1473 POSTER

Tocosol® paclitaxel and cremophorel®-paclitaxel: the pharmacokinetic comparison shows that a new paclitaxel formulation leads to increased drug exposure

A.-R. Hanauske[1], L. Goedhals[2], H. Gelderblom[3], Y. Lee[4], A. Awada[5], J.B. Vermorken[6], C. Bolling[1], A. Ruiz-Garcia[7], J. Pratt[7], M.B. Stewart[7]. [1]*AK St. George, Hamburg, Germany;* [2]*National Hospital Bloemfontain, South Africa;* [3]*Leiden University Medical Center, Leiden, Netherlands;* [4]*Harborview Hospital Baltimore, Baltimore, USA;* [5]*Institut Jules Bordet, Brussels, Belgium;* [6]*Antwerp University Hospital, Edegem, Belgium;* [7]*Sonus Pharmaceuticals, Botham, USA*

Background: TOCOSOL® Paclitaxel (TP) is a novel tocopherol-based cremaphor-free formulation that allows a 15 minute infusion time. Phase II clinical trials have demonstrated antitumor activity in bladder, ovarian, and non-small cell lung cancers. The pharmacokinetic (PK) comparison of TP and CremophorEL®-formulated paclitaxel (P) is important for further development of TP.
Methods: 36 patients with solid tumors without proven treatment option were enrolled. Each patient was randomized to receive as first course either a single infusion of 175 mg/m^{2}of TP given over 15 minutes q 21 days or 175 mg/m^2 of P given over 3 hours q 21 days. In a second cycle the other formulation was administered so that each patient served as his/her own control. Blood samples for PK of free and total paclitaxel were obtained up to 120 hours after each dose; non-compartmental and compartmental PK analyses were performed. Adverse events and complete blood counts were assessed every 3 days.
Results: TP given over 15 minutes produced a mean 67% higher exposure to free paclitaxel and a mean 108% higher exposure to total paclitaxel compared to P administered over 3 hours. The geometric mean ratio of free paclitaxel AUC values after equal 175 mg/m^2 doses of TP and P was 1.67 (90% CI 1.56–1.79, $p < 0.0001$) and that for total paclitaxel AUC values was 2.08 (90% CI 1.97–2.20, $p < 0.0001$). Mean values for paclitaxel terminal elimination t2 were similar for TP and P. TP produced more neutropenia than P, but without causing clinical complications. Grade 1–3 non-hematologic adverse events were comparable between the two drugs.
Conclusions: TP offers a significant increase of drug exposure with similar tolerability. The differences in exposure may reflect differences in rate and/or extent of dissociation of paclitaxel from the tocopherol emulsion formulation compared to cremaphor-ethanol micelles. TOCOSOL® Paclitaxel is easily administered and may provide a profoundly higher drug exposure if given on a weekly basis. Since this may result in improved clinical activity compared to P further development is warranted.

1474 POSTER

A Phase I, open-label study with escalating doses of intravenous PTK787/ZK 222584, a multi-VEGF receptor inhibitor, followed by multiple oral daily dosing to assess the absolute bioavailability of PTK787/ZK 222584 after single and multiple doses in advanced cancer patients

M. Pavel[1], D. Schuppan[2], L. Jost[3], A. Harris[4], D. Kerr[5], T. Jalava[6], E. Masson[7], M.L.D. Palacay-Radona[7], C. Köhne[8]. *1. University of Erlangen-Nuernberg, Erlangen, Germany;* [2]*Beth Israel Deaconess Medical Center, Harvard Medical school, Boston, MA, USA;* [3]*Medizinische Universitätsklinik, Kantonsspital Bruderholz/Onkologie, Switzerland;* [4]*Medical Oncology Unit, Oxford, United Kingdom;* [5]*University of Oxford, Oxford, United Kingdom;* [6]*Schering AG, Berlin, Germany;* [7]*Novartis Pharmaceuticals, East Hanover, New Jersey, USA;* [8]*Klinikum Oldenburg, Germany*

