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ORIGINAL ARTICLE

D. Ouellet · A. P. Periclou · R. D. Johnson
J. R. Woodworth · R. L. Lalonde

Population pharmacokinetics of pemetrexed disodium (ALIMTA) in patients with cancer

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Abstract *Purpose*: To evaluate the population pharmacokinetics of pemetrexed disodium in cancer patients enrolled in four different open-label, multicenter, nonrandomized phase II studies. *Methods*: Pemetrexed disodium was administered as a 10-min intravenous infusion (600 mg/m^2) every 21 days. A total of four blood samples were to be collected each cycle per patient ($n = 103$ patients) during cycles 1 and 3. Plasma concentration-time data were analyzed by nonlinear mixed-effect modeling using NONMEM to estimate pemetrexed disodium pharmacokinetic parameters (mean, and between- and within-patient variability) as well as relationships between the pharmacokinetic parameters and various patient-specific factors (demographic and physiologic data). *Results/Conclusions*: The pharmacokinetics of pemetrexed disodium were best characterized by a two-compartment model with initial distribution and terminal elimination half-lives of 0.63 h and 2.73 h, respectively. The typical value of systemic clearance (CL) in liters per hour included a relationship to creatinine clearance (CrCL) with a slope of 0.0292. Typical values of central volume (V_c), distributional CL (Q), and peripheral volume (V_p) were 11.3 l, 3.21 l/h, and 5.20 l, respectively. Between-patient variability was 19.6%, 15.6%, and 21.7% for CL, V_c, and V_p, respectively. A combined additive/proportional error model was used to describe residual variability, with a coefficient of variation of 23.7% for the proportional component and a standard deviation of 0.0410 μg/ml for the additive component. Significant patient-specific factors on CL were calculated CrCL, body weight, and to a lesser extent alanine transaminase and folate deficiency. Gender and body weight were significant factors on V_c while both body surface area and albumin were significant factors on V_p. In conclusion, population pharmacokinetic modeling revealed relationships between pharmacokinetic parameters and various patient specific factors.

Key words ALIMTA · Pemetrexed disodium · Population pharmacokinetics · NONMEM

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D. Ouellet · A. P. Periclou · R. L. Lalonde
Phoenix International Life Sciences Inc., Saint-Laurent, Québec, Canada

R. D. Johnson (✉)[1] · J. R. Woodworth
Lilly Research Laboratories, Lilly and Company, Indianapolis, IN, USA

Contact address:
[1] Lilly Laboratory for Clinical Research,
Indiana University Hospital and Outpatient Center,
Rm UH6134, 550 North University Boulevard,
Indianapolis, IN 46202, USA, e-mail: johnson_r_d@lilly.com
Tel.: +1-317-2771717; Fax: +1-317-2778831

Introduction

Pemetrexed disodium (ALIMTA, LY231514) is a novel multitargeted antifolate that inhibits multiple enzymes in the folate cascade such as thymidylate synthase (TS), dihydrofolate reductase (DHFR), and glycinamide ribonucleotide formyltransferase (GARFT) [1, 2]. TS is involved in the de novo synthesis of thymidylates for DNA synthesis while GARFT is involved in the de novo synthesis of purines for DNA synthesis. In preclinical models, pemetrexed has shown antitumor activity against different tumor types including thymidine- and hypoxanthine-deficient murine tumor cell lines as well as human tumor xenograft models. Pemetrexed, an excellent substrate for folypolyglutamate synthase (FPGS), is metabolized to polyglutamates intracellularly. The pentaglutamate form of pemetrexed is the predominant intracellular form and is more potent in its ability to inhibit TS than the monoglutamate [1].

The pharmacokinetics of single-agent pemetrexed in humans have been evaluated separately in three phase I dose-escalation studies. Pemetrexed disodium was

administered as a 10-min intravenous infusion every 21 days [3], as a 10-min infusion every 7 days for 4 weeks followed by 2 weeks of rest [4], and as a 10-min infusion once a day for five consecutive days followed by 16 days of rest [5]. Pemetrexed was shown to have a small steady-state volume of distribution (V_{ss}, about 15 l) suggesting that the compound has limited tissue distribution. Pemetrexed is eliminated rapidly from plasma with terminal elimination half-life values that range from about 2 to 5 h at doses of 525 to 700 mg/m^2. The primary route of elimination is by urinary excretion of unchanged drug (70% to 90% of dose). Pemetrexed exhibits dose-proportional increases in plasma concentrations, since C_{max} and $AUC_{0-\infty}$ values increased linearly with dose in each study.

Based upon results from phase I studies, a 600-mg/m^2 dose administered intravenously over 10 min every 21 days was selected as the phase II dosage regimen. Limited blood samples were collected from cancer patients in phase II studies for pharmacokinetic assessments. The patient population for this analysis consisted of patients with colorectal, pancreatic, breast, and esophageal cancers. A pharmacokinetic analysis was performed on plasma concentration-time data from these initial phase II studies to quantitatively assess systemic pemetrexed exposure. A population approach was used to characterize the pharmacokinetics of pemetrexed using relatively sparse plasma concentration-time data. The objective of this investigation was to construct a pharmacostatistical model to identify the important sources of variability (relevant demographic or pathophysiologic factors) in the pharmacokinetic parameters and estimate the magnitude of the residual variability. Measures of systemic exposure were used in a subsequent safety analysis to help guide further clinical development of this compound.

