PHARMACOKINETICS AND DRUG DISPOSITION

A pharmacokinetic-pharmacodynamic disease model to predict in vivo antiviral activity of maraviroc

Background: **The viral dynamics of human immunodeficiency virus (HIV) infection has been widely studied and expressed as mathematic equations. For most of the current registered antiretroviral drugs, the pharmacokinetics is well characterized and some relationships with the viral load–time profiles in plasma from HIV patients have been established. The integration of these models in a pharmacokinetic (PK)–pharmacodynamic (PD)–disease model can help toward a better understanding of the complexity of the interactions, as well as in the identification and clarification of the current model assumptions.**

Methods: **This work describes the development of a generic PK-PD disease model for a short-term (10 days) monotherapy phase IIa study with a novel anti-HIV drug, maraviroc (UK-427,857). The disease component of the model was based on the model published by Bonhoeffer et al, which was adapted for short-term treatment and for the new mechanism of action, CCR5-receptor antagonism. The model parameters were derived from the literature, as well as from a model-based analysis of available phase IIa clinical data from another investigational antiretroviral drug. The PD component that links the plasma concentrations of maraviroc to the inhibition of virus replication was based on in vitro measurements of drug potency and took into account the difference in the in vitro and in vivo protein binding and the uncertainties regarding the interpretation of the in vitro to in vivo extrapolation of the 50% inhibitory concentration. Finally, the PK component was based on information obtained from a single-dose study in healthy volunteers.**

Results: **The integrated PK-PD disease modeling allowed prediction of the effect on viral load of different maraviroc doses given as monotherapy to drug-naive patients.**

Conclusions: **By making use of the available PK-PD disease model, the possible range of active oral doses for maraviroc in HIV-positive patients was estimated by simulation before any clinical trials were taking place. The use of a model-based approach for selecting doses for clinical phase IIa has improved and accelerated the drug's development. This model was a powerful tool for assisting in the design of clinical studies on new agents for treating HIV/acquired immunodeficiency syndrome. (Clin Pharmacol Ther 2005;78:508-19.)**

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Maraviroc is a novel CCR5-receptor antagonist that is being developed for the treatment of human immunodeficiency virus (HIV) infection. It is a potent antagonist of the CC-chemokine receptor CCR5, which acts as a coreceptor for HIV entry into target cells.[1] Mathematic models have been widely used to describe viral

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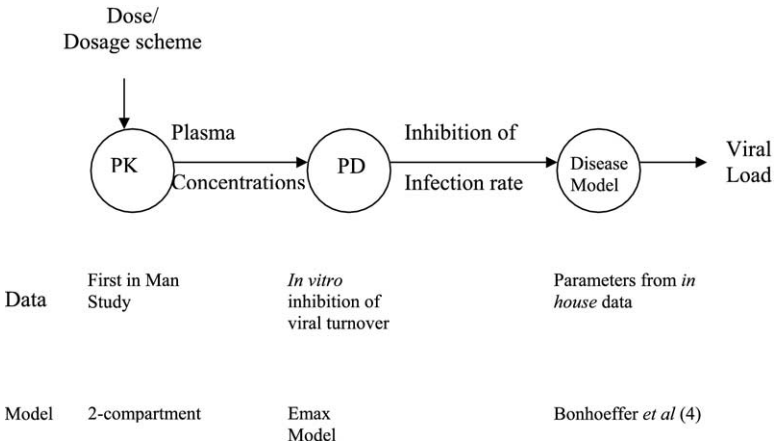

Fig 1. Schematic representation of pharmacokinetic (PK)–pharmacodynamic (PD)–disease model for an antiretroviral drug. E_{max}, Maximum effect.

and cellular dynamics during HIV-1 infection for the purpose of understanding the biologic process of the disease and to gain insights into the dynamics of HIV-1 infection in the presence and absence of antiretroviral drugs.[2-4] The dynamics of HIV-1–infected cells changes in a nonlinear fashion with respect to time and plasma concentration of antiviral drugs.[4] The impact of these different factors on the system can be assessed only through simulations by use of an integrated pharmacokinetic (PK)–pharmacodynamic (PD)–disease model.

The basic reproductive ratio (R_0) concept is widely used in the field of infectious disease dynamics and the evolution of pathogens but to date has not been applied to the interpretation of data from clinical studies. R_0 can be defined as the capacity of viral replication within the patient at a given time. This can be defined as the average number of offspring (or infected cells) generated by a single virus particle, in the absence of constraints.[4] To maintain infection within the host, R_0 has to be greater than 1, because below this value, the pathogen is unable to produce enough offspring to maintain infection. Depending on the R_0 in the test tube (not known), the 90% effective concentration (EC_{90}) could be translated to different values of receptor inhibition and, therefore, to different values of 90% inhibitory concentration (IC_{90}).

The purpose of this report is to describe the modeling and simulation activities carried out to investigate and assess the impact of maraviroc in vitro antiviral efficacy in the design of a short-term monotherapy study in HIV-1–infected patients. We address how a published mathematic modeling framework developed for antiretroviral drugs with different mechanisms of action, such as protease inhibitors and reverse transcriptase inhibitors, was adapted for CCR5-receptor antagonists. We then describe how the cell dynamics model (disease model) was linked to an in vitro inhibitory model (PD model) and then linked to a PK model to form an integrated PK-PD disease model.

Furthermore, we illustrate how this PK-PD disease model was used to aid translational pharmacology in extrapolating in vitro to in vivo information. We also show how modeling and simulation were used to identify limitations and uncertainties and to help in optimizing the design of a monotherapy study before any clinical efficacy data from maraviroc were available.

METHODS

The PK-PD disease model described in this report consists of the following 3 components: a disease model, a PD model, and a PK model. The PD model links the PK model with the disease model. The modular structure of the model allows for the integration of information from different sources such as the literature and in vitro and in vivo data. Fig 1 shows a schematic representation of how the 3 models are linked.

