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ORIGINAL ARTICLE

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Modelling of ftorafur and 5-fluorouracil pharmacokinetics following oral UFT administration. A population study in 30 patients with advanced breast cancer

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Abstract *Purpose*: The pharmacokinetics of ftorafur, 5-fluorouracil (5FU) and uracil were investigated in order to built a population pharmacokinetic model for the anticancer drug UFT, administered with leucovorin and vinorelbine. *Methods*: A total of 31 patients with metastatic breast cancer were treated with escalating oral doses of UFT (300 to 500 mg per day) plus leucovorin (90 mg per day) in combination with intravenous vinorelbine (15 to 25 mg/m^2). Concentration-time data were obtained on days 1, 8, 15 and 21 of cycle 1. *Results*: Of the 31 patients treated, 30 were available for the pharmacokinetic analysis. Ftorafur, 5FU and uracil appeared rapidly in plasma and showed large interpatient variations. Ftorafur concentrations were higher than those of 5FU and uracil. AUC significantly increased between day 1, and days 8, 15 and 21. Ftorafur C_{max} and AUC values were proportional to UFT dose, whereas C_{max} and AUC values of 5FU and uracil were not linearly related to UFT dose. The pharmacokinetics of ftorafur were ascribed to a two-compartment open model in which 5FU was produced from the central compartment. The absorption and exponential distribution rate constants were assumed equal. The effect of uracil on 5FU elimination was straightforward, since no reasonable curve-fitting could be obtained for 5FU data when this covariate was not taken into account. The uracil concentration inducing a 50% reduction in 5FU elimination was 2.67 μmol.l^{-1}. This result confirms the important role played by uracil as a competitive inhibitor of 5FU catabolism. *Conclusion*: A pharmacokinetic model for ftorafur and 5FU was developed and should be useful to further study drug interactions and establish dosing guidelines.

Keywords UFT · Oral fluoropyrimidine · Population pharmacokinetics

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Introduction

UFT is a combination of ftorafur (tegafur) and uracil at a molar ratio of 1:4. Ftorafur is a prodrug of 5-fluorouracil (5FU). It is slowly metabolized to 5FU by cytochrome P450 in the liver, thymidine phosphorylase in tumour tissue, and spontaneous degradation [16, 31]. The coadministration of uracil increases the concentration of 5FU in tumours, and the resulting antitumor activity of ftorafur [22]. The mechanism for the improved cytotoxicity in tumour cells is due to competitive inhibition of the activity of dihydropyrimidine dehydrogenase (DPD), the main enzyme involved in 5FU catabolism [11].

Oral administration of UFT results in 5FU drug levels which are comparable to the levels attained with continuous infusion [2, 17]. Biochemical modulation of 5FU with folinic acid (leucovorin, LV) has demonstrated significant benefit in terms of efficacy [1, 25], and combination of 5FU and vinorelbine has proven high clinical activity in metastatic breast cancer patients [12]. A phase I trial of oral UFT/LV in a schedule of 21 consecutive days in combination with vinorelbine was investigated for the treatment of metastatic breast cancer.

The pharmacokinetics of UFT in patients receiving oral UFT alone or in combination with LV have been investigated in numerous studies [2, 14, 17, 21, 28], all of which have shown wide inter- and intrapatient variations of ftorafur, 5FU and uracil concentrations. To

investigate the pharmacokinetics of UFT in breast cancer patients, we performed a pharmacokinetic study in parallel to the current clinical phase I study. The methodology was based on individual analysis of plasma concentrations of ftorafur, 5FU and uracil on days 1, 8, 15 and 21 of the first cycle of treatment. Thereafter, we analysed the data according to a population approach in order to model the effect of uracil on 5FU pharmacokinetics, describe more accurately the inter- and intra-individual pharmacokinetic variabilities and reveal the influence of patient characteristics.

Methods

Patients and treatment

Between February 1998 and May 2000, 31 patients were entered into the multicentre pharmacokinetic prospective study with UFT administered in combination with LV and vinorelbine. Written informed consent from each patient and ethics committee approval were obtained before beginning the treatment. Eligibility criteria included: histologically proven metastatic breast cancer, objectively measurable and/or evaluable disease; age ≥18 years, World Health Organization (WHO) performance status of 0–2, adequate haematological parameters (granulocytes $\geq 2.0\times10^9$/l; platelets $\geq 100\times10^9$/l), hepatic parameters (total bilirubin not more than 1.5 times the upper limit of normal, AST and ALT not more than 2 times the upper limit of normal), and renal function (serum creatinine not more than 1.25 times the upper limit of normal).

UFT was supplied as 100-mg capsules (referring to 100 mg ftorafur plus 224 mg uracil). The first UFT dose level was 300 mg per day, and the daily dose was increased by 100 mg at subsequent levels. LV was supplied as 30-mg tablets, and administered as a total daily dose of 90 mg to all patients. UFT and LV were administered together with 150–300 ml water in three divided doses every 8 h daily for 21 consecutive days, followed by a 7-day rest period. Doses were scheduled at 0700, 1500 and 2300 hours. No food was permitted from 1 h before to 1 h after dosing. If an uneven dose was required, the higher dose was given in the morning. Vinorelbine was given as a 5-min i.v. infusion on days 1, 8 and 15 or on days 1 and 8. In cycle 1, vinorelbine was given 6 h after the first dose of UFT plus LV. In cycle 2, vinorelbine was given before the first dose of UFT plus LV. For subsequent cycles, vinorelbine was given immediately after the first dose of UFT plus LV. The total administered dose of vinorelbine was diluted in 50 ml 0.9% NaCl solution, starting at 15 mg/m^2. Treatment was continued unless there was evidence of disease progression or unacceptable toxicity, or until patient refusal occurred.

