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**MODEL-BASED OPTIMAL DESIGN IN PHARMACOMETRICS USING ROBUST AND
ADAPTIVE APPROACHES WITH APPLICATION IN ONCOLOGY**

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Abstract (French)

Le choix de protocole de prélèvements dans un essai clinique ou préclinique est une étape cruciale car un protocole non informatif peut conduire à des études non concluantes. Des approches basées sur la matrice d'information de Fisher (FIM) pour les modèles non linéaires à effets mixtes ont donc été développées pour l'évaluation et l'optimisation de protocoles dans des études pharmacocinétiques (PK)/pharmacodynamiques (PD) de population, i.e. en pharmacométrie. La FIM est utilisée pour quantifier l'information attendue provenant d'un protocole donné et prédire les erreurs standard relatives (pRSE) des paramètres de population.

Le choix de protocole optimal dépend de la connaissance préalable sur les modèles et les paramètres, ce qui peut être partiellement erroné. Cependant, les protocoles de prélèvements sont souvent fixés tout au long d'un essai avec des données analysées seulement à la fin.

Des approches des protocoles robustes ont été développées pour tenir compte de l'incertitude sur les paramètres, en attribuant aux paramètres des distributions à priori. Des approches alternatives sont les designs adaptatifs (AD), qui consistent en des designs qui utilisent l'accumulation d'informations pour modifier les aspects prédéfinis de l'étude après chaque nouvelle cohorte d'individus. Ces deux approches ont fait l'objet d'une évaluation plus approfondie dans le cadre de ce doctorat en utilisant des exemples de PKPD en oncologie pour des données continues et une nouvelle évaluation de la FIM pour des études avec des données longitudinales discrètes.

En outre, la planification statistique de protocoles était rarement appliquée aux études précliniques. Dans cette thèse, nous avons appliqué la théorie du design pour le modèle d'inhibition de la croissance tumorale de Simeoni afin d'évaluer l'importance d'inclure des mesures au cours de la phase de croissance de la tumeur des expériences de xénogreffe pour une meilleure estimation des paramètres du modèle.

En conclusion, cette thèse présente différents contextes et l'utilisation de stratégies de planification de protocoles basées sur la FIM qui sont des approches puissantes qui permettent d'améliorer la qualité d'une étude, en garantissant sa fiabilité et une meilleure précision de l'estimation des paramètres.

Abstract (Italian)

La scelta di un buon design (numero di individui, tempi di campionamento) è essenziale nella pianificazione di uno studio in ambito clinico o preclinico, in quanto un design non informativo può portare a conclusioni sbagliate, o perlomeno imprecise. Per la valutazione e l'ottimizzazione del design, i.e. "optimal design", è stato considerato un approccio che si basa sulla matrice di informazione di Fisher (FIM) per modelli di popolazione, detti modelli non lineari a effetti misti. In particolare, il contesto è quello di studi farmacocinetici (PK) / farmacodinamici (PD), ossia nell'ambito della farmacometria. La matrice di Fisher è utilizzata per il calcolo dell'errore standard atteso di ciascun parametro di popolazione, rispetto ad un determinato design.

La scelta del design ottimale dipende dalle informazioni note a priori sul modello e sui parametri del modello, informazioni che possono essere imprecise o addirittura in parte sbagliate. Tuttavia, il design solitamente rimane fissato durante tutto il periodo dello studio, e i dati vengono analizzati solo al termine.

Approcci basati su design robusti sono stati sviluppati in modo da introdurre l'incertezza dei parametri, assumendo una distribuzione a priori per ciascun parametro. Un approccio alternativo è l'optimal adaptive design, un metodo flessibile che consente di modificare alcuni aspetti predefiniti attraverso l'informazione accumulata nel corso dello studio. Entrambi gli approcci sono stati oggetto di una valutazione approfondita in questo dottorato, per la quale è stato utilizzato un modello PKPD in ambito oncologico per dati continui e un nuovo metodo per ottenere la matrice di Fisher per un modello con dati longitudinali discreti.

Inoltre, l'optimal design è stato raramente applicato a studi in ambito preclinico. In questa tesi, la teoria del design è stata utilizzata in esperimenti *in vivo* sugli xenograft, al fine di valutare quantitativamente la diversità, in termini di precisione della stima dei parametri, nel caso in cui venissero o meno prese le misure della massa tumorale anche dopo il termine del trattamento.

Per concludere, questa tesi presenta diversi contesti per l'utilizzo di approcci legati all'optimal design basato sulla matrice di Fisher, mostrando che sono approcci potenti che consentono di migliorare la qualità di uno studio, garantendone l'affidabilità e una migliore precisione della stima dei parametri.

Chapter 1

Introduction

Drug disease model resources (DDMoRe)

This thesis was funded by the DDMoRe consortium (<http://ddmore.eu>), a five-years Innovative Medicines Initiative (IMI) project, since October 2013, in which EFPIA, Academic and Small Medium Enterprise (SME) partners are involved to provide with new tools to facilitate modeling and simulation in model-based drug development (Harnisch et al., 2013).

Within the passed five years, three main DDMoRe open sources products have been released. These are: i) the DDMoRe model repository, i.e. a public repository where models can be published, are supported by peer review, and can be downloaded; ii) the DDMoRe interoperability framework, i.e. an integrated infrastructure that provides with exchange and integration of models across modeling languages and existing or new tools through the Pharmacometrics Markup Language (PharmML), developed by Swat et al. (2015), and the standard model description language MDL (human readable language); iii) the DDMoRe training, i.e. courses about how to use DDMoRe products and how to apply them in drug development.

1.1 Pharmacometrics

Pharmacometrics is a relatively young bridging discipline that links biostatistics and pharmacology (Barrett et al., 2008; Pfister and D'Argenio, 2010). First introduced by L.B. Sheiner, it was defined by van der Graaf (2012) as "the science of developing and applying mathematical and statistical methods to characterize, understand, and predict a drug's pharmacokinetic and pharmacodynamic behavior".

Pharmacometrics's methods are widely used in drug development, and their benefit over traditional statistical approaches was reported in (Jonsson and Sheiner, 2002; Bhattaram et al., 2007).

1.1.1 Pharmacokinetics and pharmacodynamics

Pharmacometrics is mainly used for analysis of pharmacokinetic (PK) and pharmacodynamic (PD) data. As reported in (Ette and Williams, 2006), "pharmacokinetics is what the body does to the drug; pharmacodynamics is what the drug does to the body". Indeed, pharmacokinetics is the study of the movement of the drug into, through and out various areas of the body through the mechanism of the ADME of the drug, where ADME stands for absorption, distribution, metabolism and elimination (Gabrielsson and Weiner, 2006; Rowland and Tozer, 1995). Drug concentration measurements over time help to characterize the PK of the drug. Figure 1.1 (panel A) shows an example of the plasma drug concentration profile over time, after an oral administration. During the phase of absorption, concentration increases until reaching a maximum (C_{\max}). It then decreases in the elimination phase. The measurement that helps quantifying the total exposure to the drug of an individual is the so called "area under the curve", AUC , which is the area underlying the drug concentration curve over time. The parameter related to the drug distribution is the volume of distribution, V , whereas the drug elimination is characterized by the clearance, Cl . From the elimination phase it is possible to derive the half-life, $t_{1/2}$, i.e. the time needed by the organism for eliminating half of the quantity of administered drug. In case of oral administration, V and Cl are estimated over a constant F , i.e. the bioavailability, which is the fraction of active pharmaceutical ingredient reaching the systemic circulation. F can be estimated by comparing concentration measurements obtained after oral and intravenous administration. In case of single dose administration and linear pharmacokinetic, Cl can be derived knowing the AUC , F and the dose D , with the following: $AUC = \frac{FD}{Cl}$ (Gabrielsson and Weiner, 2006).

The PK models used in this thesis are based on the assumption that the organism is composed by one or more compartments in which the drug enters, is distributed, metabolized and eliminated. In figure 1.2 a one-compartment first order absorption and elimination model for an oral administration is presented, with k_a being the rate constant of absorption of the drug and $k_e = Cl/V$ the first order rate constant of elimination.

Compartmental models can be written either using differential equations or with analytic expression, if the analytic solution exists. The analytic expression of the model in figure 1.2 can be defined with two different parameterizations: $(k_a, k_e, V/F)$ or $(k_a, Cl/F, V/F)$. Using the first parameterization, the structural model f is described by the following equation:

$$f(\theta, t) = \frac{FD}{V} \frac{k_a}{k_e - k_a} (e^{-k_a t} - e^{-k_e t}) \quad (1.1)$$

where θ is the vector of model parameters $(k_a, k_e, V/F)$.

In order for the drug to produce the desired effect, it is necessary to reach concentrations that are pharmacodynamically active in the biophase– a compartment for the site of action

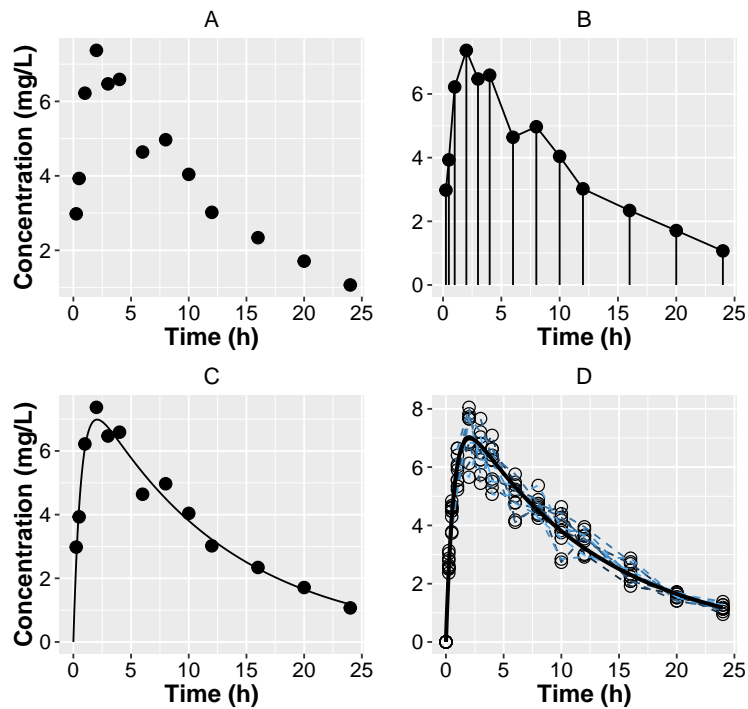


Figure 1.1: Example of plasma drug concentration profiles over time: (A) Measurements in one individual; (B) Non-compartmental analysis - area under the curve performed with the trapezoidal rule; (C) Nonlinear regression using one-compartment first order absorption and elimination PK model, where the black line represents model predictions; (D) Repeated measurements observed in several individuals (empty dots linked by dashed lines) which are analyzed using nonlinear mixed-effect model: the solid line represents the "population" profile obtained from the estimated fixed-effects.

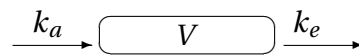


Figure 1.2: Schematic illustration of the one-compartment model with first order absorption and elimination.

of the drug— regardless the route of administration used. Pharmacodynamics, indeed, is the study of the effect of a drug.

In presence of a direct link between concentrations C and effect E , with drug increasing the response, the usual model that describes this link is the E_{\max} model. The sigmoid E_{\max} model is expressed with the following equation:

$$E = E_0 + \frac{E_{\max} C^\gamma}{C_{50}^\gamma + C^\gamma} \quad (1.2)$$

where E_0 is the baseline value, E_{\max} is the maximal drug effect, C_{50} is the drug concentration needed to achieve the 50% of the maximal effect ($E = E_0 + E_{\max}/2$) and γ is a sigmoidicity factor which controls the steepness of the concentration-response curve. It was introduced by Hill as extension to the traditional E_{\max} model, that is obtained for $\gamma = 1$. Sometimes the response E is directly explained by the dose D . The equation 1.2 can then be written

by replacing C with D and C_{50} with D_{50} . If the effect of the drug causes a decrease in the response, the model is called I_{\max} . The sigmoid I_{\max} model is expressed with the following equation:

$$E = E_0 \left(1 - \frac{I_{\max} C^\gamma}{C_{50}^\gamma + C^\gamma} \right) \quad (1.3)$$

where I_{\max} is maximal inhibitory effect and C_{50} is the drug concentration needed to achieve the 50% of the maximal inhibition.

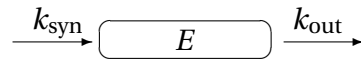


Figure 1.3: Schematic illustration of a turnover model of the effect E .

Several drugs do not act directly on the effect E . In that case, E is produced and eliminated by the input synthesis rate, k_{syn} , and the output elimination rate constant, k_{out} , respectively. These kind of models have a similar structure to the one presented in figure 1.3. The corresponding differential equation is expressed with the following:

$$\frac{dE}{dt} = k_{\text{syn}} - k_{\text{out}} E \quad (1.4)$$

In absence of treatment, $E(t) = \frac{k_{\text{syn}}}{k_{\text{out}}}$, which is the initial condition before the start of the treatment. By introducing the drug, its action can impact the input (k_{syn}) or the output (k_{out}). For instance, a turnover model with drug inhibiting the input is expressed with:

$$\frac{dE}{dt} = k_{\text{syn}} \left(1 - \frac{I_{\max} C}{C_{50} + C} \right) - k_{\text{out}} E \quad (1.5)$$

Figure 1.4 displays the concentration and effect curves over time, related to the mean profiles obtained from a one compartment PK model with first order absorption and elimination and a turnover PD model, as the one described in equation 1.5. The simulated parameters are the following : $k_a = 2 \text{ h}^{-1}$, $V = 8 \text{ L}$, $Cl = 0.1 \text{ L h}^{-1}$, $I_{\max} = 1$, $k_{\text{syn}} = 6$, $k_{\text{out}} = 0.04 \text{ h}^{-1}$, $C_{50} = 1 \text{ mg L}^{-1}$. A documentation of various PK and PD models is available in (Gabrielsson and Weiner, 2006; Dubois et al., 2014)

1.1.2 Modeling

In the context of population PKPD studies, repeated samples are taken over time from several individuals. Various approaches have been developed for analyzing this type of data. One method is the naïve average data (NAD) (Magni et al., 2014) in which the model can be fitted using standard nonlinear regression. In this approach, the average of the subjects observations is computed at each sampling time to obtain the typical response, leading to a more straightforward parameter estimation. However, NAD does not model all sources of

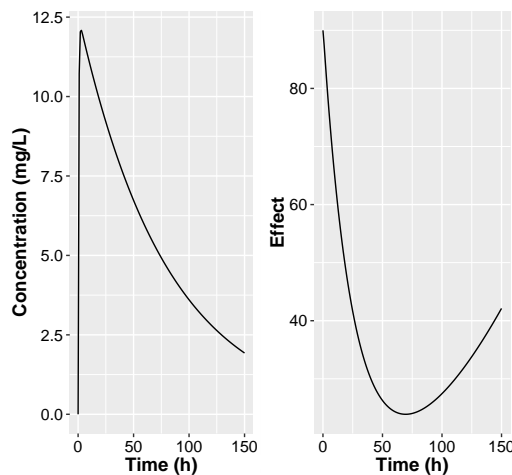


Figure 1.4: PK (left) and PD (right) simulated mean profiles for a total daily dose of 150 mg.

variability, therefore no information about the differences between subjects is considered. NAD is suitable for case studies in which all individuals are similar, with a small inter-individual variability. On the other hand, if between subject variability is high, NAD might lead to biased results.

A more practical method than NAD is the naïve pooled data (NPD) (Magni et al., 2014). In NPD repeated subjects observations are fitted simultaneously and considered as measured from the same subject. As for NAD, NPD works better in cases with low inter-individual variability. Moreover, NPD is more flexible than NAD as the experiment and number of samples can differ among subjects. Nonetheless, this may cause biases when a different amount of data is available for each subject, as individuals with rich data are more influential than those with few observations, i.e. sparse data.

An alternative approach that can be used when naïve methods are not feasible is the standard two-stage (STS) (Steimer et al., 1984; Magni et al., 2014). In the first step parameters are estimated from each individual, whereas population parameters can be derived in the second stage using the summary statistics from the individual estimates. It is simple to apply but it leads to overestimation of the variability, and it requires a rich design.

Nonlinear mixed effects models (NLMEMs) (Sheiner et al., 1972; Sheiner and Steimer, 2000; Lalonde et al., 2007; Smith and Vincent, 2010; Asín-Prieto et al., 2014; Lavielle, 2014) are a popular alternative approach. This method is in general more complex, but it has been increasingly used in the biomedical field as it keeps all the data of all subjects, and it allows to model all sources of variability.

NLMEM is the main approach adopted in this PhD. For the preclinical example in oncology – discussed in Chapter 2 – NPD, and therefore standard nonlinear regression, was also applied.

1.1.3 Nonlinear mixed effects models

A NLMEM incorporates both fixed effects that describe a typical individual, and random effects, which include the variability between individuals, assessing the distribution for the respective model parameters, and residual unexplained variability (error). Some or all of the fixed and random effects occur non-linearly in the model function (Pinheiro and Bates, 2000). Additionally NLMEM allows different structures for the residual errors – like additive and proportional.

Continuous outcome Let y_{ik} be the n_i -vector of k continuous responses (e.g. concentrations and effect, for $k = 2$) measured from subject i ($i = 1, \dots, N$) at times $\xi_i = (t_{i1}, \dots, t_{in_i})$. Assuming $k = 1$, and removing the index k for simplicity, y_i is denoted with:

$$y_i = f(\theta_i, \xi_i) + \epsilon_i \quad (1.6)$$

where f is the nonlinear structural model and ξ_i is the elementary design of individual i composed of n_i sampling times. Of note, in this thesis the attention was restricted to sampling times, but other design variables may be of interest, such as the doses. θ_i is the vector of p individual parameters and ϵ_i is the vector of residual error following a normal distribution $\epsilon_i \sim N(0, \Sigma(\theta, \xi_i))$ with $\Sigma(\theta, \xi_i) = \text{diag}(\sigma_{\text{inter}} + \sigma_{\text{slope}} \times f(\theta_i, \xi_i))^2$. σ_{inter} is the parameter for the additive error and σ_{slope} for proportional error. The individual parameters θ_i for the i^{th} subject is modeled as a function g , such that $\theta_i = g(\mu, \eta_i)$. Often it is assumed that θ_i follows a normal distribution, then the function g is additive:

$$g(\mu, \eta_i) = \mu + \eta_i \quad (1.7)$$

or it can be assumed that θ_i follows a log-normal distribution, then g is exponential:

$$g(\mu, \eta_i) = \mu \times e^{\eta_i} \quad (1.8)$$

where μ is the vector of p fixed effects similar in all individuals and η_i is the random effect associated to subject i which follows a normal distribution $\eta_i \sim N(0, \Omega)$, where Ω is a $p \times p$ variance-covariance matrix, that is assumed diagonal with diagonal elements the variances $\omega_s^2, s = 1, \dots, p$. We assume that $\epsilon_i | \eta_i$ are independent between subjects and, for each individual, ϵ_i and η_i are independent.

Part of variability can be explained by subject-specific characteristics, as the age, weight, gender, type of treatment received etc., which can be expressed by introducing covariates in the model. Covariates can be either continuous or discrete, and they may or may not vary between occasions. Let us consider a binary covariate C_i for the individual i , which does not vary during time, ($C_i = 0$ for the reference class otherwise $C_i = 1$), the individual parameter

vector θ_i , assuming that it is normally distributed, is defined as follows:

$$\theta_i = \mu + \beta C_i + \eta_i \quad (1.9)$$

or for log-normal distribution of θ_i :

$$\log(\theta_i) = \log(\mu) + \beta C_i + \eta_i \quad (1.10)$$

where μ is the vector of fixed effects related to the reference class of the covariates, β is the effect of the covariate C , i.e. the increase of θ_i with respect to μ when $C_i = 1$.

If C_i is a continuous covariate, the values that it takes are in the continuous scale. By considering a reference individual, and assuming log-normal distribution for the parameter θ_i , the model for continuous covariates can be written as following:

$$\log(\theta_i) = \log(\mu) + \beta(\log(C_i) - \log(C_{pop})) + \eta_i \quad (1.11)$$

where μ is the reference value of the parameter, and C_{pop} is the covariate set to its reference value, e.g. observed median or mean. In equation 1.11, the variability is decomposed in the following: i) in the term $\beta(\log(C_i) - \log(C_{pop}))$ the variability is expressed by the variation of the covariate $\log(C_i)$ with respect to the reference $\log(C_{pop})$; ii) the component η_i , that is the remaining variability, describes the between subject variability for individuals that have same value for the covariate.

Let λ be the vector of variances terms $(\omega_1^2, \dots, \omega_p^2, \sigma_{inter}, \sigma_{slope})$, the vector of population parameters to be estimated is defined as $\Psi = (\mu', \beta', \lambda')'$, which is of size P .

Figure 1.1 (D) displays the individual profiles over time of plasma concentration and the corresponding mean profile for an oral administration of a certain compound. The prediction of the mean profile is obtained from the PK function f which is the one-compartment model with first order absorption and elimination, and the fixed effects estimated from the data.

Discrete outcome For discrete data, the probability for all observations $j = 1, \dots, n_i$ from an individual i ($i = 1, \dots, N$) at times $\xi_i = (t_{i1}, \dots, t_{in_i})$, is denoted with:

$$P(y_i, \Psi | \eta_i) = \prod_{j=1}^{n_i} h(y_{ij}, \theta_i, \xi_i) \quad (1.12)$$

where h is a known link function that describes the probability model of observing y_i at times ξ_i . Of note, as opposed to the definition of models for continuous outcomes, the residual error term ϵ is not present in the expression of model for discrete outcomes.

Discrete data can be classified mainly into two groups: count data and noncount data. The former are repeated measurements of events occurring in specific time intervals, whereas the latter are either grouped binary data, i.e. numbers of successes (or failures)

obtained from a Bernoulli process, or ordinal data. The Poisson model is often used to describe count data, whereas a binomial distribution better describes the grouped binary data. More details can be found in (Plan, 2014), an extensive tutorial on modeling of count data.

An example of NLMEM for repeated binary response was proposed by Riviere et al. (2016); Ueckert and Mentré (2016) and it is expressed as follows:

$$\text{logit}(\pi) = \theta_1 + \theta_2(1 - \mu_3\delta)t \quad (1.13)$$

where π is the probability of success.

The fixed effects vector $\mu = (\mu_1, \mu_2, \mu_3)$ is given by the intercept μ_1 , the slope μ_2 and the treatment effect μ_3 . An additive random effect is associated to θ_1 and θ_2 , such that $\theta = g(\mu, \eta) = \mu + \eta$. δ is the treatment group indicator, i.e. $\delta = 0$ in control group and $\delta = 1$ in treatment group.

1.1.4 Estimation methods

Most estimation methods are based on the maximum likelihood estimation (MLE). In NLMEM, the likelihood of the vector of observations y of an individual i is defined as the integral over the random effects of its conditional likelihood:

$$L(y_i; \Psi) = \int_{\mathbb{R}^p} p(y_i | \eta_i; \Psi) p(\eta_i; \Psi) d\eta_i = \int_{\mathbb{R}^p} p(y_i, \eta_i; \Psi) d\eta_i \quad (1.14)$$

where $p(y|\eta; \Psi)$ is the conditional probability density function (p.d.f.) of y given the random effects η , $p(\eta; \Psi)$ is the p.d.f. of η and $p(y, \eta; \Psi)$ is the joint likelihood of the data y and the random effects η . Because of the non-linearity of the structural model f on the parameters, there is no closed form solution for the likelihood. However, several methods have been developed to approximate it.

Plan et al. (2012) proposed a review on the estimation algorithms in NLMEM existing in the literature (Pillai et al., 2005). These methods are implemented in various software, such as NONMEM (Beal et al., 2009), MONOLIX (Team MONOLIX, 2012), R (R Core Team, 2014), SAS (SAS Institute Inc, 2004) etc., and they will be introduced in this section.

The first developed algorithms are based on the first order Taylor expansion of f . The linearization leads to a linear mixed effect model, whose likelihood has a closed form. These algorithms are the first order (FO) and first order conditional estimation (FOCE) developed by Sheiner et al. (1972) and Lindstrom and Bates (1990), respectively. FO linearizes the model around the expectation of the random effects, i.e. 0, whereas FOCE linearizes the function around the individual estimates of the random effects $\hat{\eta}_i$. Both algorithms are implemented in NONMEM (Beal et al., 2009), the first software developed to analyze NLMEM, which is widely adopted by the pharmacometrics community. The algorithm FO is also implemented in the NLMIXED procedure in the SAS software, whereas the nlme function available in R

and S-plus (Pinheiro and Bates, 2000) implements an approximation of FOCE. In presence of large individual variability or with a number of observations per subjects that grows slower than the number of subjects, FO and FOCE tend to produce biased estimators (Ge et al., 2004; Vonesh, 1996).

More precise methods that are not based on linearization, have followed. Of note, the gain in precision requires more intensive computation time. Laplacian approximation, proposed in (Wolfinger, 1993) is based on second order Taylor expansion around the random effects, and the method is implemented in NONMEM and SAS.

Adaptive Gaussian quadrature (AGQ) (Pinheiro and Bates, 1995) computes $L(y; \Psi)$ numerically, as a weighted average of $p(y|\eta; \Psi)$ and $p(\eta; \Psi)$ with predefined number of nodes and weights. AGQ with one node corresponds to the Laplacian approximation. Moreover, AGQ is an extension of Gaussian quadrature (GQ) (Golub and Welsch, 1969) as the nodes instead of being centered on the expectation of the random effects (i.e. 0), they are centered on the estimated conditional modes of the random effects. As standard GQ does not consider the nature of the integrand, it requires a high number of nodes for obtaining precise estimates. AGQ instead requires a smaller number of nodes than GQ as it centers and scales the standard quadrature nodes, based on the areas of high density, and therefore it improves the approximation. Nevertheless, the method is not suitable for models with high number of random effects, as the computational complexity increases exponentially with the number of random effects of the model. This approach is implemented in the NLMIXED procedure of SAS.

The expectation maximization (EM) algorithm is an iterative algorithm, first developed by Dempster et al. (1977), used for maximizing the likelihood by treating as missing data the unknown individual random effect. Let $\hat{\Psi}_{k-1}$ be the population parameters vector at the beginning of the k^{th} iteration. In the E-step, the conditional expectation of the log-likelihood of the complete data is evaluated with respect to the distribution of unknown random effect η given the observations y and $\hat{\Psi}_{k-1}$:

$$Q_k(\Psi|\hat{\Psi}_{k-1}) = E[\log p(y, \eta; \Psi) | y, \hat{\Psi}_{k-1}] \quad (1.15)$$

In the M-step $\hat{\Psi}_{k-1}$ is updated for the next step by maximizing the expectation Q of the k^{th} iteration:

$$\hat{\Psi}_k = \underset{\Psi}{\operatorname{argmax}} Q_k(\Psi|\hat{\Psi}_{k-1}) \quad (1.16)$$

In this thesis, a stochastic version of the EM algorithm known as the stochastic approximation expectation maximisation (SAEM) algorithm, was used. It was developed by Delyon et al. (1999), and then modified in (Kuhn and Lavielle, 2004) by introducing a Markov chain Monte Carlo (MCMC) approach. In this method, the E-step of the k^{th} iteration is divided into two parts: first missing data (unknown individual random effect) are simulated from the conditional distribution $p(\cdot | y; \hat{\Psi}_{k-1})$ using the Markov chain Monte Carlo (MCMC)

algorithm. Then, in the second part, the stochastic approximation is computed by replacing the integral of the likelihood with a sufficient statistic, which is then maximized for updating the vector of population parameters Ψ . The updated parameter vector will then be used for the first part of the next iteration until convergence, which should be reached at a local or global maximum of the likelihood, under general conditions (Delyon et al., 1999).

The SAEM algorithm was extended by (Panhard and Samson, 2009) to allow for the estimation of the inter-occasion variability and it is implemented in the MONOLIX software, as well as in NONMEM (since version 7), in S-ADAPT (since version 1.56) and in the R package `saemix` (www.saemix.biostat.fr) since 2011 (Comets et al., 2016).

1.2 Optimal design

The development of estimation software and methods in NLMEM helps carrying out pharmacometrics analysis. Nevertheless, the definition of a good study design is still a problem that needs to be investigated.

The study of the influence of design on experiments goes back to the 20th century (Fisher and Mackenzie, 1923). By that time, R.A. Fisher discovered that there is a link between the information obtained from an experiment and the Fisher information matrix (FIM) (Box, 1980). Around twenty years later, Cramer and Rao observed that the variance of any unbiased estimator is greater than the inverse of FIM, which is defined independently of any estimation method (Cramer, 1946; Rao, 1992). Cramer-Rao inequality is the principle on which the optimal design theory described in this section is based.

A drawback of optimal designs is the fact that it requires previous information, both on models and parameters (Pronzato and Pázman, 2013a), which can be partially wrong as they may be difficult to guess. Currently, population designs are often fixed for the whole study with data analyzed at its end. Local optimal designs are optimized based on a set of parameters values defined prior to the study. As alternative to local designs, robust designs or adaptive designs can be applied. These two approaches are expanded in subsections 1.2.3 and 1.2.5, respectively.

1.2.1 Design definition

In the approximate design theory, a continuous design ξ is considered as a series of sampling times with a specific weight assigned, and the sum of these weights must be 1. Herein, without considering the weights, we define an elementary design for the subject i as ξ_i , which is given by the number of individual observations n_i and the sampling times t_{i1}, \dots, t_{in_i} . A population design Ξ is defined by the total number N of individuals and the set of individual elementary designs to be performed in each individual: ξ_1, \dots, ξ_N , with a total number of observations $n_{tot} = \sum_{i=1}^N n_i$. A special case is when the same elementary design is performed in all individuals ($\xi_i = \xi$ for $i = 1, \dots, N$), then $n_{tot} = n \times N$ and $\Xi = \{\xi; N\}$. Often

population designs are composed of Q individual groups with same elementary design ξ_q in each group of N_q subjects ($q = 1, \dots, Q$). The population design in this case is defined as $\Xi = \{[\xi_1, N_1]; \dots; [\xi_Q, N_Q]\}$, with $N = \sum_{q=1}^Q N_q$ and $n_{tot} = \sum_{q=1}^Q n_q \times N_q$. For the projects in this thesis the focus was on optimizing the allocation in time of the sampling times, for some fixed N and n .

Defining an appropriate design requires a good balance between the total number of subjects, the number of groups, the proportion of subjects associated to each group, the allocation in time of the sampling times, the number of observations per subject. All this factors, which are part of the design, are influential for the precision of parameter estimates of the model selected for the study. This has been shown very early by simulation studies (Al-Banna et al., 1990; Hashimoto and Sheiner, 1991; Jonsson et al., 1996).

This can be essential in cases of sparse design, such as in studies with children, old patients, or other studies. Making an adequate choice on the design thus leads to more informative and more ethical studies.

There are two main approaches to design evaluation. The first, by trial simulation, involves parameter estimation using numerous simulated datasets and thus is a time-consuming method (Holford et al., 2010). The second approach, which avoids simulation, relies on the Cramer-Rao inequality, thus on the expected FIM.

1.2.2 Fisher information matrix in NLMEM

From the individual likelihood expressed in equation 1.14, it is possible to define the individual Fisher information matrix $M_F(\Psi; \xi_i)$, for the subject i and the elementary design ξ_i , which can be defined as:

$$M_F(\Psi; \xi_i) = E \left(\frac{\partial \log L(y_i; \Psi)}{\partial \Psi} \times \frac{\partial \log L(y_i; \Psi)}{\partial \Psi'} \right) \quad (1.17)$$

or equivalently as:

$$M_F(\Psi; \xi_i) = -E \left(\frac{\partial^2 \log L(y_i; \Psi)}{\partial^2 \Psi} \right) \quad (1.18)$$

Because of the assumption that individuals are independent, the population FIM $M_F(\Psi, \Xi)$ is defined as the sum of N elementary FIMs:

$$M_F(\Psi, \Xi) = \sum_{i=1}^N M_F(\Psi, \xi_i) \quad (1.19)$$

In case of Q elementary designs it can be defined as:

$$M_F(\Psi, \Xi) = \sum_{q=1}^Q N_q M_F(\Psi, \xi_q) \quad (1.20)$$

As previously mentioned in the estimation methods section 1.1.4, because of the nonlinearity of the structural model f in the parameters, there is no analytic expression for

the likelihood, therefore also for FIM. Indeed, several approximations were developed for the evaluation of FIM in both continuous and discrete NLMEM.

Continuous outcome The first approximation of FIM for single response NLMEMs with continuous data was provided by Mentré et al., in the case of additive residual error model, with normally distributed errors with 0 mean and homoscedastic variance (Mentré et al., 1997). The method consisted in a linearization of f around the random effects, i.e. 0, using the FO approximation. The development of FIM was extended by Retout et al. (2002) who introduced the variability parameters of the combined residual error model in the vector of parameters to be estimated, allowing for heteroscedastic error models. The new method was tested by a simulation study using real data from the pharmacokinetics of the enoxaparin (Retout et al., 2002; Retout and Mentré, 2003). The FIM can then be expressed either as full matrix or as block diagonal. The former is more complex than the latter because it requires calculation of the second derivatives of the model, however, there is no clear consensus on which is the best approximation. Some comparison studies have been performed by (Nyberg et al., 2015) by simulation, for a simple PK model and a more complex PKPD model. They showed that the block-diagonal matrix provided with results that were closer to those obtained using the full matrix. These conclusions were also supported by (Mielke, 2012). Moreover, the observed FIM is implemented as block-diagonal in the MONOLIX software. The FIM's expression in NLMEM was later extended to multiple response models, using the same linearization method (Hooker and Vicini, 2005). The new method was validated in a PKPD simulation study by Bazzoli et al. (2009). Dumont et al. (2013) proposed an extension of that matrix by taking into account the correlations among random effects, i.e. the full variance-covariance matrix Ω . Nevertheless, the linearization of the FIM by FO approximation has some limits. It performs well when the random effects variances are small, or in presence of moderate model nonlinearity (Jones et al., 1999; Jones and Wang, 1999).

Several new methods that avoid the FO approximation have been developed. For instance, methods based on the FOCE estimation algorithm were proposed by Retout and Mentré (2003), for which the linearization of the model around the individual random effects was performed, or by Nyberg et al. (2010), in which the linearization was computed around the expected mode of the marginal likelihood. The results obtained with FOCE are similar to those obtained by FO, but FOCE takes higher computational time because it requires MC simulations. Other approaches based on simulations were proposed by Vong et al. (2012) who used the Laplacian approach, or by Mielke (2012), who implemented MC integration in the design evaluation. These approaches provide correct predictions of the precision of the parameter estimates, but they are computationally expensive. Nguyen and Mentré (2014) used a method based on MC-AGQ to approximate FIM for normally distributed responses. They showed the superiority of their approach compared to FO, when the nonlinearity of the

model increases. Finally, Riviere et al. (2016) proposed an alternative method to compute the asymptotically exact FIM, based on MC–MCMC. This method is available in the R package *MIXFIM* and it uses functions that are written in the probabilistic language STAN (Carpenter et al., 2016), for Bayesian inference.

Discrete outcome In case of discrete outcomes, a closed form of FIM is not available in both generalized linear mixed effects models (GLMEM) and discrete NLMEM. Some pre-existing methods for the approximation of FIM for discrete data in GLMEM are available from the literature, but most of them have the limitation of being response-specific (Ueckert and Mentré, 2016). For instance, the method by Niaparast (2009) is limited to Poisson models with random intercept. Waite and Woods (2015) were the first to calculate FIM for general GLMEM, using marginal quasi-likelihood (MQL), which is similar to FO (Retout and Mentré, 2003) and penalized quasi-likelihood approximations, avoiding a full maximum likelihood estimation.

For discrete NLMEM Ogungbenro et al. (2005) developed a method based on generalized estimating equations and MQL approximation for count, binary and ordinal responses; Nyberg et al. (2009) used an approach based on second-order approximation of the likelihood and applied it to binary and count responses. The method of Nguyen and Mentré (2014) based on MC–AGQ for continuous response was recently extended by Ueckert and Mentré (2016) to discrete response mixed effects model, and the program for the implementation was written in R. This new method does not use any approximation and it allows to numerically compute FIM by calculating the derivatives of the exact conditional likelihood, however it should be noted that for more than three random effects the method becomes very computationally expensive. Finally, the method proposed by Riviere et al. (2016), based on MC–MCMC, can also be applied to NLMEMs with discrete outcome.

1.2.3 Optimality criteria

From equations (1.17,1.18) it is clear that the FIM in the context of NLMEMs depends on the design and on the vector of population parameters. When performing optimal design, we would like to obtain the most precise parameter estimates for the selected model, which means to "minimize" the estimation variances, i.e. "maximizing" the FIM. Several optimality criteria based on some scalar functions were developed for comparing FIMs in NLMEMs, by extending those already available in classic nonlinear regression (Fedorov, 1972; Atkinson et al., 1992). These criteria are also called "alphabetic" criteria as their name is an alphabetic letter (Atkinson et al., 1992).

The most important criterion, and widely used in pharmacometrics is the D-optimality (Fedorov, 1972), which consists in maximizing the determinant of FIM, allowing for minimizing the generalized variance of the parameter estimates. An extension of D criterion is the D_s criterion, which is useful when the interest is on estimating a subset s of

parameters. Another criterion is the A-optimality, for which the inverse of the trace of the FIM is minimized, therefore the average variance is minimized. E-optimality consists in maximizing the least eigenvalue of FIM, whereas C-optimality is based on minimizing the variance of a linear combination of the parameter estimates. These criteria were developed for performing "local" optimality, for which a design is optimized for a specific set of model parameters values, that is defined prior to the study.

Robustness with respect to parameters As opposed to local optimality, the principle of robust approaches with respect to parameters consists in assigning a prior distribution to population parameters, instead of constraining them to a fixed value. Several robust criteria based on D-optimality have been developed first in classic nonlinear regression (D'Argenio, 1981; Pronzato and Walter, 1985, 1988), and later they were adopted in pharmacometrics (Dodds et al., 2005). Optimal designs obtained using robust criteria are called robust designs (RD).

Robust criteria first proposed by Pronzato and Walter (1985) are the ED- and EID-optimality (Walter and Pronzato, 1987) which consist in maximizing the expectation of the determinant of the FIM and the inverse of the expectation of the inverse of the determinant, respectively. DE-optimality criterion, i.e. the determinant of the expectation of the FIM, is not equivalent to ED-optimality in nonlinear regression models. ED-optimal designs might have poor performance for parameters that are associated to small values of probability density function. Pronzato and Walter (1988) thus proposed the maxmin criterion (MM-optimality), which requires knowledge of a prior feasible region for population parameters Ψ . Indeed MM implies maximization of the least value of the determinant of FIMs associated to the different parameters that lie in that domain. Other robust criteria were further proposed. The ELD-optimality, or API (D'Argenio, 1990), consists in maximizing the expectation of the logarithm of the determinant of the FIM. A simplified version of these criteria– which does not require the whole prior parameters distribution but instead it uses only the 2.5th and 97.5th percentiles of that distribution (Pronzato and Pázman, 2013b) – are the so called HCDs criteria, developed for the hypercube D-optimal designs (HCDs) (Foo and Duffull, 2010). By using HCD the time for the computation is considerably reduced.

In the passed decades various studies have been performed using a robust design approach. Tod et al. (1998) used the EID robust criterion to find the optimal design in a population pharmacokinetics study; Dodds et al. (2005) showed in two-case studies that RDs, obtained using ED-optimality, are better than D-optimal designs in cases of misspecified prior information; Hennig et al. (2009) used ED-optimality to optimize treatment length in a disease progression study; Foo and Duffull (2010) compared in a PK study HCDs versus some robust designs, highlighting how HCDs designs are computed much faster than traditional RDs, being an approximation of traditional RDs. Nyberg and Hooker (2012) also compared standard robust criteria and HCDs for an E_{max} model and for an

exponential decay model. Parameter uncertainty was also taken into account in the context of generalized linear models (Woods et al., 2006)

Robustness with respect to models T-optimality (Atkinson and Fedorov, 1975b,a) can be used for taking into account of uncertainty around several models. T-optimality used in combination with D-optimality leads to compound optimality (McGree et al., 2008), which consists in combining opposing criteria. Indeed, D-optimality concerns parameter estimation, whereas T-optimality accounts for model discrimination. Jansen et al. (2013) applied T-optimality to discriminate among models that describe the individual patient's parasite-time profile; Nguyen et al. (2016) used compound optimality as weighted criterion for combining optimality criteria of several models to find informative sampling times for two different drugs while studying the influence of the model weight on the optimization results. (Woods et al., 2006) took into account models uncertainty in the context of generalized linear models.

1.2.4 Optimization algorithms

Several algorithms were developed for the optimization of a design. As explained in subsection 1.2.1, a population design for Q groups of N_q subjects with same elementary design is defined as $\Xi = \{[\xi_1, N_1]; \dots; [\xi_Q, N_Q]\}$, with $N = \sum_{q=1}^Q N_q$ and $n_{tot} = \sum_{q=1}^Q n_q \times N_q$. It is possible to attribute a rational weight to each group, $m_q = \frac{N_q}{N}$, corresponding to the proportion of subjects m_q who have same elementary design ξ_q such that $\sum_{q=1}^Q m_q = 1$. Ξ can thus be defined as $\Xi = \{[\xi_1, m_1]; \dots; [\xi_Q, m_Q]\}$, which is a statistical design.

A widely used algorithm is the Fedorov-Wynn (FW), which provides the optimal statistical design, no matter the initial design used for the initialization of the algorithm (Fedorov, 1972; Wynn, 1972; Walter and Pronzato, 1997). The FW algorithm was initially developed in a context of nonlinear regression (Kiefer and Wolfowitz, 1959), and then extended to NLMEMs by Mentré et al. (1997) in order to determine D-optimal designs. FW is an iterative algorithm that can determine the best elementary design combination among all possible predefined finite times or doses, which maximizes the FIM. Indeed, the FW algorithm optimizes the number of Q elementary designs, and the proportions of subjects m_q who are assigned to each elementary design (ξ_q). All these features constitute the structure of the population design Ξ .

