immunofluorescence were fixed in freshly prepared 4% paraformaldehyde in cacodylic buffer dehydrated through a series of graded ethanol to xylol (Fisher Scientific) and then embedded in paraffin for sectioning. Immunofluorescence was performed using antibodies to AML-1 (Cell Signaling Technologies), Ki-67 (Abcam), PCNA (Cell Signaling Technologies), and VCAM-1 (Santa Cruz).

Results:

Human Osteoarthritic Clones Express VCAM-1, A Marker for Mesenchymal Progenitor Cells

In bovine articular cartilage, and in a mouse model of osteoarthritis, our group has shown that Runx1 is expressed in the same population of cells as VCAM-1, which is a marker for mesenchymal progenitor cells. To address the possibility that human OA clones represent a population of mesenchymal progenitor cells, we stained sections from human OA cartilage with an antibody to VCAM-1. We show here that chondrocytes in human OA clones express VCAM-1, indicating that they are a mesenchymal progenitor cell population (Figure 1).

Select Human Osteoarthritic Clones Demonstrate Proliferative Potential

Previously, our group has shown that Runx1 is expressed at higher levels in osteoarthritic cartilage when compared to normal adult articular cartilage. Immunofluorescence analysis shows that, in osteoarthritic cartilage, Runx1 is localized to chondrocytes in OA clones. In a mouse model of osteoarthritis, immunohistochemical data suggests that Runx1 is associated with proliferating chondrocytes. To test whether chondrocytes in OA clones represent cells that had proliferated, we looked for presence of the proliferation markers Ki-67 and PCNA within human OA cartilage. We show that some, but not all OA clones are Ki-67 positive (Figure 2) or PCNA positive (Figure 3), indicating that these cells have proliferative potential.

Discussion: We have characterized the proliferative state of the chondrocyte clones that are characteristic of advanced human osteoarthritis. We show that these cells express the transcription factor Runx1, the markers of proliferation Ki-67 and PCNA, and a marker of mesenchymal progenitor cells, VCAM-1. Immunofluorescence analysis of non-OA cartilage from the lateral side of the same joint does not show chondrocytes that are VCAM-1, Ki-67, PCNA, or Runx1 positive. We hypothesize that this population of chondrocytes found in OA clones could be of therapeutic value and represent a source of chondrocyte committed mesenchymal progenitor cells. Our studies indicate that the fibrillated cartilage found in osteoarthritis is a rich source of an expandable population of chondrocytes that could be used for research and tissue engineering.

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