Disease model. A mathematic model was used to simultaneously characterize the viral dynamics in different HIV-1–infected cell compartments as a function of time. This mathematic model was based on a model previously developed by Bonhoeffer et al.[4,5] From the model of viral dynamics described in this report, only 3 types of cells were considered, as follows: target $CD4^+$ cells (T), actively infected cells (A), and latently infected cells (L), as well as the virus particles (V). The persistently infected cells (P) with a very long half-life (>1000 days) do not affect the viral load response

Table I. Estimated parameter values used in disease model

Parameter	*Symbol*	*Unit*	*Value*	*Comments and reference*
Birth rate constant of uninfected cells	b	cells/mL · d	0.16-8.8	Estimated
Death rate constant of uninfected cells	d_1	d^{-1}	0.006	Fixed, 0.002-0.01 (Funk et al[6])
Infection rate of activated $CD4^+$ cells	i	mL/virion	0.00002-0.0003	Estimated, 2×10^{-8} to 3×10^{-7} (Funk et al[6])
Death rate constant of actively infected cells	d_2	d^{-1}	0.49-1.79	Estimated (Funk et al[6] and Perelson et al[2,3])
Death rate constant of latently infected cells	d_3	d^{-1}	0.0132	Fixed, limits (0.002-0.12) (Funk et al[6])
Activation rate constant of latently infected cells	a	d^{-1}	0.037	Fixed, 0.004-0.007 (Funk et al[6])
Fraction of newly infected cells that become actively infected cells	f_1		91%-97.9%	Estimated, 75%-99.8% (Funk et al[6])
Fraction of newly infected cells that become latently infected cells	f_2		$1 - f_1$	Estimated, 0.022 (Funk et al[6])
Virus production rate	p	Particles · d^{-1}	1240/10	Fixed, 230-2500 (Funk et al[6])
Clearance rate of free virus	c	d^{-1}	35/10	Fixed, 28-42 (Funk et al[6])
Basic reproductive ratio	R_0		1.3-17.6/ 1.1-11.6	Estimated, 2.46
σ^2	0.04	Y = F · exp(EPS)		

exp(EPS), Exponential errors model.

during only 10 days of therapy and, therefore, were not included in our model. The relationship between infectious virus and each type of cell was mathematically characterized by differential equations (equation 1). Target uninfected cells are activated at constant rate b, from a pool of $CD4^+$ cells, and die with death rate constant d_1. The virus infects activated uninfected $CD4^+$ cells to produce infected cells (actively and latently) at rate constant i. Actively and latently infected cells are cleared at rate constants d_2 and d_3, respectively. The designations f_1 and f_2 represent the fractions of infected $CD4^+$ cells that become actively and latently infected ($f_1 + f_2 = 1$). Latently infected cells can become reactivated at rate constant a. The virus is produced from actively infected cells with rate constant p and is cleared with rate constant c.

$$\begin{aligned} dT/dt &= b - d_1T - iVT \\ dA/dt &= f_1iVT - d_2A + aL \\ dL/dt &= f_2iVT - d_3L - aL \\ dV/dt &= pA - cV \end{aligned} \quad (1)$$

After the start of treatment with an antiretroviral drug (eg, maraviroc), a new parameter had to be introduced into the equations. This parameter reflects the degree by which maraviroc inhibits the infection rate constant. Entry inhibitors act by reducing the infection rate constant, i, by factor INH. The effectiveness of maraviroc at blocking viral replication is then given by the expression 1 − INH. During treatment, equation 1 can thus be presented as follows:

$$\begin{aligned} dT/dt &= b - d_1T - (1 - INH)iVT \\ dA/dt &= f_1(1 - INH)iVT - d_2A + aL \\ dL/dt &= f_2(1 - INH)iVT - d_3L - aL \end{aligned} \quad (2)$$

INH represents the inhibition fraction by a CCR5-receptor antagonist as described in the PD model and can take values between 0 and 1. It assumes a value of 0 in the absence of the drug and a value of 1 when the drug reaches its maximal inhibition. If the drug reaches maximal inhibition, the virus eventually becomes extinct. In-house clinical data from another antiretroviral investigational compound were used to derive the variability associated with disease model parameters. The individual HIV-1 ribonucleic acid (RNA) (copies per milliliter) measurements were organized in a longitudinal data set. Some unidentifiable model parameters were fixed to values in the literature,[6] as follows: $d_1 = 0.006\ d^{-1}$, $d_3 = 0.0132$