For obtaining optimal exact designs, i.e. without accounting for proportions of subjects, based on D-optimality, some specific algorithms were developed, based on the Fedorov exchange algorithm. Let time be the design variable, the principle of these algorithms is to improve at each iteration the initial design by exchanging one or more sampling times with better times, i.e. times that lead to higher determinant (Fedorov, 1972; Wynn, 1972; Mitchell, 1974). Ogungbenro et al. (2005) proposed a modified version Fedorov exchange algorithm, in order to obtain exact D-optimal design with respect to the design variable. They showed

the good performance of the algorithm but computational time is higher with respect to the classical exchange algorithm.

Other optimization algorithms are the gradient or the Newton algorithms, which are based on the computation of first and second derivatives, respectively. The simplex algorithm can also be used in this context, and it does not require any derivative computation. However, these algorithms are local algorithms that do not guarantee to find a global optimum but just a local one, i.e. an optimum in the neighborhood of the initial point. The random search algorithm allows to find a global optimum and it does not require derivative calculation. A two-stage approach of this algorithm based on the method of stochastic gradient, also called method of Broyden-Fletcher-Goldfarb-Shanno (Broyden, 1970) was described by (Foracchia et al., 2004). Simulated annealing is a global optimization algorithm, more often used than simplex and the random search algorithms, for more complex design situations (Duffull et al., 2002).

Retout et al. (2007) compared FW and simplex algorithm, and showed a better performance of the former, in case of complex design structures. More recently (Retout and Mentré, 2003) proposed an extension FW algorithm by incorporating cost functions and showed the applicability for multiple-response models. The costs were based on clinical practice, e.g. cost of the elementary design defined by the number of samples to be taken; cost of including a new patient in the study etc.

1.2.5 Adaptive design in pharmacometrics

Adaptive designs (ADs) are promising alternatives to local or robust designs (Dodds et al., 2005). As opposite to traditional clinical studies, ADs are clinical trial designs that use accumulating information in order to decide how to modify predefined aspects of the study during its implementation instead of leaving them fixed until the end (Chang, 2007; Pronzato, 2009; Foo and Duffull, 2012). This is very important, for instance when designing a clinical trial, having only prior information on preclinical data, or designing a study in children from adults information. ADs are useful to provide some flexibility but were rarely used for NLMEM (Zamuner et al., 2010). Nevertheless, Maloney et al. (2007) showed that ADs are less sensitive to miss-specification of the initial design. Moreover, Leonov and Miller (2009) implemented ADs for the E_{max} model, with an application of dose findings in Alzheimer's disease study, whereas (Zamuner et al., 2010) applied ADs in positron emission tomography studies.

According to a survey by Mentré et al. (2013), adoption of adaptive design approach for population PKPD studies is promising in pharmaceutical industries. Indeed, ADs in NLMEMs were recently used in different context: Foo and Duffull (2012) showed the applicability of ADs for a PK bridging study; Strömberg and Hooker (2015) implemented stopping criteria defined by the Food and Drug Administration in pediatric PK bridging studies; Ryznik et al. (2016) applied ADs in a dose finding study for time to event responses

with adaptive choice of the number of patient based on the rate of response. Finally Bogacka (2016) presented an application of ADs in clinical trials optimizing both doses and PK parameters.

It was shown, in previous studies not concerning NLMEM, that two-stage designs could be more efficient than fully adaptive designs (Fedorov et al., 2012) when the adaptation is performed after each patient. (Chen, 1997) compared three- versus two-stage AD. They found that the gain of extending a two-stage design approach to three-stage design is not as relevant as compared to the advantage of using two-stage instead of one-stage design. Moreover, two-stage designs are easier to implement in clinical practice as only one adaptation is performed. Some important steps in AD are choosing the number of stages one wants to compute, i.e., the number of times parameters estimation and adaptations will be performed, and the cohort size to be set for each stage. For NLMEM, Dumont et al. (2016) implemented the optimization of the determinant of FIM for two-stage adaptive designs. In that paper (Dumont et al., 2016) a simulation study that mimicked the design of a pediatric PK trial was used and it was analyzed through NLMEM. For the first stage, parameters were guessed from adults. Simulations of one- and two-stage designs were evaluated assuming that some parameters were different than the initial ones. The influence of the size of the cohorts in two-stage designs was also investigated. They showed the applicability and usefulness of the approach that we wished to further investigate in this thesis in a more complex example.

1.2.6 Optimal design software

Several software that allow for computing the FIM and performing optimal design based on the D-criterion have been developed in the past years. These are: PFIM (Mentré et al., 2014), PopED (Nyberg et al., 2012), PopDes (Gueorguieva et al., 2007), POPT (Duffull, 2006), MIXFIM (Riviere and Mentré, 2015). PFIM, PopED and MIXFIM are available in R, the other software are implemented in MATLAB. A MATLAB version of PopED is also available. Besides POPT, all the software can be freely downloaded online.

A recent study (Mentré et al., 2013) showed that all these optimal design software, excluding MIXFIM as it was not already built at that time, provide with the same answers using the same FO approximation of FIM. PopED is the only software presently which allows robust design. The new release of PFIM, PFIM 4.0, was the first software allowing to perform adaptive design (Dumont et al., 2016), as the FIM can be saved and prior FIM can be uploaded. Andrew Hooker has recently developed an R prototype for adaptive design, "MBAOD" (Model Based Adaptive Optimal Design), and it was already used by Ryznik et al. (2016). MBAOD allows to perform adaptive design in R through sequential use of PopED and NONMEM, the former for optimizing design, and the latter for generating data and estimating parameters.

1.3 Pharmacometrics and oncology

Modeling in clinical pharmacology and translational medicine research in oncology has put a big effort in building models for simplifying and describe complex biological systems (Rew, 2000; Quaranta et al., 2005). Indeed, mathematical modeling is a powerful discipline that can be used to derive quantitative measures of biological functions for performing predictions and prognosis (Della Pasqua, 2011).

First models in oncology were developed to describe the simple exponential growth of solid tumors, with cell accruing at a constant rate (Laird, 1964). Subsequently, the mathematics behind these models became more complex, in order to account for the process of tumor cells spread; the development of metastasis, and for considering that some tumor cells are not immortal (Deisboeck et al., 2001). Nowadays, population PKPD modeling is a key tool in oncology drug development, for describing and quantifying dose-response relationships (Bender et al., 2015; Venkatakrishnan et al., 2015). The main modeling approaches concern tumor growth studies, tumor marker/biomarker and survival analysis.

Figure 1.5 by Bender (2016) displays a schema of modeling in oncology drug development. Everything starts with defining a population PK model. Subsequently, the PK is linked to the PD leading to PKPD models for various outcomes, such as tumor and biomarker responses etc., which can help predicting the overall survival (OS). More specifically, the efficacy of anti-tumor treatment is often determined using tumor bearing mice, whereas in clinical studies, treatment effect for solid tumors is assessed by tumor sum of longest diameter (SLD) measurements. Other indicators of treatment effect are the biomarkers associated with drug mechanism of action. Chemotherapy-induced myelosuppression or other adverse effects can be recognized in cancer treatments. Treatment success is established using progression-free survival (PFS) and overall survival (OS) clinical endpoints.

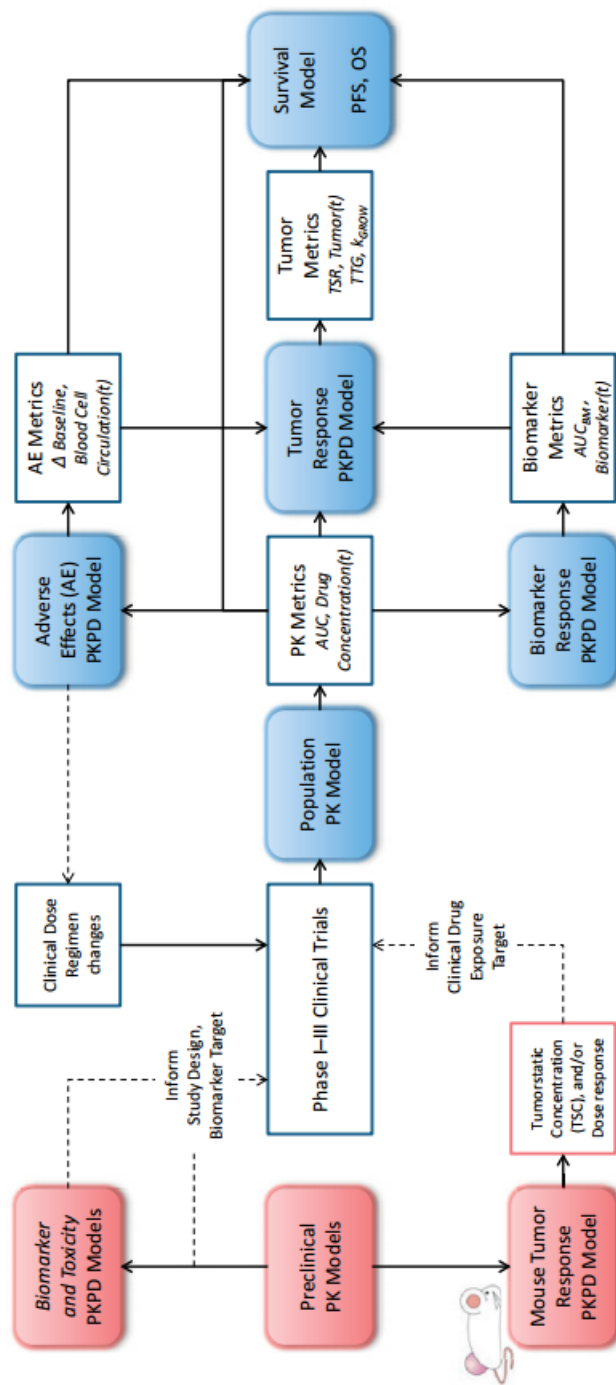


Figure 1.5: (Bender, 2016): schematic representation of modeling in oncology in drug development. Δ Baseline: change from baseline; AUC: area under the curve; AUC_{BM} : area under biomarker-time curve; K_{growth} : tumor growth rate constant parameter; OS: overall survival; PFS: progression-free survival; PKPD: pharmacokinetic-pharmacodynamic;(t): time-course; TSR: tumour size ratio; TTG: time to tumour growth.

1.3.1 *In vivo* preclinical studies

Xenografts experiments Among *in vivo* preclinical studies, the most popular is the xenograft model (Kelland, 2004), for which the common practice is either to implant tumor fragments subcutaneously or to inject tumor cells into the flank of athymic mice (Simeoni et al., 2004a). When the tumor has reached the desired volume, mice are randomized into control and treatment arms receiving either a vehicle (control arm) or the compound to be tested at different dose levels or with different drug administration schedules (treatment arms). Tumor sizes are measured at repeated time points during the study and the collected

data may be analyzed by various methods, such as NPD or NLMEM (see subsection 1.1.2)

Until now preclinical studies have been rarely designed using optimal design strategies. Moreover, *in vivo* tumor size measurements in xenograft experiments are often taken only during and shortly after treatment (Nagy et al., 2015; Chang et al., 2016), possibly preventing correct identification of some parameters of tumor growth inhibition (TGI) models (Rocchetti et al., 2013). It has been proved that the time efficacy index (TEI), which is a secondary parameter of the Simeoni TGI model (Simeoni et al., 2004a; Magni et al., 2006) that is linked to the drug potency parameter, is better estimated when considering regrowth (Magni et al., 2006). Moreover, many institutions have already had the common intuition that sampling during tumor regrowth would allow for more informative studies, nevertheless, to our knowledge, this was never proven before.

Tumor growth inhibition models Several tumor growth inhibition (TGI) models have been developed for describing drug action on tumor cells. Among those, the Gompertz model (Simpson-Herren and Lloyd, 1970) was one of the first developed and largely used model and it has been successfully used for both *in vitro* and *in vivo* preclinical studies, for different tumors in various unperturbed situations, i.e. without drug administration (Bonate, 2011). The Gompertz equation can be expressed as reported in Ribba et al. (2014):

$$\frac{dy}{dt} = \alpha \cdot y \cdot \log\left(\frac{y}{\theta}\right) \quad (1.21)$$

where, y is the tumor size, α is the deceleration rate related to the natural death of the tumor cells and θ is the maximum value that y can reach. In perturbed scenarios, the Gompertz model can be used for the net growth process, i.e. the difference between growth and natural death, in which the saturation size (θ) varies as a function of different parameters, especially of a drug effect parameter (Ribba et al., 2014).

TGI models can be divided in three categories, based on their formulation (Ribba et al., 2012): i) models with structure similar to E_{max} model, i.e. models that assume first-order exponential net growth process with a term for the drug-induced decay (e.g. Gompertz model), which is function of the concentration; ii) models with a second compartment that has back-and-forth exchange with the first compartment, based on the assumption that the drug acts on a tumor cells fraction; iii) models with four transit compartments, which are introduced for accounting on the assumption of a delay in the action of the drug. Of note, there is lack of experimental data supporting the assumption for formulation ii).

A model that is presently widely used by industry for the *in vivo* evaluation of antitumor effect in xenograft experiments, is the Simeoni tumor growth inhibition (TGI) model (Simeoni et al., 2004a, 2013).

The Simeoni TGI model is a pharmacokinetic-pharmacodynamic (PKPD) model in which the dose of an anticancer compound is linked, through concentration, to the inhibition of tumor growth. It has been used for analysis of hundreds of single-agent

experiments in preclinical studies in the industry (Simeoni et al., 2004a; Terranova et al., 2013) and was recently used to evaluate interaction effects when drugs are administered in combination (Terranova et al., 2013). Figure 1.6 displays tumor growth curves related to control and treated arms. The Simeoni model is further described in Chapter 2.

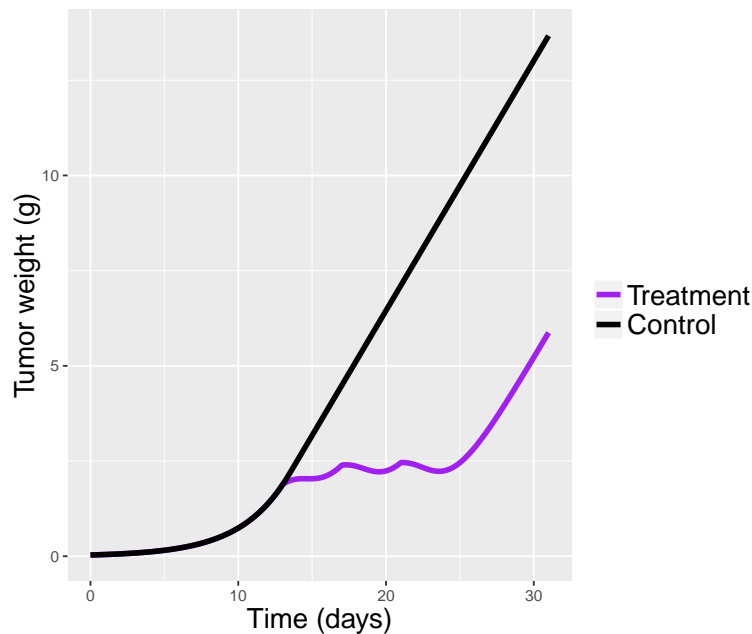


Figure 1.6: Simeoni tumor growth inhibition model. *Black* and *purple* curves are the tumor weight (g) over time in control arm and treatment arm, respectively.

1.3.2 Tumor growth models in clinical studies

The Response Evaluation Criteria in Solid Tumors (RECIST) is used as guideline in solid tumor clinical trials (Eisenhauer et al., 2009). The main issue is that RECIST criteria evaluation is performed at discrete time points, not allowing for taking into account the dynamic of tumor growth, treatment-related shrinkage, and development of resistance (Ribba et al., 2014). Indeed, according to the US Food and Drug Administration Critical Path Initiative, modeling should be used for taking better decisions in drug development (U.S. Food and Drug Administration, 2004).

Several tumor growth models have been developed with algebraic equations for describing the tumor size curve as a combination of linear growth and exponential decay (Wang et al., 2009; Stein et al., 2011; Bonate and Suttle, 2013). Other models were expressed by differential equations, such as the Gompertz model (Simpson-Herren and Lloyd, 1970). Gompertz model, described in subsection 1.3.1, was also applied to clinical studies, such as a myeloma study (Sullivan and Salmon, 1972) and a survival study to analyze data of untreated breast cancer female patients (Norton, 1988).

As reported by Ribba et al. (2014), in clinical studies, longitudinal analysis of the tumor size in eight different therapeutic indications were proposed since 2008. These

are: colorectal cancer (Bruno and Claret, 2009; Claret et al., 2013), non-small cell lung cancer (Tham et al., 2008; Wang et al., 2009), renal cell carcinoma (Houk et al., 2010; Stein et al., 2012; Maitland et al., 2013; Bonate and Suttle, 2013), thyroid cancer (Claret et al., 2010), metastatic breast cancer (Frances et al., 2011), prostate cancer (Stein et al., 2011), gastrointestinal stromal tumor (Houk et al., 2010; Hansson et al., 2013), and low-grade glioma (Ribba et al., 2012).

1.3.3 Models for tumor markers and biomarkers

In certain types of cancer, tumor markers are used instead of tumor size measurements for diagnosing cancer or for monitoring the response to treatment (Bender et al., 2015). Tumor markers are produced in the body by cells that can be either or not related to cancer, and they are measured in blood. An example of marker is the prostate specific antigen (PSA) and it reaches high levels in men with prostate cancer. You et al. (2009) used population models to describe PSA levels after prostatectomy. Other markers are the M-protein in myeloma, cancer antigen 125 (CA-125) in ovarian cancer and carcinoembryonic antigen (CEA) in colorectal cancer. A joint model for tumor size and CA-125 kinetics was developed by Wilbaux et al. (2014).

Biomarkers are molecules that are present in the blood or other fluids, and they can be used to assess how the body has reacted to treatment. Turnover models (Sharma and Jusko, 1998) are often used for characterizing the PD when the effect is produced by an indirect mechanism of the drug that induces the production or inhibition of some factors, i.e. biomarkers, which directly control the effect.

1.3.4 Other models in oncology

Adverse effects occurring during a therapy can also be modeled (Trocóniz et al., 2012). One important example is the myelosuppression model developed by Friberg et al. (2002). Myelosuppression is considered as adverse effect for the majority of cancers, and it is caused by certain cancer treatments. The model can be freely downloaded on the Drug Disease Model Resources (DDMoRe) repository (<http://repository.ddmore.eu/model/DDMODEL00000186>).

To conclude, all the former models presented, such as TGI models, turnover models and those for the adverse effects, can be used to predict the overall survival (OS), by linking these metrics to time to event (TTE) models (Bender et al., 2015). A nice example is the one by Desmée et al. (2015) who proposed NLMEMs for prostate-specific antigen kinetics linked to OS in metastatic prostate cancer.

1.4 Aims

In this thesis we aimed at evaluating robust and adaptive designs in pharmacometrics with application in oncology. Our goal was to analyze optimal setups for these trials based on models developed on previous knowledge on the disease and the respective drug. We also aimed at applying optimal design to preclinical studies in oncology to allow for a better translation into clinical studies.

Chapter 1 was dedicated to the introduction to the basic concepts of pharmacometrics, optimal design in pharmacometrics and pharmacometrics in oncology.

In Chapter 2 a proof of concept study about optimal design in xenograft experiments is presented. Indeed, xenografts experiment were rarely conducted using optimal design strategies. One relevant objective of that project was to use optimal design in several xenograft experiments that were already published in (Simeoni et al., 2004a).

Chapter 3 is dedicated to an adaptive design simulation study, where the aim was to compare different cohort sizes of two-stage designs for an oncology example, but also to investigate the gain of performing more than one adaptation.

In Chapter 4 a comparison study of robust design criteria is illustrated, with an evaluation for two examples with longitudinal clinical data. In that study we aimed at comparing all the robust design criteria existing in the literature in the context of NLMEM, and to apply optimal and robust designs to a NLMEM with discrete outcome. To our knowledge, it was never performed before.

As this thesis was supported by the drug disease model and resources (DDMoRe), part of the PhD was dedicated to help integrating the optimal design software developed in the lab in Paris (IAME UMR-S 1137, INSERM and Université Paris Diderot) to the DDMoRe platform. This work is described in Chapter 5.

Finally, in Chapter 6 the overall conclusions and perspectives of this thesis are drawn.

Chapter 2

Optimal design in xenograft experiments

2.1 Summary

Experimental models in rodents have been developed in recent decades to evaluate *in vivo* the antitumor effect of a new compound (Bernard et al., 2012). Among *in vivo* preclinical studies, the most popular is the xenograft model (Kelland, 2004), that is the human tumor transplanted into animals. When the tumor has reached the desired volume, mice are randomized into control and treatment arms. Tumor sizes are measured at repeated time points during the study and specific metrics are then calculated to assess tumor activity in the different arms (Bissery et al., 1996).

The first objective of this study was to assess quantitatively in two-arm experiments, *i.e.* one control and one treatment arm, the difference in terms of precision of parameter estimation of taking samples only during and shortly after the treatment administration, or also later, during the tumor regrowth phase, by using standard nonlinear regression or NLMEM. The second objective of this work was to investigate the influence of including arms with different doses or different drug administration schedules in the trials, as well as the number of treated and control mice to be used in the experiments, when using NLMEMs.

We used the Simeoni tumor growth inhibition (TGI) model. Ten xenograft experiments were selected from the literature (Simeoni et al., 2004a,b) involving various drugs with various modes of action, different drug administration schedules and cell lines. For each experiment two scenarios were considered: a "short study", in which it was possible to measure only until three days after the end of the treatment period in order to mimic the frequently adopted setting of those preclinical studies, and a "long study", in which measurements could be taken also later, after the end of the treatment period, during the tumor regrowth phase, but for ethical reasons only up to a tumor weight of 6 grams. Optimization of sampling times from the allowed sampling window was performed simultaneously in control and treatment arms for each selected experiment and for short and long studies. For the second objective, optimal design was applied in the context of four-arm designs, *i.e.* one control and three treatment arms, with either different doses per arm or different drug administration schedules per arm.

The optimal design approach used in this study was the one based on the Fisher information matrix (FIM), and the D-optimality criterion was chosen as the optimal design metric. To compute FIM, first-order (FO) linearization method was used from the PFIM program and optimal measurement times were found using the Fedorov-Wynn algorithm, also implemented in PFIM.

We found that in long studies, some optimal times were always located in the regrowth phase of treatment arms, highlighting the importance of continuing the experiment after the end of the treatment. These results were obtained for all two-arm experiments, i.e. one treatment and one control arm, either when only one subject was assigned per arm, or when more mice were included in each arm. Moreover, predicted relative standard errors (%) of the Simeoni TGI model parameters obtained in long studies were better than those obtained in the short study of the corresponding experiment. Making measurements during tumor regrowth should become a general rule for informative preclinical studies in oncology, independently of adopting standard nonlinear regression or an NLMEM approach. In the four-arm designs, the results showed that the proportions of control and treated mice can differ.

This work was a first attempt to provide some general guidelines for preclinical xenograft experiments and proved that optimal design can help us perform more informative preclinical tumor growth inhibition studies, by measuring tumor growth for longer after the end of treatment, thus ensuring more precise model parameter estimation and hence better translation into clinical studies.

2.2 Article I (published)

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Research Article

Optimal Design for Informative Protocols in Xenograft Tumor Growth Inhibition Experiments in Mice

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Abstract. Tumor growth inhibition (TGI) models are increasingly used during preclinical drug development in oncology for the *in vivo* evaluation of antitumor effect. Tumor sizes are measured in xenografted mice, often only during and shortly after treatment, thus preventing correct identification of some TGI model parameters. Our aims were (i) to evaluate the importance of including measurements during tumor regrowth and (ii) to investigate the proportions of mice included in each arm. For these purposes, optimal design theory based on the Fisher information matrix implemented in PFIM4.0 was applied. Published xenograft experiments, involving different drugs, schedules, and cell lines, were used to help optimize experimental settings and parameters using the Simeoni TGI model. For each experiment, a two-arm design, *i.e.*, control *versus* treatment, was optimized with or without the constraint of not sampling during tumor regrowth, *i.e.*, “short” and “long” studies, respectively. In long studies, measurements could be taken up to 6 g of tumor weight, whereas in short studies the experiment was stopped 3 days after the end of treatment. Predicted relative standard errors were smaller in long studies than in corresponding short studies. Some optimal measurement times were located in the regrowth phase, highlighting the importance of continuing the experiment after the end of treatment. In the four-arm designs, the results showed that the proportions of control and treated mice can differ. To conclude, making measurements during tumor regrowth should become a general rule for informative preclinical studies in oncology, especially when a delayed drug effect is suspected.

KEY WORDS: Fisher information matrix; oncology; optimal design; pharmacodynamic; tumor growth inhibition models.

INTRODUCTION

The *in vivo* evaluation of antitumor effect is an important step of preclinical drug development in oncology. Experimental models in rodents have been developed in recent decades to assess the antitumor effect of a new compound (1). Among *in vivo* preclinical studies, the most popular is the xenograft model (2), that is the human tumor transplanted into animals. Most recent xenograft experiments have been performed on athymic nude mice (3), which are mice with reduced capacity to reject foreign cells. The common practice is either to implant tumor fragments subcutaneously or to inject tumor cells into the flank of athymic mice (4). When

the tumor has reached the desired volume, mice are randomized into control and treatment arms receiving either a vehicle (control arm) or the compound to be tested at different dose levels or with different drug administration schedules (treatment arms). Tumor sizes are measured at repeated time points during the study and specific metrics are then calculated to assess tumor activity in the different arms (5). The main purpose of those studies is to quantify the inhibition of tumor growth provided by the treatment, which depends on dose, time, and dosing regimen (4, 6). For this reason, mathematical models able to dissect system-related from drug-related parameters have been proposed to describe and predict the antitumor effect of a new compound. We chose to use the Simeoni tumor growth inhibition (TGI) model (7, 8) for *in vivo* evaluation of antitumor effect in xenografted mice. The Simeoni TGI model is a pharmacokinetic-pharmacodynamic (PKPD) model in which the dose of an anticancer compound is linked, through concentration, to the inhibition of tumor growth. By searching for papers that cite the model and looking at author affiliations, we observed that the model is

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widely used in both industry and academia. It has been used for analysis of hundreds of single-agent experiments in preclinical studies in the industry (4, 9) and was recently used to evaluate interaction effects when drugs are administered in combination (9). Xenograft data may be analyzed by various methods. One method is the “naïve pooled data” (NPD) approach (10) in which the model can be fitted using standard nonlinear regression. In this approach parameter estimation is straightforward but does not model all sources of variability, so no information about the differences between subjects is considered. Nonlinear mixed effects models (NLMEMs) (11–13) are a popular alternative approach. This method is more complex than NPD, but has been increasingly used in the biomedical field as it keeps all the data of all subjects.

Before performing the experiment, it is important to define an appropriate design, which implies there has to be a good balance between the number of subjects, the number of samples per subject, and the timing of sampling and the doses, in line with the experimental conditions. As reported in (4), until now, preclinical studies have been rarely designed using optimal design strategies. When planning a study, it is crucial to choose a good design as it can play an important part in parameter estimation (14), and poor design can lead to inconclusive studies. As recommended in (4), designing experiments with optimal design strategies would lead to more informative studies and ensure more precise parameter estimates for the selected model.

There are two main approaches to design evaluation. The first, by trial simulation, involves parameter estimation using numerous simulated datasets and thus is a time-consuming method (15). The second approach, which avoids simulation, relies on the Cramer-Rao inequality, which states that the inverse of the Fisher information matrix (FIM) is the lower bound of the variance-covariance matrix of any unbiased parameter estimate. Several criteria based on FIM have been proposed. One is the D-optimality criterion, which consists in maximizing the determinant of FIM, considering all model parameters. In NLMEMs, there is no closed form of the likelihood and thus of FIM; therefore, an approximation method must be used. Mentré *et al.* (16) first introduced the first-order (FO) linearization method and further approximations have been developed since. FIM is implemented in several optimal design software programs for NLMEMs (17, 18). These are as follows: PFIM (19, 20), PopED (21), PopDes (22), and POPT (23). These tools allow for design evaluation/optimization by computing FIM and the D-optimality criterion (24), and it was shown in (17) that they all provide the same answer when using the same FO approximation of FIM.

The aim of this study was thus to apply optimal design strategies to the xenograft experiments analyzed with the Simeoni TGI model, to derive some general rules that could be used as guidelines for future preclinical studies in this field,

and to assess the impact of suboptimal design. More specifically, *in vivo* tumor size measurements in xenograft experiments are often taken only during and shortly after treatment (25, 26), possibly preventing correct identification of some parameters of TGI models (27). The first objective of this study was to assess quantitatively in two-arm designs, *i.e.*, one control and one treatment arm, the difference in terms of precision of parameter estimation of taking samples only during and shortly after the treatment administration, or also later, during the tumor regrowth phase, by using standard nonlinear regression or an NLMEM. The main idea behind this thought is that the time efficacy index (TEI), which is a secondary parameter of the Simeoni TGI model that is linked to the drug potency parameter, is estimated better when considering regrowth (8). Although many institutions have already had the common intuition that sampling during tumor regrowth would allow for more informative studies, to our knowledge, this was never proven before.

The second objective of this work was to investigate the influence of including arms with different doses or different drug administration schedules in the trials, as well as the number of treated and control mice to be used in the experiments, when using NLMEMs.

MATERIALS AND METHODS

Experimental Settings

Several xenograft experiments were selected from the literature (7, 28) involving various drugs with various modes of action, different drug administration schedules, and cell lines. The compounds tested in the experiments considered were paclitaxel and 5-fluorouracil (5-FU), which are available on the market, and two drugs (drug A and drug B) synthesized by Pharmacia (7). The A2780 human ovarian carcinoma and HCT116 colon carcinoma cell lines were used. Tumor fragments were subcutaneously implanted into the left flank of athymic nude mice and, one week after tumor inoculation, mice bearing a palpable tumor were randomized into control and treatment arms. Tumor dimensions were measured using calipers and the tumor masses w were calculated at each time t with the following formula reported in (7):

$$w = \frac{l \cdot z}{2} \rho \quad (1)$$

where l is the length of the tumor measured in millimeters and z is the tumor squared width expressed in square millimeters. It was assumed that the density ρ was equal to 1 mg/mm³ for tumor tissue such that the tumor weight w is expressed in milligrams.

A schematic description of selected experiments is reported in Table I. Experimental settings for experiments 1, 2, 9, and 10 were taken from (28), whereas the information for the other experiments was found in (7).

Optimal Design in Tumor Growth Experiments in Mice

Table 1. Experimental and Optimal Designs Settings and Results for “Long” (First Row) and “Short” (Second Row) Studies in the Two-Arm Designs, i.e., Control and Treatment Arm, Selected from Reference (7) and (28)

Experimental settings										Optimal design settings and results									
Exp	Drug	Daily Dose (mg/kg)	Treatment		Dosing frequency	TGI Parameters ^a			Ref	n	Sampling window treatment/control arms (days)	Optimal designs in treatment arm (days)	Optimal design in control arm (days)						
			Starting day	Last day		Fixed effects ($\lambda_0, \lambda_1, w_0, k_1, k_2$)	Variance of random effects ($\omega_0^2, \omega_1^2, \omega_{k_1}^2, \omega_{k_2}^2$)	St. deviation of residual error ($\sigma_{inter}, \sigma_{slope}$) ^b											
1	Paclitaxel	30	8	16	every 4 days	0.238, 0.14, 0.049, 0.117, 6.3910 ⁻⁴	0.138, 0.063	(28)	1	8-68/8-48	8, 28, 63, 68	8, 12, 44, 48							
2	Drug B	120	9	12	bid	0.269, 0.397, 0.022, 0.631, 2.7210 ⁻⁴	0.118, 0.07	(28)	1	9-33/9-27	8, 12, 18, 19	8, 11, 18, 19							
3	Paclitaxel	30	13	21	every 4 days	0.311, 0.656, 0.033, 0.968, 6.2910 ⁻⁴	0, 0.063	(7)	1	8-15/8-15	9, 15, 23, 33	8, 9, 14, 15							
4	Paclitaxel	30	8	16	every 4 days	0.273, 0.814, 0.055, 0.968, 6.2910 ⁻⁴	0.138, 0.063	(7)	1	9-31/9-19	9, 15, 23, 27	9, 13, 18, 19							
5	5-FU	50	8	29	every 4 weeks	0.215, 0.412, 0.065, 0.056, 20.2110 ⁻⁴	0.138, 0.063	(7)	1	8-30/8-18	14, 18, 26, 30	8, 9, 15, 18							
6	Drug A	15	9	19	qd	0.349, 0.363, 0.010, 0.405, 3.4510 ⁻⁴	0.118, 0.07	(7)	1	8-47/8-24	8, 23, 41, 47	8, 9, 15, 18							
7	Drug B	30	13	17	bid	0.309, 0.796, 0.034, 0.517, 2.8910 ⁻⁴	0, 0.07	(7)	1	9-37/9-26	8, 24, 28, 32	8, 12, 22, 24							
8	Drug B	83	9	15	7-day infusion	0.369, 0.511, 0.016, 0.615, 2.9310 ⁻⁴	0, 0.063	(7)	1	9-22/9-22	9, 10, 19, 22	9, 13, 25, 26							
9	Paclitaxel	30	8	16	every 4 days	0.238, 0.14, 0.049, 0.117, 6.3910 ⁻⁴	0.138, 0.063	(28)	4	9-20/9-18	9, 15, 19, 23	9, 14, 17, 18							
10	Drug B	120	9	12	bid	0.269, 0.397, 0.022, 0.631, 2.7210 ⁻⁴	0.118, 0.07	(28)	7	8-32/8-20	8, 18, 22, 24	8, 12, 18, 20							
							0.002, 0.177, 0.053, 1.580			8-18/8-18	8, 16, 17, 18	8, 12, 17, 18							
							2.080, 0.360			8-68/8-48	27, 44, 48, 68	8, 11, 12, 49							
										8-19/8-19	10, 16, 22, 33	11, 12, 18, 19							
										9-33/9-27	11, 13, 14, 15	10, 16, 26, 27							
										8-15/8-15	11, 13, 14, 15	11, 13, 14, 15							

For experiments 1–8, an individual approach was used therefore $n=1$ mouse was included in each arm. For the population experiments 9 and 10, $n=4$ and $n=7$ mice were included in each arm, respectively. The route was i.v. bolus for all experiments except for experiment 8, which was i.v. infusion. Inter individual variability on parameter w_0 was set to 0
^aTGI parameters units are as follows: $\lambda_0(\text{day}^{-1}), \lambda_1(\text{gday}^{-1}), w_0(\text{g}), k_1(\text{day}^{-1}), k_2(\text{ng}^{-1} \text{ml day}^{-1})$
^bParameter values for σ_{inter} and σ_{slope} were only available for the experiments in reference (28); therefore, they were arbitrarily defined for the experiments from (7)

Tumor Growth Inhibition Model

The Simeoni TGI model is built based on the assumption that tumor growth follows an exponential curve in the early phases followed by linear growth (5, 7). The unperturbed growth model for untreated mice is expressed with the following differential equation (7):

$$\frac{dw(t)}{dt} = \frac{\lambda_0 \cdot w(t)}{\left[1 + \left(\frac{\lambda_1}{\lambda_0} \cdot w(t)\right)^\psi\right]^{1/\psi}} \quad (2)$$

$$w(0) = w_0$$

where w_0 is the tumor weight at inoculation time, and λ_0 and λ_1 are parameters describing the rate of exponential and linear growth, respectively. The parameter ψ is set to 20 ensuring a sharp system transfer from first-order growth (exponential) to zero-order growth (linear) (7). With this model, if the tumor weight $w(t)$ is small, the denominator of Eq. 2 can be approximated to 1 and then the tumor growth is given by $\lambda_0 \cdot w(t)$; whereas when $w(t)$ is large, 1 can be neglected in the denominator and the tumor growth is approximated by λ_1 . The system-related parameters of the unperturbed model are therefore: λ_0 , λ_1 , and w_0 .

In the treatment arms, the action of the antitumor compound prevents the proliferation of a proportion of tumor cells, which go through a three-stage chain at a constant rate of damage k_1 , with different levels of impairment, and then, eventually, die. The antitumor potency is characterized by the constant parameter k_2 , which is proportional to $c(t) \cdot x_1(t)$, where $c(t)$ is the drug concentration, which can be predicted with a PK model, and $x_i(t)$ are the proliferating cells in $w(t)$. A schematic representation of the perturbed growth model is reported in Fig. 1.

The perturbed growth model for treated mice is described with the following system of differential equations (7):

$$\frac{dx_1(t)}{dt} = \frac{\lambda_0 \cdot x_1(t)}{\left[1 + \left(\frac{\lambda_1}{\lambda_0} \cdot w(t)\right)^\psi\right]^{1/\psi}} - k_2 \cdot c(t) \cdot x_1(t)$$

$$\frac{dx_2(t)}{dt} = k_2 \cdot c(t) \cdot x_1(t) - k_1 \cdot x_2(t)$$

$$\frac{dx_3(t)}{dt} = k_1[x_2(t) - x_3(t)] \quad (3)$$

$$\frac{dx_4(t)}{dt} = k_1[x_3(t) - x_4(t)]$$

$$w(t) = x_1(t) + x_2(t) + x_3(t) + x_4(t)$$

$$c(t) = 0 \quad \text{for } 0 < t \leq t_0$$

where t_0 represents the time of beginning of treatment. Before the start of treatment, the concentration $c(t)$ is equal to 0 and the model follows an unperturbed growth, with $w(t) = x_1(t)$. After time t_0 , the model follows a perturbed growth and $w(t) = x_1(t) + x_2(t) + x_3(t) + x_4(t)$.

The parameters of the perturbed growth model therefore are: λ_0 , λ_1 , w_0 , and k_1 , k_2 , which are the system-related and drug-related parameters, respectively.

For each proposed experiment, a PK model was used in order to predict the $c(t)$ needed for the treatment arm in the TGI model. The PK model and its parameter

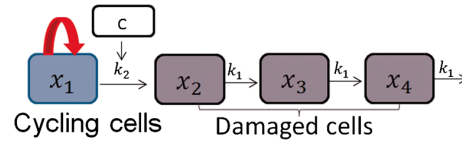


Fig. 1. Schematic representation of the Simeoni TGI model. The first box represents the proliferating cells for which the tumor growth is characterized by an exponential growth followed by a linear growth. k_2 is the parameter related to drug potency, C is the plasma concentration of the anticancer agent and k_1 is the parameter related to the rate of death of the tumor cells

values were taken from the literature (7, 28). For experiments 1 to 7 of Table I, a two-compartment model with intravenous (i.v.) bolus was used, whereas in experiment 8, a two-compartment model with continuous i.v. infusion was used.

Figure 2 shows the simulated PK model and TGI model for experiments 1 and 9 of Table I.

Statistical Model

In standard nonlinear regression, the vector y_i of observations y_{i1}, \dots, y_{in_i} measured from the subject i at times $\xi_i = (t_{i1}, \dots, t_{in_i})$ is defined with $y_i = f(\theta_i, \xi_i) + \varepsilon_i$ where f is the nonlinear structural model describing the tumor growth over time and ξ_i is the elementary design composed of n_i sampling times. θ_i is the vector of p individual parameters and ε_i is the vector of residual error following a normal distribution $\varepsilon_i \sim N(0, \Sigma(\theta, \xi_i))$ with $\Sigma(\theta, \xi_i) = \text{diag}(\sigma_{\text{inter}} + \sigma_{\text{slope}} \times f(\theta_i, \xi_i))^2$. σ_{inter} is the parameter for the additive error and σ_{slope} for proportional error. The individual parameters vector is $\gamma' = (\theta_i, \sigma_{\text{inter}}, \sigma_{\text{slope}})$.

In NLMEMs, all samples of all individuals are analyzed simultaneously. The individual parameters θ_i for the i^{th} subject is modeled as $\theta_i = \mu e^{\eta_i}$ with a fixed effect μ , which represents the median values of the parameter in the population, and a random effect $\eta_i \sim N(0, \Omega)$, where $\Omega = \text{diag}(\omega_1^2, \dots, \omega_p^2)$ accounts for the between-subject variability. The population parameter vector is $\Phi' = (\mu_1, \dots, \mu_p, \omega_1^2, \dots, \omega_p^2, \sigma_{\text{inter}}, \sigma_{\text{slope}})$.

All parameter values used in this article are given in Table I and were estimated from real experiments in (7) for experiments 3 to 8, and in (28) for experiments 1, 2, 9, and 10. Moreover, experiments 1 and 2 have the same mean parameter values as experiments 9 and 10, respectively, and experiments A and B in Table II have same TGI model parameters as those of experiments 9 and 10 in Table I.

Optimal Design Strategy

Criterion and Optimization

For optimal design, we used the D-optimal criterion, which corresponds to the maximization of the determinant

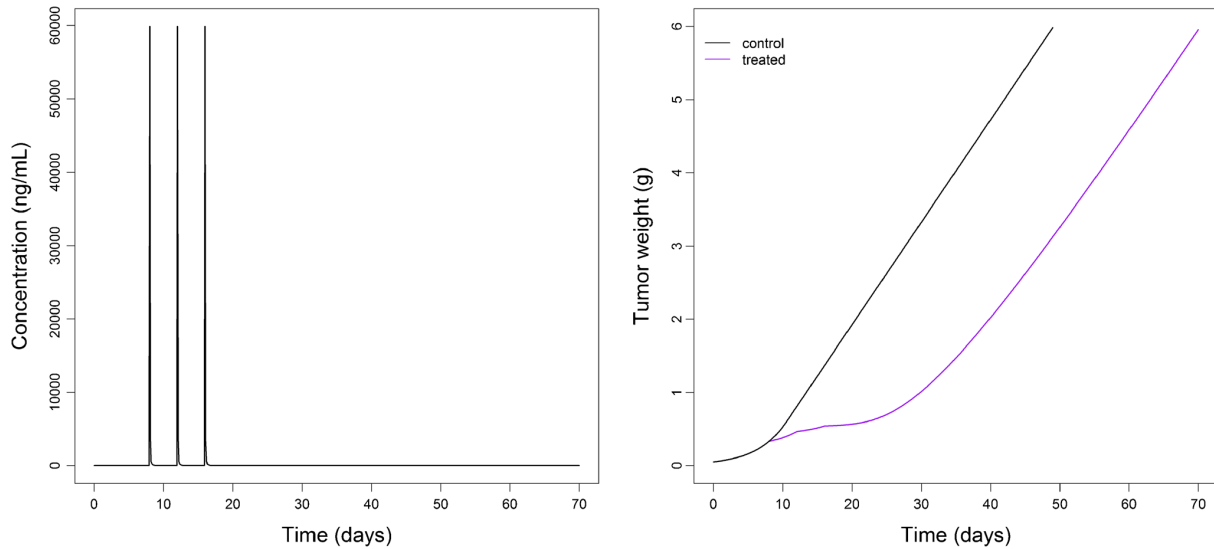


Fig. 2. Simulated PK (left) and PD (right) for experiments 1 and 9 of Table I. The PK is a two compartment model i.v. bolus with multiple doses administration (of 30 mg/kg given every 4 days for three times). The PD is the TGI model for a two-arm experiment with control arm (black curve) and treatment arm (purple curve)

of the individual or population FIM, corresponding to the minimization of the confidence region associated with the individual or population parameter estimates. The reported optimality criterion is the determinant to the power $1/p$, where p is the total number of estimated parameters in the model. A search for optimal measurement times was performed using the Fedorov-Wynn algorithm implemented in the R function PFIM, in the PFIM 4.0 program (19, 29), in which both individual and population FIMs can be computed.

