A limited sampling strategy to estimate the pharmacokinetic parameters of irinotecan and its active metabolite, SN-38, in patients with metastatic digestive cancer receiving the FOLFIRI regimen

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Abstract. In this study we propose for the first time a limited sampling strategy to estimate the individual pharmacokinetic parameters of both irinotecan and SN-38 in patients treated with the irinotecan plus 5-fluorouracil (FOLFIRI) regimen. The pharmacokinetics of irinotecan and SN-38 were studied in 74 patients with advanced inoperable digestive cancer. Plasma concentrations were taken during and up to the 42 h following a 90-min infusion of irinotecan (180-225 mg/m^2). Data splitting was used to create model-building and validation data sets, and data were analysed with the NONMEM program. The disposition of SN-38 was dependent on the disposition of irinotecan. The estimated pharmacokinetic parameters of irinotecan [terminal half-life (t), 11.5 h; total clearance (CL), 25.0 l h^{-1}; area under curve (AUC), 14.9 mg x h l^{-1}] and SN-38 (terminal t , 32.2 h; AUC, 0.42 mg x h l^{-1}) were similar to those determined in other studies. The protocol involving two sampling times, at 1 and 24 h following the beginning of the infusion, allowed for a precise and accurate determination of the individual pharmacokinetic parameters of the two drugs. The limited sampling strategy developed in this study ought to facilitate future studies on the pharmacology and toxicity of irinotecan-based therapy.

Introduction

Colorectal cancer is the third most common cancer in both men and women and the third most prevalent cause of cancer-related deaths in the world (1). When colorectal cancer is detected early, it is highly curable. However, in more than 60% of cases the cancer is detected at an advanced stage. For these patients, 5-fluorouracil (5-FU) remains, more than 40 years after its introduction, the first-line chemotherapy. The modulation of 5-FU therapy by folinic acid has shown a significant benefit in terms of tumour response rate versus single-agent 5-FU (2).

The development of two recent anticancer drugs, irinotecan and oxaliplatin, has modified the management of colorectal cancer. Irinotecan-based combination therapy sets a new survival standard for the treatment of this life-threatening disease (3-5).

Irinotecan {7-ethyl-10-[4-(1-piperidino)-1-piperidinol]-carbonyloxy-camptothecin, or CPT-11} is a semi-synthetic derivative of camptothecin. It is extensively metabolised in the liver into various metabolites (6-8) and is cleaved by carboxylesterases to form SN-38 (7-ethyl-10-hydroxycamptothecin), a compound 100- to 1000-fold more cytotoxic. SN-38 is further conjugated in the human liver to an inactive ß-glucuronide derivative (SN-38G). The cytochrome P450 3A4 enzymes are responsible for the formation of two other metabolites, 7-ethyl-10-[4-*N*-(5-aminopentanoic acid)-1-piperidino]-carbonyloxycamptothecin and 7-ethyl-10-(4-amino-1-piperidino)-carbonyloxycamptothecin. Irinotecan and SN-38 bind to the topoisomerase DNA complex, preventing the re-ligation of single-strand breaks in the DNA molecule. These drugs are believed to exert their cytotoxic effect during the S-phase of the cell cycle.

Both irinotecan and SN-38 undergo pH-dependent reversible hydrolysis from active closed-ring lactones to open inactive carboxylate forms. Rivory *et al* (9) found this interconvertion to be of low variability, suggesting that a simple assay of their total forms is as informative as an assay of their lactone forms.

The pharmacokinetics of irinotecan and its metabolites in humans have been widely studied (10-23). The terminal disposition phases of the parent drug and its two oxidation metabolites are quite similar, suggesting that a formation rate-limitation of the metabolite disposition occurrs. The terminal disposition phases of SN-38 and SN-38G are

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delayed compared with the elimination rates of irinotecan and cytochrome P-450-mediated metabolites, suggesting that SN-38 disposition may involve an elimination rate-limiting process (17-19). However, the pharmacokinetic analyses performed in the majority of these studies treated the metabolites as independent of the parent compound. Only Klein *et al* (13) developed a population pharmacokinetic model to describe the disposition of both irinotecan and its metabolites, SN-38 and SN-38G.

Since it is very important to limit the number of blood samples taken from cancer patients, and since irinotecan and SN-38 both contribute to the pharmacological and toxic effects of irinotecan-based therapy, our first objective was to devise a limited sampling strategy that enabled us to estimate individual pharmacokinetic parameters. In this way, by developing and validating a population pharmacokinetic model which simultaneously accounted for irinotecan and SN-38 concentrations, we were able to individualise therapies and achieve a target systemic exposure for both irinotecan and SN-38. The predictive performance of this Bayesian procedure was evaluated by comparing predicted irinotecan and SN-38 concentrations with measured concentrations using an independent group of patients. The study was carried out on a population of patients receiving both 5-FU and irinotecan.

