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In vivo efficacy of BI 2536, a potent and selective inhibitor of the mitotic kinase Plk1, in human hematopoietic cancers

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Background: Polo-like kinase 1 (Plk1), a key regulator of cell cycle progression and accurate spindle assembly, is an attractive target for cancer drug discovery. We have previously shown that BI 2536, a potent and selective small-molecule inhibitor of Plk1, induces a distinct mitotic arrest phenotype in prometaphase ("polo-arrest") with subsequent apoptosis in cell lines derived from a broad range of human epithelial cancers, irrespective of their oncogenome signatures. When used in vivo, BI 2536 given i.v. shows excellent activity in xenograft models of human epithelial cancers in nude mice at well-tolerated doses. BI 2536 is the first specific Plk1 inhibitor in clinical development and has demonstrated encouraging results in phase I trials. The present study was designed to extend the analysis of BI 2536 to human leukemias and lymphomas.
Material and Methods: A range of malignant human cell lines of hematopoietic and lymphoid derivation (Raji Burkitt's lymphoma; HL-60 acute myeloid leukemia; THP-1 acute monocytic leukemia; Granta-519 B-cell lymphoma; DOHH2 B-cell lymphoma) was used to evaluate the anti-tumor effects of BI 2536 in vitro using Alamar Blue™ assays. To assess the in vivo efficacy of BI 2536 in models of hematological cancers, C.B-17 SCID mice were inoculated intravenously with Granta-519 or DOHH2 B-cell lymphoma cells, respectively. Mice were treated with BI 2536 i.v. once weekly (50 mg/kg) or twice weekly on consecutive days (40 and 50 mg/kg). Body weight and clinical signs of progressive disease were monitored.
Results: BI 2536 potently inhibited proliferation in vitro with EC_{50} values of 6−14 nM. Cell cycle analysis by propidium iodide staining showed a G2/M arrest of BI 2536-treated leukemia and lymphoma cells at concentrations that matched closely the EC_{50} values in Alamar Blue™ assays. Immunocytochemical analysis of cytospin preparations of Granta-519 cells, using tubulin staining to visualize mitotic spindles and DAPI staining to visualize DNA, showed that cells arrest in mitosis with abnormal monopolar spindles.
In both lymphoma models tested, treatment with BI 2536 showed good efficacy with significant delay in the appearance of clinical signs of disease and prolonged survival compared to control animals.
Conclusion: BI 2536 is a potent inhibitor of leukemia and lymphoma cell proliferation in vitro and shows remarkable efficacy in two in vivo models of human B-cell lymphoma at well tolerated doses.

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Molecularly targeted immunochemotherapeutic formulation (SEVINA) composed of pegylated trispecific disulfide linked Fv (sdFv) targeting epitopes of EGFR, PTHrP and RANKL conjugated covalently with SATA to vinorelbine eradicated osteolytic metastases of multiple myeloma inhibiting proliferation of tumor associated and bone microvascular endothelial cells inducing ADCC, AMP and type I, II, III PCD

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Background: Bone metastasis is a common complication of multiple myeloma, and one for which only palliative treatment is available. We aim to inhibit the vicous cycle between tumor cells and the bone microenvironment, which results in increased tumor burden and bone destruction.
Materials and Methods: Multiple myeloma obtained from a metastatic patient were inoculated into the left cardia ventricle of nude mice. After osteoclastic metastases were observed radiographically, the animals were treated with pegylated trispecific disulfide linked Fv (sdFv) targeting epitopes of PTHrP, RANKL and EGFR conjugated covalently with SATA to vinorelbine-tartrate.
Results: Post-treatment, we observed autocrine inhibition of multiple myeloma cells, and paracrine inhibition of tumor associated endothelial cells, and bone microvascular endothelial cells. Downregulation of EGFR led to inhibition of expression of AP1 blocking transcription of VEGF, and its receptors KDR and Flt-1 exerting an angiocidal effect. EGFR downregulation led to reduced activity of osteoclasts, and inhibition of PI3K, MAPK and MMP-9. Downregulation of PTHrP and RANKL inhibited osteoclastogenesis. Subsequently, release of growth factors TGF-a and IGF from the bone matrix was blocked. Vinorelbine destabilised microtubule (MT) dynamics of tumor, and endothelial cells blocking cell cycle at G2/M. It also phosphorylated bcl-2 blocking uPAR and SP1 DNA binding activity through ERK signaling pathway. This led to inhibition of osteoclast proliferation by inhibiting hydrolysis of IGF-binding proteins, and downregulation of MMP-1, 3, 9. We observed antibody dependent cellular toxicity, and antibody mediated phagocytosis. In multiple myeloma cells, tumor associated and bone derived endothelial cells, we observed inhibition of DNA synthesis, and metabolic activity by BrdU and MTT analysis. Transmission electron microscopy exhibited nuclear PCD type III with pyknosis, and zeiosis leading to secondary necrosis. Type I PCD which is caspase-3 dept, and autophagic type II PCD led to a bystander killing effect of tumor and endothelial cells. There was not radiographic evidence of osteolytic bone metastases of multiple myeloma cells.
Conclusion: These findings suggest that this molecularly targeted immunochemotherapeutic formulation named SEVINA (under patent) may provide a novel therapeutic approach to inhibiting the processes involved in bone metastases from multiple myeloma.