Background: PTK/ZK is a novel oral small molecule that blocks the tyrosine kinase activity of all known VEGF receptors and inhibits tumor angiogenesis and lymphangiogenesis, slowing tumor growth and spread.
Material and Methods: This Phase I study was performed to evaluate the safety and pharmacokinetics (PK) of single intravenous (IV) doses of PTK/ZK and the absolute bioavailability of the oral (PO) PTK/ZK after single (SD) and multiple doses (MD). Patients had histologically confirmed advanced cancer, adequate organ function and WHO PS of 0–2. The study was done in two parts. In Part 1 cohorts of 3 pts received single IV PTK/ZK doses of 22.5 mg, 45 mg and 90 mg on day 1 and PK samples were taken. Pts were monitored for safety until day 8. In Part 2 pts received a single IV dose of PTK/ZK 90 mg on day 1, and 1250 mg/d PO from days 8 to 22 and second IV dose of PTK/ZK 90 mg on day 23. PK was taken on days 1, 8, 22 and 23. Levels of PTK/ZK and it's metabolites were measured by HPLC/UV. Pts could continue with PO PTK/ZK until disease progression or unacceptable toxicity. The results of follow-up will be reported later.
Results: 26 pts were enrolled (9 to Part 1 and 17 to Part 2). PTK/ZK was the major active analyte in plasma. PK profiles of PTK/ZK following SD and MD oral administration with a dose of 1250 mg/d were similar to those observed in previous studies. Average PK parameters following PTK/ZK IV dose of 90 mg and PO dose of 1250 mg is shown in table 1:

Parameters (Unit)	Day 1 SD IV N = 20	Day 23 MD IV N = 12	Day 8 SD PO N = 17	Day 22 MD PO N = 12
Cmax (ng/mL)	2074	2462	9146	6457
AUC0-t (ng*h/mL)	5579	5018	63334	25307
Clearance(L/h)	18.5	25.2	31.6	71.5
Vz (L)	65.5	104	229	509
$t_{\frac{1}{2}}$ el.(h)	2.90	3.28	7.00	5.31

The mean (CV%) absolute bioavailability of PTK/ZK was 0.58 (55%) after SD and 0.37 (53%) after MD and was significantly lower after MD as compared to SD [Geo. mean ratios (90% CI); 1.6 (2.22 –2.10)]. PTK/ZK was well tolerated. No significant toxicities were reported in any of IV dose cohorts. The most frequent AEs with IV administration were nausea and fatigue and with PO administration dizziness, nausea and vomiting. Most of AEs were grade 1/2. No grade 4 AEs were observed. Two pts had grade 3 PTK/ZK related SAEs. Five pts discontinued Part 2 of the study because of AEs.
Conclusions: Both IV and PO PTK/ZK were well tolerated. The absolute bioavailability of PTK/ZK was 0.58 after SD, and declined to 0.37 after MD due to increase in oral clearance following multiple oral doses of PTK/ZK.

1475 POSTER

ErbB3 expression predicts tumor cell radiosensitization induced by Hsp90 inhibition

H. Dote[1], D. Cerna[1], W. Burgan[1], K. Camphausen[2], F. Tofilon[1]. [1]*NIH/NCI, Molecular Radiation Therapeutics Branch, Bethesda, USA;* [2]*NIH/NCI, Radiation Oncology Branch, Bethesda, USA*

The ability to identify tumors that are susceptible to a given molecularly targeted radiosensitizer would be of clinical benefit. Towards this end, we have investigated the effects of a representative Hsp90 inhibitor, 17-(dimethylaminoethylamino)-17-demethoxygeldanamycin (17DMAG), on the radiosensitivity of a panel of human tumor cell lines. 17DMAG was previously shown to enhance the radiosensitivity of a number of human cell lines, which correlated with the loss of ErbB2. We now report on cell lines in which 17DMAG induced the degradation of ErbB2, yet had no effect on radiosensitivity. In a comparison of ErbB family members, ErbB3 protein was only detectable in cells resistant to 17DMAG-induced radiosensitization. To determine whether ErbB3 plays a casual role in this resistance, siRNA was used to knock down ErbB3 in the resistant cell line AsPC1. Whereas individual treatments with siRNA to ErbB3 or 17DMAG had no effect on radiosensitivity, the combination, which reduced both ErbB2 and ErbB3, resulted in a significant enhancement in AsPC1 radiosensitivity. In contrast to siRNA to ErbB3 or 17DMAG treatments only, AsPC1 cell exposure to the combination also resulted in a decrease in ErbB1 kinase activity. These results indicate that ErbB3 expression predicts for tumor cell susceptibility to and suggest that the loss of ErbB1 signaling activity is necessary for 17DMAG-induced radiosensitization. However, for cell lines sensitized by 17DMAG, treatment with siRNA to ErbB2, which reduced ErbB1 activity, had no effect on radiosensitivity. These results suggest that, whereas the loss of ErbB1 signaling may be necessary for 17DMAG-induced radiosensitization, it is not sufficient.