Patients and methods

This study was conducted according to the Declaration of Helsinki as amended in the 41st World Medical Assembly (Hong Kong, 1989) and was reviewed and approved by the Regional Ethics Committee. Patients included in the study had given their informed consent.

Study design. Patients with advanced inoperable histologically-proven digestive cancer were included in the trial and were admitted to the Medical Oncology Service of the Anticancer Centre (Montpellier, France). Prior to admission, full clinical histories were recorded and each patient received a physical examination. The selection criteria for inclusion were: i) age of 18-75 years; ii) WHO performance score of 0-3; iii) adequate bone marrow (absolute neutrophil count ≥2,000/mm^3, platelet count ≥100,000/mm^3 and haemoglobin ≥10 g/dl), hepatic function (bilirubin level <1.5 times normal, prothrombin time ≥50% and alkaline phosphatase level <5 times normal) and kidney function (serum creatinine level <135 μM); iv) life expectancy of ≥3 months.

All patients received methylprednisolone and the FOLFIRI regimen (irinotecan-LV/5FU simplified).

The LV/5FU simplified regimen was administered at a fixed dose [leucovorin (LV), 200 mg/m^2] by intravenous infusion over 2 h followed by a 5-FU loading dose (400 mg/m^2) and then 5-FU (2,400 mg/m^2) in a continuous 46-h infusion). Irinotecan was administered during the infusion of LV at a dose of 180-225 mg/m^2 in 250 ml of 5% dextrose over a 90-min intravenous infusion. Blood samples were collected in heparinised glass tubes before drug administration, then at 0.5, 1, 1.5 (end of infusion), 4, 8, 24 and 42 h from the start of the infusion. Immediately after collection, samples were centrifuged (1500 g) at 4˚C for 10 min, and plasma samples were then immediately frozen at -80˚C until assay.

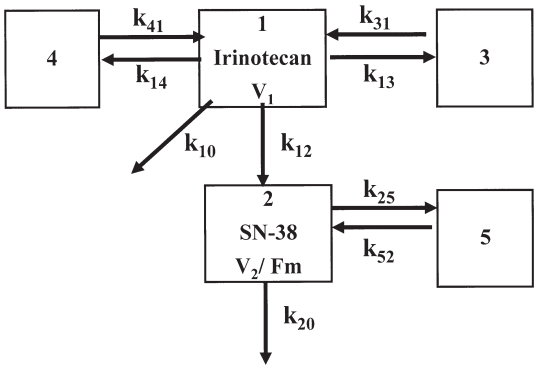

Figure 1. Structural pharmacokinetic model for irinotecan and SN-38. V_1, volume of the central compartment for irinotecan; k_{14} and k_{41}, transfer rate constants from the central (irinotecan) (1) to the fast-equilibrating tissue compartment (4); k_{13} and k_{31}, transfer rate constants from the central (irinotecan) (1) to the slow-tissue equilibrating compartment (3); k_{12}, elimination of irinotecan by the formation of SN-38; k_{10}, elimination of irinotecan by other routes; V_2/F_m, volume of the central compartment for SN-38 (2); k_{25} and k_{52}, transfer rate constants from the central (SN-38) to the tissue equilibrating compartment (5), k_{20}, elimination of SN-38 from the central compartment.

Irinotecan and SN-38, as a total of their lactone and carboxylate forms, were simultaneously assayed in human plasma by high performance liquid chromatography with fluorescence detection (24). For both compounds, the limit of quantitation was 0.5 ng/ml. The inter-assay precision varied 2.6-10.8%. The inter-batch accuracy ranged from 92.8 to 111%.

Population pharmacokinetic analysis. Patients were randomly distributed between a population (model-building) group (n=43) and a test (validation) group (n=31). Potentially explanatory covariates such as patients' age, weight, gender, body surface area, serum creatinine, serum ALAT and serum ASAT were included in the original data files.

Pharmacokinetic model-building and model-validation analyses were performed using the subroutines ADVAN-6 and TOL-5 from the library of programs provided with the NONMEM-PREDPP package (Version 5.0) (25) through the Visual-NM graphical interface (26). Compartmental analysis was performed by processing the parent drug and its metabolite simultaneously. The population characteristics of the pharmacokinetic parameters (fixed and random effects) were estimated using the first-order method. As previously reported (13,19), the disposition of irinotecan was described with a linear three-compartment model and that of SN-38 with a linear two-compartment structural model, with SN-38 as a first-order formation of irinotecan. The structural model is represented in Fig. 1. In this model, we estimated both the elimination of irinotecan by the formation of SN-38 (k_{12}) and its elimination by other routes (k_{10}), with the sum of k_{10} and k_{12} representing the elimination of irinotecan from the central compartment. Because the metabolised fraction (F_m) of the irinotecan dose into SN-38 was unknown for the patient population, the central volume of distribution (V_2) and the total clearance (CL) divided by the F_m had to be estimated.

