Exosomes containing CD39 and CD73 were very potent in reducing pro-inflammatory cytokine production in in vitro inflammation assays (IC50 in pM range). Transgenic exosomes are a promising approach for the delivery of membrane bound therapeutic enzymes and may have potential for the treatment of disease, including rheumatoid arthritis.

P049

Bacterial vectors for gene therapy have beneficial effects in a murine model of colitis

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The non-specific effects of bacterial vectors for gene therapy are largely unknown. Inflammatory bowel diseases are associated with changes in gut microflora. Bacteria-mediated gene therapy might be suitable for this disease as the target tissue is accessible for the bacterial vectors. The aim of our study was to prove whether a bacterial vector for gene therapy has any effects on dextran sodium sulfate (DSS) colitis in mice. Colitis was induced in male mice using 2,5% DSS in the drinking water for 9 days with one day recovery. Live or heat-inactivated attenuated Salmonella typhimurium strain SL7207 ($1 \times 10E9$) were administered via gavage every other day. Weight and stool consistency were monitored daily. After sacrifice colon weight/length and spleen length/weight was measured and samples were taken for histology, biochemistry and gene expression analysis. DSS induced diarrhea, weight loss, increased spleen weight and reduced colon length in comparison to the control group. Live, but not inactivated Salmonella partially but significantly prevented colitis-associated changes in the observed parameters. Although biochemical and histological outcomes are yet to be determined, based on the clinical variables it is clear that the severity of colitis was lower in the live Salmonella-treated groups in comparison to the DSS group receiving inactivated Salmonella and DSS group without treatment. These results point towards a possible bias in experiments using bacterial vectors for gene transfer in vivo. Although the mechanism is unclear, the design of the experiments should include appropriate control groups to distinguish gene-specific and non-specific effects of bacteria-mediated gene therapy.

P050

Enhancement of osteogenic vector

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The work was focused on the improvement of osteoinductive vectors to compensate low transfection efficiencies. Here we describe improvements of an osteogenic vector for significantly enhanced mediation of osteogenic differentiation of target cells and tissues. Codon optimization, addition and mutation of cis and trans acting elements, as well as screening for more potent secretion signal in order to improve BMP2 secretion were under investigation of this study. Furthermore, we describe a space saving hybrid vector capable of overexpressing bone morphogenic protein 2 (BMP2) while simultaneously down regulating several previously elucidated inhibitory genes by inserted shRNAs (either alone or in clusters) in order to enhance therapeutical effect. Various enhancements showed several fold higher impact in osteogenic differentiation of target cell lines and mesenchymal stem cells. By combining the most promising improvements, even higher differentiation rates could be achieved even with low transfection efficiencies. Finally, combinations of BMP heteromers (BMP2 to BMP7 vector ratio) revealed ideal ratio for the mediation of enhanced osteogenic differentiation. Altogether, we present several improved and highly efficient vectors and their impact in osteogenic differentiation which can be used in viral as well as non-viral gene therapeutical applications, and furthermore can easily be transfered to other fields of gene therapy.

P051

Efficient and non-invasive plasmid-DNA administration into the tibialis cranialis muscle of dwarf mice

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Our group investigated an alternative treatment for growth hormone deficiency based on hGH-DNA plasmid administration followed by electrotransfer. Aiming at improving this strategy, hGH plasmid-DNA (50 μg) were administered into the exposed quadriceps or non-exposed tibialis cranialis muscle of immunodeficient dwarf (lit/scid) mice. For the quadriceps, we utilized our previous optimized electrotranfer conditions (eight 90 V/cm pulses of 20 ms with 0.5 s intervals) while for the tibialis a new combination of high/low (H/L) voltage pulses (one 800 V/cm pulse of 100 μs and one 100 V/cm pulse of 400 ms) was used. After 3 days, blood was withdrawn and hGH determined, both groups showing similar results: 5.0 ± 2.2 and 3.5 ± 0.9 ng/mL ($P > 0.05$) of serum for quadriceps and tibialis treatment, respectively. A second experiment (28-day assay) was carried out in order to compare the growth parameters relative to the two groups: the slopes of the body weight variation curves were thus found similar (0.066 and 0.063 g/mouse/day) while the final body weight increase was 16.1% for the quadriceps and 18.9% for the tibialis group, respectively. Tail and nose-to tail length increase was 4.5% and 7.1% for the quadriceps and 4.8 and 4.6% for the tibialis group. Circulating mIGF-1 levels were also determined, being 126 ± 46.5 and 106 ± 93.2 ng/ml ($P > 0.05$) for quadriceps and tibialis treatment, respectively. Based on these results, hGH-DNA administration into the tibialis muscle followed by HV/LV electrotransfer proved to be an efficient and less aggressive treatment, much more suitable for pre-clinical testing since it avoids muscle exposition.

P052

Development and Therapeutic Efficacy of the DNA Complex-Releasing Systems Comprising Injectable Auto-Forming Alginate Gel

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