STATISTICS IN MEDICINE
Statist. Med. 2003; **22**:833–846 (DOI: 10.1002/sim.1448)

Population pharmacokinetics/pharmacodynamics relationships of an anticancer drug

Cécile Ané and Didier Concordet*,†

UMR 181, Physiopathologie et Toxicologie Expérimentales, Ecole Vétérinaire de Toulouse, 23 chemin des Capelles, 31076 Toulouse Cedex, France

SUMMARY

This paper proposes a method for studying the toxicity of an anticancer drug with a delayed effect. The goal is to predict a dosage regimen with controlled toxicity. To this end, a semi-physiological model is used. A limit of toxicity is demonstrated, which is intrinsic to the model. It reduces the effect of high drug concentrations. This limit explains the mixed behaviour of the drug: time-dependence and concentration-dependence, according to the dose actually administered. A population analysis is performed to estimate the parameters of the model, and to predict a safe dosage regimen. Copyright © 2003 John Wiley & Sons, Ltd.

KEY WORDS: pharmacokinetics; pharmacodynamics; non-linear mixed effects models; toxicity

1. INTRODUCTION

A primary goal when using an anticancer drug is to control its toxicity on patients. Since these drugs usually have a narrow therapeutic index, the dose has to be adjusted carefully to find an admissible regimen which is effective and yet not too toxic. Topotecan is an anticancer drug given by IV infusion to women with ovarian cancer, as second-line therapy (see reference [1] for instance). The major toxic effect of topotecan is a decrease in neutrophil counts that occurs 8 to 15 days after drug administration (Figure 1). A primary index used to measure this is the time the neutrophil count remains below the fixed limit: 500 PN/mm^3. Another index is the minimum neutrophil count reached.

The aim of this paper is first to propose a semi-physiologic explanation of the toxicity observed in women. Then, using this model, a second aim is to predict the occurrence and magnitude of the toxicity in patients. It has been shown in mice [2] that this toxicity varies not only with the dose given, but also importantly with the duration of the exposure to the drug. Thus, when the total dose is given in a single infusion, a low toxicity is observed, but

* Correspondence to: Didier Concordet, UMR 181, Physiopathologie et Toxicologie Expérimentales, Ecole Vétérinaire de Toulouse, 23 chemin des Capelles, 31076 Toulouse Cedex, France.
† E-mail: d.concordet@envt.fr

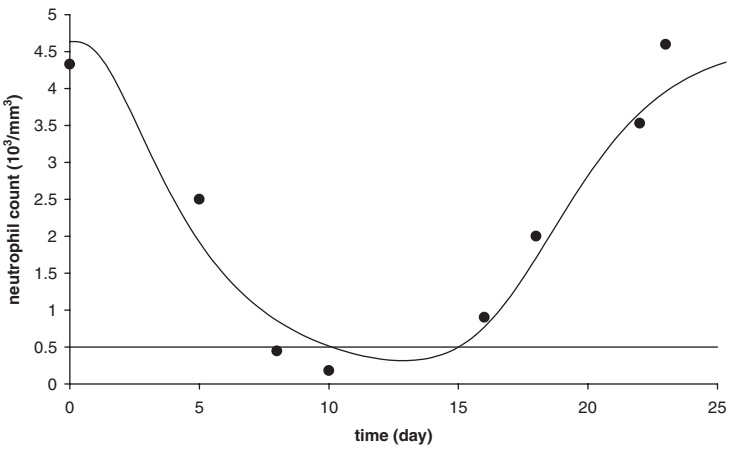

Figure 1. An example of observed and fitted neutrophil profiles. The horizontal line, at 0.5×10^3 PN/mm^3, allows one to define the primary index of toxicity (time spent below).

when the same total dose is fractionated over 5 days, toxicity is high. Surprisingly, when it is fractionated over 20 days or more, toxicity is reduced.

There are many PK/PD models that directly link the effect to the exposure. However, as has been already described for mice, a single measure of exposure cannot directly explain the toxicity. These kinds of models (direct PK/PD models) are too simple to describe properly the toxicity in mice and in women. It is thus natural to look for another family of models that is rich enough to explain for different toxicities with the same total dose. Among possible choices, the indirect models are probably most often used. There is a huge literature on the subject. Among the authors, Jusko [3, 4] described the properties of a large number of models, and Minami [5] and Zamboni [6] proposed use of these models. These models have two interesting properties in relation to our study. First, they allow one to describe the whole time course of the neutrophil counts. Secondly, there is an intrinsic delay between drug administration and the effect. However, the main drawback of these models for topotecan is that they do not give an accurate description of the drug action. Thus, we propose using another family of PK/PD models that has already been used by Karlsson [7, 8] for anticancer drugs. These semi-physiologic models macroscopically mimic the action of topotecan on cells. A careful analysis of the properties of these models enables one to explain the phenomenon observed in mice. They also give a PK/PD parameter that is easy to interpret and can be useful when planning the dosage regimen. To prevent toxicity on a fixed percentage of women, it is suitable to describe the interpatient variability. Population analysis (non-linear mixed effects models) is a natural way to reach this goal.

The remainder of the paper is organized as follows. Section 2 is devoted to the kinetics of topotecan. Section 3 deals with the pharmacodynamic (PD) model: its choice and description are followed by the study of the property which explains the curious toxicity pattern in mice. The population pharmacodynamic model is then estimated. Finally, in Section 4, simulations are carried out to predict admissible dosage regimens.

