Cancer Chemother Pharmacol (2007) 59:139–147
DOI 10.1007/s00280-006-0267-9

CLINICAL TRIAL REPORT

A phase I study of the safety and pharmacokinetics of edotecarin (J-107088), a novel topoisomerase I inhibitor, in patients with advanced solid tumors

Herbert I. Hurwitz · Roger B. Cohen · J. Patrick McGovren · Samit Hirawat · William P. Petros · Yutaka Natsumeda · Tomoko Yoshinari

Received: 16 June 2005 / Accepted: 9 May 2006 / Published online: 4 July 2006

Abstract
Purpose To assess the maximum tolerated dose, safety, and pharmacokinetic (PK) profile of escalating doses of the novel topoisomerase I (topo I) inhibitor edotecarin (J-107088) given as a 2-h intravenous (IV) infusion once every 21 days in patients with advanced solid tumors who had not responded to standard therapy.
Patients and methods Twenty-nine patients (18M:11F) received a 2-h IV infusion of edotecarin in doses of 6, 8, 11, 13, or 15 mg/m^2 every 21 days (with an additional 1–2 weeks permitted for recovery) and were evaluated for safety, PK, and tumor response.
Results The most common non-hematologic toxicities were grade 1–2 nausea, fatigue, anorexia, vomiting, and fever. The most common hematologic toxicities were grade 1–2 thrombocytopenia and grade 3–4 neutropenia, leukopenia, and anemia. No grade 3–4 diarrhea was reported. Dose-limiting toxicities were observed in four patients at the 15 mg/m^2 dose and one patient at the 13 mg/m^2 dose. These toxicities included grade 3 nausea, vomiting, headache, and fatigue, as well as grade 4 neutropenia and febrile neutropenia. The maximum tolerated dose was declared at 15 mg/m^2. One patient with bladder cancer had a confirmed partial response at a dose of 13 mg/m^2. There was a trend to dose-proportional increases in edotecarin peak plasma concentrations and area under the curve values. Renal excretion of edotecarin was minimal (3–8% of the dose).
Conclusion The recommended Phase II dose of edotecarin is 13 mg/m^2 once every 21 days. The toxicities in this study were those typical of cytotoxic chemotherapy and less severe than those associated with other topo I inhibitors. The observed safety profile and preliminary evidence of clinical benefit warrant further investigation of this drug as monotherapy or part of combination therapy in patients with solid tumors.

Keywords Edotecarin · J-107088 · Pharmacokinetics · Maximum tolerated dose · Topoisomerase I inhibitor · Solid tumor

Presented at the American Society of Clinical Oncology 2000 Annual Meeting, abstract #767.

H. I. Hurwitz (✉) · W. P. Petros
Division of Hematology and Oncology,
Duke University Medical Center, P.O. Box 3052,
Durham, NC 27710, USA
e-mail: hurwi004@mc.duke.edu

R. B. Cohen
University of Virginia Health System,
Charlottesville, VA, USA

J. P. McGovren
Pfizer Inc., New York, NY, USA

S. Hirawat
PTC Therapeutics, South Plainfield, NJ, USA

Y. Natsumeda · T. Yoshinari
Banyu Pharmaceutical Co. Ltd., Tokyo, Japan

Introduction

Topoisomerase I (topo I) is a nuclear enzyme that relaxes supercoiled DNA during replication by

transiently breaking and rejoining DNA strands. Complex molecular interactions between topo I, DNA, and topo I inhibitory drugs ultimately result in double-strand breaks in DNA and tumor cell death in experimental cancer models. Irinotecan and topotecan are semisynthetic derivatives of the plant alkaloid camptothecin. The clinical activity of topo I inhibitors is well established. For metastatic colorectal cancer, irinotecan (CPT-11) is effective in the second-line setting as monotherapy and in the first-line setting when combined with 5-fluorouracil [1–4]. Topotecan has demonstrated activity in advanced ovarian cancer [5] and small-cell lung cancer [6, 7].

