Monitoring Adipose-derived Human Mesenchymal Stem Cells in vivo: Urine Gaussia Luciferase Expression

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Introduction: Luciferase-mediated bioluminescent imaging is a powerful ex vivo technique to assess in vivo biological processes, providing a noninvasive means to account for cell viability. The purpose of this study was to evaluate Gaussia luciferase (Gluc) in monitoring implanted adipose-derived human mesenchymal stem cells (Ad-HMSCs, xenograft) viability in vivo and after recombinant human bone morphogenic protein (rhBMP-2) exposure. Ad-HMSCs and their survival are of interest because they have the propensity to differentiate into osteoblasts and generate bone. The following study investigates our hypothesis that higher photon measurement of Gluc activity in urine is associated with increased cell viability in vivo.

Methods: Ad-HMSCs were isolated using standard cell culture techniques, maintained in preadipocyte media, and introduced to a lentivirus encoding Gluc. Posterolateral spinal fusion was performed in rats. Cells were implanted into the paraspinal muscle bed across the L4-L5 transverse processes. Urine was collected by experimenter bladder expression daily over the first two weeks, then every other day until sacrifice. Urine Gluc expression was assessed via a luminometer in relative light units per second (RLU/s).

Results: Results showed that the average RLU/s values from rats with 5x10^6 Ad-HMSCs engineered with 0.003 mg/ml rhBMP-2 on ACS were the highest, followed by the 5x10^6 Ad-HMSCs exposed to rhBMP-2 on ACS, then the 5x10^6 Ad-HMSCs/ACS (no rhBMP-2), and finally the rhBMP-2/ACS only (no cells, control) (p< 0.05). In vitro results were similar to in vivo study findings, wherein Gluc expression was only observed during the first 2-3 weeks after infection. Data suggest that decreased Gluc expression was possibly due to deletion of Gluc (target DNA), cell mutation, limited integration, or unstable infection rather than cell death.

Conclusion: Gluc urine-based assays facilitate frequent interval measurements in longitudinal studies and avoid the invasive procedure of terminal tissue harvest to evaluate the stem cells.