245 IMMUNE TARGETING OF THE PHOSPHOPROTEOME IN HEMATOLOGY-MALIGNANCY

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Introduction: CD8+ T cells have been identified as potent effectors of the adaptive anti-tumor immune response. So far, only a small number of tumor antigens have been objectively linked to the oncogenic processes. Alteration in phosphorylation status of cellular proteins is a hallmark of malignant transformation and a proven important therapeutic target. Phosphorylated antigens thus represent attractive immunotherapeutic targets.

Methods: Using a mass spectrometry approach phosphopeptide display was analyzed in CLL, AML, ALL, HCL and MCL samples. X-ray crystallography studies were performed on phosphopeptides on the local. Phosphopeptide-specific T cells were generated from lab donors using DCs and tested for activity in xenogenic models of lymphoma.

Results: Eight phosphopeptides have been characterized many derived from phosphoproteins known to function in signaling cascades implicated in neoplastic transformation including c-Myc, NFAT and Bcl-11 with many representing novel phosphorylation sites. Six HLA-phosphopeptide structures were resolved and demonstrate upward facing phosphate moiety with potential for direct recognition by the TCR. Phosphopeptide-specific primary T cell lines were also generated ex-vivo from healthy lab donors which bound HLA-phosphopeptide tetramers and recognized HLA-matched primary tumor samples. Xenograft studies revealed activity of adoptively transferred T cells into tumor-bearing NOD/SCID mice.

Conclusion: This work characterizes phosphopeptides that are differentially presented on primary leukemia and lymphoma samples by class I MHC molecules. These phosphopeptides therefore represent novel candidates for future cancer immunotherapy.

246 CLINICAL AND IMMUNOLOGIC RESPONSES TO A NOVEL IN SITU LYMPHOMA VACCINE MANEUVER: RESULTS OF A PHASE II TRIAL OF INTRA-TUMORAL CGP- [PF-3512676]

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Background: Therapeutic lymphoma vaccines are an appealing concept, but their cohort will guide the development of subsequent iterations of this trial. We demonstrated feasibility and safety. Objective clinical responses have our T cell response assays. There is excellent correlation between the different measures whom are clinically evaluable. There were no significant adverse events. Tumor at the As of January 2008, all of the planned 15 patients have been enrolled, 14 of these patients were used to design a method for multi-color flow cytometric analysis of T cell mediated, tumor-specific immune responses have been difficult to reproduce and clinically validate. A more rigorous approach in which patient responsiveness.

Methods: Patients with recurrent, low-grade lymphoma, received a vaccine maneuver consisting of 2 Gy irradiation to a single site of lymphoma and 10 weekly intra-tumoral injections of CGP-[PF-3512676] at the same site. Peripheral blood samples from these patients were used to design a method for multi-color flow cytometric detection of T cells. Independent markers of tumor-specific CD8 T-cell immunity: intracellular IFN-Y, CD3 and CD107.

Results: As of January 2008, all of the planned 15 patients have been enrolled, 14 of whom are clinically evaluable. There were no significant adverse events. Tumor at the treated site almost always responded to the local therapy. Evaluation of un.injected sites showed little or no change. We have been able to use a panel of B cell lymphoma lines and primary tumor specimens as allo-stimulators to establish ex-vivo T-cell response assays. There is excellent correlation between the different measures of CD8 T cell responsiveness. These assays will be used in an attempt to correlate clinical outcome with T cell response.

Conclusions: This is the first report of an intra-tumoral CGP based vaccine for lymphoma. We demonstrated feasibility and safety. Objective clinical responses have been documented. Correlation of immunologic findings and clinical responses in this cohort will guide the development of subsequent iterations of this trial.

** - authors contributed equally to this work

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247 THE IN VIVO IMMUNOGENICITY OF HUMAN IDIOTYPE-KLH VACCINES IS MARKEDLY ENHANCED BY MALEIMIDE CONJUGATION

