SCIENTIFIC ABSTRACT

There is an ongoing, intensive search for novel therapies to improve the prognosis of patients with the most common and aggressive form of primary brain cancer; high grade glioma (HGG; Grade III or IV glioma). Gene transfer is one such approach. Early gene-transfer studies with replication incompetent vectors showed this approach to be generally safe, but ineffective due to limited transduction of the tumor. More recently gene transfer has been attempted with oncolytic, replicating viruses. However these viruses are rapidly cleared by the immune system due to the inflammatory response generated from the virus and its lytic process. Toca 511 uses a retroviral replicating vector (RRV) to overcome the limitations of previous gene transfer protocols. This platform has the following advantages: 1) The vector only infects dividing cells, 2) The virus stably integrates into the genome of the tumor cells allowing for long-term control of the tumor, 3) The virus is not intrinsically oncolytic, and 4) The virus has been engineered to express the prodrug-activator enzyme, cytosine deaminase (CD), that catalyzes the intracellular conversion of the antifungal drug, flucytosine (5-fluorocytosine, 5-FC) to the cytotoxic drug 5-fluorouracil (5-FU). In both xenograft and syngeneic intracranial mouse tumor models the Toca 511/5-FC combination was able to significantly increase the survival of treated animals. The goal of the current clinical development program is to demonstrate the safety and efficacy of Toca 511 administered intratumorally to patients with recurrent HGG followed by cyclic treatment with the oral prodrug 5-FC. The vector used in this gene-transfer platform is a retroviral replicating vector derived from a cloned Moloney murine leukemia virus (MLV). The original ecotropic envelope gene has been replaced with an amphotropic envelope gene enabling the virus (referred to as ampho-MLV) to infect human cells. (Logg CR 2001) A modified, yeast-derived cytosine deaminase (CD) gene has been inserted into this vector. The vector and CD gene construct is classified as a prodrug-activator form of gene transfer. The final formulation of the vector/CD construct is referred to as Toca 511.

Multiple studies have been conducted in both tumor-bearing and non tumor-bearing mice with Toca 511 intracranial doses ranging from 1.7x10^3 to 4.7x10^6 TU/g of brain, with and without 5-FC. Durable, statistically significant increases of up to 3-6 fold survival advantages have been observed in a human xenograft intracranial mouse brain-cancer model, and two immune-competent mouse brain-cancer models. The lowest intracranial Toca 511 dose at which benefit was observed in any mouse model was at 1.7x10^3 TU/g, which was approximately the starting dose in the previous first-in-human study, and slightly below the proposed starting dose in the new study (9.5 x 10^3 TU/g). When Toca 511 was administered intracranially, which approximates the intended clinical route of administration (intratumoral-intracavitary), with subsequent administration of 5-FC, there were no treatment-related organ weight, macroscopic, or microscopic findings at any vector dose out to 180 days. When not followed by administration of 5-FC, administration of high dose Toca 511 to BALB/c mice (4.7x10^5 TU/g or higher) was associated with a low incidence (10 – 15%) of late-occurring (~6 months) lymphomas.- Lymphoma is a known spontaneous finding in mice and is also an expected consequence of MLV infection in BALB/c mice. No lymphomas were observed in this strain (BALB/c) or a second mouse strain (B6C3F1) when Toca 511 administration was followed by administration of 5-FC. In the absence of any other findings in other target sites or in other species, the relevance of lymphoma in mice to human carcinogenic risk is unknown (Wogan,

1984). Given that lymphomagenesis in the BALB/c mouse involves a number of species- and strain-specific mechanisms (immune tolerance, biodistribution, defective viral restriction and recombination with endogenous murine retroviruses) and that human subjects will also be receiving 5-FC, the risk of lymphoma in humans is felt to be quite low. No other Toca 511-related safety findings were identified in the preclinical studies. Biodistribution studies of Toca 511 in BALB/c mice have demonstrated widespread vector biodistribution, especially to lymphoid tissues, with no evidence of horizontal transmission to treatment naïve cagemates. Viral distribution in the B6C3F1 mice was limited, with low copy number of provirus within lymphoid tissues across all timepoints and no shedding of virus detected.

These preclinical data suggest that the risk-benefit ratio favors treating patients with recurrent HGG with Toca 511 and 5-FC as such patients have a projected life expectancy of approximately 6 months. A Phase 1 clinical, ascending dose study evaluating the safety and tolerability of single doses of Toca 511 administered intratumorally via stereotactic, transcranial injection and followed by repeated cycles of orally administered 5-FC has previously been reviewed by NIH-RAC and is currently enrolling subjects. (Protocol 0904-976). In this ongoing study, 3 subjects in the first dosing cohort have received Toca 511 injected once into the tumor and followed 3-4 weeks later by 6 days of oral 5-FC. 5-FC was repeated monthly until tumor progression. There were no DLTs or product-related SAEs reported for the first dosing group, and overall safety and tolerability were excellent. The new proposed clinical study is similar to the ongoing clinical study, but will evaluate the safety and clinical effects of ascending doses of Toca 511 administered to subjects with recurrent HGG at the time of resection, followed by treatment with repeat cycles of oral 5-FC. This study will evaluate intracavitary injection of a single dose of Toca 511 administered by multiple injections into the walls of the resection cavity at the time of craniotomy and tumor resection. Up to four dose levels of Toca 511 will be studied, with three subjects per dose level, and 6 subjects will be enrolled at the maximum tolerated dose Approximately 7 weeks (± 1 week) later, subjects will begin treatment with oral 5-FC for 8 days. On the 6th, 7th or 8th day of dosing the trough 5-FC serum concentration will be determined and the dose of subsequent 5-FC cycles adjusted to maintain the concentration in the therapeutic range. If tolerated, these 8-day courses of 5-FC will be repeated approximately every 8 (± 1 week) weeks until study completion. All three subjects in a dosing cohort must complete at least one cycle of 5-FC before dose escalation can occur. Subjects will undergo MRI scanning approximately every 8 weeks. Tumor response will be assessed using the Macdonald criteria. Safety assessments will include flucytosine blood levels, monitoring of blood, saliva and urine for virus, clinical chemistries and hematology at selected time points, and recording of adverse events throughout. All patients will be followed for 6 months in this study. All patients who receive Toca 511 treatment may elect to roll into a continuation protocol that will record long-term follow up for safety and viral biodistribution.