Edotecarin (J-107088) is a structurally novel topo I inhibitor that is related to indolocarbazole antibiotics, such as rebeccamycin and BE13793C [8, 9]. Edotecarin differs from camptothecin in several respects. First, camptothecin and its analogs cause DNA cleavage at a T↓G/A site whereas edotecarin mediates cleavage at a C/T↓G site [10]. Second, the ternary complex formed between DNA, topo I, and edotecarin appears to be more stable than that formed by camptothecin. As a result, edotecarin inhibits topo I enzyme at lower concentrations of drug and is eightfold more potent at inducing DNA cleavage than camptothecin [11]. The persistence of cleavable complexes in vitro long after drug removal may explain its relative lack of schedule dependence in preclinical models. In preclinical studies, edotecarin had an improved therapeutic index compared with topotecan, doxorubicin, cisplatin, and paclitaxel [10]. Like the camptothecins, the cytotoxic effects of edotecarin are not significantly altered by the multidrug resistance phenotype because edotecarin is not a substrate for the p-170 glycoprotein drug efflux pump [12, 13]. In preclinical studies in animal species, edotecarin is largely eliminated as unchanged parent drug via biliary excretion, in marked contrast to the camptothecin analog irinotecan, which is subject to very complex disposition and metabolic pathways. The unique in vitro and in vivo pharmacological profiles of edotecarin relative to other topo I inhibitors makes this compound a potentially useful antineoplastic agent.

Materials and methods

Patient population

Patients were eligible for this study if they were ≥18 years of age, had a histologically or cytologically confirmed solid tumor that was either resistant to standard therapy or was previously untreated due to: lack of an effective therapy; a life expectancy of at least 12 weeks; adequate bone marrow function defined as an absolute neutrophil count (ANC) ≥1,500/mm^3, a platelet count ≥100,000/mm^3, hemoglobin ≥9 g/dL; creatinine <1.5 mg/dL; adequate liver function defined as a total bilirubin within normal limits, and alanine transaminase, aspartate transaminase, and alkaline phosphatase <2.0x upper limit of normal; and an Eastern Cooperative Oncology Group (ECOG) performance status of ≤2. The eligibility criteria were: any chemotherapy, radiotherapy, or immunotherapy within 4 weeks prior to the first dose of edotecarin; within 6 weeks of receiving mitomycin C or nitrosoureas; within 2 weeks of major surgical intervention; any central nervous system metastases; radiation treatment to more than one-third of the bone-marrow; patient at high risk of complications due to significant co-morbid conditions, such as clinically significant cardiac, pulmonary, renal or hepatic disease, or there was evidence of an uncontrolled, or chronic, active infection, such as HIV. Patients with any medical, social, or psychological factors that might increase safety risks or prevent compliance or who were pregnant were excluded. Routine prophylactic use of colony-stimulating factors was not permitted. Also, patients who were being treated with corticosteroids, anticoagulants, immunotherapy, biological response modifiers, other investigational agents, or who had received a bone marrow stem cell transplant were excluded.

This study was conducted in accordance with the Declaration of Helsinki (1996) [14] and the International Conference on Harmonization (ICH) Guideline for Good Clinical Practice (1996) [15]. At each center, the protocol was reviewed and approved by the local Ethics Committee and a signed informed consent was obtained from all patients before they participated in the study.

Study design

This was an open-label, uncontrolled, dose-escalating, Phase I, single-agent study. Patients were treated once every 21 days in the absence of dose-limiting toxicity (DLT) and tumor progression. Toxicity was assessed according to the National Cancer Institute Common Terminology Criteria (NCI CTC), version 2.0. DLT was defined by events in cycle 1, as follows: (1) any grade 3 or 4 non-hematologic toxicity (except nausea and grade 3 vomiting that responded to symptomatic management; fever that was managed by maximum standard medical treatment; fatigue that responded to standard supportive measures; and grade 3 catheter-related infection); (2) grade 4 neutropenia (<500/mm^3) that lasted for five or more days, (3) grade 4

thrombocytopenia of any duration; (4) febrile neutropenia, defined as grade 4 neutropenia with fever of ≥39.1°C and/or infection requiring antibiotic or antifungal therapy; or (5) failure of the patient's peripheral blood counts to recover to ANC ≥1,500/mm^3 and/or platelet count ≥100,000/mm^3 within 35 days of day 1 dosing.

The starting dose of edotecarin was 6 mg/m^2, administered by a 2-h intravenous (IV) infusion every 21 days with 1 to 2 weeks permitted for recovery from toxicities, if needed. Successive cohorts of patients received higher dose levels with escalation in approximately 33% increments. The actual doses administered were 6, 8, 11, 13, and 15 mg/m^2. Escalation to the next dose level was not permitted until all patients treated at the previous dose level had completed follow-up (21–35 days). If any of the first three patients treated at a given dose level experienced DLT, additional three patients were treated at that level. The maximum tolerated dose (MTD) was defined as the dose at which two or more patients in a cohort (i.e., >1/3 of patients) experienced a DLT. Once the MTD was reached, an additional six patients were treated at the immediately preceding dose, which was defined as the recommended Phase II dose.