2. THE PHARMACOKINETIC MODEL

Topotecan was administrated by 30-minute IV infusions to $N = 42$ women, on five consecutive days. For each patient, the three first daily doses were fixed *a priori*, and the two last were adjusted to reach a total AUC within a targeted range (37 500–75 000 nM min). Figure 2 gives an example of a concentration profile in one patient.

The PK data were analysed [9] on a subsample of $N_1 = 31$ women. Let us recall their results briefly. Total topotecan plasma levels were analysed according to a two-compartment model with linear elimination from the central compartment. The resulting individual parameters were the clearance Cl of the central compartment, the volume V_c of the central compartment, the volume V_p of the peripheral compartment and the clearance Q between compartments. This clearance Q was assumed to be fixed in the population, and estimated by $\hat{Q} = 46.6$ l/h. The individual PK parameters $\Phi_i = (\ln Cl_i, \ln V_{ci}, \ln V_{pi})$ were assumed to be independent identically distributed (i.i.d.), drawn from a Gaussian $N(\mu, \Omega)$ distribution, and the covariance matrix Ω was assumed to be diagonal. The estimation of (μ, Ω) obtained in [9] from the subsample of size 31 is given in Table I.

Finally, the individual parameters Φ_i were predicted by the Bayesian estimates (maximum *a posteriori*), for all 42 patients. There was good agreement between model-predicted and observed concentrations for each patient.

In the rest of this paper, the kinetic profiles are considered as known and fixed.

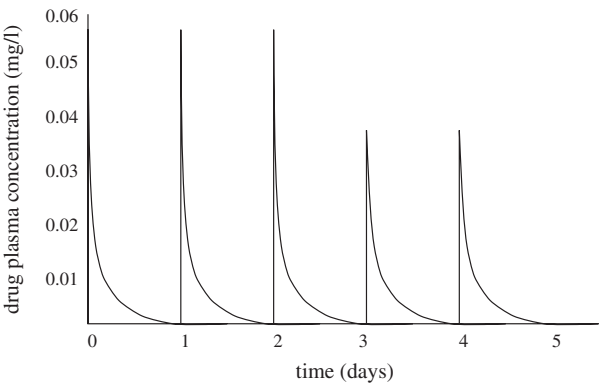

Figure 2. An example of kinetics.

Table I. PK parameter estimates. Clearance Cl is expressed in l/h and volumes V_c, V_p in l.

	$\ln Cl$	$\ln V_c$	$\ln V_p$
$\hat{\mu}$	2.99	3.66	3.35
$\sqrt{\hat{\Omega}}$	0.42	0.55	0.38

3. THE PHARMACODYNAMIC MODEL

The discussion concerning the pharmacodynamic model is divided into three parts. First, a physiologically based structural model is chosen. Then we consider the properties and choice of the function describing the drug action on cells. Finally, a non-linear mixed effect model is set up, and its parameters are estimated.

3.1. The choice of the structural model

Topotecan is a drug that acts during the replication of DNA. It binds to topoisomeras I when this enzyme is unwinding DNA. At this stage, replication is stopped, DNA is broken, and the cell dies. Thus, each dividing cell is likely to be killed when topotecan is present. From a macroscopic point of view, the bone marrow produces progenitor stem cells that divide rapidly, and so can be killed. If these cells survive, they continue to mature without obstacle in the bone marrow. Finally, they migrate into blood, the observed pool, as white blood cells.

When no drug is given, this system is at equilibrium, and can be described using the family of compartmental models given in Figure 3.

There is a large choice for the number of sensitive compartments, the number of non-sensitive compartments, and the type of exchanges between compartments. Actually, when all the compartments in the same region (sensitive or non-sensitive) share the same parameters (rates of exchange), the different models that could be considered by changing the number of compartments or their order of exchanges are not nested. Therefore, no criterion of choice of model (except the BIC criterion in a Bayesian framework) can be used.

Bone marrow constitutes a non-observed part of the life of these cells. In order to get information about this part, a large number of different outputs (neutrophil profiles), obtained with a large number of different inputs (kinetics) are necessary. We have at our disposal roughly a single shape of PK profile. Remember that all the women received five consecutive daily infusions, and that the two last doses were adjusted so as to reach a target AUC. Consequently, the data are not rich enough to allow a precise description of the actual model. Therefore, we deliberately chose to use a model built on numerous data for 5-fluorouracil in rats [8]. As this drug acts at the same stage as topotecan, it appears reasonable to take the structure of the rat model, and to scale it to the human, although will see that slight modifications are necessary.

The model used in [8], represented in Figure 4, contains five compartments. Two compartments are sensitive to the drug, two are non-sensitive, and the last compartment is the blood pool. The exchanges are second-order exchanges, except for what leaves the blood, which is

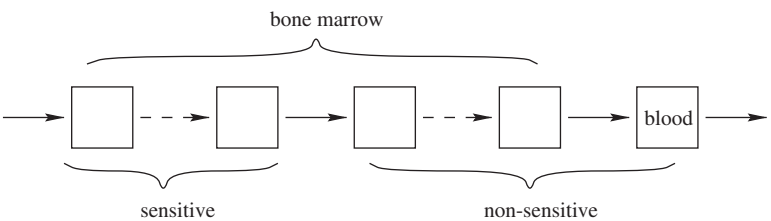

Figure 3. General compartmental model.

