How Human Gene Therapy Has Treated Brain Cancer Since 2000

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ABSTRACT

Disorders and diseases can be treated with a variety of methods, including conventional medicine and human gene therapy. The primary distinction between these two methods lies in their respective approaches. Biomedical engineering is an emerging field that seeks to focus on the advances to improve human health and health care with genetic engineering, which is a field involving genetics, as a primary method as one of the subfields. Human gene therapy, falling underneath the field of genetic engineering, targets the root cause of the ailment, resulting in long-lasting effects. Human gene therapy has been accessible since the early 1970s, with a primary focus on the treatment of genetic disorders and diseases (1992). Scientists have made significant efforts to address a wide range of ailments, including various types of cancer, sickle cell disease, hemophilia, and others. This article specifically highlights the remarkable accomplishments of human gene therapy treatments for animal models of brain cancer over the past 25 years. Through an extensive examination of existing literature, we review an article where mice were observed using in vivo experimental work when evaluating baculoviral vectors as a potential solution for brain cancer. Furthermore, we review the various techniques employed in human gene therapy, thereby ascertaining their potential applicability as a viable treatment modality. Based on this data, the author has concluded that human gene therapy, facilitated by the utilization of recombinant DNA technology, has effectively treated brain cancer.

INTRODUCTION

Biomedical engineering is a broad, burgeoning field combining biology and engineering to improve human health. The field encompasses sub-specialties such as prosthetics, biomedical instrumentation, synthetic biology, and genetic engineering. Genetic engineering, usually in

reference to recombinant DNA technology, is the artificial modification, manipulation, or assimilation of DNA in order to modify an organism. Some significant advancements are the hepatitis B vaccine, the human growth hormone, human insulin, as well as disease-resistant plants (2019). Human gene therapy is an emerging field in genetic engineering. Human gene therapy is a technique that modifies a person's genome to treat or cure a disorder or disease, vector/gene integration, or the presence of exogenous copies of a gene to a person without modifying the person's DNA itself. There are multiple techniques that can be used to prevent disease such as plasmid DNA, viral vectors, bacterial vectors, CRISPR gene editing, and patient-derived cellular gene therapy products(Bansal et. al, 2000). These techniques may be applied in-vitro, a technique used in human gene therapy by using cell cultures in a laboratory, ex-vivo, a technique used in human gene therapy using a tissue not created artificially instead taken directly from the organism, and in-vivo, a technique used in human gene therapy which takes place inside the organism.

Human gene therapy has been in existence since the early 1970s, with the primary objective of treating various genetic disorders and diseases, including sickle cell disease, hemophilia, and cancer. One type of cancer being treated is brain cancer. Brain cancer treatment is a potential use for these techniques. Brain cancer is a life-threatening disease due to the changes it causes to the vital structures in the brain. As of 2020, an estimated 251,329 people died of primary brain cancer and central nervous system (CNS) tumors(2012). In pediatric patients, malignant brain tumors kill one within 5 years of diagnosis. The relative 5-year survival rate for people younger than 15 is around 75%. For those ages 15-39, the 5-year survival rate nears 72% and for those 40+, the survival rate is about 21% (2012). Common treatments for patients with brain cancer include radiology, surgery, and chemotherapy. The

primary benefit to human gene therapy is the effects. With a focus on two major papers, this article aims to describe various treatment methods for brain cancer using human gene therapy as there needs to be more effective methods for treating brain cancer. This article also reviews human gene therapy techniques for brain cancer in mice studies.

METHODS

The author employed several methodologies to identify suitable papers for review. The author conducted an initial literature search on Google Scholar and PubMed using the keyword phrase "human gene therapy treated brain cancer." After articles were identified, they were filtered to display only results that were accessible in the English language, returning an initial total of 322 articles from the PubMed database and approximately 1.15 million results from Google Scholar. From the multiple options, only two major articles centered the message aimed to be presented by the author. From this extensive pool, the author meticulously selected two articles that were pertinent to the research topic. The following papers were selected due to the relevance for the topic of this article.

