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Clinical report

Phase I and pharmacokinetic study of the orally administered farnesyl transferase inhibitor R115777 in patients with advanced solid tumors

CJA Punt,[1] L van Maanen,[1] CJJG Bol,[2] WF Seifert[2] and DJTh Wagener[1]

[1]Department of Medical Oncology, University Medical Center St Radboud, 6500 HB Nijmegen, The Netherlands. [2]Janssen Research Foundation, 2340 Beerse, Belgium.

R115777 is a novel selective inhibitor of farnesyl transferase, an enzyme that is involved in the proliferation of the malignant cell type. This study was designed to determine the toxicity, maximal tolerated dose and pharmacokinetics of R115777 when given orally b.i.d. for 28 days followed by 1–2 weeks of rest. Patients with advanced solid tumors for whom no standard therapy was available could enter the study. The starting dose of R115777 was 200 mg/dose and inter- as well as intra-patient dose escalations were performed with increments of 100 mg/dose. Nine patients entered the study and received in total 23 treatment cycles. A dose of 300 mg b.i.d. proved feasible with grade 4 neutropenia occurring in one of six patients who completed the first treatment cycle. Other toxicities were infrequent. Pharmacokinetic analysis demonstrated that peak plasma concentrations of 881 ± 393 ng/ml were reached within 1–5 h. No accumulation of R115777 was observed over a 28-day period. The study was terminated based on these results together with the observation from a related phase I study in which higher doses of R115777 were associated with the frequent occurrence of grade 3–4 myelosuppression. We conclude that the recommended dose of R115777 given for 28 days followed by 1–2 weeks of rest is 300 mg b.i.d. Myelosuppression is the dose-limiting toxicity. [© 2001 Lippincott Williams & Wilkins.]

Key words: Farnesyl transferase inhibitor, pharmacokinetics, phase I, R115777, *ras*, solid tumors

Introduction

The need for new chemotherapeutic agents in cancer is evident from the limited capacity of existing agents to cure or significantly prolong the survival of patients with advanced solid tumors. Laboratory studies over the last several years have demonstrated the importance of post-translational protein prenylation in the functioning of malignant cells. The most well-characterized system of protein prenylation is that involving the Ras proteins.

ras proto-oncogenes produce 21 kDa GTP binding proteins that are involved in the signal transduction pathways regulating cell growth and differentiation. Ras is synthesized as a cytosolic precursor that ultimately localizes to the cytoplasmic face of the plasma membrane after a series of post-translational modifications.[1] This process is initiated by the post-translational attachment of a farnesyl moiety to the protein and is catalyzed by farnesyl protein transferase (FTPase). Farnesylation of Ras is required for activity and is the rate-limiting step in post-translational modification of the protein. As potent and selective farnesyl transferase inhibitors became available, they were shown to have a broad spectrum of antitumor activity in human tumor cell lines and xenografts. The activity did not correlate with the presence or absence of *ras* mutations.[2–6]

R115777 is a non-peptidomimetic, competitive inhibitor of FTPase. R115777 shows anti-proliferative effects in a wide variety of cell lines both *in vivo* and *in vitro* independent of the presence or absence of *ras* mutations (End DW *et al.*, submitted). We performed a dose-escalation study of R115777 with a twice daily oral administration (b.i.d.) for 28 days followed by 7–14 days of rest in patients with advanced solid tumors. Previous pharmacokinetic data showed that a b.i.d. dosing regimen reached plasma concentrations which correspond to levels associated with antitumor effects in pre-clinical models.[7] The objectives of the current study were to assess the safety and the maximal tolerated dose

Correspondence to CJA Punt, Department of Medical Oncology, University Medical Center Nijmegen St Radboud, PO Box 9101, 6500 HB Nijmegen, The Netherlands.
Tel: (+31) 24 3615215; Fax: (+31) 24 3540788;
E-mail: c.punt@onco.azn.nl

(MTD), and to determine the pharmacokinetics of R115777 at this schedule.