Treatment

Patients were assigned to a dose cohort at enrollment. Edotecarin (Banyu Pharmaceutical Co. Ltd., Tokyo, Japan) was administered by IV infusion over 2 h at a concentration of 0.15 mg/mL in 5% dextrose solution. The dose was controlled by varying the infusion rate (i.e., 0.63–1.56 mL/min). In cycle 1, patients experiencing nausea and vomiting within 24 h of drug administration were treated with granisetron. For nausea and vomiting that occurred more than 24 h after drug administration, patients could receive standard antiemetics. In subsequent cycles, granisetron could be administered prophylactically for nausea and vomiting and dexamethasone could be administered prophylactically for fatigue attributable to edotecarin. During the study, the protocol was amended (11 mg/m^2 dose level) to permit pre-medication with dexamethasone and granisetron in cycle 1.

Patients were discontinued from the study if they had progressive disease, a DLT, withdrew consent, or were not expected to have further clinical benefit from the treatment in the judgment of the investigator. In general, no dose modifications were allowed. The only exception was for patients who achieved an objective response and who experienced a DLT. These patients could continue treatment at a lower dose level once ANC and platelet counts recovered to grade 1 and any other toxicity had resolved to grade 2 or less.

Patients without signs of tumor progression or DLT were eligible for additional cycles of edotecarin therapy for the duration of the study (12 months).

Evaluations

Patients were monitored for safety (vital signs, hematology, serum chemistry, and urinalysis) during each cycle of therapy. Physical examinations were performed before treatment, weekly during cycle 1, and before drug administration on day 1 of each subsequent cycle. Adverse events were monitored throughout the study.

Clinical and radiographic tumor assessments were performed before the first treatment and after every second cycle of therapy. The overall tumor response was categorized using the WHO response criteria for solid tumors. Complete or partial responses were to be confirmed at least 4 weeks after the first observed response.

Pharmacokinetic sampling schedule

Edotecarin plasma concentrations were measured during cycle 1. Peripheral blood plasma samples were collected before infusion, at 15 and 60 min after the start of the infusion, immediately prior to completion of the infusion, and at 5, 15, 30, 45, 60, and 90 min and at 2, 3, 4, 6, 8, 12, 24, 48, and 72 h after completion of the infusion. Blood was mixed by repeated inversion and placed in an ice and water bath, then centrifuged at 1,100–1,300 *g* for 10 min in a refrigerated centrifuge to separate plasma from formed elements. One milliliter of plasma was then pipetted into each of the two separate vials and immediately frozen at −70°C. Urine samples were collected before the start of the infusion and for 0–24, 24–48, and 48–72 h after completion of the infusion; aliquots of urine were stored at −70°C until analysis. Upon collection of the final pharmacokinetic (PK) sample (at the 72-h time point), all samples for that patient were shipped on dry ice via overnight courier to the bioanalytical facility.

Measurement of edotecarin concentration in plasma and urine

Plasma and urine samples were analyzed by high-performance liquid chromatography (HPLC). Briefly, 0.1 mL aliquot of each plasma sample, 0.1 mL of internal standard solution (300 ng/mL J-109404; Banyu), and 0.2 mL of 0.05 M $NaH_2PO_4 \cdot 7H_2O$ buffer (pH 7.0)

were vortexed in glass tubes and loaded into Bond Elut CH solid-phase extraction cartridges (Varian, Harbor City, CA, USA), which had been preconditioned using dichloromethane, methanol, and water. The tubes were washed with water and 20% methanol followed by elution with 0.25-mL methanol using centrifugation (1,800 *g* for 5 min at 4°C). Eluents were evaporated under a nitrogen stream, reconstituted with 0.2-mL mobile phase, filtered (Ultrafree-MC; 0.22 μm; Millipore, Bedford, MA, USA) using centrifugation (4,500 *g* for 5 min at 4°C) and subsequently transferred to vials for injection via an autosampler (Model 712; Waters Associates, Milford, MA, USA).

Aliquots of 100 μL were chromatographed on a Waters HPLC system consisting of a model 510 solvent pump, a temperature control/column heater module (40°C), and a model 490 programmable wavelength detector. Separation was performed with a Superiorex ODS, 5 μm, 4.6 x 250-mm column (Shiseido Co., Tokyo, Japan) using a mobile phase of acetonitrile, methanol, water, and trifluoroacetic acid (22/15/63/0.1, v/v/v/v) flowing at 1.0 mL/min. Peaks were detected at a wavelength of 334 nm and integrated with Waters Maxima software. Retention times for edotecarin and the internal standard were 4.9 and 6.1 min, respectively. The lower limit of quantification of the assay for edotecarin in plasma was 5 ng/mL. Assay linearity was evident over the standard concentration range of 5–1,000 ng/mL. The inter-day and intra-day assay coefficients of variation were both less than 5%.