1476 POSTER

Phase I study of S-1 plus cisplatin (CDDP) in patients with advanced non-small-cell lung cancer (NSCLC): a 2-week course of S-1

H. Sakai[1], Y. Ichinose[2], K. Kubota[2], H. Kunitoh[2], K. Nakagawa[2], K. Takeda[2], N. Yamamoto[2], N. Saijo[2], M. Ariyoshi[2], M. Fukuoka[2]. [1]*Saitama Cancer Center, Thoracic Oncology, Saitama, Japan;* [2]*S, .S-1 Cooperative Study Group*

Background: A combination of S-1 (tegafur, oxonic acid, and CDHP), an oral drug active against advanced NSCLC, plus CDDP resulted in a 47% response rate in our previous study, which used a relatively low dose intensity (DI) of CDDP (12 mg/m^2/week) [Clin Cancer Res 10: 7860, 2004]. This study examined the maximum tolerated dose (MTD) and recommended dose (RD) of CDDP when combined with a 2-week course of S-1 to increase the DI of CDDP in patients with advanced NSCLC.

Material and methods: The eligibility criteria included either histologically or cytologically documented stage IIIB or IV NSCLC, a performance status 0 or 1, an age of ⩽74 years, adequate organ function, no prior chemotherapy, and written informed consent. S-1 was administered orally at a dose of 40 mg/m^2 bid (80 mg/m^2/day) on days 1–14 of a 21-day cycle. CDDP was administered intravenously on day 1 of each cycle (level 0, 1, 2: 60, 70, 80 mg/m^2). Treatment was started at level 1 (70 mg/m^2), and the dose was increased to level 2 (80 mg/m^2) on the basis of DLT. However, if severe toxicity or other safety-related problems occurred, the dose was reduced to level 0 (60 mg/m^2). The MTD and RD were decided by globally evaluating toxicity, completion of treatment according to protocol, and response.
Results: Eighteen patients were enrolled (12 men; median PS = 1). One patient had DLT at level 1 (S-1/CDDP at 80/70). No DLT occurred at level 2 (S-1/CDDP at 80/80). However, some adverse events meeting the criteria for DLT occurred after 2 cycles, and 6 additional patients were given level 0 (S-1/CDDP at 80/60). No DLT occurred at level 0. On the basis of these results, level 1 was determined to be the MTD, and 60 mg/m^2 the RD of CDDP.
Conclusions: Level 0 (S-1/CDDP at 80/60) is the RD for S-1 plus CDDP in patients with advanced NSCLC. This is an active combination against advanced NSCLC, and a multicenter phase II study is now in progress (supported by Taiho Pharma Japan).

1477 POSTER

Jasmonates can overcome drug resistance induced by p53 mutations and P-glycoprotein expression

E. Flescher[1], D. Reischer[1], O. Fingrut[2], A. Harel[2], I. Zan-Bar[1]. [1]*Tel Aviv University, Human Microbiology, Tel Aviv, Israel;* [2]*Sepal Pharma, Ness-Zionna, Israel*