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Tumor-specific immunoglobulin (idiotype, Id) can serve as a target for therapeutic vaccination against B cell malignancies. While Id chemically-coupled to the immunogenic foreign carrier protein keyhole limpet hemocyanin (Id-KLH) has shown promising results in phase III clinical trials, many immunized patients fail to mount anti-Id immune responses, and one phase III trial recently failed in its primary efficacy endpoint. Id is traditionally coupled to KLH using glutaraldehyde (glut). We developed an alternative Id-KLH conjugation method employing maleimide (mal) chemistry, that markedly improved anti-tumor efficacy in 3 different murine lymphoma models (A20, M513, and BCL-1) owing to increased induction of tumor-specific CD8+ T cells and antibodies. To move towards clinical application, we conjugated human monoclonal IgG (IgG1 or IgG3) to KLH using mal or glut and tested their immunogenicity in vivo. The immunoreactivity of polyclonal anti-human IgG, towards various conjugates was measured by ELISA. Surprisingly, glut conjugates of IgG1 and IgG3 showed only 1.16%-5% of the reactivity of free IgG, while that of mal conjugates was highly preserved (91.7-99.9%), indicating a substantial reduction in antigenic content after glut conjugation. Mice were vaccinated with various conjugates plus GM-CSF and sera tested for binding to each immunogen. In all four cases, mal-Ig-KLH resulted in higher (2.1 to 14.2-fold) anti-Id antibody titers than glut Ig-KLH (Table below). In conclusion, like murine lymphoma Id proteins, human mal-Ig-KLH conjugates are more immunogenic than glut conjugates. The observed destruction of human IgG antibody epitopes by glut may help explain the unfavorable results seen in some Id-KLH trials. Thus, maleimide Id-KLH therapeutic vaccines deserve testing in human lymphoma trials.

Table: Anti-id antibody levels in vaccinated mice (mcg/ml)

<table>
<thead>
<tr>
<th>IgG1 (Rituximab)</th>
<th>IgG1 (Trastuzumab)</th>
<th>IgG3 (Kappa)</th>
<th>IgG3 (Lambda)</th>
</tr>
</thead>
<tbody>
<tr>
<td>136</td>
<td>1950</td>
<td>454</td>
<td>420</td>
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248 ANTITUMORAL EFFECT OF VACCINATION WITH CD40L-GENETICALLY MODIFIED DENDRITIC AND TUMOR CELLS FUSIONS IN MYELOMA

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Introduction: Fusions of dendritic cells (DCs) and tumor cells represent a novel approach that combines the expression of molecules needed for immune stimulation with the presentation of a repertoire of tumor antigens. Several studies have shown the importance of triggering CD40 molecules to efficiently the efficiency of DC as antigen presenting cells. Fusions of dendritic and tumor cells stimulated through the CD40 pathway could enhance their capacity to stimulate specific antitumor T cells. We have studied the antitumoral effect of the administration of cell fusions transduced with a recombinant adenosine encoding the CD40L gene (AdvCD40L) in a murine model of myeloma.

Material and Methods: DCs obtained from bone marrow of Balb/c mice were fused with tumor cells, a syngenic murine myeloma cell line (4T10O). Fusion cells (FC) were generated in polyethylene glycol and selected after culturing in HAT medium plus GM-CSF for 7 days. FC were quantified by determining the percentage of cells that coexpress specific DCs (CD11c+) and tumor (CD138+) markers. FC were transduced with AdvCD40L (PC-CD40L) or a recombinant adenosine encoding green fluorescent protein (FC-GFP) as control. For vaccination, mice (n=10 per group) were injected i.v. with 2.5x10^6 tumor cells and treated with irradiated FC, PC-GFP or PC-CD40L (1x10^6 cells each, i.v.) on days 2, 6 and 10 after tumor challenge.

Results: Mean fusion efficiency was 30% (range, 20-40%). FC expressed moderate levels of CD80, CD86, CD54, and MHC class II. In contrast, PC-CD40L showed a significant increase of expression of costimulatory molecules (CD80, CD86, CD54, and MHC class II) compared to FC-GFP (p<0.01). 40% of mice treated with FC-CD40L showed a significant increase of expression of costimulatory molecules (CD80, CD86, CD54, and MHC class II) compared to FC-GFP (p<0.01). In parallel, mice treated with mixed cells (DC+Tumor cells without fusing), mix transduced with GFP or mix transduced with CD40L did not show a significant antitumor effect.
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Conclusions: Dendritic-tumor cells fusions transduced with recombinant viruses encoding CD40, gene stimulates antitumor immune responses in vivo and may provide a strategy for treating patients with myeloma or lymphoma.

249 ADVOCATE TRANSFER OF AUTOLOGOUS CD25-DEPLETED,
CD3/CD28-COSTIMULATED T-CELLS (ACTC) AFTER CYCLOPHOSPHAMIDE – FLUDARABINE CHEMOTHERAPY (CF)
ENHANCES LYMPHOCYTE RECOVERY AND REDUCES T
REGULATORY CELLS IN PATIENTS (PTS) WITH LOW-GRADE
FOLLICULAR LYMPHOMA (FL)

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Introduction: CF is effective therapy for pts with FL but causes immunosuppression, possibly limiting immunologic control of residual disease. CD4+CD25+FOXP3+ T regulatory cells (Trgs) can suppress cellular immune responses. Adoptive transfer of ACTC may improve disease control after CF by accelerating T-cell recovery and reducing Trgs.