Patients and methods

Patients were required to meet the following entry criteria: pathological confirmation of advanced solid tumor for which no potentially curative therapy is available, life expectancy of $\geqslant 3$ months, age $\geqslant 18$ years, ECOG performance status (PS) 0–2, adequate unassisted oral intake, previous chemo- or radiotherapy completed >4 weeks prior to study entry (6 weeks for nitrosureas/mitomycin C), negative pregnancy test for females with reproductive potential, normal serum bilirubin, serum creatinine $<1.5\times$ the normal upper limit (ULN), serum transaminases $<2\times$ ULN ($<5\times$ ULN in the case of liver metastases), WBC $>3.5\times10^9$/l, ANC $>1.5\times10^9$/l, platelets $>100\times10^9$/l and written informed consent. Patients with previous high-dose chemotherapy with bone marrow or stem cell rescue were not eligible. Concurrent radiation therapy, chemotherapy, hormonal therapy or immunotherapy was not allowed. Patients who participated in a clinical trial involving an investigational drug in the past 30 days or had been enrolled concurrently in another investigational trial were not eligible. The study was approved by the review board of the institution. Patients received R115777 as oral capsules b.i.d. immediately after a meal for 28 consecutive days (defined as one cycle) followed by a rest period of 7–14 days (depending on the recovery of toxicity). R115777 was provided by Janssen Research Foundation. Toxicity was scored according to NCIC Expanded Common Toxicity criteria. Patients were considered evaluable if they had completed at least one treatment cycle. Dose-limiting toxicity (DLT) was defined as grade 3 or 4 toxicity for both hematologic and non-hematologic events or treatment interruption more than 14 days, and a reasonable possibility that the event could be attributed to the drug. DLT determination was not limited to any particular cycle due to the intra-patient dose escalation design. If DLT was observed all intra- and inter-patient dose escalation was to be stopped. At least one patient was treated at each dose level. Dose escalation was performed at increments of 100 mg/dose. In the case of DLT in a patient, a maximum of six patients were to be treated at that dose level. In the absence of a DLT, intra-patient dose escalation was allowed in subsequent cycles with increments of 100 mg/dose. The MTD was protocol-defined as the dose level preceding the level at which equal two or more of six patients experienced DLT. The initial starting dose of 200 mg b.i.d. was selected since this dose at this schedule had already proven feasible in a related study.[8] Patients were followed for toxicity weekly during the first cycle and biweekly during subsequent cycles. Tumor evaluation was performed pre-study, after the first cycle and thereafter every two cycles. Since in a 3-month rat toxicity study (but not with other species) a routine ophthalmological examination revealed lens opacities an ophthalmologic examination was planned at the same time points at which tumor evaluation was performed.

Pharmacokinetics

Plasma samples were collected during the first cycle on day 1 and 28 at 0 (pre-dose), 0.5, 1, 2, 3, 5, 8 and 12 h after the first dose, and a pre-dose sample was collected at day 2, 7, 14 and 21. During the second cycle a pre-dose sample was collected at day 14 and 28. Heparinized blood was centrifuged immediately after collection and stored at -20°C until analysis. The plasma concentration of R115777 was determined by a validated HPLC method.[7] The following pharmacokinetic parameters were calculated by standard procedures: maximum plasma concentration (C_{max}), time to maximum plasma concentration (t_{max}), minimum concentration in plasma (C_{min}) and area under the plasma concentration–time curve over a 12 h dosing interval calculated by trapezoidal summation (AUC_{12h}). Elimination half-lives were not determined, because wash-out of R115777 was only followed for 12 h. The accumulation ratio was calculated as the AUC_{12h} ratio of day 28 and 1.

Results

Patients

Nine patients entered the study, six males and three females. Median age was 53 years (range 29–65), median PS 1 (0–2), tumor types were colorectal (n=3), soft tissue sarcoma (n=2), duodenal (n=1), gastric (n=1), renal (n=1) and medullary thyroid carcinoma (n=1). All patients had received prior chemotherapy: six patients with one, two with two and one with three regimens. Also two patients had received prior immunotherapy and one patient prior hormonal therapy. The number of cycles per patient at a given dose level is summarized in Table 1. Patient were treated with R115777 doses between 200 and 500 mg b.i.d.. The calculated dose intensity for these doses R115777 $\times$ 28 days followed by 7–14 days of rest is between 1.9 and 5.6 g/week, respectively.