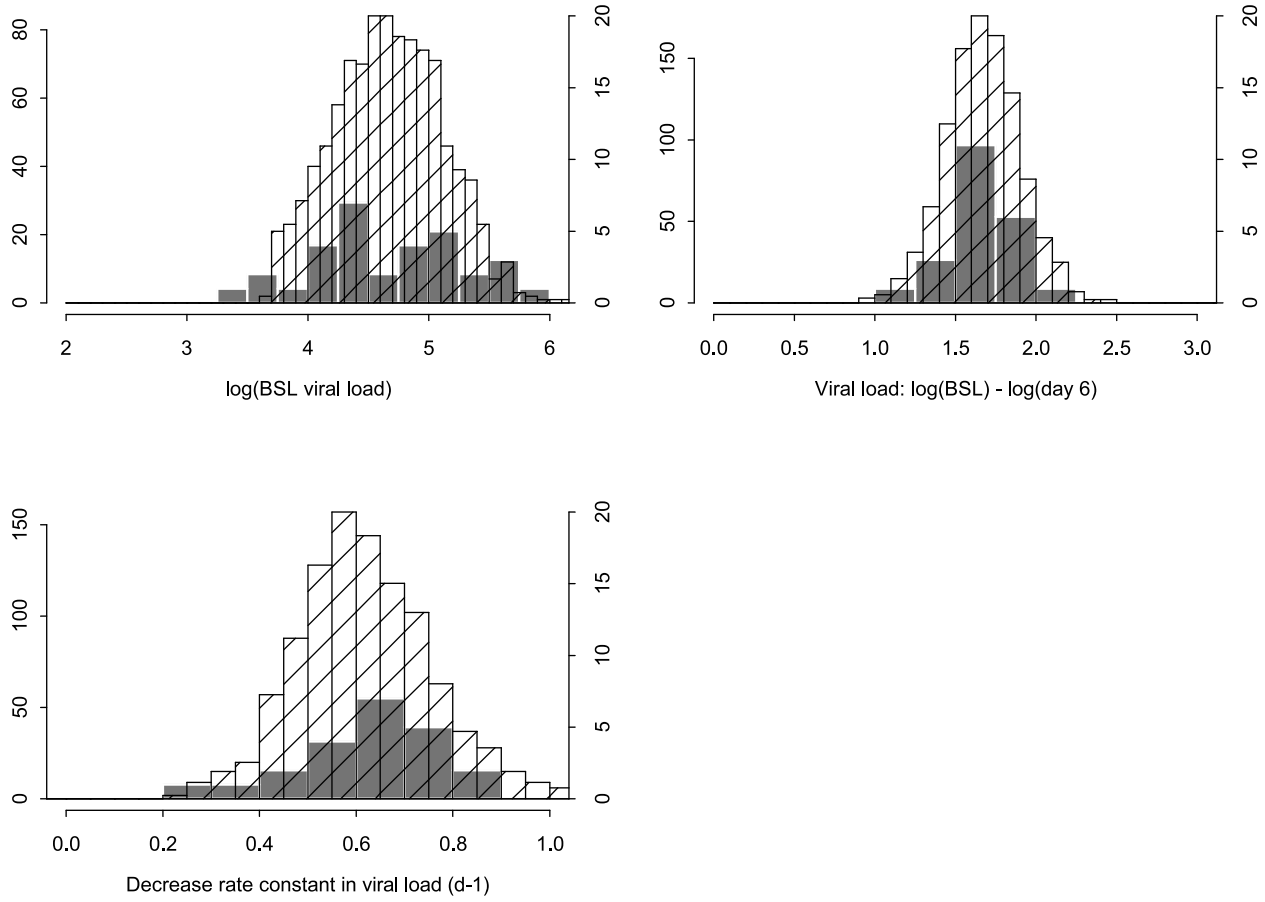

Fig 2. Posterior predictive check of disease model. *Blue bars* represent results from simulations by use of the model, and *green bars* represent measured viral load from an investigational drug. BSL, Baseline.

Table II. Mean values of in vitro antiviral assessment of maraviroc in various cell cultures

	Minimum	*Mean*	*Maximum*
In vitro*			
EC_{50} (nmol/L)	0.08	0.20	0.56
EC_{90} (nmol/L)	0.40	1.05	3.10
In vitro†			
EC_{50} (ng/mL)‡	0.15	0.38	1.04
EC_{90} (ng/mL)	0.74	1.94	5.74

EC_{50} and EC_{90}, 50% and 90% effective concentrations, respectively.
*Not protein binding–adjusted, expressed as free concentrations.
†Protein binding adjusted from in vitro to in vivo, expressed as total concentrations.
‡Molecular weight, 513.7.

d^{-1}, a = 0.037 d^{-1}, p = 1240/10 particles · d^{-1}, and c = 35/10 d^{-1}. The parameters b, i, f_1, f_2, and d_2 were estimated in a stepwise manner. First, the individual model parameters were estimated for each

Table III. Estimated IC_{50} and IC_{90} for different values of basic reproductive ratio for observed EC_{90} of 5.74 ng/mL

R_0 in vitro	*IC_{50} (ng/mL) of infection rate*	*IC_{90} (ng/mL) of infection rate*
1.5	11.5	103
2	**5.74**	51.7
3	2.87	25.8
4	1.91	17.2
5	**1.44**	12.9
6	1.15	10.3
7	0.96	8.61
8	0.82	7.38
9	0.72	6.46
10	**0.64**	5.74

The IC_{50} values used in simulated scenarios are shown in boldface type.
IC_{50} and IC_{90}, Drug concentrations that give 50% and 90%, respectively, of the inhibition of the viral replication; R_0, basic reproductive ratio.

Table IV. Posterior predictive check of pharmacokinetic model

Dose (mg)	*Observed C_{min} (ng/mL)*			*Simulated C_{min} (ng/mL)*		
	Minimum	*Geometric mean*	*Maximum*	*5th percentile*	*Geometric mean*	*95th percentile*
10	0.39	0.50	0.78	0.41	0.62	0.96
30	0.61	1.59	3.74	1.13	1.91	3.26
100	4.55	6.16	7.46	3.14	5.77	11.1
300	10.5	18.1	24.9	10.0	19.0	37.7
900	26.1	46.5	70.3	31.9	59.6	118
1200	31.1	65.6	136	45.7	81.9	174

C_{min}, Minimum concentration; C_{max}, maximum concentration.

patient by use of a nonlinear least squares approach with NONMEM.[7] Second, the geometric means from individual parameters were calculated and are presented in Table I. Finally, the model was fitted simultaneously to all patients, and the parameters b, i, f_1, f_2, and d_2 were set to the geometric means obtained from the individual fits. The mean and covariance parameters, which were used in simulations for sampling from a multivariate normal distribution, were computed from POSTHOC individual parameters through log transformation of b, i, and d_2 and logit transform of f_1 and f_2 parameters.

A posterior predictive check of the data used for parameter estimation was performed to evaluate the predictive performances of the disease model. Fig 2 shows that simulated data are in good agreement with the observed data.