The eleven-dimensional vectors, θ, of the kinetic parameters considered in the population analysis are presented in Fig. 1.

We used an exponential interindividual variability error model to estimate individual deviations in the pharmacokinetic parameters from the estimated population average values. We tested various error models and selected a proportional error one, with different errors assigned for irinotecan and SN-38.

After the selection of the basic structural and statistical models, we conducted a preliminary assessment of covariate influence by plotting individual Bayesian pharmacokinetic estimates against all the preselected potential covariates. When a relationship emerged, the covariate was included in the subsequent stage of analysis. The change in the NONMEM objective function produced by the inclusion of a covariate term (χ^2 test) was used to compare alternative models. If the objective function did not vary significantly, the relationship between the covariate and the pharmacokinetic parameter was ignored.

Several secondary pharmacokinetic parameters were calculated from the individual (Bayesian estimates) primary pharmacokinetic parameters. In the case of irinotecan, these were the CL, the steady-state volume of distribution (V_{ss}), the half-lives (t) of the λ_1, λ_2 and λ_3 hybrid constants and the plasma concentration versus time area under curve (AUC). For SN-38 they were the CL/F_m, the t of the λ_1 and λ_2 hybrid constants and the plasma concentration-time AUC.

Structural model validation. The individual pharmacokinetic parameters of patients in the validation group (n=31, not included in the calculation of population parameters) were calculated based on the Bayesian approach. From the resulting individualised parameter values, we calculated plasma irinotecan and SN-38 concentrations at each sampling time, as well as the secondary pharmacokinetic parameters (described above).

Validation of a limited sampling strategy. The individual pharmacokinetic parameters of the 31 patients in the validation group were estimated, based on Bayesian estimates, from a limited number of samples. Two- (1/24, 1.5/24, 1/42, 1.5/42 h) or three-sample (1/4/24, 1.5/4/24, 1/4/42, 1.5/4/42 h) schedules were tested. The database consisted of data gathered from the validation group. From the resulting individual parameters we calculated, for each patient, plasma irinotecan and SN-38 individual predicted (IPRED) concentrations at all sampling times, as well as the secondary pharmacokinetic parameters.

For patients from whom blood samples were not available at the scheduled times (4 h for 2 patients, 24 h for 5 patients and 42 h for 1 patient), concentrations were estimated using the posthoc option in the NONMEM program during the structural model-validation step.

Statistical analysis

Model acceptance (population group). At each step of the model building, closeness to and randomness along the line of unity on the observed (DV) versus predicted (PRED) concentration plot, as well as randomness along the residual and weighted residual (WRES) zero line of the PRED concentration or time versus residual or WRES plot, were considered qualitative evidence of goodness of fit. Moreover, IPRED concentrations were plotted versus DV concentrations and the results were compared to the reference line of slope = 1 and intercept = 0. PRED concentrations were computed based on population parameter estimates, and IPRED concentrations based on individual parameter estimates. Descriptive statistics were used to compare mean residual values to zero and to calculate the 95% confidence intervals. Given the wide range of concentrations, the residual value was calculated as follows: (DV-IPRED)/DV. The model was accepted when i) plots showed no systematic pattern and ii) descriptive statistics did not show any systematic deviation from the initial hypothesis (mean assumed to be zero).

Performance of Bayesian individual parameter estimates (validation group). In the case of both irinotecan and SN-38, the performance of the Bayesian estimates was assessed in the validation group (n=31) by comparing the DV concentrations to the IPRED ones estimated by the Bayesian approach. Given the wide range of irinotecan concentrations, the error was defined as relative. Bias and precision were calculated as follows:

i) bias or mean relative predictor error:

$$\text{Bias} = \frac{1}{N}\sum_{i=1}^{i=N} [\text{DV - IPRED}] / \text{DV}$$

ii) precision or root mean relative square error:

$$\text{Precision} = \sqrt{\frac{1}{N}\sum_{i=1}^{i=N} [(\text{DV} - \text{IPRED}) / \text{DV}]^2}$$

In these equations, the 'i' index refers to concentration number and N to the sample size. The 95% confidence interval for bias was computed and the t-test was used to compare the bias to zero.

For SN-38, bias and precision were calculated according to Sheiner and Beal (27,28).

Computing of a limited sampling strategy (validation group). To evaluate the reliability of the parameter estimates for each combination, i) the IPRED concentrations, at all sampling times and calculated using a limited sampling strategy, were compared to the DV concentrations (as described above) and ii) the pharmacokinetic parameters (only CL and AUC for irinotecan and CL/F_m and AUC for SN-38 were considered for this purpose due to their clinical interest), estimated using Bayesian methodology and a limited sampling strategy, were compared to the ones estimated using Bayesian methodology and the entire set of data. These comparisons were performed by computing the bias and precision (27,28).

Results

Patients. Seventy-four patients, between October 2001 and August 2006, were included. For 25 of them, difficult venous access prevented us from obtaining all the necessary blood samples; only 5-8 samples per patient were collected. Patient characteristics are listed in Table I.