The extraction and analysis methods for urine samples were identical to those described previously for the plasma with the following exceptions. The internal standard stock concentration was 1,500 ng/mL. The solid-phase extraction cartridges were Bond Elut CN SPE. The mobile phase consisted of acetonitrile/water and trifluoroacetic acid at a 25/75/0.1 v/v/v ratio. The sample washing procedure involved a water wash followed by two washes with 10% methanol. Urine sample extracts were analyzed on a Waters 2690 HPLC system equipped with a model 996 photodiode array detector using Millennium software. The data from the 430-nm wavelength was used for standard and sample concentration calculations. Retention times for edotecarin and the internal standard were 5.5 and 7.3 min, respectively. The lower limit of quantification of this assay in urine was 50 ng/mL. Assay linearity was evident over the standard concentration range of 50–5,000 ng/mL. The inter-day and intra-day assay coefficients of variation of the assay were both less than 7%.

Edotecarin concentrations were found to be stable in spiked plasma standards frozen at –70°C for at least 66 weeks and in spiked urine for at least 18 weeks. Adequate freeze/thaw and autosampler stability were demonstrated for both plasma and urine samples.

PK analysis

PK parameters of edotecarin were calculated by non-compartmental modeling [16] using WinNonlin Pro version 3.1 pharmacokinetic software (Pharsight Corporation, Mountain View, CA, USA). The maximum plasma concentration (C_{max}) and the time of the maximum plasma concentration (t_{max}) were reported as the observed values. The area under the concentration–time curve from time zero to the last concentration–time point ($AUC_{(0\text{-last})}$) was estimated using the linear-log trapezoidal rule.

Statistical methods

Summary statistics for determination of the MTD, recommended Phase II dose, and PK were based on data from cycle 1. Continuous variables were summarized by mean, standard deviation, median, and minimum and maximum values. Categorical variables were summarized by the number and percent of patients in each category. For selected continuous variables, the mean and range of responses at different doses were compared graphically.

Results

Patient characteristics

Twenty-nine patients were enrolled in the study between February 1999 and November 2000 at two sites in the United States Patient characteristics are summarized in Table 1. The mean age of the patients treated in the study was 56 years (range 36–80 years). There were 18 (62%) males and 11 (38%) females. Twenty-seven of the 29 patients (93%) were Caucasian. All patients had an ECOG performance status score of ≤2, with 21 patients (72%) having a score of 1.

The most common type of primary tumor was colorectal (10 patients, 34%). Most patients had received extensive prior treatment for their disease. Twenty-seven (93%) patients had received prior chemotherapy, with the most common therapies being 5-fluorouracil (17 patients in total) administered either alone (five patients) or in combination with leucovorin (12 patients).

The number of patients treated in each cycle is summarized in Table 2. Patients received a total of 119 cycles of edotecarin. Seventeen cycles were delayed to

Table 1 Patient characteristics

Number of patients treated	29
Males:females	18:11
Age, years (mean ± SD)	56 ± 10.5
ECOG performance status	
0	7
1	21
2	1
Primary tumor type	
Colorectal	10
Lung	3
Bladder	2
Renal cell	2
Malignant mesothelioma	2
Cecal adenocarcinoma	2
Other[a]	8
Race	
Caucasian	27
African-American	1
Asian	1
Prior anti-cancer therapies	
Chemotherapy and surgery	15
Chemotherapy, radiation, and surgery	12
Radiation and surgery	1
Surgery only	1
Prior chemotherapy[b]	
5-Fluorouracil ± leucovorin	17
Gemcitabine	8
Paclitaxel	7
Cisplatin	6
Irinotecan	9

[a] Includes GIST, appendiceal pseudomyxoma, angiosarcoma, carcinoma of the thymus, adenocarcinoma of the pancreas, breast cancer, and unspecified adenocarcinoma (two cases)

[b] Number of chemotherapy medications taken singly or in combination

day 28, and six cycles were delayed between 29–35 days. Delays in the majority of cases were to permit patients to recover from toxicity, such as neutropenia.

All 29 patients received at least one dose of edotecarin. One patient, who received initial treatment at the 6 mg/m^2 level, discontinued due to progressive disease after cycle 4 and then re-entered the study at the 11 mg/m^2 level, with the agreement of the sponsor, under a different patient number and was permitted to continue for cycles 5–8. In the safety analysis, this patient was treated as two separate patients. Thus, the total number of patients in the safety analysis is reported as 30. Twenty-eight patients were evaluated for tumor response. One patient at the 15 mg/m^2 dose level was not included in the efficacy analysis as the patient did not have at least one post-baseline tumor assessment. Twenty-nine patients were evaluated for PK and pharmacodynamics. The most common reason for patient discontinuation was progressive disease (21/30 patients, 70%).