Together with the optimal design, PFIM returns the predicted relative standard error (pRSE) in percent for each model parameter, and pRSE is defined as:

$$pRSE(p) = \frac{SE(\hat{\phi})}{\phi} \times 100 \quad (4)$$

where $SE(\hat{\phi})$ is the expected standard error of parameter ϕ .

Two-Arm Designs

All experiments described in Table I were used for the first objective of the present study, that is to assess quantitatively the difference in precision of parameter estimates in two-arm designs, when taking four measurement samples only during and shortly after the treatment period, compared with taking measurements also later, in the tumor regrowth phase. In experiments 1 to 8, only one mouse per arm was included in order to perform individual design optimization. This can be viewed as an NPD approach where mean data are fitted, hence assuming that they come from a single mouse. In experiments 9 and 10, four and seven mice were assigned to each arm, respectively, such that population design optimization could be computed.

For each experiment, two scenarios were thus considered: a “short study,” in which it was possible to measure only until 3 days after the end of the treatment period in order to mimic the frequently adopted setting of those preclinical

Table II. Experimental and Optimal Designs Settings and Results in Four-Arm Designs from Reference (28)

Experimental settings					Optimal design settings and results							
Exp	Drug	Daily Dose (mg/kg)	Treatment		Dosing frequency (route: i.v. bolus)	Allowed sampling window (days)	Optimal design (scenario 1)			Optimal design (scenario 2)		
			Starting day	Last day			Optimized times (days)	n (optimized)	D-optimal criterion	Optimized times (days)	n (fixed)	D-optimal criterion
A	Paclitaxel	0			every 4 days	8–48	11, 26, 29, 41, 47, 68	6	2100	8, 11, 14, 17, 47, 49	4	1924
		20	8	16	every 4 days	8–68		0		8, 11, 14, 32, 56, 68	4	
		30	8	16	every 4 days	8–68		0		8, 11, 23, 44, 47, 68	4	
		40	8	16	every 4 days	8–68	8, 11, 14, 17, 47, 49	10		11, 23, 26, 29, 41, 68	4	
B	drug B	0			bid	9–27	9, 11, 17, 23, 25, 27	10	4708	9, 10, 11, 16, 26, 27	7	4643
		60	9	12	bid	9–33	9, 11, 15, 23, 31, 33	9		10, 11, 16, 22, 32, 33	7	
		60	9	9	tid	9–29		0		9, 12, 14, 20, 28, 29	7	
		60	9	19	qd	9–35	9, 11, 13, 21, 27, 35	9		11, 14, 22, 27, 34, 35	7	

TGI parameters for experiment A and B are those of experiment 9 and 10 of Table I, respectively

studies, and a “long study,” in which measurements could be taken also later, after the end of the treatment period, during the tumor regrowth phase, but for ethical reasons only up to a tumor weight of 6 g. Note that 6 g is an overestimated threshold adopted to emphasize the impact on precision of parameter estimates in short *versus* long studies. See the “DISCUSSION” section for more details. The two scenarios therefore only differ in the chosen range of allowed sampling times, in the optimal design settings. The parameters were used to simulate the tumor weight with and without treatment for each experiment, to define the time when a tumor weight of 6 g was reached. Allowed sampling time ranges for the two scenarios are reported in Table I under the “Sampling window” column for both treatment and control arms, first line for the short study and second line for the long study. Note that in the long study, treatment arms should have larger allowed sampling time windows compared with those for the corresponding control arms, as treated mice reached a tumor weight of 6 g later, because of the treatment effect.

The allowed sampling times were chosen based on the time ranges defined for each experiment. In short studies, a dense grid for each day in the range was set as the allowed sampling times, except for experiment 5 in which the allowed times were allocated every other day. In long studies, the allowed sampling times were allocated every other day, if the last time in the range was before day 38, every 3/4 days if the last time in the range was before day 50 and every 4 or 5 days for the long studies of experiments 1 and 9, in which the last allowed time was day 68.

Optimization of sampling times from the allowed sampling window was performed simultaneously in control and treatment arms for each selected experiment and for short and long studies. In the individual design optimization, for experiments 1 to 8, the model was implemented as a two-response model, each response corresponding to one arm. For experiments 9 and 10 and population design optimization, as the Fedorov-Wynn algorithm would also optimize the proportion of subjects per arm, two steps were used to get the optimal times with a fixed number of mice per arm. The model was implemented as a single response model with a two-arm design. As currently in PFIM, the same allowed sampling window should be set for the two arms—the treatment arm window was chosen, as it includes all the possible times for both arms. Optimal design was performed and the optimal times obtained were stored. For the control arm, optimal times exceeding the allowed range were replaced by the last possible times. We then performed two-arm design evaluation of all possible combinations of four times among the optimal times found in the previous step for a fixed number of mice in each arm. The two-arm design with the combination that led to the highest criterion was defined as the optimal design.

pRSE (%) were used to compare long and short studies in all the experiments, knowing that the lower pRSE (%) would guarantee the more precise parameter estimates for the study considered.

Graphs of sensitivity functions, *i.e.*, partial derivatives of the Simeoni TGI model prediction with respect to each parameter, were used to visualize the impact of each model parameter through time. Sensitivity functions with respect to each parameter provide knowledge on the design values (*e.g.*,

tumor weight measurement time) leading to the greatest information, *i.e.*, the times for which the square of the sensitivity function is maximal. Figure 3 displays the graph of sensitivity functions for experiments 1 and 9. The PFIM input and output files for experiment 1 are reported in the supplementary material.

Four-Arm Designs

The second objective of this study was to apply optimal design in the context of four-arm designs, *i.e.*, one control and three treatment arms, with (i) different doses per arm or (ii) different drug administration schedules per arm. The experiments used for this purpose were experiments A and B reported in Table II, which were analyzed by an NLMEM. Experiment A had three different doses of 20, 30, and 40 mg/kg in each treatment arm, respectively, whereas experiment B had three different drug administration schedules: *i.v.* bolus bid, *i.e.*, twice a day, in treatment arm 1; *i.v.* bolus tid, *i.e.*, three times a day, in treatment arm 2; and *i.v.* bolus qd, *i.e.*, once daily, in treatment arm 3. The allowed sampling time ranges for treatment and control arms were defined as before, using the criterion of stopping the study when the tumor weight reached 6 g.

In a first scenario, six sampling times from the allowed sampling times were optimized with the Fedorov-Wynn algorithm. The initial design for experiment A consisted of four mice per arm at six times; 8, 17, 20, 26, 29, 35 days, arbitrarily chosen. The same was true for experiment B, where the initial design consisted of seven mice per arm at the six times: 11, 13, 15, 17, 19, 21 days. Using the Fedorov-Wynn algorithm in PFIM for optimization could lead to some arms being dropped from the experiment. The model was implemented as a one-response model and in a first scenario the four-arm design was optimized using the same approach that was applied to optimize the two-arm design for experiments 9 and 10 of Table I, with the exception that the arms and number of mice per arm were those found to be optimal with the Fedorov-Wynn algorithm instead of being fixed.

As it might be useful to keep all the arms, in a second scenario a different approach was used. First, three two-arm designs were optimized (control and treatment arms 1, 2, or 3) with four mice and seven mice in each arm of experiments A and B, respectively. Optimal design of the three two-arm designs was obtained using the same approach as that used for experiments 9 and 10 of Table I. The four-arm design with the previously obtained optimal times for each arm could then be evaluated, keeping fixed the number of mice per arm.

RESULTS

Two-Arm Designs

Optimal sampling times obtained simultaneously in the control and treatment arms of all the selected experiments in short and long studies are reported in Table I. In long studies, some optimal times were located in the regrowth phase of treatment arms, highlighting the importance of continuing the experiment after the end of the treatment. These results were obtained for all two-arm experiments, *i.e.*, one treatment and one control arm, either when only one subject was assigned

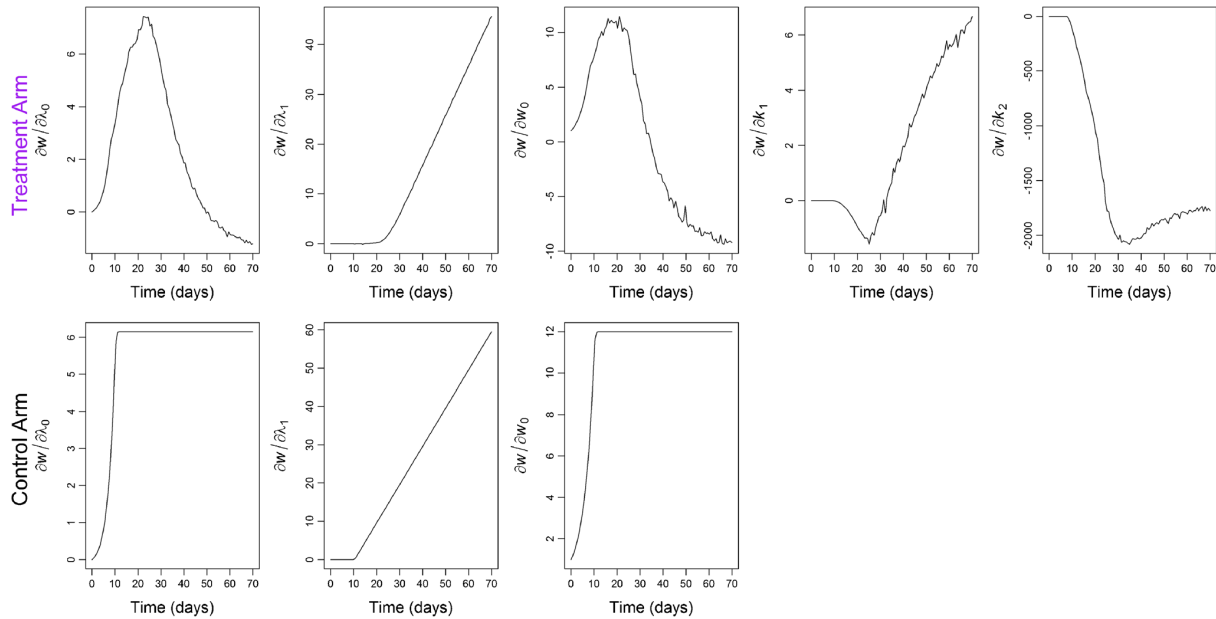


Fig. 3. Graphs of sensitivity functions of tumor weight (w) versus time for TGI parameters in treatment arm and control arm obtained for experiments 1 and 9 of Table I

per arm, as for experiments 1 to 8, or when more mice were included in each arm, as for experiments 9 and 10, using a population approach.

Graphs showing results for experiment 1 are reported in Fig. 4. In the graph on the left, it is noticeable that two out of four optimal sampling times obtained in the long study lie on the last times of the curve. In the graph on the right, it is evident that pRSE (%) of the Simeoni TGI model parameters obtained in long studies (blue bars) were better than

those obtained in the short study (red bars) of the corresponding experiment. Sensitivity graphs related to experiments 1 and 9 (Fig. 3) provide a visual check of the impact of model parameters through time for those experiments. When focusing on the fixed effects of the drug-related parameters k_1 and k_2 , it is noticeable that they start to be informative, *i.e.*, the curve stops being constant at 0, when the treatment is given (day 8), and they both reach an optimum after the end of treatment, at around day 25 and day 35, respectively.

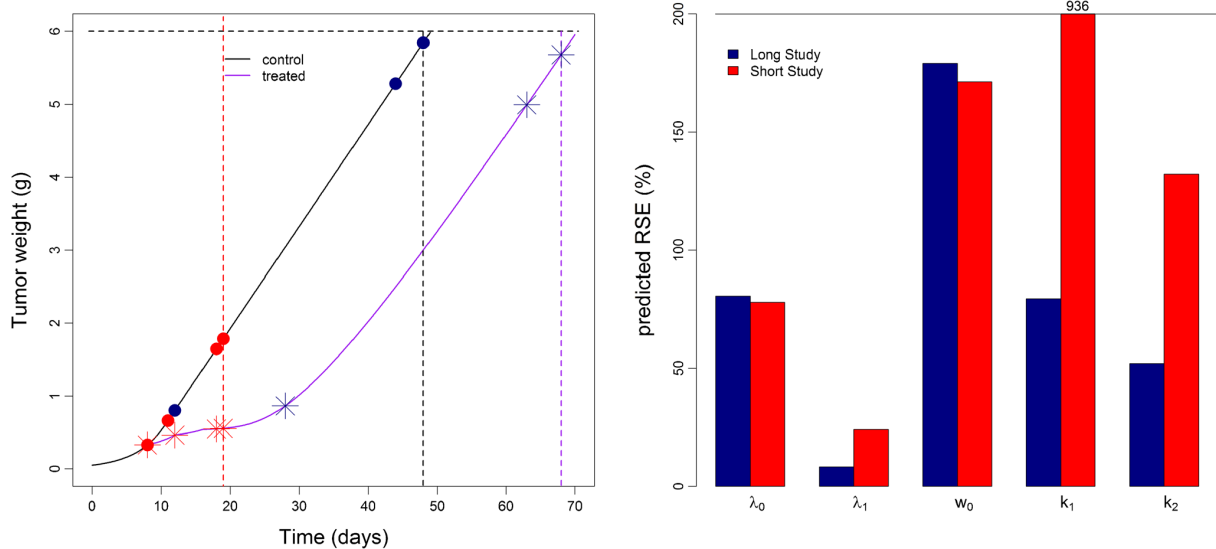


Fig. 4. Optimal times (left) in the long study (dark blue symbols) and short study (red symbols) and predicted RSE (%) in TGI model parameters (right) for experiment 1 of Table I. In the left panel red dashed lines define the end of the short study; dashed black and purple lines define the end of the long study for control and treatment arm, respectively

Similar reasoning regarding sensitivity graphs was possible for the other experiments, but it was chosen not to report all the graphs in the article.

Ratios of pRSE (%) from short to long studies (Fig. 5) were calculated for all experiments reported in Table I for the treatment-related parameters k_1 and k_2 . The ratio was greater than 1 in all experiments, proving that pRSE (%) in long studies were better than the corresponding pRSE (%) in short studies.

Four-Arm Designs

For experiment *A* in Table II, not surprisingly, it was found that the highest dose (40 mg/kg) gave the greatest information, indeed although the initial design had four arms with four mice per arm, the design obtained after the optimization of FIM was a two-arm design with six control mice, 37.5% of the total number, and ten treated mice, 62.5% of the total number, at the highest dose (40 mg/kg). The criterion obtained for the optimal design was 2100, which is much higher than the criterion obtained for the initial design of 1255, *i.e.*, an increase of efficiency of 1.67. The criterion obtained for the four-arm design with four mice per arm was 1924, *i.e.*, a small loss of efficiency of 0.92 from the optimal design, but a gain of efficiency of 1.53 from the initial design. An evaluation of the first optimal two-arm design was performed including eight mice in each arm to investigate the loss of having 50% of control and treated mice instead of 37.5% and 62.5%, respectively. The criterion found was 2049, *i.e.*, a small loss of efficiency of 0.98. Graphs showing the initial design, the two-arm design and the four-arm design are in Fig. 6, top panel.

For experiment *B*, the criterion obtained with the initial design was 2765 and the optimal design obtained was a three-arm design, thus only one arm was “excluded” from the optimal design: treatment arm 2 with tid treatment. The proportions of treated and control mice were approximately the same as for the first example: 64% treated, of which nine mice in both treatment arms 1 and 3, and 36% controls, which is ten mice. The criterion obtained was 4708, *i.e.*, an increase of efficiency of 1.70. The criterion obtained for the four-arm design with seven mice per arm was 4643, *i.e.*, a small loss of efficiency of 0.99 from the optimal design, but a gain of efficiency of 1.68 from the initial design. Graphs showing the

initial design, the three-arm design, and the four-arm design are in Fig. 6, bottom panel.

DISCUSSION

Optimal design strategies have rarely been applied to experiments concerning the *in vivo* evaluation of antitumor effect in xenografted mice (4), and none of them, to our knowledge, have approached the question using optimal design theory. Some work on design of xenograft experiments has been published (30), but the focus was on comparing treatment and control groups, disregarding the correct estimation of drug-related parameters. The use of optimal design will ensure more informative studies and provide measurement times that guarantee the smallest pRSE for the model parameters. In the present work, optimal design was performed for the Simeoni TGI model, for different sets of experiments, in order to answer some important questions arising from these studies.

Optimal designs strategies applied in two-arm designs, *i.e.*, control *versus* treatment, showed that studies curtailed three days after the end of treatment administration led to a much higher pRSE of the drug-related parameters k_1 and k_2 , compared with those obtained in longer studies, where for ethical reasons tumor growth measurements could only be taken until tumor weight reached 6 g. This result proved that short studies would likely prevent correct identification of parameters k_1 and k_2 , whereas long studies should ensure more precise estimates of Simeoni TGI model parameters, leading to a more informative study. This can be considered as a proof-of-concept study, where the hypothesis has been stated in the past by many laboratories and in (27), but has never been proved before, to our knowledge. Graphs of sensitivity functions through time (Fig. 3) showed the time points in which the Simeoni TGI model parameters were more informative in each experiment, underlying the fact that parameters k_1 and k_2 increase their impact starting from the treatment period, and continuing also afterwards, during the regrowth phase. These results were observed in all the experiments of Table I, where either individual or population approaches were used. Moreover, those experiments were performed for different cell lines, drugs, and drug administration schedules. Furthermore, the optimal sampling times reported in Table I were located in the last part of the regrowth phase at least two out of four times, which partially

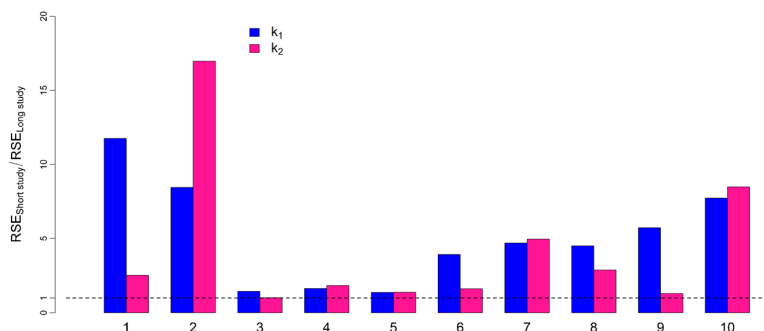


Fig. 5. Ratio of predicted RSE (%) of short study to long study in two-arm designs for drug-related parameters k_1 (blue bars) and k_2 (pink bars) for the experiments listed in Table I

Optimal Design in Tumor Growth Experiments in Mice

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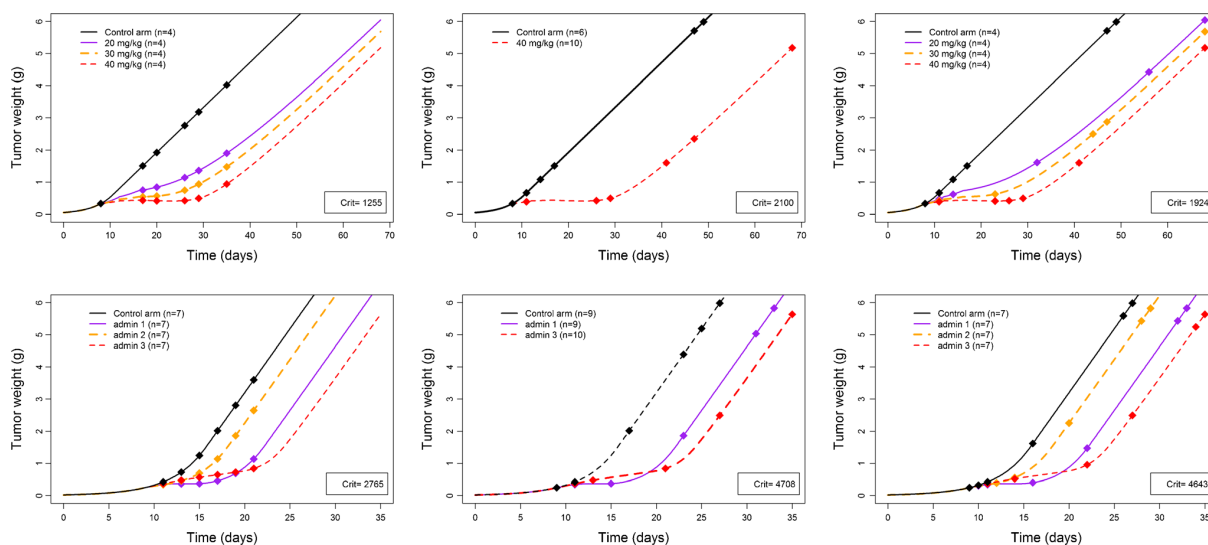


Fig. 6. From left to right: initial design, optimal design for first scenario, and optimal design for second scenario (*i.e.*, with fixed proportions of mice per arm) for Exp A (*top panel*) and Exp B (*bottom panel*). D-optimal criteria (“Crit”) obtained for each design are specified inside each corresponding graph

supports the initial intuition that sampling during the tumor regrowth phase would ensure more informative studies.

In some laboratories, experiments stop when the tumor reaches 2 g (5), disregarding the fact that the regrowth rate may be well visible and then model parameters could be well estimated. Furthermore, other researchers stop analyzing when the tumor weight in the control group reaches the maximum allowed by the Institutional Animal Care and Use Committee (IACUC) (31), which corresponds to 4 g. This may be an issue for the estimation of drug-related parameters, in cases where the tumor in the treatment group has not started regrowing by the time the control group has reached a tumor weight of 4 g. Note that 6 g is an overestimated threshold adopted to emphasize the impact on precision of parameter estimates in short *versus* long studies. However, results are substantially confirmed also choosing lower limits. It should also be considered that longer studies may cost more than short studies. A cost-efficacy analysis could be performed in order to define a good balance between information acquisition and cost, as envisaged in our study.

The conclusion drawn from the results discussed in this first part is that making measurements during tumor regrowth should become a general rule for informative preclinical studies in oncology, independently of adopting standard nonlinear regression or an NLMEM approach.

Using a population approach, we further investigated the application of optimal design strategies to four-arm designs in two experiments with several mice included in each arm, considering for the three treatment arms either three different doses or three different drug administration schedules and the same dose. The former optimal design experiment led to the expected conclusion that the treatment arm with the highest dose would provide the greatest information, and the other treatment arms were excluded from the optimal design. We may suspect that in cases where the relationship

between drug concentration and cell killing effect is not linear, intermediate doses might be selected by the optimal design, but these analyses were not performed in this work. In preclinical trials, intermediate doses are often studied to answer other experimental questions, *e.g.*, toxicology studies. We therefore evaluated an optimal four-arm design including also intermediate doses and using the same number of mice in each arm. We found that keeping the intermediate doses did not lead to a large loss of information, compared with the optimal two-arm design obtained with the highest doses. For the latter experiment with different dosing regimens, the same optimal design strategy was employed. In the optimal design, one treatment arm with a “poor” drug administration schedule was excluded, but in the optimal four-arm design evaluation with same number of mice per arm it was again found that this experiment did not greatly reduce the information yielded by the study. For both experiments, the proportions of mice were approximately 63% treated mice (divided in the treatment arms) and 37% control mice. As computation of FIM is independent of the number of subjects, these proportions should remain the same when performing the same experiment with a different number of mice in the initial design.

In this work, the design variable to be optimized was the sampling time, but dose and dosing regimen could also be considered as design variables. It would be interesting for future studies to investigate the impact of dosing for longer compared with shorter times.

In optimizing design, it is necessary to define the model and parameter values for the model at the beginning of the analysis. In practice, the values for the parameters can be taken from previous experiments. The estimation of parameters from well-planned experiments based on these D-optimal designs provides better estimates than the initial ones and optimal design can be

generated with this new set of parameters for subsequent studies. For laboratories in which xenograft experiments are routinely performed, optimal design could be applied in a sequential manner, to fine-tune the experimental design. A limitation of this work is that the model parameters were assumed to be true and error-free. Indeed, the D-optimality used here does not handle uncertainty in parameters. To introduce uncertainty in the parameters, several robust design criteria (32–34) have been developed in recent years, based on the assumption of assigning prior distributions for the parameters, rather than constraining them to a fixed value. An alternative to robust design methods could be to apply two-stage adaptive design (35), *e.g.*, starting the experiment with a small number of mice in the first cohort and then performing a second part of the experiment with a better design. Moreover, considering the model as true could also be a limitation. The concept of robustness could be extended across models, leading to model averaging criteria (36), which could be considered for application in future studies.

We would like to stress the fact that in the present work the performance of the optimal designs in long and short studies was compared based on the predicted precision of the model parameters. Complementary to this analysis, Monte Carlo simulations could be performed to obtain the empirical precision and biases of the parameter estimates, considering that parameter estimation would be computed for a certain number of simulated datasets, for which responses are simulated at the optimal times found in the optimal design analysis. The Monte Carlo approach could thus be considered for use in future studies.

An additional limitation of this work is that the number of sampling times to be optimized was kept low, *i.e.*, either four or six times for each experiment described in Tables I and II, respectively. But it may be important for researchers to take more measurements in long studies, to keep track of tumor size more frequently. In practice, measurements are taken at a higher frequency, also considering the fact that caliper readings are relatively inexpensive.

Another limitation of our work relates to software limitations. It would be interesting to introduce more flexibility into the design optimization settings by requiring optimal time ranges instead of optimal fixed times.

CONCLUSION

To conclude, this work was a first attempt to provide some general guidelines for preclinical xenograft experiments and proved that optimal design can help us perform more informative preclinical tumor growth inhibition studies, by measuring tumor growth for longer after the end of treatment, thus ensuring more precise model parameter estimation and, hence, better translation into clinical studies.

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Chapter 3

Adaptive design in pharmacometrics

3.1 Summary

Adaptive designs are a promising alternative to robust design, for taking uncertainty in the model parameters into account. In traditional "local" designs, parameters are fixed during the study; adaptive designs (ADs) are more flexible designs as they allow to modify predefined aspects – such as parameter values – while the study is still ongoing, using the accruing information (Chang, 2007; Foo and Duffull, 2012).

Some important steps in AD are choosing the number of stages one wants to compute, i.e. the number of times parameters estimation and adaptations will be performed, and the cohort size to be set for each stage.

The example used, concerns a PKPD model in oncology, based on the SMAD phosphorylation (pSMAD) biomarker, and developed for a novel oral transforming growth factor β (TGF- β) inhibitor (Gueorguieva et al., 2007; Bueno et al., 2008). This example was proposed by the EFPIA, for DDMoRe. In this study the aim was at evaluating ADs by clinical trial simulation and to compare them with one-stage designs, i.e., when no adaptation is performed, using wrong prior parameters.

The PKPD model was assumed known. Two sets of population parameters were considered: wrong "prior" parameters (Gueorguieva et al., 2014) Ψ_0 , and "true" parameters, Ψ^* . We evaluated several designs of $N = 50$ patients: two one-stage designs ξ_0 and ξ_* optimized with Ψ_0 and Ψ^* , respectively, and various two-stage designs with different cohort sizes at each stage ($N_1 + N_2 = N$). Two-stage designs have N_1 patients with design ξ_0 in the first cohort and $N_2 = N$ patients with design ξ_2 in the second cohort, where ξ_2 is optimized using parameter estimates from data collected after first stage. We finally compared a two-stage adaptive design with $N_1 = 10$ patients in the first cohort with 2 three- and 1 five-stage adaptive designs with the same number of patients in the first cohort as in the two-stage design. Design optimization was performed using the FW optimization algorithm and the determinant of FIM, which was computed by FO linearization of the structural model around the random effects (Mentré et al., 1997) with block diagonal expression (Mielke and Schwabe, 2010; Nyberg et al., 2015), in PFIM 4.0. For the adaptive designs, prior information

obtained after each stage was incorporated in the evaluation of FIM.

We simulated 100 datasets for each scenario using true parameters Ψ^* . Parameters were then estimated – after each cohort, for adaptive designs – using the SAEM algorithm in MONOLIX 4.3 (Lavielle, 2014).

Results showed that adaptive design is useful in case of wrong prior information on parameters, allowing to improve the design and obtaining meaningful results, almost as good as the optimal design if good priors were assumed.

Comparison of two-stage designs of different cohort size showed that the balanced design, with same size for the two cohorts, performed better. Two-stage designs with a small initial cohort could be reasonable to be used in some situation, for instance in early phases for ethical and safety reasons. Estimation results for designs with a large first cohort were less satisfactory compared to the other two-stage designs, because the few patients included in the second stage were not enough to correct for the wrong initial design. In case of small first cohort, more adaptations are needed but these designs are more complex to implement in clinical practice.

To conclude, this study confirmed for a PKPD simulation study in oncology that two-stage design can be useful when the correct prior information is not available. Moreover, it can be more suitable in clinical practice than designs with more stages, as only one adaptation is needed.

For further information on adaptive design method, various publications may be examined, as those here listed: Gautier and Pronzato (1999); Dette et al. (2013); Pronzato and Pázman (2013b); Lane et al. (2014).

3.2 Article II (published)

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RESEARCH PAPER

Influence of the Size of Cohorts in Adaptive Design for Nonlinear Mixed Effects Models: An Evaluation by Simulation for a Pharmacokinetic and Pharmacodynamic Model for a Biomarker in Oncology

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ABSTRACT

Purpose In this study we aimed to evaluate adaptive designs (ADs) by clinical trial simulation for a pharmacokinetic-pharmacodynamic model in oncology and to compare them with one-stage designs, i.e., when no adaptation is performed, using wrong prior parameters.

Methods We evaluated two one-stage designs, ξ_0 and ξ_* , optimised for prior and true population parameters, Ψ_0 and Ψ^* , and several ADs (two-, three- and five-stage). All designs had 50 patients. For ADs, the first cohort design was ξ_0 . The next cohort design was optimised using prior information updated from the previous cohort. Optimal design was based on the determinant of the Fisher information matrix using PFIM. Design evaluation was performed by clinical trial simulations using data simulated from Ψ^* .

Results Estimation results of two-stage ADs and ξ_* were close and much better than those obtained with ξ_0 . The balanced two-stage AD performed better than two-stage ADs with different cohort sizes. Three- and five-stage ADs were better than two-stage with small first cohort, but not better than the balanced two-stage design.

Conclusions Two-stage ADs are useful when prior parameters are unreliable. In case of small first cohort, more adaptations are needed but these designs are complex to implement.

KEY WORDS adaptive design · Fisher information matrix · nonlinear mixed effects model · optimal design · pharmacokinetic-pharmacodynamic

ABBREVIATIONS

AD	Adaptive design
FIM	Fisher information matrix
NLMEM	Nonlinear mixed effects model
PD	Pharmacodynamic
PK	Pharmacokinetic
REE	Relative estimation error
RRMSE	Relative root mean squared error
TGF- β	Transforming growth factor β

INTRODUCTION

Nonlinear Mixed Effects Models (NLMEM) [1] are increasingly performed for analysis of pharmacokinetic and pharmacodynamic (PKPD) data of preclinical or clinical studies [2, 3] in drug development and drug use.

The design of a so-called population PKPD study, that is the number of patients, the sampling times for each patient and their allocation in time, plays an important role on parameter estimation [4]. Choosing a good design when planning a study is essential and it is a crucial step as poor design can lead to inconclusive studies. For the evaluation and optimisation of population designs, the first approach was clinical trial simulation, which involves parameter estimation and thus is a time-consuming method [5]. The approach that avoids simulations, based on the Fisher information matrix (FIM) for NLMEM utilizing first-order (FO) linearisation method [6], has been first proposed by Mentré *et al.* in 1997 and several developments have been done since then. Different software are in use to perform optimal design in NLMEM [7, 8]; these are: PFIM [9, 10], PopED [11], PopDes [12], POPT [13]. Within those, one can evaluate the different designs by computing FIM and perform design optimisation based on the D-optimality criterion, i.e., maximization of the determinant of FIM. Furthermore, it was shown in [8] that all software

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provided with the same answer using the same FO approximation of FIM.

Optimal designs depend on *a priori* information, both on models and parameters, which can be partially wrong as they may be difficult to guess. Currently, population designs are often fixed for the whole study with data analyzed at its end. Local optimal designs are optimised based on a set of parameters values known *a priori*, whereas in robust optimal designs [14–16], a prior parameter distribution is defined.

Adaptive designs (ADs) are promising alternatives to local or robust designs [17]. As opposite to traditional clinical studies, ADs are clinical trial designs that use accumulating information in order to decide how to modify predefined aspects of the study during its implementation instead of leaving them fixed until the end [18, 19]. This is very important, for instance when designing a clinical trial, having only prior information on preclinical data, or designing a study in children from adults information. ADs are useful to provide some flexibility during the design but were rarely used for NLMEM [20]. Nevertheless, according to a survey by Mentré *et al.* [7], adoption of adaptive design approach for population PKPD studies is promising in pharmaceutical industries. It was shown, in previous studies not concerning NLMEM, that two-stage designs could be more efficient than fully adaptive designs [21] when the adaptation is performed after each patient. Chen *et al.* [22] compared three- versus two-stage AD. They found that the gain of extending a two-stage design approach to three-stage design is not as relevant as compared to the advantage of using two-stage instead of one-stage design. Moreover, two-stage designs are easier to implement in clinical practice as only one adaptation is performed.

Some important steps in AD are choosing the number of stages one wants to compute, i.e., the number of times parameters estimation and adaptations will be performed, and the cohort size to be set for each stage.

For NLMEM, Dumont *et al.* [23] implemented the optimisation of the determinant of FIM for two-stage adaptive designs. In that paper [23] a simulation study that mimicked the design of a pediatric PK trial was used and it was analyzed through NLMEM. For the first stage, parameters were guessed from adults. Simulations of one- and two-stage designs were evaluated assuming that some parameters were different than the initial ones. They showed the applicability and usefulness of the approach that we wish to further investigate in this study in a more complex example.

The example used for this study concerns a PKPD model in oncology, based on the SMAD phosphorylation (pSMAD) biomarker, and developed for a novel oral transforming growth factor β (TGF- β) inhibitor [12, 24]. TGF- β , plays an important role on regulation of many physiological processes. TGF- β signalling leads to phosphorylation of SMAD complexes which stimulates transcription of TGF- β responsive gene. The compound LY2157299, that is a small molecule

TGF- β inhibitor, inhibits the TGF- β induced Smad phosphorylation [25].

The objective of the present work is to evaluate by clinical trial simulation two-stage ADs for this model and compare them with one-stage designs, i.e., when no adaptation is performed, when wrong prior parameters are used. We then studied the influence of the size of each cohort in two-stage ADs. Finally we studied extensions of two-stage AD, not yet investigated in NLMEM, as three- and five-stage ADs. We used the new release of PFIM 4.0, where prior information can be incorporated on FIM evaluation and/or optimisation in order to perform adaptive design [10, 23].

MATERIALS AND METHODS

Standard NLMEM

In NLMEM, the vector of observations y_i for the i^{th} individual is described by a function f that depends nonlinearly on the p -sized vector of individual parameters ϕ_i and on the elementary design ξ_i of n_i sampling times $(t_{i1}, \dots, t_{in_i})$ then $y_i = f(\phi_i, \xi_i) + \epsilon_i$. The model can also be defined as $y_i = f(g(\beta, b_i), \xi_i) + \epsilon_i$, with $\phi_i = g(\beta, b_i)$, where β is the p -sized vector of fixed effects parameters, b_i are the random effects assumed normally distributed with zero mean and variance Ω . The standard functions for g are $g(\beta, b_i) = \beta + b_i$ and $g(\beta, b_i) = \beta \times \exp(b_i)$, corresponding to additive or exponential random effects, respectively. Here it is further assumed that Ω is a $p \times p$ diagonal matrix with diagonal elements the variances $\omega_{s,s}^2, s = 1, \dots, p$ each one corresponding to the variance of the s^{th} component of the vector b_i . The n_i -vector of residual errors ϵ_i is normally distributed with zero mean and variance equal to $\Sigma(\beta, b_i, \sigma_{\text{inter}}, \sigma_{\text{slope}}, \xi_i) = \text{diag}(\sigma_{\text{inter}} + \sigma_{\text{slope}} \times f(g(\beta, b_i), \xi_i))^2$, where σ_{inter} and σ_{slope} are the standard deviations of the additive and proportional components respectively.

The vector of the population parameter Ψ is composed of the vector of fixed effects β and the vector of variance terms $\lambda' = (\omega_1^2, \dots, \omega_p^2, \sigma_{\text{inter}}, \sigma_{\text{slope}})$, such that $\Psi' = (\beta', \lambda')$.

Adaptive Population Design

A fixed population design Ξ , i.e., a one-stage design, is defined by the total number N of individuals and the set of individual elementary designs to be performed in each individual: ξ_1, \dots, ξ_N with a total number of observations $n_{\text{tot}} = \sum_{i=1}^N n_i$, so that $\Xi = \{\xi_1, \dots, \xi_N\}$. A special case is when the same elementary design is performed in all individuals ($\xi_i = \xi$ for $i = 1, \dots, N$), then $n_{\text{tot}} = n \times N$ and $\Xi = \{\xi; N\}$. In the case of K -stage design, K population designs are defined for N_1, \dots, N_K groups of individuals ($N_1 + \dots + N_K = N$) and are denoted by Ξ_1, \dots, Ξ_K .

In this work, the following assumptions were made: i) the same elementary design ξ_k is performed for all individuals

within each cohort k ; ii) the size of each cohort N_k ($k=1, \dots, K$) is fixed, and iii) the number of sampling times n in each elementary design is fixed and is the same for each cohort.

The K -stage adaptive design schema is shown in Fig. 1 and the approach can be defined as follows. For the first stage, data Y_1 are collected for N_1 individuals with a prior design ξ_1 optimised from prior parameters Ψ_0 . Population parameters $\hat{\Psi}_1$ are then estimated from the collected data. At the k^{th} stage, data Y_k are collected for N_k individuals with design ξ_k , where ξ_k is optimised using parameter estimates $\hat{\Psi}_{k-1}$. Population parameters $\hat{\Psi}_k$ can thus be estimated with both data Y_1, \dots, Y_k gathered together from $N_1 + \dots + N_k$ individuals. The process of adaptation continues until the last step, that is the K^{th} stage, where the final parameters $\hat{\Psi}_K$ are estimated using data $Y_1, \dots, Y_k, \dots, Y_K$, collected for $N_1 + \dots + N_k + \dots + N_K = N$ individuals.

Several approaches can be used for design optimisation, here D-optimality criterion was used, which is the maximisation of the determinant of the Fisher information matrix.

Fisher Information Matrix

Let $l(\Psi|y)$ be the log-likelihood of the vector of observations y for an individual (the index i is omitted for simplicity) for the population parameters Ψ . The elementary FIM for that individual with design ξ is defined as

$$M_F(\Psi, \xi) = E\left(-\frac{\partial^2 l(\Psi|y)}{\partial \Psi \partial \Psi'}\right). \tag{1}$$

Because of nonlinearity of the model $f(g(\beta, b), \xi)$, there is no analytical expression of the log-likelihood and therefore of FIM. Several approaches have been developed in the years to compute FIM. Although there is no clear consensus on what is the best approximation, in this approach by FO linearization of the structural model around the random effects it is assumed the choice of block diagonal expression [8, 26].

As shown in Dumont *et al.* [23], in adaptive design, the population FIM in the first stage can be written as

$$M_F^1 = M_F(\Psi_0, N_1 \xi) = N_1 M_F(\Psi_0, \xi) \tag{2}$$

A design ξ_1 maximises the determinant of M_F^1 .

At the k^{th} stage, using parameters estimates from the previous stage ($\hat{\Psi}_{k-1}$), the design ξ_k corresponds to the maximum of the determinant of Fisher information matrix M_F^k , where

$$\begin{aligned} M_F^k &= M_F(\hat{\Psi}_{k-1}, N_1 \xi_1 + \dots + N_{k-1} \xi_{k-1} + N_k \xi) \\ &= N_1 M_F(\hat{\Psi}_{k-1}, \xi_1) + \dots + N_{k-1} M_F(\hat{\Psi}_{k-1}, \xi_{k-1}) \\ &\quad + N_k M_F(\hat{\Psi}_{k-1}, \xi) \end{aligned} \tag{3}$$

so that

$$\xi_k = \underset{\xi}{\operatorname{argmax}} (\det(M_F^k)) \tag{4}$$

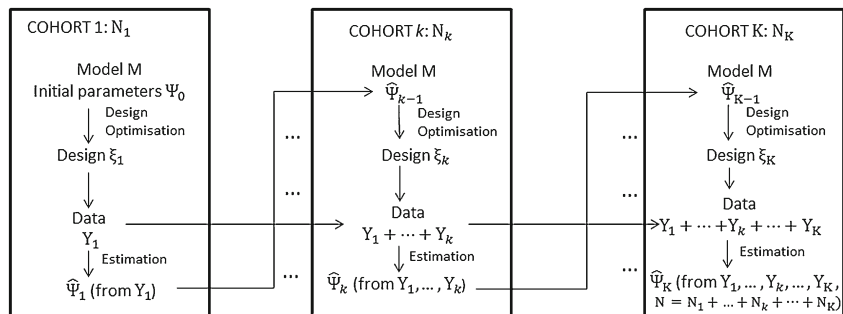
The process continues until the last stage (K) where the population FIM can be written as:

$$\begin{aligned} M_F^K &= M_F(\hat{\Psi}_{K-1}, N_1 \xi_1 + \dots + N_{K-1} \xi_{K-1} + N_K \xi) \\ &= N_1 M_F(\hat{\Psi}_{K-1}, \xi_1) + \dots + N_{K-1} M_F(\hat{\Psi}_{K-1}, \xi_{K-1}) \\ &\quad + N_K M_F(\hat{\Psi}_{K-1}, \xi) \end{aligned} \tag{5}$$

and ξ_K is the optimal design for M_F^K :

$$\xi_K = \underset{\xi}{\operatorname{argmax}} (\det(M_F^K)) \tag{6}$$

Fig. 1 Schema of K -stage adaptive design.