Population parameters. Plasma concentration-time profiles in the patient population group are presented in Fig. 2. The population database consisted of 441 concentrations from 43 patients. No relationship was found between the covariates

Table I. Patient characteristics.

	Population group (n=43)		Validation group (n=31)	
	Mean (CV %) (min-max)	No. of patients	Mean (CV %) (min-max)	No. of patients
Sex				
Male		23		18
Female		20		13
Age (years)	61.1 (13.7) (38-75)		61.8 (13.8) (45-81)	
Weight (kg)	63.8 (17.0) (46-84)		70.7 (16.7) (49-97)	
Body surface area (m^2)	1.7 (9.9) (1.38-2.02)		1.8 (10.3) (1.5-2.19)	
WHO Performance status				
0		30		21
1		12		10
3		1		0
Metastasis				
Liver alone		32		21
Peritoneum		3		4
Lymph nodes		2		-
Lung		5		4
Bones		1		2
Line of chemotherapy				
First line		42		30
Second line		1		1
WBC count (10^9/l)	7.9 (22.0) (4.7-11.6)		6.5 (33.5) (2.9-10.8)	
Neutrophil count (10^9/l)	5.5 (32.6) (2.8-12.9)		4.7 (53.4) (1.4-12)	
Haemoglobin (g/dl)	12.5 (12.0) (9.3-17.4)		11.8 (11.5) (8.6-14)	
Platelet count (10^9/l)	355 (34.2) (144-741)		396 (6.3) (140-1200)	
Bilirubin (μM)	9.9 (52.4) (2.7-27.9)		11.6 (35.1) (5.1-20.4)	
AST (U/l)	26.1 (68.0) (4-93)		25.3 (81.6) (2-9.7)	
ALT (U/l)	28.4 (73.1) (8-108)		22.0 (80.3) (4-79)	
Serum creatinine (μM)	66.4 (29.4) (35-126)		63.8 (22.9) (32-95)	
Total proteins (g/l)	77.1 (8.3) (66-91)		73.9 (8.5) (56-86)	
Serum albumin (g/l)	39.6 (10.5) (31-48.1)		38.3 (18.5) (23.1-67)	

CV, coefficient of variation; WBC, white blood cells; AST, aspartate transferase; ALT, alanine transferase.

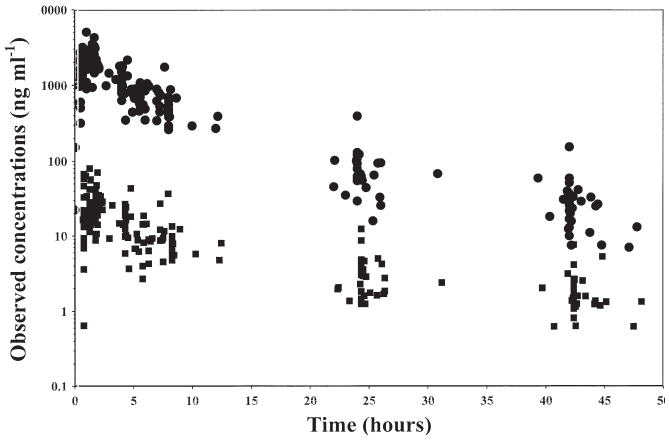

Figure 2. Irinotecan (●) and SN-38 (■) concentrations from 43 patients (population group).

Table II. Population parameters.

	Model-building group (n=43)		Validation group (n=31)		Total population (n=74)	
	Mean	IIV (%)	Mean	IIV (%)	Mean	IIV (%)
Irinotecan						
k_{14}, h^{-1}	8.20	48.4	15.1	25.3	15.7	35.1
k_{41}, h^{-1}	1.24	36.1	1.22	20.0	1.26	20.2
k_{13}, h^{-1}	0.534	58.1	0.571	33.2	0.698	38.7
k_{31}, h^{-1}	0.0758	9.73	0.076	7.30	0.0842	13.5
k_{10}, h^{-1}	0.215	84.2	0.221	58.0	0.195	69.2
k_{12}, h^{-1}	1.91	3.61	1.93	3.47	2.38	15.8
V_1, l	10.7	7.87	10.5	6.86	12.3	9.39
SN-38						
k_{25}, h^{-1}	2.60	4.2	2.60	1.38	2.58	13.1
k_{52}, h^{-1}	0.0371	55.1	0.0631	65.0	0.0426	61.3
k_{20}, h^{-1}	3.07	31.5	2.60	20.0	2.52	28.0
V_2/F_m, l	374	38.1	353	27.0	349	37.4
Objective function	3377		-		6881	
Interindividual variability (%)	ε_1, 31.6		-		ε_1, 31.5	
	ε_2, 36.7				ε_2, 31.1	

V_1, volume of the central compartment; k_{14} and k_{41}, transfer rate constants from the central to the fast-equilibrating tissue compartment; k_{13} and k_{31}, transfer rate constants from the central to the slow-tissue equilibrating compartment; k_{12}, elimination of irinotecan by formation of SN-38; k_{10}, elimination of irinotecan by other routes; V_2/F_m, volume of the central compartment; k_{25} and k_{52}, transfer rate constants from the central to the tissue equilibrating compartment; k_{20}, elimination of SN-38 from the central compartment.

and the kinetic parameters (θ) considered in the population analysis. Population parameters are presented in Table II.