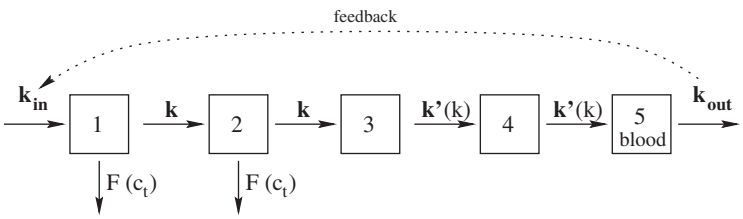

Figure 4. Model given in reference [8] and final structural model. Bold characters stand for parameters of our final model (k and k'), whereas the characters in brackets are for the model in [8] (k only). The dotted line represents the feedback of the original model.

of order 1. All bone marrow compartments share the same second-order constant, k. Recall that when the exchanges are of order 2, the behaviour of a given cell depends on the size of the compartment. The more cells in this compartment, the more rapidly this particular cell will move. This type of exchange roughly mimics the birth of new cells in the compartment, and describes adequately the observed neutrophil profiles, which quickly leave and come back to baseline.

In the original model [8] the production rate is assumed to be constant, up to a feedback mechanism. More precisely, if N_{base} is the baseline neutrophil count, and $N(t)$ the neutrophil count at time t, then the production rate is set to $k_{\text{in}}N_{\text{base}}/N(t)$. Finally, the action of the drug is modelled by a first-order killing rate on each of the two sensitive compartments. This killing rate is assumed to be linked directly to the drug concentration c_t at time t: it is set to $F(c_t) = k_e c_t$.

Applying this model to data on women, the individual curve fitting was not satisfactory for two reasons. The feedback leads to a rebound, which is pronounced in rats. As it is missing in human patients, this feedback mechanism is inappropriate for our study. Without feedback, the model used in reference [8] appears to be too constrained to allow a rapid decrease or increase of the curve. In this model, cells have to spend the same time in the two different states (sensitive/non-sensitive). For this reason, we chose two different rates of exchange (k and k') as shown in Figure 4. The mean residence time of a cell in the non-sensitive region is then approximately proportional to the number of non-sensitive compartments, two, times k'^{-1}. Similarly, the mean residence time of a cell in the sensitive region is proportional to $2k^{-1}$. In other respects, we notice that increasing the number of non-sensitive compartments does not determine another dynamic behaviour, since an increase in k' can compensate. The same argument applies to the number of sensitive compartments and the constant k.

In summary, there are two differences between the previous model in reference [8] and ours: feedback is discarded and two rates of exchange are used instead of one. The differential equations driving the selected model are now detailed. The neutrophil profile in blood is the solution $X_5(t)$, $t \geqslant 0$, of the system

$$\begin{cases} \partial X_1/\partial t = k_{\text{in}} - kX_1^2 - F(c_t)X_1 \\ \partial X_2/\partial t = kX_1^2 - kX_2^2 - F(c_t)X_2 \\ \partial X_3/\partial t = kX_2^2 - k'X_3^2 \\ \partial X_4/\partial t = k'X_3^2 - k'X_4^2 \\ \partial X_5/\partial t = k'X_4^2 - k_{\text{out}}X_5 \end{cases}$$

starting, when $t=0$, at equilibrium, that is, $X_1(0)=X_2(0)=\sqrt{(k_{in}/k)}$, $X_3(0)=X_4(0)=\sqrt{(k_{in}/k')}$ and $X_5(0)=k_{in}/k_{out}$. In the following, the neutrophil profile $X_5(t)$ is also denoted by $N(t)$ for convenience, or $N(F)(t)$ to emphasize the dependence on F. In the final model, $F(c_t)$ is set to $k_e c_t$. The following section deals with the rationale of this choice.

3.2. Drug action: the PK/PD link

The choice of the drug action, modelled by the killing function F, has not been yet discussed. Here, we first give the properties of F, depending on its shape and filtered by the model. We derive then two consequences: the choice F linear in c_t, and a qualitative explanation of the strange toxic behaviour observed in mice in reference [2]. The killing function $F(c)$ has some obvious properties: it cancels at $c=0$ (no drug, no action), and it is increasing (more drug, greater effect). Intuitively, the shape of F determines the drug action: when F is convex, low concentrations give little toxicity, but when F is concave, low concentrations rapidly give toxicity. Consequently, for a fixed total dose, it seems that when F is convex a low target toxicity can be reached with a large number of small doses, whereas when F is concave the same target toxicity will be reached with a small number of high doses. Thus, the shape of F seems to be of major importance, especially in the context of this paper.

Actually, the impact of the shape of F is reduced by the existence of a limit of toxicity. This limit is intrinsic to this family of catenary models. It is reached when the sensitive compartments are emptied (by the drug action). In that case, whatever the killing rate, the drug cannot kill more cells than those arriving in these compartments. Even if the killing rate $F(c_t)$ is very high, its effect on the system is very similar to the one that would be obtained with a smaller killing rate. Two different mechanisms drive the toxicity: when $F(c_t)$ is low (below the limit of toxicity), the shape of F determines the toxicity; when $F(c_t)$ is high, only the time it spends above the limit of toxicity governs the effect. These intuitive considerations need to be formalized in order to quantify the maximal effects.

To this end, let Δ be a fixed length of time and $0<\varepsilon<1$.