Table 1. Treatment-related grade 3 and 4 toxicities

Patient no.	Dose (b.i.d.)	Cycle	Grade 3–4 toxicities
1	200	1	
	300	2	grade 3 diarrhea[b] and vomiting[b]
	200[a]	3–4	
2	300[c]	1	grade 4 leukopenia and neutropenia, grade 3 fatigue
3	300	1	
	400[c]	2	
4	300	1	
	400	2–5	
	500	6	grade 3 leukopenia and grade 4 neutropenia
	400[d]	7–9	
5	300[a]	1	grade 3 nausea[b]
6	300[c]	1	grade 3 fatigue[b]
7	300[a,e]	1	
8	300	1	grade 3 dyspnea[b] and fatigue[b]
	400[c]	2	
9	300	1	
	400[c]	2	

[a]First cycle not completed.
[b]Relationship to study drug considered doubtful or absent.
[c]Treatment was stopped at the end of cycle due to progressive disease.
[d]Treatment was stopped due grade 2 dyspepsia, doubtful if related to drug medication.
[e]Treatment was stopped due to grade 2 abdominal pain, possibly related to drug medication.

Toxicity

One patient experienced DLT at 300 mg b.i.d. in the first cycle (grade 4 leukopenia and neutropenia) and therefore the number of patients at this dose level was expanded to six (Table 1). In four of these patients grade 3 diarrhea and vomiting, grade 3 nausea, grade 3 fatigue, and grade 3 dyspnea and fatigue occurred, respectively, but the relationship to R115777 was considered doubtful or absent by the investigator. Other grade 3–4 toxicities did not occur at this dose level. Two patients did not complete the first cycle of 300 mg b.i.d. because of rapid disease progression and withdrawal of consent, respectively. One patient experienced grade 3 leukopenia and grade 4 neutropenia at 500 mg b.i.d. which did not recur after dose reduction to 400 mg b.i.d. In both patients with grade 4 neutropenia a complete recovery was documented within 2 weeks without febrile episodes. Fatigue, nausea, vomiting, anorexia and diarrhea were the most frequent non-hematological toxicities which were generally mild and not dose limiting. Follow-up ophthalmologic examinations were performed in five patients, including one patient with pre-existing glaucoma, and no abnormalities or change from baseline were observed. Based on these results, together with the results from a related, interdose escalation study[8] in which a dose of 400 mg b.i.d. for 13 and 25 days had resulted in DLT (myelosuppression), it was decided to close the study prematurely.

Tumor response

One patient with metastatic gastric carcinoma (Table 1, patient 4) who had progressive disease at study entry after two previous chemotherapy regimens remained stable for a period of 9 months. All other patients had disease progression after 2–16 weeks of treatment.

Pharmacokinetic analysis

The first dose data on day 1 (n=9), steady-state data on day 28 (n=6), and pre-dose data on days 2, 7, 14 and 21 were analyzed. The individual and mean ($\pm$SD) pharmacokinetic parameters are listed in Table 2. A representative concentration–time profile is presented in Figure 1. Peak plasma concentrations (C_{max}) were reached within 1–5 h after drug intake, and ranged from 511 to 1552 ng/ml on day 1 and from 431 to 1503 ng/ml on day 28. Pre-dose data (C_{min}) before the morning dose on day 2 and 28 averaged 95.7 and 72.4 ng/ml, respectively. The mean accumulation ratio was 0.85 ± 0.10, indicating no accumulation. The results indicate that steady state was maintained throughout the 28-day dosing period (see also Figure 1).

Discussion

In this trial we attempted to define the MTD of the new farnesyl transferase inhibitor, R115777, when administered orally b.i.d. for 28 days to patients with advanced solid tumors. A dose of 300 mg b.i.d. proved feasible, with grade 4 neutropenia occurring in one of six patients who completed the first cycle. Diarrhea was the only other grade 3 toxicity that could definitely be related to treatment. Since myelosuppression also occurred at a higher dose levels of 400 and 500 mg b.i.d. in a related study,[8] our study was terminated prior to the formal MTD determination since it was deemed unlikely that further dose escalation to 400 mg would be feasible.