Materials and methods

Patients

Pharmacokinetic assessments were performed in patients with metastatic colorectal cancer, pancreatic cancer, locally/regionally recurrent or metastatic breast cancer, or esophageal cancer. Male and nonpregnant, nonlactating, female patients, 18 years of age or older were enrolled if they met the following criteria: (1) performance status of 0 to 2 on the ECOG (Eastern Cooperative Oncology Group) scale; (2) a life expectancy of at least 12 weeks; (3) adequate bone marrow reserve as defined by a white blood cell count $\geq 3.5 \times 10^9$/l, platelets $\geq 100 \times 10^9$/l, hemoglobin ≥ 9 g/dl, and absolute granulocyte count $> 2.0 \times 10^9$/l. Patients with inadequate liver function, defined as bilirubin more than 1.5 times above the normal range, aspartate transaminase (AST) and alanine transaminase (ALT) greater than three times normal, with the exception that AST and ALT could be elevated to five times normal in patients with known metastatic disease in the liver, and patients with a calculated creatinine clearance (CrCL) of less than 45 ml/min were excluded from these studies. CrCL was calculated using a modified version of the method of Cockcroft and Gault based upon lean body weight where:

$$\mathrm{CrCL} = [140 - \mathrm{age(years)}] \times \text{lean body weight } (kg) / [71 \times \text{serum creatinine(mg/dl)}]$$

This formula was generated from data collected at the Lilly Clinic, Eli Lilly and Company, and has been used extensively throughout clinical development of pemetrexed.

Drug administration

Pemetrexed disodium was administered as a 10-min intravenous infusion of 600 mg/m^2 every 21 days. The same dosing regimen was used in all patients. A maximum total dose was limited to 1800 mg for patients whose BSA exceeded 3 m^2. Dosage adjustments were permitted at the start of a subsequent course of therapy based on nadir white blood cell count or maximal nonhematologic toxicity from the preceding course of therapy.

Study design

A total of eight blood samples (10 ml) for analysis of pemetrexed in plasma were collected during the first cycle (day 1, four blood samples) and third cycle (day 42, four blood samples). Based upon results from a phase I study using the same dosing schedule [3], blood sampling times were determined and used to construct the blood-sampling paradigm used in this study [6]. Each blood sample was collected according to a randomization schedule during each of four collection intervals which encompassed the optimal sampling times: 0 to 2, 2 to 6, 6 to 12, and 12 to 36 h after the start of the infusion. A predose sample was collected prior to drug administration on the third cycle only. The investigators were asked to document the dose, date, and time of day of pemetrexed administration and sample collection.

Pharmacokinetics and statistical analysis

Plasma pemetrexed concentrations were determined by reverse-phase high-performance liquid chromatography (HPLC). A volume of 0.5 to 1 ml plasma was extracted on a preconditioned solid-phase extraction (SPE) cartridge (Bond Elut Certity II, part no. 1210–2080; Varian, Harbor City, Calif.). The SPE cartridges were preconditioned with 2 ml HPLC-grade methanol, followed by 2 ml of a pH 7.0 phosphate buffer. Immediately following the addition of the sample, the column was washed with 2 ml of the pH 7.0 phosphate buffer, and then with 2 ml methanol. The absorbed pemetrexed was eluted with 2 ml 40% acetonitrile and 60% buffer solution. The eluate was evaporated to dryness under nitrogen. The residues were reconstituted with 200 µl distilled, deionized water and then filtered through 0.1-µm Ultrafree-MC centrifuge filters (Millipore, Bedford, Mass.). The extraction efficiency of pemetrexed from plasma was 60%. The chromatographic procedure consisted of injecting 150 µl of the filtrate onto an octadecyl column (YMCbasic, 25 cm × 4.6 mm; YMC, Wilmington, N.C.) preceded by a YMC basic precolumn (23 cm × 4 mm). The mobile phase consisted of 14% acetonitrile and 86% pH 3.0 phosphate buffer solution, pumped at a rate of 0.8 ml/min, and monitored by UV detection at 250 nm. The internal standard used was dideazatetrahydrofolate (Lometrexol; Eli Lilly, Indianapolis, Ind.), with a retention time of approximately 13 min. The retention time for pemetrexed was approximately 17 min. Two calibration curves were used in the assay of the plasma samples. A low concentration range (10 to 400 ng/ml) was used for the 1-ml plasma samples, and a high concentration range (400 to 20,000 ng/ml) for the 0.5-ml plasma samples. Both concentration curves were linear over their respective ranges, with a correlation coefficient of more than 0.96. The lower limit of quantitation of pemetrexed in plasma was 10 ng/ml.

Plasma concentration-time data were analyzed using a nonlinear mixed-effects model (NONMEM version V) [7] to estimate population pharmacokinetic parameters (mean, between-patient, and residual variability) as well as relationships between the pharmacokinetic parameters and various patient-specific factors (demographic or physiologic data).

The minimum value of the NONMEM objective function (MOF) was used to discriminate between various models during

the model-building process. Since the difference in MOF between a proposed model with one additional parameter and a reduced reference model approximates a χ^2 distribution, any decrease that exceeds 3.8 in the MOF was considered statistically significant. A visual representation of the goodness of fit was assessed by visual inspection of appropriate plots. The following approach, based on discussions by Mandema et al. [8], Maitre et al. [9], and Ette and Ludden [10], was used to identify the final pharmacokinetic model.

