after the injection of ONYX-015. Twenty patients were enrolled, 19 were eligible. Serious toxicities (> grade 2) were uncommon, and included hepatic (3), anemia (1), infection (1), and cardiac (1, atrial fibrillation). Sixteen patients were evaluable for response. Among these evaluable patients, 1/16 (6.3%) had a partial response, 1/16 (6.3%) had prolonged disease stabilization (49 weeks), and 8/16 (50%) had a >50% reduction in tumor markers. Among the 19 eligible patients,18 (94.7%) had specimens available for p53 analysis. Fifteen/18 (83.3%) had evidence of p53 mutation by one or both methods, although the methods correlated poorly. Viral shedding was confined to bile (2/2) and ascites (4/4). Pretreatment adenoviral antibodies were present in 14/14 patients and increased by 33.2% after ONYX-015 treatment. In the course of the trial, a patient with paired abdominal wall implants from a primary gall bladder carcinoma was injected with ONYX-015, followed by sequential excision of the lesions at 37 h and 7 days. Tissue sections were analyzed for evidence of viral replication using a novel assay, in situ RT-PCR to measure expression of hexon, a viral gene, which is expressed late during viral replication. Strong signals were obtained in gland-forming tumor cells both at 37 h and at 7 days, indicating that the virus was both present and replicating at those time points. Of interest, signal was also observed in adjacent normal stromal cells which were presumably wild-type for p53. Intralesional treatment with ONYX-015 in patients with hepatobiliary tumors is safe and well-tolerated, and some patients had evidence of an anticancer effect.

469

Phase II Study of the DNA Methyltransferase I (DNMT1) Inhibitor MG98 in Patients (Pts) with Renal Cell Carcinoma (RCC). A Trial of the National Cancer Institute of Canada Clinical Trials Group (NCIC CTG)

E. Winquist[1], J. Knox[2], J.-P. Ayoub[3], L. Wood[4], N. Wainman[5], L. Pearce[6], A. Shah[7], E. Eisenhauer[8]. [1]*London Regional Cancer Centre, Medical Oncology, London ON, Canada;* [2]*Princess Margaret Hospital, Medical Oncology, Toronto ON, Canada;* [3]*CHUM Hopital Notre Dame, Medical Oncology, Montreal QC, Canada;* [4]*Queen Elizabeth II Health Sciences Complex, Medical Oncology, Halifax NS, Canada;* [5]*NCIC CTG, Kingston ON, Canada;* [6]*MethylGene Inc, Montreal QC, Canada;* [7]*MGI Pharma Inc, Bloomington MN, USA;* [8]*NCIC CTG, Kingston ON, Canada*

MG98, a 2nd generation phosphorothioate antisense oligodeoxynucleotide, is a specific inhibitor of human DNMT1 mRNA. Hypermethylation by DNMT1 is postulated to inactivate tumor suppressor genes leading to neoplastic transformation. Thus, inhibition of this enzyme might restore normal growth control. Two phase I trials of MG98 have been conducted by the NCIC CTG: 21-day continuous IV and a 2-hour twice weekly 3 out of 4 weeks schedule. The latter was selected for phase II evaluation because it was well tolerated up to 360 mg/m^2/dose and a pt with RCC had an objective response. This 2-stage phase II study of MG98 evaluating this dose and schedule was conducted in pts with recurrent, unidimensionally measurable RCC. Accrual to the second stage of this trial would occur if 1/15 pts had an objective response or if 11 or more pts showed stable disease for a minimum of 8 weeks. Pharmacokinetic (PK) evaluation and assessments of DNMT1 mRNA in PBMCs were done in all patients. 17 eligible pts were entered with the following characteristics: prior nephrectomy: 12; prior chemo- or immunotherapy: 0; ECOG PS 0/1: 6/11; male: 11. All pts were evaluable for toxicity and 15 for response. Toxic effects graded by CTC v 2.0 were: fatigue 16 pts (4 gr 3, 1 gr 4), nausea 13 pts (1 gr 3), anorexia 12 pts (1 gr 3), fever 9 pts (1 gr 3), neurosensory changes 6 pts (0 gr 3). No grade 3 or 4 neutropenia or thrombocytopenia was seen. Elevations in serum biochemistry were noted as follows: creatinine: 9 (0 gr 3), ALT: 13 (6 gr 3, 1 gr 4), AST 13 (1 gr 3). All creatinine changes and all grade 3 ALT and AST elevations were in the subset of patients (n=12) with prior nephrectomy. Pharmacokinetic evaluation is currently ongoing and will be available for presentation. Best response was stable disease in 7 pts and progressive disease in 8 pts. PBMC data are awaited. The trial closed to accrual after the first stage. Study supported by Grants from the National Cancer Institute of Canada and MethylGene Inc.