Proposition 1
Let $T=1/\sqrt{(k_{in}k)}$ (respectively $T'=1/\sqrt{(k_{in}k')}$) be the mean residence time in the sensitive compartments (respectively non-sensitive compartments) at steady state. Let $N_b=k_{in}/k_{out}$ be the neutrophil count at steady state (neutrophil count at baseline). Set

$$K_0=\max\left\{\frac{1}{\Delta}\ln\left(\frac{20}{\varepsilon}\sqrt{(k'/k)}\right);\frac{1}{\varepsilon\min\{T,T'\}}\frac{4}{3}\left(6+4\frac{\Delta}{T'}\right)\right\}$$

For all killing rate functions $F(c_t)$, the following property is true: if $F(c_t)\geqslant K_0$ on an interval of length Δ, say $[t_0,t_0+\Delta]$, then decreasing F to K_0 on the interval $[t_0,t_0+\Delta]$ does not change the effect more than εN_b. More precisely

$$\sup_{t\geqslant 0}|N(F)(t)-N(F^{(K_0)})(t)|\leqslant\varepsilon N_b$$

where $F^{(K_0)}=F$ outside $[t_0,t_0+\Delta]$, and $F^{(K_0)}=K_0$ on $[t_0,t_0+\Delta]$.

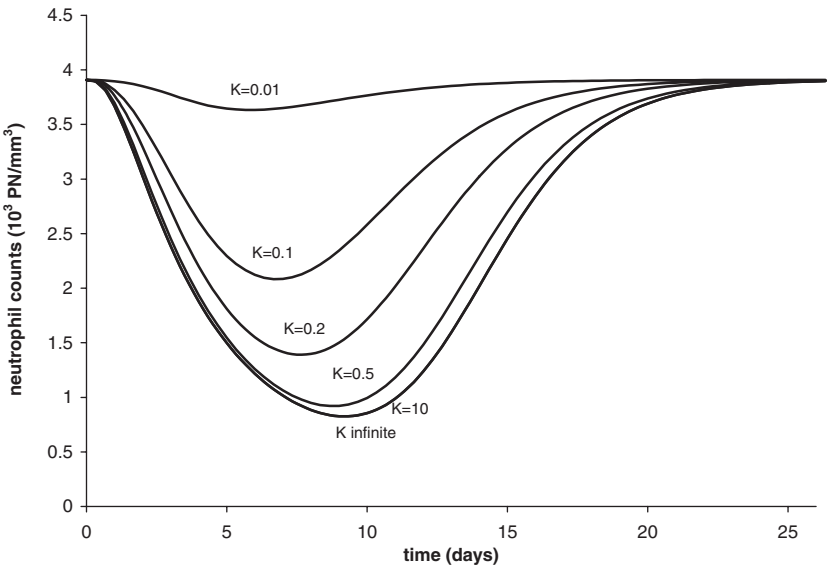

Figure 5. Simulation of the neutrophil count profile $N(F^{(K)})$ for different values of K (per hour) and c_0 set at 0.02 mg/l. When K increases, the curve of $N(F^{(K)})$ tends to the curve of $N(F^{(\infty)})$.

The proof of this result is deliberately omitted, since it is too long and without practical interest. Notice that K_0, as previously defined, does not depend on F. The main consequence of this proposition is that all shapes of F above K_0 give nearly the same toxicity. A simple way to explain this property is to consider the following example. Let us take the first patient of this study. Assume that her kinetics $(c_t)_{t\geqslant 0}$, as well as her structural parameters $(k_{\text{in}}, k, k', k_{\text{out}})$ are known. Assume now that the killing function F is a step function

$$F^{(K)}(c)=\begin{cases} 0 & \text{if } c<c_0 \\ K & \text{if } c\geqslant c_0 \end{cases}$$

and that c_0 is known. Since $(c_t)_{t\geqslant 0}$ and c_0 are fixed, the lapse of time Δ during which $c_t \geqslant c_0$ is fixed (for example, $\Delta = 5 \times 1$ hours). The proposition says just that for all $\varepsilon < 1$, there exists K_0 such that, for all $K \geqslant K_0$, $\|N(F^{(K)}) - N(F^{(\infty)})\|_\infty \leqslant \varepsilon N_{\text{b}}$. This last inequality means, first, that toxicity is limited by the one given by $F^{(\infty)}$. Secondly, it implies that when K becomes large, the shape of the neutrophil curve becomes constant. Figure 5 shows the simulation of neutrophil count profiles $N(F^{(K)})$ for several values of K.

We now give the first consequence of Proposition 1, namely an estimability problem and the resulting choice of F. Even if the map $K \mapsto N(F^{(K)})$ remains injective, its derivative tends to zero (uniformly on time) when K is large. Thus, if the actual K of the considered individual is large (more than K_0), the Fisher information matrix is nearly degenerate, which implies that the maximum likelihood estimator of such a K has too a large variance to be useful in practice. For instance, let us consider the following model:

$$Y_j = N(F^K)(t_j) + \sigma\varepsilon_j$$

where Y_j is the observed neutrophil count at time t_j, and ε_j are i.i.d. $N(0,1)$. Then, the asymptotic variance of the ML estimator of K is proportional to

$$\left[\sum_j \left(\frac{\partial N(F^K)(t_j)}{\partial K}\right)^2\right]^{-1}$$

and tends to infinity when K tends to infinity. This example illustrates a general property implied by Proposition 1: whatever the shape of F above K_0, it cannot be properly estimated with a reasonable variance. Thus, whatever the chosen parameterization for F, there exists an area of this parameter space where the estimation is difficult. We mention, however, that in this particular model, if the kinetics is known, as well as k_{in}, k, k' and k_{out}, then K is identifiable. Indeed, the function $K \to N(F^K)(t_1)$ is strictly decreasing, and injective (as soon as the kinetics c_t crossed c_0 before t_1).