PD model. Antiviral activity of a drug can be assessed in vitro by the degree of viral replication inhibition induced by the drug in a cell-based assay system.[8] In vitro assessment of maraviroc antiviral activity was determined in various cell cultures that represented the target cells for the virus in humans. The test used the measurement of the drug-dependent decrease in reverse transcriptase activity in infected cell supernatants after 5 days of incubation at various concentrations of UK-427,857. The 50% inhibitory concentration (IC_{50}) and IC_{90} values of maraviroc were determined from the observed concentration-replication inhibition curves. The mean, minimum, and maximum reported values (HIV Ba-L in pooled peripheral blood lymphocytes with monocytes) of IC_{50} and IC_{90} were corrected for protein binding in the incubation medium, as well as in human plasma. The bound fractions in human plasma and in incubation medium were 0.79 and 0.243, respectively. A scaling factor of 3.6 was used to correct for protein binding differences. The maximum values for IC_{50} and IC_{90} were estimated to be 1.04 and 5.74 ng/mL, respectively (Table II). This information was used in the PD model by use of a sigmoid maximum effect (E_{max}) model to express the inhibition fraction obtained by maraviroc at a given plasma concentration. It is represented by the following equation:

$$INH = \frac{I_{max} \cdot Conc^{\gamma}}{IC_{50\gamma} + Conc^{\gamma}} \quad (3)$$

where INH represents the viral replication inhibition fraction by maraviroc; I_{max} is the maximal inhibition, which is assumed to be equal to 1 (assuming no escape routes); Conc is either the predicted or the measured maraviroc plasma concentration that evolves in the function of time; IC_{50} is the estimated maraviroc plasma concentration that gives 50% inhibition of the infection rate; and γ is the sigmoidal factor. In the particular case of a drug that acts by blocking receptors, such as maraviroc, IC_{50} has an interpretation similar to that of the dissociation constant (K_D). For the purpose of the work described in this report, it is assumed that the inhibition (INH) is equal to the functional occupancy of the CCR5 receptor by the antagonist. No individual variability was taken into account in the PD model parameters, because the uncertainty around clinical interpretation of IC_{50} had a bigger impact than the actual variability associated with the PD parameter estimation (which is discussed later).

Interpretation of IC_{50}/EC_{50}. The extrapolation from inhibitory concentration (IC) to efficacious concentration (EC) is associated with caveats.[9] In the in vitro experiment, the IC_{50} and the IC_{90} (drug concentrations that give 50% and 90%, respectively, of the inhibition of the viral replication) have been estimated to be 1.04 and 5.74 ng/mL, respectively (Table II). Actually, if the IC_{90} is interpreted mechanistically, it is defined as EC_{90}, the concentration of the compound that brings the R_0 in the test tube just below 1.[9] Depending on the R_0 in the test tube (unknown at this stage), the EC_{90} could be translated to different values of receptor inhibition

Observed C_{max} (ng/mL)			*Simulated C_{max} (ng/mL)*		
Minimum	*Geometric mean*	*Maximum*	*5th percentile*	*Geometric mean*	*95th percentile*
1.24	2.24	8.34	1.35	1.86	2.80
3.93	8.94	42.1	9.08	12.6	18.1
132	135	229	62.6	82.8	112
417	456	970	284	371	507
881	1449	2550	990	1333	1769
1980	2327	3960	1401	1856	2560

and, therefore, to different values of IC_{90}. For example, if the R_0 is equal to 2, an inhibition of 50% will bring the reproductive ratio to 1 and the viral load will not increase. In an in vitro test, this will be translated to an almost full inhibition of viral growth. In this case the measured EC_{90} will correspond to the IC_{50}. On the other hand, if the R_0 is equal to 10, an inhibition of 90% will bring the reproductive ratio to 1. In this case the measured EC_{90} will correspond to the IC_{90}. Table III gives the possible IC_{50} and IC_{90} values for different values of R_0 for an observed EC_{90} of 5.74 ng/mL. The IC_{90} could range between 5.74 and 103 ng/mL, corresponding to IC_{50} values of 0.64 and 11.5 ng/mL, respectively. This uncertainty linked to the interpretation of the EC_{90} was taken into account during simulations of dose-response curve.

PK model. Maraviroc plasma concentrations from 24 healthy male volunteers who received ascending single doses of maraviroc of 1, 3, 10, 30, 100, 300, 900, and 1200 mg, administered orally as a solution under fasting conditions, were available for the PK model. A 2-compartment model assuming first-order absorption was used to fit the concentration-time median data. A correction factor was also introduced in the model to describe the apparent increase in absorption rate with increasing dosage. A saturable first-pass metabolism was implemented to describe the more than dose-proportional increase in area under the plasma concentration curve (AUC/F) at low doses. For simulations, an arbitrary variability of 15%, expressed as coefficient of variation, on the PK parameters was used to cover the apparent variability observed in the data. The observed double absorption peak was not modeled. A posterior predictive check was performed on the PK model and is presented in Table IV. The model describes the data well.

Model assumptions. To implement the model in the maraviroc program, several assumptions were made: The maximum effect will be reached at the highest dose tested; the inhibition of the infection rate constant is equal to the functional receptor occupancy by the antagonist; the pharmacologic action is described by a "noncompetitive" antagonism, where the amount of virus does not play a significant role[1]; no escape routes from the antagonism of the CCR5 receptor will occur in the first 11 days of treatment; and there would be no resistance in naive patients.

No induction or inhibition was assumed in the PK model.