Background: Jasmonates are plant stress hormones which we discovered as novel anti-cancer agents in vitro and in vivo. Given the urgent need for new chemotherapeutic agents to combat drug-resistant tumors, we decided to evaluate the activity of jasmonates against various types of drug-resistant cancer cells. We chose to study two major mechanisms of drug resistance: p53 mutations, occurring in more than half of human cancers, and multi-drug resistance mediated by P-glycoprotein (P-gp) expression.
Methods: Various drug resistant cells were studied. In addition, we compared the response of two pairs of clones: 1. B lymphoma cells – a clone expressing wild type p53 (29M6.2, wt p53) versus a clone expressing mutated p53 (29M6.10, mut p53); 2. Melanoma cells (B16-F10.9) – a clone expressing low levels of p-gp (low P-gp) versus a clone expressing high levels of P-gp (high P-gp). p53 and P-gp expression were determined by Western blotting analysis. Cytotoxicity was measured by a tetrazolium-based assay, and necrotic versus apoptotic death was determined by flow cytometry.
Results: Methyl jasmonate (MJ) killed efficiently prostate adenocarcinoma, glioblastoma and histiocytic lymphoma cells expressing mutated p53, as well as breast adenocarcinoma cells expressing both mutated p53 and high levels of P-gp. Jasmonic acid (JA) and MJ were each equally cytotoxic towards the wt p53 and mut p53 clones, whereas the mut p53 clone was resistant to treatment with the radiomimetic agent neocarzinostatin, as well as to bleomycin, and cisplatin. Neocarzinostatin and bleomycin induced an elevation in the p53 levels in the wt p53 clone whereas MJ did not. MJ induced mostly necrotic death (and apoptotic death in about 20% of the cells) in the wt p53 clone, but only necrotic death in the mut p53 clone. JA and MJ were each equally cytotoxic to the low P-gp and high P-gp clones, whereas the high P-gp clone was resistant to treatment with colchicine, doxorubicin and vinblastine. Inhibition of P-gp in the resistant cells, with the P-gp inhibitor PSC-833, sensitized the cells to the chemotherapeutic agents but did not increase the cytotoxicity of MJ.
Conclusions: Our findings suggest that jasmonates can circumvent the resistance of mutant p53- and P-gp-expressing cells towards chemotherapy. Jasmonates can induce p53-independent cell death, and do not appear to be P-gp substrates. Jasmonates are therefore promising candidates for the treatment of drug-resistant tumors.

1478 POSTER

Dual inhibition of mTOR and of the Insulin like Growth Factor Receptor-1 completely blocks the PI3K/Akt/mTOR pathway and results in a supra-additive anti-tumor effect: a mechanistic-based combination approach

S. Di Cosimo[1], M. Scaltriti[1], F. Rojo[1], D. Val[1], M. Guzman[1], J. Seoane[1], J. Jimenez[1], J. Arribas[1], F. Cognetti[2], J. Baselga[1]. [1]*Vall d'Hebron University Hospital, Medical Oncology, Barcelona, Spain;* [2]*Regina Elena Cancer Institute, Medical Oncology "A", Rome, Italy*

RAD001, an oral derivative of rapamycin, inhibits mTOR, a protein kinase downstream of PI3K and Akt, involved in the regulation of cell growth, proliferation and survival. We and others have previously shown that RAD001 has anti-tumor activity *in vitro* and *in vivo*. In our preclinical studies as well as in ongoing phase I pharmacodynamic study, RAD001 administration results in an increase in activated (phosphorylated) Akt (p-Akt). In prior studies (Di Cosimo, AACR 2005) we reported that gefitinib, an anti-epidermal growth factor receptor TKI, has the capacity to inhibit pAKT when given alone but does not fully reverse RAD001-induced pAkt suggesting that Akt activation as result of mTOR inhibition occurs via an alternative mechanism/s.
In exploratory studies, we observed a RAD001 dose-dependent induction of insulin receptor substrate-1 (IRS-1), which serves as a docking protein for SH2 domain containing proteins, including PI3K, both in prostate (DU145) and breast (MCF-7) cancer cell lines. The RAD001-induced increased expression of p-Akt and IRS-1 was also seen *in vivo* by immunohistochemical analysis of MDA-MB-468 human breast cancer xenografts. Since IRS-1 is tightly regulated by insulin-like growth factor receptor 1 (IGF-1R), we analyzed IGFR-1R signaling in RAD001 treated cells. First, in DU145 cells, treatment with IGF-1 resulted in a strong induction and posterior degradation of IRS-1. Similarly, p-Akt was rapidly induced (30 minutes) and then gradually decreased (12 hours). Conversely, the addition of RAD001 resulted in a sustained induction of both IRS-1 and p-Akt, for up to 36 hours. In order to dissect the mTOR-dependent pathways that regulate Akt activation, experiments with siRNA demonstrated that a 70% knock down of the down-stream effector of mTOR S6K mimics the effect of RAD001 by increasing the levels of p-Akt and preventing the degradation of IRS-1. These results suggest that the inhibition of S6K by RAD001 results in a sustained activation of IGF-1R signaling which in turn results in the activation of Akt. Next, we demonstrated that the IGF-1R TKI NVP-AEW-541, at a dose of ⩾1 μM inhibits the IGF-1R pathway and prevents the activation of Akt induced by either siRNA-S6K or RAD001. Combined treatment with RAD001 (3, 6 and 12 nM) and NVP-AEW-541 (1.5, 3 and 6 μM) resulted in supra-additive growth inhibitory effects in a panel of different cancer cell lines (DU145, BT-474, MDA-MB-468), as demonstrated by the median effect analysis of growth curve.
In summary, the IGF-1R pathway is involved in the activation of Akt by RAD001 and dual mTOR and IGF-1R targeting is a mechanistic-based combined anti-tumor strategy. Our findings provide a rationale for combining the inhibitors of mTOR with agents targeting growth factor receptors as a novel anticancer approach.