The estimation quality (variance of the estimator) of the parameters governing the shape of F below K_0 depends on the quantity of information available below this limit. If this information is poor, a large variance of the estimator is expected whatever the parameterization of F. In such a case, the simplest shape for F is to be preferred, that is, a linear shape. As already mentioned, all women of the study have very similar kinetic profiles, so that the information of the data only concerns a narrow range of concentrations. We are thus confronted with two possibilities: either the drug concentrations lead to killing rates above K_0, and then we have no information about the shape of F for small concentrations, or the drug concentrations lead to small killing rates, but since the range of these concentrations is narrow, the shape of F can be documented for only a small interval of concentrations. In both cases, the linear killing rate $F(c) = k_e c$ has to be chosen.

The second consequence drawn from Proposition 1 is a qualitative explanation of the toxic behaviour observed in mice [2]. Recall that with a single dose the observed toxicity is low; since concentrations reach high values, they lead to high killing rates (above K_0), but only for a short time. When this dose is fractionated over 5 days, concentrations are lower, but high enough to lead to killing rates above K_0. Since the total time spent by the killing rate above K_0 is long, toxicity is high. Finally, when the total dose is fractionated over 20 days, concentrations are too low to give high killing rates and high toxicity. Our model implies thus a high correlation between toxicity and the time spent by the killing rate above K_0. That is exactly, what is observed in reference [2]; a high correlation between toxicity and the time the drug concentration remains above 0.7μM.

3.3. The population PK/PD analysis

As can be seen on a patient-by-patient analysis, both the response curves and the individual PD parameter $\Psi = (k_{\text{in}}, k, k', k_e)$ vary widely. A natural way to capture and evaluate this variability is to use a non-linear mixed effects model. The PD analysis relies on the concentration time course. Recall that the individual PK profiles are known. We assume here, as have others [5, 6], that k_{out} does not vary among patients and is equal to 0.1 per hour.

Table II. PD population parameter estimates.

	$\ln k_{in}$	$\ln k$	$\ln k'$	$\ln k_e$
m	-1.02 (8.4×10^{-3})	-10.1 (0.14)	-7.92 (0.30)	1.55 (0.076)
$\sqrt{D}$	0.39 (0.98)	2.07 (0.82)	0.62 (0.23)	0.84 (0.37)
σ^2	0.370 (0.17)			

A family of non-linear mixed effects PD models can be described as

$$\begin{cases} Y_{ij} = N_i(t_{ij}; \Psi_i) + \sigma[N_i(t_{ij}; \Psi_i)]^{\alpha}\varepsilon_{ij} \\ \Psi_i = (k_{in}^i, k^i, k'^i, k_e^i) \\ \ln \Psi_i \sim_{\text{iid}} \mathrm{N}(m, D) \\ \varepsilon_{ij} \sim_{\text{iid}} \mathrm{N}(0, 1) \end{cases}$$

where Y_{ij} is the observed neutrophil count in the ith individual at time t_{ij} ($j \in [1 \ldots n_i]$) and Ψ_i is the unobserved vector of its PD parameters. $N_i(t, \Psi_i)$ denotes the neutrophil count at time t of the ith individual, whose PD parameter is Ψ_i, as described in Section 3.2. It is indexed by i to keep track of the dependence on the known kinetics in the women. The Ψ_i are assumed to be independent of the PK individual parameters Φ_i. Since all the individual parameters are positive, we assumed they are distributed according to a log-normal distribution with mean m and variance D. The population parameter to be estimated is $\theta = (\sigma^2, m, D)$.

With regard to the number of patients involve in the trial, we chose to take D as a diagonal matrix. In other words, we assumed that individual parameters k_{in}, k, k', k_e are mutually independent. The estimation method used is FOCE [10]. It provides an asymptotically Gaussian estimator $\hat{\theta} = (\widehat{\sigma^2}, \hat{m}, \hat{D})$ of θ whose asymptotic variance will be denoted by $V(\theta)$.

Table II gives the estimation of σ^2, m, $\sqrt{D}$, as well as their asymptotic standard error (in brackets). It turns out that the optimized criterion (FOCE) has a large number of local minima, reflecting a large distance from the asymptotic framework. Therefore the asymptotic variance-covariance matrix of the estimator should be interpreted with care.

An example of an individual fitted curve was given in Figure 1.

4. SIMULATIONS AND TOXICITY PREDICTIONS

This section is devoted to the second goal of the paper, namely, to give a whole set of dosage regimens with a controlled toxicity. Recall that the primary measure of toxicity is the time spent by the neutrophil counts below 500 PN/mm^3. When this time is longer than 7 days, toxicity is considered as intolerable. First, we give an index that allows one to decide qualitatively whether, for a given regimen, the toxic behaviour of the drug is rather concentration-dependent (toxicity is linked to the daily dose) or time-dependent (toxicity is linked to the time spent by the drug concentration above a limit). Finally, we give quantitative results that determine sets of dosage regimens with acceptable toxicity.

