Conclusions: ^{89}Zr-bevacizumab PET visualizes tumor lesions in RCC patients. Tumor quantification shows large differences within and between patients, whereas differences in normal tissue uptake between patients are small. With its high tumor-to-background ratio in target lesions this technique is expected to perform well in serial VEGF quantification during treatment with angiogenesis inhibitors.

222 POSTER

A first-in-human, phase I trial of the anti-DLL4 antibody (OMP-21M18) targeting cancer stem cells (CSC) in patients with advanced solid tumors

D.C. Smith[1], P. Eisenberg[2], R. Stagg[3], G. Manikhas[4], A. Pavlovskiy[5], B. Sikic[6], A. Kapoun[7], S.E. Benner[3]. *[1]University of Michigan, Department of Medicine, Ann Arbor MI, USA; [2]California Cancer Care, NA, Greenbrae CA, USA; [3]OncoMed Pharmaceuticals, Clinical Development, Redwood City CA, USA; [4]St. Petersburg State Healthcare Institution, Oncology Department, St. Petersburg, Russian Federation; [5]Federal State Institution Russian Research Center, Operative Surgery Department, Leningrad Region, Russian Federation; [6]Stanford University, Department of Medicine, Palo Alto CA, USA; [7]OncoMed Pharmaceuticals, Translational Medicine, Redwood City CA, USA*

Background: DLL4 is a ligand that activates the Notch pathway which is important for CSC survival. OMP-21M18 is a humanized, anti-DLL4 antibody that has been shown to inhibit tumor growth and decrease CSC frequency in minimally passaged human xenograft models, using an in vivo tumorigenicity limiting dilution assay. Inhibition of DLL4 has also been shown to cause dysfunctional sprouting of new vessels resulting in an antiangiogenic effect.
Methods: Patients with advanced solid tumors were enrolled in escalating dose cohorts and received OMP-21M18 weekly or every other week. The primary study objective was to determine the safety profile of OMP-21M18. Other objectives included: immunogenicity, pharmacokinetics, antitumor activity and biomarkers of Notch signaling and CSCs in blood, hair follicles and tumor cells.
Results: As of June 4, 2010, 33 patients were treated and 27 were evaluable for safety. Patients were treated with 0.5, 1, 2.5, or 5 mg/kg weekly or 2.5, 5 or 10 mg/kg every other week. The median age was 60 years and the patients had received a median of 4 prior chemotherapy regimens. The maximum tolerated dose (MTD) was not reached. Adverse events were predominantly grade 1 and unrelated to study drug. Adverse events reported as related to study drug and occurring in at least 3 patients included: hypertension (12), asthenia/fatigue (7), nausea (5), headache (3), and abdominal pain (3). Six of the 12 subjects who developed hypertension requiring oral antihypertensive treatment, had a diagnosis of hypertension at study entry. GI bleeding was observed in 4 patients and isolated BNP elevations were also observed. The half-life of OMP-21M18 was 12 days and administration of OMP-21M18 was associated with little immunogenicity. OMP-21M18 was shown to alter Notch signaling in blood and hair follicle cells. A waterfall plot of the % change in RECIST tumor measurements/patient suggests antitumor activity, in this heavily pretreated patient group, at doses of 5 mg/kg weekly and 10 mg/kg every other week and an unconfirmed partial response was observed in a patient with pancreatic cancer.
Conclusion: OMP-21M18 was generally well tolerated with asymptomatic hypertension being the most common drug related toxicity. The MTD was not reached at 10 mg/kg every other week. Encouraging early evidence of biologic and clinical activity has been observed. Enrollment is ongoing and updated results will be presented.

223 POSTER

Trastuzumab-DM1: mechanisms of action and mechanisms of resistance

G. Li[1], C.T. Fields[1], K.L. Parsons[2], J. Guo[1], G.D. Lewis Phillips[1]. *[1]Genentech Inc., Research Oncology, South San Francisco, USA; [2]Genentech Inc., Translational Oncology, South San Francisco, USA*

