62.5 mg/kg for 3 consecutive days (days 0, 1, 2). PET imaging was carried out on days −1 (pretreatment baseline), 1 (24 h after dose 1) and 3 (24 h after dose 3).
Results: During the first treatment cycle, mean tumor volume in the placebo group increased by approximately 49% vs a 6% decrease and 7% increase in the groups treated with GSK923295A at 125 mg/kg and 62.5 mg/kg treated groups, respectively. Subsequently (day 7 onwards), tumor regression (>50% decrease in tumor volume relative to pre-treatment volume) was observed in 11/13 mice dosed at 125 mg/kg, but only 4/14 dosed at 62.5 mg/kg. FDG-PET imaging showed significant decreases in mean FDG standardized uptake values (SUV) relative to placebo at both dose levels. On day 3, the mean SUV's in GSK923295A-treated tumors decreased by approximately 25% and 30% at 62.5 and 125 mg/kg respectively. The day 3 SUV's for both treatment groups were significantly lower than either day −1 (pre-treatment) ($p < 0.05$) or placebo ($p < 0.001$).
Conclusion: The results demonstrate that in a preclinical setting, GSK923295A treatment significantly affected FDG uptake early after onset of therapy and prior to tumor regression. FDG-PET may provide a means of evaluating pharmacodynamic activity in patients treated with GSK923295A.

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Preclinical validation of the TrpM8 ion channel as a cancer target

D.F. Duncan[1], N. Provost[2], O. Moreno[1], M.W. Frohlich[3], D. Urdal[4].
[1]Dendreon Corporation, Chemistry, Seattle, USA; [2]Dendreon Corporation, Product Development, Seattle, USA; [3]Dendreon Corporation, Clinical Affairs, Seattle, USA; [4]Dendreon Corporation, Research, Seattle, USA

Background: TrpM8, a transmembrane cation channel protein, is expressed in cancers including prostate, breast, lung, and colon. In normal tissues, its expression is primarily confined to prostate and a subset of sensory neurons. We have previously reported that small molecule agonists of TrpM8 can selectively kill cells that express TrpM8.
Methods: Activity and selectivity of agonists were measured in vitro in cell killing assays using CHO cells and CHO cells stably transfected to express TrpM8 (CHO/TrpM8). Cell viability was used to determine EC_{50} values. Plasma pharmacokinetics (PK) was determined in mice. Compounds were assessed in vivo in murine xenograft models using CHO and CHO/TrpM8 cells. Tumor growth inhibition (%TGI) was defined as the difference between the control and treated tumor volumes as a % of control. The plasma PK of the lead compound was evaluated in rats and dogs. In vivo activity was evaluated in human prostate cancer (LuCap) xenograft models and in a rat model of androgen-induced, benign prostate hyperplasia (BPH).
Results: Several compounds were identified that were potent and selective in vitro and efficacious in vivo via oral administration. Compound D-3263 proved to be the most potent and preliminary PK studies conducted in mice, rats and dogs suggest the plasma half-life ranges from 10 hrs (mice and rats) to 27 hrs (dogs). The compound appears to inhibit LuCap growth in Nude mice ($p = 0.004$, 123.7 mm^3 for treated vs 207.6 mm^3 for vehicle on Day 4) and inhibited androgen-induced hyperplasia of normal rat prostates (mean prostate weights for treated 620.1 g vs 1009.9 g for untreated; $p = 0.004$).

Table 1

Compound	MW	*In vitro* EC50 (uM)		*In vivo* %TGI
		CHO/TrpM8	CHO	
D-3263	373	0.003	>10	70%
D-3457	368	0.01	>10	62%
D-3517	346	0.01	>10	48%

Conclusions: These results establish TrpM8 as a tractable therapeutic target and endorse the likelihood of an agonist demonstrating clinical activity against cancers that express TrpM8. The orally bioavailable small molecule agonist D-3263 has been selected for clinical development.