Methods: We initiated a phase I study in pts with relapsed FL (grade 1 or 2). After leukapheresis, pts receive 4 cycles of CF. Four weeks after last CF, responding pts receive escalating doses of ACTC prepared from autologous T-cells collected before CF. ACTC were then infused at escalating doses for 3 cycles after CF.

Results: 14 pts have been enrolled: median age 49 (32–68), median prior therapies 2 (1–3). For 10 evaluable pts completing CF and ACTC, 8 pts achieved CR and 2 pts achieved PR after CF. 5 pts received 1–3 x 10^9 and 5 pts 3–5 x 10^9 CD3+ ACTC. There have been no adverse events due to ACTC. Following CF, median CD4 and CD8 counts were 91 (15–169) and 64 (19–273); four weeks after ACTC, median CD4 and CD8 counts were 592 (156–1053) and 309 (186–1300); median %CD4+FOXP3+ blood cells before CF was 15.8% (n=5; 3.07–37.1%); after CF/before ACTC 21.4% (n=7; 6.15–56.4%); on day 60 after ACTC 3.3% (n=8; 1.5–20.4%) (p=.01 for both pre ACTC and post ACTC comparisons).

Conclusions: ACTC after CF chemotherapy: (1) accelerates recovery of CD4+ and CD8+ lymphocyte numbers compared to historical controls; (2) enhances recovery of lymphocyte function; (3) results in reduction of peripheral blood FOXP3+ Trgs; (4) can result in a longer progression-free survival compared to traditional therapy.

250 IDIOTYPE VACCINATION OF UNTREATED B-CELL LYMPHOMA IS ASSOCIATED WITH DURABLE FREEDOM FROM CYTOTHERAPY
AND CELLULAR IMMUNE RESPONSES

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Tumor-specific immune responses induced by idiotype immunization of B-NHL patients correlate with prolonged remission and survival rates after cytotoxic chemotherapy. Conventional idiotype vaccines are coupled to the carrier KLH and are administered subcutaneously with adjuvant. Intradermal administration of recombinant idiotype Fab fragment with lipid-based adjuvant and subcutaneous coadministration of GM-CSF has excellent immunogenicity in advanced B-NHL patients (Bertiintetti et al., 2006). In a subsequent trial, 19 patients with untreated indolent B-NHL (13 follicular [FL], 3 nodal marginal zone [nMZL], 3 mantle cell [MCL]) and without immediate need for cytoreduction received at least 6 monthly idiotype vaccinations. After a median follow-up of 32 months, 9 patients (47%) are in progression-free survival (PFS), and 11 (53%) had no requirement of cytoreductive therapy (TFS). 5 patients (26%; only FL or nMZL) achieved an objective partial remission. Of these, 4 cases are in continuing remission after 18-37 months. IFNγamma ELISPot indicated a cellular immune response to the idiotype in 10/13 analyzed patients (77%). These responses were associated with superior PFS (p<0.05), and all 3 nonresponders eventually required cytoreductive therapy. 6/15 analyzed patients (40%) developed anti-idiotype IgG or IgM antibodies as assessed by ELISA. These responses were associated with the combined immune response rate was 85%. In a parallel cohort of 19 indolent lymphoma patients vaccinated in remission after cytoreductive therapy, 6 relapses (progressions) (32%) occurred during a median follow-up of 21 months, including 4 MCL. Cellular and humoral immune response rates in this group were 78% and 83%, respectively, but not associated with PFS.

In conclusion, intradermal idiotype vaccination has an excellent cellular immune response rate. Since durable responses and long-term freedom of remission for conventional therapy were observed after this active immunotherapy in otherwise untreated FL and nMZL, this vaccination warrants comparison to no treatment or passive immunotherapy. Given the possible importance of cellular immune responses, combined active and passive immunotherapy may be envisioned to act synergistically.

251 SYNERGISTIC EFFECT OF T-CELL ADOPTIVE IMMUNOTHERAPY WITH ANTI-CD19 OR ANTI-CD38 CHIMERIC RECEPTOR ON B-CELL LYMPHOMA IN CONJUNCTION WITH RITUXIMAB

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Introduction: Using artificial receptors it is possible to redirect the specificity of immune cells to tumor-associated antigens, a strategy that holds great potential as a cancer therapy. Since B-cell non-Hodgkin’s lymphoma (B-NHL) cells invariably express CD19 and CD38, these antigens are suitable molecular candidates for immunotherapy.

Material and methods: We transduced human peripheral T cells or T cell lines with an anti-CD19-chimeric receptor (CR) or anti-CD38-CR containing 4-1BB as well as anti-CD19 or anti-CD38 antibody-derived single-chain variable domain respectively.

