BACKGROUND

Oncolytic viruses are unique immunotherapeutic agents for the treatment of cancer. Amgen's oncolytic Herpes Simplex Virus 1 (HSV1) is FDA-approved for the treatment of melanoma, and several companies are developing oncolytic HSV1 variants including Johnson and Johnsons through their recent acquisition of BeneVir Biopharm. The Canadian company Turnstone Biologics, in a license agreement with AbbVie, initiated phase II clinical trials with the rhabdovirus Maraba MG1. Notably, Boehringer Ingelheim recently acquired the rights to the rhabdovirus vesicular stomatitis virus (VSV) for their clinical studies. Clearly there is strong incentive from big pharmaceutical companies to develop oncolytic viral platforms for cancer therapies. All these viruses are attenuated, even over-attenuated, to be safely administered to patient. *However, each oncolytic viral platform is faced with this critical limitation: Poor virus replication within tumours tissues and rapid clearance during treatment.* Thus, to improve oncolysis and stimulate greater anti-tumour immune responses, strategies to potentiate and sustain viral replication within tumour tissues are intensively pursued.

PROGRESS REPORT

We have discovered that the compound BI-D1870 is a small molecule that potently increases the replication of HSV1 and oncolytic rhabdoviruses in brain cancer cells. We have found that BI-D1870 treatment in a dose dependent manner strongly augments infection and spread of the oncolytic HSV1-1716 and HSV1-dICP0, and the oncolytic rhabdoviruses Maraba MG1 and VSVΔ51 in multiple cancer cell lines including brain cancer cells. In contrast, normal human fibroblasts, or normal murine breast epithelial cells, are unaffected by this pharmacoviral approach, as no increase in viral propagation is observed in these normal cells. We also examined the effects of BI-D1870 on the replication of other oncolytic viruses such as Vaccinia virus, Myxoma virus and Adenovirus in the glioblastoma cell line U343. Interestingly, BI-D1870 renders U343 glioblastoma cells refractory to these three viruses. The differential effects obtained between normal and cancer cells, and the complete opposite responses in viral potentiation between specific oncolytic viral platforms, offers clues into the mechanism of action of this pharmacoviral strategy that we are currently following.

Investigations *in vivo* also demonstrate that BI-D1870 potentiates viral propagation within brain and mammary tumour tissues. In addition to performing experiments aimed at determining the mechanism of action, we initiated investigations in murine models of cancer. BI-D1870 was previously shown to be well tolerated in mice at doses of 100 mg/kg with no toxicities, and notably, the compound can cross the blood-brain barrier. We performed three independent experiments using a luciferase expressing VSVΔ51-FLuc in combination with BI-D1870 administered intraperitoneally (i.p.) at 100mg/kg. A striking augmentation of viral replication/luciferase counts within tumour tissues was observed when VSVΔ51-Fluc was administered either intratumorally (i.t., at high titer - 5E8 pfu or at low titer - 1E6 pfu) in a subcutaneous mammary cancer model. We also measured increased luciferase counts in brain tumours of mice bearing $NF1^{+/-}$ $p53^{+/-}$ mouse glioma cells from VSVΔ51-Fluc administered intravenously (i.v. 5E8 pfu). These significant results demonstrate that BI-D1780 displays efficacy *in vivo*, even in a brain tumour model, with tolerable dosing in mice of 100 mg/kg. *However, solubility of the molecule is challenging, and its bioavailability, particularly the reported poor pharmacokinetics/dynamics (PK/PD) and limited half-life in tissues, is a concerning liability*. While remarkable *in vivo* response on viral propagation can still be obtained with BI-D1870, optimizing dose schedules and routes of administration, but also synthesizing derivatives that display peak activity, PK/PD, and bioavailability could further enhance therapeutic efficacy. We are currently testing a novel analog of BI-D1870 that we synthesized. We anticipate that this derivative will have even stronger effects in combination with oncolytic viruses for the treatment of brain malignancies.