470

Molecular imaging in the development of efficient gene therapy for human glioma

A.H. Jacobs[1], R. Reszka[3], A. Gossmann[4], C. Dittmar[1], C. Kaestle[1], R. Richter[1], K. Wienhard[1], W.D. Heiss[1], J. Voges[2]. [1]*MPI for Neurological Research, Laboratory for Gene Therapy and Molecular Imaging, Köln, Germany;* [2]*University of Cologne, Department of Stereotactic and Functional Neurosurg, Köln, Germany;* [3]*Max Delbrück-Center for Molecular Medicine, Berlin, Germany;* [4]*University of Cologne, Department of Radiology, Köln, Germany*

Objective: To non-invasively evaluate the safety and efficiency of vector delivery and gene transduction after gene therapy of patients with recurrent glioblastoma by positron emission tomography (PET) and magnetic resonance imaging (MRI).
Methods: 8 patients (age: 49-67) received a stereotactically guided Gd-DTPA infusion with subsequent MRI and intratumoral convection-enhanced delivery (CED; max. flow: 0.6 ml/h, volume 30-60 ml) of a liposome-gene-complex (LGC; DAC-Chol/DOPE [w:w; 30:70]) transducing herpes simplex virus type 1 thymidine kinase (HSV-1-TK) as part of a Phase I/II clinical trial. To determine the transduction efficiency, PET was performed after injection of [124I]-2-fluoro-2-deoxy-1-β-D-arabinofuranosyl-5-iodo-uracil ([124I]FIAU), a specific marker substrate for HSV-1-TK. Ganciclovir (GCV) treatment (2 × 5 mg/kg/bw; 14 days) was started four days after LGC-infusion. Treatment response was recorded by means of MRI, [18F]-2-fluoro-2-deoxy-D-glucose (FDG) and [11C]-methionine (MET) PET.
Results: Infusion of LGC was tolerated well. In 1/8 patient specific [124I]FIAU-accumulation was observed as indication for HSV-1-TK expression in coregistration to signs of necrosis after GCV treatment as determined by FDG- and MET-PET. In 4/8 patients [18F]FDG- and [11C]MET-uptake was focally decreased in areas coregistering to the distribution volume of Gd-DTPA. Two patients showed transient reduction of the methionine positive tumor-volume by more than 50 %. All patients developed tumor relapses outside areas with reduced tracer activity.
Conclusions: Intratumoral convection-enhanced delivery of LGC is safe and leads to focal alterations of tumor activity. However, overall therapeutic efficacy is low indicating that more efficient vectors have to be engineered. Non-invasive imaging of vector distribution and vector-mediated gene expression by PET and MRI shall contribute to the development of standardized gene therapy protocols and improve efficiency and safety of vector applications in humans.

471

TNFerade, an adenovector encoding the human tumor necrosis factor alpha gene, in soft tissue sarcoma in the extremity. safety and early efficacy data

A. Mundt[1], J. Nemunaitis[2], S. Vijayakumar[3], A. Sandler[5], N. Hanna[5], T. Peabody[1], N. Senzer[2], H. Rasmussen[6], T. Das Gupta[3], R. Weichselbaum[1]. [1]*University of Chicago, Cell and Radiation Oncology, Chicago;* [2]*US Oncology, Dallas, TX, USA;* [3]*University of Illinois, Radiation Oncology, Chicago;* [4]*Vanderbilt University, Hematology and Oncology, Nashville;* [5]*University of Kentucky, General Surgery, Lexington;* [1]*GenVec, Inc., Gaithersburg, USA*

Background: TNFerade is a second-generation replication deficient adenovector, carrying the transgene encoding for human TNFa, regulated by the radiation-inducible promoter Egr-1. Purpose: The purpose of the study was to identify the maximum tolerated dose (MTD) of TNFerade in combination with radiation and to assess potential biologic activity in patients with extremity soft tissue sarcoma, receiving radiation pre-operatively or for palliation.
Method: Standard Phase Ib study, exploring in a dose-escalating fashion 3 dose levels of TNFerade (4×10^9 - 4×10^{11} pu), given in 1 log increments by intra-tumoral injections, twice weekly during week 1, then once weekly during weeks 2-5, concurrent with radiation (~50Gy).
Results: Accrual into cohorts 1-2 has completed with 7 patients enrolled. TNFerade + radiation was well tolerated with no dose limiting toxicities (DLTs) and no drug-related serious adverse events (SAEs). Plasma-TNFa levels remained low in all patients (<5pg/ml); no patients had virus detected in cultures from blood or urine. Of the 6 patients evaluable for response assessments, 4/6 received TNFerade + radiation pre-operatively, 2/6 received TNFerade + radiation for palliation. Of the pre-operative patients, 3/4 showed a complete pathologic response and 1/4 showed >95% necrosis (PR). This is remarkable as these patients had very large tumors (baseline volumes of 837, 2985, 4056 and 2142cm3, respectively). One palliation patient demonstrated radiologic PR, the other stable disease. However, CT