The first dose level was UFT 300 mg/day for 21 days and LV 90 mg/day plus vinorelbine 15 mg/m^2 on days 1, 8 and/or 15, followed by a 1-week rest period. The dose levels of UFT/LV/vinorelbine investigated were the following: (1) 300/90/15, (2) 300/90/20, (3) 400/90/20, (4) 400/90/25, and (5) 500/90/20. At levels 1 and 2, vinorelbine was administered on days 1, 8 and 15. Because of significant dose-limiting toxicity (DLT) reported at dose level 2, which was considered the maximum tolerated dose (MTD), the study was amended and the administration of vinorelbine on day 15 was removed at levels 3 to 5. Three patients were treated at each dose level. Escalation to the next dose level was not attempted before at least two patients had completed two cycles. If one out of three patients developed a DLT, three more patients were entered at the same dose level.

Analytical method

The pharmacokinetics of UFT (ftorafur, 5FU and uracil) were examined during cycle 1 on days 1, 8, 15 and 21, after the first administration of UFT. Blood samples were collected through an indwelling catheter inserted in the arm. Blood samples (5 ml) were collected for ftorafur, 5FU and uracil into heparinized tubes at the following times: (1) immediately before UFT administration; (2) on day 1 30 min, 1 h, 1 h 30 min, 2 h, 2 h 30 min, 4 h and 6 h after administration of UFT; and (3) on days 8, 15 and 21 30 min, 1 h, 2 h, 2 h 30 min, 4 h and 6 h after administration of UFT. All samples were immediately centrifuged at 1000 *g* for 10 min at 4°C and stored at −70°C until analysis.

Plasma concentrations were assessed using high-performance liquid chromatography with UV detection. Ftorafur concentrations were determined using a modification of the method of Creaven et al. [6]. Ftorafur and 5-bromouracil, used as an internal standard, were extracted from plasma with ethylacetate. The ethylacetate layer was dried at 50°C under nitrogen. The residue was reconstituted in 2.5 m*M* acetate buffer and injected onto a Spherisorb ODS2 (5 μm, 250×4.6 mm) column. Ftorafur was eluted with an isocratic buffer comprising 2.5 m*M* acetate ammonium plus methanol, pH 5 (78:22 vol/vol) and detected at 265 nm. The limit of quantification of the method was 0.25 μmol.l^{-1} using a 0.5-ml plasma specimen. The interassay precision for standard curve (STD) and quality assurance (QA) samples (overall CV%) was less than 15% in the calibration range 0.5–50 μmol.l^{-1}.

5FU and uracil were assayed separately using the method of Christophidis et al. [5] with modification. Briefly, the compounds with 5-fluorocytosine as an internal standard were extracted from plasma by diethylether/propanol-1 (80:20 vol/vol) with saturated sodium sulphate solution to precipitate proteins. The aqueous phase was removed and the organic fraction was mixed with potassium phosphate buffer, pH 11. The aqueous layer containing 5FU and uracil was neutralized with 1 *N* H_2SO_4. Liquid chromatography was carried out using a μBondapak C18 (10 μm, 300×3.9 mm) and an Inertsil ODS-2 (5 μm, 250×4.6 mm) column for 5FU and uracil, respectively. Compounds were eluted with an isocratic buffer of 50 m*M* potassium phosphate (pH 3) at a flow rate of 1.2 ml/min (5FU) or 1.0 ml/min (uracil) and detected at 262 nm. For 5FU, the limit of quantification of the method was 0.04 μmol.l^{-1}, using a 0.5-ml plasma specimen. For uracil, the limit of quantification was not determined because of basal uracil concentrations ranging from 0.01 to 0.40 μmol.l^{-1}. For 5FU, the interassay precision for STD and QA samples (overall CV%) was less than 15% in the calibration range 0.04–0.76 μmol.l^{-1}. For uracil, the interassay precision for STD and QA samples (overall CV%) was less than 15% in the calibration range 0.11–8.92 μmol.l^{-1}.

Non-compartmental pharmacokinetics

The pharmacokinetics of ftorafur, 5FU and uracil were first described in terms of C_{max} (the maximal observed concentration after dosing) and T_{max} (the time at which C_{max} was observed). The linear trapezoidal method was then used to calculate AUC from t_{dosing} to $t_{\text{last measured concentration after dosing}}$.

Population pharmacokinetic modelling

Concentration-time data were analysed using the non-linear mixed effects modelling approach [29], implemented in the program MP2 (Micropharm Population, INSERM, Paris, France). Ftorafur data were analysed according to one- and two-compartment pharmacokinetic models with first-order absorption and elimination, and different hypotheses regarding relative values of absorption and distribution rates. When the pharmacokinetic model of ftorafur was established, ftorafur and 5FU data were simultaneously analysed according to the pharmacokinetic model depicted in Fig. 1. Parameters of the final structural model were ftorafur CL (systemic clearance), V_1 and V_2, (central and peripheral compartment volumes), Q (intercompartmental clearance), f_m/V_m (fraction of ftorafur metabolized to 5FU-to-volume ratio), and CL_m/V_m, (5FU clearance-to-volume ratio). The distribution volume of 5FU, V_m,