RESULTS

Techniques of Gene Therapy Used for Brain Cancer Treatment

The article entitled "Gene Therapy for Brain Tumors" presents findings that illustrate how diverse approaches in human gene therapy can effectively diminish the size of brain tumors (Bansal et al., 2000). There are various methods by which gene therapy can be employed to eradicate undesirable cells, such as cancer cells. These methods encompass gene transfer-based immunotherapy, gene-directed enzyme prodrug therapy (GDEPT), and oncolytic virus therapy. These techniques have demonstrated efficacy in laboratory settings by effectively killing tumor cells and reducing tumor growth in living organisms (Bansal et al., 2000).

There are numerous human gene therapy techniques including the utilization of plasmids and calcium phosphate transfection (Bansal et al., 2000). Plasmids, which are small circular DNA molecules capable of independent replication within cells, can be injected into tissue at high concentrations. The surrounding cells take up the DNA, resulting in the expression of the inserted gene as mRNA and protein. Plasmids are considered safe for gene therapy due to their non-inflammatory and non-immunogenic nature, as well as their ability to function without cell division. However, only a small percentage of exposed cells typically express the gene. Another technique used is calcium phosphate transfection(Bansal et. al., 2000). Calcium phosphate transfection involves the addition of DNA and calcium phosphate to cultured cells, where the DNA adheres to cells and likely enters through active endocytosis(Bansal et. al., 2000. Electroporation employs electromagnetic waves to render the cell membrane permeable to DNA, leading to gene expression, although the DNA remains primarily extrachromosomal.

Gene therapy utilizes various viral packages, such as adenovirus, herpes simplex virus, adeno-associated virus, and retroviruses, to deliver therapeutic genes into the body(Bansal et. al., 2000). These viruses are modified to be incapable of replication and must be cultivated in helper cell lines(Bansal et. al., 2000). The selection of a viral vector is crucial, as it can impact the therapy's effectiveness(Bansal et. al., 2000). Different viral vectors have different advantages and disadvantages. Viral vectors offer advantages such as tissue selectivity, integration into the host genome, and stability. However, they also possess drawbacks, including tissue toxicity, immune reactions, and limited gene size. Due to their size, viruses may encounter difficulties in penetrating the brain, but efforts are underway to enhance delivery by intra-arterial injection with blood-brain barrier disruption. Overall, gene therapy utilizing viral vectors has yielded promising results (Bansal et al., 2000).

The results from the article ‘Evaluation of Baculoviruses as Gene Therapy Vectors for Brain Cancer’ show that using cell cultures, animal models, human gene therapy, adenoviral vectors, and baculoviral vectors can be used to treat brain cancer. The following text discusses the objective of the study as well as the outcomes. This study conducted by (Fallit et al., 2023) discusses about the significance of assessing the immune response to blood vessels as a means to comprehend their efficacy as vectors for gene therapy. The conducted study entailed the administration of BVs(baculoviruses) to mice, thereby inducing a pre-existing immune response. The primary objective of this study was to investigate the efficacy of BVs not solely as therapeutic agents for brain tumors, but also as carriers for gene delivery into the CNS.

The text discusses a study that used viral vectors to transfer genetic material into normal and tumoral glial cells. The researchers used different vectors expressing fluorescent proteins to assess their effectiveness. Mice were pre-immunized with the BV expressing dTomato and then challenged with the BV expressing citrine to ensure immunity was directed against the viral vector. The human cell line U251-MG was incubated with the BV encoding citrine and the AdV vector encoding GFP. The AdV vectors had a high transduction efficiency, easily transducing over 90% of cells PFU/cell. PFU verifies the presence of viral vectors within a cellular environment. The BV had a lower transduction efficiency but still transduced over 40% of cells (Fallit et al., 2023).