Results: Retroviral transduction led to anti-CD19-CR or anti-CD38-CR expression in T cells with high efficiency. T cell line HuT78 retrovirally transduced with anti-CD19-CR or anti-CD38-CR possessed strong cytotoxic activity against the B-NHL cell lines BL and HT and in vitro simultaneously. To determine the synergy of two chimeric receptors, we incubated HuT78 anti-CD19 CR and/or anti-CD38 CR in the presence of HT cells. Interestingly, we found that only two sets of chimeric receptors made an additive cytotoxic effect on HT cells in vitro. To confirm the mutual effect of T cells with these chimeric receptors on lymphoma cells in vivo, human peripheral T cells expressing either anti-CD19-CR or anti-CD38-CR were synergistically effective in NOD-SCID mice inoculated subcutaneously with HT cells. Intriguingly, we found that T cells with either anti-CD19-CR or anti-CD38-CR enhanced cytotoxicity against HT cells in xenografted mice in conjunction with rituximab.

Conclusion: We demonstrated that simultaneous immunotherapy against different antigens augmented cytotoxicity to lymphoma cells in vitro and in vivo. These results may provide a rationale for clinical testing of autologous T cells with anti-CD19 CR or anti-CD38 CR in the presence of rituximab in patients with aggressive or relapsed B-NHL refractory to conventional therapy.

252 FILTERING OUT REGULATORY T CELLS AND CURING LARGE LYMOPHOMA TUMORS USING SYNGENIC IMMUNOTRANSPLANTATION

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Background: We previously described a vaccine maneuver against murine B-cell lymphoma combining chemotherapy (CTX) and in vivo injection of an immunostimulant – CpG, which induced T-cell immunity and eradicated established tumors. An ongoing clinical trial of this maneuver has demonstrated complete and partial responses in lymphoma patients, though immune-regulatory factors such as regulatory T cells (Treg) and limiting amounts of “homeostatic” cytokines might prevent even more powerful anti-tumor immunity. Transfer of tumor-specific lymphocytes into lymphodepleted patients may elicit these immune-regulatory targets. The clinical potency of this approach has been seen with ex vivo activated tumor-infiltrating lymphocytes, though, the use of in vivo vaccine-primed anti-tumor lymphocytes could make it sufficiently feasible for broader clinical application.

Methods: Donor mice were immunized with the CTX/Cpg vaccination and recipient mice were conditioned with lethal irradiation followed by transfer of syngenic bone marrow and immunized donor splenocytes, i.e. “immunotransplant”. Peripheral blood tumor-specific T cell responses were measured by flow cytometric analysis of intracellular IFNγ up-regulation on exposure to irradiated tumor. Transplanted animals were tested for tumor response.

Results: We show that transfer of T cells into the lymphodepleted recipient selectively filters against Treg, and doubles the transferred Treg/Teff ratio. Transferred tumor-specific CD8 T cells preferentially expand in the lymphodepleted recipient.

Immunotransplant cures large subcutaneous (> 100mm2) and metastatic tumors in 100% of recipients and enhances tumor-specific T cell memory. The addition of a vaccine “boost” to the immunotransplant maneuver, results in an additional 3-fold increase in tumor-specific immunity.

Conclusions: As hematopoietic stem cell transplantation is a standard therapy for lymphoma patients, pre-transplant vaccination followed by post-transplant transfer of tumor-specific T cells could be tested in clinical trials. Our model suggests that this immunotransplant approach would allow the benefit of cancer vaccines to be revealed and might lead to cure of otherwise resistant malignancies.
253 T CELL MODULATION COMBINED WITH INTRATUMORAL INJECTIONS OF CpG Oligodeoxynucleotides CURES LARGE AND SYSTEMIC LYMPHOMA TUMORS IN MICE

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Introduction/Background: In a murine B cell lymphoma model, we have previously shown that the combination of intratumoral CpG and chemotherapy can cure metastatic tumors (Li et al., J Immunol 2007). Although CpG induces an anti-tumor T cell response through the activation of antigen presenting cells, it requires the combination with chemotherapy to be effective. We now ask whether these immune responses can be enhanced by using T cell-specific antibodies, thereby eliminating the need for chemotherapy.

Material and Methods: Mice were challenged with A20 tumors at 2 different sites (right and left abdomen). Only one site was injected with CpG allowing us to evaluate the systemic anti-tumor response at the distant site. Treatment started when tumors became palpable. CpG was administered intratumorally and all monoclonal antibodies used to modulate the T cell responses were administered i.p. - Regulatory T cells were depleted using anti-Folate receptor 4 (FR4) antibodies or functionally blocked using anti-GITR antibodies. Effector T cells were triggered through their co-stimulatory molecules using anti-CD40 or anti-4-1BB antibodies, while inhibitory signals were blocked using anti-CTLA4 or anti-PD-L1 antibodies.

