

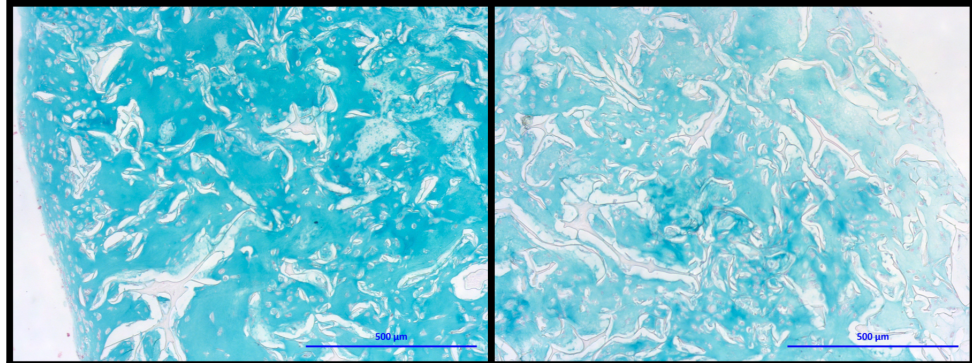
UCB-MSCs

BT

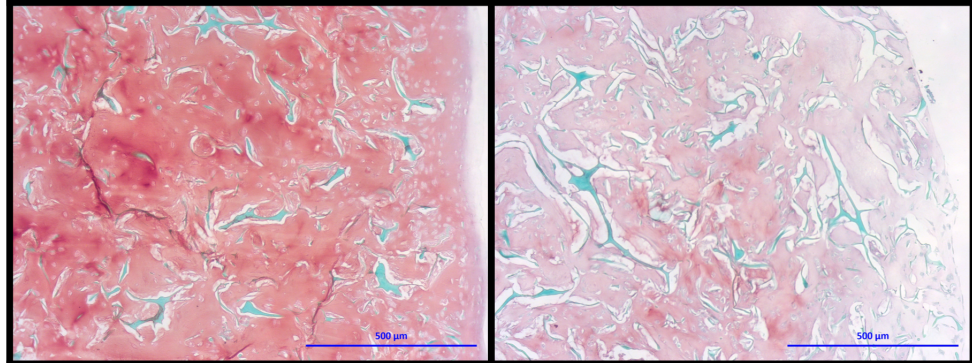
Normoxia

Hypoxia

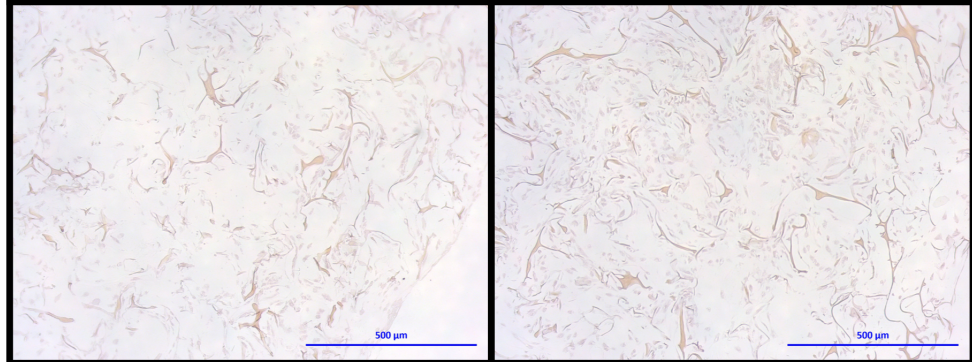
Alcian
blue



Safranin-O



Alizarin
red S



Hypoxia influence on the nature of the matrix newly synthesized by UCB-MSCs under BT treatment.

UCB-MSCs were cultured in type I/III collagen scaffolds at 21% or 3-5% O₂ during 28 days in ICM supplemented with BMP-2 (50 ng/ml) and TGF-β1 (10 ng/ml). The qualitative nature of the ECM was evaluated by alcian blue (sulfated glycosaminoglycans), O-safranin (sulfated proteoglycans) and alizarine red S stainings (extracellular matrix calcification). A representative example from three samples for UCB-MSCs are shown (magnification: ×10, scale bar: 500 μm).

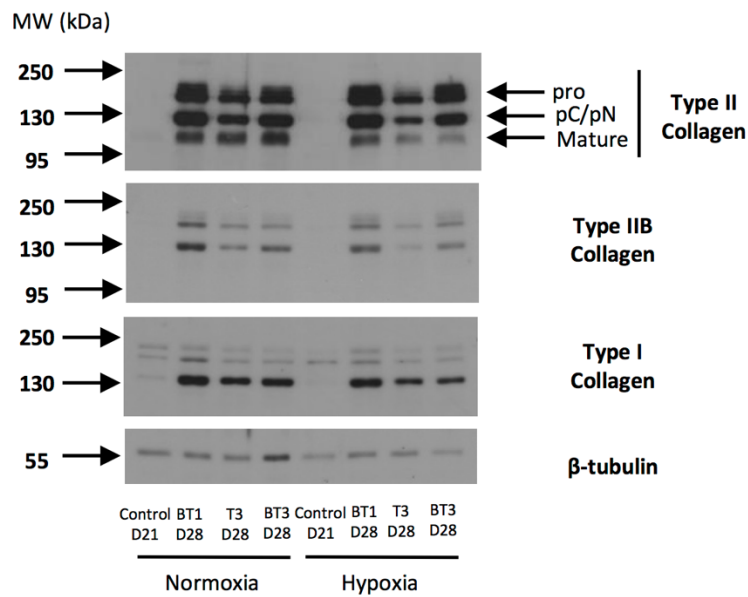


Figure. Comparison between oxygen tension and TGF- β 1 or TGF- β 3 in addition to BMP-2 on protein expression of equine umbilical cord blood-derived mesenchymal stem cells (eUCB-MSCs) differentiated into chondrocytes. eUCB-MSCs at passage 4 were cultured in type I/III collagen sponges for 21 (D21) and 28 days (D28) in normoxia or in hypoxia, in the absence (control), or in the presence of 50 ng/ml of BMP-2 and 10 ng/ml of TGF- β 1 (BT1) or 10 ng/ml of TGF- β 3 (T3) or BMP-2 and TGF- β 3 (BT3). Protein extracts were analyzed in western blots for type II, type IIB and type I *versus* β -tubulin. Representative blots are shown of two independent experiments. Type II collagen shows different levels of maturation forms such as type II procollagen (pro), with only C- or N-terminal propeptides (pC/pN) and the doubly cleaved form (mature form). MW: Molecular Weight, kDa: kilodaltons.