1. Identify a basic structural pharmacokinetic model (base model) with no patient-specific factors and determine the best model to estimate interpatient and residual variability. Both open one- and two-compartment models were tested. From the basic model, obtain individual Bayesian estimates of pharmacokinetic parameters for each patient using the POSTHOC option in NONMEM.

2. Identify initial patient-specific factors that may be important determinants of the pharmacokinetics using a generalized additive model (GAM) approach [8] and plots of individual pharmacokinetic parameter estimates versus each patient-specific factor. Following identification of the basic structural pharmacokinetic model, individual estimates of each pharmacokinetic parameter were estimated in NONMEM. Using a GAM procedure in SPLUS (StatSci Division, MathSoft, Seattle, Wash.), individual pharmacokinetic parameter estimates were subjected to stepwise regression analysis starting with CrCL, age, weight (WGT), gender, origin, BSA, albumin (ALB), total protein, AST and ALT as initial patient-specific factors (backward elimination). Selection of statistically significant patient-specific factors for the GAM analysis were identified using the Akaike Information Criterion (AIC).

3. Each significant patient-specific factor from the GAM analysis was evaluated one at a time in NONMEM to obtain a full pharmacostatistical model. Patient-specific factors that produced a statistically significant ($P < 0.05$) decrease in MOF were sequentially added to the model. All continuous variables, with the exception of CrCL, were evaluated as mean/median-centered values of the patient-specific factors. For example, BSA was included on the typical value of volume (TVV) as follows:

$$TVV = \theta_V + \theta_{BSA} \cdot (BSA - 1.8)$$

where θ_V is the population estimate of volume for a typical individual with an average BSA of 1.8 m^2, and θ_{BSA} is the leading linear coefficient of the relationship between volume and BSA.

The effect of CrCL on total systemic pemetrexed CL was modeled as follows:

$$TVCL = \theta_V + \theta_{CrCL} \cdot CrCL$$

where θ_{CL} is the intercept of the total systemic CL (portion of the systemic CL not dependent on calculated CrCL, i.e. when CrCL is equal to 0) and $\theta_{CrCL} \times CrCL$ is the portion of the total systemic CL that is attributed to renal function as assessed by the calculated CrCL with θ_{CrCL} as the leading linear coefficient for the relationship between renal CL and calculated CrCL.

The effects of categorical variables were modeled as indicated below for the effect of gender on TVV:

$$TVV = \theta_V(1 - \theta_{GDR} \cdot GDR)$$

where GDR is an indicator variable equal to 0 in males and equal to 1 in females. Thus, θ_V represents the TVV in male patients and a fraction of that value $(1 - \theta_{GDR})$ represents the TVV in female patients.

4. Attempt to reduce the full model by eliminating each patient-specific factor one at a time from the full model to determine if a model with fewer parameters will describe the data. Model discrimination was based upon the magnitude of the increase in MOF. The *P*-value for statistical significance was adjusted for multiple comparisons ($P < 0.005$). The final model included only those parameters that produced an increase in the objective function of that exceeding 7.879 for one degree of freedom when they were excluded. The error model for residual variability was tested again during the final analysis.

Between-patient (η) variability was modeled using a proportional error structure and reported as percent coefficient of variation (%CV). Between-patient variability was calculated as $100 \cdot \sqrt{\varpi^2}$, where ϖ^2 is the variance of the normal probability distribution of η. The 95% CI for interpatient variability was calculated as $100 \cdot \sqrt{\varpi^2 \pm 2 \cdot SE}$.

Residual variability was most appropriately modeled as a combined additive/proportional error structure as follows:

$$Y = F + \sqrt{(F^2 + \Theta_{err}^2)} \cdot \varepsilon_1$$

where F is the predicted plasma concentration, ε_1 is the proportional component to the residual variability, and Θ_{err} is the proportionality constant between the coefficient of variation of the proportional error term and the standard deviation of the additive term.

Model building was performed using the NONMEM first-order method (FO). The final analysis was performed using the first-order conditional estimation (FOCE) method. For comparison with the final model, parameters of the basic pharmacokinetic model were also estimated using the FOCE method to examine the effect of patient-specific factors on the interpatient variability.

The 95% confidence intervals (CI) of the estimates of fixed-effect pharmacokinetic parameters were calculated by objective function mapping [11]. The overall shape of the parameter space, confirmed the absence of local minima, and identified 95% CIs. The analysis was performed by fixing the parameter of interest to ± 5, 10, 15, 20, 30, 40, 50, and 60% of the population estimate and allowing NONMEM to estimate all other parameters. Changes in the objective function were used to assess the effect of altering the parameter value on the overall fit of the plasma concentration versus time data. The curve produced by the objective function versus parameter value relationship was fitted using polynomial regression to obtain a 95% CI. Assuming a chi-squared distribution, the values which produce a change in the objective function of 3.841 represent the 95% CI for that parameter.

Results

Patient characteristics

A total of 632 plasma concentrations that were obtained from 103 patients who received their first dose of pemetrexed disodium between September 1995 and November 1996 were included in the pharmacokinetic data analysis. Patients received multiple courses of pemetrexed disodium ranging from 1 to 12 courses. Only pharmacokinetic data obtained during the first and third (if applicable) cycles of therapy were used. However, three patients deviated from the standard blood sampling protocol and had additional blood samples collected on the second ($n = 2$) and fourth ($n = 1$) cycles of therapy. These data were also used in the analysis.