Results: Treatment with CpG alone cured no mice as expected, while the combination of CpG with each antibody alone only partially increased the cure rate (range: 0-40%). Selected antibodies (anti-CD40, anti-CTLA4, anti-FR4, and anti-GITR) were then used in various combinations, in conjunction with CpG. Three antibody combos showed strong efficacy, curing between 80 to 100% of the mice (anti-OX40+anti-CTLA4, anti-OX-40+anti-FR4, and anti-CTLA4+anti-FR4), whereas other combinations did not provide any significant improvement (anti-OX40+anti-GITR, anti-CTLA4+anti-FR4, anti-FR4+anti-GITR).

Conclusions: Our results show that the addition of specific antibodies against different functional T cell targets greatly enhances the therapeutic potency of intratumoral vaccination with CpG. Antibodies against these human targets are becoming available for clinical trials.

254 ANTI-CD19 BITE ANTIBODY MT103 (MEDI-358) INDUCES DURABLE OBJECTIVE RESPONSES IN PATIENTS WITH RELAPSED NON-HODGKIN’S LYMPHOMA (NHL): UPDATE FROM ONGOING PHASE I STUDY MT103-104

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Introduction: MT103/MEDI-358, a BITE antibody targeting the CD19 antigen, is a member of a novel class of molecules that direct T cells to target cells. A Phase 1 dose escalation study is conducted in patients with advanced NHL.

Methods: Relapsed incurable NHL patients requiring treatment were included. Most patients were heavily pre-treated with a median of 4 previous chemo/immuno therapy regimens. A classical 3+3 dose escalation was employed. To date, 6 dose levels ranging from 0.5 to 60 mg/m2 per day have been tested. MT103 was continuously infused as single agent over a period of 4-8 weeks. Objective responses assessed by anti-tumor activity in bone marrow and liver was assessed by histochemical analysis of biopsies obtained before and after treatment with MT103.

Results: To date 37 patients have been treated. Most common AE’s included lymphopenia, leukopenia and pyrexia. The majority of the AE’s improved or resolved during treatment. Few patients experienced fully reversible CNS events in terms of confusion and cerebellar symptoms. Dose-dependent responses in mantle cell lymphoma; follicular lymphoma and CLL were observed in 9 out of 25 patients starting at the dose level of 15 mg/m2/24hr. Four patients had complete and 5 had partial responses. Histochemical analysis of biopsies showed removal of target cells from bone marrow and liver. All responders had been pre-treated with rituximab-based combination regimens. At the highest dose level of 60 mg/m2/24hr administered to date, 5 out of 5 patients have shown objective responses. The first response observed in mantle cell lymphoma has been ongoing for 11 months and none of the responders at the two highest dose levels of 30 mg and 60 mg/m2/24hr has experienced treatment failure to date.

Conclusions: MT103/MEDI-358 as single agent induces durable responses in pre-treated NHL patients and is well tolerated. Dose escalation is ongoing.

255 PHASE I STUDY OF KW-0761, A HUMANIZED ANTI-CCR4 ANTIBODY, IN PATIENTS WITH RELAPSED ADULT T-CELL LEUKEMIA-LYMPHOMA (ATL) AND PERIPHERAL T-CELL LYMPHOMA (PTCL)

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Introduction: KW-0761 is a defucosylated humanized IgG1 monoclonal Ab against CC chemokine receptor 4 (CCR4) with enhanced ADCC activity. Previous studies revealed that CCR4 was overexpressed on tumor cells from 88% of pts with ATL and 38% of pts with PTCL, and was associated with poor prognosis. These results suggest that CCR4 could be a reasonable molecular therapeutic target.

Methods: A multicenter phase I study of KW-0761 is being conducted for relapsed pts with CCR4-positive ATL and PTCL to evaluate its safety, pharmacokinetics, immunogenicity and efficacy. Pts were planned to receive 4 weekly iv infusions of KW-0761 at 0.1, 0.5, 1.0 and 2.0 mg/kg.