Safety

The most common toxicities in this study are presented in Table 3. Fatigue and nausea, which occurred in 26 (87%) patients each, were the most frequent adverse events and tended to be mainly of grade 1 or grade 2. Neutropenia (22 patients, 73%) and leukopenia (20 patients, 67%) were the most frequent hematologic toxicities with neutropenia tending to be more severe (mainly grade 3 or 4) than leukopenia (mainly grade 2 or 3). Nine patients experienced thrombocytopenia, predominantly of grade 1 severity (7 patients, 23%). Myelosuppression was more common at the higher doses (i.e., 13 and 15 mg/m^2). Nadir counts generally occurred within seven days after the infusion with recovery by days 21–28.

Pre-medication with dexamethasone and granisetron was permitted at the discretion of the investigator following an amendment implemented during the conduct of the study. Thus, dexamethasone and granisetron were administered at cycle 1 and subsequent

Table 2 Patient disposition by dose level and cycle

Dose level (mg/m^2)	Characteristic	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle ≥5
6 (*n*=3)	Patients entered	3	3	1	1	0
	Discontinued therapy	0	2	0	1	0
8 (*n*=3)	Patients entered	3	3	1	1	1
	Discontinued therapy	0	2	0	0	1
11 (*n*=6[a])	Patients entered	5	5	2	1	1
	Discontinued therapy	0	3	1	1	1
13 (*n*=9)	Patients entered	9	7	4	4	3
	Discontinued therapy	2	3	0	1	3
15 (*n*=9)	Patients entered	9	6	3	3	2
	Discontinued therapy	3	3	0	1	2
Total patients entered		29	24	11	10	7

[a] Includes one patient who discontinued from the 6-mg/m^2 group after cycle 4 and then re-entered the study at the 11-mg/m^2 level in cycle 5

Table 3 Toxicities, # patients/total

	Dose (mg/m^2)				
	6	8	11	13	15
Hematologic toxicities					
Neutropenia					
Gr 1/2	0/3	2/3 (67%)	2/6 (33%)	1/9 (11%)	0/9
Gr 3/4	0/3	0/3	2/6 (33%)	6/9 (67%)	9/9 (100%)
Leukopenia					
Gr 1/2	0/3	1/3 (33%)	2/6 (33%)	4/9 (44%)	2/9 (22%)
Gr 3/4	0/3	0/3	1/6 (33%)	3/9 (33%)	7/9 (78%)
Anemia					
Gr 1/2	0/3	0/3	3/6 (50%)	3/9 (33%)	1/9 (11%)
Gr 3/4	0/3	0/3	0/6	3/9 (33%)	2/9 (22%)
Thrombocytopenia					
Gr 1/2	0/3	0/3	2/6 (33%)	1/9 (11%)	5/9 (56%)
Gr 3/4	0/3	0/3	0/6	1/9 (11%)	0/9
Non-hematologic toxicities					
Fatigue					
Gr 1/2	2/3 (67%)	3/3 (100%)	4/6 (67%)	7/9 (78%)	6/9 (67%)
Gr 3/4	0/3	0/3	1/6 (33%)	0/9	0/9
Nausea					
Gr 1/2	3/3 (100%)	2/3 (67%)	6/6 (100%)	6/9 (67%)	7/9 (78%)
Gr 3/4	0/3	0/3	0/6	1/9 (11%)	1/9 (11%)
Anorexia					
Gr 1/2	1/3 (33%)	3/3 (100%)	5/6 (83%)	4/9 (44%)	6/9 (67%)
Gr 3/4	0/3	0/3	0/6	0/9	1/9 (11%)
Vomiting					
Gr 1/2	3/3 (100%)	1/3 (33%)	3/6 (50%)	3/9 (33%)	5/9 (56%)
Gr 3/4	0/3	0/3	0/6	1/9 (11%)	1/9 (11%)
Fever					
Gr 1/2	3/3 (100%)	1/3 (33%)	1/6 (17%)	4/9 (44%)	5/9 (56%)
Gr 3/4	0/3	0/3	0/6	0/9	0/9

cycles to 3/6 patients in the 11-mg/m^2 group, all nine patients in the 13-mg/m^2 group, and 6/9 patients in the 15-mg/m^2 group.

Five patients experienced DLTs during this study: one patient at the 13-mg/m^2 and four others at the 15-mg/m^2 dose levels. Grade 4 neutropenia occurred at 13 mg/m^2. At the 15-mg/m^2 dose level, DLTs were grade 3 nausea, vomiting, headache, and fatigue in two patients who had not received antiemetic prophylaxis. One patient at 15 mg/m^2 experienced grade 4 neutropenia and a grade 3 urinary tract infection and one patient had grade 4 febrile neutropenia.