The patient characteristics at the time of their first dose of pemetrexed disodium are listed in Tables 1 and 2 for continuous and categorical variables, respectively.

Plasma concentrations

Detectable plasma pemetrexed concentrations ranged from 0.0101 to 225 μg/ml. Scatter plots illustrating the observed pemetrexed plasma concentrations versus time profiles following the 10-min intravenous infusion during cycles 1 and 3 are shown in Fig. 1.

During the third cycle (about day 42), plasma pemetrexed concentrations were measured immediately prior to administration of the dose (predose sample). Residual plasma pemetrexed concentrations were

Table 1 Patient characteristics: continuous variables (values of the variables at the time of administration of the first dose)

Continuous variables	Mean ± SD[a]	Median	Range
Age (years)	59.6 ± 10.2	58.4	33.9–79.1
Weight (kg)	72.0 ± 16.9	71.7	34.0–138.3
Body surface area (m^2)	1.82 ± 0.24	1.80	1.26–2.49
Creatinine clearance (ml/min)	78.6 ± 20.2	76.8	46.0–141.0
Albumin (g/dl)	3.7 ± 0.5	3.7	2.2–4.9
Total protein (g/dl)	7.1 ± 0.5	7.1	5.8–8.8
AST (U/l)	29 ± 20	23	13–159
ALT (U/l)	30 ± 24	22	5–166
ALK (mU/ml)	141 ± 121	96	32–713
HCYS (μmol/l)	12.1 ± 13.4	10.2	4.7–132.4
MMA (μmol/l)	296 ± 912	132	29–8507
CYST (μmol/l)	241 ± 191	183	50–1303

[a] $n = 103$ patients with the exception of Weight ($n = 100$), HCYS ($n = 90$), MMA ($n = 90$), CYST ($n = 90$)

Table 2 Patient characteristics: categorical variables

Categorical variables	Categories	Count (%)[a]
Gender	Male	54 (52.4)
	Female	49 (47.6)
Ethnic origin	Caucasian	89 (86.4)
	African-American	10 (9.7)
	Hispanic	2 (1.9)
	Southeast Asian	1 (1.0)
	Other	1 (1.0)
Performance status	0	43 (42.6)
	1	50 (49.5)
	2	8 (7.9)
Primary tumor	Colorectal	37 (35.9)
	Pancreatic	40 (38.8)
	Breast	13 (12.6)
	Esophageal	13 (12.6)
Previous radiotherapy	No	86 (83.5)
	Yes	17 (16.5)
Baseline liver metastases	No	60 (58.3)
	Yes	43 (41.7)
Folate deficiency[b]	No	101 (98.1)
	Yes	2 (1.9)

[a] $n = 103$ patients with the exception of Performance status ($n = 101$)
[b] Value of the variable at the time of administration of the first dose

above the minimum limit of quantitation in 20 out of 57 patients at approximately 3 weeks after administration of the previous dose. These predose plasma concentrations ranged from 0.011 to 0.3253 μg/ml (mean 0.0522 μg/ml, median 0.0223 μg/ml) which is in the same range as the plasma concentrations observed 24–36 h postinfusion. Inclusion of these plasma concentrations resulted in a very prolonged terminal phase which may represent a very deep distribution compartment or slow release from a tight binding site. Considering the magnitude of the predose concentrations relative to those obtained during the first 36 h postinfusion, this apparent long terminal phase would have little impact on the overall disposition of pemetrexed. Therefore, these predose concentrations were excluded from the analyses.

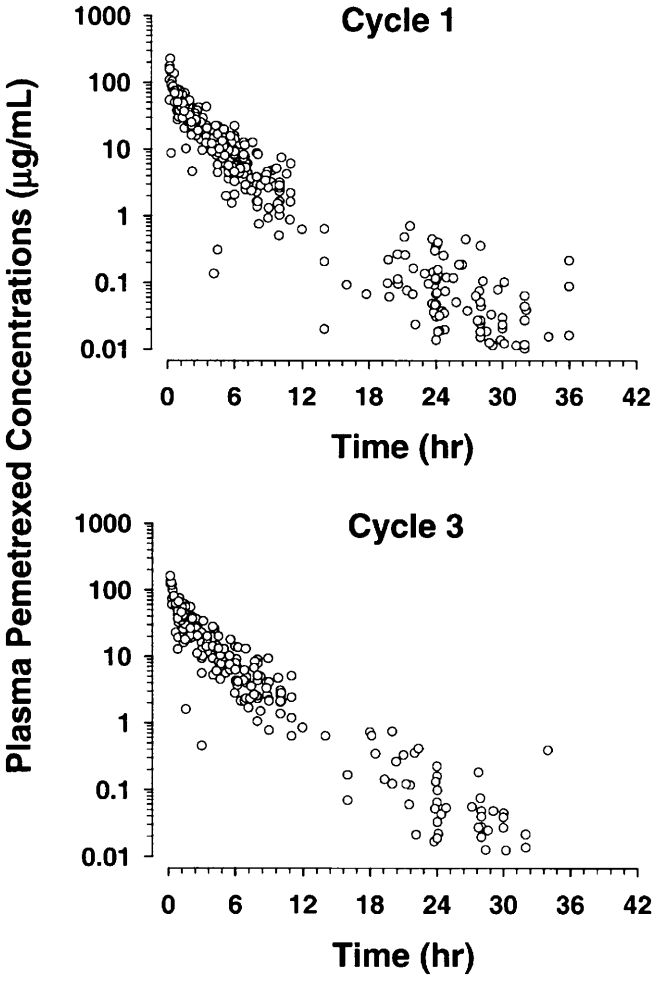

Fig. 1 Observed plasma pemetrexed concentrations vs time profiles (pemetrexed concentrations were measured on the second cycle of drug administration in two patients and on the fourth cycle of drug administration in one patient)