Simulation Study

The example used in this study is based on a PKPD model published in [12] for the compound LY2157299, a small molecule TGF-β inhibitor. A single oral daily dose of 80 mg was considered.

In the model reported in the literature, the PK was modelled by a one-compartment first-order absorption model given by:

$$C(t) = \frac{D}{V} \frac{k_a}{k_a - (CL/V)} \left(e^{-(CL/V)t} - e^{-k_a t} \right) \tag{7}$$

where D is the dose, k_a is the first order rate constant of absorption, V is the volume of distribution, CL is the clearance. Please note that CL and V are apparent volume and clearance. As there is no data after intravenous administration, bioavailability of typical individual was set to 1. The inhibition of TGF-β signalling by the treatment is represented by a turnover model [12], that is a simplification of the semi-mechanistic model developed by Bueno *et al.*[24]:

$$\frac{dR(t)}{dt} = k_{syn} \left(1 - I_{max} \frac{C(t)}{C(t) + IC_{50}} \right) - k_{out} R(t) \tag{8}$$

where R(t) is the quantity of pSMAD (correlated to TGF-β activity); k_{syn} and k_{out} are a zero order rate constant of synthesis and a first order rate constant of degradation of pSMAD, respectively, and IC_{50} is the concentration necessary to achieve 50% maximum inhibition. The maximum inhibitory response I_{max} was set to 1. A graphical representation of the model is shown in Fig. 2.

The PD used in the modelling is I(t), the relative inhibition of TGF-β defined by:

$$I(t) = \frac{R_0 - R(t)}{R_0} \tag{9}$$

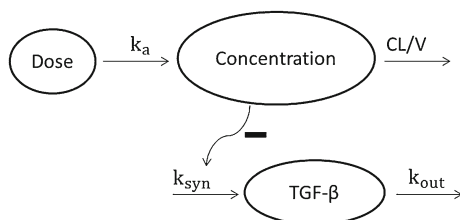


Fig. 2 Graphical representation of the PKPD model.

where R_0 is the baseline pSMAD equal to $\frac{k_{syn}}{k_{out}}$. The PD model is therefore rewritten as follow:

$$\frac{dI(t)}{dt} = k_{out} I_{max} \frac{C(t)}{C(t) + IC_{50}} - k_{out} I(t) \tag{10}$$

Two sets of PK and PD parameter values were defined for this study (Table 1): prior (wrong) parameters Ψ_0 and true parameters Ψ^* . Prior parameters for PK were those coming from a PK analysis for the clinical study [25], whereas for the PD, similar values to those obtained in a preclinical study [24] were assumed. Concerning the true parameters Ψ^* , it was assumed that prior values were correct except for CL and k_{out} which were set to be four fold smaller and ten folds smaller, respectively. The PKPD model for the two sets of parameters is displayed in Fig. 3.

Exponential random effect model was chosen for all parameters with similar inter-individual variability of 70% except for k_a whose variability was set to 0. Proportional error model and additive error model were assumed for PK and PD respectively, with σ_{prop} and σ_{inter} set to 0.2.

Evaluated Designs

Several one-stage designs, i.e., non-adaptive designs, various two-stage ADs, two three-stage and one five-stage ADs were considered for a total number of N=50 patients.

The evaluated one-stage designs were: first a rich design, ξ_{rich} , with $n=6$ sampling times, 0.1, 0.5, 1.5, 4, 6, 12 h for both

Table 1 PK and PD Parameter of the Oncology Model Used in the Simulation Studies: Prior (Ψ_0) and True (Ψ^*)

PK Parameters	Prior (Ψ_0)	True (Ψ^*)
k_a (h^{-1})	2	2
V (L)	100	100
CL ($L \cdot h^{-1}$)	40	10
$\omega_{k_a}^2$	0	0
ω_V^2	0.49	0.49
ω_{CL}^2	0.49	0.49
$\sigma_{inter,PK}$	0	0
$\sigma_{slope,PK}$	0.2	0.2
PD Parameters		
k_{out} (h^{-1})	2	0.2
IC_{50} ($mg \cdot L^{-1}$)	0.3	0.3
$\omega_{k_{out}}^2$	0.49	0.49
$\omega_{IC_{50}}^2$	0.49	0.49
$\sigma_{inter,PD}$	0.2	0.2
$\sigma_{slope,PD}$	0	0

PK and PD; then two optimal designs with $n=3$ samples among the $n=6$ sampling times in ξ_{rich} that could differ between PK and PD. These designs are: ξ_0 , D-optimal design for the prior parameters Ψ_0 and ξ_* , D-optimal design for the true parameters Ψ^* . For ξ_0 the optimal sampling times were for PK: 0.1, 4, 12 h and for PD: 0.5, 1.5, 4 h. For ξ_* the optimal sampling time were for PK: 0.1, 4, 12 h and for PD: 4, 6, 12 h. It should be noted that they are similar for PK and rather different for PD. A mixed design ξ_{0*} with $N_1 = 25$ patients with design ξ_0 and $N_2 = 25$ patients with design ξ_* was also evaluated. ξ_{0*} can be considered as a special case of two-stage design that would occur in the ideal case of estimating after the first stage the exact set of true parameters values, that is $\hat{\Psi}_1 = \Psi^*$. Graphs of PK and PD simulated models with parameters Ψ_0 and Ψ^* and optimal designs ξ_0 and ξ_* are shown in Fig. 3.

The two-stage designs evaluated in this study were: first a balanced design ξ_{25-25} with the same cohorts size, i.e., same number of patients in the two cohorts: $N_1 = N_2 = 25$; then various unbalanced designs with different sizes in the two cohorts: ξ_{10-40} ; ξ_{15-35} ; ξ_{35-15} ; ξ_{40-10} , where the first two designs have fewer patients in the first cohort whereas the second two designs have greater first cohort size.

The two three-stage designs considered have 10 patients in the first cohort: $\xi_{10-20-20}$ and $\xi_{10-10-30}$, whereas the five-stage design is $\xi_{10-10-10-10-10}$ with $N_1 = N_2 = N_3 = N_4 = N_5 = 10$ patients.

All adaptive designs (two-, three- and five-stage) start by having the first design ξ_1 equal to the prior design ξ_0 for the first cohort.

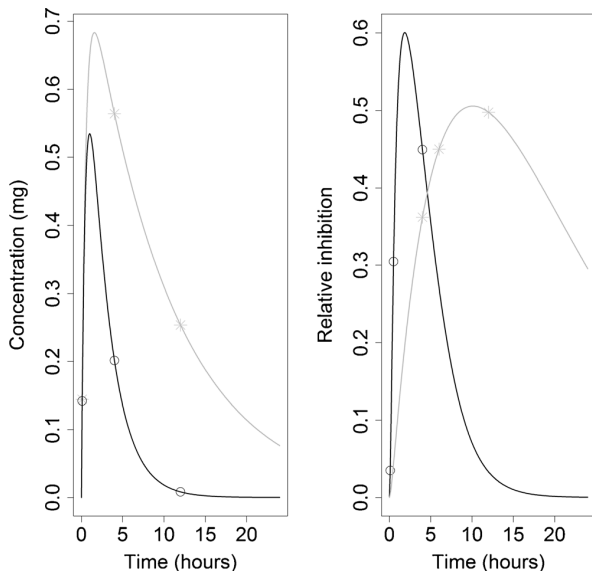


Fig. 3 PK (left) and PD (right) simulated models for a total daily dose of 80 mg, with prior parameters Ψ_0 , in dark, and optimal sampling times ξ_0 , in \circ , and with true parameters Ψ^* , in grey, and optimal sampling times ξ_* , in $*$.

Clinical Trial Simulation and Designs Comparison

One hundred datasets of $N=50$ patients were simulated with the true parameters Ψ^* and design ξ_{rich} described in the section above. In order to get datasets for the other designs only the corresponding sampling times were selected from the dataset of ξ_{rich} .

Design optimisation was performed using PFIM 4.0 [10, 23]. Adaptive designs were implemented in PFIM 4.0 [10, 23] thanks to the new features that allow for saving FIM and for considering previous information, i.e., previous FIM, in the calculation of FIM.

Parameters estimation was performed with the Stochastic Approximation Expectation Maximisation (SAEM) algorithm in the software MONOLIX 4.3 [1], with five chains and initial parameters estimates Ψ_0 . By linking through an R code PFIM 4.0 and Monolix 4.3, it was thus possible to perform K-stage adaptive design.

In order to compare the designs in terms of the precision of parameter estimates, relative estimation error (REE) and relative root mean squared error (RRMSE) were calculated from the $R=100$ final parameters estimates for 50 patients and for each design considered in this study:

$$REE(\Psi_q^r) = \frac{\hat{\Psi}_q^r - \Psi_q^*}{\Psi_q^*} \times 100 \tag{11}$$

$$RRMSE(\Psi_q) = \sqrt{\frac{1}{R} \sum_{r=1}^R \left(\frac{\hat{\Psi}_q^r - \Psi_q^*}{\Psi_q^*} \right)^2} \times 100 \tag{12}$$

In those expressions, $\hat{\Psi}_q^r$ represents the q^{th} estimated population parameter from the r^{th} simulated dataset, and Ψ_q^* is the correspondent true parameter value.

Furthermore, standardized RRMSEs [23] were computed for each design and each population parameter using as reference the RRMSE obtained with the best optimal one-stage design ξ_* optimised with the true parameters Ψ^* . More precisely the RRMSE associated to each parameter for a given design was divided by the corresponding RRMSE obtained with ξ_* , optimised with the true parameters Ψ^* . Means of the standardized RRMSEs across all parameters were also calculated and the closer they are to 1 the better is the design performance.

Estimated Optimal Designs for Various Adaptive Designs

For the 100 clinical trial simulations and for each studied adaptive design, the optimised designs of each cohort (except the first one which is fixed to ξ_0) were studied and it was also

compared how many elementary designs differed and how many were equal to the optimal design ξ_* .

RESULTS

One- and Two-Stage designs

Boxplots of the REEs for PK parameters k_a and CL, and PD parameters k_{out} and IC_{50} , for the various one-stage designs and the balanced two-stage adaptive design ξ_{25-25} are presented in Fig. 4. As expected, as design ξ_0 and ξ_* for PK are similar, estimation of PK parameters was found good for all designs and medians are close to 0. Different conclusions were drawn from the boxplots for PD parameters, where the medians REEs for the one-stage design ξ_0 in parameters k_{out} and IC_{50} were very large (about 30% and 50%, respectively),

showing a systematic bias. Good results were obtained for ξ_* as expected, and also for the mixed design ξ_{0*} and for the balanced two-stage design ξ_{25-25} . Those results were confirmed by the RRMSE values and the standardized RRMSEs to those of ξ_* (Table II). RRMSE values for PK parameters were similar to those of ξ_* in all designs. For PD parameters, large RRMSE values were obtained for design ξ_0 , with value larger than 2 for mean standardized RRMSE, whereas good results were observed for design ξ_{25-25} and design ξ_{0*} , similarly to those obtained with ξ_* , except somehow for IC_{50} and $\omega_{IC_{50}}^2$.

Influence of the Size of each Cohort in Two-Stage Adaptive Designs

Boxplots of REEs of PK and PD parameters for the various two-stage adaptive designs are presented in Fig. 5 and RRMSEs are presented in Table III. As before, there is a good

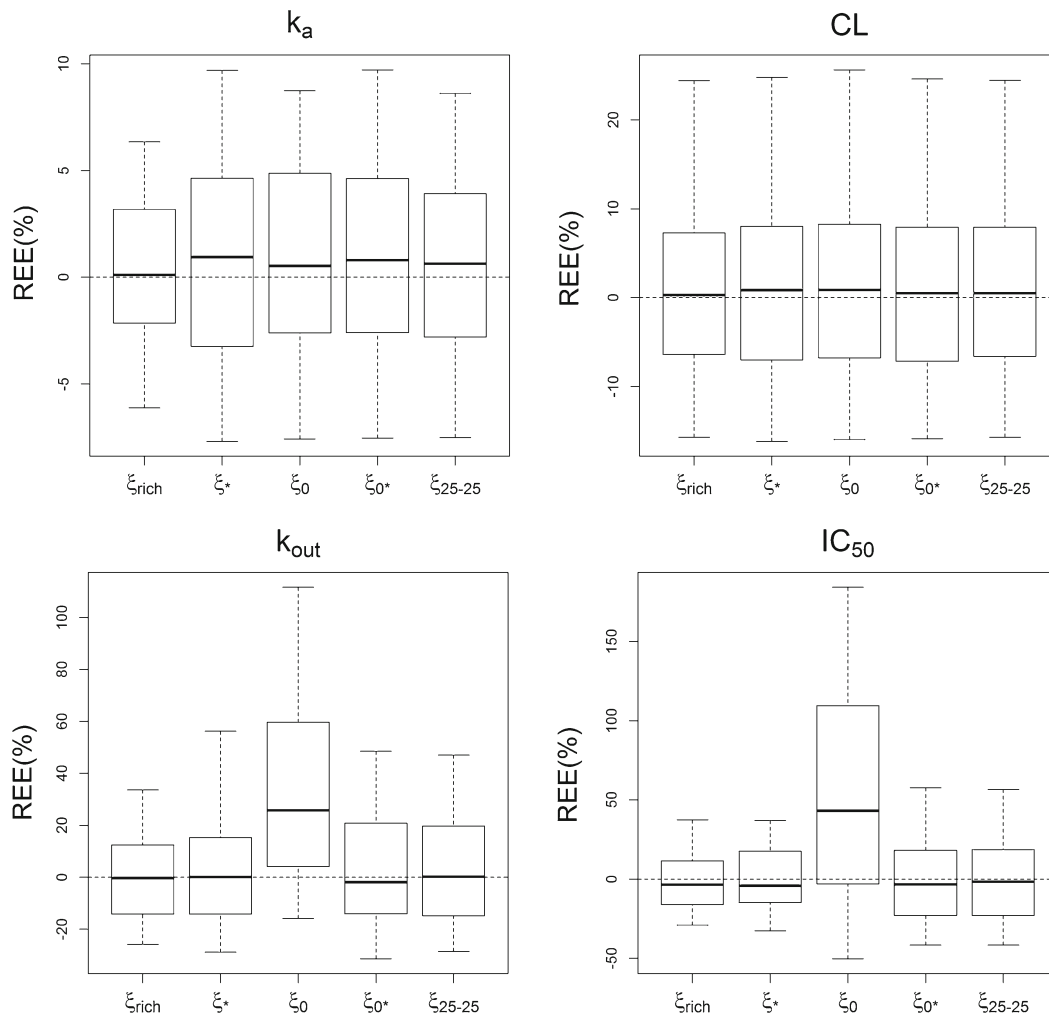


Fig. 4 Boxplot of relative estimation error (REE) for PK parameters k_a and CL (top panel) and PD parameters k_{out} and IC_{50} (bottom panel) in four one-stage designs and a balanced two-stage adaptive design.

Table II RRMSE % (and Standardized RRMSE with Respect to ξ_*) of Final Estimated Parameters in One-Stage Design and in Balanced Two-Stage Adaptive Design (N = 50 Patients)

Parameters	RRMSE % (standardized RRMSE)			
	ξ_*	ξ_0	ξ_{0*}	ξ_{25-25}
k_a (h^{-1})	5.8	5.6 (0.97)	5.7 (0.98)	5.0 (0.86)
V (L)	9.9	9.9 (1.00)	9.9 (1.00)	9.3 (0.94)
CL ($L h^{-1}$)	12.5	12.4 (0.99)	12.5 (1.00)	12.5 (1.00)
ω_V^2	22.8	22.5 (0.99)	22.5 (0.99)	22.2 (0.97)
ω_{CL}^2	24.6	24.7 (1.00)	24.4 (0.99)	24.3 (0.99)
$\sigma_{slope,PK}$	10.1	10.2 (1.01)	10.0 (0.99)	9.9 (0.98)
k_{out} (h^{-1})	23.8	54.5 (2.29)	25.4 (1.07)	24.1 (1.01)
IC ₅₀ ($mg L^{-1}$)	22.1	91.3 (4.13)	30.4 (1.38)	30.3 (1.37)
$\omega_{k_{out}}^2$	76.0	59.5 (0.78)	59.2 (0.78)	60.9 (0.80)
$\omega_{IC_{50}}^2$	72.2	709.8 (9.83)	95.3 (1.32)	98.6 (1.37)
$\sigma_{inter,PD}$	7.3	6.4 (0.88)	6.3 (0.86)	6.2 (0.85)
Mean Standardized RRMSE	1.00	2.17	1.03	1.01

RRMSE in *bold* have at least a two-fold increase standardized RRMSE

precision of PK parameters estimates among all designs, whereas for PD parameters some differences between designs are noticeable, with a better result for the balanced two-stage adaptive design ξ_{25-25} . Results of RRMSEs and standardized RRMSEs for the various two-stage adaptive designs (Table III), confirmed a better performance of design ξ_{25-25} , and worst performance of designs with a larger sample size in the first cohort (ξ_{35-15} and ξ_{40-10}), where indeed the design adaptation is performed in only a small number of patients.

Two-, Three- and Five-Stage Adaptive Designs

Finally we compared two-, three- and five-stage ADs all with same number of patients in the first cohort ($N_1 = 10$). Mean of standardized RRMSEs was smaller with the three-stage design $\xi_{10-20-20}$ and the five-stage design $\xi_{10-10-10-10-10}$, compared to the two-stage design ξ_{10-40} and the three-stage design $\xi_{10-10-30}$ (Table IV). Overall, ξ_{10-40} performed less well than the two three- and one five-stage designs considered, but those three- and five-stage designs were not better than the balanced two-stage design ξ_{25-25} . Furthermore, the five-stage design was not better than the best three-stage design $\xi_{10-20-20}$.

Comparison of Optimal Designs at each Step in Adaptive Design

Considering all adaptive designs, the number of different designs for the second cohort was the largest (12 different designs in 100 datasets) when the first cohort had only 10 patients, and was only 6 for design with larger first cohort (Table V).

For the two-stage ADs, the greatest number of datasets with optimal designs ξ_2 equal to ξ_* was obtained in the balanced two-stage AD ξ_{25-25} (Table V). Large numbers were

also obtained for designs ξ_{35-15} and ξ_{40-10} but only a small sample of patients was affected in the second stage (15 and 10 patients, respectively), which explains the bad performance of the two designs (Table III).

In three-stage designs the greatest number of simulated datasets with optimal designs ξ_2 equal to ξ_* was obtained for designs with smaller sample size in the second cohort ($\xi_{10-10-30}$) (Table V), whereas a greater number of optimal designs ξ_3 equal to ξ_* was obtained for the design that performed better, that is $\xi_{10-20-20}$.

Considering the first three stages of the five-stage design, results are similar to those obtained with the three-stage design $\xi_{10-10-30}$ with the same greatest number of simulated datasets with optimal designs ξ_2 equal to ξ_* and with only one design ξ_3 equal to ξ_* less, but in five-stage design fewer patients were used (only 30 in the first three stages, *versus* the 50 patients included in the three-stage design). For the fourth stage of five-stage designs the number of ξ_4 equal to ξ_* is smaller than the number of ξ_3 equal to ξ_* in the three-stage design $\xi_{10-20-20}$, whereas the number of ξ_5 equal to ξ_* in the fifth stage is slightly bigger than the number of optimal designs equal to ξ_* in the third stage of $\xi_{10-20-20}$, and this could explain why results of RRMSEs for the five-stage designs were similar to those obtained with $\xi_{10-20-20}$.

DISCUSSIONS

Two-stage AD in NLMEM was developed and implemented by Dumont *et al.* [23]. They have compared by simulation one-stage design, i.e., no adaptive design, with two-stage design, for a PK model in paediatrics simulation study, showing the importance of the adaptive design method if poor prior information is available. One point of discussion in their work was the

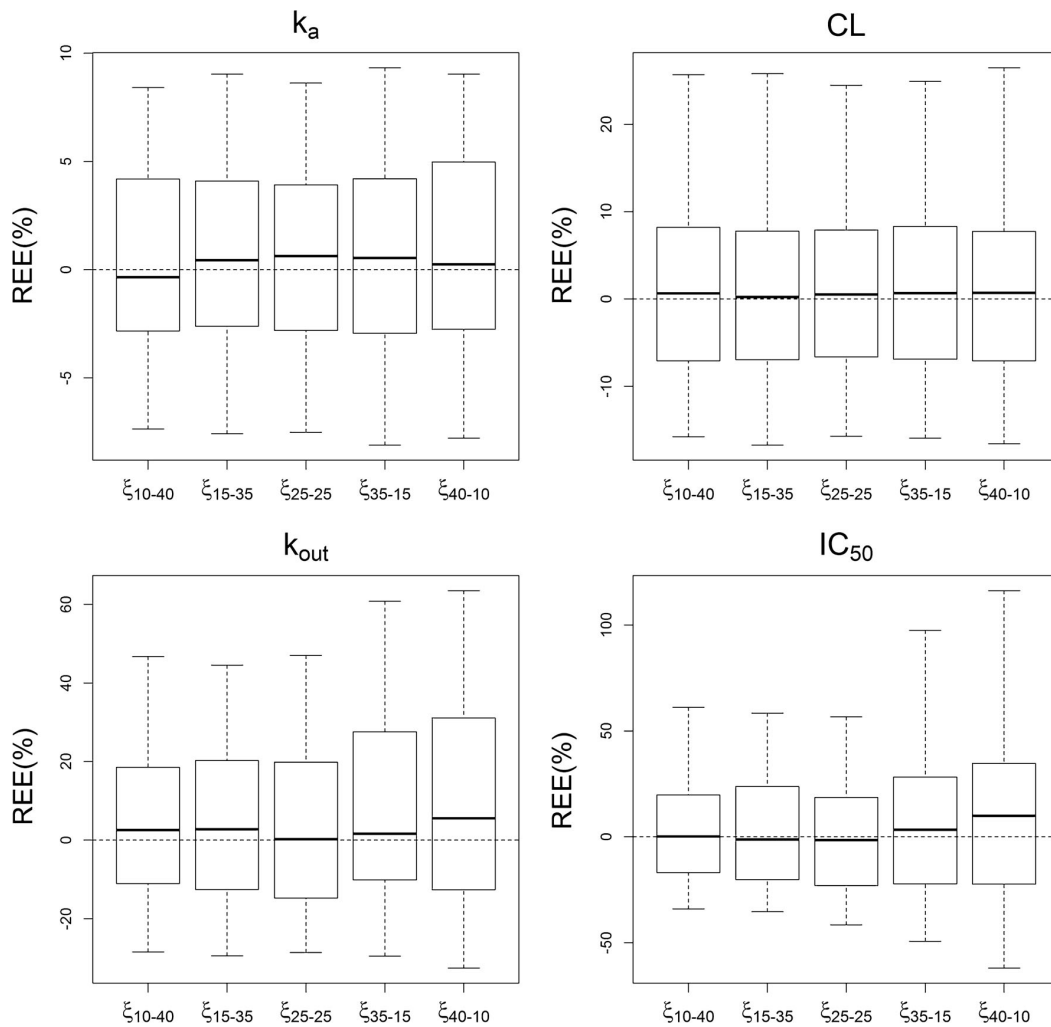


Fig. 5 Boxplot of relative estimation error (REE) for PK parameters k_a and CL (top panel) and PD parameters k_{out} and IC_{50} (bottom panel) in various two-stage adaptive designs.

fact of having used a small number of simulated dataset after the first stage, which was due to software connection limitation that comported manual work to import estimated parameters after first cohort needed for the optimisation of the design of the second stage. The need of additional studies to provide further validation of the approach was therefore necessary.

In the present study, we compared by clinical trial simulation various adaptive designs for a PKPD model in oncology [12], where the model is defined by ordinary differential equation. Design optimisations were performed in PFIM 4.0 [10], thanks to the new features that allow for saving the FIM into a text file and using FIM as prior information for the evaluation or optimisation of a design. For each design we simulated 100 clinical trials and parameters were estimated after each cohort with MONOLIX 4.3. Parameter estimates were imported in R and iteratively used in PFIM through an R loop for design optimisation of the next cohort.

We first compared one-stage design and two-stage AD as confirmatory analysis of the previous work by Dumont *et al.* Although with the prior design ξ_0 based on wrong prior parameters, there is evident bias in PD parameters, estimation results with two-stage designs were close to those with the optimal design ξ_* and much better than those with ξ_0 . Two-stage AD thus improved the design after the first stage and is therefore useful when the correct prior information is not available. We also compared various two-stage designs of different cohort size. The choice of two-stage designs with a small initial cohort is reasonable in some situation, for instance in early phases for ethical and safety reasons. Estimation results for designs with a large first cohort were less satisfactory compared with the other two-stage designs, because only few patients are then included in the second stage which is not enough to correct for the wrong initial design. In both Dumont *et al.* [23] and our study, results obtained with two-

Adaptive Design for a PKPD model in oncology

Table III RRMSE % (and Standardized RRMSE with Respect to ξ_0) of Final Estimated Parameters in Balanced and Various Unbalanced Two-Stage Adaptive Designs (N = 50 Patients)

Parameters	RRMSE % (standardized RRMSE)				
	ξ_{10-40}	ξ_{15-35}	ξ_{25-25}	ξ_{35-15}	ξ_{40-10}
k_a (h^{-1})	5.5 (0.95)	5.5 (0.95)	5.0 (0.86)	5.3 (0.91)	5.6 (0.97)
V (L)	9.4 (0.95)	9.7 (0.98)	9.3 (0.94)	9.5 (0.96)	9.7 (0.98)
CL ($L h^{-1}$)	12.4 (0.99)	12.5 (1.00)	12.5 (1.00)	12.5 (1.00)	12.5 (1.00)
ω_V^2	22.4 (0.98)	22.0 (0.96)	22.2 (0.97)	22.1 (0.97)	22.3 (0.98)
ω_{CL}^2	24.1 (0.98)	24.8 (1.01)	24.3 (0.99)	24.2 (0.98)	25.1 (1.02)
$\sigma_{slope,PK}$	10.6 (1.05)	10.0 (0.99)	9.9 (0.98)	9.9 (0.98)	9.9 (0.98)
k_{out} (h^{-1})	28.7 (1.21)	26.4 (1.11)	24.1 (1.01)	32.0 (1.34)	33.1 (1.39)
IC ₅₀ ($mg L^{-1}$)	49.1 (2.22)	36.0 (1.63)	30.3 (1.37)	45.8 (2.07)	57.2 (2.59)
$\omega_{k_{out}}^2$	60.5 (0.80)	63.8 (0.84)	60.9 (0.80)	58.8 (0.77)	62.9 (0.83)
$\omega_{IC_{50}}^2$	104.5 (1.45)	102.5 (1.42)	98.6 (1.37)	197.6 (2.74)	246.5 (3.41)
$\sigma_{inter,PD}$	6.2 (0.85)	6.7 (0.92)	6.2 (0.85)	6.4 (0.88)	6.4 (0.88)
Mean Standardized RRMSE	1.13	1.07	1.01	1.24	1.37

RRMSE in *bold* have at least a two-fold increase standardized RRMSE

stage designs were better than those obtained with ξ_0 . Moreover, a balanced two-stage design, i.e., with same number of patients in the two cohorts, provided the smaller mean standardized RRMSE, and therefore was preferable within the different cohort sizes of two-stage designs. The present results confirmed the results obtained in Dumont *et al.* [23].

We then investigated adaptive designs with more stages, which, according to our knowledge, were never evaluated in NLMEM. We considered designs with a small number of patients in the first cohort ($N_1 = 10$). Results on RRMSEs were better in the three- and five-stage design considered, than in the two-stage design, but not much better than those obtained for the balanced two-stage design. Of note, the three-stage design that performed best, $\xi_{10-20-20}$, is again the balanced

design of the remaining 40 patients after the first stage. More stage designs with larger size in the first cohort were not taken into account in this study. Further studies on feasibility of adaptive designs with more stage in clinical practice should be performed, especially if the prior guess is very far from the true value of parameters. But it should be noted that with more stages the practical implementation is more complex, hence two-stage AD seems a good approach [21].

There are several limitations in the scope of the simulation study performed here, limitations that were also present in [23]. First we assumed that the same elementary design was performed in patients belonging to the same cohort, with a fix number of sampling times. Second, we assumed that the

Table IV RRMSE % (and Standardized RRMSE with Respect to ξ_0) of Final Estimated Parameters in a Two-Stage Design, Two Three-Stage Designs and Five-Stage Design (N = 50 Patients). All Designs are Characterized by Having 10 Patients in the First Cohort

Parameters	RRMSE % (standardized RRMSE)			
	ξ_{10-40}	$\xi_{10-20-20}$	$\xi_{10-10-30}$	$\xi_{10-10-10-10-10}$
k_a (h^{-1})	5.5 (0.95)	5.6 (0.97)	5.6 (0.97)	5.5 (0.95)
V (L)	9.4 (0.95)	9.7 (0.98)	9.8 (0.99)	9.7 (0.98)
CL ($L h^{-1}$)	12.4 (0.99)	12.5 (1.00)	12.4 (0.99)	12.4 (0.99)
ω_V^2	22.4 (0.98)	22.4 (0.98)	22.2 (0.97)	22.2 (0.97)
ω_{CL}^2	24.1 (0.98)	24.3 (0.99)	24.3 (0.99)	23.9 (0.97)
$\sigma_{slope,PK}$	10.6 (1.05)	10.7 (1.06)	10.7 (1.06)	10.8 (1.07)
k_{out} (h^{-1})	28.7 (1.21)	22.3 (0.94)	25.6 (1.08)	23.0 (0.97)
IC ₅₀ ($mg L^{-1}$)	49.1 (2.22)	27.1 (1.23)	31.8 (1.44)	26.3 (1.19)
$\omega_{k_{out}}^2$	60.5 (0.80)	65.8 (0.87)	72.4 (0.95)	73.0 (0.96)
$\omega_{IC_{50}}^2$	104.5 (1.45)	96.6 (1.34)	95.2 (1.32)	92.2 (1.28)
$\sigma_{inter,PD}$	6.2 (0.85)	6.5 (0.89)	6.5 (0.89)	6.4 (0.88)
Mean Standardized RRMSE	1.13	1.02	1.06	1.02

RRMSE in *bold* have at least a two-fold increase of standardized RRMSE

Table V Number of Different Elementary Design (n_{ξ}) and Number of Datasets When the Elementary Design is Equal to ξ_* (n_{ξ}^*), for the Various Stages, of the Two-, Three- and Five-Stage Studied Adaptive Designs

	2nd Stage		3rd Stage		4th Stage		5th Stage	
	n_{ξ}	n_{ξ}^*	n_{ξ}	n_{ξ}^*	n_{ξ}	n_{ξ}^*	n_{ξ}	n_{ξ}^*
Two-stage design								
ξ_{10-40}	12	24						
ξ_{15-35}	8	35						
ξ_{25-25}	6	49						
ξ_{35-15}	6	47						
ξ_{40-10}	6	45						
Three-stage design								
$\xi_{10-20-20}$	12	27	5	71				
$\xi_{10-10-30}$	12	28	6	61				
Five-stage design								
$\xi_{10-10-10-10-10}$	12	28	7	60	4	69	4	76

structural model was known, correct and similar for all the stages. As in early phases there usually is no certainty about the model whereas there are various possible models that can be considered, we suggest using a model averaging approach for future studies [27]. Third, we assumed that the dose was fixed and identical in all patients in all cohorts and that design optimisation was performed only on sampling times. It would be interesting to expand the approach also for dose findings, optimising for instance the maximum tolerated doses in addition to the sampling times.

In adaptive design, as in optimal design, it is also necessary to define at the beginning of the analysis some prior parameters values for the model, which are usually difficult to guess when correct information is not available. Prior parameters shown in Table I were assumed to be error free. Indeed D-optimality used here at each stage does not handle uncertainty in parameters. However it should be noted that here, the changes in prior and true parameters (four fold for CL and tenfold for k_{out}) do not intend to represent estimation uncertainty but are mimicking a change from preclinical to clinical parameters. In that case taking into account estimation uncertainty using a robust design criteria, for instance for the first cohort, is expected to make only few changes. Further studies are needed to analyze the impact of the use of robust criteria in adaptive designs. Furthermore, to introduce this uncertainty in the parameters, several robust designs criteria were developed for optimal designs of fixed experiments in previous studies [17, 28, 29]. The common characteristic that links these methods is the assumption of assigning prior distributions for the parameters, rather than constraining them to a fixed value. A perspective of this work could thus be to use a robust design approach for defining prior information in the first stage of adaptive design.

In this work, we only changed two parameters, one in the PK model (CL) and one in the PD model (k_{out}) and we made rather large change in order to clearly see a variation of the shape of PK and PD curves with respect to time. In addition, by changing IC_{50} to four fold bigger or smaller, the optimal PD design did not change. When reducing inter-individual variability on PK parameters (30% instead of 70%), the design ξ_* was only slightly changed for PK (0.1, 1.5, 12 h), whereas ξ_0 was unchanged. The full simulation study with that lower variability was not performed. Inter-patient variability on k_a was not considered in the present study.

To conclude, two-stage designs provided satisfactory results close to those of the design optimised with true parameters, which allowed compensating the poor information of the prior design. The balanced two-stage design seems the best option, as in Dumont *et al.* [23], although a two responses (PKPD) model in oncology was used. Furthermore, in case of small first cohort, more adaptations may be performed but those designs are more complex to implement in clinical practice.

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Chapter 4

Robust design in pharmacometrics

4.1 Summary

Optimal design requires prior information on model and parameters which may be difficult to guess. D-optimal criterion, which maximizes the determinant of FIM, is often used as optimal design metric in local optimality for a given value of the parameters, but it does not consider parameter uncertainty. Robust designs methods can be used for taking into account uncertainty on model parameters (Pronzato and Walter, 1985; Dodds et al., 2005; Nyberg and Hooker, 2012), by assuming prior distributions for the parameters, rather than constraining them to a fixed value.

Robust criteria, such as DE, ED, EID, ELD or API, MM, are described in subsection 1.2.3 and they are all based on the determinant of FIM. Robust criteria were never systematically compared in NLMEM, according to our knowledge. Our aim was indeed at comparing robust criteria in continuous and discrete NLMEMs through two examples: the PKPD model that was already used for the AD study described in Chapter 3 and published in (Lestini, 2016), and a longitudinal binary model that was previously used by Riviere et al. (2016); Ueckert and Mentré (2016). For the former model, 50 patients were considered and 3 observations were allowed to be optimized among 11 discrete times, assuming same elementary design all individuals; for the latter, a total of 100 patients, half treated and half control, were considered and four observations were allowed, but the first and last times were kept fixed, and therefore only the two central sampling times could be optimized among a set of 11 discrete times, also assuming same design in all individuals.

The models for the two examples were known, and values of model parameters, Ψ^* , were defined. Moreover, prior distributions were assumed in both continuous and discrete NLMEMs, for two fixed-effects parameters, in order to take into account parameters uncertainty.

Robust criteria were computed through MC simulations. Indeed, a finite set of K population parameter vectors Ψ_1, \dots, Ψ_K was simulated from the prior distribution $p(\Psi)$, and FIM was computed for each Ψ_k and each possible elementary design ξ . For FIM calculation, for the model with continuous outcome, the approximation method used was

the FO linearization of the structural model around the random effects (Mentré et al., 1997) with block diagonal expression (Mielke and Schwabe, 2010; Nyberg et al., 2015). For the example with discrete outcome, the new method developed by Ueckert and Mentré (2016) that allows to derive the exact FIM was used. This method is based on quasi random Monte Carlo (QRMC) simulations and the adaptive Gaussian quadrature (AGQ) which are used to perform the derivatives of the exact conditional likelihood (Ueckert and Mentré, 2015).

Once all possible FIMs were obtained, robust criteria were computed and optimal RDs were derived. For design comparison, D-efficiency and D-criterion were then computed for each Ψ_k and for the various designs of interest: the optimal RDs, the D-optimal design, and a fixed design. A clinical trial simulation study was also performed for both examples to assess each robust design. More specifically, for each evaluated design, one dataset of N subjects was simulated for each population parameters set $\Psi_k, k = 1, \dots, 1000$. Population parameters were then estimated from each simulated dataset using the SAEM algorithm in the software MONOLIX 4.3.

For both examples, the various robust criteria lead to different optimal designs. D-efficiencies suggest that the design obtained from the expectation of the logarithm of the determinant (ELD criterion) is the more robust. Moreover, the D-optimal design performed quite well, whereas the fixed design was poor, in both examples. From CTS the difference of estimation performances between robust designs was less clear. Most designs showed rather good properties in those two examples, as also the D optimal design for the mean parameters.

To conclude, various robust criteria lead to various optimal designs, design ELD preserving best the loss of efficiency across parameter values. However, the clinical trial simulation did not find such a difference in estimation performances of these designs, perhaps due to the limited uncertainty in the prior distribution and the rather rich elementary designs used.

4.2 Article III (in preparation)

Comparison of robust design criteria in model-based analysis of longitudinal clinical data

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Robust approaches have been developed for taking into account the uncertainty of parameters when optimizing study designs. In this work we compared various robust criteria for designing studies in nonlinear mixed effect models (NLMEM), assuming a prior distribution on the parameters. Those criteria are based on the determinant of the Fisher information matrix (FIM), using different ways to account for the prior distribution of the parameters. We used two examples: (i) a population pharmacokinetic (PK)/pharmacodynamic (PD) model, considering uncertainty on two PD parameters, and (ii) a longitudinal binary model, adapted from a previous study, for which the uncertainty was considered on two fixed-effect parameters. For the PKPD model we computed the FIM using first order approximation implemented in PFIM; whereas for longitudinal binary model, we computed the FIM using a new method based on adaptive Gaussian quadrature and quasi random Monte Carlo. For each example, 1000 population parameter vectors from the prior distribution were simulated to compute the robust criteria by Monte Carlo. For both examples, the various robust criteria lead to different optimal designs. Using loss of efficiencies, compared to the optimal design for each simulated vector of population parameters, suggest that the design obtained from the expectation of the logarithm of the determinant (ELD criterion) is the most robust. We also performed a clinical trial simulation to assess each robust design. We found that the difference of estimation performances between robust designs was less clear. Most designs showed rather good properties in those two examples, as also the D optimal design for the mean parameters.

In conclusion, various robust criteria led to various optimal designs, design ELD preserving best the loss of efficiency in the extreme cases. However, the clinical trial simulation did not find such a difference in estimation performances of these designs.

Key words: Fisher information matrix; Nonlinear mixed effect model; Optimal design; Pharmacokinetic-pharmacodynamic; Robust design.

1 INTRODUCTION

Longitudinal analyses of clinical data are increasingly performed. Those studies provide repeated continuous or discrete data over time in several individuals. The main statistical tool used to analyze longitudinal data through models, is the nonlinear mixed effects model (NLMEM) (Lavielle, 2014), which allows for estimation of population parameters and inter-individual variability. Choosing a good design for a planned study is essential but it is not always straightforward. It is a crucial step as poor designs can lead to inconclusive studies. For this reason, optimal design approaches have been developed for evaluation and optimization of the design of these studies (Retout et al., 2001; Waterhouse et al., 2005; Gueorguieva et al., 2006). The optimal design approach used, which avoids simulation, relies on the Fisher information matrix (FIM) as its inverse is the lower bound of the variance-covariance matrix of any unbiased parameter estimator, as stated in the Cramer-Rao inequality. The calculation of FIM in continuous and discrete NLMEM, but also in generalized linear mixed effect models (GLMEM) is quite challenging due to the fact that there is no closed form of FIM. For this reason, different approximation methods were developed in the last decades, such as the first order (FO) linearization, which was first proposed by Mentré et al. (1997) for continuous data, and later on it was extended to various scenarios. Waite and Woods (2015) were the first to calculate FIM for general GLMEM, using marginal quasi-likelihood (MQL) and penalized quasi-likelihood approximations. For discrete NLMEM Ogungbenro et al. (2005) developed a method based on generalized estimating equations and MQL approximation for count, binary and ordinal responses; Nyberg et al. (2009) used an approach based on second-order approximation of the likelihood and applied it to binary and count responses. The method of Nguyen and Mentré (2014) based on Monte Carlo (MC) simulations and adaptive Gaussian quadrature (AGQ) for continuous response was recently extended by Ueckert and Mentré (2016) to discrete response mixed effects model. This new method does not use any approximation and it allows to numerically compute FIM by calculating the derivatives of the exact conditional likelihood. Finally, Riviere et al. (2016) proposed an alternative method to compute the asymptotically exact FIM for both continuous and discrete NLMEMs, based on MC and Hamiltonian Monte Carlo (HMC). This method is available in the R package *MIXFIM* and it uses functions that are written in the probabilistic language STAN (Carpenter et al., 2016), which was developed for Bayesian inference.

Optimal design requires prior information on model and parameters that can be partially wrong as it may be difficult to guess. D-optimal criterion, which maximizes the determinant of FIM, is often used as optimal design metric for local optimality, as it provides with the highest precision of parameter estimates, but it does not consider parameter uncertainty (Atkinson et al., 1992). Robust design (RD) methods can be used for taking into account uncertainty on model parameters (Pronzato and Walter, 1985; Dodds et al., 2005), by assuming prior distributions for the parameters, rather than constraining them to a fixed value. Several robust criteria were developed in order to compute D-optimality and they are known in the literature. Robust criteria first proposed by Pronzato and Walter (1985) are the ED and EID criteria (Walter and Pronzato, 1987) which consist in maximizing the expectation of the determinant of FIM and the inverse of the expectation of the inverse of the determinant, respectively. Pronzato and Walter (1988) then proposed the maxmin criterion

(MM), which requires knowledge of a prior feasible region for population parameters. Indeed MM-optimality implies maximization of the smallest value of the determinant of FIMs associated to the different parameters that lie in that domain. Other robust criteria were further proposed. The ELD, or API (D'Argenio, 1990; Nyberg and Hooker, 2012), consists in maximizing the expectation of the logarithm of the determinant of FIM. A simplified version of these criteria – which does not require the whole prior parameters distribution but instead it uses only the 2.5th and 97.5th percentiles of that distribution – are the so called HCDs criteria, developed for the hypercube D-optimal designs (HCDs) by Foo and Duffull (2010). Finally, parameter uncertainty was also taken into account in the context of generalized linear models (Woods et al., 2006).