Results: As of Jan 8 2008, 8 pts (7 ATL and 1 PTCL) were treated with KW-0761 at 1.0 (N=3), 0.5 (N=4), and 0.5 mg/kg (N=1). 1 pt was withdrawn without dose-limiting toxicities. Major toxicities included: hematologic: lymphopenia (G4: N=1, G3: N=2, G2: N=3), neutropenia (G3: N=1, G2: N=2), eosinophilia and thrombocytopenia (G2: N=1 each), non-hematologic: herpes zoster (3 months after the 4th dosing, G3: N=1), acute infusion reaction/cytokine release syndrome (G3: N=1, G2: N=3), constipation, rash, QTc prolongation and ALT elevation (G2: N=1 each). One pt was withdrawn due to early disease progression. PK analysis showed that Cmax at 0.01 and 0.1 mg/kg after the 4th dosing were 324±657 and 1835±438 ng/ml, respectively. T1/2 of each pt was between 60 and 80 hr. No anti-KW-0761 Ab has been detected. Preliminary investigator-assessed responses in 8 pts included 1 CR (1.0 mg/kg, PB & skin), 2 PRs (0.01 mg/kg, CR in PB & PR in LN, 0.1 mg/kg, CR in PB & PR in skin) and 3 SDs.

Conclusions: KW-0761 is a promising new Ab therapy for CCR4-positive ATL and PTCL. Pts accrual is ongoing and the updated results will be presented.
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257 PROMISING RESULTS OF EPRTUZUMAB AND RITUXUMAB IN COMBINATION WITH CYCLOPHOSPHAMIDE, DOXORUBICIN, VINCristine AND Prednisone CHEMOTHERAPY (ER-CHOP) IN PATIENTS WITH PREVIOUSLY UNtREATED DIFFUSE LARGE B-CELL LYMPHOMA

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Background: A priori pilot study of eprtuzumab (ImmunoMedics) and rituximab in combination with CHOP chemotherapy (ER-CHOP) in untreated patients with DLBCL demonstrated feasibility and safety. This multicenter phase II trial was carried out to assess efficacy.

Methods: Patients received immunochemotherapy on the following schedule: eprtuzumab 360 mg/m2, rituximab 375 mg/m2, and standard dose CHOP every 3 weeks for 6 cycles. Primary endpoint was the occurrence of any serious AE during treatment (FSU12).

Secondary endpoints were complete response rate (CR), overall response rate (ORR). An interim analysis was planned when the first 34 eligible patients were evaluable for EFSU12.

Results: This study has met full accrual, with 107 patients (pts) entered between Feb 2006 and Aug 2007. The median age was 62 years (range: 19-77), 67 were male. 60 pts had stage 3 or 4 disease; 57% had Ann Arbor stage IV, 43% stage III. 83% had bone marrow involvement. 72% had B symptoms, 45% had Stage III disease; 65% had prior chemotherapy. The ORR was 89% (21 CR, 10 PR), the CR rate was 76%. The median time to progression was 37 months (95% CI 32-42) and median OS was not reached. 1 pt had a serious AE of pulmonary embolism, with 85% of pts having some form of serious AE.

Conclusions: The combination of eprtuzumab and CHOP chemotherapy is feasible and active in this population. Further assessment of this combination is warranted. A randomized, Phase II comparison study is underway.

259 VORinostAT PROVIDES PROLONGED SAFETY AND CLInICAL BENEFIT TO PATIENTS WITH ADVANCED CTCL

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Background: Vorinostat (Zolinza®), a histone deacetylase inhibitor, was approved in 2006 by the FDA for the treatment of the cutaneous manifestations of CTCL in patients (pts) with progressive, persistent or recurrent disease on or following 2 prior systemic therapies.

Methods: Pts with stage ≥ IB CTCL who had received ≥ 2 prior therapies received oral vorinostat 400 mg daily, until intolerable toxicity or progressive disease (PD), in an open-label, Phase IB trial. The primary endpoint was objective response rate. This post hoc subset analysis reports results from patients who received vorinostat for ≥ 2 years.

Results: As of November 1 2007; 6 of 74 pts had received vorinostat for ≥ 2 years: median age 65 years (range 57-74), median number of prior therapies 2.5, median time from diagnosis to enrollment 1.8 years (range 0.7-5.9). 1 pt had a complete response, 4 a partial response and 1 stable disease. Diarrhea (100%), nausea (83%), fatigue (67%) and alopecia (38%) were the most common drug-related AEs. 1 pt had a serious AE of pulmonary edema, resolving in 7 days. This pt is continuing treatment as of Day 955. 1 pt who experienced serious AEs of increased creatinine phosphokinase (CPK) [Day 490] and rash (Day 455) remained on the study until Day 948. The other grade 3 or 4 AEs were anorexia (n=1) and thrombocytopenia (n=1); pt discontinued due to PD, 1 discontinued due to PD, rash and increased CPK, and 4 are continuing therapy. Updated data will be presented.