Pharmacokinetic analysis

The plasma concentration-time data was best fitted to an open two-compartment model. Prior to evaluation in NONMEM, individual estimates of CL, V_c, and V_p were subjected to a GAM analysis to prescreen patient-specific factors. Based upon the results of the GAM analysis, those patient-specific factors that were selected by the GAM analysis for each parameter were added one at a time to the NONMEM model in the following order: CrCL and WGT on CL, Gender and WGT on V_c, WGT and BSA on V_p, ALB, AST, and ALT on CL, and ALB and TP on V_p. Each continuous or categorical patient-specific factor was added in NONMEM as described earlier. Alternative order of addition of these patient-specific factors as well as other patient-specific factors [e.g. folate deficiency (FOL), alkaline phosphatase (ALK), baseline liver metastases (BLM)] were also tested in NONMEM. Folate deficiency was defined by the following criteria: homocysteine (HCYS) > 13.9 μM, cystathionine (CYS) > 342 nM, and

methylmalonic acid (MMA) >73 nM and <271 nM. In addition to testing folate deficiency as a categorical variable, vitamin metabolite concentrations (HCYS, CYS, and MMA) were evaluated. The effects of the addition of HCYS, CYS, and MMA on systemic CL were not statistically significant, and these variables were excluded from the model.

Since there were several missing values in these parameters, the model was tested both by excluding patients with missing data, and by assigning a value (median value or value at another time-point). Since the results obtained from testing the model with excluded patients were consistent with those obtained with assigned values, the final analysis included all patients with missing values replaced with their respective assigned values.

After completing the model-building process, statistically significant patient-specific factors in the full model included CrCL, WGT, ALT, FOL, and ALK on CL, GDR and WGT on V_c, as well as BSA and ALB on V_p. A final model was developed by removing each patient-specific factor one at a time from the full model as described above. During the model reduction process, the increase in the MOF was found to be not statistically significant when ALK was removed from the model for CL. Therefore, this was the only factor from the full model that was not included in the final model.

Estimates of the population pharmacokinetic parameters, i.e. V_c, Q, and V_p, interpatient variability on CL, V_c and V_p, and residual variability are shown in Tables 3 and 4. The population predicted versus observed concentrations and individual predicted versus observed concentrations (Fig. 2) were plotted to examine the goodness of fit of the model. In this patient population, individual estimates of CL, V_c and V_p ranged from 2.68 to 9.63 l/h, 5.02 to 19.8 l, and 2.77 to 11.7 l, respectively.

The final model produced the following relationships between TVCL, TVV_c, and TVV_p, and the patient-specific factors:

$$\mathrm{TVCL} = [\theta_{\mathrm{CL}} + \theta_{\mathrm{CrCL}} \cdot \mathrm{CrCL} + \theta_{\mathrm{WGT}} \cdot (\mathrm{WGT} - 70) + \theta_{\mathrm{ALT}} \cdot (\mathrm{ALT} - 30.5)] \cdot (1 - \theta_{\mathrm{FOL}} \cdot \mathrm{FOL})$$

Table 4 Population estimates of the random effects of the final model (see text for a description of the final pharmacokinetic model)

Variability	Estimate (95% CI)
Interpatient	
CL	19.6 (14.6, 23.6)
V_c	15.6 (0, 23.7)
Q	–
V_p	21.7 (11.3, 28.5)
Residual[a]	
Additive	0.0411 (0.0224, 0.0537)
Proportional	23.7 (19.4, 27.3)

[a] Standard deviation (μg/ml)

where FOL is an indicator variable equal to 0 in patients with normal functional folate status and equal to 1 in patients with folate deficiency.

$$\mathrm{TVV_c} = [\theta_{\mathrm{V_c}} + \theta_{\mathrm{WGT2}}(\mathrm{WGT} - 70)] \cdot (1 - \theta_{\mathrm{GDR}} \cdot \mathrm{GDR})$$

where GDR is an indicator variable equal to 0 in male patients and equal to 1 in female patients.

$$\mathrm{TVV_p} = \theta_{\mathrm{V_p}} + \theta_{\mathrm{BSA}}(\mathrm{BSA} - 1.8) + \theta_{\mathrm{ALB}} \cdot (\mathrm{ALB} - 3.67)$$

Since CL is inversely proportional to overall systemic exposure as assessed by the area under the plasma concentration-time curve, the relationships between CL and those important covariates that affect overall systemic exposure (CrCL and WGT) are illustrated in Fig. 3.

Estimates of the leading linear coefficients for the relationships between each patient-specific factor and the pharmacokinetic parameters are also listed in Table 3. In addition, the pharmacokinetic parameter values at the minimum and maximum value of each patient-specific factor are included in Table 3 to provide an estimate of the expected range of possible values for each parameter. These ranges were determined after fixing the values of all other patient-specific factors to their respective mean/median-centered values and fixing the calculated CrCL equal to the population average value of 78.6 ml/min.