Different software are in use to perform optimal design, which are adopted by pharmaceutical industry (Mentré et al., 2013). These are: PFIM (Mentré et al., 2014), popED (Nyberg et al., 2012), PopDes (Gueorguieva et al., 2007), POPT (Duffull, 2006). They provided with similar results when using the same (FO) approximation of FIM. PopED is the only one that presently has implemented also a robust criterion, more specifically the ED criterion.

All the proposed robust criteria were never systematically compared in NLMEM, according to our knowledge. Our aim was indeed at comparing robust criteria in continuous and discrete NLMEM through two examples: a PKPD model and a longitudinal binary model. We compared both optimal designs obtained and their results through an extensive simulation study.

2 METHODS

2.1 Continuous and discrete NLMEM

For continuous NLMEM, the n_i -vector y_i of observations for subject i ($i = 1, \dots, N$) at times $\xi_i = (t_{i1}, \dots, t_{in_i})$, is given by $y_i = f(g(\mu, \eta_i), \xi_i) + \epsilon_i$, where f is the nonlinear structural model and ξ_i is the elementary design composed of n_i sampling times. The vector of p individual parameters is expressed by the function g of the vector of fixed effects μ and of the vector of random effects η_i . ϵ_i is the vector of residual error following a normal distribution $\epsilon_i \sim N(0, \Sigma(\theta, \xi_i))$ with $\Sigma(\theta, \xi_i) = (\text{diag}(\sigma_{\text{inter}} + \sigma_{\text{slope}} \times f(\theta_i, \xi_i)))^2$. σ_{inter} and σ_{slope} are the additive and proportional error model parameters, respectively. It is assumed that η_i follows a normal distribution $\eta_i \sim N(0, \Omega)$, where Ω is a $p \times p$ variance-covariance matrix with diagonal elements the variances ω_s^2 , $s = 1, \dots, p$, each one corresponding to the variance of the s^{th} component of the vector η_i . The vector of the population parameter Ψ is composed of the vector of fixed effects μ and the vector of variance terms $\lambda' = (\omega_1^2, \dots, \omega_p^2, \sigma_{\text{inter}}, \sigma_{\text{slope}})$, such that $\Psi' = (\mu', \lambda')'$.

For discrete NLMEM, the probability of the n_i -vector y_i of observations for subject i ($i = 1, \dots, N$) at times $\xi_i = (t_{i1}, \dots, t_{in_i})$, is given by $P(y_i, \Psi | \eta_i) = \prod_{j=1}^{n_i} h(y_{ij}, g(\mu, \eta_i), \xi_i)$, where h is a known link function that describes the probability model of observing y_i at times ξ_i . The vector of individual parameters is given by g , the function of the fixed effects μ and of the random effects η_i . It is assumed that η_i follows a normal distribution $\eta_i \sim N(0, \Omega)$, where Ω is a $p \times p$ variance-covariance matrix with diagonal elements the variances ω_s^2 , $s = 1, \dots, p$, each one corresponding to the variance of the s^{th} component of the vector η_i . The vector of the population parameter Ψ is composed of the vector of fixed effects μ and the vector of variance terms $\lambda' = (\omega_1^2, \dots, \omega_p^2)$, such that $\Psi' = (\mu', \lambda')'$.

2.2 Population design

The elementary design for subject i , ξ_i , is given by the number of individual observations n_i and the sampling times t_{i1}, \dots, t_{in_i} . A population design Ξ is defined by the total number N of individuals and the set of individual elementary designs to be performed in each individual: ξ_1, \dots, ξ_N , with a total number of observations $n_{tot} = \sum_{i=1}^N n_i$. In the present work the same elementary design was performed in all individuals ($\xi_i = \xi$ for $i = 1, \dots, N$), then $n_{tot} = n \times N$ and $\Xi = \{\xi; N\}$.

2.3 Fisher information matrix in NLMEM

The individual FIM, $M_F(\Psi; \xi_i)$, for subject i and the elementary design ξ_i , is defined as:

$$M_F(\Psi; \xi_i) = -E \left(\frac{\partial^2 l(y_i; \Psi)}{\partial^2 \Psi} \right) \quad (1)$$

where $l(y_i; \Psi)$ is the log-likelihood of the vector of observations y_i for the individual i , for the population parameters Ψ . It is assumed that individuals are independent, therefore the population FIM $M_F(\Psi, \Xi)$ is defined as the sum of N elementary FIMs:

$$M_F(\Psi, \Xi) = N \times M_F(\Psi, \xi) \quad (2)$$

Because of non-linearity in $f(\theta_i, \xi_i)$ or h , there is no analytical expression of the log-likelihood, thus of FIM, therefore approximation methods need to be used.

2.4 Optimal design criteria

The criterion most often used in optimal design is the D-criterion. For a given *a priori* vector of population parameters Ψ^* , the D-optimal design is the design ξ_D that maximizes the determinant of the FIM:

$$\xi_D = \underset{\xi}{\operatorname{argmax}} |M_F(\Psi^*, \xi)| \quad (3)$$

When performing RD, a distribution $p(\Psi)$ is assumed for Ψ . The various robust optimality criteria to account for this prior are: DE, ED, EID, ELD, MM. Corresponding optimal robust designs are defined below:

$$\xi_{DE} = \underset{\xi}{\operatorname{argmax}} |E_{\Psi}(M_F(\Psi, \xi))|; \quad (4)$$

$$\xi_{ED} = \underset{\xi}{\operatorname{argmax}} E_{\Psi} |M_F(\Psi, \xi)|; \quad (5)$$

$$\xi_{EID} = \underset{\xi}{\operatorname{argmax}} (E_{\Psi} |M_F(\Psi, \xi)|^{-1})^{-1}; \quad (6)$$

$$\xi_{ELD} = \underset{\xi}{\operatorname{argmax}} E_{\Psi} [\log |M_F(\Psi, \xi)|]; \quad (7)$$

$$\xi_{MM} = \underset{\xi}{\operatorname{argmax}} \underset{\Psi}{\operatorname{min}} |M_F(\Psi^*, \xi)| \quad (8)$$

3 EVALUATION ON TWO EXAMPLES

3.1 Examples specifications

3.1.1 Continuous NLMEM

The example used is based on a PKPD model published by Gueorguieva et al. (2007) for the compound LY2157299, a small molecule TGF- β inhibitor. The model was already used for an adaptive design study (Lestini et al., 2015) and it was published in the drug disease models resources (DDMoRe) model repository (Lestini, 2016). A single oral daily dose of 80 mg was considered. The PK was a one-compartment model with first-order absorption:

$$C(t) = \frac{D}{V/F} \frac{k_a}{k_a - CL/V} (e^{-(CL/V)t} - e^{-k_a t}) \quad (9)$$

where D is the dose, k_a is the first order rate constant of absorption, V is the apparent volume of distribution, CL is the apparent clearance. Of note, CL and V are in reality CL/F and V/F . Because of absence of data after intravenous administration, the absolute bioavailability F was assumed to be 1. The inhibition of TGF- β signaling by the treatment is represented by a turnover model (Lestini et al., 2015), that is a simplification of the semi-mechanistic model developed by Bueno et al. (2008):

$$dI(t)/dt = k_{out} I_{max} \left(\frac{C(t)}{C(t) + IC_{50}} \right) - k_{out} I(t) \quad (10)$$

$I(t) = (R_0 - R(t))/R_0$ is the relative inhibition of TGF- β , where $R_0 = k_{syn}/k_{out}$ is the pSMAD baseline and $R(t)$ is the quantity of pSMAD, which is correlated to TGF- β activity. k_{syn} and k_{out} are a zero order rate constant of synthesis and a first order rate constant of degradation of pSMAD, respectively, and IC_{50} is the concentration necessary to achieve 50% maximum inhibition. The maximum inhibitory response I_{max} was set to 1.

The fixed effects vector μ is given by $\mu = (\mu_{k_a}, \mu_V, \mu_{CL}, \mu_{k_{out}}, \mu_{IC_{50}})$. An exponential random effect model was assumed, such that $g(\mu, \eta) = \mu \exp(\eta)$, where $\eta \sim N(0, \Omega)$, and Ω is the diagonal matrix of elements $\omega_s^2 = Var(\eta_s)$. The values Ψ^* of the model parameters are similar to those in (Lestini et al., 2015) and are displayed in Table 1.

Prior parameter distribution A multivariate log-normal distribution was assumed as prior parameter distribution, $p(\Psi)$, for two PD parameters $\mu_{k_{out}}$ and $\mu_{IC_{50}}$, with mean the vector of the logarithm of the corresponding Ψ^* values presented in Table 1, i.e. $M = ((\log(0.2), \log(0.3)))'$, and variance

$$V = \begin{pmatrix} 0.8^2 & 0 \\ 0 & 0.8^2 \end{pmatrix}$$

such that

$$(\mu_{k_{out}}, \mu_{IC_{50}})' \sim \log MVN(M, V) \quad (11)$$

Design constraints A total number of $N = 50$ subjects was considered. For PK, times were fixed to 0.1, 4 and 12 hours. For PD, $n = 3$ sampling times could be optimized among the possible times: 1, 2, 3, 4, 6, 9, 10, 15, 22, 23, 24 hours, i.e. $\binom{11}{3} = 165$ possible elementary

designs.

A design " ξ_{ES} ", with sampling times fixed to 1, 10 and 24 hours, was also considered for design comparison.

3.1.2 Discrete NLMEM

The discrete model is a logistic model for repeated binary response with treatment increasing the slope of the logit of the response with time, and it is similar to the one in (Ogungbenro and Aarons, 2011; Ueckert and Mentré, 2016; Riviere et al., 2016) and it is expressed as follows:

$$\text{logit}(\pi) = \theta_1 + \theta_2(1 + \mu_3\delta)t \quad (12)$$

where π is the probability of success.

The fixed effects vector $\mu = (\mu_1, \mu_2, \mu_3)$ is given by the intercept μ_1 , the slope μ_2 and the treatment effect μ_3 . An additive random effect model is associated to θ_1 and θ_2 , such that $\theta = g(\mu, \eta) = \mu + \eta$ where $\eta \sim N(0, \Omega)$, and Ω is the diagonal matrix of elements $\omega_s^2 = \text{Var}(\eta_s)$. δ is the treatment group indicator, i.e. $\delta = 0$ in control group and $\delta = 1$ in treatment group.

The values Ψ^* of the model parameters are displayed in Table 2 and they were arbitrarily chosen such that treatment group at the end of the study ($t = 12$) have a high probability of success (99%), whereas control group have a much lower probability of success (29%).

Prior parameter distribution A multivariate normal distribution was assumed as prior parameter distribution, $p(\Psi)$, for two fixed effects μ_2 and μ_3 , i.e. the treatment slope and the treatment effect parameters, with mean the vector of the corresponding values of Ψ^* displayed in Table 2, i.e. $M = (0.09, 5)'$, and variance

$$V = \begin{pmatrix} 0.2^2 & 0 \\ 0 & 2^2 \end{pmatrix}$$

such that

$$(\mu_2, \mu_3)' \sim MVN(M, V) \quad (13)$$

Design constraints In this study $N_t = 50$ and $N_c = 50$ for treatment and control group, respectively. Assuming $n = 4$ and the first and last time fixed to 0 and 12, respectively, two sampling times could be optimized among the following times: 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11 months, i.e. $\binom{11}{2} = 55$ possible elementary designs.

An equispaced design ξ_{ES} of times 0, 4, 8, and 12 months, i.e. a design for which samples are taken in time intervals of equal distance, was also considered for the design comparison.

3.2 Design optimization

The expectation on the prior distribution of the parameters in the robust criteria was computed by MC simulations. For each example, $K = 1000$ sets of parameters values were simulated from the prior distribution $p(\Psi)$: $\Psi_k, k = 1, \dots, 1000$.

For the continuous NLMEM FO linearization of the structural model around the random effects (Mentré et al., 1997) with block diagonal expression (Mielke and Schwabe, 2010; Nyberg et al., 2015) was applied using the PFIM program (Mentré et al., 2014) to compute the

FIM for each Ψ_k . For the discrete NLMEM, the first MC-AGQ method (Ueckert and Mentré, 2016) was improved on the MC part (Ueckert and Mentré, 2015) and we chose here quasi random MC (QRMC). We used the following settings: 500 integration samples for QRMC and 3 nodes for AGQ computation.

For each of the 165 (55) designs for the continuous (discrete) example, robust criteria were computed as the average over all $\Psi_k, k = 1, \dots, 1000$, of the FIM for Ψ_k to get the corresponding optimal design. Optimal design using the standard D-criterion for Ψ^*, ξ_D , and for each $\Psi_k, \xi_{D,k}$, was also obtained.

3.3 Design Comparison

Optimal robust designs and loss of efficiency For each example, optimal designs for the various criteria were compared and the loss of efficiency of each optimal design, for each of the criteria, was computed.

We then compared each robust design with all designs $\xi_{D,k}$, which are indeed the best D-optimal design if the value Ψ_k for the parameters were known *a priori*. We therefore defined for each optimal design, and each Ψ_k the loss of efficiency as:

$$Eff_k^D(\xi) = \left(\frac{|M_F(\Psi_k, \xi)|}{|M_F(\Psi_k, \xi_{D,k})|} \right)^{1/p} \quad (14)$$

For each design, boxplots of these loss of efficiencies over the $K=1000$ values of the parameters were computed, and we focused mainly on the lower 5th percentile. Indeed it represents the "greatest" loss of efficiency, compared to the D-optimal design if the Ψ_k value were known.

We also computed for each design, the D criterion for each simulated vector of parameters $\Psi_k, k = 1, \dots, 1000$ and similarly drew boxplots.

Clinical trial simulations CTS include simulating the PKPD observations of a study, and estimating the population parameters based on these observations.

CTS were performed for both examples using the same simulation strategy. For each design one dataset of N subjects was simulated for each population parameters set $\Psi_k, k = 1, \dots, 1000$. Population parameters were then estimated for each simulated dataset using the stochastic approximation expectation maximisation (SAEM) algorithm in the software MONOLIX 4.3 (Lavielle, 2014) with five chains and initial parameter estimates Ψ^* .

For continuous NLMEM, relative estimation errors (REE) (%) and the 95th percentile of the absolute REE(%) distribution were calculated from the 1000 population parameters estimates for each design considered in this study.

$$REE_k = \frac{\hat{\Psi}_k - \Psi_k}{\Psi_k} \quad (15)$$

For the discrete NLMEM, estimation errors (EE) were computed instead of REE, to avoid infinite values obtained for values of Ψ_k that were close to 0. The 95th percentile of the absolute EE distribution were calculated from the 1000 population parameters estimates for each design considered in this study: $EE_k = \hat{\Psi}_k - \Psi_k$.

4 RESULTS

4.1 Optimal robust designs and loss of efficiency

Figure 1 displays the time locations of each optimal robust design and the most frequent (at least 10%) D-optimal designs across the $K=1000$ Ψ_k vectors. For the continuous example, the various robust optimality criteria led to rather different designs. For the discrete example, optimal design ξ_{DE} was identical to ξ_{ELD} and optimal design ξ_{MM} was identical to ξ_{EID} . Design ξ_D , D-optimal for Ψ^* , has high frequency among $\xi_{D,k}$ for both examples. Especially for the discrete examples it stands for 25% of the K designs, and only 12% for the continuous example.

Of note, the evaluation of FIM in this discrete NLMEM with the new proposed approach was rather fast, allowing its first application for design optimization.

Loss in efficiency of the optimal designs across the criteria for continuous and discrete examples are displayed in Table 3 and 4. Surprisingly, none of the optimal designs was good across all criteria, with dramatic loss of efficiency for some criteria.

Boxplot of loss of D-efficiencies across the 1000 values Ψ_k are displayed in top of Figure 2 for the various designs for both continuous and discrete examples. It can be seen that some robust design led to rather great loss of D-efficiency across Ψ_k values. The highest value of the 5th percentile, i.e. the lower whisker of the boxplot, was obtained for design ξ_{ELD} , in both examples. Moreover, for the discrete NLMEM, the 5th percentile of D-efficiency for ξ_{ELD} ($=\xi_{DE}$) was above 50%. Boxplot of D-criteria across the 1000 values Ψ_k are displayed in bottom of Figure 2 for the various designs for both continuous and discrete examples. Rather similar results, although less strong, were obtained for the boxplot related to D-criterion. Highest median was obtained by the ξ_{ELD} design in continuous example and by ξ_{ELD} ($=\xi_{DE}$) and ξ_{EID} ($=\xi_{MM}$) designs in the discrete example. Indeed, in that example, the couples of identical designs $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$ are very similar, with only one different sampling time. The similarity of those designs can be due to the design constraints of the discrete NLMEM, for which the first and last sampling times were kept fixed and only the two central times had to be optimized.

Based on these results, ξ_{ELD} seems to be more robust in both examples, compared to the other designs. Furthermore, the non-optimised ξ_{ES} design performed worse than most of the optimal designs.

4.2 Clinical trial simulation results

Boxplot of REE(%) for the continuous NLMEM are presented in Figure 3 showing the impact of each evaluated design in the PD parameters $\mu_{k_{out}}$ and $\mu_{IC_{50}}$, i.e. the parameters on which uncertainty was accounted for. The values of 95th percentile of absolute REE(%), are reported in Table 5. It is noticeable that on average no design performed better than ξ_{ELD} . CTS study confirmed the results expected for continuous NLMEM, reported in subsection 4.1.

Boxplot of EE related to the discrete NLMEM are presented in Figure 5, for the three fixed effect parameters μ_1 , μ_2 , and μ_3 , but uncertainty was accounted only for μ_2 , and μ_3 . Corresponding values of 95th percentile of absolute EE are displayed in Table 5. The designs performance based on EE does not vary considerably within the various designs.

The differences in loss of D-efficiencies of the various designs illustrated above, had few consequences in the estimation performance in this CTS, which was close to the results seen

in the D-criterion distribution displayed in Figure 2, bottom part.

5 DISCUSSION

Optimal designs obtained with five different robust criteria were compared together with D-optimal design and a design with fixed sampling times, in two longitudinal models for continuous and for discrete data. All these robust designs were never systematically compared in NLMEM. Moreover, optimal design was applied to discrete NLMEM for the first time, using a new method, based on QRMC and AGQ, that allows to compute the exact FIM rather quickly, and it was implemented in R by Ueckert and Mentré (2016). For continuous NLMEM, uncertainty was accounted for two PD parameters that were assumed to be log-normally distributed, whereas for the discrete NLMEM uncertainty was assumed for the slope and treatment effect parameters, and they were assumed to be normally distributed.

The prior distributions assumed in continuous and discrete NLMEM were centered to the "true" means. This was the first intuitive guess, but other scenarios may also be considered in future studies such as, non-central prior distribution dispersed over some regions of the true distribution.

All the criteria led to different optimal sampling times in the continuous example, whereas in the discrete example robust criteria DE and EID led to same optimal designs as those obtained with ELD and MM, respectively. For the continuous example, three sampling times were optimized among eleven possible times, whereas in the discrete example each design had four sampling times with only the two central times to be optimized, as the first and last times were kept fixed. That constraint may explain why some criteria led to the same optimal designs in the NLMEM with discrete outcome. Allowing all four sampling times to be optimized, without keeping fixed any time, would be an important perspective of this work, as it may lead to stronger differences in the optimal designs that are obtained using the same criteria.

D-efficiency boxplots were used to evaluate the robust designs, compared to the one we would get if the true value of the parameters Ψ_k was known. Results showed that the highest 5th percentile was obtained in both examples by robust design ξ_{ELD} ($= \xi_{DE}$ for the discrete example). ξ_{ELD} can be considered the most robust design, as it guarantees higher efficiency than the other designs for the "worst" values of the parameters.

CTS were performed as complementary study to the previous analysis. Surprisingly, the estimation performances across the various designs were rather similar. An explanation could be related to the limited uncertainty that was assumed in the priors and the fact that it was centered on Ψ^* . Also, elementary designs for each example were rather rich (three and four sampling times in continuous and discrete example, respectively). Another limitation of this CTS study is the fact that only one dataset was simulated for each of the 1000 population parameter sets. It would be interesting to simulate more datasets, e.g. one hundred, from each set of population parameters but this might be too computationally expensive unless a subgroup of simulated population parameter sets would be selected.

Some limitations of this work are based on some designs aspects, such as the assumption of optimizing only times, whereas other design variables may also be of interest, or the fact of assuming the same design in all patients. Moreover, the two NLMEM models were considered "true" models. In order to account for model uncertainty, model averaging techniques may be applied (Hoeting et al., 1999).

Finally, another perspective of this work could be to combine adaptive design (Lestini

et al., 2015) and robust design. Indeed, adaptive design requires previous information on the parameters for the first cohort. A prior distribution could be assumed for model parameters in the first stage of adaptive design, instead of assigning fix values, and robust criteria may thus be applied. For the second stage, the information accrued from the first stage may be sufficient and local optimality may be used, or robust criterion using the uncertainty in the estimated parameters after the first cohort may be applied. In this framework, model averaging approaches (Hoeting et al., 1999) may also be integrated, at each stage of adaptive design.

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Table 1: PK and PD parameters of continuous example.

PK parameters	Ψ^*
$\mu_{k_a}(h^{-1})$	2
$\mu_V(L)$	100
$\mu_{CL}(Lh^{-1})$	10
ω_V^2	0.49
ω_{CL}^2	0.49
$\sigma_{inter,PK}$	0
$\sigma_{slope,PK}$	0.2
PD parameters	
$\mu_{k_{out}}(h^{-1})$	0.2
$\mu_{IC_{50}}(mgL^{-1})$	0.3
$\omega_{k_{out}}^2$	0.49
$\omega_{IC_{50}}^2$	0.49
$\sigma_{inter,PD}$	0.2
$\sigma_{slope,PD}$	0

Table 2: Parameters of discrete example.

Parameters	Ψ^*
μ_1	-2
$\mu_2(\text{months}^{-1})$	0.09
μ_3	5
ω_1^2	0.49
$\omega_2^2(\text{months}^{-2})$	0.03

Table 3: Optimum design obtained, across robust design criteria, for continuous example.

Criteria	Designs												
	ζ_{DE}	ζ_{ED}	ζ_{EID}	ζ_{ELD}	ζ_{MM}	ζ_D	ζ_{ES}	6, 9, 24	1, 4, 15	3, 9, 24	1, 3, 15	4, 6, 22	1, 10, 24
DE	1.00	0.72	0.33	0.82	0.27	0.86	0.38						
ED	0.85	1.00	0.12	0.73	0.11	0.67	0.36						
EID	≈ 0	≈ 0	1.00	≈ 0	0.88	≈ 0	0.08						
ELD	0.97	0.97	0.98	1.00	0.97	0.99	0.98						
MM	≈ 0	≈ 0	0.63	≈ 0	1.00	≈ 0	0.04						
D (for parameters Ψ^*)	0.98	0.98	0.91	0.97	0.89	1.00	0.85						

Table 4: Optimum design obtained, across robust design criteria, for discrete example. Of note, $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$.

	Designs										
	ξ_{DE}	ξ_{ED}	ξ_{EID}	ξ_{ELD}	ξ_{MM}	ξ_D	ξ_{ES}	ξ_{DE}	ξ_{ED}	ξ_{EID}	ξ_{ELD}
Times (months)	0, 1, 3, 12	0, 2, 7, 12	0, 1, 2, 12	0, 1, 3, 12	0, 1, 2, 12	0, 2, 3, 12	0, 4, 8, 12	0, 1, 3, 12	0, 2, 7, 12	0, 1, 2, 12	0, 1, 3, 12
Criteria											
DE	1.00	0.71	0.95	1.00	0.95	0.97	0.41	1.00	0.71	0.95	1.00
ED	0.82	1.00	0.72	0.82	0.72	0.80	0.70	0.82	1.00	0.72	0.80
EID	0.95	0.42	1.00	0.95	1.00	0.67	≈ 0	0.95	0.42	1.00	0.95
ELD	1.00	0.92	1.00	1.00	1.00	0.98	0.66	1.00	0.92	1.00	1.00
MM	0.52	0.12	1.00	0.52	1.00	0.32	≈ 0	0.52	0.12	1.00	0.52
D (for parameters Ψ^*)	0.95	0.93	0.97	0.95	0.97	1.00	0.82	0.95	0.93	0.97	0.95

Table 5: 95th percentile of absolute REE(%) of estimated PD fixed effects parameters from CTS study for continuous example.

Designs	ξ_{DE}	ξ_{ED}	ξ_{EID}	ξ_{ELD}	ξ_{MM}	ξ_D	ξ_{ES}
Parameters							
$\mu_{k_{out}}$	71	70	65	55	64	67	66
$\mu_{IC_{50}}$	49	44	68	49	66	56	52
Average	60	57	66.5	52	65	61.5	59

Table 6: 95th percentile of absolute EE of estimated fixed effects parameters from CTS study for discrete example. Of note, $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$.

	Designs	ξ_{DE}	ξ_{ED}	ξ_{EID}	ξ_{ELD}	ξ_{MM}	ξ_D	ξ_{ES}
Parameters								
μ_1		0.5	0.6	0.5	0.5	0.5	0.6	0.6
μ_2		1.8	1.5	1.9	1.8	1.9	1.8	1.3
μ_3		3.7	3.6	3.8	3.7	3.8	3.7	3.7
	Average	2.0	1.9	2.1	2.0	2.1	2.0	1.9

Legend to Figures

Figure 1 Robust designs (*red dots*), D-optimal and equispaced designs (*blue dots*) in continuous (*top*) and discrete (*bottom*) NLMEMs. D-optimal designs $\xi_{D,k}$ with frequencies higher than 10% over the 1000 population parameters vectors are reported with *black dots* with size proportional to the frequency. For the discrete NLMEM, the first and last time have light color dots to underline that they were kept fixed during the optimization. Of note, for discrete example, $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$.

Figure 2 Boxplot of D-efficiency (*top*) and D-criteria (*bottom*) obtained across 1000 Ψ_k values of the parameters in continuous (*left*) and discrete (*right*) NLMEMs, for robust designs (*red*) and for D-optimal and equispaced designs (*blue*). The whiskers represent the 5th and 95th percentiles of the data. Of note, for discrete example, $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$.

Figure 3 Boxplot of relative estimation error (REE)(%) in the clinical trial simulation, for PD parameters $\mu_{k_{out}}$ and $\mu_{IC_{50}}$ of continuous NLMEM, in robust designs (*red*), D-optimal and equispaced designs (*blue*). The whiskers represent the 5th and 95th percentiles of the data.

Figure 4 Boxplot of estimation error (EE) in the clinical trial simulation, for parameters μ_1 , μ_2 and μ_3 of discrete NLMEM in robust designs (*red*), D-optimal and equispaced designs (*blue*). The whiskers represent the 5th and 95th percentiles of the data. Of note, $\xi_{DE} = \xi_{ELD}$, and $\xi_{EID} = \xi_{MM}$.

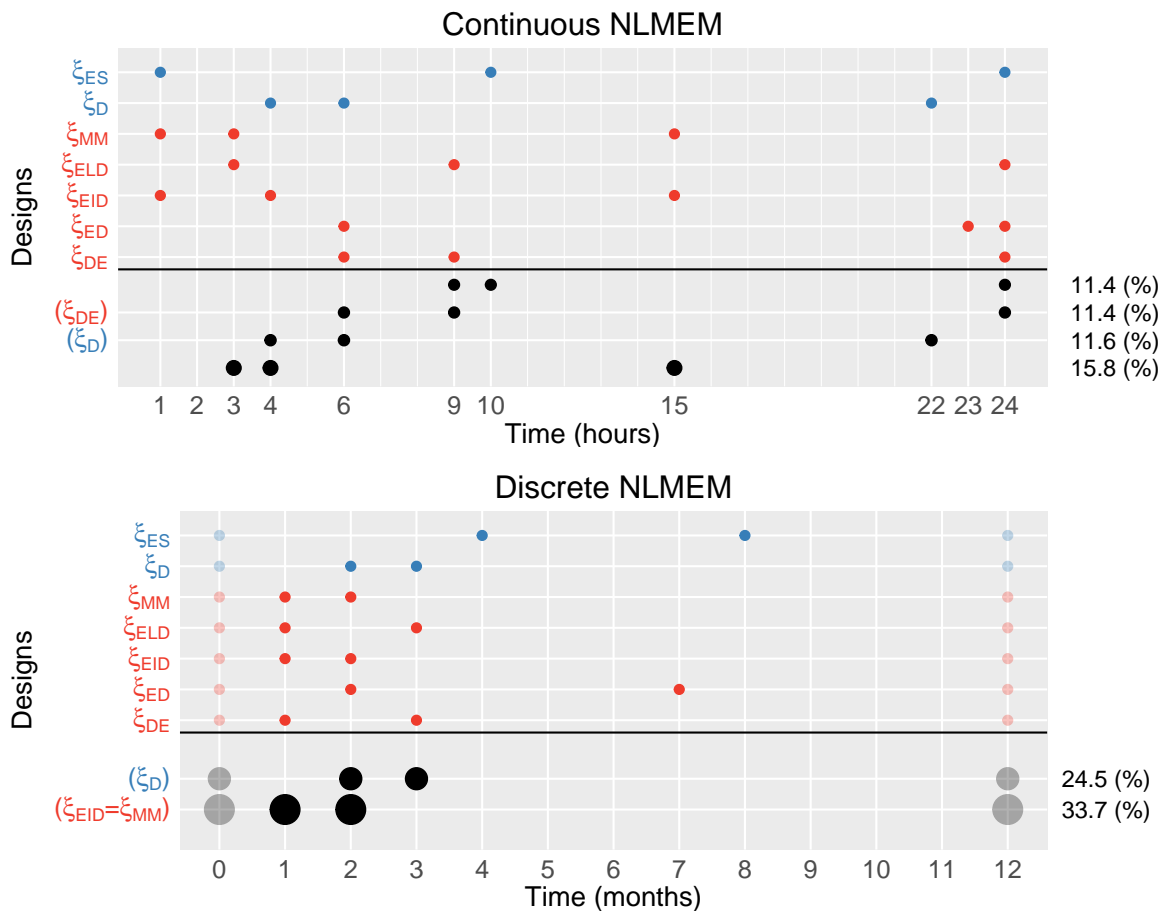


Figure 1

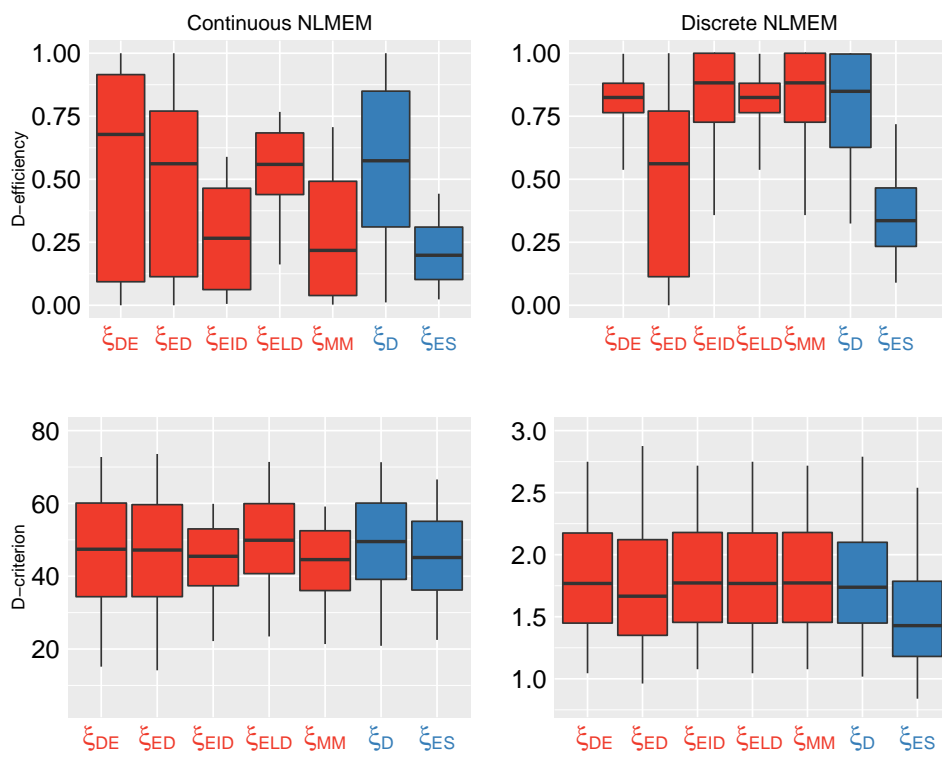


Figure 2

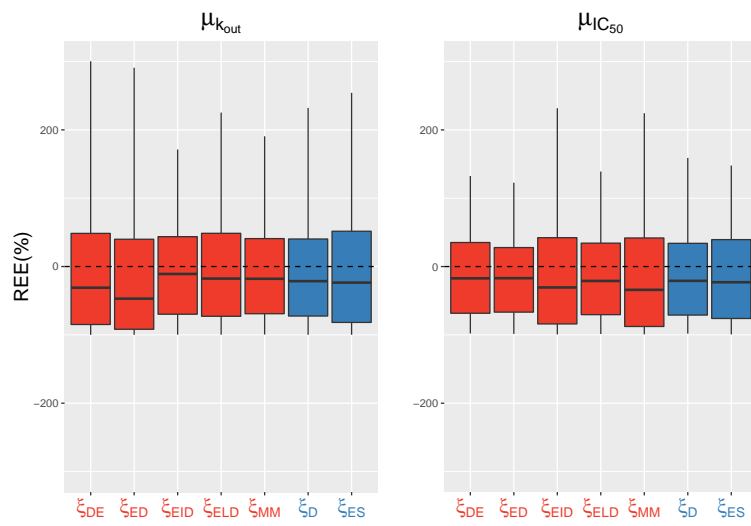


Figure 3

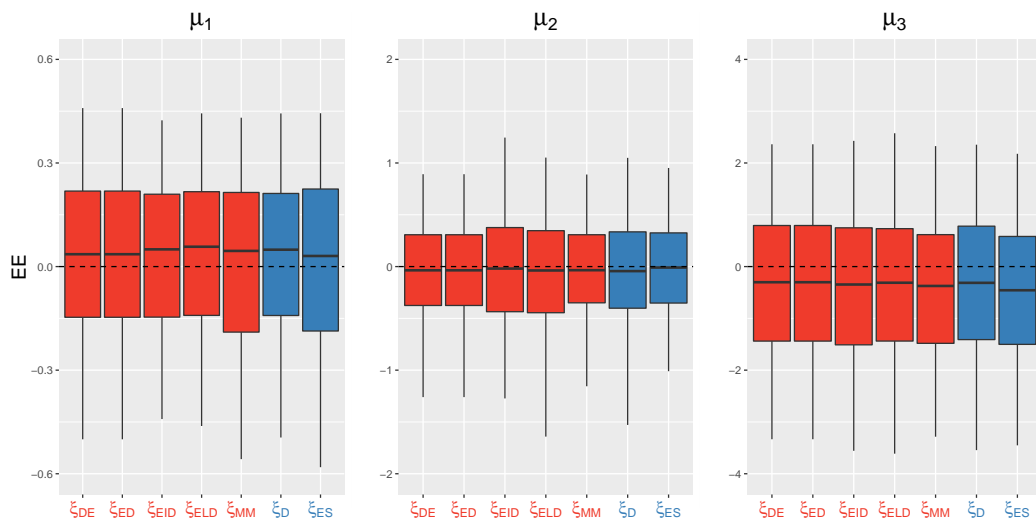


Figure 4

Chapter 5

Contribution to optimal design software development

5.1 Optimal design software PFIM

PFIM (www.pfim.biostat.fr) is a collection of functions in R, used to evaluate and/or optimize population or individual design and it enables the calculation of the FIM in continuous NLMEMs (Mentré et al., 1997; Retout et al., 2002; Retout and Mentré, 2003; Retout et al., 2007; Bazzoli et al., 2009; Nguyen et al., 2012). Several optimal design programs are available and they all provided with similar results when compared using the same FO approximation (Nyberg et al., 2015). The new release, PFIM 4.0, allows to perform adaptive optimal design, as the FIM can be saved into a text file and used as prior information for the evaluation or optimization of a design. This new version of PFIM also allows to compute individual or bayesian design (Combes et al., 2013) and models can be written in a new user-defined form.

As member of the PFIM group, part of my task was to alpha-test PFIM 4.0 before the release, and I contributed in writing the documentation of examples and implementing the corresponding examples that show all the new features of PFIM 4.0. Similarly, I also contributed to the release of PFIM Interface 4.0., the new version of the PFIM graphical user interface. PFIM 4.0 documentation of examples and PFIM Interface 4.0. documentation are reported in the Appendices 1.1 and 1.2, respectively.

5.2 DDMoRe

The main contribution of this thesis was performed for the work package (WP) 6.2., which concerns model-based adaptive optimal design (MBAOD). One of the goals of WP6.2 was to define methodologies and software tools appropriate for implementing MBAOD. Indeed, a project on adaptive design methods using an EFPIA example was worked on during the first year of PhD, and it was published in a peer-reviewed journal. That project is described in Chapter 3. For the robust design criteria comparison study presented in

Chapter 3-4, the same EFPIA example was used, and that project can also be considered a contribution for WP6.2. Indeed, robust design could be used in the first cohort of adaptive design, to account for prior parameters uncertainty.

Furthermore, Andrew Hooker implemented the MBAOD program in R, that allows for performing MBAOD by calling in R two different programs used for performing optimal design, simulating data and for model parameters estimation. Together with Sebastian Ueckert, I helped testing the MBAOD software tool, using the same adaptive design example in Chapter 4, and results were close to those previously obtained, although MBAOD was using different programs with respect to those applied in the previous study.

This PhD is a joint PhD with the University of Pavia, which is also part of DDMoRe, Work Package 1 (WP1). Indeed, the second PhD project, which is presented in Chapter 2, was worked on under the supervision of Professor Magni and Professor Mentré, i.e. my co-supervisor and supervisor, respectively, and it was published on a peer-reviewed journal. For that project, optimal design was applied to xenograft experiments – using a model in oncology of expertise of the lab in Pavia – and one of those examples was used for the demonstration of the DDMoRe interoperability framework, that was presented at PAGE meeting of Lisbon, 2016.

The objective of WP1 is to prepare a selection of approved models relevant to diabetes, oncology and diseases of other therapeutics areas, and to integrate them into the DDMoRe model repository. In that respective, I contributed by submitting a model to the DDMoRe model repository, which is available for download (Lestini, 2016).

In addition, I contributed to connect PFIM to the DDMoRe framework, by providing extensive PFIM examples and by translating into PFIM files some models that were written in the MDL language and that were provided by the MDL development group.

Chapter 6

Discussion and conclusions

Optimal design methodologies were applied in pharmacometrics to investigate the use and benefit of the approach in different phases of model based drug development in oncology. Indeed, optimal design was applied to preclinical experiments and to a clinical Phase 1 case study.

Part of this PhD focused on methods that allow to take into account model parameters uncertainty by either assuming prior distributions for the parameters, and these are the robust designs approaches, or by applying adaptive designs that consist in integrating the information of previous stages into the following stages, while the study is ongoing. Moreover, optimal and robust designs were applied for the first time in an NLMEM with discrete outcome, using a new method for computing the FIM (Ueckert and Mentré, 2016).

Discussion

Optimal design in xenograft experiment Optimal design strategies have rarely been applied to experiments concerning the in vivo evaluation of antitumor effect in xenografted mice (Simeoni et al., 2013), and none of them, to our knowledge, have approached the question using optimal design theory. In the project presented in Chapter 2, optimal design strategies were applied to published two-arm design experiments, i.e. control vs treatment, and results showed that studies curtailed three days after the end of treatment administration led to much higher pRSE of the drug-related parameters of Simeoni TGI model, compared with those obtained in longer studies, where for ethical reasons tumor growth measurements could only be taken until tumor weight reached 6 grams. This result proved that short studies would likely prevent correct identification of drug-related parameters, whereas long studies should ensure more precise estimates of Simeoni TGI model parameters, leading to a more informative study. This can be considered as a proof-of-concept study, where the hypothesis has been stated in the past by many laboratories and in (Rocchetti et al., 2013), but has never been proved before, to our knowledge.

Graphs of sensitivity functions through time showed the time points in which Simeoni TGI model parameters were more informative in each experiment, underlying the fact

that drug-related parameters increase their impact starting from the treatment period, and continuing also afterwards, during the regrowth phase. These results were observed in several experiments, where either individual or population approaches were used. Moreover, those experiments were performed for different cell lines, drugs and drug administration schedules. The conclusion drawn from the results discussed in this first part is that making measurements during tumor regrowth would be recommended for informative preclinical studies in oncology, independently of adopting standard nonlinear regression or an NLMEM approach.

Moreover, using a population approach, we further investigated the application of optimal design strategies to four-arm designs in two experiments with several mice included in each arm, considering for the three treatment arms either three different doses or three different drug administration schedules and the same dose. The former optimal design experiment led to the expected conclusion that the treatment arm with the highest dose would provide the greatest information, and the other treatment arms were excluded from the optimal design. We may suspect that in cases where the relationship between drug concentration and cell killing effect is not linear, intermediate doses might be selected by the optimal design, but these analyses were not performed in this work. In preclinical trials intermediate doses are often studied to answer other experimental questions, e.g. toxicology studies. We therefore evaluated an optimal four-arm design including also intermediate doses and using same number of mice in each arm. We found that keeping the intermediate doses did not lead to a large loss of information, compared with the optimal two-arm design obtained with the highest doses. For the latter experiment with different dosing regimens, the same optimal design strategy was employed. In the optimal design one treatment arm with a "poor" drug administration schedule was excluded, but in the optimal four-arm design evaluation with same number of mice per arm it was again found that this experiment did not greatly reduce the information yielded by the study. For both experiments the proportions of mice were approximately 63% treated mice (divided in the treatment arms) and 37% control mice. As computation of FIM is independent of the number of subjects, these proportions should remain the same when performing the same experiment with a different number of mice in the initial design. In this work, the design variable to be optimized was the sampling time, but dose and dosing regimen could also be considered as design variables. It would be interesting for future studies to investigate the impact of dosing for longer compared with shorter times.