Table III. Mean (coefficient of variation %) secondary pharmacokinetic parameters.

	Model building group (n=43) (%)	Validation group (n=31) (%)	Total population (n=74) (%)
Irinotecan			
CL, l/h	21.7 (11.3)	22.5 (11.9)	25.0 (16.4)
[a]AUC (mg x h/l)	15.4 (29.9)	14.9 (20.5)	14.9 (24.3)
V_{ss}, l	352 (25.3)	388 (19.8)	397 (16.5)
t λ_3, h	11.2 (19.6)	11.9 (14.7)	11.5 (18.3)
SN-38			
CL/F_m, l/h	1083 (30.7)	948 (40.3)	950 (32.0)
[a]AUC (μg x h/l)	340 (58.2)	444 (76.2)	422 (73.4)
t λ_2, h	34.0 (16.9)	27.2 (29.0)	32.2 (44.4)

CL, total clearance; AUC, area under curve; V_{ss}, steady-state volume of distribution; t λ, half-life of the terminal part of the plasma concentration versus time curve; F_m, fraction of the irinotecan dose metabolised into SN-38. [a]Normalised to a 330 mg administered dose.

Mean secondary pharmacokinetic parameters, calculated from the individual primary pharmacokinetic parameters, are given in Table III.

For irinotecan, the mean half-lives corresponding to the hybrid constants λ_1, λ_2 and λ_3 were 0.0455, 2.91 and 11.2 h, respectively. For SN-38, they were 0.175 h for λ_1 and 34.0 h for λ_2. The distribution of irinotecan AUC was close to normal. Concerning the SN-38 AUC, there was no clear evidence of bimodal distribution characteristic of a genetic polymorphism.

The plot of model-predicted versus DV concentrations, obtained from the final model based on population parameter estimates, is shown in Fig. 3A. Given the wide difference in concentrations between irinotecan and SN-38, concentrations are presented in ln-ln coordinates. Various statistical tests carried out showed that i) there was no significant difference when the regression line of IPRED versus DV concentrations (slope = 1.01, S.E. = 0.009; intercept = 14.6 ng/ml, S.E. = 8.43, respectively) was compared to the reference line (slope = 1, intercept = 0), and that ii) the frequency of the distribution histogram of the normalised residuals was as expected (normal with zero mean and unitary variance) (Fig. 3B). The vast majority of the WRES lay within 2 U of perfect agreement and were symmetrically distributed around the zero ordinate (Fig. 3C). The mean relative error (DV vs. IPRED) was low, -0.0407 ng/ml (bias -0.0827/0.0013).

Evaluation of Bayesian pharmacokinetic parameter prediction. Data consisted of 404 concentrations from 31 patients. Mean pharmacokinetic parameters are presented in Tables II and III. For irinotecan, the mean half-lives of the hybrid constants were 0.0410, 3.60 and 11.9 h for λ_1, λ_2 and λ_3, respectively. For SN-38, they were 0.133 h for λ_1 and 27.2 h for λ_2. For both irinotecan and SN-38, the regression lines of IPRED and DV concentrations did not differ significantly