In the final model, after accounting for the effects of the different patient-specific factors, interpatient vari-

Table 3 Population estimates of the fixed effects in the final model (see text for a description of the final pharmacokinetic model)

Parameter	Estimate (95% CI)	Range of value of parameter (range of value of covariate)
θ_{CL} (l/h)[a]	2.82 (2.315, 3.33)	
θ_{CrCL}	0.0292 (0.0226, 0.0358)	3.77–7.37 l/h (33–156 ml/min)
θ_{WGT}	0.0475 (0.0368, 0.0590)	3.40–8.35 l/h (34.0–138.3 kg)
θ_{ALT}	0.0041 (0.0023, 0.0058)	5.00–7.01 l/h (5–495 U/dl)
θ_{FOL}	0.344 (0.212, 0.479)	5.11 or 3.35 l/h (1 = No or 2 = Yes)
$\theta_{\mathrm{V_c}}$ (l)	11.3 (9.9, 13.01)	
θ_{GDR}	0.325 (0.222, 0.418)	11.3 or 7.64 l (1 = M or 2 = F)
θ_{WGT2}	0.105 (0.046, 0.162)	7.52–20.3 l (34.0–138.3 kg)
θ_{Q} (l/h)	3.21 (2.46, 4.19)	
$\theta_{\mathrm{V_p}}$ (l)	9.78 (7.69, 12.06)	
θ_{BSA}	4.28 (2.662, 6.028)	2.67–8.15 l (1.21–2.49 m^2)
θ_{ALB}	−1.25 (−0.732, −1.793)	7.91–3.66 l (1.5–4.9 U/dl)

[a] Represents the nonrenal clearance

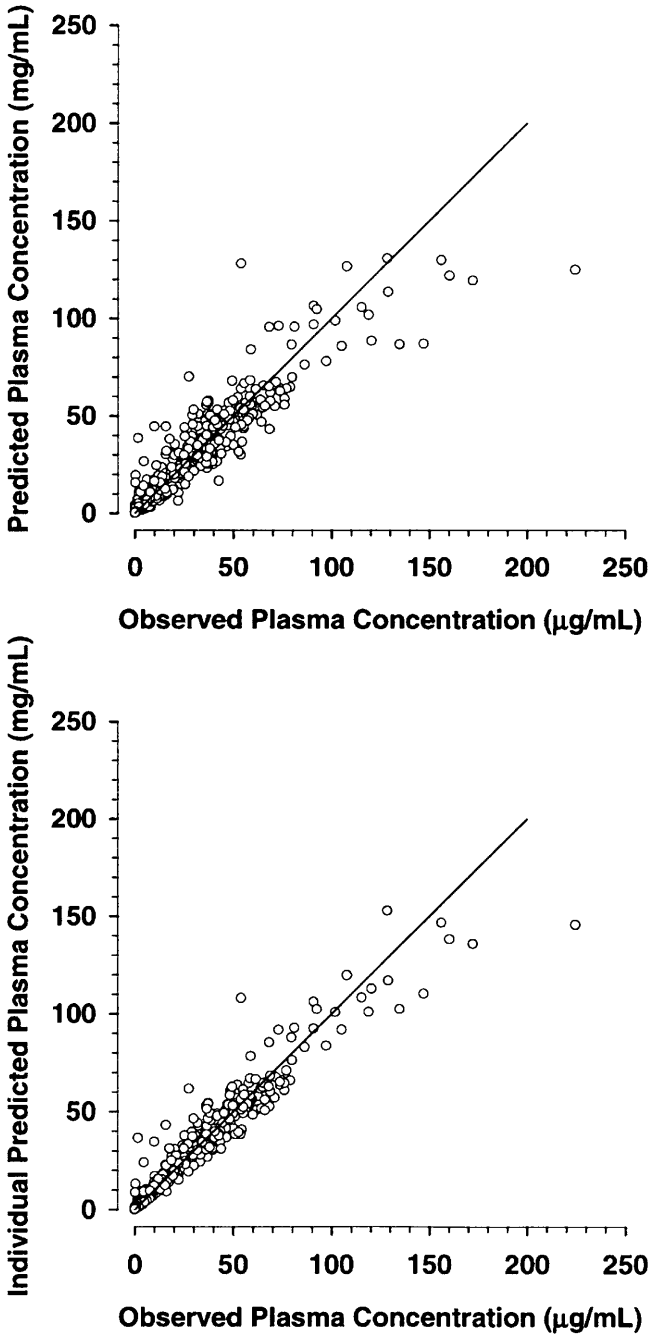

Fig. 2 Observed plasma pemetrexed concentrations vs population predicted concentrations (*top*) and individual predicted concentrations (bottom). The *solid lines* are the lines of identity

ability was 19.6%, 15.6%, and 21.7% for CL, V_c, and V_p, respectively. The addition of patient-specific factors to the basic pharmacokinetic model produced decreases in interpatient variability (%CV) in CL, V_c, and V_p of 4.6%, 7.6% and 14.8%, respectively, from the basic pharmacokinetic model (FOCE method). The proportional portion of the residual variability decreased by 3.5% while the additive component (θ_{err}) of residual variability increased slightly from 0.142 to 0.173.

