Kai Xiao
Dr. med

Properties of Human Mesenchymal Stem Cells (MSC) Derived from Reamer-Irrigator-Aspirator (RIA) and Bone Marrow and the Effect of Bone Morphogenetic Protein-2 on Osteogenesis of RIA-derived MSCs (RMSC) and Bone Marrow-derived MSCs (BMSC)

Einrichtung: Orthopädie

Doktorvater: Priv.-Doz. Dr. med. Arash Moghaddam-Alvandi

Although the RIA autologous bone graft has been applied in clinic as an alternative method to treat nonunion or bone defects, there is still not enough evidence to elucidate the osteogenesis compared to iliac crest at the molecular and cellular levels. Meanwhile, in recent decades, BMP-2 is extensively used in research and in the clinic as a growth factor to improve bone formation. However, there is still no consensus on the role of BMP-2 in osteogenesis of MSCs.
In our study, we confirmed the multipotency of MSC-derived from RIA (RMSC) compared to MSC-derived from bone marrow aspirate (BMSC) obtained from iliac crest and investigated the effect of exogenous BMP-2 on osteogenesis of RMSCs and BMSCs *in vitro* and *in vivo* experiments. *In vitro*, RMSCs and BMSCs at passage 2 were identified by flow cytometry. The differentiations of MSCs were observed in an adipogenic, chondrogenic and osteogenic induction medium, respectively. Meanwhile, BMP-2 (0.2 or 1.0ug/ml) was used as a supplement in an osteogenic induction medium to evaluate the effect of BMP-2 on osteogenic differentiation of MSCs. ALP activity and calcium deposition were used to evaluate the osteogenic differentiation at day 1, day 7, day 14 and day 21. The expression of osteoblast markers including ALP, OPN, OCN and Runx2 were measured by real time PCR to evaluate osteogenic differentiation. *In vivo*, osteogenesis was performed subcutaneously in SICD mice by implanting β-TCP with RMSCs/BMSCs with or without BMP-2 (4 or 20ng), and histochemistry was used to observe new bone formation by H＆E staining after 8 weeks of implantation under a confocal laser scanning microscope.
Our findings showed that RMSCs possess similar multipotency with BMSCs *in vitro*. However a superior osteogenesis was found in RMSCs *in vivo*. Dexamethasone (D), Ascorbate (A) and β-glycerophosphate (G) as the osteogenic medium supplements (DAG) seem to influence exogenous BMP-2. BMSCs or RMSCs in the presence of BMP-2 in DAG did not increase the level of osteogenic markers in the initial (ALP activity, ALP mRNA expression), or final (Calcium deposition, OCN and OPN mRNA expression) phases. Data suggest that the addition of exogenous BMP-2 did not improve osteogenesis of human BMSCs and RMSCs *in vitro*. *In vivo*, BMP-2 presented down-regulation in bone formation in RMSCs and up-regulation in BMSCs; however, the capability of osteogenesis in RMSC itself is not inferior to BMSC treated with BMP-2.

In summary, the present study suggests that the use of MSCs from RIA materials in orthopedic is reasonable and can be used as an additional cell-based therapeutic strategy in combination with the gold standard treatment of iliac crest autograft. There is no consensus in the literature between results on BMP-2 *in vitro* and *in viv*o, which demonstrates that BMP-2 as a strategy for treatment in the clinic requires further consideration of the attending physicians.