1479 POSTER

ZK-EPO, a novel epothilone B derivative, significantly inhibits tumor growth in a wide range of human tumor models

U. Klar[1], B. Buchmann[1], W. Schwede[1], W. Skuballa[1], R. Lichtner[2], J. Hoffmann[1]. [1]*Schering AG, Berlin, Germany;* [2]*Signature-Diagnostics, Potsdam, Germany*

Background: Epothilones represent a novel class of natural products that stabilize microtubules and exhibit marked antiproliferative effects. ZK-EPO is a novel epothilone analog that has shown marked preclinical activity.
Material and methods: A broad panel of cell lines derived from normal cells or a variety of human tumors (eg, colorectal, prostate, breast) were exposed *in vitro* to different concentrations of ZK-EPO and compared with standard cytotoxic compounds (eg, paclitaxel, adriamycin, cisplatin, and camptothecin). *In vivo*, the efficacy of ZK-EPO was compared with standard cytotoxic compounds (eg, paclitaxel, cisplatin, doxorubicin) in human tumor xenografts. These included breast and pancreatic cancer, melanoma, ovarian (OVAR Ca-2, OVCAR 8, A2780/AD10), cervical (A431), and non-small-cell lung (NCI-H460) carcinomas that were grown in nude or SCID mice.
Results: *In vitro*, ZK-EPO showed significantly greater activity in all human tumor cell lines examined compared with paclitaxel, adriamycin, cisplatin or camptothecin, with a mean IC_{50} value of <1 nM after 3 days' incubation. ZK-EPO also exhibited significant activity compared with paclitaxel, and all other agents examined, in cell lines expressing marked resistance to the standard cytotoxics. ZK-EPO showed no toxicity against quiescent non-malignant cells (HUVEC, HaCat) and, unlike paclitaxel, showed high accumulation in tumor cell nuclei *in vitro*. *In vivo*, ZK-EPO was highly effective in a wide spectrum of human tumor models, both non-resistant (OVCAR 8, OVCAR 3, MaTu, OVAR Ca-2) and resistant (MaTu/Dox, NCI/ADR, A2780/AD10), exhibiting superior activity compared with paclitaxel. It was also active in melanoma and pancreatic cancer models representing tumors intrinsically resistant to almost every chemotherapeutic agent.
Conclusions: ZK-EPO exhibits significantly better activity compared with paclitaxel, and other agents, in a range of *in vitro* and *in vivo* tumor models. ZK-EPO is the first fully synthetic epothilone in clinical trials designed to overcome multi-drug resistance and to combine high efficacy with an improved therapeutic window. ZK-EPO's ability to circumvent resistance mechanisms in preclinical models suggests that it may postpone or prevent the development of resistance when used in the clinic.