Analysis of pharmacokinetic parameters showed no accumulation of R11577 over the 28-day period of dosing. Our results on R115777 pharmacokinetics were comparable to the previously reported data on R115777 given for 5 days b.i.d. followed by 9 days of

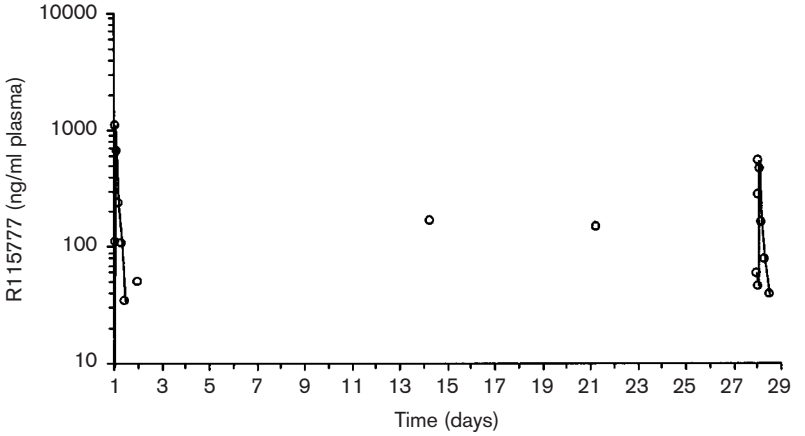

Figure 1. The concentration–time profile of R115777 for patient 9 after receiving 300 mg R115777 b.i.d. for 28 days. It should be noted that the scheduled pre-dose samples on day 14 and 21 were actually taken after drug intake.

Table 2. Pharmacokinetic parameters of R115777

Parameter	Patient									*n*	Mean ± SD
	1	2	3	4	5	6	7	8	9		
Day 1											
b.i.d. dose (mg)	200	300	300	300	100	300	300	300	300		300
t_{max} (h)	2.1	5.0	2.0	3.0	2.1	3.0	5.0	2.0	2.0	7	3.1 ± 1.3
C_{max} (ng/ml)	800	681	568	601	584	1164	511	1552	1092	7	881 ± 393
AUC_{12h} (ng·h/ml)	2713	4580	2333	3397	2157	5240	3675	5427	3216	7	3981 ± 1138
Day 28											
b.i.d. dose (mg)	200	300	300	300	300	300	300	300	300		300
t_{max} (h)	2.0	NA	2.0	3.3	NA	3.0	NA	1.0	2.0	5	2.3 ± 0.9
C_{max} (ng/ml)	431	NA	578	555	NA	1284	NA	1503	545	5	893 ± 464
AUC_{12h} (ng·h/ml)	1998	NA	1956	2823	NA	4814	NA	5245	2269	5	3421 ± 1508
Accumulation ratio AUC_{12h}	0.74	–	0.84	0.83	–	0.92	–	0.97	0.71	5	0.85 ± 0.10

[a]Patient 5 received 100 mg b.i.d. on day 1 and 2, thereafter 300 mg b.i.d., and is therefore excluded from the descriptive statistics.
NA, not available.

rest.[7] We observed no obvious correlation between pharmacokinetic data and toxicity.

The calculated dose intensity for our recommended dose of 300 mg b.i.d. R115777 × 28 days followed by 7–14 days of rest is between 2.8 and 3.4 g/week. This is consistent with the results of a study in which R115777 was given b.i.d. for 21 days followed by 7 days of rest, which had a MTD of 300 mg b.i.d. resulting in a calculated dose intensity of 3.1 g/week.[9] The DLT observed in that trial was also myelosuppression. A third trial using a treatment schedule of R115777 for 5 days followed by 9 days of rest yielded a lower dose intensity.[7] The protocol-defined MTD was not achieved in this trial (only one DLT of grade 3 peripheral neuropathy in six patients who received R115777 at 1300 mg). Analysis of toxicity and pharmacokinetic data from all cycles resulted in a recommended dose of 500 mg b.i.d. for 5 days every 14 days. The calculated dose intensity would be 2.5 g/week with that schedule. Objective tumor responses were not documented although the disease stabilization of 9 months in a pre-treated patient with gastric cancer was remarkable. In conclusion, the recommended dose of R115777 given orally b.i.d. for 28 days every 5–6 weeks is 300 mg b.i.d.

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