Simulation plan. The proposed phase IIa study was a placebo-controlled 10-day monotherapy study of maraviroc in HIV-1–infected patients naive to therapy or not receiving therapy for at least 8 weeks. The main objective of the modeling and simulation project was to determine the dose or dose range of maraviroc that would allow the characterization of a dose-response curve in a limited number of patients with reasonable precision. The metric on which the treatment activity/efficacy could be evaluated was assessed through simulations (results not shown) and was defined as the difference between the $\log_{10}$ HIV-1 RNA observed at baseline and that observed at day 6. For phase IIb/III dose selection, the ED_{90} was defined as the dose that gives 90% of the maximum mean effect (on viral load) in a population. The PK-PD disease model was implemented in the Trial Simulator computer program and was used to simulate outcomes.[10]

The main goals of the simulations were as follows:

1. To identify and explore the impact of critical factors and model uncertainties on the predicted dose-response relationship of viral load on day 6 of a 10-day treatment with maraviroc given as monotherapy.
2. To evaluate/optimize the design of the monotherapy study in terms of trial strategy, number of treatment arms, and number of patients per treatment arm and to select a dose range to be studied to

describe the dose-response relationship and the estimation of the ED_{90} with reasonable precision.
3. To interpret the value of the predictions made in light of emerging data by comparing measured and simulated results.

When the first goal was considered, the main uncertainty resided in the measured in vitro IC_{50} (Table III), and it was estimated to range between 0.64 and 5.74 ng/mL. Therefore we simulated 1000 patients per treatment arm for 3 different scenarios, each scenario using different values for IC_{50}, as follows: low (0.64 ng/mL), medium (1.44 ng/mL), and high (5.74 ng/mL). These IC_{50} values were arbitrarily selected. The investigated doses were 12.5, 25, 50, 100, 200, 400, and 600 mg twice daily. Maraviroc was administered for 10 days and HIV-1 RNA was measured daily for 16 days after the start of treatment, as defined in the study protocol. The difference in $\log_{10}$ of HIV-1 RNA between the baseline and day 6 was derived for each simulated patient. Then, for each scenario, the viral load decline per dose group was analyzed with an E_{max} model by use of the nonlinear least squares function in S-PLUS.[11]

Regarding the second aim of the simulation plan, the precise estimate of the dose-response curve was achieved by determining the appropriate dose range of maraviroc that allows the characterization of the dose-response curve and the estimation of the ED_{90} with reasonable precision. The performance of each trial scenario was evaluated on the basis of the bias and precision of the estimated ED_{90} dose. The bias of the ED_{90} estimate was computed as the median (50th percentile) of the ratio of the dose derived from simulated results and the "true" dose used in the model to achieve ED_{90}. The precision of the ED_{90} estimate was determined by the 10th and 90th percentiles of the distribution of the ED_{90} from the simulation results. The characteristics of the HIV drug-naive patient population (eg, distribution of R_0) in the simulated trial were similar to those from the monotherapy study performed with another in-house antiretroviral investigational drug. For each proposed trial design, 200 replicates were simulated. Several trial designs were simulated to evaluate the impact of the number of treatment arms, dose range, number of patients per arm, and model uncertainties, such as the IC_{50} value, on the dose-response curve. Two different trial strategies, parallel and sequential, were also tested as part of the trial design optimization. The sequential design proposed here refers to updating an estimated dose response on the basis of the results from a previous cohort. The updated dose-response curve is then used to determine the dose and number of patients to be tested in the next cohort. Table V shows the dosage options tested during this analysis. Dosage options 1 to 4 were tested in a parallel trial design, whereas options 5 to 8 were tested under a sequential trial design. Doses for dosage options to be tested in a sequential design were selected to cover a range of IC_{50} values. For each dosage option presented in Table V, 3 different scenarios were simulated with 6, 8, and 10 patients assumed per treatment arm. On the basis of the safety and tolerability data from the ascending single-dose study, it appeared that 1000 mg twice daily was a possible maximum dose that could be used in a monotherapy study. The dose that would give a trough concentration corresponding to the in vitro IC_{90} of 5.74 ng/mL was 100 mg twice daily. Finally, the dose that would give a trough concentration corresponding to the in vitro IC_{50} of 1.44 ng/mL was around 30 mg twice daily.

Finally, regarding the third aim from the simulation plan, when new clinical data became available, these predictions were qualified, which allowed resolution of part of their uncertainty. Because the major uncertainty identified was around the IC_{50} values, a series of simulations were performed by use of various IC_{50} values. Predicted viral load–time profiles were compared visually with those observed in the study, and the best fit was selected.

Statistical methods. The HIV-1 RNA decline as a function of time (disease model) was fitted by use of a nonlinear mixed-effects approach as implemented in NONMEM, version V, computer software.[7] The PREDPP library subroutine ADVAN6 was used with the first-order estimation method. Maraviroc plasma concentrations were modeled by use of WinNonmix software.[12] Simulations were carried out in with Trial Simulator software, version 2.1.2.[10] The statistical package S-PLUS Professional edition was used for statistical analysis.[11] The simulated viral load data for maraviroc-treated subjects were analyzed by an E_{max} model by use of the nls function in S-PLUS.

RESULTS

Trial outcomes were simulated for various design strategies based on the PK-PD-disease model described in the Methods section. The simulations were used to evaluate the options proposed by colleagues on the maraviroc development team and to help evaluate whether a sequential design would enable the characterization of the dose-response relationship on the biomarker (viral load in plasma).

