

The scientific abstract:

An adenoviral vector, BG00001, which expresses the human interferon-beta (hIFN- β) gene under the direction of the cytomegalovirus (CMV) immediate-early promoter, is being developed as a locally administered therapy for glioblastoma multiforme (GBM). The phase I study described below will include adult patients with resectable GBM that has recurred or progressed after initial therapy. In animal models the local production and secretion of hIFN- β protein following gene transfer has demonstrated potent tumoricidal effects that extend beyond the transfected cells. It is hoped that local delivery of the hIFN- β gene will have similar anti-tumor activity in humans with GBM.

BG00001 is a human serotype 5 adenovirus vector from which most of the E1 and E3 genes have been deleted, and into which the hIFN- β gene has been inserted. The E1 deletion renders the vector replication-incompetent. BG00001 is derived from a Master Seed Stock of virus H5.010CMVhIFN- β , which was generated through a collaboration between Biogen and the University of Pennsylvania. BG00001, and similar adenovirus vectors delivering the human or murine interferon-beta genes (including H5.010CMVhIFN- β) have been tested by direct injection into various murine tumor models, including subcutaneous xenograft models with human glioma cell lines. These experiments have consistently demonstrated a potent anti-tumor effect. In an orthotopic xenograft model of human prostate cancer, high intra-tumor concentrations of IFN- β (100-1000 ng/g tumor tissue) have been achieved with little or no detectable circulating IFN- β . These results suggest that gene transfer therapy with BG00001 has the potential to result in anti-tumor activity without producing the toxicities caused by high circulating concentrations of IFN- β .

Under the sponsorship of the Institute for Human Gene Therapy (IHGT), a single patient with recurrent high-grade glioma has been treated in a phase I study with H5.010CMVhIFN- β . The patient received a dose of 3×10^{10} viral particles at each of two administrations, and tolerated the therapy well. More than 12 months following treatment the patient is fully functional, and remains free of detectable disease. Adenoviral vectors carrying other genes, including herpes simplex virus thymidine kinase and p53 have generally been well tolerated when delivered by intracranial injection to patients with high-grade gliomas. However, serious drug-related adverse events have been observed in a minority of patients treated at doses comparable to the highest dose-level proposed in the study described below.

As described below, BG00001 will be manufactured at BioReliance (Rockville, MD) under GMP. Prior to initiation of the phase I clinical study, a thorough non-human primate toxicology study will be performed under GLP. The described phase I clinical study will be conducted at the University of Pennsylvania (Philadelphia, PA), under the direction of Stephen Eck, M.D., Ph.D. In order to enroll the required number of patients within the planned 1.5 year period, additional clinical sites will be added. No patient will be enrolled at a new clinical site prior to submission of the Principal Investigator curriculum vitae, IRB-approved informed consent, and IBC and IRB approvals to the Recombinant DNA Advisory Committee, in accordance with NIH guidelines.

The proposed phase I study is a multi-center, open label, two part, dose escalation by cohort protocol. The primary objective is to determine the major toxicities of intra-tumor injection of BG00001 in subjects with recurrent or progressive GBM. The secondary objectives are as follows: determine the maximum tolerated dose (MTD) of BG00001; assess 6 month progression-free survival (PFS); assess overall survival at 12 months; assess pathological evidence of anti-tumor activity; assess pathologic evidence of gene transfer in resected tumor specimens; determine serum, cerebrospinal fluid (CSF) and brain tissue interferon-beta levels; assess immune response to adenoviral vector and hIFN- β .

Approximately 32 subjects (range: 3-44 subjects) will participate. Subjects will be ≥ 18 years of age, with a diagnosis of GBM, and recurrent or progressive tumor following prior therapy, in whom surgical resection is clinically indicated.

The study will be divided into two parts. Part I will be a dose escalation phase to determine the MTD or maximum achievable dose (3×10^{12} viral particles/administration). Part II will enroll an additional 14 – 17 subjects at the MTD or maximum achievable dose, in order to treat a total of 20 subjects. Up to five cohorts of 3 – 6 subjects will be enrolled into Part I. The starting dose will be 3×10^{10} viral particles/administration, and the dose will escalate approximately three fold between cohorts.

After informed consent is obtained subjects will undergo screening and baseline evaluations. On Day 1 eligible patients will be hospitalized, and undergo stereotactic, intracranial administration of BG00001 to five sites in their tumor. Subjects will remain hospitalized at least until Day 5 (post-operative day 4). Subjects will be readmitted to the hospital on Day 8, and undergo surgical resection of their brain tumor, followed by intraoperative administration of BG00001 to ten sites in the tumor bed and residual tumor. Subjects will remain hospitalized at least until Day 12 (post-operative day 4). The remainder of study follow-up will be on an outpatient basis. Subjects will follow-up on Days 15, 22, 29, 57 (8 weeks), 120 (17 weeks), 183 (26 weeks), and 365 (52 weeks). Surviving subjects will undergo brain MRI (magnetic resonance imaging) every 6 months after Day 365, until disease progression, and telephone follow-up to document survival every 6 months, after disease progression.

Safety will be monitored by periodic physical examinations, laboratory tests, and questioning about adverse events. Study-specific laboratory tests to assess safety will include head CT (computed tomography) the day after stereotactic drug administration, periodic brain MRIs beginning the day after tumor resection, interferon-beta levels in blood, cerebrospinal fluid (CSF), and brain tissue (at resection), anti-adenovirus antibodies in blood, and blood levels of the cytokines IL-6 and IL-10. Samples of blood, CSF, and nasal swabs will be banked for future analysis for adenovirus by polymerase chain reaction (PCR), if necessary.

If available, autopsy data will be collected. This will include the autopsy report (including brain), histopathological examination of brain (injection site), and PCR for adenovirus DNA in brain (injection site), lung, heart, liver, spleen, and bone marrow. Subjects will be asked to consider agreeing to an autopsy, when they give consent to participate in this study. However, subjects may enroll in this study whether or not they agree to an autopsy.

Efficacy measures will include the following: time to tumor progression; time to death; pathological evidence of anti-tumor effect; evidence for gene transfer in tumor specimens by *in situ* hybridization; evidence of effect on mitotic index and apoptosis. Mitotic index will be measured by staining for proliferating cell nuclear antigen (PCNA), and apoptosis will be measured by terminal deoxynucleotidyl transferase dUTP nick end-labeling (TUNEL).

With 20 patients treated at the MTD (or Level-5 dose), if the observed 6-month progression-free survival is 50%, then the 95% confidence interval is 28% - 72%. This will provide sufficient evidence of anti-tumor activity to support further evaluation of BG00001 in additional clinical studies of patients with GBM.