Overexpression of the HER2 receptor tyrosine kinase (RTK) occurs in approximately 20−25% of human breast cancer. Patients with elevated HER2 levels show more rapid disease progression and worse survival than breast cancer patients with HER2-negative disease. Trastuzumab (Herceptin®) is a humanized antibody developed specifically to treat HER2-positive breast cancer and is used in both the adjuvant and metastatic setting. Although trastuzumab is efficacious for many patients, a number of patients will have disease progression through trastuzumab therapy. We have recently developed an antibody–drug conjugate comprised of trastuzumab covalently linked through a stable linker to the microtubule inhibitory agent DM1, as an alternative treatment for patients whose cancers overexpress HER2. Trastuzumab-MCC-DM1 (T-DM1) is currently undergoing clinical testing in metastatic HER2-positive breast cancer patients. We performed cell culture studies to compare the potency of T-DM1 to chemotherapeutic agents and to determine T-DM1 mechanisms of action (MOA). Viability and clonogenic assays on HER2-amplified breast cancer cells show that T-DM1 is more potent than taxane or vinca alkaloid agents. Treatment with T-DM1 results in G2/M cell cycle arrest, as measured in cell cycle experiments or by Western analysis of the G2/M markers cyclin B1 and phospho-histone H3. Subsequently, G2/M-arrested cells undergo apoptosis as indicated by PARP cleavage and loss of XIAP expression. Like trastuzumab, T-DM1 suppresses signaling through the PI3 kinase pathway. However, T-DM1 inhibits PI3 kinase signaling in trastuzumab-insensitive cells; this activity was shown to be mediated by the DM1 component of T-DM1. Anti-proliferative activity of T-DM1 was also compared to T-DM1 components (trastuzumab, DM1, SMCC linker) and identified catabolites (Lys-MCC-DM1 and MCC-DM1). Only T-DM1 and DM1 potently inhibit growth of HER2-overexpressing breast cancer cells. Mechanisms of resistance to T-DM1 were investigated in 3 cell lines developed to have acquired T-DM1 resistance. One T-DM1-resistant line shows upregulation of multi-drug resistance (MDR) transporters. Increased expression of EGFR and other RTKs, as well as several erbB ligands was also observed. Mutations in beta 1-tubulin were not detected. These studies demonstrate the potent anti-tumor activity of T-DM1 compared to conventional chemotherapeutic agents, show multiple MOA for T-DM1, and shed light on potential mechanisms of resistance.

224 POSTER

Her3 as an emerging target for lung tumor initiating cells

G. Ciliberto[1], E. Marra[2], Z. Gunes[3], A. Noto[3], G. Roscilli[4], R. Mancini[3], M.R. Giovagnoli[3], A. Ricci[3], E. Giarnieri[3], L. Aurisicchio[2]. *[1]University Of Catanzaro "Magna Graecia", Experimental and Clinical Medicine, Catanzaro, Italy; [2]Biogem Scarl, Molecular Oncology, Ariano Irpino (AV), Italy; [3]University "La Sapienza", Experimental and Clinical Medicine, Rome, Italy; [4]Biogem, Molecular Oncology, Ariano Irpino (AV), Italy*

Background: Lung cancer is the leading cause of cancer mortality in the world. Current therapy is relatively ineffective and survival rate is approximately 15%. The human ErbB (or HER) receptor family plays multiple roles in normal cell functions as well as in tumor development and maintenance of lung cancer. Mutational activation and/or gene-amplification of ErbB family members is observed in several human malignancies. Given this key role in oncogenic signalling, several agents, antibodies and small molecules designed to target EGF/HER receptors, have been developed clinically. These include the antibodies cetuximab, panitumumab (EGFR), trastuzumab and pertuzumab (HER2) and the small molecule tyrosine kinase inhibitors (TKIs) gefitinib, erlotinib (targeting EGFR) and lapatinib (targeting EGFR/HER2). However, clinical efficacy for all these agents has been less pronounced than predicted based on preclinical models and responses are generally transient leading to the emergence of drug resistance. Intra-tumoral heterogeneity possibly underlies resistance of lung cancers to current therapies. Malignant pleural effusions (MPE) represent an opportunity to culture a wide variety of cancer cells from a single individual and possibly predict the response to a given therapy.
Materials and Methods: HER3 represents a crucial player in signal transduction and acquired resistance to TKIs, and a potential new drug target. In fact, HER3 has been shown to influence maintenance and survival of tumor cells through activation of the PI3K/AKT pathway. Since HER3 is devoid of kinase activity, it cannot be targeted by small molecule inhibitors. In contrast, antibodies are a viable strategy for pharmacological intervention. We have recently generated and started to characterize a set of five novel hybridomas directed against different epitopes of HER3. These antibodies significantly affected ligand-dependent signal transduction, viability and maintenance of cancer cells of different origins *in vitro*.
Results: In this study, we have characterized MPE primary cultures for ErbB receptors expression, phosphorylation and expression of the relevant ligands. In particular, HER3 was expressed at high levels and thus could be considered a relevant target for cancer stem-like cells. Importantly, anti-HER3 antibodies were capable of blocking signal transduction, cell proliferation and spheroid-forming activity.
Conclusions: We are currently establishing predictive *in vitro* and *in vivo* systems to test our anti-HER3 antibodies for their capability to block tumorigenicity of MPEs and revert acquired resistance to other antibodies and TKIs. Our results may have a significant impact on current standard therapies for lung cancer.