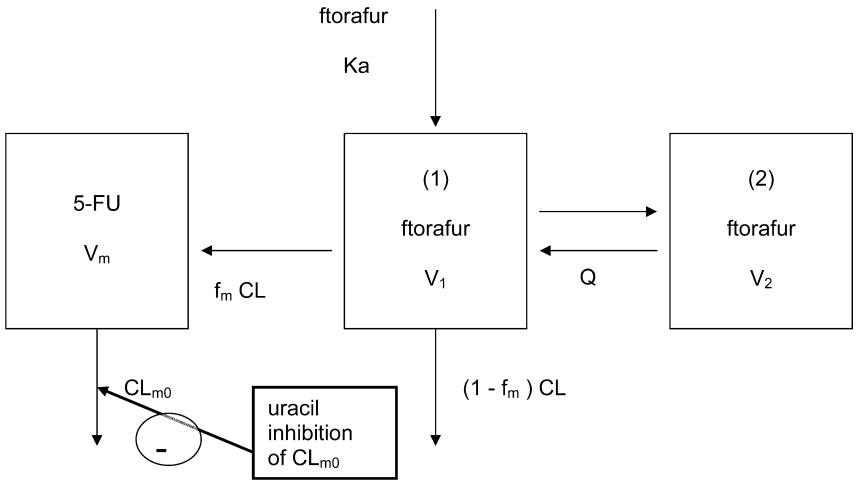

Fig. 1 Scheme of the pharmacokinetic compartment model for the simultaneous prediction of ftorafur and 5FU plasma concentration after UFT oral administration. Ftorafur can exchange between compartments 1 and 2, and can undergo irreversible biotransformation from compartment 1 to produce 5FU compartment *m* (*V* volume term, *CL* clearance term, *Q* intercompartmental clearance; *subscripts 1*, *2* and *m* refer to the ftorafur and 5FU species, respectively)

was not identifiable in this model. The ftorafur parameters were apparent parameters, since they included the bioavailability fraction, F.

Several error models were investigated (i.e. proportional error model with constant coefficient of variation and additive random effects model) to describe interpatient and residual variability. An extensive graphical analysis of predicted versus observed (PRED vs OBS) concentrations was performed to test the value of each model. Comparison between the means of the individual Bayesian (i.e. POSTHOC) parameter estimates and the population estimates was also used to discriminate between the error models.

The influences of individual patient covariates on CL, V_1, V_2, Q, f_m/V_m and CL_{m0}/V_m were systematically tested using a generalized additive modelling. Individual covariates included age, body weight, body surface area (BSA), ASAT, ALAT, alkaline phosphatase and total bilirubin. Full and reduced models (one parameter fewer) were compared by the chi-squared test of the difference between their respective objective function values. A change of at least 7 ($P < 0.01$, one degree of freedom) was required for the addition of a single parameter in the model. The effect of a covariate was considered to have improved the fit if there was a significant decrease in the objective function value of at least 7 compared to the basic pharmacokinetic model (with no covariate). An intermediate multivariate model was then obtained including all significant covariates. In order to keep only those covariates with the largest contribution to predict UFT pharmacokinetics in a final multivariate model, a change of 11 ($P < 0.001$, one degree of freedom) in the objective function was required for the retention of a single parameter during backward stepwise multiple regression analysis.

Results

Clinical results

Enrolled in the study were 31 patients with second-line metastatic breast cancer. All patients were evaluable for safety. When vinorelbine was administered on days 1, 8 and 15, 4 patients out of the 12 treated at dose levels 1 and 2 experienced the same DLT, which was inability to receive vinorelbine on days 8 and/or 15, because of grade 2–3 neutropenia. The protocol was amended, and vinorelbine injection on day 15 was removed. Using this new schedule, the MTD was reached at dose level 5 (UFT/LV/vinorelbine 500/90/25, Table 1). The DLTs were neutropenia and anaemia. The most frequent gastrointestinal symptom was nausea/vomiting, but the incidence of grade 3–4 was low (19% of patients). Grade 3 and 4 diarrhoea was reported in only 6% of patients. Therefore, the recommended dose was UFT 400 mg/day plus LV 90 mg/day over 21 days and vinorelbine 25 mg/m^2 on day 1 and day 8.

Of the 31 patients, 26 were evaluable for efficacy. There were five objective responses (one complete, four partial) and ten stable diseases.

Pharmacokinetics results

Of the 31 patients, 30 (ranging in age from 34 to 68 years) were available for pharmacokinetic evaluation. Their characteristics are listed in Table 2.

Non-compartmental pharmacokinetics

The database included 603, 490 and 613 plasma concentration-time values for ftorafur, 5FU and uracil, respectively (Fig. 2). Plasma concentration-time values

Table 1 Dose-limiting toxicities

Dose level	UFT (mg/day) (days 1–21)	Vinorelbine		Number of patients	Number of cycles (mean)	Number of patients with a DLT
		Dose (mg/m^2)	Schedule (days)			
1	300	15	1, 8, 15	6	2.6	1
2	300	20	1, 8, 15	6	4.1	3
3	400	20	1, 8	3	4.6	0
4	400	25	1, 8	9	4.0	1
5	500	25	1, 8	7	3.2	3

Table 2 Characteristics of the 30 female patients studied

Characteristic	Mean	Median	Range
Age (years)	53	53.5	34–68
Body weight (kg)	61	58	41–90
Body surface area (m^2)	1.63	1.61	1.38–2.03
Serum Creatinine ($\mu mol.l^{-1}$)	78	79	53–119
Aspartate amino transferase (IU)	35.5	33	11–77
Alanine amino transferase (IU)	30	28	7–91
Alkaline phosphatase (IU)	123	112	42–310
Total bilirubin ($\mu mol.l^{-1}$)	5.7	5.5	2–11