Conclusions: Vorinostat has demonstrated prolonged safety and clinical benefit in these pts with advanced CTCL.
261 LOW DOSE PRALATREXATE (PDX) IS ACTIVE IN CUTANEOUS T-CELL LYMPHOMA: PRELIMINARY RESULTS OF A MULTI-CENTER DOSE FINDING TRIAL

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Background: PDX is a novel antifolate with activity at a range of doses in rel/ref T-cell lymphoma (TCL). The maximum tolerated dose (MTD) from our phase I study was 30 mg/m² weekly for 67 weeks. In that study, responses were seen in patients (pts) with cutaneous T-cell lymphoma (CTCL). To explore this activity, we designed a trial of PDX in pt with CTCL. CTCL is more indolent than peripheral TCL and treatment paradigms use maintenance approaches. We sought a best tolerated, yet active dose and schedule for these unique pts by designing a dose reduction scheme.

Methods: Eligibility includes CTCL types: mycosis fungoides (MF), Sezary syndrome (SS), cutaneous anaplastic large cell lymphoma (ALCL) and progression of disease after 2 systemic therapy. The dosing scheme uses 2 schedules: 3/4 weekly and 2/3 weekly. Doses are reduced in sequential cohorts based on toxicity. Optimal dose and schedule is defined as activity without Grade (Gr) 4 heme tox, Gr 3-4 infection, or febrile neutropenia.

Results: From 80/7-1/08, 12 pts have enrolled. 11 with MF and/or SS, 1 with ALCL. 7 had prior therapies. The ORR was 56% (14/25) with all responses being PR; 9 additional pts exhibited tumor reductions including a CR and 3 PRs in DLBCL pts; with progression free survival (PFS) for responders ranging from 168 to > 336 days. Five DLBCL pts with stable disease had PFS ranging 112 to > 336 days. One of 10 FL pts with tumor reassessment had a PR. Comparison of the 85 and 110 mg cohorts revealed 16% & 53% of patients, respectively, with 2 grade 3 toxicity. PK evaluation revealed Tmax =1 hr, with biphasically elimination & plasma t 1/2 =6 hrs. Similar Cmax & AUC were observed at 85 (n=8) and 110 (n=17) mg, suggesting a trend towards saturable absorption. Inhibition of HDAC activity in PBMCs was seen in 13/18 pts and was similar between the 85 & 110 mg groups.

Conclusion: The preliminary activity of PDX in pts with CTCL at much lower doses than used for aggressive lymphomas. This study is ongoing to see how doses of PDX can result in maintained responses with minimal toxicity and will be updated.

262 ORAL MTOR INHIBITION WITH EVEROLIMUS IN RELAPSED T CELL AND HODGKIN LYMPHOMA

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Introduction: mTOR inhibition has demonstrated promising activity in phase II trials for aggressive B-cell non-Hodgkin lymphomas, including mantle cell and diffuse large B-cell lymphoma (DLBCL). MGCD0103 (2010) is a potent and selective inhibitor of mTOR (2010) with activity in preclinical models of both solid tumors and hematopoietic malignancies. This study tested oral everolimus in a phase II trial for patients with T cell lymphoma or Hodgkin lymphoma.

Methods: Open-label, Phase II trial (Trial 008) in adults with relapsed or refractory diffuse large B-cell lymphoma (DLBCL) or follicular lymphoma (FL). Eligibility required measurable disease (≥ 2.0 cm), ECOG 0 or 1. MGCD0103 given 3x/week starting at 110 mg after 32 patients pts) an 85 mg starting dose was evaluated. Response rate, PFS and toxicity were updated.

Results: 50 pts received treatment; 33 DLBCL & 17 FL median age [range]; 61.5 years [32-80]; male, n=27; prior rituximab therapy: 96%; prior stem cell transplant: 33%. The median age was 47.3 years. The patients were heavily pre-treated with a median of 6 prior therapies. Sixty percent of all patients (87.5% of HL) had undergone prior stem cell transplant. The ORR was 56% (14/25) with all responses being PR; 9 additional pts exhibited tumor reductions including a CR and 3 PRs in DLBCL pts; with progression free survival (PFS) for responders ranging from 168 to > 336 days. Five DLBCL pts with stable disease had PFS ranging 112 to > 336 days. One of 10 FL pts with tumor reassessment had a PR. Comparison of the 85 and 110 mg cohorts revealed 16% & 53% of patients, respectively, with 2 grade 3 toxicity. PK evaluation revealed Tmax =1 hr, with biphasically elimination & plasma t 1/2 =6 hrs. Similar Cmax & AUC were observed at 85 (n=8) and 110 (n=17) mg, suggesting a trend towards saturable absorption. Inhibition of HDAC activity in PBMCs was seen in 13/18 pts and was similar between the 85 & 110 mg groups.