A limitation of this work is that the model parameters were assumed to be known and true. Indeed, the D-optimality used here does not handle uncertainty in parameters. To introduce uncertainty in the parameters, robust approaches may be applied. The concept of robustness could be extended across models, leading to model averaging criteria (Hoeting et al., 1999), which could be considered for application in future studies.

An additional limitation of this work is that the number of sampling times to be

optimized was kept low, i.e. either 4 or 6 times for each experiment described in Table I and Table II, respectively. But it may be important for researchers to take more measurements in long studies, to keep track of tumor size more frequently. In practice, measurements are taken at a higher frequency, also considering the fact that caliper readings are relatively inexpensive. Another limitation of our work relates to software limitations. It would be interesting to introduce more flexibility into the design optimization settings by requiring optimal time ranges instead of optimal fixed times.

To conclude, this work was a first attempt to provide some general guidelines for preclinical xenograft experiments and proved that optimal design can help us perform more informative preclinical tumor growth inhibition studies, by measuring tumor growth for longer after the end of treatment, thus ensuring more precise model parameter estimation and hence better translation into clinical studies.

Adaptive design in pharmacometrics Two-stage AD in NLMEM was developed and implemented by Dumont et al. (2016). They have compared by simulation one-stage design, i.e. no adaptive design, with two-stage design, for a PK model in paediatrics simulation study, showing the importance of the adaptive design method if poor prior information is available. One point of discussion in their work concerned the way CTS were computed, that was by using a small number of simulated dataset after the first stage, due to software limitation. The need of additional studies to provide further validation of the approach was therefore necessary.

In the study presented in Chapter 4, we compared by CTS various ADs for a PKPD model in oncology (Gueorguieva et al., 2007; Bueno et al., 2008), where the model is defined by ordinary differential equation, and it was published in the DDMoRe model repository (Lestini, 2016). Design optimization was performed in PFIM 4.0 (Mentré et al., 2014). For each design one hundred clinical trials were simulated and parameters were estimated after each cohort with MONOLIX 4.3.

We first compared one-stage design and two-stage ADs. Although the prior design in ADs was based on "wrong" prior parameters, estimation results with two-stage designs were close to those with the optimal design based on "true" parameters and much better than those with the optimal design based on "wrong" parameters. Two-stage AD thus improved the design after the first stage and it is therefore useful when the correct prior information is not available. We also compared various two-stage designs of different cohort size. The choice of two-stage designs with a small initial cohort is reasonable in some situation, for instance in early phases for ethical and safety reasons. Estimation results for designs with a large first cohort were less satisfactory compared with the other two-stage designs, because only few patients are then included in the second stage which is not enough to correct for the wrong initial design. In both (Dumont et al., 2016) and our study, results obtained with two-stage designs, of any cohort size, were better than those obtained with optimal design based

on "wrong" parameters. Moreover, a balanced two-stage design, i.e. with same number of patients in the two cohorts, provided the smaller mean standardized RRMSE, and therefore was preferable within the different cohort sizes of two-stage designs. The present results confirmed the results obtained in (Dumont et al., 2016).

We then investigated adaptive designs with more stages, which, according to our knowledge, were never evaluated in NLMEM. We considered designs with a small number of patients (10 patients) in the first cohort. Results on RRMSEs were better in the three- and five-stage design considered, than in the two-stage design, but not much better than those obtained for the balanced two-stage design. Further studies on feasibility of adaptive designs with more stage in clinical practice should be performed, especially if the prior guess is very far from the true value of parameters. But it should be noted that with more stages the practical implementation is more complex, hence two-stage AD seems a good approach (Fedorov et al., 2012).

There are several limitations in the scope of the simulation study performed here, limitations that were also present in (Dumont et al., 2016). First we assumed that the same elementary design was performed in patients belonging to the same cohort, with a fix number of sampling times. Second, we assumed that the structural model was known, correct and similar for all the stages. As in early phases there usually is no certainty about the model whereas there are various possible models that can be considered, we suggest using a model averaging approach for future studies (Hoeting et al., 1999). Third, we assumed that the dose was fixed and identical in all patients in all cohorts and that design optimization was performed only on sampling times. It would be interesting to expand the approach also for dose findings, optimizing for instance the maximum tolerated doses in addition to the sampling times.

Finally, a perspective of this work could be to combine adaptive design and robust design. Indeed, adaptive design, as optimal design, requires previous information on the parameters. A prior distribution could be assumed for model parameters in the first stage of adaptive design, instead of assigning fix values, and robust criteria may thus be applied. For the second stage, the information accrued from the first stage may be sufficient and local optimality may be used, or estimated uncertainty. In this framework, model averaging approaches may also be applied, at each stage of adaptive design.

Robust design in pharmacometrics Five robust optimal designs were compared together with D-optimal design and a design with equispaced sampling times, in two examples: a continuous and a discrete NLMEM, where the continuous NLMEM is the same PKPD model that was used for an adaptive design study presented in Chapter 3. All these robust designs were never systematically compared in NLMEM. Moreover, optimal and robust designs were applied to discrete NLMEM for the first time, using a new method, based on QRMC and AGQ, that allows to compute the exact FIM rather quickly, and it was implemented in R by

Ueckert and Mentré (2016). For continuous NLMEM, uncertainty was accounted for two PD parameters that were assumed to be log-normally distributed, whereas for the discrete NLMEM uncertainty was assumed for the slope and treatment effect parameters, and they were assumed to be normally distributed.

The prior distributions in continuous and discrete NLMEM were centered to the "true" means. This was the first intuitive guess, but other scenarios may also be considered in future studies such as non-central prior distribution dispersed over some regions of the true distribution.

All the criteria led to different optimal sampling times in the continuous example, whereas in the discrete example robust criteria DE and EID led to same optimal designs as those obtained with ELD and MM, respectively. For the continuous example, three sampling times were optimized among eleven possible times, whereas in the discrete example each design had four sampling times with only the two central times to be optimized among 11 times, as the first and last times were kept fixed. That constraint may explain why some criteria led to the same optimal designs in the discrete NLMEM. Allowing all four sampling times to be optimized, without keeping fixed any time, would be an important perspective of this work, as it may lead to stronger differences in the optimal designs that are obtained using the same criteria.

Evaluation of the designs through D-efficiency showed a better performance of the ELD design in both NLMEMs with continuous and discrete outcome. Moreover, the D-optimal design performed quite well, and it was selected D-optimal in more than 10% of the 1000 MC simulated population parameters sets in both continuous and discrete NLMEMs.

CTS were performed as complementary study to the previous analysis. Surprisingly, the estimation performances across the various designs were rather similar. These results can be explained by the fact that the initial values of the parameters for the estimation were Ψ^* , which is the center of the prior distribution and therefore already a good estimate for all parameters with no uncertainty in the prior. Another explanation could be related to the limited uncertainty that was assumed in the priors and the fact that it was centered on Ψ^* . Moreover, one limitation of this CTS study is the fact that only one dataset was simulated for each of the 1000 population parameter sets. It would be interesting to simulate more datasets, e.g. one hundreds, from each set of population parameters but this might be too computationally expensive unless a subgroup of simulated population parameter sets would be selected.

Other limitations of this study are based on some designs aspects, such as the assumption of optimizing only times, whereas other design variables may also be of interest, or the fact of assuming the same design in all patients. Finally, the two NLMEM models were considered "true" models. In order to account for model uncertainty, model averaging techniques may be applied (Hoeting et al., 1999).

Contribution to optimal design software development PFIM 4.0 release was successful: many world-wide users has downloaded either the R version or the graphical interface. Thanks to the documentation of examples and the evaluated examples that are downloaded together with the program, one can easily learn how to apply all the new features of the program. Moreover, PFIM 4.0 is also used by some pharmaceutical companies.

Finally, PFIM 4.0. was used in all projects presented in this thesis, underlying its ability to fulfill different optimal design questions.

The Simeoni model implemented in R and used in PFIM in Chapter 3, was successfully used for a demonstration of the DDMoRe interoperability framework at the international conference PAGE, 2016, in which an application of optimal design was shown.

General conclusions

The three PhD projects led to some interesting results: i) optimal design was applied to several xenograft experiment and it showed that it could help reaching more informative preclinical xenografts studies; ii) two-stage ADs were applied to a PKPD model in oncology and performed better than fix designs with wrong prior parameters, confirming the results obtained in a different study. In case of small first cohort, more adaptations may be needed but these designs are complex to implement; iii) robust design criteria were compared in continuous and discrete NLMEMs and showed better performance of ELD design based on D-efficiency. Moreover, the release of the optimal design software PFIM 4.0 was successful, allowing the software to be used world-wide.

To conclude, this PhD project presents different contexts and use of optimal design strategies and it shows that these are powerful approaches which allow for improving the quality of a study, guarantying its reliability and a better precision of the acquired information.

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Appendix

1.1 PFIM 4.0. Documentation of examples



PFIM 4.0

PFIM Group

IAME UMR1137, INSERM and Université Paris Diderot, Paris, France

April 2014

www.pfim.biostat.fr

Examples

Written by Giulia Lestini

PFIM 4.0 is free library of functions.
The Université Paris Diderot and INSERM are the co-owners of this library
of functions.

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This documentation contains a series of examples for **EVALUATION** (first section) and **OPTIMISATION** (second section) of design in pharmacokinetics (PK) and pharmacodynamics (PD).

We have tried to illustrate all the features of PFIM 4.0 in this choice of examples. In example 1 of each section we have more specifically illustrated all the new features in this version 4.0 of PFIM.

All the input, model and output codes used for this example are available when PFIM is downloaded. In this document we have copied the specific part of the code on the illustrated features. Please remember to update the directories in the `PFIM.R` file before running each example.

EVALUATION

1. Example 1: PK Model

The purpose is to evaluate a design for a one compartment first order absorption PK model with parameters k_a , V and k after single dose administration.

1.1. Model Files

Four possible and exchangeable ways for defining models are available in PFIM. These are listed below:

- A. Model Library
- B. Expression
- C. User-defined Model
- D. Ordinary Differential Equations (ODE)

For completeness, this model was written in the four available forms.

A. Model Library:

```
source(file.path(directory.program,"LibraryPK.r"))  
  
formA<-orall_1cpt_kaVk()[[1]]  
  
form<-c(formA)
```

B. Expression:

```
formA <-expression(dose/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))  
  
form<-c(formA)
```

C. User-defined Model:

```
form<-function(t,p,X){  
ka<-p[1]  
k<-p[2]  
V<-p[3]  
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))  
return(y)  
}
```

D. choiceODE:

```
formED<-function(t,y,p)
{
ka<-p[1]
k<-p[2]
V <-p[3]
yd1<-ka*y[2]-k*y[1]
yd2<--ka*y[2]
list(c(yd1,yd2),c(y[1]/V))
}
```

NB 1: Derivatives

A new feature for specifying the derivatives is available under the Analytical Model Option (in the “stdin.R” file). This option is available only if the model is written as a function using User-defined Model form. The code for this feature in the stdin.R file is:

```
#Numerical derivatives (Yes=T, No=F)

#If 'Yes', specify the model function "form" in the model file

#If 'No', specify the object "form" which is a vector of expressions in the model
file

#-----
NUM<-F
```

NB 2: Graphs

Another new feature allows one to display graphs of the simulated model and sensitivity functions without evaluation of the Fisher Information Matrix (FIM). This can help for a better initial understanding of the case study and to check the model code.

In order to do this, one can set in the stdin.R file the “graph.only” option equals to true (“T”). Then for the evaluation of FIM it is necessary to switch the `graphic.only` option back to false and re-run PFIM.

1.2. Population Fisher Information Matrix (P-FIM)

Evaluation of the Population Fisher information matrix (P-FIM) for several designs, all patient have a dose of 100.

1.2.1. One group with Elementary Design ξ_1

200 subjects who have the same elementary design composed of 4 sampling times:

$$\xi_1 = (0.33, 1.5, 5, 12)$$

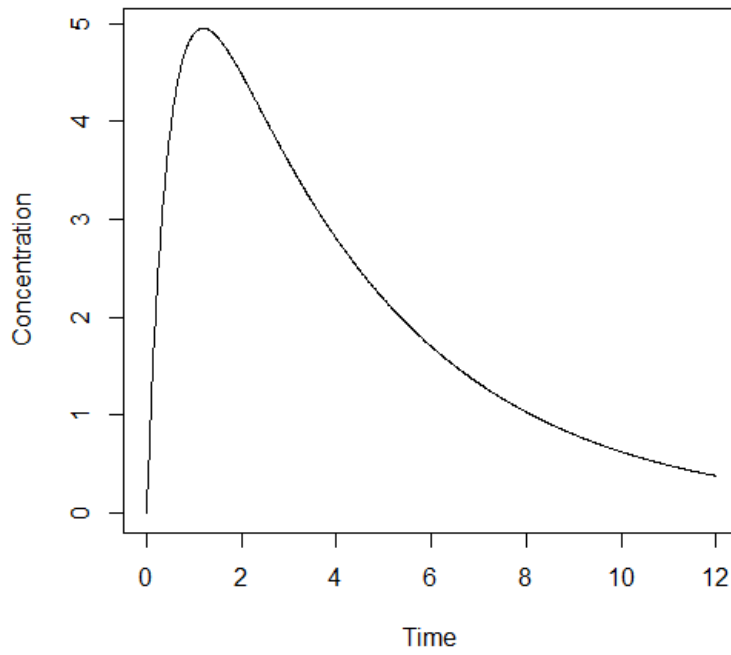
For this first example, we report the PFIM input files (stdin.R) for each possible model form (see section 1.1). We therefore repeated the example four times, using four input files. Note that all the outputs are identical so only one is given.

For other features in EXAMPLE 1, the model was written always in the “User-Defined” form.

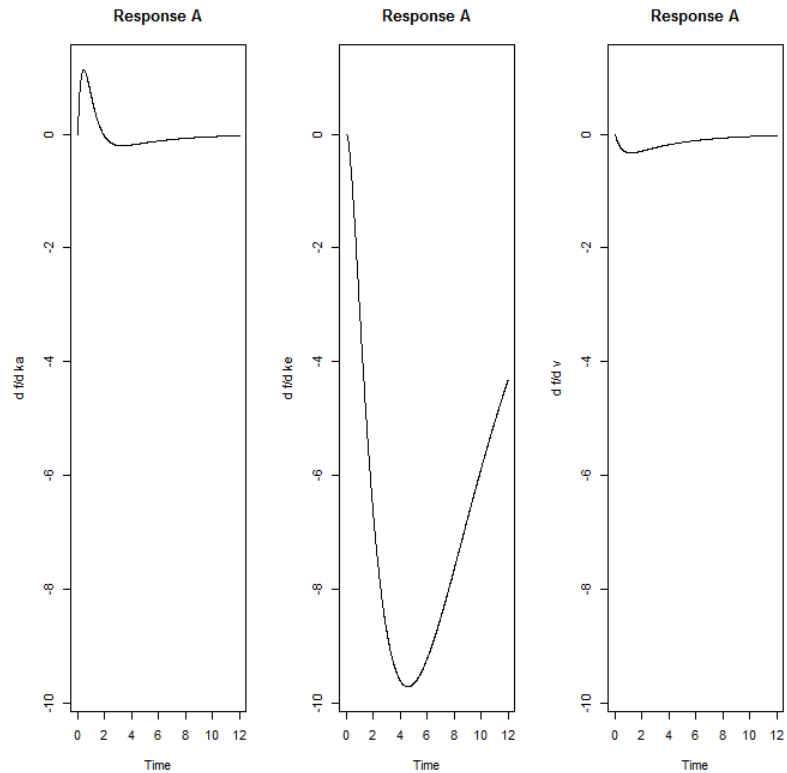
Graphs pre-evaluation

Graphs were obtained using the graphical option “`graphic.only`” showed in section 1.1.

Simulated model



Sensitivity functions with respect to each parameter



INPUT FILE; model called from the library

```
#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"1.1_ModLibrary_1.2.1"

#Name of the file containing the PK or PD model
#-----
file.model<-"model.R"

#Name of the output file for the results and for the Fisher information matrix
#-----
output<-"Stdout.r";
outputFIM<-"FIM.txt";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-""
```

```

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"EVAL"

#To display only graphs of models and/or sensitivity functions before computing
the Fisher Information matrix
graph.only<-F

#Block diagonal Fisher information matrix (option<-1) or complete Fisher
information matrix (option<-2)
#-----
option<-1

#Number of responses
#-----
nr<-1

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"AF"

##### ANALYTICAL MODEL OPTION #####
#####

#Identical dose in each elementary design (Yes=T, No=F)
#-----
dose.identical<-T

# If 'Yes', enter the value of the dose,
# else, enter the vector of the dose values for each elementary design
#-----
dose<-c(100)

#Vector of the times intervals of each expression
#-----
boundA<-list(c(0,Inf))

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-F

##### END ANALYTICAL MODEL OPTION #####
#Name of the fixed effects parameters
#-----
parameters<-c("ka","k","v")

#Fixed effects parameters values
#-----
beta<-c(2,0.25,15)

#Some parameters may not be estimated (not estimated = T, estimated = F)

```

```

#-----
beta.fixed<-c(F,F,F)

#Number of occasions
#-----
n_occ<-1

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(1,0.25,0.1))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0,0,0))

#Standard deviation of residual error (sig.inter+sig.slope*f)^2:
#-----
sig.interA<-0.5
sig.slopeA<-0.15

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.33,1.5,5,12))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(200)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

#If 'proportions of subjects' give the total number of samples
#-----
#Ntot<-40
##### GRAPH SPECIFICATION OPTION #####

#graphical representation of the model (Yes=T, No=F)
#-----
graph.logical<-T

#graphical representation of sensitivity functions (Yes=T, No=F)
#-----
graphsensi.logical<-T

#Vector of Names on X axes for each response
#-----
names.datax<-c("Time")

#Vector of Names on Y axes for each response
#-----

```

```

names.datay<-c("Concentration")

#Controls logarithmic axes for the graphical representation of the model
#Values "xy", "x", or "y" produce log-log or log-x or log-y axes.
#(For standard graphic, log.logical<-F)
#-----
log.logical<-F
#log.logical<-'y'

#Vector of lower and upper sampling times for the graphical representations
#-----
graph.infA<-c(0)
graph.supA<-c(16)

#Vector of lower and upper concentration for the graphical representations
#-----
y.rangeA<-NULL # default range
#y.rangeA<-c(1,10)

```

INPUT FILE; model written in expression form

Same stdin.R as in the sub-section above, where the model is called from the library.

```

#Name of the project
#-----
project<-"1.1_ExpressionMod_1.2.1"

```

INPUT FILE; model written in User-defined model form

```

#Name of the project
#-----
project<-"1.1_UserDefMod_1.2.1"

```

Change in the stdin.R code

```

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-T

```

INPUT FILE; ODE Model

```

#Name of the project
#-----
project<-"1.1_ModeOde_1.2.1"

##### MODEL OPTION #####

```

```

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"DE"
##### DIFFERENTIAL EQUATION OPTION #####
#####

#Initial time for which initial conditions are given
#-----
time.condinit<-0

#Identical initial conditions in each elementary design (Yes=T, No=F)
#-----
condinit.identical<-T

# If 'Yes', enter once the expression of the initial values of the system at the
initial time
# else, enter the vectors of the initial conditions for each elementary design
# If initial values depend on the parameters to be estimated,
# enter this parameter into the expression without any quotation marks
#-----
condinit<-expression(c(0,100))

# Error tolerance for solving differential equations
#-----

RtolEQ<-1e-08
AtolEQ<-1e-08
Hmax<-Inf

##### END DIFFERENTIAL EQUATION OPTION #####

```

OUTPUT FILE

```

PFIM 4.0

Project: 1.1_ModeOde_1.2.1

Date: Mon Jan 27 13:34:58 2014

***** INPUT SUMMARY *****

Differential Equations form of the model:

function(t,y,p)
{
ka<-p[1]
k<-p[2]
V <-p[3]
yd1<-ka*y[2]-k*y[1]
yd2<--ka*y[2]
list(c(yd1,yd2),c(y[1]/V))
}

Design:
Sample times for response: A

```



```

times subjects
1 c(0.33, 1.5, 5, 12)      200

Initial Conditions at time 0 :

0 100

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Error tolerance for solving differential equations system: RtolEQ = 1e-08 , AtolEQ
= 1e-08 , Hmax = Inf

Computation of the Population Fisher information matrix: option = 1

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]
[1,] 38.46761  82.86692 -3.770150  0.000000  0.000000  0.000000  0.000000
[2,] 82.86692 8310.88573 77.977974  0.000000  0.000000  0.000000  0.000000
[3,] -3.77015  77.97797  4.938229  0.000000  0.000000  0.000000  0.000000
[4,] 0.00000  0.00000  0.000000  59.190272  4.291829  31.98157  28.15945
[5,] 0.00000  0.00000  0.000000  4.291829  674.519743  213.76984  193.67427
[6,] 0.00000  0.00000  0.000000  31.981572  213.769845  3086.36662  295.74231
[7,] 0.00000  0.00000  0.000000  28.159451  193.674273  295.74231  1208.60605
[8,] 0.00000  0.00000  0.000000  85.786203  226.638153  1167.39334  1544.00251
      [,8]
[1,]  0.0000
[2,]  0.0000
[3,]  0.0000
[4,] 85.7862
[5,] 226.6382
[6,] 1167.3933
[7,] 1544.0025
[8,] 4118.3997

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta  StdError  RSE
ka  2.00  0.17480763  8.740382 %
k   0.25  0.01239415  4.957658 %
V   15.00  0.52291111  3.486074 %

----- Variance of Inter-Subject Random Effects -----

      omega^2  StdError  RSE
ka  1.00  0.13203570  13.20357 %
k   0.25  0.03977275  15.90910 %
V   0.10  0.01933249  19.33249 %

```

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.04077340	8.154681 %
sig.slopeA	0.15	0.02293716	15.291443 %

***** DETERMINANT *****

2.930398e+20

***** CRITERION *****

361.7144

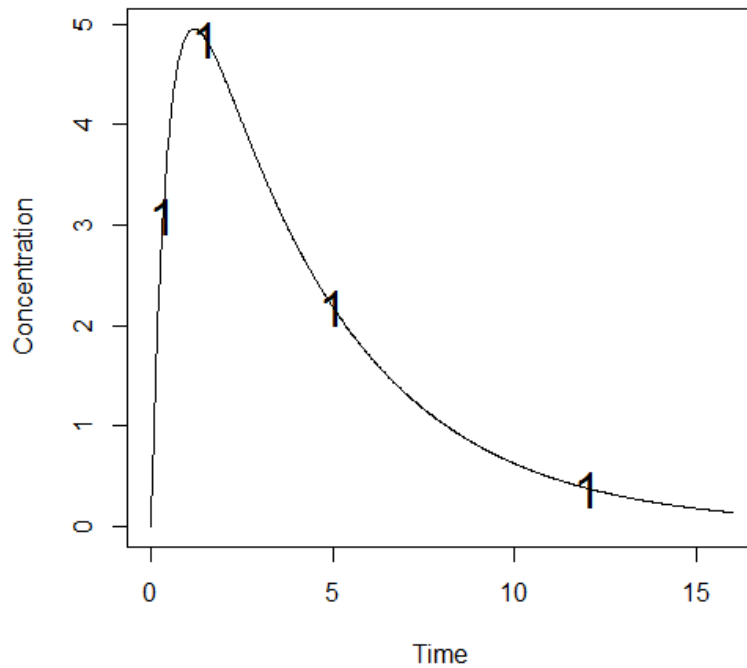
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	2490.782065	3.598669
max	8312.446904	702.614499
max/min	3.337284	195.242883

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	1.0000000	-0.2836925	0.3614018	0.0000000000	0.0000000000	0.00000000
[2,]	-0.2836925	1.0000000	-0.4466787	0.0000000000	0.0000000000	0.00000000
[3,]	0.3614018	-0.4466787	1.0000000	0.0000000000	0.0000000000	0.00000000
[4,]	0.0000000	0.0000000	0.0000000	1.0000000000	0.0008945392	-0.01707707
[5,]	0.0000000	0.0000000	0.0000000	0.0008945392	1.0000000000	-0.13147269
[6,]	0.0000000	0.0000000	0.0000000	-0.0170770725	-0.1314726865	1.00000000
[7,]	0.0000000	0.0000000	0.0000000	0.0186040065	-0.1824110351	0.12863649
[8,]	0.0000000	0.0000000	0.0000000	-0.1283096902	0.0574454505	-0.31004595
	[,7]	[,8]				
[1,]	0.00000000	0.00000000				
[2,]	0.00000000	0.00000000				
[3,]	0.00000000	0.00000000				
[4,]	0.01860401	-0.12830969				
[5,]	-0.18241104	0.05744545				
[6,]	0.12863649	-0.31004595				
[7,]	1.00000000	-0.68199073				
[8,]	-0.68199073	1.00000000				

Time difference of 0.539031 secs



1.2.2. One group with Elementary Design ξ_2

200 subjects who have the same elementary design composed of 3 sampling times:

$$\xi_2 = (1, 3, 8)$$

INPUT FILE

```
#Name of the project
#-----
project<-"1.2.2"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1,3,8))
```

OUTPUT FILE

```
PFIM 4.0

Project: 1.2.2

Date: Mon Jan 27 15:05:21 2014

***** INPUT SUMMARY *****

Analytical function models :
```

```

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
      times subjects doses
1 c(1, 3, 8)          200  100

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Population Fisher information matrix: option = 1

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]      [,8]
[1,] 24.824352 114.28686 -3.961996  0.000000  0.000000  0.000000  0.000000  0.0000
[2,] 114.286857 8098.01333 80.527748  0.000000  0.000000  0.000000  0.000000  0.0000
[3,] -3.961996  80.52775  5.116281  0.000000  0.000000  0.000000  0.000000  0.0000
[4,]  0.000000  0.000000  0.000000  24.649938  8.163429  35.31918  33.35118  147.8426
[5,]  0.000000  0.000000  0.000000  8.163429  640.408398  227.97837  186.69165  204.3552
[6,]  0.000000  0.000000  0.000000  35.319180  227.978372  3312.94177  284.04661  1079.4933
[7,]  0.000000  0.000000  0.000000  33.351180  186.691652  284.04661  455.36961  971.8908
[8,]  0.000000  0.000000  0.000000  147.842611  204.355180  1079.49330  971.89080  2577.3614

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta  StdError      RSE
ka  2.00  0.24120620 12.060310 %
k   0.25  0.01361296  5.445183 %
V  15.00  0.55940163  3.729344 %

----- Variance of Inter-Subject Random Effects -----

      omega^2  StdError      RSE
ka   1.00  0.37079715 37.07971 %
k    0.25  0.05288198 21.15279 %
V    0.10  0.02260288 22.60288 %

----- Standard deviation of residual error -----

```

```

          Sigma   StdError   RSE
sig.interA  0.50 0.18804055 37.60811 %
sig.slopeA  0.15 0.09283442 61.88961 %

***** DETERMINANT *****
1.230099e+18

***** CRITERION *****
182.4914

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

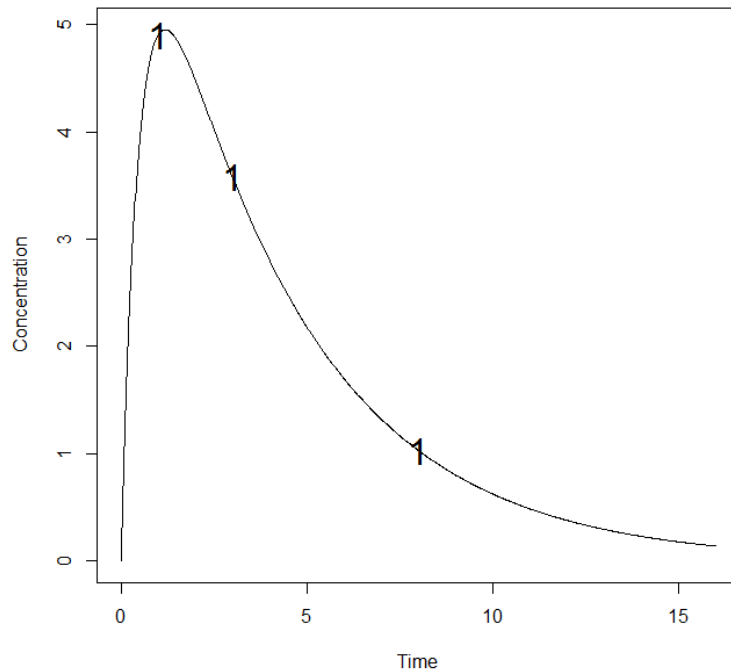
      FixedEffects VarianceComponents
min      2020.201440          3.027094
max      8100.430663          627.104778
max/min   4.009714           207.163955

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]      [,8]
[1,]  1.0000000 -0.4582328  0.5094178  0.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[2,] -0.4582328  1.0000000 -0.5360254  0.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[3,]  0.5094178 -0.5360254  1.0000000  0.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000 -0.4660810  0.4704245  0.7368611 -0.8188563
[5,]  0.0000000  0.0000000  0.0000000 -0.4660810  1.0000000 -0.4194565 -0.6482711  0.5995498
[6,]  0.0000000  0.0000000  0.0000000  0.4704245 -0.4194565  1.0000000  0.5456326 -0.6075711
[7,]  0.0000000  0.0000000  0.0000000  0.7368611 -0.6482711  0.5456326  1.0000000 -0.9589960
[8,]  0.0000000  0.0000000  0.0000000 -0.8188563  0.5995498 -0.6075711 -0.9589960  1.0000000

Time difference of 0.02700186 secs

```



1.2.3. Two Groups Design

Two group population design of 400 subjects: 200 with elementary designs ξ_1 and 200 with elementary design ξ_2 .

INPUT FILE

```
#Name of the project
#-----
project<-"1.2.3"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<- list(c(0.33,1.5,5,12),c(1,3,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(200,200)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1
```

OUTPUT FILE

```
PFIM 4.0

Project: 1.2.3

Date: Mon Jan 27 15:37:15 2014
```

***** INPUT SUMMARY *****

Analytical function models :

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Design:

```
Sample times for response: A
                times subjects doses
1 c(0.33, 1.5, 5, 12)      200  100
2          c(1, 3, 8)      200  100
```

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Computation of the Population Fisher information matrix: option = 1

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	63.291953	197.1538	-7.732147	0.00000	0.00000	0.00000
[2,]	197.153796	16408.8991	158.505718	0.00000	0.00000	0.00000
[3,]	-7.732147	158.5057	10.054510	0.00000	0.00000	0.00000
[4,]	0.000000	0.0000	0.000000	83.84019	12.45526	67.30076
[5,]	0.000000	0.0000	0.000000	12.45526	1314.92814	441.74820
[6,]	0.000000	0.0000	0.000000	67.30076	441.74820	6399.30871
[7,]	0.000000	0.0000	0.000000	61.51064	380.36592	579.78890
[8,]	0.000000	0.0000	0.000000	233.62885	430.99333	2246.88658
	[,7]	[,8]				
[1,]	0.00000	0.0000				
[2,]	0.00000	0.0000				
[3,]	0.00000	0.0000				
[4,]	61.51064	233.6288				
[5,]	380.36592	430.9933				
[6,]	579.78890	2246.8866				
[7,]	1663.97567	2515.8934				
[8,]	2515.89337	6695.7614				

***** EXPECTED STANDARD ERRORS *****

```

----- Fixed Effects Parameters -----

      Beta      StdError      RSE
ka  2.00  0.141381876  7.069094 %
k   0.25  0.009077767  3.631107 %
V   15.00  0.377988684  2.519925 %

----- Variance of Inter-Subject Random Effects -----

      omega2      StdError      RSE
ka  1.00  0.11577052  11.57705 %
k   0.25  0.02893401  11.57360 %
V   0.10  0.01357748  13.57748 %

----- Standard deviation of residual error -----

      Sigma      StdError      RSE
sig.interA  0.50  0.03908612  7.817223 %
sig.slopeA  0.15  0.02065427  13.769511 %

***** DETERMINANT *****
1.665585e+22

***** CRITERION *****
599.3712

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min      4693.602414      6.806441
max      16412.806404      1313.606918
max/min      3.496846      192.994680

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,]  1.0000000 -0.3572238  0.4228893  0.0000000  0.0000000  0.0000000
[2,] -0.3572238  1.0000000 -0.4813562  0.0000000  0.0000000  0.0000000
[3,]  0.4228893 -0.4813562  1.0000000  0.0000000  0.0000000  0.0000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000 -0.02281874  0.03522493
[5,]  0.0000000  0.0000000  0.0000000 -0.02281874  1.0000000 -0.14572956
[6,]  0.0000000  0.0000000  0.0000000  0.03522493 -0.14572956  1.0000000
[7,]  0.0000000  0.0000000  0.0000000  0.11792720 -0.24609424  0.16443852
[8,]  0.0000000  0.0000000  0.0000000 -0.28514144  0.12142535 -0.33126615

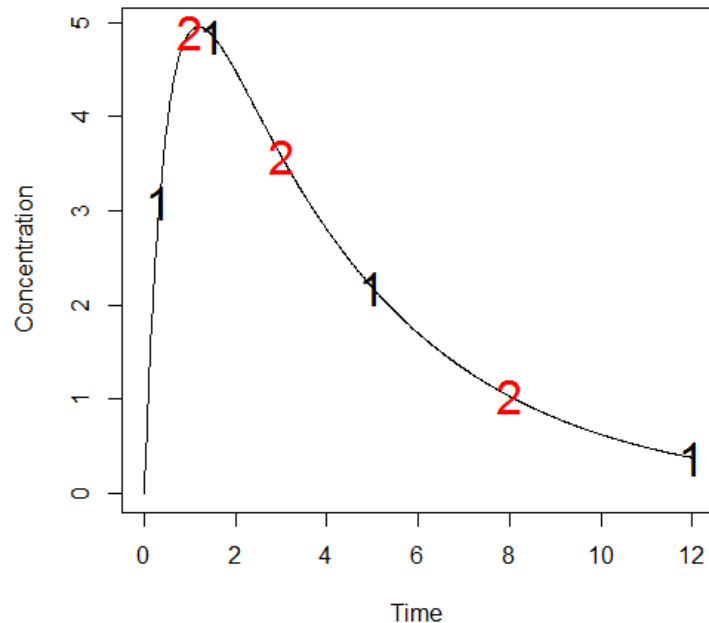
      [,7]      [,8]
[1,]  0.0000000  0.0000000
[2,]  0.0000000  0.0000000
[3,]  0.0000000  0.0000000
[4,]  0.1179272 -0.2851414
[5,] -0.2460942  0.1214254
[6,]  0.1644385 -0.3312662

```



```
[7,] 1.0000000 -0.7482048
[8,] -0.7482048 1.0000000
```

Time difference of 0.04000306 secs



1.2.4. Previous Information on PFIM

A new feature in v4.0 is the possibility to evaluate a population design with previous information stored in a file. Here we evaluate a design with 200 subjects with elementary design ξ_2 in addition to a previous information stored in the file "FIM_Prev_Info.txt". This file was produced from the example in section 1.2.1 (with the name of "FIM.txt") which corresponds to the FIM evaluated for 200 subjects with the elementary design ξ_1 .

INPUT FILE

```
#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"FIM_Prev_Info.txt"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1,3,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(200)
```

OUTPUT FILE

PFIM 4.0

Project: 1.2.4

Date: Mon Jan 27 16:23:13 2014

***** INPUT SUMMARY *****

Analytical function models :

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Design:

```
Sample times for response: A
      times subjects doses
1 c(1, 3, 8)      200  100
```

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Computation of the Population Fisher information matrix: option = 1

Previous FIM from file FIM_Prev_Info.txt

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

	V1	V2	V3	V4	V5	V6
[1,]	63.291953	197.1538	-7.732147	0.00000	0.00000	0.00000
[2,]	197.153796	16408.8991	158.505718	0.00000	0.00000	0.00000
[3,]	-7.732147	158.5057	10.054510	0.00000	0.00000	0.00000
[4,]	0.000000	0.0000	0.000000	83.84019	12.45526	67.30076
[5,]	0.000000	0.0000	0.000000	12.45526	1314.92814	441.74820
[6,]	0.000000	0.0000	0.000000	67.30076	441.74820	6399.30871
[7,]	0.000000	0.0000	0.000000	61.51064	380.36592	579.78890
[8,]	0.000000	0.0000	0.000000	233.62885	430.99333	2246.88658
	V7	V8				
[1,]	0.00000	0.0000				
[2,]	0.00000	0.0000				
[3,]	0.00000	0.0000				
[4,]	61.51064	233.6288				
[5,]	380.36592	430.9933				

```
[6,] 579.78890 2246.8866
[7,] 1663.97567 2515.8934
[8,] 2515.89337 6695.7614
```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE	
ka	2.00	0.141381876	7.069094	%
k	0.25	0.009077767	3.631107	%
V	15.00	0.377988684	2.519925	%

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE	
ka	1.00	0.11577052	11.57705	%
k	0.25	0.02893401	11.57360	%
V	0.10	0.01357748	13.57748	%

----- Standard deviation of residual error -----

	Sigma	StdError	RSE	
sig.interA	0.50	0.03908612	7.817223	%
sig.slopeA	0.15	0.02065427	13.769511	%

***** DETERMINANT *****

1.665585e+22

***** CRITERION *****

599.3712

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents	
min	4693.602414	6.806441	
max	16412.806404	1313.606918	
max/min	3.496846	192.994680	

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
V1	1.0000000	-0.3572238	0.4228893	0.0000000	0.0000000	0.0000000
V2	-0.3572238	1.0000000	-0.4813562	0.0000000	0.0000000	0.0000000
V3	0.4228893	-0.4813562	1.0000000	0.0000000	0.0000000	0.0000000
V4	0.0000000	0.0000000	0.0000000	1.0000000	-0.02281874	0.03522493
V5	0.0000000	0.0000000	0.0000000	-0.02281874	1.0000000	-0.14572956
V6	0.0000000	0.0000000	0.0000000	0.03522493	-0.14572956	1.0000000
V7	0.0000000	0.0000000	0.0000000	0.11792720	-0.24609424	0.16443852
V8	0.0000000	0.0000000	0.0000000	-0.28514144	0.12142535	-0.33126615

```

      [,7]      [,8]
V1  0.0000000  0.0000000
V2  0.0000000  0.0000000
V3  0.0000000  0.0000000
V4  0.1179272 -0.2851414
V5 -0.2460942  0.1214254
V6  0.1644385 -0.3312662
V7  1.0000000 -0.7482048
V8 -0.7482048  1.0000000

```

Time difference of 0.1180069 secs

Comment on results

When comparing the results from the output with those obtained in section 1.2.2, it is noticeable that the criterion increased when prior information on FIM is used. It increased from 182.5 to 599.3. There are evident improvements also in the RSE of both fixed and random effects, which are lower here than in point 1.2.2.

Comparing the output of one group design using prior information with the output of the two groups design in section 1.2.3, it is evident that results are the same. Indeed, the prior information on FIM used was the FIM obtained for the first group design, group that was evaluated in section 1.2.1.

1.3. Individual Fisher Information Matrix (I-FIM)

Evaluation of the Individual Fisher Information Matrix for the elementary design ξ_1 and then ξ_2 .

1.3.1. Elementary Design ξ_1

INPUT FILE

```

#Name of the project
#-----
project<-"1.3.1"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.33,1.5,5,12))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(1)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

```

OUTPUT FILE

```
PFIM 4.0

Project: 1.3.1

Date: Mon Jan 27 16:33:26 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
                times subjects doses
1 c(0.33, 1.5, 5, 12)          1    100

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Individual Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 1.3302192  1.159014 -0.2522055  0.000000  0.000000
[2,] 1.1590137 214.546128  3.6213766  0.000000  0.000000
[3,] -0.2522055  3.621377  0.1479803  0.000000  0.000000
[4,] 0.0000000  0.000000  0.0000000 12.85799 21.91468
[5,] 0.0000000  0.000000  0.0000000 21.91468 66.59115

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta StdError      RSE
ka  2.00 1.4531150 72.65575 %
k   0.25 0.1228738 49.14952 %
V   15.00 5.6733768 37.82251 %

----- Standard deviation of residual error -----
```

```

          Sigma StdError      RSE
sig.interA 0.50 0.4208513 84.17025 %
sig.slopeA 0.15 0.1849298 123.28655 %

***** DETERMINANT *****

3317.958

***** CRITERION *****

5.060276

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min          5.053743          0.02977512
max          214.613529          1.38102331
max/min      42.466252          46.38178541

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 1.0000000 -0.6884871  0.8014327  0.0000000  0.0000000
[2,] -0.6884871  1.0000000 -0.8305558  0.0000000  0.0000000
[3,]  0.8014327 -0.8305558  1.0000000  0.0000000  0.0000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000 -0.7489284
[5,]  0.0000000  0.0000000  0.0000000 -0.7489284  1.0000000

Time difference of 0.02400112 secs

```

1.3.2. Elementary Design ξ_2

INPUT FILE

```

#Name of the project
#-----
project<-"1.3.2"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1,3,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(1)

```

OUTPUT FILE

```
PFIM 4.0

Project: 1.3.2

Date: Mon Jan 27 17:02:58 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
      times subjects doses
1 c(1, 3, 8)          1  100

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Individual Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 0.35331883  1.538575 -0.08983448  0.000000  0.000000
[2,] 1.53857495 214.538102  3.84156478  0.000000  0.000000
[3,] -0.08983448  3.841565  0.13399054  0.000000  0.000000
[4,] 0.00000000  0.000000  0.00000000  7.836682 17.89450
[5,] 0.00000000  0.000000  0.00000000 17.894502 60.29574

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta StdError      RSE
ka  2.00 2.7633469 138.16734 %
k   0.25 0.1464156  58.56623 %
V  15.00 6.3313534  42.20902 %

----- Standard deviation of residual error -----
```

```

          Sigma  StdError      RSE
sig.interA  0.50 0.6291957 125.8391 %
sig.slopeA  0.15 0.2268342 151.2228 %

***** DETERMINANT *****
279.0063

***** CRITERION *****
3.08406

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min          2.314023          0.02215148
max          214.617932          0.38532766
max/min      92.746652          17.39511865

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,]  1.0000000 -0.7437627  0.7857500  0.0000000  0.0000000
[2,] -0.7437627  1.0000000 -0.8806589  0.0000000  0.0000000
[3,]  0.7857500 -0.8806589  1.0000000  0.0000000  0.0000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000 -0.8232092
[5,]  0.0000000  0.0000000  0.0000000 -0.8232092  1.0000000

Time difference of 0.02400088 secs

```

1.4. Bayesian Fisher Information Matrix (B-FIM)

Evaluation of the Bayesian Fisher Information Matrix for the elementary design ξ_1 and then ξ_2 .