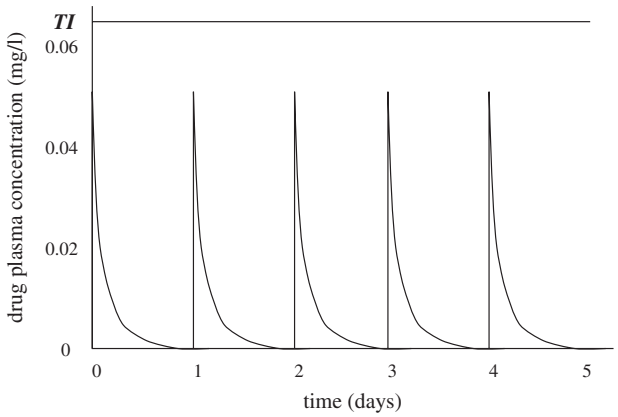

Figure 6. Toxicity index for a qualitative description of the drug toxic behavior.

4.1. Simulation of a PK toxicity index

The limit of toxicity K_0, defined in Proposition 1, gives information on exposure to the drug. It can be used as a surrogate that gives some useful complementary information on the toxic behaviour. Indeed, as previously seen, when drug concentrations are low, the drug toxicity is concentration-dependent, but as soon as the limit K_0 is reached, toxicity depends on the time spent above K_0, and is thus rather time-dependent.

Let us define a toxicity index (TI). For a chosen duration Δ and a chosen $\varepsilon > 0$ (these choices are discussed later), we set $\mathrm{TI} = K_0/k_e$. The main advantage of this index is that it is homogeneous to a drug plasma concentration and can be compared directly to the kinetic profile. Since both K_0 and k_e vary amongst patients, it is possible to simulate their distribution and then to derive the 5 per cent percentile of TI. In other respects, as has been shown [9], it is possible to use covariates such as creatinine clearance to predict for each patient the PK profile for a dosage regimen chosen *a priori*. An example of such an expected kinetic profile and the 5 per cent percentile of TI is presented in Figure 6.

This shows that the expected concentration remains below the 5 per cent quantile of TI. It means that the early sensitive compartments are not emptied by the action of the drug, with a probability of 95 per cent. This implies that the toxicity is concentration-dependent with this dosage regimen. It would not be so if the concentration time course had crossed the index TI for longer than Δ.

Let us detail the rationale for the choice of ε and Δ. As can be seen in Proposition 1, K_0 depends on maturation times T and T', whose values are about 260 and 90 hours. Therefore, K_0 is about $(120\varepsilon)^{-1}(6 + 0.044\Delta)$. Moreover, it is natural to set Δ below 24 hours, the delay between two consecutive infusions. Thus the influence of the term 0.044Δ is low compared to 6. We set $\Delta = 5\,\mathrm{h}$. It remains to choose ε, which is a proportion of the baseline neutrophil count. If the difference between two neutrophil profiles is of the same order as the critical threshold, then the resulting toxicities are similar. Recall that the critical threshold ($500\,\mathrm{PN/mm^3}$) is around 10 times less than the baseline neutrophil count. These considerations lead us to choose $\varepsilon = 0.05$ (5 per cent). Even if this graphical method does not tell us directly

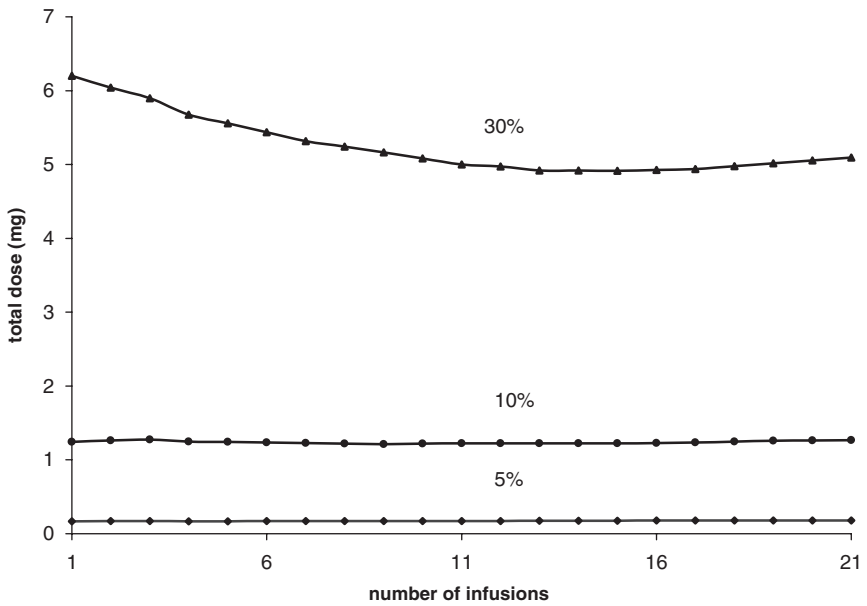

Figure 7. Curves giving the set of dosage regimens with the same fixed occurrence of toxicity (5, 10 and 30 per cent).

whether or not an intolerable toxicity is reached, it gives qualitative information about the toxic behaviour.