Residual variability was characterized by a proportional error component with a coefficient of variation of 23.7% and a standard deviation for the additive error term of 0.0410 μg/ml. Thus, at higher concentrations, the proportional error component would dominate with an overall residual variability of approximately 23.7%. At lower concentrations, especially those approaching the minimum limit of quantitation for the drug assay, the influence of the additive term would become dominant. The coefficient of variation was calculated for different plasma pemetrexed concentrations. For plasma pemetrexed concentration of 0.01, 0.1, 1, and 10 μg/ml,

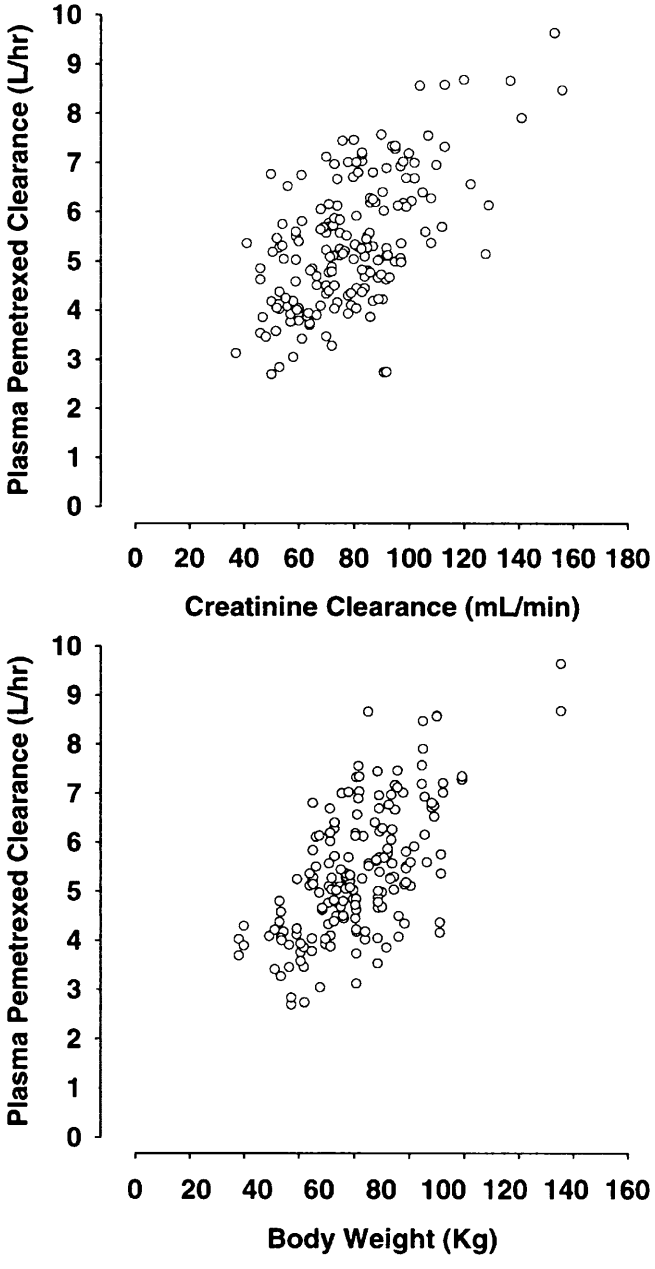

Fig. 3 Relationship between total plasma CL, CrCL (*top*), and body weight (*bottom*)

the residual variability is expected to be 410.8%, 47.4%, 24.1%, and 23.7%, respectively.

Discussion

The population pharmacokinetics of pemetrexed administered as a 10-min intravenous infusion in cancer patients were adequately characterized by a two-compartment model. Based on the final population parameter estimates, pemetrexed pharmacokinetics were demonstrated to have an initial/distribution half-life of 0.63 h, an effective terminal/elimination half-life of 2.73 h, and a volume of distribution at steady-state of 16.5 l. These values are consistent with those from a previous phase I study in which a terminal half-life of 2 h and a steady-state volume of distribution of 6 l/m^2 (about 11 l) were found. Interpatient variability of the pharmacokinetic parameters was relatively small with coefficients of variation of 22% or less. Therefore, the pharmacokinetics of pemetrexed behaved in a highly predictable fashion.

The patient-specific factors tested in NONMEM were preselected based on results from a GAM analysis, which represents an advantage in terms of time of analysis. One limitation to the above approach is that only the value of each patient-specific factor observed at the time of the first dose was used for this analysis, even if the value of the variable changed with time for a patient. During the NONMEM analysis, the numerical value of each patient-specific factor that corresponded to the time and date of the actual administered dose was used. Hence, time-dependent changes in patient-specific factors were incorporated into the model-building process.

The typical value of CL was found to be dependent on CrCL, body weight, alanine transferase, and folate deficiency. At the population mean CrCL of 78.6 ml/min, total systemic CL of pemetrexed was 5.11 l/h. Of the total CL, 45% was attributed to renal function. The proportion of the systemic CL that could be attributed to renal function in the population analysis was lower than found in a phase I study in which 70% of the dose was excreted into urine as unchanged drug [3]. In this patient population, the lowest value of CrCL was 33 ml/min. Thus, extrapolation of systemic CL when the CrCL is near 0 may not be accurate. Based on the results of this analysis, the systemic CL would be 2.81 l/h in patients with no renal function.

The coefficient for folate deficiency indicated that patients who were folate deficient had a systemic CL that was 67.3% of the CL observed in patients with no folate deficiency. Only 2 out of 103 patients were categorized as being folate deficient and 1 additional patient changed from no deficient to being folate deficient between the first and third course. Since these results were generated based on a very small number of patients, the effect of folate deficiency on systemic CL is inconclusive prompting further evaluation in future studies.