1.4.1. Elementary Design ξ_1

INPUT FILE

```

#Name of the project
#-----
project<-"1.4.1"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.33,1.5,5,12))

```



```

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(1)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

```

OUTPUT FILE

```

PFIM 4.0

Project: 1.4.1

Date: Mon Jan 27 17:09:24 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
           times subjects doses
1 c(0.33, 1.5, 5, 12)      1  100

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Bayesian Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

           [,1]      [,2]      [,3]
[1,]  1.5802192   1.159014 -0.2522055
[2,]  1.1590137 278.546128  3.6213766
[3,] -0.2522055   3.621377  0.1924248

***** EXPECTED STANDARD ERRORS *****
----- Fixed Effects Parameters -----

```

```

      Beta   StdError   RSE Shrinkage
ka  2.00 0.96051649 48.02582 % 23.06480 %
k   0.25 0.07402618 29.61047 % 35.07121 %
V   15.00 3.16229457 21.08196 % 44.44492 %

***** DETERMINANT *****

43.88159

***** CRITERION *****

3.527179

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min      9.700529e-02          NA
max      2.785981e+02          NA
max/min  2.871988e+03          NA

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]
[1,]  1.0000000 -0.3642010  0.5585526
[2,] -0.3642010  1.0000000 -0.5855403
[3,]  0.5585526 -0.5855403  1.0000000

Time difference of 0.02100205 secs

```

1.4.2. Elementary Design ξ_2

INPUT FILE

```

#Name of the project
#-----
project<-"1.4.2"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1,3,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(1)

```

OUTPUT FILE

```
PFIM 4.0

Project: 1.4.2

Date: Tue Jan 28 08:26:36 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
      times subjects doses
1 c(1, 3, 8)          1  100

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Bayesian Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]
[1,] 0.60331883 1.538575 -0.08983448
[2,] 1.53857495 278.538102 3.84156478
[3,] -0.08983448 3.841565 0.17843498

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

Beta StdError      RSE Shrinkage
ka 2.00 1.419173 70.95865 % 50.35130 %
k 0.25 0.075761 30.30440 % 36.73427 %
V 15.00 3.090213 20.60142 % 42.44184 %

***** DETERMINANT *****

17.34976
```

```

***** CRITERION *****
2.588796

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min      0.1004813          NA
max     278.5996083          NA
max/min 2772.6525284          NA

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]
[1,] 1.0000000 -0.3321737  0.4065396
[2,] -0.3321737  1.0000000 -0.6046221
[3,]  0.4065396 -0.6046221  1.0000000

Time difference of 0.02000093 secs

```

Comment on results

Comparing examples 1.3 and 1.4, it is noticeable that the evaluation of B-FIM leads to smaller RSE particularly for the parameter k_a , which in the evaluation of I-FIM is very high, above all for the second elementary design, reaching the value of 138 %.

Comparing the two elementary designs, for both I-FIM and B-FIM, results are overall better with ξ_1 than with ξ_2 , as the variability on k_a is kept lower and the criterion is slightly higher. For B-FIM, shrinkage values are provided and they show that with ξ_1 it is obtained more information than with ξ_2 .

1.5. Delta Method

Using the delta method available in the R package “car”, we can compute the standard error (SE) of any derived parameters.

Here we evaluate the SE of the Clearance ($Cl = k \cdot V$) for the Population, Individual and Bayesian Fisher Matrix using design ξ_1 . We use M_F stored in the `outputFIM` for each of these three cases, or the one directly obtained in R console after running `PFIM()`.

1.5.1. P-FIM

```

setwd("C:\\Users\\Guilia\\Desktop\\ExamplesPFIM4\\EVALUATION\\EXAMPLE
1\\1.1_UserDefMod_1.2.1")

Beta<-c(2,0.25,15) # provided from the stdin.r file in example
#1.1_UserDefMod_1.2.1.
names(Beta) <- c("ka","k","V")

fishmat <- read.table("FIM.txt",sep="")

```

```
colnames(fishmat) <- paste(1:8)

varcov<-solve(fishmat)[1:3,1:3]#we only include the columns of the fix effects

DMcl <- deltaMethod(Beta,"k*V",vcov.=varcov)
> DMcl
      Estimate      SE
k * V      3.75 0.1730348
```

1.5.2. I-FIM

```
directory<-"C:\\Users\\Guilia\\Desktop\\ExamplesPFIM4\\EVALUATION\\EXAMPLE
1\\1.3.1"
directory.program<-"C:\\Users\\Guilia\\Documents\\PFIM4.0\\Program"

result<-PFIM()
fishmat<-result$mfisher
colnames(fishmat) <- paste(1:5)

varcov<-solve(fishmat)[1:3,1:3]
DMcl <- deltaMethod(Beta,"k*V",vcov.=varcov)
> DMcl
      Estimate      SE
k * V      3.75 1.032634
```

1.5.3. B-FIM

```
directory<-"C:\\Users\\Guilia\\Desktop\\ExamplesPFIM4\\EVALUATION\\EXAMPLE
1\\1.4.1"

colnames(fishmat) <- paste(1:3)

varcov<-solve(fishmat)[1:3,1:3]
DMcl <- deltaMethod(Beta,"k*V",vcov.=varcov)
> DMcl
      Estimate      SE
k * V      3.75 0.9110153
```

1.6. Evaluation of FIM with Fixed Parameter

Another new feature of v4.0 is to assume that a parameter is known (fixed) and not estimated.

Here we evaluated Population, Individual and Bayesian Fisher Information Matrix for the design ξ_1 assuming that the parameter k_a is fixed (and has no variability).

1.6.1. P-FIM

INPUT FILE

```
#Name of the project
#-----
project<-"1.6.1"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"
```

```

#Name of the fixed effects parameters
#-----
parameters<-c("ka","k","v")

#Fixed effects parameters values
#-----
beta<-c(2,0.25,15)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
omega<-diag(c(0,0.25,0.1))

```

OUTPUT FILE

```

PFIM 4.0

Project:  1.6.1

Date:  Mon Jan 27 17:15:03 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
                times subjects doses
1 c(0.33, 1.5, 5, 12)      200  100

Random effect model: Trand =  2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Computation of the Population Fisher information matrix: option =  1

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]    [,2]    [,3]    [,4]    [,5]    [,6]
[1,] 8906.33251 50.88726  0.00000  0.00000  0.00000  0.00000
[2,]  50.88726  6.17076  0.00000  0.00000  0.00000  0.00000
[3,]  0.00000  0.00000 774.63632  91.03759 212.6390 298.6706
[4,]  0.00000  0.00000  91.03759 4819.28232 362.6152 1318.2543
[5,]  0.00000  0.00000 212.63905 362.61524 1502.9442 2413.8017
[6,]  0.00000  0.00000 298.67055 1318.25428 2413.8017 6880.7738

```

```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta      StdError      RSE
k  0.25  0.01085503  4.342010 %
V  15.00  0.41239269  2.749285 %

----- Variance of Inter-Subject Random Effects -----

      omega2      StdError      RSE
k  0.25  0.03667690  14.67076 %
V  0.10  0.01482801  14.82801 %

----- Standard deviation of residual error -----

      Sigma      StdError      RSE
sig.interA  0.50  0.03957486  7.914972 %
sig.slopeA  0.15  0.01861316  12.408776 %

***** DETERMINANT *****

8.000286e+17

***** CRITERION *****

963.4982

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects  VarianceComponents
min      8350.060448      5.879818
max      8906.623447      795.196613
max/min      1.066654      135.241698

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,]  1.0000000  -0.2170653  0.0000000  0.0000000  0.0000000  0.0000000
[2,]  -0.2170653  1.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[3,]  0.0000000  0.0000000  1.0000000  -0.02729577  -0.15390246  0.03342554
[4,]  0.0000000  0.0000000  -0.02729577  1.0000000  0.06116443  -0.19591071
[5,]  0.0000000  0.0000000  -0.15390246  0.06116443  1.0000000  -0.74204223
[6,]  0.0000000  0.0000000  0.03342554  -0.19591071  -0.74204223  1.0000000

Time difference of 0.02500105 secs

```

Comment on results

Comparing the output of this example with the one in section 1.2.1, we can see that the standard errors are slightly reduced in this example.

1.6.2. I-FIM

INPUT FILE

```
#Name of the project
#-----
project<-"1.6.2"

outputFIM<-"FIM.txt";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"

##### END ANALYTICAL MODEL OPTION #####
#Name of the fixed effects parameters
#-----
parameters<-c("ka","k","v")

#Fixed effects parameters values
#-----
beta<-c(2,0.25,15)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
```

OUTPUT FILE

```
PFIM 4.0

Project:  1.6.2

Date:  Mon Jan 27 17:21:44 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
                times subjects doses
1 c(0.33, 1.5, 5, 12)          1    100

Variance error model response A : ( 0.5 + 0.15 *f)^2
```


Computation of the Individual Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]
[1,]	214.546128	3.6213766	0.000000	0.000000
[2,]	3.621377	0.1479803	0.000000	0.000000
[3,]	0.000000	0.0000000	12.85799	21.91468
[4,]	0.000000	0.0000000	21.91468	66.59115

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
k	0.25	0.08911405	35.64562 %
V	15.00	3.39316156	22.62108 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.4208513	84.17025 %
sig.slopeA	0.15	0.1849298	123.28655 %

***** DETERMINANT *****

7006.011

***** CRITERION *****

9.148875

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	74.395393	0.08682947
max	214.607279	0.08682947
max/min	2.884685	1.00000000

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]
[1,]	1.0000000	-0.6427045	0.0000000	0.0000000
[2,]	-0.6427045	1.0000000	0.0000000	0.
[3,]	0.0000000	0.0000000	1.0000000	-0.74892840000000
[4,]	0.0000000	0.0000000	-0.7489284	1.0000000

Time difference of 0.02300096 secs

Comments on the results

As for PFIM, fixing ka resulted in slight improvements of the Relative Standard Errors with respect to the results in section 1.3.1.

1.6.3. B-FIM

INPUT FILE

```
#Name of the project
#-----
project<-"1.6.3"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
```

OUPTUT FILE

```
PFIM 4.0

Project: 1.6.3

Date: Mon Jan 27 17:27:30 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Design:
Sample times for response: A
                times subjects doses
1 c(0.33, 1.5, 5, 12)      1 100

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2
```

Computation of the Bayesian Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

```
          [,1]      [,2]
[1,] 278.546128 3.6213766
[2,]   3.621377 0.1924248
```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE	Shrinkage
k	0.25	0.06894209	27.57684 %	30.41928 %
V	15.00	2.62302564	17.48684 %	30.57895 %

***** DETERMINANT *****

40.48481

***** CRITERION *****

6.362767

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	0.1453187	NA
max	278.5932338	NA
max/min	1917.1190240	NA

***** CORRELATION MATRIX *****

```
          [,1]      [,2]
[1,] 1.0000000 -0.4946462
[2,] -0.4946462 1.0000000
```

Time difference of 0.02200103 secs

2. Example 2: PK and immediate response PD model using the libraries of PK and PD Models (ODE)

The purpose is to evaluate a population design using a PKPD model. The PK model is a one compartment model with an infusion of 1 hour and a Michaelis-Menten elimination. The PD model is an immediate response model with linear drug action and no baseline. The PKPD model is described by a differential equation system using the function `create_formED` implemented in the file `CreateModel_PKPDdesign.r`.

The design to be evaluated is composed of one group of 100 subjects with a dose of 100 and sampling times at 0.5, 2, 30, 49, 180 for the PK and 0.5, 2, 14, 110, 150 for the PD.

2.1. Model File

```
source(file.path(directory.program, "CreateModel_PKPDdesign.r"))
create_formED(infusion_lcpt_VVmkm, immed_lin_null, dose=100, TInf=1)
# The differential equation system is created in the file model_created.r
```

2.2. Input File (P-FIM)

```
#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"EXAMPLE 2"

#Name of the file containing the PK or PD model
#-----
file.model<-"model.R"

#Name of the output file for the results and for the Fisher information matrix
#-----
output<-"Stdout.r";
outputFIM<-"";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"EVAL"

#To display only graphs of models and/or sensitivity functions before evaluating
the Fisher Information matrix
graph.only<-F
```

```

#Block diagonal Fisher information matrix (option<-1) or complete Fisher
information matrix (option<-2)
#-----
option<-1

#Number of responses
#-----
nr<-2

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"DE"
##### DIFFERENTIAL EQUATION OPTION #####
#####

#Initial time for which initial conditions are given
#-----
time.condinit<-0

#Identical initial conditions in each elementary design (Yes=T, No=F)
#-----
condinit.identical<-T

# If 'Yes', enter once the expression of the initial values of the system at the
initial time
# else, enter the vectors of the initial conditions for each elementary design
# If initial values depend on the parameters to be estimated,
# enter this parameter into the expression without any quotation marks
#-----
condinit<-expression(c(0,0))

# Error tolerance for solving differential equations
#-----

RtolEQ<-1e-08
AtolEQ<-1e-08
Hmax<-Inf

##### END DIFFERENTIAL EQUATION OPTION #####

#Name of the fixed effects parameters
#-----
parameters<-c("V", "Vm", "km", "Alin")

#Fixed effects parameters values
#-----
beta<-c(12.2,0.082,0.37,0.1)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(F,F,F)

```

```

#Number of occasions
#-----
n_occ<-1

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0.25,0.25,0,0.25))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0,0,0))

#Standard deviation of residual error (sig.inter+sig.slope*f)^2:
#-----
sig.interA<-0
sig.slopeA<-0.2

sig.interB<-0.1
sig.slopeB<-0

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5, 2, 30, 49, 180))
protB<-list(c(0.5, 2, 14, 110, 150))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(100)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

#If 'proportions of subjects' give the total number of samples
#-----
#Ntot<-40
##### GRAPH SPECIFICATION OPTION #####

#graphical representation (Yes=T, No=F)
#-----
graph.logical<-T

#Vector of Names on Y axes for each response
#-----
names.datax<-c("Time","Time")

#Vector of Names on Y axes for each response
#-----
names.datay<-c("Concentration","Effet")

#Controls logarithmic axes for the graphical representation.
#Values "xy", "x", or "y" produce log-log or log-x or log-y axes.

```

```

#(For standard graphic, log.logical<-F)
#-----
#log.logical<-'y'
log.logical<-F

#Vector of lower and upper sampling times for the graphical representation
#-----
graph.infA<-c(0)
graph.supA<-c(180)
graph.infB<-c(0)
graph.supB<-c(180)

#Vector of lower and upper concentration for the graphical representation
#-----
y.rangeA<-NULL # default range
#y.range<-c(0,10)

##### END OF GRAPH SPECIFICATION OPTION #####

```

2.3. Output File

```

PFIM 4.0

Project:  EXAMPLE 2

Date:  Fri Mar 21 15:57:00 2014

***** INPUT SUMMARY *****

Differential Equations form of the model:

function(t,y,p){
V<-p[1]
Vm<-p[2]
km<-p[3]
Alin<-p[4]
pk<-y[1:1]
pd<-y[2:2]
conc<-y[1]
if(t<=1){
dpk1<-(100/(1*V))+(-Vm)*pk[1]/(km+pk[1])}
else{
dpk1<-(-Vm)*pk[1]/(km+pk[1])}
dpd1<-0
pdIm<-Alin*conc
return(list(c(dpk1,dpd1),c(pk[1],pdIm)))
}

Design:
Sample times for response: A
                times subjects
1 c(0.5, 2, 30, 49, 180)      100

Sample times for response: B
                times subjects
1 c(0.5, 2, 14, 110, 150)     100

```

```

Initial Conditions at time 0 :

0 0

Random effect model: Trand = 2

Variance error model response A : ( 0 + 0.2 *f)^2
Variance error model response B : ( 0.1 + 0 *f)^2

Error tolerance for solving differential equations system: RtolEQ = 1e-08 , AtolEQ
= 1e-08 , Hmax = Inf

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]      [,8]      [,9]
[1,]  2.6770341  9.444202  -57.39048  -0.7254162  0.000000e+00  0.0000000  0.000000e+00  0.000000e+00  0.0000000
[2,]  9.4442023 50919.213161  7982.73905  657.4076389  0.000000e+00  0.0000000  0.000000e+00  0.000000e+00  0.0000000
[3,] -57.3904833  7982.739050 33057.82792 -6714.1078293  0.000000e+00  0.0000000  0.000000e+00  0.000000e+00  0.0000000
[4,] -0.7254162  657.407639 -6714.10783  38772.8151987  0.000000e+00  0.0000000  0.000000e+00  0.000000e+00  0.0000000
[5,]  0.0000000  0.0000000  0.000000  0.0000000  7.938111e+02  0.4463216  3.916193e-03  2.546794e+00  8.097541
[6,]  0.0000000  0.0000000  0.000000  0.0000000  4.463216e-01  586.1230244  1.453005e-01  9.449987e+01  301.234913
[7,]  0.0000000  0.0000000  0.000000  0.0000000  3.916193e-03  0.1453005  7.516656e+02  8.332548e-01  116.540931
[8,]  0.0000000  0.0000000  0.000000  0.0000000  2.546794e+00  94.4998735  8.332548e-01  1.725786e+04  1714.876645
[9,]  0.0000000  0.0000000  0.000000  0.0000000  8.097541e+00  301.2349133  1.165409e+02  1.714877e+03  64572.948196

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta      StdError      RSE
V      12.200  0.624822332  5.121495 %
Vm     0.082  0.004534793  5.530235 %
km     0.370  0.005841524  1.578790 %
Alin   0.100  0.005182493  5.182493 %

----- Variance of Inter-Subject Random Effects -----

      omega^2      StdError      RSE
V      0.25  0.03549293  14.19717 %
Vm     0.25  0.04137028  16.54811 %
Alin   0.25  0.03647948  14.59179%

----- Standard deviation of residual error -----

      Sigma      StdError      RSE
sig.slopeA  0.2  0.007625037  3.812519 %
sig.interB  0.1  0.003945487  3.945487 %

***** DETERMINANT *****

5.994606e+31

***** CRITERION *****

3395.176

```


***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

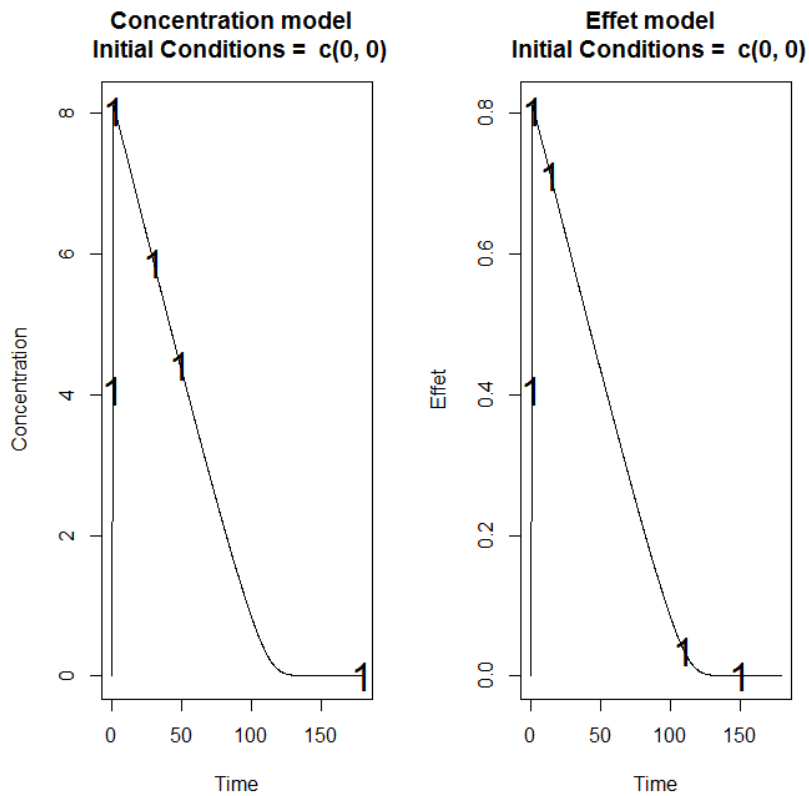
```

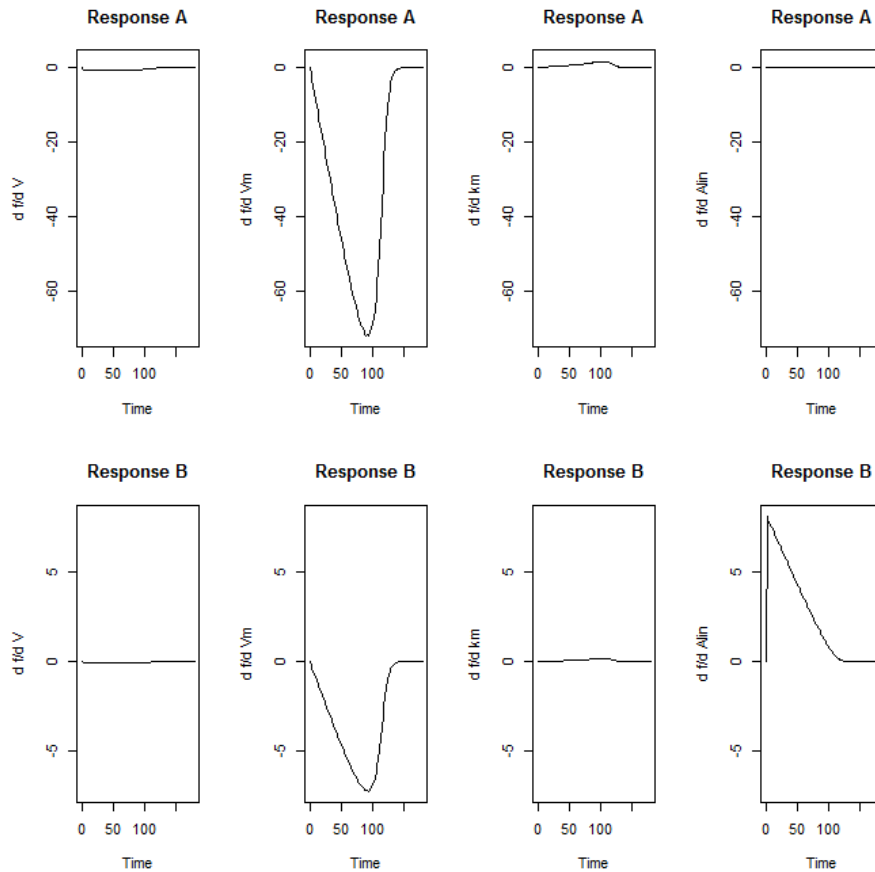
FixedEffects VarianceComponents
min      26521.376944      2.561446
max      64636.681018      17196.209993
max/min   2.437154      6713.478513
    
```

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]	[,7]	[,8]	[,9]
[1,]	1.00000000	-0.06785010	0.2062542	0.04352023	0.000000e+00	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[2,]	-0.06785010	1.00000000	-0.2101514	-0.05600795	0.000000e+00	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[3,]	0.20625422	-0.21015142	1.00000000	0.19876898	0.000000e+00	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[4,]	0.04352023	-0.05600795	0.1987690	1.00000000	0.000000e+00	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[5,]	0.00000000	0.00000000	0.00000000	0.00000000	1.000000e+00	-0.0005826757	1.311462e-05	-0.0006146867	-0.001068259
[6,]	0.00000000	0.00000000	0.00000000	0.00000000	-5.826757e-04	1.0000000000	5.840399e-04	-0.0272653545	-0.047524772
[7,]	0.00000000	0.00000000	0.00000000	0.00000000	1.311462e-05	0.0005840399	1.000000e+00	0.0006127203	-0.016746938
[8,]	0.00000000	0.00000000	0.00000000	0.00000000	-6.146867e-04	-0.0272653545	6.127203e-04	1.0000000000	-0.050000216
[9,]	0.00000000	0.00000000	0.00000000	0.00000000	-1.068259e-03	-0.0475247719	-1.674694e-02	-0.0500002164	1.0000000000

Time difference of 3.551203 secs





3. Example 3: PK model with inter-occasion variability

The purpose of this example is to evaluate a design for a PK model including inter-occasion variability. The PK model is a one compartment oral model with first order absorption and first order elimination. The inter-occasion variability was 15% for the three parameters. The design to be evaluated is composed of one group of 40 subjects with a dose of 30 and sampling times at 0.5, 2, 4, 8.

3.1. Model File

```
source(file.path(directory.program,"LibraryPK.r"))
formA<-orall_1cpt_kaVCl()[[1]]
form<-c(formA)
```

3.2. Input File (P-FIM)

```
#####
##                INPUT FILE FOR PFIM 4.0                ##
#####

#Name of the project
#-----
```

```

project<-"EXAMPLE 3"

#Name of the file containing the PK or PD model
#-----
file.model<-"model.R"

#Name of the output file for the results and for the Fisher information matrix
#-----
output<-"Stdout.r";
outputFIM<-"";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"EVAL"

#To display only graphs of models and/or sensitivity functions before evaluating
the Fisher Information matrix
graph.only<-F

#Block diagonal Fisher information matrix (option<-1) or complete Fisher
information matrix (option<-2)
#-----
option<-1

#Number of responses
#-----
nr<-1

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"AF"

##### ANALYTICAL MODEL OPTION #####
#####

#Identical dose in each elementary design (Yes=T, No=F)
#-----
dose.identical<-T

# If 'Yes', enter the value of the dose,
# else, enter the vector of the dose values for each elementary design
#-----
dose<-c(30)
#Vector of the times intervals of each expression

```

```

#-----
boundA<-list(c(0,Inf))

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-F
##### END ANALYTICAL MODEL OPTION #####
#Name of the fixed effects parameters
#-----
parameters<-c("ka","V","Cl")

#Fixed effects parameters values
#-----
beta<-c(1,3.5,2)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(F,F,F)

#Number of occasions
#-----
n_occ<-2

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0.09,0.09,0.09))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0.0225,0.0225,0.0225))

#Standard deviation of residual error (sig.inter+sig.slope*f)^2:
#-----
sig.interA<-0.1
sig.slopeA<-0

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5,2,4,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(40)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1
#If 'proportions of subjects' give the total number of samples
#-----
#Ntot<-40

```

3.3. Output File

```
PFIM 4.0

Project:  EXAMPLE 3

Date:  Tue Jan 28 11:40:35 2014

***** INPUT SUMMARY *****

Analytical function models :

dose/V * ka/(ka - (Cl/V)) * (exp(-(Cl/V) * t) - exp(-ka * t))

Design:
Sample times for response: A
      times subjects doses
1 c(0.5, 2, 4, 8)      40   30

Number of occasions: 2

Random effect model: Trand = 2

Variance error model response A : ( 0.1 + 0 *f)^2

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 326.376227 -15.9864218  3.8839055  0.0000000  0.0000000  0.0000000
[2,] -15.986422  28.3366743  0.9574449  0.0000000  0.0000000  0.0000000
[3,]  3.883905  0.9574449  98.1347183  0.0000000  0.0000000  0.0000000
[4,]  0.000000  0.0000000  0.0000000 1331.5180159  39.1334952  0.7542361
[5,]  0.000000  0.0000000  0.0000000  39.1334952 1506.1906549  0.5614793
[6,]  0.000000  0.0000000  0.0000000  0.7542361  0.5614793 1926.0845868
[7,]  0.000000  0.0000000  0.0000000  665.7590079  19.5667476  0.3771180
[8,]  0.000000  0.0000000  0.0000000  19.5667476  753.0953275  0.2807396
[9,]  0.000000  0.0000000  0.0000000  0.3771180  0.2807396  963.0422934
[10,] 0.000000  0.0000000  0.0000000  486.6656278  340.8242080  22.4031198

      [,7]      [,8]      [,9]      [,10]
[1,]  0.000000 0.000000e+00 0.000000e+00  0.000000
[2,]  0.000000 0.000000e+00 0.000000e+00  0.000000
[3,]  0.000000 0.000000e+00 0.000000e+00  0.000000
[4,]  665.759008 1.956675e+01 3.771180e-01  486.66563
[5,]  19.566748 7.530953e+02 2.807396e-01  340.82421
[6,]  0.377118 2.807396e-01 9.630423e+02  22.40312
[7,] 11180.419592 5.410321e+03 9.653853e+01  2203.93606
[8,]  5410.320690 1.766938e+04 8.459735e+01  1682.30579
[9,]  96.538535 8.459735e+01 3.679038e+04  634.05703
[10,] 2203.936057 1.682306e+03 6.340570e+02  21694.43805

***** EXPECTED STANDARD ERRORS *****
```

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	1.0	0.05615162	5.615162 %
V	3.5	0.19055327	5.444379 %
Cl	2.0	0.10099429	5.049714 %

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE
ka	0.09	0.02797418	31.08242 %
V	0.09	0.02614241	29.04712 %
Cl	0.09	0.02293631	25.48479 %

----- Variance of Inter-Occasion Random Effects -----

	gamma ²	StdError	RSE
ka	0.0225	0.010507710	46.70093 %
V	0.0225	0.008278751	36.79445 %
Cl	0.0225	0.005249319	23.33031 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.1	0.006890589	6.890589 %

***** DETERMINANT *****

4.122587e+32

***** CRITERION *****

1826.068

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	18665.494014	27.46247
max	36847.167079	8088.68560
max/min	1.974079	294.53599

***** CORRELATION MATRIX *****

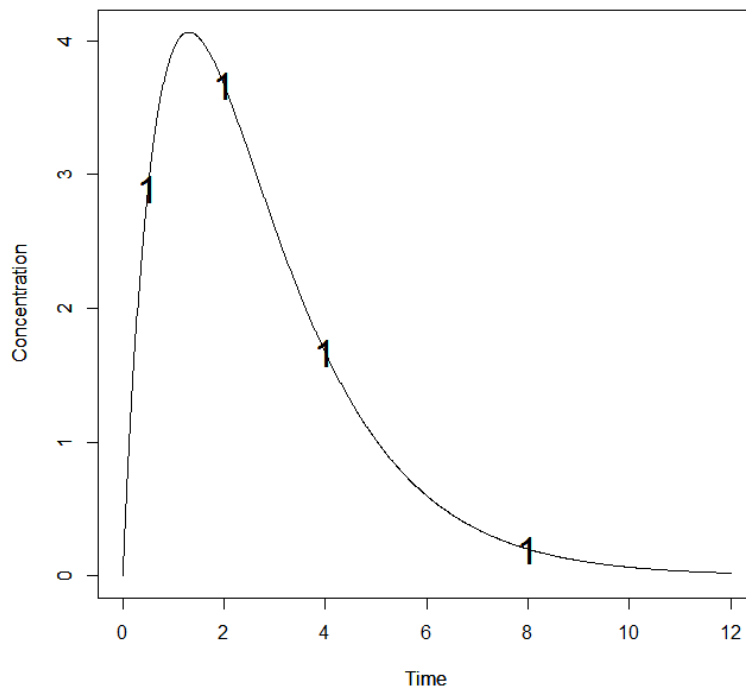
	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	1.00000000	0.16669388	-0.02507303	0.000000000	0.000000000
[2,]	0.16669388	1.00000000	-0.02207619	0.000000000	0.000000000
[3,]	-0.02507303	-0.02207619	1.00000000	0.000000000	0.000000000
[4,]	0.00000000	0.00000000	0.00000000	1.000000000	-0.0347716680
[5,]	0.00000000	0.00000000	0.00000000	-0.0347716680	1.000000000
[6,]	0.00000000	0.00000000	0.00000000	-0.0004678731	-0.0002900774
[7,]	0.00000000	0.00000000	0.00000000	-0.1783934889	0.0676233782

```

[8,] 0.00000000 0.00000000 0.00000000 0.0756870609 -0.1565953140
[9,] 0.00000000 0.00000000 0.00000000 0.0022115605 0.0013941467
[10,] 0.00000000 0.00000000 0.00000000 -0.0684619703 -0.0523532439
      [,6]      [,7]      [,8]      [,9]     [,10]
[1,] 0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
[2,] 0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
[3,] 0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
[4,] -0.0004678731 -0.1783934889 0.0756870609 0.0022115605 -0.0684619703
[5,] -0.0002900774 0.0676233782 -0.1565953140 0.0013941467 -0.0523532439
[6,] 1.0000000000 0.0005358684 0.0002096836 -0.1143575380 -0.0009338365
[7,] 0.0005358684 1.0000000000 -0.3886303379 -0.0014888252 -0.1064630705
[8,] 0.0002096836 -0.3886303379 1.0000000000 -0.0009173038 -0.0305013497
[9,] -0.1143575380 -0.0014888252 -0.0009173038 1.0000000000 -0.0218405019
[10,] -0.0009338365 -0.1064630705 -0.0305013497 -0.0218405019 1.0000000000

```

Time difference of 0.04000306 secs



4. Example 4: PK model with inter-occasion variability and covariate effects (Equivalence test)

The purpose of this example is to evaluate a design for a crossover PK trial with two periods, two sequences: 20 subjects receive treatment A at period 1 then treatment B at period 2; 20 subjects receive treatment B at period 1 then treatment A at period 2. The PK model is a one compartment oral model with first order absorption and first order elimination. We add a gender effect which does not change with the occasion on the volume of distribution (V) and a treatment effect changing with the occasion on the clearance (Cl). The dose is fixed to 30 for the 40 subjects with the same sampling times at 0.5, 2, 6 and 8. With $\alpha=0.05$, we then compute the expected power of the Wald test for

equivalence on the interval $[\ln(0.8)$ and $\ln(1.25)]$ and the number of subjects needed for a given power of 0.9.

4.1. Model File

```
#User-defined model
form<-function(t,p,X){
ka<-p[1]
V<-p[2]
Cl<-p[3]
y<-X/V * ka/(ka - (Cl/V)) * (exp(-(Cl/V) * t) - exp(-ka * t))
return(y)
}
```

4.2. Input File (P-FIM)

```
#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"EXAMPLE 4"

#Name of the file containing the PK or PD model
#-----
file.model<-"model.R"

#Name of the output file for the results and for the Fisher information matrix
#-----
output<-"Stdout.r";
outputFIM<-"";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"EVAL"

#To display only graphs of models and/or sensitivity functions before evaluating
the Fisher Information matrix
graph.only<-F

#Block diagonal Fisher information matrix (option<-1) or complete Fisher
information matrix (option<-2)
#-----
option<-1

#Number of responses
#-----
nr<-1
```



```

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"AF"

##### ANALYTICAL MODEL OPTION #####
#####

#Identical dose in each elementary design (Yes=T, No=F)
#-----
dose.identical<-T

# If 'Yes', enter the value of the dose,
# else, enter the vector of the dose values for each elementary design
#-----
dose<-c(30)

#Vector of the times intervals of each expression
#-----
boundA<-list(c(0,Inf))

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-T

##### END ANALYTICAL MODEL OPTION #####
#Name of the fixed effects parameters
#-----
parameters<-c("ka","V","Cl")

#Fixed effects parameters values
#-----
beta<-c(1,3.5,2)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(F,F,F)

#Number of occasions
#-----
n_occ<-2

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0.09,0.09,0.09))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0.0225,0.0225,0.0225))

```

```

#Standard deviation of residual error (sig.inter+sig.slope*f)^2:
#-----
sig.interA<-0.1
sig.slopeA<-0

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5,2,4,6,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(40)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

#If 'proportions of subjects' give the total number of samples
#-----
#Ntot<-40

#####
#                                     #
#               Covariate model       #
#                                     #
#####

#####
# Covariates not changing with occasion #
#####

#Add covariate to the model (Yes==T No==F)
#-----
covariate.model<-T

#Vector of covariates
#-----
covariate.name<-list(c("Sex"))

#Categories for each covariate (the first category is the reference)
#-----
covariate.category<-list(Sex=c("M","F"))

#Proportions of subjects in each category
#-----
covariate.proportions<-list(Sex=c(0.5,0.5))

#Parameter(s) associated with each covariate
#-----
parameter.associated<-list(Sex=c("V"))

# Values of covariate parameters in covariate model

```

```

# (values of parameters for all other categories than the reference category (for
which beta=0)
# covariate is additive on parameter if additive random effect model (Trand=1)
# covariate is additive on log parameters if exponential random effect model
(Trand=2)
#-----
beta.covariate<-list(Sex=list(c(log(1.2))))

#####
#Covariates changing with occasion #
#####

#Add covariate to the model   (Yes==T No==F)
#-----
covariate_occ.model<-T

#Vector of covariates depending on the occasion
#-----
covariate_occ.name<-list( c("Treat") )

#Categories for each covariate (the first category is the reference)
#-----
covariate_occ.category<-list( Treat=c("A","B") )

#Sequences of values of covariates at each occasion
#Specify as many values in each sequence as number of occasions (n_occ) for each
covariate
#-----

covariate_occ.sequence<-list( Treat=list(c("A","B"),c("B","A")) )

#Proportions of elementary designs corresponding to each sequence of covariate
values
#Specify as many values of proportion as number of sequences defined in
covariate_occ.sequence for each covariate
#-----
-----
covariate_occ.proportions<-list( Treat=c(0.5,0.5) )

#Parameter(s) associated with each covariate
#-----
parameter_occ.associated<-list( Treat=c("C1") )

# Values of covariate parameters in covariate model
# (values of parameters for all other categories than the reference category (for
which beta=0)
# covariate is additive on parameter if additive random effect model (Trand=1)
# covariate is additive on log parameters if exponential random effect model
(Trand=2)
#-----
beta.covariate_occ<-list( Treat=list(c(log(1.1))) )

#Type one error alpha
#-----
alpha<-0.05

```

```

#Compute expected power for comparison test (Yes=T, No=F)
#-----
compute.power<-F

#Compute the number of subjects needed for a given power for comparison test(Yes=T,
No=F)
#-----
compute.nni<-F

#Equivalence interval
interval_eq<-c(log(0.8),log(1.25))

#Compute expected power for equivalence test (Yes=T, No=F)
#-----
compute.power_eq<-T

#Compute the number of subjects needed for a given power for equivalence test
(Yes=T, No=F)
#-----
compute.nni_eq<-T

#Set value the given power
#-----
given.power<-0.9

```

4.3. Output File

```

PFIM 4.0

Project:  EXAMPLE 4

Date:  Tue Jan 28 11:59:42 2014

***** INPUT SUMMARY *****

Analytical function models :

function(t,p,X){
ka<-p[1]
V<-p[2]
Cl<-p[3]
y<-X/V * ka/(ka - (Cl/V)) * (exp(-(Cl/V) * t) - exp(-ka * t))
return(y)
}

Design:
Sample times for response: A
                times subjects doses
1 c(0.5, 2, 4, 6, 8)      40      30

Number of occasions: 2

Random effect model: Trand = 2

Variance error model response A : ( 0.1 + 0 *f)^2

```

Covariate model :

NB: Covariates are additive on log parameters

Covariates not changing with occasion

Covariate 1 : Sex (V)

Categories References Proportions

(1)	M	*	0.5
(2)	F		0.5

Covariates changing with occasion

Covariate 1 : Treat (C1)

Categories References

(1)	A	*
(2)	B	

Sequences Proportions

(1)	A B	0.5
(2)	B A	0.5

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

						s2
	339.888866	-12.1244029	2.2403232	-17.9762700	1.8294873	0.0000000
	-12.124403	29.3831440	0.5085979	52.3319426	0.4561115	0.0000000
	2.240323	0.5085979	98.2790307	0.8587853	98.1435338	0.0000000
s2	-17.976270	52.3319426	0.8587853	183.1617993	0.8270805	0.0000000
	1.829487	0.4561115	98.1435338	0.8270805	953.4932901	0.0000000
	0.000000	0.000000	0.000000	0.000000	0.000000	1444.480034
	0.000000	0.000000	0.000000	0.000000	0.000000	23.0348957
	0.000000	0.000000	0.000000	0.000000	0.000000	0.2509671
	0.000000	0.000000	0.000000	0.000000	0.000000	736.6832299
	0.000000	0.000000	0.000000	0.000000	0.000000	34.0127520
	0.000000	0.000000	0.000000	0.000000	0.000000	32.8694695
	0.000000	0.000000	0.000000	0.000000	0.000000	423.7018814
	0.000000	0.000000	0.000000e+00	0.000000e+00	0.000000	0.000000
	0.000000	0.000000	0.000000e+00	0.000000e+00	0.000000	0.000000
	0.000000	0.000000	0.000000e+00	0.000000e+00	0.000000	0.000000
s2	0.000000	0.000000	0.000000e+00	0.000000e+00	0.000000	0.000000
	0.000000	0.000000	0.000000e+00	0.000000e+00	0.000000	0.000000
	23.034896	0.2509671	7.366832e+02	3.401275e+01	32.86947	423.70188
1620.000487	0.1586320	2.250149e+01	8.207294e+02	33.13093	269.48022	
0.158632	1931.7539827	1.103168e-01	4.364955e-01	966.47638	18.52264	
22.501490	0.1103168	1.214320e+04	4.704703e+03	189.35871	2594.79722	
820.729393	0.4364955	4.704703e+03	2.268081e+04	281.84596	2441.16633	
33.130931	966.4763757	1.893587e+02	2.818460e+02	39310.95582	1152.57041	
269.480223	18.5226438	2.594797e+03	2.441166e+03	1152.57041	36667.05676	

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	1.00000000	0.05466202	5.466202 %
V	3.50000000	0.26457492	7.559283 %
Cl	2.00000000	0.10650676	5.325338 %
beta_V_Sex_F	0.18232156	0.10545703	57.841231 %
beta_Cl_Treat_B	0.09531018	0.03418960	35.871924 %

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE
ka	0.09	0.02678397	29.75997 %
V	0.09	0.02510621	27.89579 %
Cl	0.09	0.02289350	25.43722 %

----- Variance of Inter-Occasion Random Effects -----

	gamma ²	StdError	RSE
ka	0.0225	0.009674645	42.99842 %
V	0.0225	0.007009914	31.15517 %
Cl	0.0225	0.005077443	22.56641 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.1	0.00527779	5.27779 %

***** DETERMINANT *****

1.124604e+38

***** CRITERION *****

1482.234

***** EQUIVALENCE TEST *****

	Beta	90 % CI	exp(Beta)	90 % CI
beta_V_Sex_F	0.18232156	[0.009;0.356]	1.2	[1.009;1.427]
beta_Cl_Treat_B	0.09531018	[0.039;0.152]	1.1	[1.04;1.164]

Type I error = 0.05

Equivalence interval = [log(0.8),log(1.25)]

	Expected_power	Number_subjects_needed (for a given power=0.9)
beta_V_Sex_F	0.1042397	2286.08074
beta_Cl_Treat_B	0.9818745	24.50351

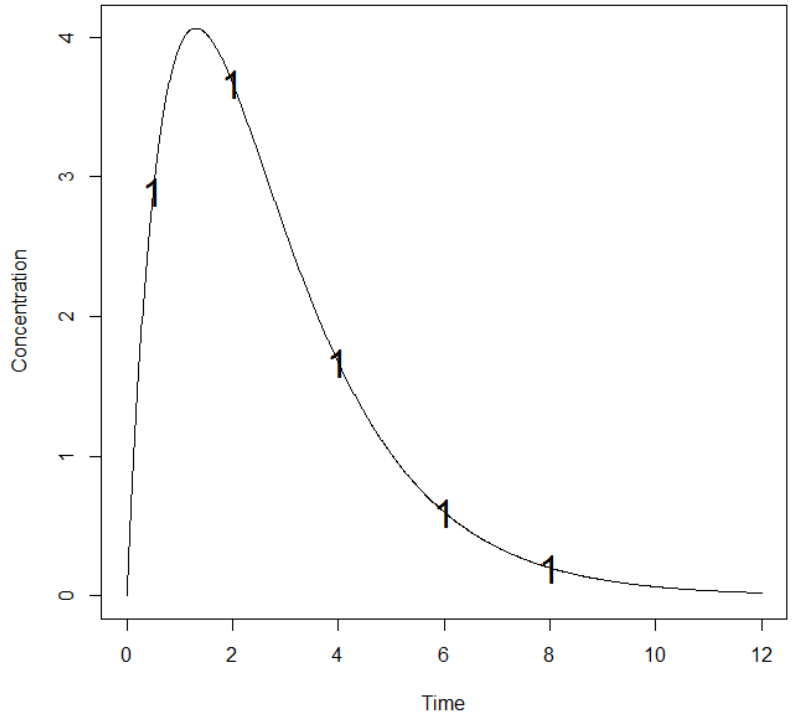
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	1906.77601	13.14078
max	39940.96957	10311.16801
max/min	20.94686	784.66932

***** CORRELATION MATRIX *****

				s2
	1.0000000000	0.1001424653	-0.0130648058	-0.0208510685
	0.1001424653	1.0000000000	-0.0080189689	-0.7116799991
	-0.0130648058	-0.0080189689	1.0000000000	0.0008174403
s2	-0.0208510685	-0.7116799991	0.0008174403	1.0000000000
	0.0008066425	0.0004664293	-0.3205794817	-0.0002392205
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
s2	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	0.0000000000	0.0000000000	0.0000000000	0.0000000000
	1.0000000000	-0.0200347674	0.0001617218	-0.177542015
	-0.0200347674	1.0000000000	0.0002346615	0.040695390
	0.0001617218	0.0002346615	1.0000000000	0.000553334
	-0.1775420153	0.0406953900	0.0005533340	1.000000000
	0.0509009583	-0.1389520685	0.0006633813	-0.282763533
	-0.0020201992	-0.0022890742	-0.1108965657	-0.002912419
	-0.0393506869	-0.0267091218	0.0010141798	-0.095641750
	0.0000000000	0.0000000000		
	0.0000000000	0.0000000000		
	0.0000000000	0.0000000000		
s2	0.0000000000	0.0000000000		
	0.0000000000	0.0000000000		
	-0.002020199	-0.03935069		
	-0.002289074	-0.02670912		
	-0.110896566	0.00101418		
	-0.002912419	-0.09564175		
	-0.005507213	-0.04970801		
	1.0000000000	-0.02893390		
	-0.028933901	1.0000000000		

Time difference of 0.09300518 secs



OPTIMISATION

We illustrate optimisation algorithms with the same examples used in the Evaluation section. As for Evaluation, Example 1 is more detailed with all the new features. For Example 2, 3 and 4, optimisation was performed only with Federow-Wynn (FW) algorithm. We therefore paste in the INPUT FILE only the part concerning optimisation.

