Gene Therapy for Head and Neck Cancer: A Murine Model

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Purpose:
The aim of this project was to develop an animal model of alloantigen gene therapy for head and neck cancer which would model the human trial of alloantigen gene therapy in progress. This animal model could then be used to determine the mechanism by which alloantigen gene therapy results in cancer regression, such that the anticancer effects can be maximized. This was to be accomplished by constructing a drug that was analogous to the human treatment, but effective in mice. The human drug causes expression of a foreign antigen on the surface of tumor cells in an attempt to heighten the immune response to cancer. The foreign antigen used in the human trial is HLA-B7, a class I major histocompatibility complex (MHC protein not present in the treated patients. For the animal model we therefore used a comparably foreign antigen, encoded by the H-2 gene complex of mice.

Methods:
For the development of a syngeneic mouse model which would mimic the human trial, the same plasmid vector backbone VR-1031 was used. However, as the murine histocompatibility complex differs somewhat from that in humans, the incorporation of a different MHC gene was required. The mouse version of the histocompatibility complex is known as H-2; the human complex is designated HLA (human leukocyte antigen). The Balb-C mice used in this study express H-2K^d. The gene for H-2K^b was therefore extracted for testing the effects of an alloantigen. Using appropriate restriction enzymes, this gene was inserted into the plasmid VR-1031, as was the gene for beta-2 microglobulin, which is required for proper expression of the MHC molecule on the cell surface. This product was then tested by sequence analysis, by an in vitro model of transfection, and in the syngeneic mouse model. The level of expression in the in vitro model was determined by flow cytometry with specific antibodies.

Results:
Sequence analysis showed the H-2K^b and P-2 microglobulin genes to be intact in the VR-1031 plasmid. However, when tested in vitro, expression of the MHC molecules was not observed. In addition, the mice failed to show tumor regression.

Discussion:
The gene therapy product created in this study failed to result in production of an MHC protein. As the insertion of the genes in the appropriate location and orientation in the vector was confirmed, the lack of expression may have been due to a faulty promoter. Therefore, the next step in this ongoing study will be to use a different promoter, in attempt to achieve adequate MHC expression.