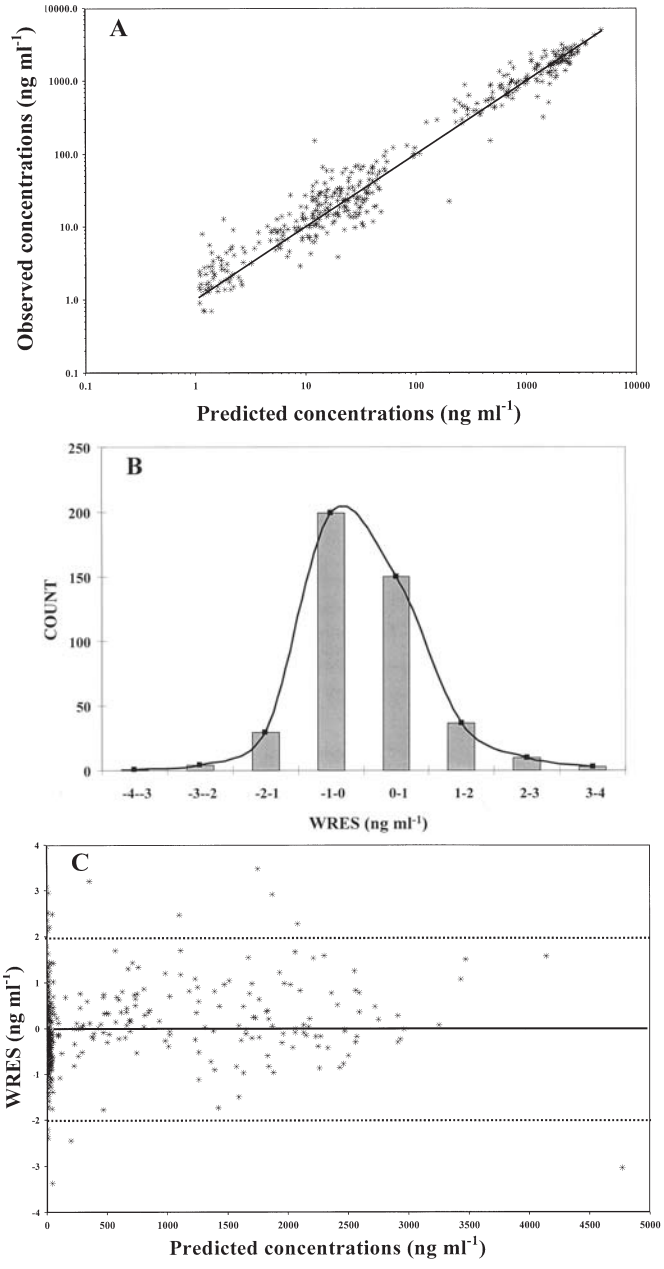

Figure 3. Model performance and diagnostic plots (43 patients, 441 concentrations). (A) Model-predicted vs. observed concentrations based on population parameter estimates (line represents line of identity). (B) Frequency distribution of weighted residuals (WRES). (C) WRES versus predicted concentrations.

from the reference line of slope = 1 and intercept = 0. Bias values were not statistically different from zero and the 95% confidence interval included the zero value (Table IV).

Validation of a limited-sampling strategy. The schedules of 1 and 24 h, 1.5 and 24 h, 1 and 42 h and 1, 4 and 24 h gave the best results in the case of both irinotecan and SN-38, combining accurate prediction with convenience. Concerning the other schedules, a small bias was observed when SN-38 concentrations were compared to IPRED concentrations. The best schedules are presented in Table IV. For practical and ethical purposes, it was important to restrict the number of samples taken to two only and to limit the time spent in

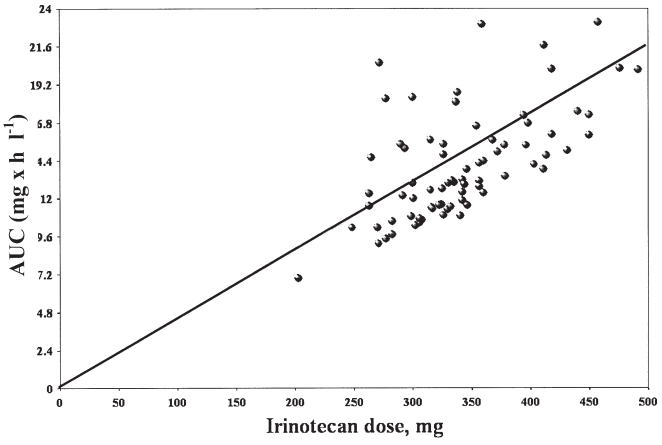

Figure 4. Irinotecan area under the plasma concentration vs. time curves (AUC) as a function of irinotecan dose (r=0.62, P<0.0001).

hospital. Moreover, in clinical practice a sample drawn at the end time of infusion is often subject to problems, which is why we selected the two-sample schedule, at 1 and 24 h after the beginning of the infusion.

Population pharmacokinetic parameters of all patients. In the final step, population pharmacokinetic parameters of irinotecan and SN-38 were determined for all patients (n=74). Mean pharmacokinetic parameters are presented in Tables II and III. For irinotecan, the mean half-lives corresponding to the hybrid constants λ_1, λ_2 and λ_3 were 0.0403, 3.92 and 11.5 h, respectively. For SN-38, they were 0.140 h for λ_1 and 28.2 h for λ_2. The calculated population parameters were similar to those calculated from patients in the population group (n=43).

During the model-building step, we found a correlation between i) irinotecan clearance and age (r=-0.2563, P=0.026) and ii) body surface area and weight and the initial distribution volume of SN-38 (r=0.24, P=0.035; r=0.33, P=0.0042, respectively). In the stepwise analysis, the influence of these covariates was not retained.

For these patients, the dose administered ranged from 203 to 491.3 mg. The irinotecan AUC increased proportionally and linearly with the administered dose (Fig. 4, r=0.62; P<0.0001).

Discussion

The pharmacokinetics of irinotecan have been extensively reported in the literature. In the present study, we propose for the first time a limited sampling strategy to simultaneously estimate the individual pharmacokinetic parameters of irinotecan and SN-38. The data complement previous findings on the pharmacokinetics of irinotecan and have important clinical implications. Indeed, for both ethical and practical reasons it is essential to select a strategy which reduces both the number of samples taken and the time spent in hospital.

