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SHORT COMMUNICATION

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Development of an optimal pharmacokinetic sampling schedule for rubitecan administered orally in a daily times five schedule

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Abstract *Purpose*: Our aim was to develop an optimal sampling strategy for the description of the pharmacokinetics of rubitecan and its active metabolite 9-aminocamptothecin (9-AC) for use in phase II/III studies with oral rubitecan administered in a daily times five schedule. *Methods*: Concentration-time data of rubitecan and 9-AC were obtained from 14 patients who had received 1.5 mg/m^2 per day rubitecan orally. Population pharmacokinetic analysis of both the parent and the metabolite was performed using the nonlinear mixed effect modelling program (NONMEM). Optimal sampling points were selected on the basis of the assessed population pharmacokinetic parameters using a D-optimality algorithm. *Results*: The pharmacokinetics of both rubitecan and 9-AC were adequately described with a one-compartment model. The absorption rate constant, apparent volume of distribution and apparent clearance of rubitecan were 0.81 h^{-1}, 50 l and 1.7 l/h, respectively. For 9-AC the corresponding values of the apparent volume of distribution and the elimination rate constant were 51 l and 0.102 h^{-1}. Interindividual variability of the pharmacokinetic parameters ranged from 38% to 49%. For the first dose, optimal sampling points were 1, 3, 5, 8 and 24 h after dosing. Monte Carlo simulations indicated that the sampling schedule produced parameter estimates which were unbiased and precise. *Conclusions*: An optimal sampling schedule was derived which allowed assessment of the pharmacokinetic parameters of both the parent compound and its metabolite 9-AC after oral administration of rubitecan.

Keywords 9-Nitrocamptothecin · 9-Aminocamptothecin · NONMEM · Pharmacokinetics · Optimal sampling strategy

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Introduction

Rubitecan (9-nitro-20(*S*)-camptothecin) is a topoisomerase I-inhibiting anticancer agent derived from the plant alkaloid camptothecin [10, 21]. In vivo it has been found that rubitecan is partly metabolized to 9-aminocamptothecin (9-AC) [9, 12]. Substantial activity of rubitecan and its metabolite 9-AC has been shown in in vivo and in vitro studies and adequate absorption of orally administered rubitecan has been shown in several species [3, 7, 11]. Oral rubitecan has recently been evaluated in several phase I and II clinical studies and clinical responses have been observed in patients with breast, ovarian and pancreatic cancers [16, 19]. The advised dose of rubitecan in further phase II/III study has been established at 1.0–2.0 mg/m^2 per day (orally) for four to five consecutive days on a weekly basis [19]. The toxicity profile of rubitecan has been shown to vary from mild to moderate in several studies with comparable administration regimens, and the main side effects include leucopenia, neutropenia, diarrhoea, and chemical cystitis [16, 19, 20]. The pharmacokinetic behaviour and pharmacokinetic-pharmacodynamic relationships of rubitecan and its metabolite 9-AC will be studied in future phase II/III studies. Extensive sampling in large numbers of patients for the assessment of the pharmacokinetics is, however, impracticable. In this study an optimal sampling strategy was developed which allowed

estimation of pharmacokinetic parameters with a limited number of samples.

Methods

Patients and pharmacokinetic study

For the development of the optimal sampling strategy, pharmacokinetic data were derived from a phase II study in which the influence of food intake on the pharmacokinetics of rubitecan was investigated. Patients were treated with 1.5 mg/m^2 rubitecan after a full night fasting period and after a high-calorie breakfast in a randomized crossover study design [14]. Blood samples were taken prior to dosing and at 30 min and 1, 2, 3, 4, 6, 8 and 24 h after dosing in order to characterize the pharmacokinetic profile of rubitecan and 9-AC. Samples were immediately centrifuged and plasma was frozen at –70°C. Analysis of rubitecan and rubitecan was performed using high-performance liquid chromatography with fluorescence and UV detection [13]. The lower limit of quantification was 10 ng/ml and 2.5 ng/ml for rubitecan and 9-AC, respectively. Further treatment consisted of 1.5 mg/m^2 per day rubitecan under fasting conditions for five consecutive days on a weekly basis. The clinical and pharmacological evaluation in this study has been described by Schöffski et al. [14]. The study protocol was approved by two independent ethics committees, and written informed consent was obtained from all patients.