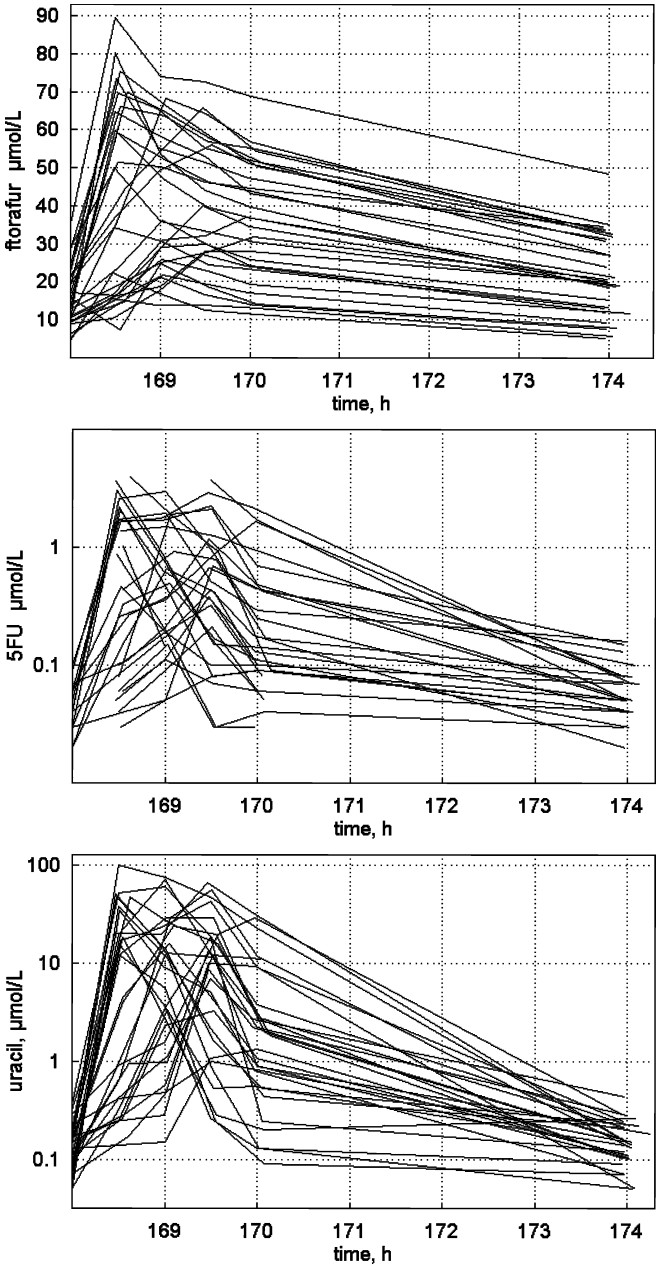

Fig. 2 Plasma concentration-time profiles of ftorafur (*top*), 5FU (*middle*) and uracil (*bottom*) in 30 breast cancer patients on day 8 of cycle 1

were obtained after administration of 100 mg (dose levels 1 and 2) or 200 mg (dose levels 3 to 5) of UFT. These data corresponded to pharmacokinetic evaluations on days 1, 8, 15 and 21 (97 pharmacokinetic courses) during the first cycle of treatment. Per pharmacokinetic course, the median number of blood samples obtained was seven (minimum five, maximum eight). Ftorafur, 5FU and uracil appeared rapidly in plasma. Ftorafur concentrations were higher than 5FU and uracil concentrations (Fig. 2). For ftorafur and 5FU, but not for uracil, the AUC values significantly increased from day 1 to days 8, 15 and 21. There were no difference in AUC values between days 8, 15 and 21 (Fig. 3). Similar results were observed for C_{max}. No differences were observed between T_{max}, either between doses or between days of treatment. Interindividual variability in pharmacokinetics of ftorafur, 5FU and uracil was high. The highest variability was observed for uracil (Fig. 3). Ftorafur dose-normalized C_{max} and AUC values were not different in the different dose groups, whereas dose-normalized C_{max} and AUC values of 5FU and uracil were significantly increased after administration of the 200-mg dose, indicating that these parameters increased more rapidly than the dose increase. Non-compartmental pharmacokinetic parameters calculated under steady-state conditions (days 8, 15 and 21) are summarized in Table 3.

Population pharmacokinetics

Error model

Interpatient and residual variability were best described by an additive error model and a mixed (proportional + additive) error model, respectively. Residual variability included intrapatient error and error related to the assay, sampling time, and model misspecification.

Ftorafur pharmacokinetic model building

A two-compartment model with linear elimination and absorption adequately described the data. However, the estimates generated higher values for the rapid exponential distribution rate, α, than for Ka. Because semilog plots of individual pharmacokinetic courses and residual plots (PRED-OBS) clearly favoured a biexponential decay and not a monoexponential decay, the data were ascribed to a two-compartment model in which the absorption and exponential distribution rate constants were equal [30]. The curve-fitting was satisfactory. PRED-OBS and residual plots were as good as those observed with the complete bicompartment model (Fig. 4). No biological or morphological parameter, including BSA or body weight, influenced ftorafur pharmacokinetics. Pharmacokinetic parameters of ftorafur are summarized in Table 4.

