Effects of Oxygen Tension on the Chondrogenic Potential of Differentially Cultured Human Adipose-derived Stem Cells.

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Introduction
Cartilage has limited capacity for self repair. Cell therapy using adult multipotent mesenchymal stem cells (MSC) has therefore been considered promising for the treatment of cartilage defect. Whilst bone-marrow derived MSC have been extensively used for tissue engineering, adipose tissue has recently been contemplated owing to its accessibility and its MSC content. Articular cartilage is an avascular tissue, therefore, oxygen tension has been suggested as a regulatory factor for chondrogenic differentiation. In this context, our work aimed at deciphering the influence of oxygen tension on the in vitro and in vivo chondrogenic differentiation potential of human adipose-derived stem cells (hATSC) when associated with a cellulose based self-setting hydrogel (Si-HPMC).

Materials and Methods
hATSC isolated from lipoaspirates were characterized for their proliferation, surface markers expression and multipotency. The effects of hypoxia on the in vitro chondrogenic potential of hATSC were sought. To decipher the effects of hypoxia on the in vitro chondrogenic potential of hATSC, cells were cultured within Si-HPMC in control or chondrogenic medium and under normal (21%) or low (5%) oxygen tension. Chondrogenic differentiation was assessed by real time PCR for the expression of chondrogenic markers (COL2A1 and ACAN). To determine whether in vitro differentiated hATSC allow cartilage formation in vivo, cells induced in monolayer culture were associated with Si-HPMC hydrogel and implanted subcutaneously in nude mice for 5 weeks. Cartilage formation was evaluated by a histological scoring of nodule density as well as intensities of Alcian Blue staining and Type II collagen immunostaining (score from 0 to 5 for each criterion).

Results
Our data demonstrate that hATSC exhibited proliferation and self-renewal abilities. hATSC also expressed typical stem cell surface markers. In addition, they were able to differentiate towards the chondro-, osteo- and adipogenic lineages. Real-time PCR analyses indicated that, in vitro, hypoxia induced an increased expression of chondrogenic markers as compared to normoxia. Besides, hATSC cultured in chondrogenic medium and under hypoxia have greater ability to form a cartilaginous tissue in vivo as evidenced by the increased presence of sulphated GAG and type II collagen.

Discussion and Conclusions
These data could help us exploit the potential of stem cells to restore the function of degenerated cartilage by cell-based regenerative medicine.

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Disclosures
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