Dexamethasone modulates BMP-2 effects on mesenchymal stem cells in vitro

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Introduction: Dexamethasone / ascorbic acid / glycerolphosphate (DAG) and bone morphogenic protein (BMP)-2 are potent agents in cell proliferation and differentiation pathways. Up to date, there are only few data in literature which elucidate the complex relationship between these factors. This study investigates the in vitro-interactions between dexamethasone and BMP-2 for an osteoblastic differentiation of mesenchymal stem cells (MSCs).

Materials and Methods: Bone marrow derived human MSCs from 3 volunteer donors were isolated and expanded over 12 d in vitro. Afterwards a total of 5,000 human bone marrow cells / 1.9 cm² were seeded into each culture dish and stimulated with DAG (group A), BMP-2 + DAG (group B), and DAG + BMP-2 combined with a porous collagen I/III scaffold (group C). RT-PCR, sandwich-ELISA, immunocytochemical stainings and flow cytometry analysis served to evaluate the osteogenic promoting potency of each of the above conditions in terms of cell morphology / viability, antigen presentation, and also gene expression.

Results: DAG induced collagen I secretion from MSCs, which was further increased by the combination of DAG + BMP-2. In comparison, the collagen scaffold and the control samples showed no significant influence on collagen I secretion of MSCs. DAG stimulation of MSCs also led also to a steady but not significant increase of BMP-2 level. A DAG and more a DAG + BMP-2 stimulation increased the number mesenchymal cells (CD105+/CD73+). RANKL+ cells were strongly promoted by the DAG + BMP-2 combination and expression was also seen in both the DAG and the DAG + BMP-2 stimulated cells. All samples showed mRNA of alkaline phosphatase (ALP), osteopontin (OP), Runx2, Twist 1 and 2, Notch 1 and 2, osteonectin (ON), osteocalcin (OC), bone sialoprotein (BSP), collagen IA1 after 28 days of in vitro culture. Culture media of all samples showed a decrease in Ca²⁺ and PO₄²⁻ concentration, whereas a collagen I peak only occurred at day 28 in DAG and DAG + BMP-2 stimulated bone marrow cells.

Discussion: We confirm the results of other investigators [1,2] who showed osteoblastic differentiation of MSCs under BMP-2 supplemented collagen I-based scaffolds. Besides species-related and individual differences, it is evident that BMP-2 regulates not only osteoblast differentiation but also influences other pathways of different human cells types. It also should be considered that glucocorticoids influence the recruitment and differentiation of other bone cells such as osteoclasts. One possible explanation for the increasing BMP-2 level under DAG could be an induction of BMP-2 genes by DAG stimulus.