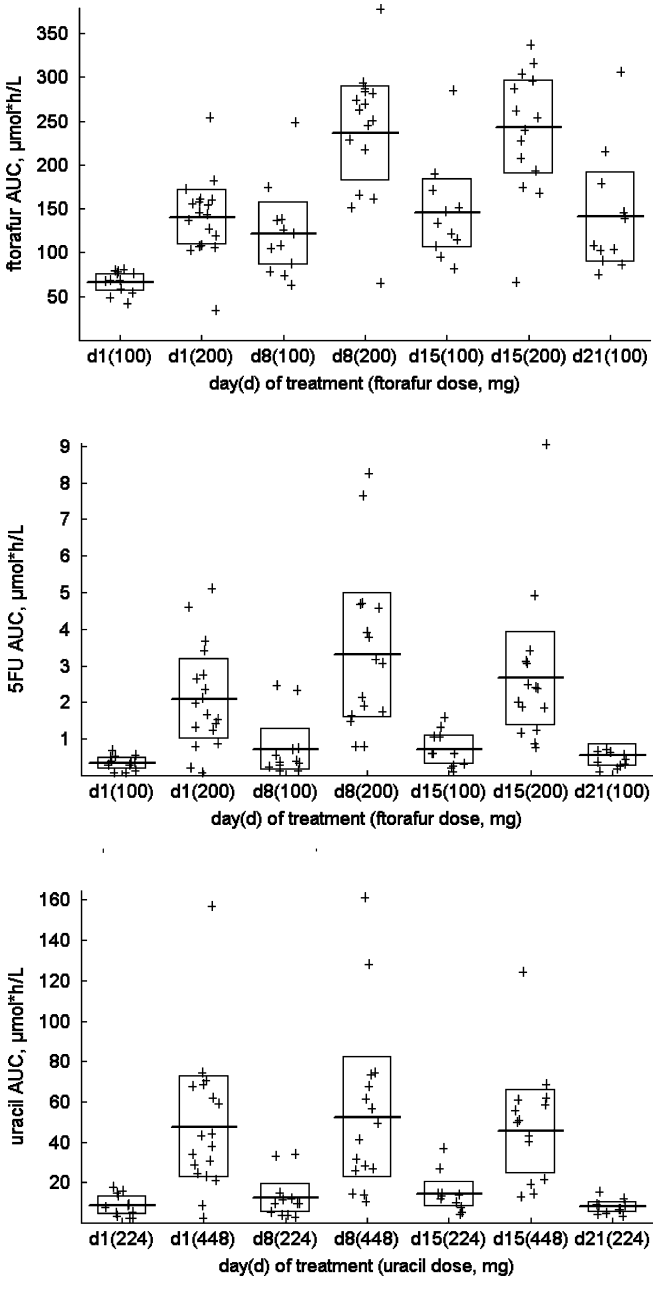

Fig. 3 Evolution of ftorafur, 5FU and uracil AUCs on days 1, 8, 15 and 21 of cycle 1 after two doses (100 and 200 mg) of UFT (*horizontal bars* means, *boxes* standard deviation)

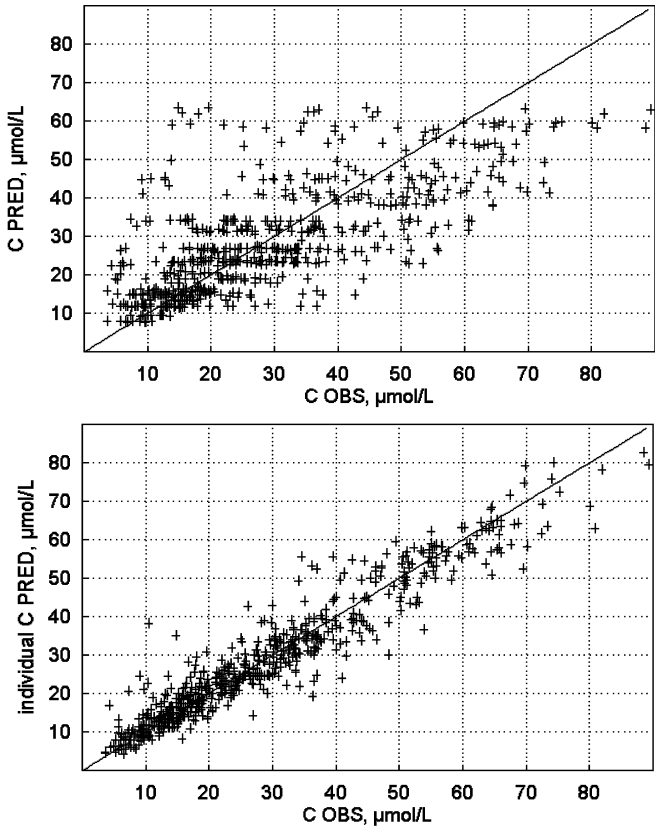

Fig. 4 a Population pharmacokinetic model: predicted (C PRED) versus observed (C OBS) plasma ftorafur concentrations. **b** Bayesian individual estimates: C PRED versus C OBS plasma ftorafur concentrations. The data are from 30 patients with advanced breast cancer

Ftorafur and 5FU pharmacokinetic model building

In a second step, 5FU pharmacokinetics were modelled as a metabolite compartment connected to the central ftorafur compartment (Fig. 1). The equations describing the metabolite concentration when the parent drug confers upon the body a two-compartment model in which the absorption and exponential distribution rate constants are equal are described in the Appendix. When the effect of uracil on 5FU elimination was not taken into account, attempts to fit the data to the compartmental model gave very poor results in terms of

Table 3 Non-compartmental pharmacokinetic parameters in patients receiving 100 mg ($n=34$) or 200 mg ($n=33$) oral UFT doses. Parameters were calculated under steady-state conditions (days 8, 15 and 21 of cycle 1). The data are presented as median (range)

	Dose (mg)	C_{max} (µmol.l^{-1})	T_{max} (h)	AUC (µmol.l^{-}.h)
Ftorafur	100	31 (21–61)	0.83 (0.46–2.06)	122 (63–306)
	200	65 (8–45)		254 (65–372)
5FU	100	0.43* (0.07–3.64)	0.80 (0.47–2.05)	0.49** (0.10–2.46)
	200	1.54* (0.44–5.46)		2.38** (0.78–9.04)
Uracil	100	12 (1–62)	1.02 (0.44–2.52)	10** (3–37.1)
	200	30 (5–49)		49** (11–161)

*$P<0.05$
**$P<0.001$, between dose-normalized values using the Mann-Whitney test

Table 4 Population pharmacokinetic parameters of ftorafur (V_1 and V_2 central and peripheral distribution volumes, Q intercompartmental clearance, $T_{1/2}$ half-life, *NA* not applicable)