4.2. Simulation of admissible dosage regimens

The aim of this part is to predict the set of admissible dosage regimens. A dosage regimen is said to be admissible if an intolerable toxicity occurs for less than a fixed percentage of patients (say 5 per cent). We limited ourselves to dosage regimens with constant daily doses, one infusion each day, and proceeded as follows. For a fixed dosage regimen, that is with fixed number of infusions and fixed daily dose, we determined the percentage of patients with intolerable toxicity, using a simulation method detailed hereafter. Then, the dosage regimen was adjusted to obtain a percentage equal to 5 per cent. Now, let us detail the simulation method. First a sample $\theta_j^* = (\sigma_j^{2*}, m_j^*, D_j^*)$ of size 200 was drawn from a $\mathrm{N}(\hat{\theta}, V(\hat{\theta}))$. Next, for each θ_j^*, two samples $(\ln \Psi_{i,j}^*)_{i=1\ldots150}$ and $(\ln \Phi_i^*)_{i=1\ldots150}$ were drawn respectively from a $\mathrm{N}(m_j^*, D_j^*)$ and from the PK population distribution $\mathrm{N}(\hat{\mu}, \hat{\Omega})$ given in Section 2. Then, $Y_{ij}^*(t) = N_i^*(t, \Psi_{ij}^*)$ was computed, as well as the time $T_{ij}^* = \int_0^\infty 1_{\{Y_{ij}^*(t) \leqslant 500\ \mathrm{PN/mm^3}\}}(t)\,\mathrm{d}t$. The proportion of patients with intolerable toxicity was then estimated as the percentage $\frac{1}{200 \times 150} \sum_{i,j} 1_{\{T_{ij}^* \geqslant 7\ \mathrm{days}\}}$.

Figure 7 gives the critical daily dose obtained with these simulations, as a function of the number of infusions. This figure shows that the toxicity occurrence increases with the total dose. Moreover, total doses that give few toxicities do not depend on the number of infusions; it is about 0.17 mg for 5 per cent toxicity and about 1.22 mg for 10 per cent toxicity. In other

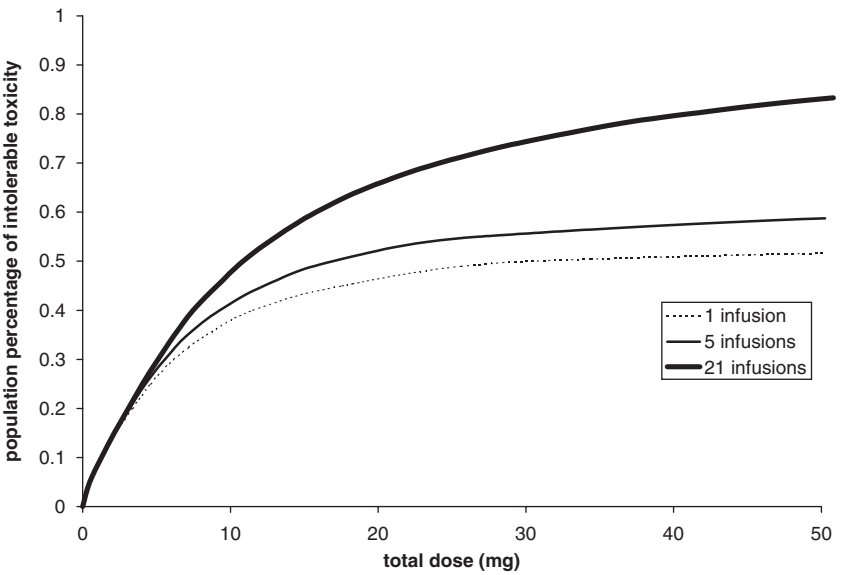

Figure 8. Cumulative distribution function of intolerable toxicity for the 1-, 5- and 21-day schedules.

words, if the target total dose is low, the number of infusions can be chosen as desired without any change of the toxicity occurrence. A striking fact shown in this figure is that the total admissible dose giving 30 per cent toxicity decreases from 6.2 mg (for one infusion) to 4.9 mg (for 15 infusions) and then it increases up to 5.1 mg for 21 infusions. Since 6.2 mg for one infusion produces the same toxic effect as 4.9 mg for 15 infusions, the one-day schedule can be considered as less toxic than the 15-day schedule (it takes more drug to produce this level of toxicity). Similarly, the 21-day schedule appears to be less toxic than the 15-day schedule. These assertions have been clinically evidenced (see reference [11] for instance). Figure 7 is a tool for comparing schedules in a rather qualitative way. Figure 8 gives the full quantitative description of toxic effects for three schedules: 1, 5 and 21 infusions.

Notice that the three curves are superposed for low doses/low toxicities. This means that when the total dose is low, the occurrence of toxicity does not depend on the number of infusions, illustrating the concentration-dependence toxic behaviour of the drug. When the total dose increases, the occurrence of toxicity tends to a plateau at a level that depends on the number of infusions, and below 100 per cent. This is a consequence of the phenomenon described in Section 3.2. When the limit of toxicity K_0 is reached (with high concentrations), the drug toxic behaviour becomes time-dependent. Of course, this study deals only with neutropenia, and many other types of toxicity may occur with high drug doses!