Development of a population pharmacokinetic model

A population pharmacokinetic model was developed for both rubitecan and 9-AC using the NONMEM program (double precision, version V, level 1.1) [2]. The pharmacokinetics of both compounds were adequately described on the basis of a one-compartment model. Estimated pharmacokinetic parameters included the absorption rate constant (k_a), apparent volume of distribution (V/F), and apparent clearance (CL/F) for rubitecan and apparent volume of distribution (Vm/Fm), elimination rate constant ($k_{e,m}$) for 9-AC. F and Fm represent the bioavailability of rubitecan and the fraction of rubitecan metabolized to 9-AC, respectively. Interpatient variability of the pharmacokinetic parameters was estimated using a proportional error model. For example, the variability in CL was estimated using $CLi = CLpop(1 + \eta i)$, in which i represents the number of the individual, CLi is the CL of the ith individual, CLpop is the population value and η is the interindividual random effect with mean 0 and variance ω^2. The difference between the jth measured concentration in the ith patient ($Cobs_{ij}$) and its respective prediction ($Cpred_{ij}$) was modelled with an additive error model: $Cobs_{ij} = Cpred_{ij} + \epsilon$, where ϵ is an independent random variable with mean 0 and standard deviation σ.

Selection of optimal time points

An optimal sampling schedule was developed with data from a study in which patients received 1.5 mg/m^2 per day. The selection of optimal sampling points was based on the estimated population pharmacokinetic parameters and the D-optimality algorithm [5], as implemented in the software package ADAPT II [6]. A sample window of 24 h was used. The validity of the sampling schedule was assessed by Monte Carlo simulation of a trial with 50 patients [4]. Individual (''true'') pharmacokinetic parameters were generated on the basis of the developed population pharmacokinetic model. Concentrations were simulated at the optimal sampling points. A (new) population model was then estimated based upon the simulated concentrations of the 50 patients. Individual Bayesian estimates were obtained for each patient and compared with the original (''true'') pharmacokinetic parameters. Bias and precision of the estimated pharmacokinetic parameters were calculated to evaluate the performance of the sampling schedule [15]. Bias and precision were averaged for 1000 simulated datasets in order to obtain estimates of the true values (Monte Carlo simulation).

Results

Population pharmacokinetic model

Plasma samples were available from 17 patients and comprised a total of 296 samples. All pharmacokinetic parameters were estimated with an acceptable coefficient of variation ranging from 12% to 34%. For rubitecan the following values for the pharmacokinetic parameters were found: $k_a = 0.81$ h^{-1} (13%), V/F = 50 l (12%), and CL/F = 1.7 l/h (27%). The calculated elimination half-life was 20 h. Interindividual variabilities of V/F and CL/F were 39% and 38%, respectively. When compared with the fasted state, the bioavailability of rubitecan was reduced by 48% when ingested with food. For 9-AC Vm/Fm was 51 l (34%) and $k_{e,m}$ was 0.102 h^{-1} (33%). Using these values, an apparent clearance (CL/Fm) of 5.2 l/h could be calculated for 9-AC. Interindividual variability of Vm/Fm was 48%. Residual variabilities for rubitecan and 9-AC were 11 and 1.7 ng/ml, respectively. Plots which indicate the goodness of fit are represented in Fig. 1.

Selection of optimal time points

Application of the D-optimality criterion provided the following optimal sampling schedule: 1, 3, 5, 8 and 24 h after the ingestion of rubitecan. The predictive performance of the optimal sampling schedule was assessed by Monte Carlo simulations ($n = 1000$). Bias and precision of the five pharmacokinetic parameters are given in Table 1. The estimates of all pharmacokinetic parameters on the basis of five samples were unbiased and had adequate precision (8–33%).

Discussion

The purpose of this study was to develop an optimal sampling strategy for the assessment of the pharmacokinetic parameters of rubitecan and the metabolite 9-AC to be used in the further clinical development of rubitecan. Since the metabolite 9-AC has shown antitumour activity in preclinical and clinical studies, it was important to include its pharmacokinetic evaluation. Several techniques have been described for the development of optimal sampling procedures [1, 5, 18]. We used a Bayesian approach which offers the advantage of the estimation of a full pharmacokinetic profile using a limited number of plasma concentration time data. Furthermore, there is no need for the sampling times to be exact. In order to use the Bayesian approach population pharmacokinetic parameters should be estab-