Parameter	Mean	Interindividual variability (%)
V_1 (l)	9.70	44.5
Clearance (l/h)	3.12	46.6
Q (l/h)	12.5	18.7
V_2 (l)	20.9	39.4
Residual error (%, proportional component)	16.0	NA
Residual error (μmol.l^{-1}, absolute component)	6.0	NA
Derived parameters		
Absorption, distribution $T_{1/2}$ (h)	0.33	NA
Terminal $T_{1/2}$(h)	7.63	NA

Table 5 Summary of 5FU and ftorafur population parameter estimates according to the pharmacokinetic model depicted in Fig. 1. (V_1 and V_2 central and peripheral distribution volumes, Q intercompartmental clearance. $T_{1/2}$ half-life, *NE* not estimated, *NA* not applicable)

Parameter	Mean estimate	Interindividual variability (%)
V_1 (l)	9.69	29.3
Clearance (l/h)	2.98	27.9
Q (l/h)	12.6	12.6
V_2 (l)	21.1	27.9
f_m/V_m (l^{-1})	0.0459	NE
θ_1 (max CL_{m0}/V_m, h^{-1})	54.6	NE
θ_2 (μmol/l)	2.67	NE
Residual error, proportional (%)		
Ftorafur	10.5	NA
5FU	11.5	NA
Residual error, additive (μmol/l)		
Ftorafur	6.0	NA
5FU	2.5	NA

adequacy between predicted and observed 5FU concentrations. The effect of uracil was then incorporated into the 5FU clearance as follows:

$$Cl_{m0} = \theta_1/(1 + [\text{uracil}]/\theta_2)$$

where θ_1 is the maximal clearance value and θ_2 is the uracil concentration that induces a 50% reduction in 5FU clearance ("IC_{50}"). The estimate was 2.67 μmol.l^{-1}. The curve fitting was satisfactory. Other formulations for uracil inhibition on CL_{m0} were also investigated (e.g. linear function, power function) but did not improve the fit. The 5FU pharmacokinetic parameters are summarized in Table 5. Model performance was estimated by comparing predicted and observed plasma concentrations, as well as individual predicted and observed plasma concentrations of 5FU (Fig. 5). Since ftorafur pharmacokinetic parameters were close to those estimated with ftorafur data alone, the PRED-OBS plots for ftorafur were similar to those depicted in Fig. 4. Figure 6 depicts two representative individual ftorafur and 5FU modelled concentration profiles. Again, no biological or morphological parameter, including BSA or body weight, influenced ftorafur or 5FU pharmacokinetics.

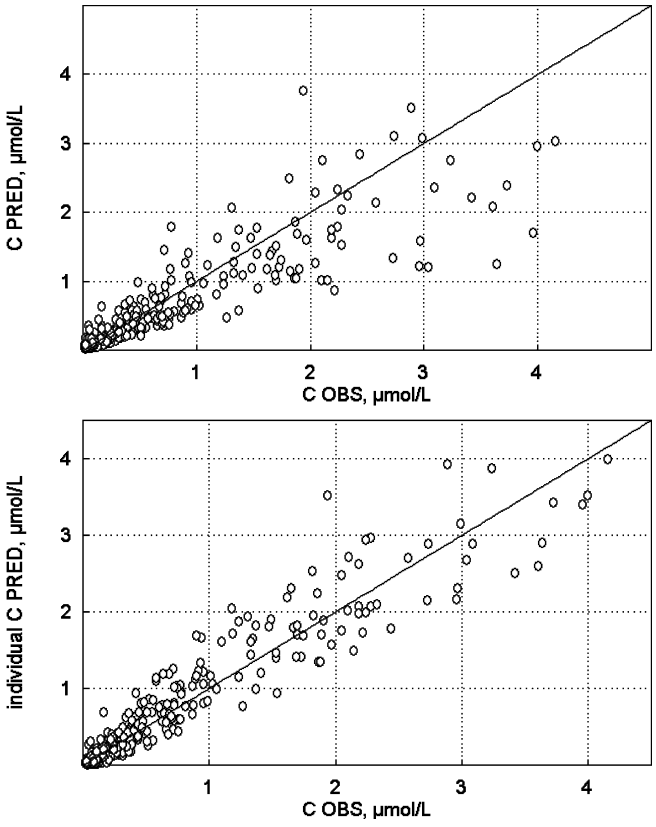

Fig. 5 a Population pharmacokinetic model: predicted (C PRED) versus observed (C OBS) plasma 5FU concentrations. **b** Bayesian individual estimates: C PRED versus C OBS plasma 5FU concentrations. The data are from 30 patients with advanced breast cancer

Discussion

We report here the analysis of pharmacokinetic data obtained in 30 patients with metastatic breast cancer during a phase I trial of oral UFT in combination with LV and vinorelbine. The pharmacokinetic analysis of ftorafur, 5FU and uracil plasma concentrations consisted of two steps: non-compartmental analysis and population model building for both ftorafur and its metabolite 5FU. Comparison of pharmacokinetics, in terms of C_{max} and AUC of ftorafur, 5FU and uracil suggested that after the repeated administration of UFT plus LV plus vinorelbine, a steady-state was attained for ftorafur and 5FU at least on day 8. Our results show that there was no further cumulative increase in the AUC of ftorafur and 5FU after 1 week of treatment with 300 or 500 mg UFT per day. As previously reported, a large interindividual variability in the pharmacokinetics of ftorafur, uracil and 5FU was observed after UFT [14, 18]. This variability was confirmed by visual inspection of concentration-time courses and non-compartmental