Patients in this study received 5-FU/LV (5-FU, 400 mg/m^2; LV, 200 mg/m^2) and irinotecan (180-225 mg/m^2). Compartmental analysis was performed by processing the parent drug and its metabolite simultaneously. The disposition of irinotecan was described with a 3-compartment model and that of SN-38 with a 2-compartment model, with first-order

Table IV. Predictive performance of Bayesian estimation of plasma concentrations, clearance and area under curves with different limited sampling strategies.

	Irinotecan (202 concentrations) IPRED[a], μg/l			Irinotecan CL, l/h			Irinotecan AUC, mg x h/l		
No.	Mean (CV %)	Bias[b] (μg/l)	Precision[b]	Mean (CV %)	Bias (1/h)	Precision	Mean (CV %)	Bias (1/h)	Precision
1	1315 (79.8)	-0.0054 (-0.026, 0.037)	0.264	22.5 (11.8)	-	-	16.6 (20.5)	-	-
2	1302 (80.4)	-0.013 (-0.055, 0.030)	0.334	22.8 (11.3)	-0.28 (-1.05, 0.48)	2.07	16.2 (15.9)	0.415 (-0.15, 0.98)	1.57
3	1388 (83.5)	-0.043 (-0.087, 0.0010)	0.371	22.3 (10.6)	0.202 (-0.52, 0.92)	1.96	16.5 (15.3)	0.0528 (-0.56, 0.66)	1.64
4	1334 (81.4)	-0.0073 (-0.045, 0.030)	0.321	22.9 (10.8)	-0.375 (-1.19, 0.44)	2.22	16.1 (15.4)	0.49 (-0.15, 1.13)	1.77
5	1323 (80.2)	-0.027 (-0.063, 0.010)	0.323	22.2 (6.93)	0.266 (-0.23, 0.76)	1.36	16.6 (16.8)	-0.024 (-0.36, 0.32)	0.91
	SN-38 (202 concentrations) IPRED[a], μg/l			SN-38 CL, l/h			SN-38 AUC, μg x h/l		
No.	Mean (CV %)	Bias	Precision	Mean (CV %)	Bias (1/h)	Precision	Mean (CV %)	Bias (1/h)	Precision
1	16.8 (78.1)	-0.80 (-1.62, 0.023)	6.0	948 (40.3)	-	-	480 (66.3)	-	-
2	16.8 (73.6)	-0.83 (-1.82, 0.16)	7.2	939 (35.6)	8.73 (-35.8, 53.2)	120	480.5 (68.4)	0.0249 (-21.8, 21.9)	48.6
3	18.3 (79.6)	-1.01 (-2.36, 0.35)	7.7	931 (37.8)	16.8 (-24.6, 58.1)	112	492 (68.6)	-11.3 (-30.7, 7.97)	53.0
4	16.5 (74.5)	-0.54 (-1.61, 0.54)	7.8	959 (35.9)	-11.3 (-62.5, 39.9)	138	463 (66.0)	17.6 (-4.72, 39.9)	62.3
5	16.9 (81.8)	-0.94 (-2.07, 0.20)	6.5	940 (40.4)	7.89 (-31.8, 47.6)	107	498.3 (69.9)	-17.8 (-40.3, 4.6)	62.8

[a]Reference, mean observed (DV) concentration values: irinotecan, 1412 μg/l (CV, 82.0%); SN-38, 14.9 μg/l (CV, 83.6%). For bias, values in parentheses are the 95% confidence interval. [b]Given the wide range of irinotecan concentrations, the error has been defined as relative. No. 1, all plasma samples; No. 2, 1 and 24 h; No. 3, 1.5 and 24 h; No. 4, 1 and 42 h; No. 5, 1, 4 and 24 h. IPRED, individual predicted; CL, total clearance; AUC, area under curve; CV, coefficient of variation.

formation of SN-38 from irinotecan. Rivory *et al* (29) and Dodds *et al* (30) suggest that non-linear processes may be involved. However, frequent sampling within the first 30 min of irinotecan administration would be necessary to detect this process.

We obtained similar pharmacokinetic parameters for irinotecan and SN-38 in the model-building group (n=43), the validation group (n=31) and the entire data set (n=74), which provides support for the pharmacokinetic model. The kinetic parameters considered in this population analysis were comparable to those published by Klein *et al* (13). Interindividual variability of the SN-38 pharmacokinetic parameters was higher than in the case of irinotecan. A possible explanation for this could be the variability between patients in the formation of SN-38 by carboxylesterases. Parameter estimates for the two compounds were comparable to those reported in other studies (Table V). As reported by Klein *et al* (13), in the total population group of 74 patients, patient age was found to significantly decrease the systemic clearance of irinotecan. Irinotecan CL decreased by 1.7 l/h between 50-60 and 60-70 years and by 2.4 l/h between 60-70 and 70-80 years. Moreover, according to Poujol *et al* (19), a relationship was found between body surface area and weight and the initial volume of distribution of SN-38. However, the change in objective functions between the naïve (no covariates) and final models was not significant and the influence of these covariates was not retained in the final model. Contrary to the results published by Chabot *et al* (11), hepatic function did not

Table V. Main pharmacokinetic parameters of irinotecan and SN-38 as reported in the literature.