Conclusion: MGCD0103 demonstrated significant anti-cancer activity in relapsed or refractory NHL (DLBCL and FL subtypes) and had a manageable side effect profile.

264 SAFETY AND TOLERABILITY OF VORINOSTAT – EXPERIENCE FROM THE VORINOSTAT CLINICAL TRIAL PROGRAM

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Background: We present the safety and tolerability data from patients (pts) who received vorinostat; a HDAC inhibitor with anticancer properties, alone or with other systemic therapies for solid and hematologic malignancies. Methods: Safety data from all pts in the Vorinostat Clinical Trial Program were collated (cut-off Dec 2007).

Results: 476 pts with CTCL, solid or hematologic malignancies received vorinostat as monotherapy (n=341) or in combination therapy (n=135). As monotherapy, the most common AEs were fatigue (68.3%), nausea (61.1%), diarrhea (35.4%), and anorexia (49.9%). Grade 3-4 AEs included thrombocytopenia (15.2%), fatigue (13.5%), dehydration (8.5%), and anemia (7.9%). There were 3 drug-related deaths (ischemic stroke, tumor hemorrhage, unspecified). Of 156 pts who received monotherapy at 400 mg q.d. (the dose approved by the FDA for advanced CTCL treatment), 13 (8.3%) discontinued due to AEs (anemia [1.9%], pulmonary embolism [0.6%]), and weight decrease [0.6%]), and 24 (15.4%) required dose modifications (commonly due to thrombocytopenia [5.8%], diarrhea [1.9%], and nausea [1.9%]. In combination therapy, the most common AEs were nausea (32.6%), diarrhea (43.0%) and fatigue (41.5%). Grade 3-4 events included fatigue (16.8%), diarrhea (5.9%), dehydration (5.2%), and nausea (5.2%). 1 drug-related death occurred (hemoptysis, NSCLC pt). 25 (18.5%) pts discontinued due to AEs (fatigue [10.0%] dehydration and nausea [both 2.2%]), and 23 (17%) required dose modifications (commonly due to fatigue [5.9%], diarrhea [1.9%] and hypertrophiccardiarnia [1.9%]).

Conclusion: Vorinostat has an acceptable safety and tolerability profile as monotherapy or in combination with other systemic therapies in cancer pts; dose modifications are usually not required in the majority of pts.
**Background:** KSP is a mitotic kinesin essential for cell cycle progression. SB-743921 (SB-921), a selective KSP inhibitor, blocks mitotic spindle assembly with cell cycle arrest in mitosis and cell death. In the first-in-humans (FIH) trial, the maximum tolerated dose (MTD) was 4 mg/m² q21 days (d), i.e., 0.2 mg/m²/d. Neutropenia was the major dose-limiting toxicity (DLT).

**Methods:** Phase I of this trial determines the MTD of SB-921 without prophylactic GCSF (pGCSF) in patients (pts) with Non-Hodgkin (NHL) or Hodgkin Lymphoma (HL). Eligible pts with relapsed or refractory lymphoma had at least 1 prior chemotherapy regimen, and had relapsed after or were not candidates for autologous stem cell transplant. SB-921 is given to cohorts of 3 on d1/d15 q28d, starting at 2 mg/m² and escalating by 1 mg/m². Cohorts expand to 6 if 1/3 pts have a DLT.

**Results:** To date, 32 pts have received ≥7 mg/m² of SB-921; 14 had HL; 18 had NHL (9 indolent; 9 aggressive). Thirty-one pts had ≥2 prior chemotherapy regimens (14 had ≥5). Cycle 1 neutropenia ≥ grade 3 occurred in 7 pts. Two DLTs at 6 mg/m² occurred with ≥ grade 3 neutropenia; one was noted after escalation to 7 mg/m², a dose tolerated through cycle 1 without DLT by 3 pts. Cycle 1 drug-related non-hematologic toxicities ≥ grade 3 were infection (1), dehydration (1) and dyspnea (1). A HL pt had a partial response (PR) for 4 cycles at 6 mg/m²; a NHL pt began at 3 mg/m² and escalated to 5 mg/m² with stable disease for 13 cycles.

**Conclusions:** SB-921 is well tolerated without pGCSF at < 6 mg/m² and possibly at ≥7 mg/m² given d1/d15 q28d in the FIH trial (possibly ≥ 0.5 vs. 0.2 mg/m²/d). The 6 mg/m² cohort was expanded and may allow re-escalation to ≥7 mg/m². A HL pt had a PR at 6 mg/m². If the DLT without pGCSF is neutropenia, the MTD will next be determined with pGCSF.