Table V. Scenarios of dose range used in simulations for study design derived from team options

Option 1 (mg, twice daily)	*Option 2 (mg, twice daily)*	*Option 3 (mg, twice daily)*	*Option 4 (mg, twice daily)*	*Option 5 (mg, twice daily)*	*Option 6 (mg, twice daily)*	*Option 7 (mg, twice daily)*	*Option 8 (mg, twice daily)*
50	25	50	25	500	500	500	500
150	100	100	75	100	100	100	100
300	300	200	200	50	50	200	200
600	600	400	400	—	25	—	400

To evaluate how the uncertainty regarding the IC_{50} was translated into the uncertainty on the ED_{90} estimate, dose-response curves were simulated under the scenarios for low, medium, and high IC_{50} values, as described in the simulation plan. The results of the simulated dose-response curves by use of the PK-PD disease model are presented in Table VI. Fig 3 represents the simulations of the dose-response curves for the scenarios tested. The results indicate that, on the basis of the knowledge at that time and the hypothesis implemented in the PK-PD disease model, the predicted "true" ED_{90} may lie between 30 and 125 mg. The results also indicate that if the true IC_{50} is 0.64 ng/mL a dose range of 25 to 500 mg twice daily (Fig 3, left panel) will only cover the top of the dose-response curve of the biomarker. Under this scenario, doses greater than 50 mg will not be differentiated because they will be on the flat part of the dose-response curve and it will be impossible, or very difficult, to accurately describe the dose-response curve. In contrast, if the "true" IC_{50} is around 5.74 ng/mL, doses of 100 mg or more need to be studied (Fig 3, right panel).

The second aim of the simulation plan was to evaluate and optimize several trial design options for the first monotherapy study. The uncertainties identified in aim 1 were incorporated in the simulations of aim 2. Simulations were performed as described in the simulation plan. The impact of the different trial attributes was assessed by calculating the bias and precision of the estimated ED_{90}. Table VII shows the results of the 10th, 50th, and 90th percentiles for the distribution of the ratio of the estimated and "true" ED_{90} of maraviroc for an IC_{50} value of 1.44 ng/mL (corresponding to an ED_{90} of 49.9 mg) and for dosage options 2 and 4. For all of the dosage options that were investigated, the effect was mainly located at the top of the dose-response curve. This observation, together with a high intersubject variability, makes the estimation of the dose that gives 50% of the maximum mean effect (on viral load) in a population (ED_{50})/ED_{90} difficult. The sigmoid model led to some unsuccessful runs in all scenarios. When the slope was fixed to 1.65, most runs were successful except in scenarios with the low IC_{50} and the lowest investigated dose equal to 50 mg. For a parallel design, options 2 and 4 with the lowest dose equal to 25 mg gave less bias and a better precision of the ED_{90} estimates (Fig 4).

Table VI. Estimated "true" ED_{50} and ED_{90} from 1000 simulated patients per arm by use of 3 different scenarios of IC_{50} values (low, medium, and high) and dose range from 12.5 to 600 mg twice daily

		Difference in log(baseline) − log(day 6)		
	Units	*Low*	*Medium*	*High*
IC_{50}	ng/mL	0.64	1.44	5.74
IC_{90}	ng/mL	5.74	12.9	51.7
ED_{50}	mg	8.78	13.9	31.0
E_{max}	copies/mL	1.33	1.33	1.30
$Hill_{ED}$		1.75	1.72	1.57
ED_{90}	mg	30.8	49.9	126

ED_{50} and ED_{90}, Doses that give 50% and 90%, respectively, of the maximum mean effect (on viral load) in a population; E_{max}, maximum effect.

Finally, when data on viral load–time profiles in the presence of a CCR5-receptor antagonist from the monotherapy study were available, these predictions were qualified, which allowed the resolution of part of their uncertainty.[13] Predicted viral load–time profiles were compared visually with those observed in the study, and the best fit was selected. Fig 5 shows that the best fit was obtained with an EC_{50} around 5.74 ng/mL, which is at the upper limit of the anticipated range of in vivo IC_{50} values based on the in vitro viral replication inhibition test.

DISCUSSION

Maraviroc is a CCR5-receptor antagonist in phase III development for the treatment of HIV-1 infection. Little information was available regarding the effect of

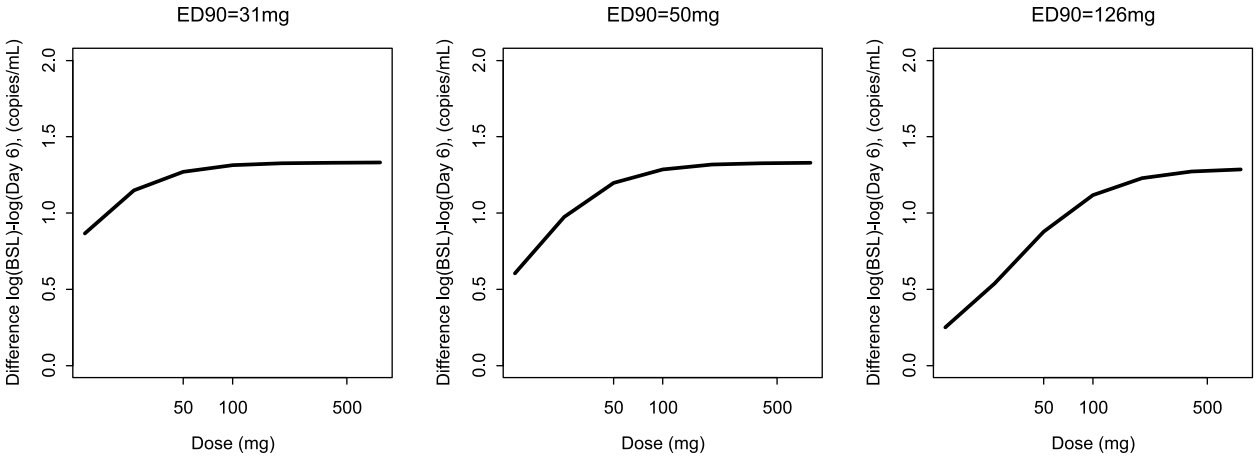

Fig 3. Results of simulation of dose-response curves by use of different scenarios of the drug concentration that gives 50% of the inhibition of the viral replication (IC_{50}), dose range from 12.5 to 600 mg twice daily, and 1000 patients per dose. *Line* represents fit obtained with a sigmoid E_{max} model. *Left panel*, IC_{50} equals 0.64 ng/mL (dose that gives 90% of the maximum mean effect [on viral load] in a population [ED_{90}] = 31 mg); *middle panel*, IC_{50} equals 1.44 ng/mL (ED_{90} = 50 mg); *right panel*, IC_{50} equals 5.74 ng/mL (ED_{90} = 126 mg).

