

# **Cobalt chloride supplementation differently affects human mesenchymal stem cells isolated from dental pulp, umbilical cord and adipose tissues in their chondrogenic potential**

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Articular cartilage is an avascular tissue without innervations, characterized by low cell density and abundant extracellular matrix (ECM). These characteristics leave articular cartilage with very limited capacity of repair and regeneration.

Multipotent stem/stromal cells (MSC) are considered promising for cartilage tissue engineering. Stem cells are resided in a special microenvironment known as the stem-cell niche, characterized by the presence of low oxygen concentration.

Previous studies have reported that hypoxic conditions could enhance the chondrogenic differentiation of mesenchymal stem cells in the presence of an inductive medium. Cobalt chloride (CoCl<sub>2</sub>) imitates hypoxia in vitro by preventing hypoxia-inducible factor-alpha (HIF- $\alpha$ ) from being destroyed by oxygen. However, the long-term hypoxic culture of stem cells is difficult and requires special attention to avoid cell death due to cobalt treatment.

In this study we investigated if CoCl<sub>2</sub> affected MSCs isolated from dental pulp, umbilical cord and adipose tissue in their potential to differentiate toward the chondrogenic phenotype.

Cells were treated with concentrations of CoCl<sub>2</sub> ranging from 50 to 400  $\mu$ M. Cell proliferation, mRNA expression of stem-cell marker and chondrogenic associated genes were analyzed by RT-PCR and Real-time PCR.

The results showed that the CoCl<sub>2</sub> supplementation had no effect on the proliferation of all the three type of cells analyzed, while the up-regulation of chondrogenic markers such as aggrecan, sox9, and type II collagen, was dependent on the cellular source.

This study shows that hypoxia induced by CoCl<sub>2</sub> treatment can differently influence the behavior of MSCs of different sources in their chondrogenic potential. These findings should be taken into consideration in the treatment of cartilage repair and regeneration based on stem cell therapies.

## Keywords

Cobalt chloride, hypoxia, chondrogenic differentiation, mesenchymal stem cells, dental pulp, umbilical cord, adipose tissue