The maximum dose administered in this study was 15 mg/m^2. At this dose, 9/9 patients experienced a study drug-related grade 3 or 4 toxicity, including grade 3 nausea, vomiting, headache, fatigue, asthenia, leukopenia, blood lactate dehydrogenase increase (two patients), mesenteric vein thrombosis, and hyperglycemia. Four of these nine patients had DLTs. The 15 mg/m^2 dose therefore exceeded the MTD. The recommended Phase II edotecarin dose is 13 mg/m^2. At this dose, a total of 48 treatment cycles were administered (range 1–14 cycles). Two patients at 13 mg/m^2 discontinued study treatment at cycle 1 (one due to DLT and the other due to progressive disease).

Tumor response

Of the 28 patients assessed for tumor response, one patient had a confirmed partial response. The confirmed partial response occurred between cycles 2 and 8 in a patient with widely metastatic transitional cell cancer of the bladder. The patient had previously failed paclitaxel/gemcitabine and second-line MVAC (methotrexate, vinblastine, doxorubicin, and cisplatin) therapies. This patient received ten cycles of edotecarin (13 mg/m^2) until progressive disease. Twelve other patients had stable disease, including three patients with an unconfirmed partial response. The median duration of stable disease was four cycles (range 1–18 cycles). Fifteen patients (51.7%) had progressive disease as best response.

Three other partial responses were initially observed, but these were not confirmed at the 4-week re-evaluation. One unconfirmed partial response was observed in a patient with metastatic colon cancer with

liver metastases. The patient was treated with edotecarin (6 mg/m^2) and had a 56% tumor shrinkage after cycle 2. The patient subsequently discontinued after four cycles of therapy because of progressive disease. Another unconfirmed partial response was observed in a patient with mesothelioma who was treated with edotecarin (8 mg/m^2). This patient had stable disease for 14 cycles and a 53% tumor shrinkage was observed after cycle 16. This patient subsequently discontinued the study after 18 cycles of therapy because of progressive disease. A patient with metastatic breast cancer treated at 15 mg/m^2 had a 51% tumor shrinkage at cycle 4 and then discontinued due to progressive disease at cycle 5.

Pharmacokinetics

Complete PK data were obtained from 29 patients. Edotecarin plasma concentration versus time profiles for the five dose cohorts are shown in Fig. 1. PK parameters are summarized in Table 4. The plasma concentration versus time profiles showed rapid drug accumulation to an apparent C_{max} either before or at the end of the infusion. Edotecarin plasma concentrations peaked during the 2-h infusion and declined steeply to levels <LLOQ of 5 ng/mL by 45 min in most patients post-end of infusion. Peak plasma levels at the MTD were 116±30.9 ng/mL (mean ± SD; $n = 9$). Given the steepness of the drop in plasma levels post-end of infusion, the apparent attainment of C_{max} at times prior to the end of the infusion in some patients may possibly be explained by variability in the infusion rate of the drug solution or termination of the infusion slightly prior to the nominal end of infusion blood draw.

Edotecarin plasma half-life and clearance (CL) values were not estimated in this study due to the short duration of quantifiable plasma concentrations post-infusion in most patients and the knowledge that the apparent terminal half-life of edotecarin is approximately 20 h, based on a separate Phase I PK study using a more sensitive bioanalytical method [17]. C_{max} values in this study (e.g., mean of 92.7 ng/mL at the recommended Phase II dose of 13 mg/m^2) were comparable with those determined in a single dose every 21-day study conducted in Japan (mean C_{max} of 103 ng/mL at the recommended Phase II dose of 13 mg/m^2) [17].

Mean cumulative edotecarin excretion in urine over the 72-h collection period was 3.3–8.3% across the five