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A phase Ib dose escalation study to evaluate safety and tolerability of the combination of the aminopeptidase inhibitor CHR-2797 and paclitaxel in patients with advanced or treatment refractory tumors

C. van Herpen[1], F. Eskens[2], M. de Jonge[2], I. Desar[1], L. Hooftman[3], E. Bone[3], A. Timmer-Bonte[1], J. Verweij[2]. *[1]Radboud University Medical Centre Nijmegen, Department of Medical Oncology, Nijmegen, The Netherlands; [2]Erasmus University Medical Center Rotterdam, Department of Medical Oncology, Rotterdam, The Netherlands; [3]Chroma Therapeutics, Oxford, United Kingdom*

Background: CHR-2797 is an orally bioavailable aminopeptidase inhibitor. In a single agent phase I study in solid tumors the recommended daily dose was 240 mg. The active metabolite, CHR-79888 accumulates intracellularly. In vitro and in vivo experiments confirmed good synergy between CHR-2797 and paclitaxel.
Methods: Patients with histologically confirmed advanced solid tumors refractory to standard therapy and performance score $\leqslant 2$ were treated every 21 days with paclitaxel given i.v. over 3 hrs (escalating from 135 mg/m^2 in the first cohort to 175 mg/m^2 in subsequent cohorts) and escalating doses of CHR-2797 (90–240 mg). The first 21 day cycle (Cy) constituted the dose finding phase. Patients received up to 6 Cy of paclitaxel and could remain on CHR-2797 therapy until evidence of PD or unacceptable toxicity.
Results: 22 patients (median age 59 years [range 34−72], 18M/4F) were treated. At the 1st dose level (DL) 3 patients were given paclitaxel 135 mg/m^2 and 90 mg CHR-2797. DLs 2−5 received paclitaxel 175 mg/m^2 and CHR-2797 at 90, 130, 180, and 240 mg (4, 3, 9, and 3 patients respectively). One DLT (dyspnea G3) was seen at DL 4. Common Grade 1−3 toxicities during treatment included alopecia and fatigue (each in 95% of patients), sensory neuropathy (59%), myalgia (50%), anorexia and dizziness (each in 45% of patients), rash (32%). Infusion reactions developed in 13 (59%) patients. At DL 4 and 5 CHR-2797 was withheld for 5 days, from d18 of each Cy, in an attempt to decrease the risk of infusion reactions. However, this had no clear effect on the number of infusion related reactions. Six patients continued CHR-2797 after discontinuation of paclitaxel. Neither agent influenced the pharmacokinetics of the other. PR was achieved in 3 patients (melanoma, non small cell lung cancer, esophageal squamous cell).
Conclusions: Except for an unexpected high number of infusion reactions to paclitaxel the combination of paclitaxel-CHR-2797 was otherwise well tolerated. Further investigation into the potential immunological mechanisms is warranted. Formal MTD was not reached. Anti-tumor activity was observed in several patients.

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Myofibrillogenesis regulator 1 as a potential target for cancer therapy

R. Shao[1], K. Ren[1], Y. Wang[1]. *[1]Institute of Medicinal Biotechnology CAMS, Department of Oncology, Beijing, China*

Background: The phosphorylation of myosin light chain-2 (MLC2) is pivotal in the regulated assembly and disassembly of focal adhesions and adherens junctions contributed to cell motility and tumor invasion. MLC kinase inhibitor abrogates MLC2 phosphorylation, cell polarization and migration, and MLC2 dephosphorylation induces apoptosis. Our previous data show that MLC2 interacts with human myofibrillogenesis regulator 1 (MR-1). Thus, we investigate whether MR-1 is associated with the proliferation and migration of cancer cells.
Material and Methods: The transcription and expression of MR-1 were detected by RT-PCR and Western blot analysis. Functional analysis of MR-1 small hairpin RNA (shRNA) was conducted in HepG2 cells using Lipofectamine-mediated gene transfer. The changes of cell spreading, adherence and migration in response to the treatments were evaluated by immunofluorescent staining, immunohistochemistry and Boyden chamber invasion assay. The tumorigenicity of HepG2 cells stably transfected with MR-1-shRNA was assessed by transplantation into nude mice.
Results: RT-PCR and Western blot analysis showed that MR-1 was overexpressed in human cancer cells and especially in hepatoma HepG2 cells. Transient treatment of cells with shRNA against MR-1 or stable transfection of cells with plasmid expressing MR-1-shRNA led to impairment of cell proliferation, adhesion and migration. Following inhibition of MR-1 by MR-1-shRNA, the phosphorylations of MLC2, focal adhesion kinase (FAK) and Akt were decreased dramatically and formation of stress fiber was destroyed. In the same condition, MLC kinase inhibitor could block exogenous MR-1-induced phosphorylations of MLC2, FAK and Akt and F-actin polymerization inhibitor also decreased phosphorylations of FAK and Akt, indicating that activation of MLC2 and intact actin cytoskeleton was upstream of FAK and Akt in MR-1 modulating pathway. *In vivo* data showed that knockdown of MR-1 markedly inhibited the tumorigenicity of human