Table VII. Bias and precision of simulated results for viral load decline given by difference of log(baseline) − log(day 6) for dosing options 2 and 4 (presented in Table V) and "true" ED_{50} of 50 mg twice daily and with 6, 8, and 10 patients assumed per arm

	Option 2					*Option 4*				
No. of patients per arm	*Percentile of estimated ED_{90} (mg)*			*Bias*	*Precision*	*Percentile of estimated ED_{90} (mg)*			*Bias*	*Precision*
	10th	*50th*	*90th*	*50%/"true"*	*90%/10%*	*10th*	*50th*	*90th*	*50%/"true"*	*90%/10%*
6	30	49	68	0.97	2.25	33	50	67	1	2.05
8	35	50	64	1.01	1.83	36	49	67	0.98	1.85
10	35	51	63	1.02	1.70	35	49	61	0.97	1.72

CCR5 antagonists in HIV-positive patients on initiation of the clinical program. However, a wealth of clinical information was available on anti-HIV drugs in the literature and was likely to be of help in the early clinical development of this new drug. PK-PD-disease modeling was believed to be an efficient way to integrate this literature information with the preclinical data on the compound. This is the reason why a modeling and simulation program was initiated at a very early stage during the preclinical development phase of the drug.

At an early stage of development, the main aim of the modeling and simulation project was to assist the development team in optimizing a proof-of-principle trial design. At this stage of development of a compound with a new mechanism of action and with limited PK information, it was decided to select a disease mechanistic model to incorporate information from several sources. Mechanistic models have several features that make them very attractive for this project. They allow the integration of information from different sources, such as the literature, other investigational compounds, and in vitro data. They can be used to propagate knowledge across compounds and, therefore, can be very useful to define a development strategy for follow-on compounds. In addition, they can predict outside the environment where they were developed so that they can also be used for late development phases of the compound. The models described in the literature for other antiretroviral drugs with different mechanisms

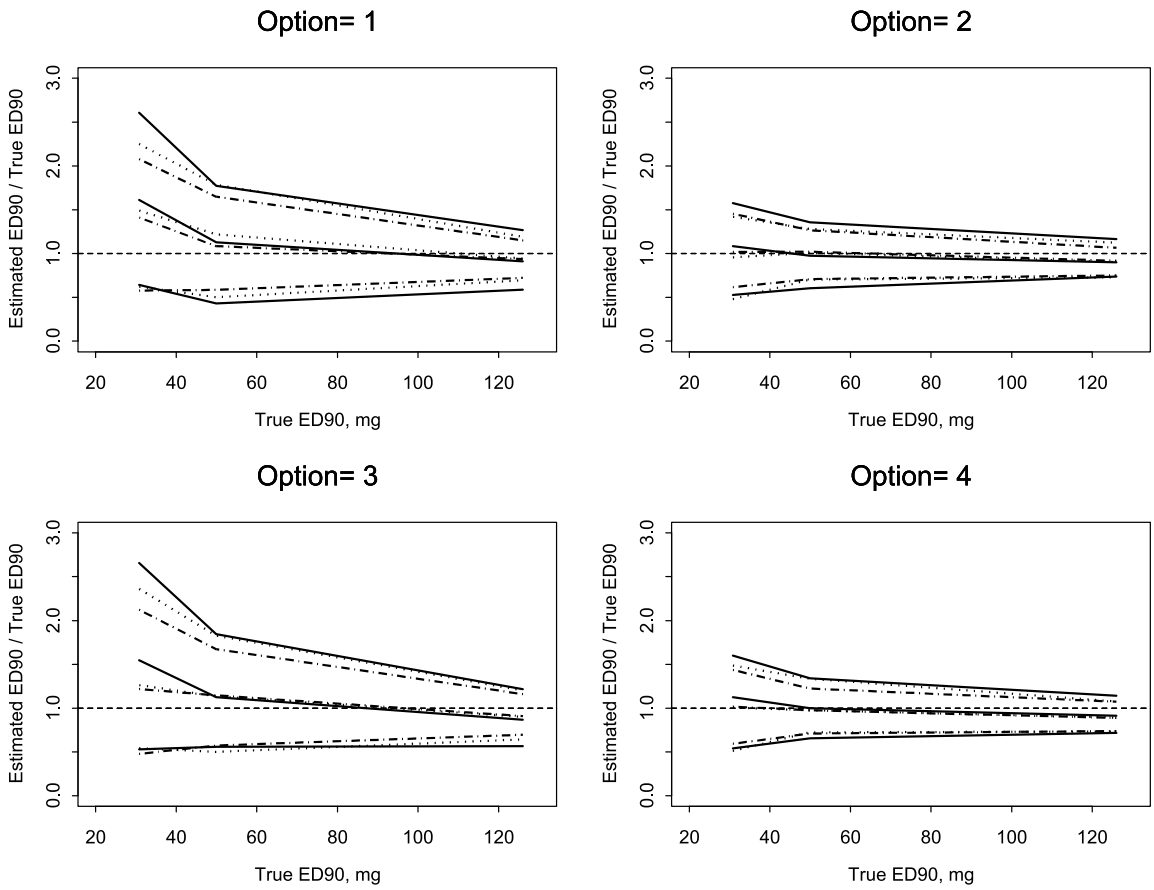

Fig 4. Bias and precision of estimated ED_{90} divided by "true" ED_{90} in function of "true" ED_{90} for viral load difference log(BSL) − log(day 6). *Solid lines* represent 6 patients per arm, *dotted lines* represent 8 patients per arm, and *broken lines* represent 10 patients per arm. Options as defined in Table V.

of action could easily be adapted for a new mechanism such as CCR5 antagonism and could easily be updated as new data become available. Finally, mechanistic models can give insight into the biologic characteristics of cell dynamics.