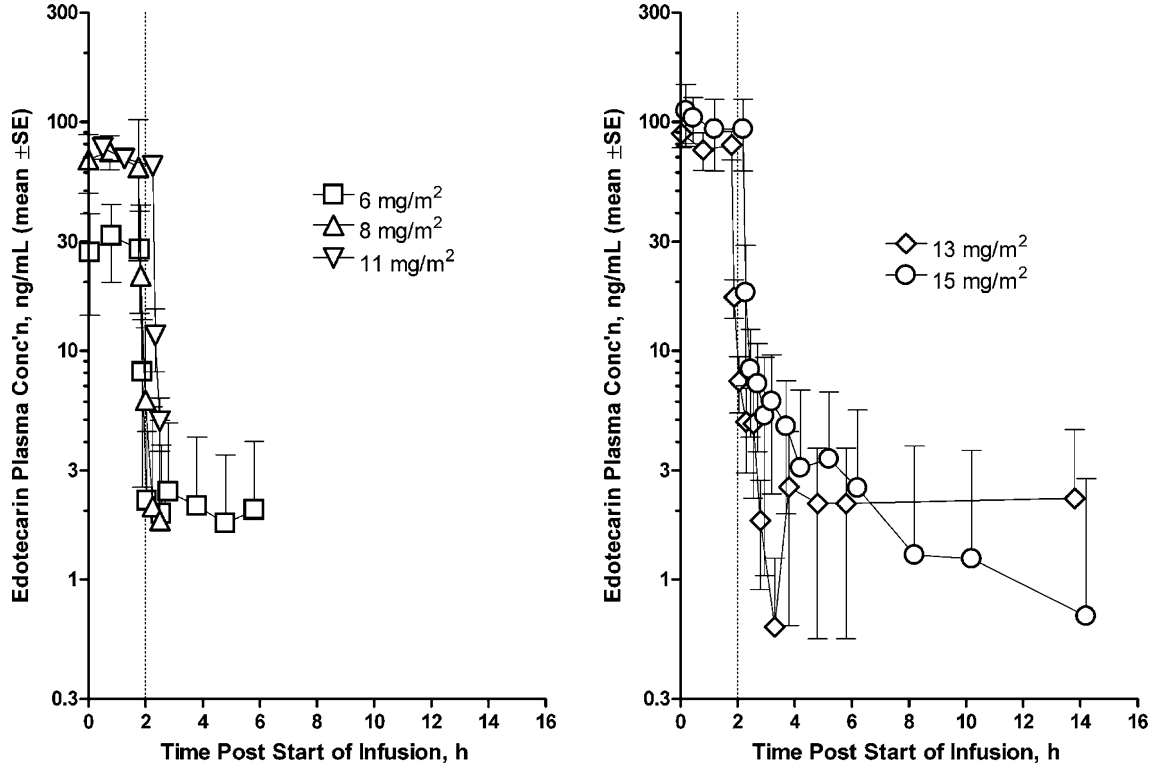

Fig. 1 Edotecarin plasma concentration (mean±SE) versus time profiles: *left panel* 6-, 8-, and 11-mg/m^2 cohorts; *right panel*, 13- and 15-mg/m^2 cohorts. Drug was infused over 2 h (*vertical dotted line* indicates end of infusion). Number of patients was three at 6 mg/m^2, three at 8 mg/m^2, five at 11 mg/m^2, and nine each at 13 and 15 mg/m^2. Data points have been offset in the *x*-direction to diminish overlapping points and error bars. The *median* plasma level was <5 ng/mL (*LLOQ*) by 30 min post-end of infusion in the 6-, 8-, and 11-mg/m^2 groups, by 45 min post-end of infusion in the 13-mg/m^2 cohort, and by 4 h post-end of infusion in the 15-mg/m^2 group

Table 4 Edotecarin pharmacokinetic parameters (mean ± SD)

	Dose (mg/m^2)				
	6	8	11	13	15
n	3	3	5	9	9
C_{max} (ng/mL)	31.7 ± 20.4	86.8 ± 19.6	78.4 ± 19.0	92.7 ± 34.7	116 ± 30.9
$AUC_{0\text{-}t(last)}$ (ng h/mL)	56.0 ± 41.8	140 ± 30.8	137 ± 14.9	157 ± 72.1	200 ± 49.3

AUC area under curve; *SD* standard deviation; C_{max} maximum concentration

dose groups. Most urinary excretion of drug occurred within the first 24 h after the end of the infusion.

Discussion

This study was designed to determine the MTD and recommended Phase II dose of edotecarin when given as a 2-h infusion once every 21 days (with an additional –2 weeks permitted for recovery) and to study the PK and pharmacodynamics of the drug. The recommended dose for single agent use in Phase II studies is 13 mg/m^2. Prolonged decreases in white blood cell, granulocyte, and platelet counts were observed at the higher doses, especially the 15 mg/m^2 dose. Grade 3–4 neutropenia and grade 2–3 leukopenia were common toxicities that occurred mainly in patients in the higher dose groups. The overall incidence of diarrhea was 23% and no grade 3 or 4 diarrhea was reported. Routine anti-diarrheal prophylaxis was not needed in this study. We observed one confirmed partial response and 12 cases of stable disease in a variety of tumor types. Three patients had stable disease for ten or more cycles.

The low extent of edotecarin urinary excretion indicates that non-renal mechanisms predominate in the total body clearance of edotecarin. Edotecarin was not significantly metabolized by the microsomal hepatic CYP450 system in vitro [unpublished data, Banyu Pharmaceutical Co.]. These findings suggest that biliary excretion plays a significant role in edotecarin clearance in man.