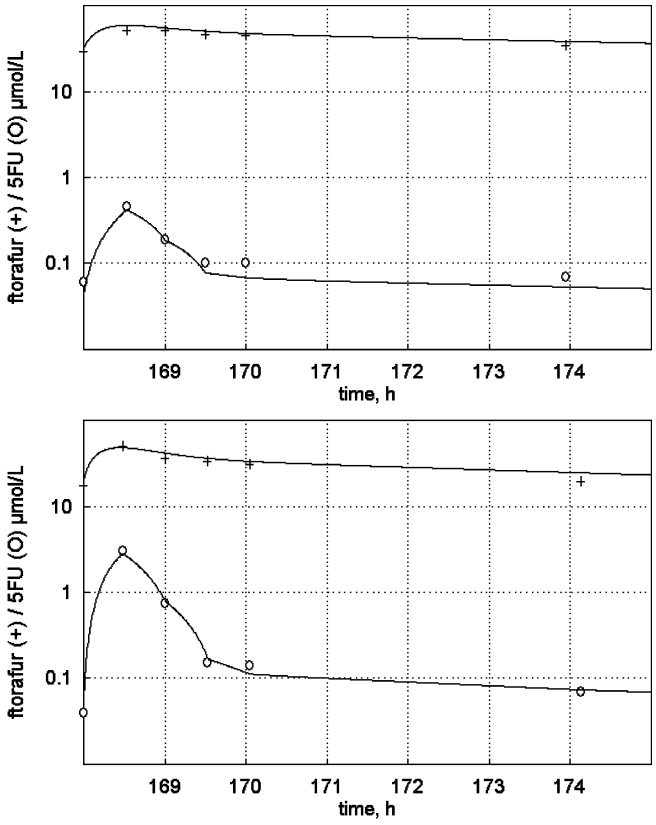

Fig. 6a, b Concentration-time profiles of ftorafur and 5FU for two doses of UFT (**a** 100 mg, **b** 200 mg). The ftorafur (+) and 5FU (○) plasma concentration curves are drawn according the model depicted in Fig. 1 modified for $Ka = \alpha$ (rapid distribution time constant; see Appendix)

pharmacokinetic parameter statistics (Table 2). In terms of non-compartmental pharmacokinetic parameters, the largest variations in C_{max}, T_{max} and AUC were observed for uracil, and the lowest for ftorafur.

One of the causes of such variations could be differences in absorption of ftorafur and uracil between individuals. A high degree of interindividual variability in pharmacokinetics is usually observed when the bioavailability of drugs administered by the oral route is low. As gastrointestinal absorption of ftorafur is generally reported to be 100% after oral UFT dosing and that concurrent administration of LV does not result in significant UFT pharmacokinetic modification [7, 18], this could not account for the variability seen. Another source of variability is ftorafur metabolism. Many enzymes are involved in the bioactivation to and inactivation of ftorafur: hepatic cytochrome P450, cytosolic thymidine phosphorylase (dThdPase) and dihydropyrimidine dehydrogenase (DPD). In normal human liver, 5FU formation from ftorafur is mainly catalysed by cytochrome CYP2A6 [15], an enzyme showing genetic polymorphism [23]. The existence of genetic polymorphism for dThdPase is not known, but it has been reported that there are large interindividual variations in dThdPase levels in various tissues and disease conditions [20]. The highest DPD activities are observed in liver and mononuclear cells, but activity is also found in most human tissues. DPD also demonstrates genetic polymorphism which is responsible for variable activity in different individuals [9, 19]. In addition to enzyme activity, a circadian rhythm in DPD activity has been generally reported in different studies [13]. In order to avoid this effect in the present pharmacokinetic study, UFT treatment was started systematically between 0700 and 0800 hours. These findings indicate that different factors could cause variations in the activation of 5FU from ftorafur and catabolism of both 5FU and uracil.

Based on non-compartmental pharmacokinetic parameters, comparison of C_{max} and AUC between the two dosage regimens suggested that uracil and 5FU pharmacokinetics were non-linear, whereas ftorafur pharmacokinetics were linear. Muggia et al. [21] have reported non-linear pharmacokinetics of ftorafur with significant increases in ftorafur AUC after escalated doses from 300 to 500 mg. In the present study, ftorafur doses were lower (100 and 200 mg) and probably below the dose levels required to saturate metabolic processes. The non-linearity of uracil pharmacokinetics was not surprising since most endogenous compounds undergo complex biotransformations and active transport systems for absorption and diffusion processes. The non-linearity of 5FU is essentially described after bolus administration, and is considered a result of saturable hepatic metabolism [9, 24]. With oral UFT, uracil is administered with ftorafur at a molar ratio of 4:1. Since uracil is a normal substrate for DPD, the apparent non-linearity of 5FU pharmacokinetics is likely to result from a competitive inhibition of DPD.

The population approach used in this study had particular relevance since only a limited number of blood samples were available for some patients. Analysis of the data from these patients using a conventional approach would be difficult because the amount of experimental data was small compared to the number of parameters to be estimated (five parameters for ftorafur in the complete model or four parameters when $Ka = \alpha$). Because the population approach uses all the data simultaneously, all patients can be described using the same model, as missing information in some patients is borrowed from other patients [27].

Ftorafur and 5FU pharmacokinetics were best described by a two-compartment open model in which the absorption and exponential distribution (α) rate constants were equal. The central compartment (1) was connected to a metabolite compartment to produce 5FU. During the model building, no significant differences were observed in the ftorafur pharmacokinetic parameter estimates between the ftorafur model and the ftorafur-5FU model, showing that incorporation of the metabolite pathway did not disturb the model.

