PAMAM DENDRIMERS USED AS VECTORS FOR GENE DELIVERY INTO MESENCHYMAL STEM CELLS

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The gene therapy area has expanded its search to find high-efficient and nontoxic nonviral gene delivery vectors. Numerous materials (such as cationic liposomes and polymers) have been studied as potential vectors for gene delivery with varying results. Polyamidoamine (PAMAM) dendrimers with primary amino surface groups were shown to have the inherent ability to associate, condense and efficiently transport DNA into a wide number of cell types[1]. A comparative study with several representative transfection systems (that included branched and linear polyethyleneimines, polyamidoamine and poly(propyleneimine) dendrimers, poly[N-ethyl-4-vinyl pyridinium bromide, as well as copolymers and lipid based reagents) has shown that transfection efficiency strongly varies with the kind of vector molecule used but also with the type of cells (10 different cell lines were tested)[2].

In the present work, our main goal was to perform a systematic evaluation of the transfection efficiency achieved by different generations of PAMAM dendrimers using mesenchymal stem cells (MSCs) due to the relevance of these cells in bone regeneration applications and in tissue engineering. A β-Galactosidase reporter gene system was used and several experimental conditions were assayed.

Since the transfection efficiency obtained was always very low, it was hypothesized that even a low transfection level could however be sufficient for the envisaged application, i.e., to improve the *in vitro* differentiation of MSCs towards the osteoblastic lineage. To confirm this possibility, MSCs were genetically engineered to express the human BMP-2 gene using PAMAM dendrimers. Differentiation of MSCs was studied through the analysis of established markers of the osteoblastic phenotype, including the activity of alkaline phosphatase, secretion of osteocalcin, and deposition of a calcified matrix. Results clearly pointed out that the systems PAMAM dendrimers/hBMP-2 plasmid DNA strongly induced *in vitro* differentiation of MSCs to the osteoblast phenotype suggesting that other low toxic and low efficient nonviral vectors can actually be used with efficacy in orthopaedics, despite their low transfection efficiency.

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