Overall, edotecarin has shown an acceptable safety profile at a dose of 13 mg/m^2 once every 21 days, with 1– 2 weeks permitted for recovery from toxicities if needed. Adverse events were similar to those seen in patients treated with irinotecan, with the notable exception that no grade 3 or 4 diarrhea was observed [18]. Given the advanced disease of the patients enrolled in this study, the observed clinical benefit to patients with advanced tumors who had failed multiple previous therapies is encouraging and suggests that Phase II studies are warranted.

Acknowledgment This study was sponsored by Banyu Pharmaceutical Co. Ltd.

References

1. Rougier P, Van Cutsem E, Bajetta E, et al. (1998) Randomised trial of irinotecan versus fluorouracil by continuous infusion after fluorouracil failure in patients with metastatic colorectal cancer. Lancet 352:1407–1412
2. Cunningham D, Pyrhonen S, James RD, et al. (1998) Randomised trial of irinotecan plus supportive care versus supportive care alone after fluorouracil failure for patients with metastatic colorectal cancer. Lancet 352:1413–1418
3. Saltz LB, Cox JV, Blanke C, et al. (2000) Irinotecan Study Group: Irinotecan plus fluorouracil and leucovorin for metastatic colorectal cancer. N Engl J Med 343:905–914
4. Douillard JY, Cunningham D, Roth AD, et al. (2000) Irinotecan combined with fluorouracil compared with fluorouracil alone as first-line treatment for metastatic colorectal cancer: a multicentre randomized trial. Lancet 355:1041–1047
5. Huinink WTB, Gore M, et al. (1997) Topotecan versus paclitaxel for the treatment of recurrent epithelial ovarian cancer. J Clin Oncol 15:2183–2193
6. Scheller JH, Kim K, Hutson P, et al. (1996) Phase II study of topotecan in patients with extensive-stage small-cell carcinoma of the lung: an Eastern Cooperative Oncology Group Trial. J Clin Oncol 14:2345–2352
7. Ardizzoni A, Hansen H, Bombemoswky P, et al. (1997) Topotecan, a new active drug in the second-line treatment of small-cell lung cancer: a Phase II study in patients with refractory and sensitive disease. J Clin Oncol 15:2090–2096
8. Bailly C (2000) Topoisomerase I poisons and suppressors as anticancer drugs. Current Med Chem 7:39–58
9. Long BH, Balasubramanian BN (2000) Non-camptothecin topoisomerase I compounds as potential anticancer agents. Exp Opin Ther Patents 10:635–666
10. Yoshinari T, Ohkubo M, Fukasawa K, et al. (1999) Mode of action of a new indolocarbazole anticancer agent, J-107088, targeting topoisomerase I. Cancer Res 59:4271–4275
11. Arakawa H, Morita M, Kodera T, et al. (1999) In vivo anti-tumor activity of a novel indolocarbazole compound, J-107088, on murine and human tumors transplanted into mice. Jpn J Canc Res 90:1163–1170
12. Arakawa H, Iguchi T, Morita M, et al. (1995) Novel antitumor indolocarbazole compound 6-N-formylamino-12,13-dihydro-1,11-dihydroxy-13-(β-D-glucopyranosyl) 5H-indolo[2,3-a]pyrrolo[3,4-c]carbazole-5,7-(6H)-dione(NB-506): Its potent antitumor activity in mice. Cancer Res 55:1316–1320
13. Komatani H, Kotani H, Hara Y, et al. (2001) Identification of breast cancer resistance protein/mitoxantrone resistance/placenta-specific, ATP-binding cassette transporter as a trans-

porter of NB-506 and J-107088, topoisomerase I inhibitors with an indolocarbazole structure. Cancer Res 61:2827–2832
14. Declaration of Helsinki (2001) Recommendations guiding medical physicians in biomedical research involving human subjects. Republic of South Africa Revision
15. International Conference on Harmonization (ICH) (1996) *Guideline for Good Clinical Practice*. As part of the ICH Harmonized Tripartite Guideline (European Union, Japan and USA)
16. Gibaldi M, Perrier D (1982) Pharmacokinetics. 2nd edn. Marcel Dekker, New York
17. Yamada Y, Tamura T, Yamamoto N, et al. (2006) Phase I and pharmacokinetic study of edotecarin, a novel topoisomerase I inhibitor, administered once every 3 weeks in patients with solid tumors. Cancer Chemother Pharmacol 58:173–182
18. Camptosar US prescribing information: http://www.pfizeroncology.com/hcp/camptosar_resources.aspx