Recall that 5 infusions were administrated and the daily dose could vary during the cycle. The total dose varied between 4.77 mg and 14.3 mg. As the total dose was not equally fractionated on the 5 days, we chose to simulate the percentage of intolerable toxicity in our sample as follows. For the ith patient, a sample $(\ln \Psi_{ij}^*)_{j=1\ldots100}$ was drawn from the PD population distribution $\mathrm{N}(\hat{m}, \hat{D})$. These 100 individuals were given the PK parameters of the ith

patient, as well as her five daily doses. The critical times $T_{ij}^{*} = \int_0^{\infty} 1_{\{N_i^*(t,\Psi_{ij}^*) \leqslant 500\ \mathrm{PN/mm^3}\}}(t)\,\mathrm{d}t$ were computed. The proportion of intolerable toxicity in the sample was then estimated as the percentage $\frac{1}{42 \times 100} \sum_{i,j} 1_{\{T_{ij}^* \geqslant 7\ \mathrm{days}\}}$. We obtained 39 per cent.

In order to see if this prediction agrees with the data, the empirical percentage of toxicity in the sample was evaluated by interpolating linearly the observed neutrophil counts for each woman. With this method, only seven women (17 per cent) showed an intolerable toxicity, which is far from 39 per cent. Actually, the empirical percentage of toxicity depends on the chosen method of interpolation. As the neutrophil profiles are convex around their minimum, the linear interpolation underestimates the time spent below 500 $\mathrm{PN/mm^3}$, especially when certain observation times are far from each other. With a smooth interpolation, 19 women (45 per cent) showed an intolerable toxicity, which is in agreement with the predicted percentage of toxicity.

In conclusion, Figures 7 and 8 enable the toxicity to be controlled. The determination of an optimal dosage regimen requires a study of efficacy as a second step. As an illustrative example, we come back to the mice study in reference [2]. Toxicity showed the same characteristic: a peak of toxicity for an intermediate schedule (5 days). Concerning efficacy, it was defined as animal survival measured by increase of lifespan. It turned out that efficacy shared the same behaviour, but shifted; peak efficacy occurred with the 20-day schedule. Thus, an optimal dosage regimen could be determined for mice (the 20-day schedule). For women, our analysis shows that for a given total dose, the toxicity does not (or not much) depend on the number of infusions. The optimal schedule may now be chosen according to efficacy.

In conclusion, we would like to emphasize that further simulations can be carried out for another purpose. The goal of our simulations was to predict the optimal schedule. With this schedule in hand (say, the 21-day schedule), one may wish to predict the optimal dose for a given patient. Assume for instance that this patient has had a first cycle of treatment, and that one wishes to choose the best dose for the second cycle of treatment. The kinetic and dynamic parameters may be estimated from the first cycle. With the hypothesis that kinetic and dynamic parameters do not change between cycles, their posterior distribution may be used, instead of the distribution in the whole population, to simulate the probability of toxicity, as above. Then, the dose may be adjusted accordingly.

REFERENCES

1. Rowinsky EK, Grochow LB, Hendricks CB, Ettinger DS, Forastiere AA, Hurowitz LA, McGuire WP, Sartorius SE, Lubejko BG, Kaufmann SH, Donhower RC. Phase I and pharmacologic study of topotecan: a novel topoisomerase I inhibitor. *Journal of Clinical Oncology* 1992; **10**:647–656.
2. Guichard S, Montazeri A, Chatelut E, Hennebelle I, Bugat R, Canal P. Schedule-dependent activity of topotecan in OVCAR-3 ovarian carcinoma xenograft: pharmacokinetic and pharmacodynamic evaluation. *Clinical Cancer Research* 2001; **7**:3222–3228.
3. Dayneka NL, Garg V, Jusko WJ. Comparison of four basic models of indirect pharmacodynamic responses. *Journal of Pharmacokinetics and Biopharmaceutics* 1993; **21**:457–478.
4. Gobburu JVS, Jusko WJ. Role of dosage regimen in controlling indirect pharmacodynamic responses. *Advanced Drug Delivery Reviews* 2001; **46**:45–57.
5. Minami H, Sasaki Y, Saijo N, Ohtsu T, Fujii H, Igarashi T, Itoh K. Indirect-response model for the time course of leukopenia with anti-cancer drugs. *Clinical Pharmacology and Therapeutics* 1998; **64**:511–521.
6. Zamboni WC, D'Argenio DZ, Stewart CF, MacVittie T, Delauter BJ, Farese AM, Potter DM, Kubat NM, Tubergen D, Egorin MJ. Pharmacodynamic model of topotecan-induced time course of neutropenia. *Clinical Cancer Research* 2001; **7**:2301–2308.
7. Friberg LE, Brindley CJ, Karlsson MO, Devlin AJ. Models of schedule dependent haematological toxicity of 2′-deoxy-2′-methylidenecytinine (DMDC). *European Journal of Clinical Pharmacology* 2000; **56**:567–574.

8. Friberg LE, Freijs A, Sandström M, Karlsson MO. Semiphysiological model for the time course of leukocytes after varying schedules of 5-Fluorouracil in rats. *Journal of Pharmacology and Experimental Therapeutics* 2000; **295**(2):734–740.
9. Montazeri A, Boucaud M, Lokiec F, Pinguet F, Culine S, Déporte-Féty R, Albin N, Laguerre B, Goupil A, Bugat R, Canal P, Chatelut E. Population pharmacokinetics of topotecan: Intraindividual variability of total drug. *Cancer Chemotherapy and Pharmacology* 2000; **46**(5):375–381.
10. Beal SL, Sheiner LB. NONMEM user's guide. *Nonlinear Mixed Effects Models for Repeated Measures Data*. University of California: San Francisco, 1992.
11. O'Reilly S. Topotecan: what dose, what schedule, what route? *Clinical Cancer Research* 1999; **5**:3–5.