The text delves into the classification of diffuse gliomas, which is based on genetic and epigenetic alterations rather than histopathological characteristics. The mutational status of the enzyme isocitrate dehydrogenase (IDH) 1 and 2 has emerged as the primary marker for prognosis and molecular classification in adult diffuse gliomas. Tumors that exhibit mutated IDH have a more favorable prognosis, whereas wild type IDH gliomas are all classified as

glioblastoma multiforme (GBM). The text also mentions to the utilization of BV and AdV to transduce neurospheres, a culture system composed of a cluster of cells from biopsies, that are derived from biopsies of GBM patients, and the correlation between the expression of certain molecules and the transduction efficiency of BV. Furthermore, explores the potential application of gene therapy for mIDH (mutated IDH) glioma patients and the transduction efficiency of BV in neurospheres that are derived from a mIDH astrocytoma biopsy (Fallit et al., 2023).

examines the comparative transduction capabilities of BV (Baculovirus) and AdV (Adenovirus) in neoplastic (cancerous) and normal astrocytes. In rat GBM cells, AdV exhibited a higher transduction efficiency than BV. Conversely, in primary cultures of astrocytes, BV demonstrated a higher transduction efficiency than AdV. Similar outcomes were observed in mouse GBM cells and primary cultures of astrocytes. Both viruses displayed a linear, dose-dependent enhancement in transduction efficiency. The incubation of murine astrocytes with these viruses did not induce any cytotoxic effects. In neurospheres derived from genetically engineered mIDH astrocytomas in mice, both viruses exhibited a linear increase in transduction efficiency, reaching approximately 60% positive cells at MOI 1000 (Fallit et al., 2023).

The study assessed the efficacy and safety of gene therapy vectors in non-neoplastic brain tissue of patients with GBM. The researchers conducted a comparative analysis of the transduction efficiency and neurotoxicity of BVs and AdVs in mice. It was observed that the highest dosage of BVs tested did not induce any neurotoxic effects, thus it was selected for administration into the brain. Conversely, the maximum tolerated dose for AdVs in the mouse brain was determined to be 107 plaque-forming units (PFU) per site, which was subsequently employed for intracranial AdV injection. The mice were subjected to injections of BV and AdV in

order to investigate the expression of transgenes within the brain. Following a period of 5 days, both vectors exhibited favorable outcomes, and the infiltration of immune cells was found to be comparable in both instances (Fallit et al., 2023).

The experiment was conducted to assess the efficacy of BVs in transducing normal and neoplastic astrocytes in mice. The experimental procedure involved the injection of BV citrine into the mice, and the subsequent observation of transgene expression in tumor cells within the tumor mass, as well as in non-neoplastic cells surrounding the tumor. The findings demonstrated a substantial reduction in transgene expression among mice that had been previously immunized. In addition, transgene expression was detected in astrocytes of naïve mice. The stability of BV-mediated transgene expression was evaluated at 7 and 21 days post-injection, and it was determined that the expression remained stable for at least 21 days, without any indications of neurotoxicity (Fallit et al., 2023).

CONCLUSION AND DISCUSSIONS

Human gene therapy is an emerging discipline that aims to address ailments that have proven resistant to conventional medical interventions. Brain cancer, a highly lethal disease, exhibits a maximum survival rate of merely five years in certain cases. Presently, human gene therapy has demonstrated efficacy in treating brain cancer in mice, revealing a reduction of tumor growth rates through several techniques. Scientists have the potential to eradicate cancer and other life-threatening diseases by leveraging these research findings in the near future.

This paper discussed the development of human gene therapy, provided background information on brain cancer, and showed human gene therapy techniques for treating brain cancer in model systems. Since 2000, human gene therapy has effectively treated brain cancer

in mice, eradicating tumor cells. Along with those results, the size of the tumor in cell cultures reduced. This article shows that brain cancer can be treated on human patients in the future. Several human clinical trials have shown a success rate in removing the tumor from the body (2022). More research is needed to understand how gene therapy techniques will be more commonly practiced for cancer treatments.

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