The major aim of this population modelling was to characterize the effect of uracil on 5FU elimination (CL_{m0}/V_m). The effect of uracil on CL_{m0}/V_m was straightforward since no reasonable curve-fitting could be obtained for 5FU data when this covariate was not taken

into account. The final model for CL_{m0}/V_m inhibition is a "dose-effect-like" relationship, which predicts the maximal possible CL_{m0}/V_m value when uracil is near zero and provides the uracil concentration that induces a 50% clearance reduction (2.67 μmol.l^{-1}). Practically, given the observed median C_{max} uracil concentrations, 12 and 30 μmol/l, the 5FU elimination rate is decreased by 82% and 92% after oral doses of 100 and 200 mg, respectively. These uracil-induced CL_{m0}/V_m variations are well illustrated in Fig. 6 and explain well the apparent "non-linearity" of 5FU pharmacokinetics deduced from non-compartmental analysis. The pharmacokinetics of intravenous 5FU have been widely studied and total body clearance estimates are high, ranging between 79 and 310 l/h per 1.70 m^2, with short half-lives of a few minutes [3]. Then 5FU rapidly disappears from plasma when infusion is stopped. The sustained 5FU concentration values observed during UFT therapy are generated by ftorafur biotransformation, whose terminal half-life is much longer than 5FU half-life and by competitive inhibition of 5FU metabolism by uracil.

As already demonstrated for other anticancer drugs including carboplatin, doxorubicin, etoposide and ifosfamide [4, 10, 26], no significant relationship was observed between ftorafur or 5FU clearances and BSA (the same kind of plot was obtained when BSA was replaced by body weight). This increases the doubt concerning the use of BSA as a normalization factor for dose administration in chemotherapy [26]. No other covariate significantly influenced the pharmacokinetics, in accordance with our criterion to finally retain a covariate in the model ($P < 0.001$).

In conclusion, this pharmacokinetic study including 30 breast cancer patients confirms previous results showing wide interindividual variability in UFT pharmacokinetics, and allowed us to define a pharmacokinetic model for ftorafur and 5FU. This present model could be useful for further studies to investigate possible interactions when UFT is combined with other cytotoxic drugs and to define a dose adjustment strategy since different studies have demonstrated a significant correlation between 5FU AUC values and toxic events. Ho et al. [14] have shown that nausea, vomiting and diarrhoea correlate significantly with high 5FU C_{max} and AUC values. We have demonstrated [8] that 5FU AUC is highly predictive of hematologic toxicity in patients treated with UFT in combination with LV and vinorelbine. A limited sampling strategy for relevant UFT pharmacokinetic parameters will be developed in order to facilitate large pharmacokinetic studies, and minimize the cost, effort and inconvenience for the patients.

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Appendix

The differential system connected with the model shown in Fig. 1 is:

$$dG/dt = -KaG \quad G = \text{Dose}, t = 0 \tag{1}$$

$$dX_1/dt = -(k_{10} + k_{1m})X_1 + k_{12}X_2 + KaG \quad X_1 = 0, t = 0 \tag{2}$$

$$dX_2/dt = k_{12}X_1 - k_{21}X_2 \quad X_2 = 0, t = 0 \tag{3}$$

$$dX_m/dt = k_{1m}X_m - k_{m0}X_m \quad X_m = 0, t = 0$$

where $CL = (k_{10} + k_{1m})V_1$, $k_{12} = Q/V_1$, $k_{21} = Q/V_2$, $k_{1m} = f_m CL$ and $k_{m0} = CL_{5FU}/V_{5FU}$ and G is the quantity of drug at the absorption site.

The Laplace transform $Z_m(s)$ of X_m is

$$Z_m(s) = \frac{DKak_{1m}(k_{21} - s)}{(s + Ka)(s + k_{m0})(s + \alpha)(s + \beta)} \tag{4}$$

Using Heaviside's formula, the solution giving the profile of the metabolite (m = 5FU) compartment is:

$$C_m = \frac{K_a k_{1m} D}{V_m}\left(\begin{array}{l}\frac{(k_{21}-Ka)e^{-Kat}}{(k_{m0}-Ka)(\alpha-Ka)(\beta-Ka)} + \frac{(k_{21}-k_{m0})e^{-Km0t}}{(Ka-k_{m0})(\alpha-k_{m0})(\beta-k_{m0})} \\ + \frac{(k_{21}-\alpha)e^{-\alpha t}}{(Ka-\alpha)(k_{m0}-\alpha)(\beta-\alpha)} + \frac{(k_{21}-\beta)e^{-\alpha t}}{(Ka-\beta)(k_{m0}-\beta)(\alpha-\beta)}\end{array}\right) \tag{5}$$

When $Ka = \alpha$, the Laplace transform becomes:

$$Z_m(s)\frac{D\alpha k_{1m}(k_{21} - s)}{(s + \alpha)^2(s + k_{m0})(s + \beta)} \tag{6}$$

and the Heaviside's formula cannot be used anymore. The solution can be obtained however by substituting $Ka = \alpha + h$ in equation 5 and evaluating C_m by determining $\lim_{h->0} C_m$. Then it comes

$$C_m = \frac{\alpha k_{1m} D}{V_m}\left(\begin{array}{l}\frac{t(k_{21}-\alpha)e^{-\alpha t}}{(k_{m0}-\alpha)(\beta-\alpha)} + \frac{\left((k_{21}-\beta)(k_{21}-k_{m0})-(\alpha-k_{21})^2\right)e^{-\alpha tt}}{(k_{m0}-\alpha)^2(\alpha-\beta)^2} \\ + \frac{(k_{21}-k_{m0})e^{-km0t}}{(k_{m0}-\alpha)^2(k_{m0}-\beta)} + \frac{(k_{21}-\beta)e^{-\alpha t}}{(k_{m0}-\beta)(\alpha-\beta)^2}\end{array}\right) \tag{7}$$

The metabolite AUC is:

$$AUC_m = Z_m(0) = Dk_{1m}k_{21}/(\alpha\beta k_{m0})$$

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