Authors (refs.)	No. of patients	Irinotecan (CV %)	SN-38 (CV %)
Rowinsky *et al* (15)	32		
Dose, mg/m^2		100-345	
Terminal t , h		5.2	5.9
V_{ss}, l/m^2		148	
CL, l/h/m^2		21.1	
De Jonge *et al* (22)	45		
Dose, mg/m^2		175-300	
Terminal t , h		12.1 (26)	22.5 (24.4)
AUC, μM x h		17.2-32.1	0.24-1.15
V_{ss}, l/m^2		151 (26.8)	
CL, l/h/m^2		17.5 (26.8)	
Chabot *et al* (11)	107		
Dose, mg/m^2		100-600	
Terminal t , h		10.5 (4.8)	10.6 (7.5)
V_{ss}, l/m^2		150 (32.7)	
CL, l/h/m^2		14.3 (28)	
Slatter *et al* (16)	8		
Dose (^{14}C), mg/m^2		125	
Terminal t , h		14.6	28.5
AUC, mg x h/l		8.80 (25)	0.40 (60.5)
V_{ss}, l/m^2		297 (40.1)	
CL, l/h/m^2		12.4 (24.4)	
Catimel *et al* (21)	21		
Dose, mg/m^2		33-115	
Terminal t , h		8.3 (57.8)	
AUC, mg x h/l		11.4-28.1	0.17-0.96
V_{ss}, l/m^2		141 (52.5)	
CL, l/h/m^2		14.3 (48.3)	
Klein *et al* (13)	78		
Dose, mg/m^2		100-340	
Terminal t , h		14.0	24.3
V_{ss}, l/m^2		151.7	
CL, l/h/m^2		14.6	
Canal *et al* (10)	47		
Dose, mg/m^2		350	
AUC, mg x h/l		25.2 (29.6)	0.496 (89.5)
CL, l/h/m^2		15.2 (26.8)	
Poujol *et al* (24)	35		
Dose, mg/m^2		180-250	
Terminal t , h		11.7 (20.5)	28.1 (26.6)
AUC, mg x h/l		13.1 (37.4)	0.319 (43.5)
V_{ss}, l/m^2		211 (33.8)	
CL, l/h/m^2		14.3 (37.9)	
Saltz *et al* (20)	26		
Dose, mg/m^2		100	
Terminal t , h		6.0	12.7
AUC, μM x h		7.38 (35)	0.166 (40.4)
V_{ss}, l/m^2		153 (98)	
CL, l/h/m^2		16.6 (74.1)	
Xie *et al* (23)	70		
Dose, mg/m^2		175-300	
Terminal t , h		18.1	24.2
V_{ss}, l/m^2			
Lactone form		257.2	
Carboxylate form		45.1	
CL, l/h/m^2			
Lactone form		39.9 (38)	
Carboxylate form		6.6 (19)	
Sparreboom *et al* (17)	10		
Dose, mg/m^2		200	
Terminal t , h		13.5 (15.3)	23.8 (32.4)
AUC, μM x h		25.6 (22.3)	1.14 (31.3)
V_{ss}, l/m^2		138 (17.4)	
CL, l/h/m^2		14 (22.5)	
This study	74		
Dose, mg/m^2		180-225	
Terminal t , h		11.5 (18.3)	32.2 (44.4)
AUC, mg x h/l		14.9(24.3)[a]	0.422 (64.5)[a]
V_{ss}, l/m^2		230 (18.9)	
CL, l/h/m^2		14.5 (18.9)	

[a]Normalised to a 330 mg administered dose. CV, coefficient of variation.

appear to influence the systemic clearance of irinotecan. These discrepancies could be explained by the fact that none of the patients in this study had elevated hepatic enzyme levels.

To determine the pharmacokinetic parameters of irinotecan and SN-38 in a clinical routine setting with minimal inconvenience to the patient, we proposed a limited sampling strategy based on Bayesian estimation and tested schedules of two or three samples. The schedule with the two sampling times of 1 and 24 h following the beginning of the infusion combined accurate prediction with convenience. In a previous study (19), we showed a good correlation between the plasma and saliva concentrations of both irinotecan and SN-38. Collecting saliva samples would be cost-effective, and would also reduce the invasiveness of the treatment.

In conclusion, the limited sampling strategy developed in this study i) grants a better quality of life to the patient, ii) reduces the risk of sepsis and iii) offers a net time gain for nursing staff.

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