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Phase I tolerability/safety of sunitinib in combination with capecitabine in patients (pts) with advanced solid tumors

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Background: Sunitinib malate (SU) is an oral, multitargeted tyrosine kinase inhibitor of VEGFR-1, -2 and -3, PDGFR-α and -β, KIT, RET, and FLT3, and is approved internationally for the treatment of advanced RCC and imatinib-resistant or -intolerant GIST. In a preclinical mouse xenograft model of human breast cancer, coadministration of sunitinib improved the antitumor activity of 5-FU. This phase I, dose-finding study assesses the maximum tolerated dose (MTD), tolerability, and safety of SU in combination with capecitabine (C) in pts with advanced solid tumors.
Materials and Methods: Entry criteria include diagnosis of an advanced solid tumor not amenable to curative therapy, previous treatment with $\leqslant$2 prior chemotherapy regimens, no prior antiangiogenic therapy, ECOG PS $\leqslant$1, and adequate organ function. C was always administered orally twice daily in 3 wk cycles (2 wks on treatment, 1 wk off). C and SU doses were alternately escalated in serial pt cohorts to determine the MTD of SU using a standard 3 + 3 design. In the first arm of the study, SU was administered once daily on a 6-wk cycle (4 wks on treatment, 2 wks off; 4/2 schedule). Following determination of the MTD on the 4/2 schedule, 2 additional arms (2/1 schedule [2 wks on treatment, 1 wk off] and a continuous dosing [CD] schedule) were assessed to determine the MTD of SU.
Results: As of Dec 2006, 48 pts have been enrolled: 28 on 4/2 schedule, 9 on 2/1 schedule, and 11 on CD schedule. Median cycles started (range) = 3 (1–9), 4 (2–9) and 2 (1–4), respectively. SU 50 mg + C 1000 mg/m^2 and SU 37.5 + C 1250 mg/m^2 on the 4/2 schedule exceeded the MTD. The MTD for the 4/2 schedule is SU 37.5 mg/d + C 1000 mg/m^2 bid; 1 dose limiting toxicity (DLT) was seen in 9 pts treated at the MTD (Grade 3 [G3] hand-foot syndrome $\geqslant$7 days). Across all doses on the 4/2 schedule, DLTs include G3 hand-foot syndrome (n = 3), G3 fatigue (n = 2), G3 myalgia (n = 1). 3 pts (thyroid, breast, NET cancers) treated on the 4/2 schedule (SU 37.5 mg/d + C 825, 1000 or 1250 mg/m^2 bid, respectively) achieved a confirmed partial response. Preliminary PK data do not indicate a drug-drug interaction. Pts continue to be accrued to the 2/1 and CD schedules.
Conclusions: In pts with advanced solid tumors, the MTD for the 4/2 schedule is SU 37.5 mg/d + C 1000 mg/m^2 bid. SU is being further studied on schedule 2/1 and CD. These preliminary data suggest that the SU + C combination is tolerable and further studies in breast cancer are warranted.

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Phase I dose-escalation study of brostallicin in combination with cisplatin (cDDP) in patients with advanced solid tumors

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Background: Brostallicin (B) is a new minor groove binder that interacts with DNA in presence of glutathione/glutathione transferase. Investigation of the combination is based on the novel mechanism of action of B, its