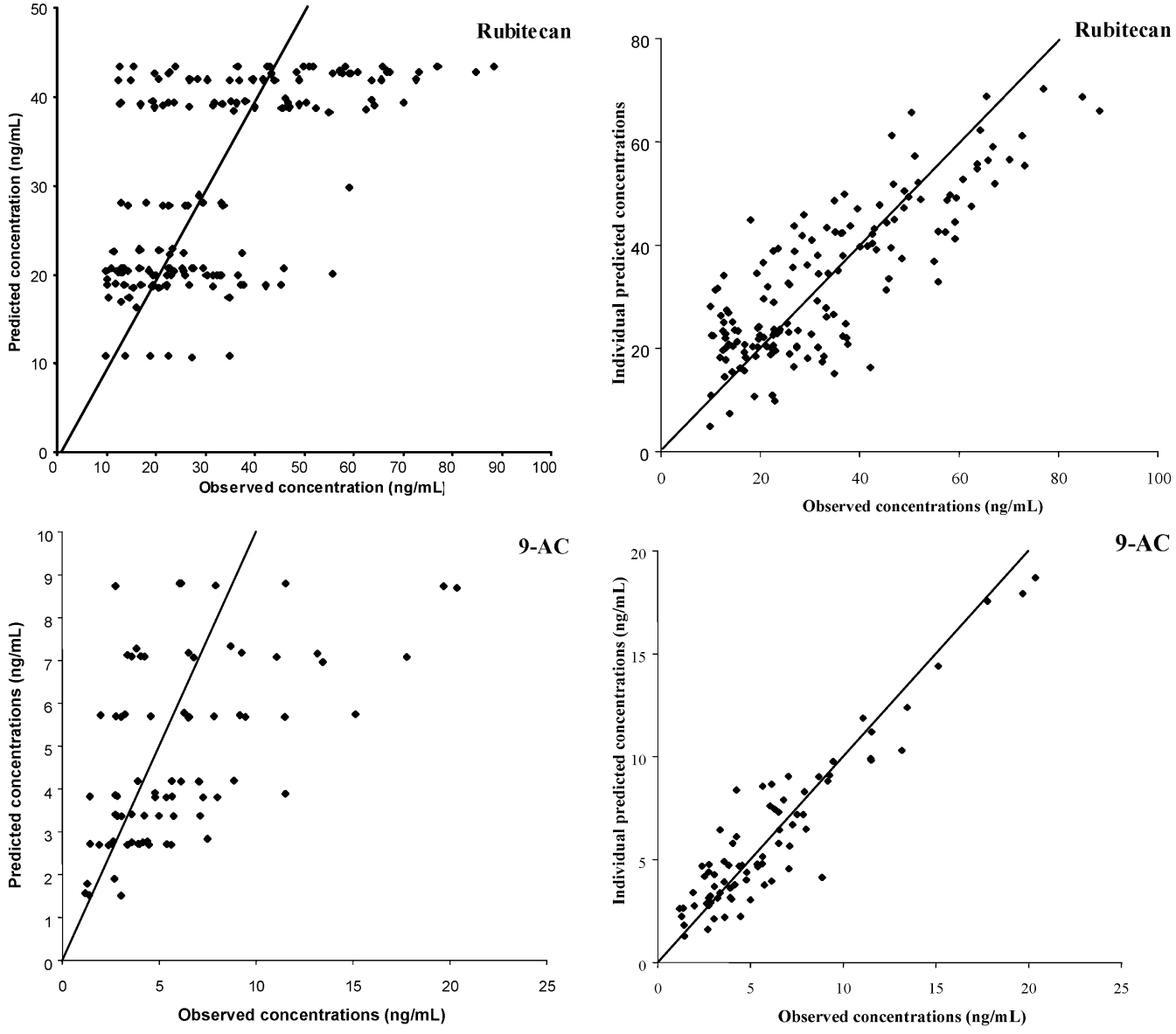

Fig. 1 The relationships between model predictions (*left*) and individual Bayesian estimates (*right*) and observed rubitecan and 9-AC concentrations with their distribution around the line of identity

Table 1 Bias and precision of pharmacokinetic parameters of rubitecan and its metabolite 9-AC assessed by Monte Carlo simulations ($n = 1000$). Values are means ± SD

Compound	Estimate	Parameter	Bias (%)	Precision (%)
Rubitecan	0.81 h^{-1}	k_a	6.1 ± 8.2	8.3 ± 6.1
	50 l	V/F	–1.7 ± 2.9	13.5 ± 1.7
	1.7 l/h	CL/F	8.5 ± 8.3	26.0 ± 5.8
9-AC	51 l	V/Fm	5.0 ± 10.1	8.8 ± 7.1
	0.102 h^{-1}	$k_{e,m}$	4.5 ± 9.7	32.5 ± 8.0

lished a priori [17]. The concentration time data of fed and fasted patients treated with 1.5 mg/m^2 per day rubitecan for five consecutive days were used for the establishment of the population pharmacokinetic parameters. For 9-AC a $k_{e,m}$ of 0.102 h^{-1} was found which corresponds to a theoretical elimination half-life of 6.8 h. The elimination half-life of rubitecan is 20 h indicating that the observed elimination rate of 9-AC is limited by its rate of formation. This apparent elimination half-life of 9-AC corresponds well with the elimination half-life after administration of intravenous 9-AC (7.0 h) [8]. Because we had data from only a limited number of patients, we used Monte Carlo simulations to validate our model and sampling strategy [4]. The model accurately described the individual pharmacokinetic profiles, as residual variability was small with acceptable coefficients of variation (12% to 39%) and interindividual variability was modest (38% to 51%). The performance of the sampling schedule was adequate with no significant bias, and precision ranging between 8% and 33%. In conclusion, we developed an optimal sampling schedule which allowed the assessment of the

pharmacokinetic parameters of rubitecan and its metabolite 9-AC with a limited number of samples.

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