Although ALT remained in the final model for CL as a statistically significant patient-specific factor, the overall effect of this enzyme level on CL was minimal. A 100-fold increase in ALT values would be expected to produce only a 1.4-fold (Table 4) increase in CL.

All three pharmacokinetic parameters CL, V_c and V_p were dependent on some measure of body size. Both CL and V_c were dependent on weight while V_p was dependent on BSA. When body weight or BSA was first introduced in the NONMEM model, the alternate body size measure was tested as well. The appropriate patient-specific factor was selected solely based on the MOF value. Since body weight is correlated to a large extent with BSA, we substituted BSA for body weight on CL in the final model to evaluate its effect. Inclusion of BSA in place of body weight resulted in a small increase in MOF (< 3 units) but still significant relative to the base model. Even though body weight was slightly better than BSA for predicting systemic CL, it appears that dosing patients based upon BSA is also an appropriate dosing strategy.

The effect of gender on V_c was not solely related to differences in body weight between male and female patients since both variables were necessary in the model. Male patients had a 1.5-fold larger volume of central the compartment relative than female patients. However, this difference is probably not clinically significant. In addition to a relationship with BSA, a reverse relationship was observed between albumin and V_p. In patients with higher albumin concentrations, the volume of distribution of the peripheral compartment would be smaller.

The primary determinant of systemic pemetrexed exposure is the area under the plasma concentration-time curve which is inversely proportional to CL. Changes in patient-specific factors that alter CL will impact overall drug exposure. Since drug toxicity is partially related to systemic exposure, factors affecting exposure may affect the toxicity profile of the drug. Patient-specific factors were evaluated with respect to the volume terms to try to explain as much of the variability in the data as possible. Therefore, relationships between patient-specific factors and the volume terms are not as clinically meaningful as those that affect CL but are included in the analysis to account for all possible sources of variability in pharmacokinetics.

In this evaluation, we characterized the pharmacokinetics of pemetrexed in patients with metastatic colorectal cancer, pancreatic cancer, locally/regionally recurrent or metastatic breast cancer, or esophageal cancer. Monitoring plasma concentrations of cancer agents has been shown to be useful in different cases where a relationship between pharmacokinetics and the efficacy and/or toxicity exists. The pharmacokinetic model described here was used to predict pemetrexed concentrations in these patients and to develop a model describing the relationship between plasma concentrations and dynamic response. In conclusion, population pharmacokinetics modeling revealed

relationships between various patient specific factors and pharmacokinetic parameters.

References

1. Shih C, Chen VJ, Gossett LS, Gates SB, MacKellar WC, Habecks LL, et al (1997) Pemetrexed disodium, a pyrrolo [2,3-d]pyrimidine-based antifolate that inhibits multiple folate-requiring enzymes. Cancer Res 57: 1116–1123
2. Jackman AL, Calvert AH (1995) Folate-based thymidylate synthase inhibitors as anticancer drugs. Ann Oncol 6: 871–881
3. Rinaldi DA, Kuhn JG, Burris HA, Dorr FA, Rodriquez G, Eckhardt SG, Jones S, Woodworth JR, Baker S, Langley C, Mascorro D, Abrahams T, Von Hoff DD (1999) A phase I evaluation of multitargeted antifolate (MTA, pemetrexed disodium), administered every 21 days, utilizing the modified continual reassessment method for dose escalation. Cancer Chemother Pharmacol 44: 372–380
4. Rinaldi DA, Burris HA, Dorr FA, Woodworth JR, Kuhn JG, Eckardt JR, Rodriguez G, Corso SW, Fields SM, Langley C, Clark G, Faries D, Lu P, Von Hoff DD (1995) Initial phase I evaluation of the novel thymidylate synthase inhibitor pemetrexed disodium, using the modified continual assessment method for dose escalation. J Clin Oncol 13(11): 2842–2850
5. McDonald AC, Vasey PA, Adams L, Walling J, Woodworth JR, Abrahams T, McCarthy S, Bailey NP, Siddiqui N, Lind MJ, Calvert AH, Twelves CJ, Cassidy J, Kaye SB (1998) A phase I and pharmacokinetic study of pemetrexed disodium, the multitargeted antifolate. Clin Cancer Res 4(3): 605–610
6. D'Argenio DZ (1981) Optimal sampling times for pharmacokinetic experiments. J Pharmacokinet Biopharm 9: 739–756
7. Beal SL, Sheiner LB (eds) (1992) NONMEM user's guides. NONMEM Project Group, University of California, San Francisco
8. Mandema JW, Verotta D, Sheiner LB (1992) Building population pharmacokinetic-pharmacodynamic models. I. Models for covariate effects. J Pharmacokinet Biopharm 20: 511–528
9. Maitre PO, Bührer M, Thomson D, Stanski DR (1991) A three-step approach combining bayesian regression and NONMEM population analysis: application to midazolam. J Pharmacokinet Biopharm 19: 377–384
10. Ette EI, Ludden TM (1995) Population pharmacokinetic modeling: the importance of informative graphics. Pharm Res 12: 1845–1855
11. Holford NH, Peace KE (1992) Methodologic aspects of a population pharmacodynamic model for cognitive effects in Alzheimer patients treated with tacrine. Proc Natl Acad Sci USA 89: 11466–11470