First, the PK-PD disease model approach was used to identify potential sources of uncertainties and critical factors that could have an impact on the estimation of the dose-response relationship of the viral load decline. Among these factors, the extrapolation of EC_{50} or EC_{90} values from in vitro to in vivo appeared to be the major source of uncertainty as a result of the fact that the reproductive ratio in the in vitro tests was unknown. Without knowledge of this reproductive ratio, an in vitro EC_{90} can correspond to a wide range of IC_{90} values (Table III). Using the mechanistic PK-PD-disease model and running simulations across IC_{90} uncertainties, we were able to quantitatively evaluate their impact on the predicted ED_{90}. The range of predicted ED_{90} values was between 30 and 125 mg. This uncertainty in the predictions had an important impact not only on the dose range to be tested in the study but also on the study design strategy itself.

Indeed, given the identified uncertainty, a parallel design, as initially suggested by the development team, would have required a larger number of doses to cover for the identified uncertainty, resulting in a costly and lengthy trial. A sequential trial design was considered to be a better option to protect against this uncertainty and avoid having to perform a larger trial. The sequential trial design allowed us to update the estimated dose-response curve with information from the first cohort before deciding about the dose and number of

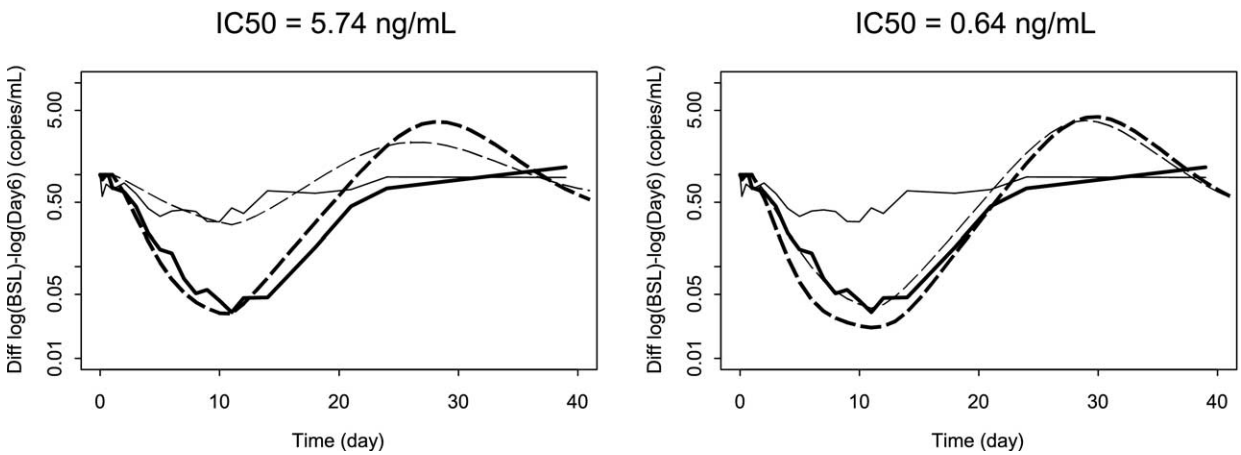

Fig 5. Simulated and observed viral load–time profiles for the 2 dosage regimens. The PD model based on the in vitro viral replication inhibition was used in the simulation. *Solid lines* represent mean measured viral load, and *dotted lines* represent simulated viral load. *Thick black lines* represent 100 mg twice daily, and *thin black lines* represent 25 mg every day.

patients to be studied in the next cohort.[14] The impact of uncertainties on the dose-response curve was reassessed through simulations each time that the model was updated with new data. Moving in a stepwise manner was then continued until the actual dose-response curve was characterized. To move faster, we suggested starting the sequential design with the highest possible dose to test whether the drug worked. Then we proposed to target the ED_{50} instead of decreasing step by step from the highest dose to accelerate the investigation of doses that were no longer at the E_{max} region of the dose-response curve and obtain an indirect estimation of the possible efficacious dose (ie, $\geq ED_{90}$).

This model-based approach was used as a tool to communicate with the development team, making the risks and assumptions clearer on several occasions during the clinical trial optimization. For example, given the knowledge from the model at that time, it would be more effective to increase the range/number of investigated doses than to increase the number of subjects per dose. The impact of increasing the numbers of patients per treatment arm is shown in quantitative terms in Fig 4.

A modeling approach was used to aid interpretation of the clinical results and to give insights into complex virus-cell interaction systems. It was also used to help to estimate the dose–viral inhibition relationship. One of the goals of the simulations was to help with the interpretation of the new data.[13] When clinical data became available, the model was updated and used to make predictions for other studies. We have put the new clinical results into the context of the preclinical uncertainties identified by the model. The simulation performed with the mechanistic PK-PD-disease model allowed the estimation of the "true" in vivo EC_{50} of around 5.74 ng/mL (EC_{90} of around 60 ng/mL).

Model-based decision making was successfully incorporated into the development program of maraviroc. Modeling and simulation were used to interpret in vitro data and as a tool to inform team discussions and decisions around risks and limitations. We were able to characterize the dose-response curve of viral load decline of a new class of HIV drug much earlier than expected and with fewer patients. Incorporating the modeling approach early in the development of the compound allowed us to proceed with confidence to phase III studies.

By making use of the available PK-PD-disease model, the possible range of active oral doses for maraviroc in HIV-positive patients was estimated by simulation before any clinical trials took place.

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Drs Rosario, Dorr, van der Ryst, and Hitchcock are full-time employees of Pfizer, and Dr Jacqmin is a full-time employee of Exprimo and was contracted to complete the analysis presented herein, and he was an employee of Pharsight (Mountain View, Calif) when he collaborated on this project.

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