1. Example 1: PK Model

1.1. Population Fisher Information Matrix (P-FIM)

Optimize a design for 200 subjects with a dose of 100.

1.1.1. Simplex algorithm

- Initial sampling times vector: $\xi_1 = (0.33, 1.5, 5, 12)$
- Time interval for the optimisation: (0,12)

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.1"
#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"OPT"

#####ONLY FOR OPTIMISATION #####

#Identical sampling times for each response
# (only if you do not have sampling times==NULL)
#-----
identical.times<-F

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
# "FW" for the Fedorov-Wynn algorithm
# "SIMP" for the Simplex algorithm
#-----

algo.option<-"SIMP"

#####
#SIMPLEX SPECIFICATION #
#####

#Optimisation of the proportions of subjects: (Yes=T, No=F)
#-----

subjects.opt<-T

#Vector of lower and upper admissible sampling times
#-----
```

```

lowerA<-c(0)
upperA<-c(12)

#Minimum delay between two sampling times
#-----

delta.time<-0

#Print iteration step (Yes=T, No=F)
#-----

iter.print<-T

#Parameter for initial simplex building (%)
#-----

simplex.parameter<-20

#Maximum iteration number
#-----

Max.iter<-5000

#Relative convergence tolerance
#-----
Rctol<-1e-6

```

OUTPUT FILE

```

PFIM 4.0

Option 1

Project: 1.1.1

Date: Wed Apr 02 11:28:22 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Initial design:

Sample times for response: A
times subjects.prop doses

```

1 c(0.33, 1.5, 5, 12) 200 100

Total number of samples (nr responses): 800

Associated criterion value: 361.7144

Window of the allowed optimised sampling times:

Upper and lower admissible samples times for the response A : [0 : 12]

Minimum delay between two sampling times: 0

Optimisation of the proportions of subjects: TRUE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

***** OPTIMISED DESIGN *****

Number of iterations: 80

Number of function evaluations: 105

Convergence Achieved

Design:

Sample times for response: A

times subjects.prop subjects
1 c(0.325, 1.632, 4.9, 12) 1 200

Associated optimised criterion: 362.4325

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]	[,7]
[1,]	38.769206	82.24941	-3.519151	0.000000	0.000000	0.00000	0.00000
[2,]	82.249405	8247.39675	79.157847	0.000000	0.000000	0.00000	0.00000
[3,]	-3.519151	79.15785	5.006781	0.000000	0.000000	0.00000	0.00000
[4,]	0.000000	0.00000	0.000000	60.122052	4.228103	27.86495	28.36476
[5,]	0.000000	0.00000	0.000000	4.228103	664.253448	220.28782	194.26206

```

[6,] 0.000000 0.00000 0.000000 27.864947 220.287825 3172.65027 302.74176
[7,] 0.000000 0.00000 0.000000 28.364762 194.262062 302.74176 1200.08551
[8,] 0.000000 0.00000 0.000000 86.421975 224.605138 1165.00625 1538.20498
      [,8]
[1,] 0.00000
[2,] 0.00000
[3,] 0.00000
[4,] 86.42197
[5,] 224.60514
[6,] 1165.00625
[7,] 1538.20498
[8,] 4050.28122

```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	2.00	0.17257305	8.628652 %
k	0.25	0.01243021	4.972085 %
V	15.00	0.51585801	3.439053 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
ka	1.00	0.13103070	13.10307 %
k	0.25	0.04013111	16.05244 %
V	0.10	0.01905418	19.05418 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.04125377	8.250754 %
sig.slopeA	0.15	0.02329525	15.530170 %

***** DETERMINANT *****

2.977264e+20

***** CRITERION *****

362.4325

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

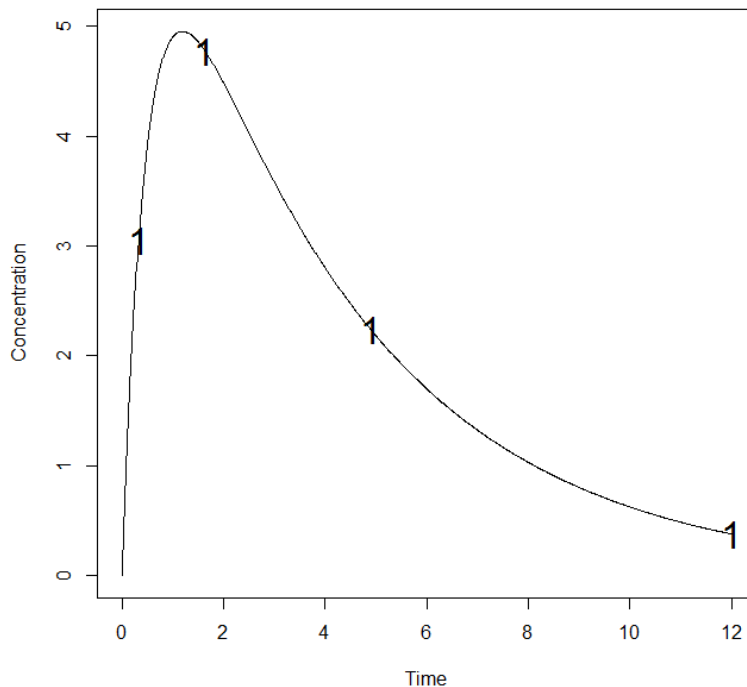
	FixedEffects	VarianceComponents
min	2543.534911	3.704497
max	8248.980105	691.463321
max/min	3.243117	186.655122

```

***** CORRELATION MATRIX *****
      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 1.0000000 -0.2736378  0.3393837  0.0000000000  0.0000000000  0.0000000000
[2,] -0.2736378  1.0000000 -0.4453064  0.0000000000  0.0000000000  0.0000000000
[3,]  0.3393837 -0.4453064  1.0000000  0.0000000000  0.0000000000  0.0000000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000000 -0.0005031654 -0.004823983
[5,]  0.0000000  0.0000000  0.0000000 -0.0005031654  1.0000000000 -0.135409227
[6,]  0.0000000  0.0000000  0.0000000 -0.0048239829 -0.1354092271  1.0000000000
[7,]  0.0000000  0.0000000  0.0000000  0.0225623149 -0.1862954540  0.127956169
[8,]  0.0000000  0.0000000  0.0000000 -0.1340088049  0.0616794121 -0.307811878
      [,7]      [,8]
[1,] 0.00000000  0.00000000
[2,] 0.00000000  0.00000000
[3,] 0.00000000  0.00000000
[4,] 0.02256231 -0.13400880
[5,] -0.18629545  0.06167941
[6,]  0.12795617 -0.30781188
[7,]  1.00000000 -0.68756593
[8,] -0.68756593  1.00000000

Time difference of 0.6150351 secs
sys.self
  0.03

```



Comment on results

The criterion associated with the initial times (ξ_1) was 361.7.

When optimizing with Simplex algorithm, the criterion associated to the optimal times (0.325, 1.632, 4.9, 12) improved of about 1 point, (362.4).

1.1.2. Fedorov-Wynn algorithm

- Allowed sampling times: 0.33,1,1.5,3,5,8,12
 - Maximum total number of points in one elementary protocol: 4
- Compare the result with the one obtained with Simplex algorithm.

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.2"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.33,1.5,5,12))

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
#####
#FEDOROV-WYNN SPECIFICATION #
#####

#Number of sampling windows
#-----
nwindA<-1

#List of vector of the allowed sampling times for each sampling window
#-----
sampwinA<-list(c(0.33,1,1.5,3,5,8,12))

#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c()

#List of vector of allowed number of points to be taken from each sampling window
#-----

nsampA<-list(c(4))

#Maximum total number of sampling times per subject
#-----

nmaxptsA<-4

#Minimum total number of sampling times per subject
#-----

nminptsA<-4
##### END OF OPTIMISATION ALGORITHM OPTION #####
```

OUTPUT FILE

PFIM 4.0

Option: 1

Project: 1.1.2

Date: Tue Jan 28 13:56:35 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

Sample times for response: A
 Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 200 100

Total number of samples: 800

Associated criterion value: 361.7144

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
 Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

Now evaluating the Fisher Information Matrix for the 35 protocols generated

BEST ONE GROUP PROTOCOL:

```

Sample times for response: A
              times freq Subjects doses
1 c(0.33, 1.5, 5, 12)      1      200   100

```

Associated criterion: 361.7144

***** OPTIMISED DESIGN *****

Optimised design:

```

Sample times for response: A
              times      freq  Subjects doses
1 c(0.33, 1, 1.5, 8) 0.35271097 70.542193   100
2 c(0.33, 1.5, 8, 12) 0.61581304 123.162608   100
3 c(0.33, 1.5, 3, 12) 0.03147599   6.295199   100

```

Associated optimised criterion: 371.3125

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]	[,7]
[1,]	37.939211	64.62921	-4.169241	0.00000	0.00000	0.00000	0.00000
[2,]	64.629208	8560.77091	62.570273	0.00000	0.00000	0.00000	0.00000
[3,]	-4.169241	62.57027	4.976251	0.00000	0.00000	0.00000	0.00000
[4,]	0.000000	0.00000	0.000000	57.57818	2.61675	39.23578	28.01875
[5,]	0.000000	0.00000	0.000000	2.61675	718.59967	138.15673	225.75506
[6,]	0.000000	0.00000	0.000000	39.23578	138.15673	3144.16863	286.25131
[7,]	0.000000	0.00000	0.000000	28.01875	225.75506	286.25131	1080.19191
[8,]	0.000000	0.00000	0.000000	89.57049	200.03981	1334.31438	1543.24600

	[,8]
[1,]	0.00000
[2,]	0.00000
[3,]	0.00000
[4,]	89.57049
[5,]	200.03981
[6,]	1334.31438
[7,]	1543.24600
[8,]	4822.65968

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	2.00	0.17491805	8.745902 %
k	0.25	0.01164345	4.657380 %
V	15.00	0.50355898	3.357060 %

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE	
ka	1.00	0.13382715	13.38272	%
k	0.25	0.03891768	15.56707	%
V	0.10	0.01919088	19.19088	%

----- Standard deviation of residual error -----

	Sigma	StdError	RSE	
sig.interA	0.50	0.04302074	8.604149	%
sig.slopeA	0.15	0.02094322	13.962147	%

***** DETERMINANT *****

3.613401e+20

***** CRITERION *****

371.3125

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	2520.328902	3.876959
max	8561.718019	2520.328902
max/min	3.397064	650.078745

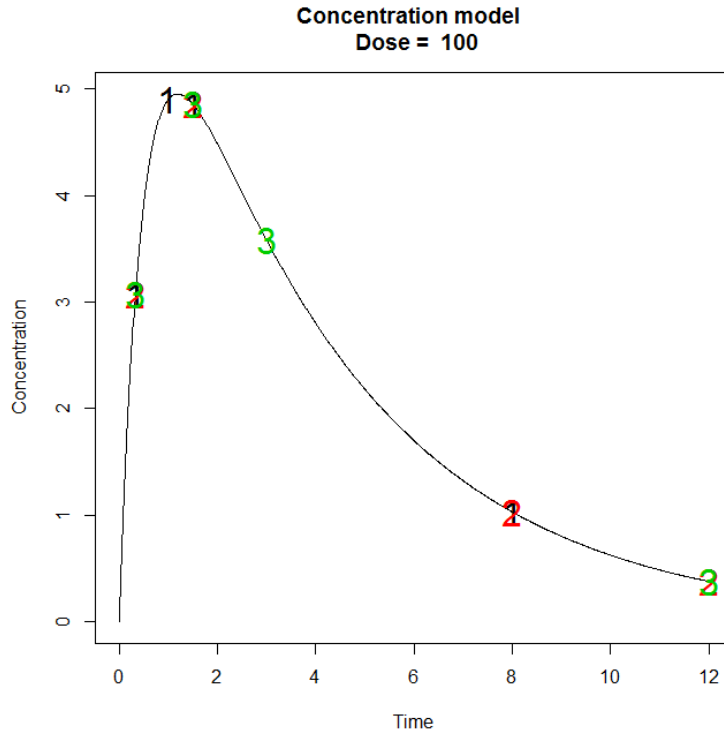
***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	1.0000000	-0.2261971	0.3567942	0.000000000	0.000000000	0.000000000
[2,]	-0.2261971	1.0000000	-0.3565652	0.000000000	0.000000000	0.000000000
[3,]	0.3567942	-0.3565652	1.0000000	0.000000000	0.000000000	0.000000000
[4,]	0.0000000	0.0000000	0.0000000	1.000000000	0.008100681	-0.03710528
[5,]	0.0000000	0.0000000	0.0000000	0.008100681	1.000000000	-0.09027489
[6,]	0.0000000	0.0000000	0.0000000	-0.037105280	-0.090274892	1.000000000
[7,]	0.0000000	0.0000000	0.0000000	-0.002578939	-0.259173157	0.12946239
[8,]	0.0000000	0.0000000	0.0000000	-0.108202443	0.115209250	-0.32726371

	[,7]	[,8]
[1,]	0.000000000	0.000000000
[2,]	0.000000000	0.000000000
[3,]	0.000000000	0.000000000
[4,]	-0.002578939	-0.1082024
[5,]	-0.259173157	0.1152093
[6,]	0.129462393	-0.3272637
[7,]	1.000000000	-0.6698686
[8,]	-0.669868635	1.0000000

Time difference of 1.401081 secs

sys.self
0.13



For this example, the optimisation with Fedorov-Wynn (FW) algorithm led to a criterion of 371.3, higher than the one obtained with the Simplex algorithm (362.4) in point 1.1.1. Relative standard errors are acceptable in both cases (below 20%).

1.1.3. Fixed Parameter

Optimize the design in 1.1 keeping the parameter k_a fixed (assuming no variability on k_a) using the constrains as in 1.1.1 and 1.1.2.

1.1.3.1. Simplex algorithm

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.3.1"
#-----
parameters<-c("ka", "k", "v")

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
omega<-diag(c(0,0.25,0.1))

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----
algo.option<-"SIMP"
```

OUTPUT FILE

PFIM 4.0

Option 1

Project: 1.1.3.1

Date: Thu Apr 17 15:50:27 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

```
Sample times for response: A
                times subjects.prop doses
1 c(0.33, 1.5, 5, 12)                200  100
```

Total number of samples (nr responses): 800

Associated criterion value: 963.4982

Window of the allowed optimised sampling times:

Upper and lower admissible samples times for the response A : [0 : 12]

Minimum delay between two sampling times: 0

Optimisation of the proportions of subjects: TRUE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

***** OPTIMISED DESIGN *****

Number of iterations: 111

Number of function evaluations: 146
Convergence Achieved

Design:

Sample times for response: A
 times subjects.prop subjects
1 c(0.939, 0.942, 8.038, 12) 1 200

Associated optimised criterion: 1081.782

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	9265.40922	35.103559	0.00000	0.00000	0.0000	0.0000
[2,]	35.10356	6.528763	0.00000	0.00000	0.0000	0.0000
[3,]	0.00000	0.000000	838.35750	43.32164	241.6202	202.8993
[4,]	0.00000	0.000000	43.32164	5394.69421	304.1137	1442.1010
[5,]	0.00000	0.000000	241.62021	304.11367	1544.1300	2123.2272
[6,]	0.00000	0.000000	202.89928	1442.10098	2123.2272	7237.6960

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
k	0.25	0.01049631	4.198526 %
V	15.00	0.39541549	2.636103 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
k	0.25	0.03543207	14.17283 %
V	0.10	0.01401500	14.01500 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.03373599	6.747199 %
sig.slopeA	0.15	0.01561541	10.410275 %

```

***** DETERMINANT *****
1.60265e+18
***** CRITERION *****
1081.782
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

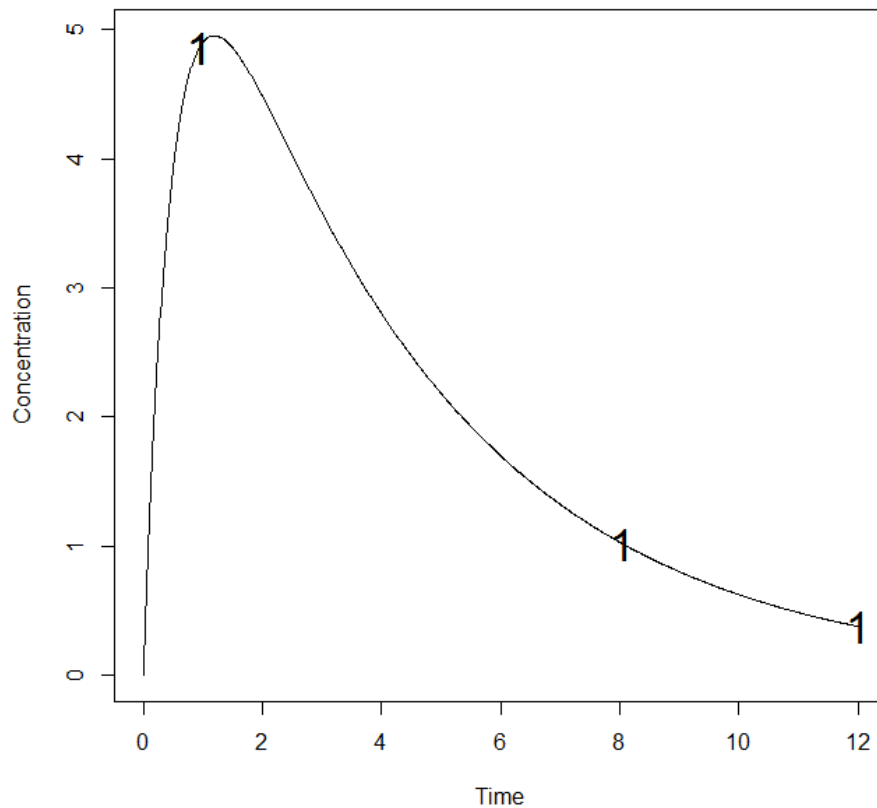
      FixedEffects VarianceComponents
min      8618.806407          6.395675
max      9265.542306          995.276798
max/min   1.075038           155.617157

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 1.0000000 -0.1427261 0.0000000 0.0000000 0.0000000 0.0000000
[2,] -0.1427261 1.0000000 0.0000000 0.0000000 0.0000000 0.0000000
[3,] 0.0000000 0.0000000 1.0000000 -0.01310911 -0.20830125 0.07075104
[4,] 0.0000000 0.0000000 -0.01310911 1.0000000 0.05635404 -0.21370973
[5,] 0.0000000 0.0000000 -0.20830125 0.05635404 1.0000000 -0.63060423
[6,] 0.0000000 0.0000000 0.07075104 -0.21370973 -0.63060423 1.0000000

Time difference of 0.8560491 secs
sys.self
0

```



1.1.3.2. Fedorov-Wynn algorithm

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.3.2"

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#      "FW" for the Fedorov-Wynn algorithm
#      "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
```

OUTPUT FILE

```
PFIM 4.0

Option: 1
```

Project: 1.1.3.2
Date: Tue Jan 28 15:07:15 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){  
ka<-p[1]  
k<-p[2]  
V<-p[3]  
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))  
return(y)  
}
```

Initial design:

Sample times for response: A
 Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 200 100

Total number of samples: 800

Associated criterion value: 963.4982

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
 Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

Now evaluating the Fisher Information Matrix for the 35 protocols generated

BEST ONE GROUP PROTOCOL:

Sample times for response: A
 times freq Subjects doses
1 c(0.33, 1.5, 8, 12) 1 200 100

Associated criterion: 713.2923

***** OPTIMISED DESIGN *****

Optimised design:

Sample times for response: A

	times	freq	Subjects	doses
1	c(0.33, 1.5, 8, 12)	0.8620492	172.40984	100
2	c(0.33, 1, 1.5, 8)	0.1379508	27.59016	100

Associated optimised criterion: 1003.861

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	9164.75153	39.514527	0.00000	0.00000	0.0000	0.000
[2,]	39.51453	6.256111	0.00000	0.00000	0.0000	0.000
[3,]	0.00000	0.000000	821.25546	54.89381	237.7609	215.400
[4,]	0.00000	0.000000	54.89381	4961.58304	359.2888	1388.113
[5,]	0.00000	0.000000	237.76094	359.28876	1546.0399	2284.918
[6,]	0.00000	0.000000	215.40005	1388.11316	2284.9178	6651.617

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
k	0.25	0.01059096	4.236382 %
V	15.00	0.40536223	2.702415 %

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE
k	0.25	0.03583385	14.33354 %
V	0.10	0.01466132	14.66132 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.03713537	7.427074 %
sig.slopeA	0.15	0.01795893	11.972620 %

***** DETERMINANT *****

1.023392e+18


```

***** CRITERION *****
1003.861

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

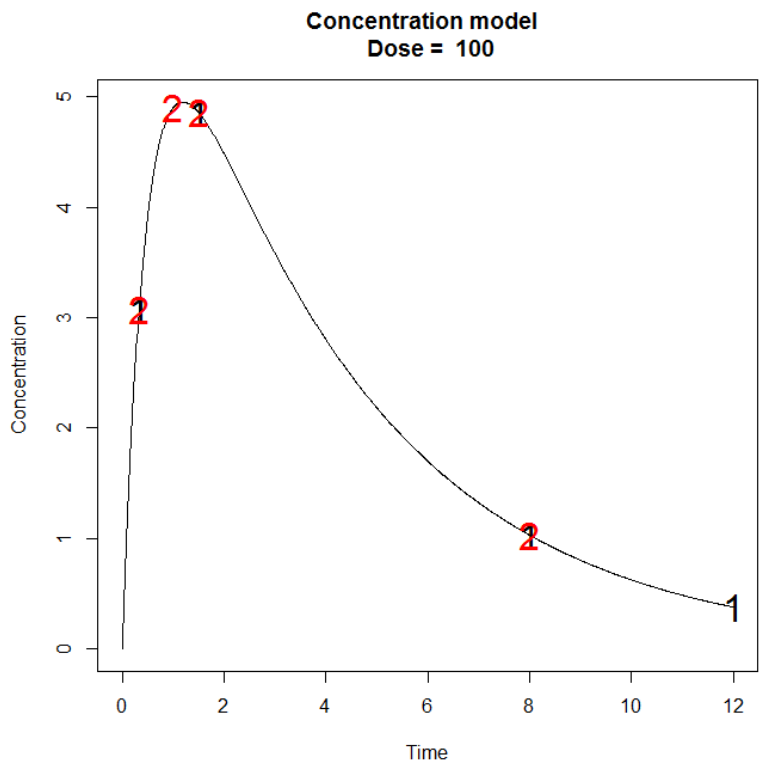
FixedEffects VarianceComponents
min      8171.126965      6.085627
max      9164.922016      8171.126965
max/min   1.121623      1342.692610

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 1.0000000 -0.1650229  0.0000000  0.0000000  0.0000000  0.0000000
[2,] -0.1650229  1.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[3,]  0.0000000  0.0000000  1.0000000 -0.01850452 -0.20874886  0.08681517
[4,]  0.0000000  0.0000000 -0.01850452  1.0000000  0.06481377 -0.21521132
[5,]  0.0000000  0.0000000 -0.20874886  0.06481377  1.0000000 -0.70786839
[6,]  0.0000000  0.0000000  0.08681517 -0.21521132 -0.70786839  1.0000000

Time difference of 0.714041 secs
sys.self
  0.13

```



1.1.4. Fixed Sampling Times

Optimize the design in 1.1 with the Fedorov-Wynn algorithm keeping fixed two sampling times (0.33, 1.5), using the constrains as in 1.1.1 and 1.1.2.

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.4"
##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#      "FW" for the Fedorov-Wynn algorithm
#      "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"

#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c(0.33,1.5)
##### END OF OPTIMISATION ALGORITHM OPTION #####
```

OUTPUT FILE

```
PFIM 4.0

Option: 1

Project: 1.1.4.2

Date: Tue Apr 22 15:30:37 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Initial design:

Sample times for response: A
          Protocol subjects doses
1 c=(0.33, 1.5, 5, 12)          200  100
```

```

Total number of samples: 800

Associated criterion value: 361.7144

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : ( 0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
    Nb of sampling points to be taken in this window, n[ 1 ]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

Now evaluating the Fisher Information Matrix for the 10 protocols generated

BEST ONE GROUP PROTOCOL:

Sample times for response: A
            times freq Subjects doses
1 c(0.33, 1.5, 5, 12)    1    200    100

Associated criterion: 361.7144

***** OPTIMISED DESIGN *****

Optimised design:
Sample times for response: A
            times      freq  Subjects doses
1 c(0.33, 1, 1.5, 8) 0.35271097  70.542193  100
2 c(0.33, 1.5, 8, 12) 0.61581304 123.162608  100
3 c(0.33, 1.5, 3, 12) 0.03147599   6.295199  100

Associated optimised criterion: 371.3125

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

            [,1]      [,2]      [,3]      [,4]      [,5]      [,6]      [,7]
[1,] 37.939211  64.62921 -4.169241  0.00000  0.00000  0.00000  0.00000
[2,] 64.629208 8560.77091 62.570273  0.00000  0.00000  0.00000  0.00000

```

```

[3,] -4.169241  62.57027  4.976251  0.00000  0.00000  0.00000  0.00000
[4,]  0.000000  0.00000  0.000000  57.57818  2.61675  39.23578  28.01875
[5,]  0.000000  0.00000  0.000000  2.61675  718.59967  138.15673  225.75506
[6,]  0.000000  0.00000  0.000000  39.23578  138.15673  3144.16863  286.25131
[7,]  0.000000  0.00000  0.000000  28.01875  225.75506  286.25131  1080.19191
[8,]  0.000000  0.00000  0.000000  89.57049  200.03981  1334.31438  1543.24600

```

[,8]

```

[1,]  0.00000
[2,]  0.00000
[3,]  0.00000
[4,]  89.57049
[5,]  200.03981
[6,]  1334.31438
[7,]  1543.24600
[8,]  4822.65968

```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	2.00	0.17491805	8.745902 %
k	0.25	0.01164345	4.657380 %
V	15.00	0.50355898	3.357060 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
ka	1.00	0.13382715	13.38272 %
k	0.25	0.03891768	15.56707 %
V	0.10	0.01919088	19.19088 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.04302074	8.604149 %
sig.slopeA	0.15	0.02094322	13.962147 %

***** DETERMINANT *****

3.613401e+20

***** CRITERION *****

371.3125

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

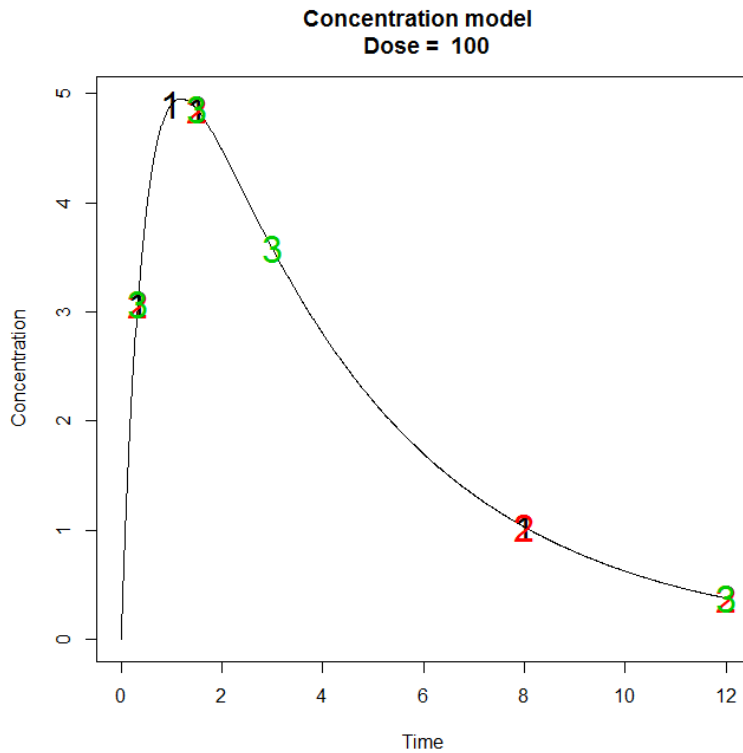
	FixedEffects	VarianceComponents
min	2520.328902	3.876959
max	8561.718019	2520.328902
max/min	3.397064	650.078745

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	1.0000000	-0.2261971	0.3567942	0.000000000	0.000000000	0.000000000
[2,]	-0.2261971	1.0000000	-0.3565652	0.000000000	0.000000000	0.000000000
[3,]	0.3567942	-0.3565652	1.0000000	0.000000000	0.000000000	0.000000000
[4,]	0.0000000	0.0000000	0.0000000	1.000000000	0.008100681	-0.03710528
[5,]	0.0000000	0.0000000	0.0000000	0.008100681	1.000000000	-0.09027489
[6,]	0.0000000	0.0000000	0.0000000	-0.03710528	-0.09027489	1.0000000
[7,]	0.0000000	0.0000000	0.0000000	-0.002578939	-0.259173157	0.12946239
[8,]	0.0000000	0.0000000	0.0000000	-0.108202443	0.115209250	-0.32726371

	[,7]	[,8]
[1,]	0.000000000	0.000000000
[2,]	0.000000000	0.000000000
[3,]	0.000000000	0.000000000
[4,]	-0.002578939	-0.1082024
[5,]	-0.259173157	0.1152093
[6,]	0.129462393	-0.3272637
[7,]	1.000000000	-0.6698686
[8,]	-0.669868635	1.0000000

Time difference of 0.2184012 secs
sys.self
0.06



1.1.5. Fixed Parameter and Fixed Sampling Times

Optimize P-FIM using the Fedorov-Wynn algorithm keeping the parameter k_a fixed (assuming no variability on k_a) and keeping fixed 2 sampling times (0.33, 1.5).

INPUT FILE

```
#####  
##                               INPUT FILE FOR PFIM 4.0                               ##  
#####  
#Name of the project  
#-----  
project<-"1.1.5"  
  
beta.fixed<-c(T,F,F)  
  
##### OPTIMISATION ALGORITHM OPTION #####  
#Character string for choice of the optimisation algorithm:  
#      "FW" for the Fedorov-Wynn algorithm  
#      "SIMP" for the Simplex algorithm  
#-----  
  
algo.option<-"FW"  
  
#####  
#FEDOROV-WYNN SPECIFICATION #  
#####  
#Fixed times (times which will be in all evaluated protocols, corresponding to  
fixed constraints)  
#-----  
fixed.timesA<-c(0.33,1.5)
```

OUTPUT FILE

```
PFIM 4.0  
  
Option: 1  
  
Project: 1.1.5  
  
Date: Tue Apr 22 15:37:32 2014  
  
***** INPUT SUMMARY *****
```

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

Sample times for response: A
 Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 200 100

Total number of samples: 800

Associated criterion value: 963.4982

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
 Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

Now evaluating the Fisher Information Matrix for the 10 protocols generated

BEST ONE GROUP PROTOCOL:

Sample times for response: A
 times freq Subjects doses
1 c(0.33, 1.5, 8, 12) 1 200 100

Associated criterion: 999.542

***** OPTIMISED DESIGN *****

```

Optimised design:
Sample times for response: A
      times      freq  Subjects doses
1 c(0.33, 1.5, 8, 12) 0.8542171 170.84342 100
2 c(0.33, 1, 1.5, 8) 0.1457829 29.15658 100

Associated optimised criterion: 1003.871

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 9157.4312 39.518203 0.00000 0.00000 0.0000 0.0000
[2,] 39.5182 6.261843 0.00000 0.00000 0.0000 0.0000
[3,] 0.0000 0.000000 819.99351 54.90407 237.9162 216.0253
[4,] 0.0000 0.000000 54.90407 4971.04579 358.6388 1386.2277
[5,] 0.0000 0.000000 237.91618 358.63875 1539.3337 2289.9023
[6,] 0.0000 0.000000 216.02528 1386.22774 2289.9023 6693.1956

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta  StdError      RSE
k 0.25 0.0105952 4.238079 %
V 15.00 0.4051770 2.701180 %

----- Variance of Inter-Subject Random Effects -----

      omega2  StdError      RSE
k 0.25 0.03587088 14.34835 %
V 0.10 0.01464165 14.64165 %

----- Standard deviation of residual error -----

      Sigma  StdError      RSE
sig.interA 0.50 0.03727137 7.454275 %
sig.slopeA 0.15 0.01792088 11.947254 %

***** DETERMINANT *****

1.023455e+18

***** CRITERION *****

1003.871

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

```



```

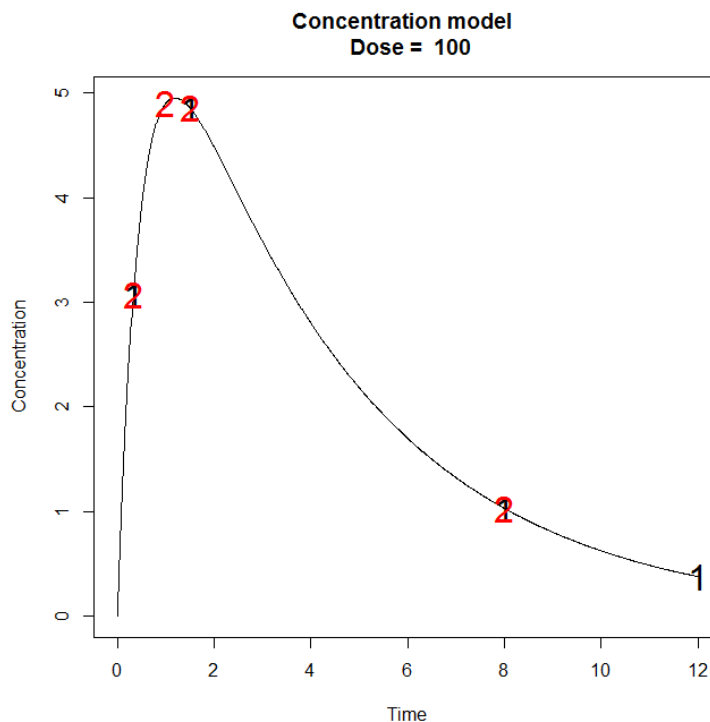
FixedEffects VarianceComponents
min      8203.777970      6.091192
max      9157.601823      8203.777970
max/min   1.116266      1346.826382

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,]  1.0000000 -0.1650286  0.0000000  0.0000000  0.0000000  0.0000000
[2,] -0.1650286  1.0000000  0.0000000  0.0000000  0.0000000  0.0000000
[3,]  0.0000000  0.0000000  1.0000000 -0.01850719 -0.20992982  0.08790201
[4,]  0.0000000  0.0000000 -0.01850719  1.0000000  0.06395325 -0.21352190
[5,]  0.0000000  0.0000000 -0.20992982  0.06395325  1.0000000 -0.70879981
[6,]  0.0000000  0.0000000  0.08790201 -0.21352190 -0.70879981  1.0000000

Time difference of 0.2028 secs
sys.self
0.03

```



1.1.6. Previous information on PFIM

1.1.6.1. Simplex algorithm

For the optimization with previous information, it was used the prior FIM "priorFIM.txt" created in the "priorInfo" folder, stored in the folder of this example.

This prior FIM was created on purpose giving early initial times: (0.33, 1, 1.5).

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.6.1"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"priorFIM.txt"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1.5,8))
#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(100)
```

OUTPUT FILE

```
PFIM 4.0

Option 1

Project: 1.1.6.1

Date: Thu Apr 17 14:00:37 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Initial design:

Sample times for response: A
      times subjects.prop doses
1 c(1.5, 8)          100    100

Total number of samples (nr responses): 200

Associated criterion value: 148.6897
```

Window of the allowed optimised sampling times:

Upper and lower admissible samples times for the response A : [0 : 12]

Minimum delay between two sampling times: 0

Optimisation of the proportions of subjects: TRUE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

***** OPTIMISED DESIGN *****

Number of iterations: 55

Number of function evaluations: 71

Convergence Achieved

Design:

Sample times for response: A

	times	subjects.prop	subjects
1	c(9.395, 12)	1	100

Associated optimised criterion: 215.3191

Computation of the Population Fisher information matrix: option = 1

Previous FIM from file priorFIM.txt

***** FISHER INFORMATION MATRIX *****

	V1	V2	V3	V4	V5	V6	V7
[1,]	19.138168	49.07954	-1.908044	0.000000	0.000000	0.000000	0.000000
[2,]	49.079541	5092.04238	68.482856	0.000000	0.000000	0.000000	0.000000
[3,]	-1.908044	68.48286	2.868817	0.000000	0.000000	0.000000	0.000000
[4,]	0.000000	0.000000	0.000000	28.530155	1.668454	20.98622	14.72601
[5,]	0.000000	0.000000	0.000000	1.668454	386.741448	166.91230	134.56302
[6,]	0.000000	0.000000	0.000000	20.986224	166.912299	1785.26280	145.47772
[7,]	0.000000	0.000000	0.000000	14.726007	134.563018	145.47772	813.66712
[8,]	0.000000	0.000000	0.000000	47.319224	139.206982	601.75932	1099.59528

v8

[1,]	0.00000
[2,]	0.00000
[3,]	0.00000
[4,]	47.31922
[5,]	139.20698
[6,]	601.75932
[7,]	1099.59528
[8,]	3764.97222

***** EXPECTED STANDARD ERRORS *****

```

----- Fixed Effects Parameters -----
      Beta   StdError   RSE
ka  2.00  0.25582962  12.791481 %
k   0.25  0.01839233   7.356932 %
V  15.00  0.79194479   5.279632 %

----- Variance of Inter-Subject Random Effects -----
      omega2  StdError   RSE
ka   1.00  0.18959249  18.95925 %
k    0.25  0.05343693  21.37477 %
V    0.10  0.02485754  24.85754 %

----- Standard deviation of residual error -----
      Sigma   StdError   RSE
sig.interA  0.50  0.04629044   9.258087 %
sig.slopeA  0.15  0.02156098  14.373985 %

***** DETERMINANT *****
4.620192e+18

***** CRITERION *****
215.3191

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects  VarianceComponents
min      1645.644667           1.561834
max      5093.437880           503.888117
max/min      3.095102           322.625898

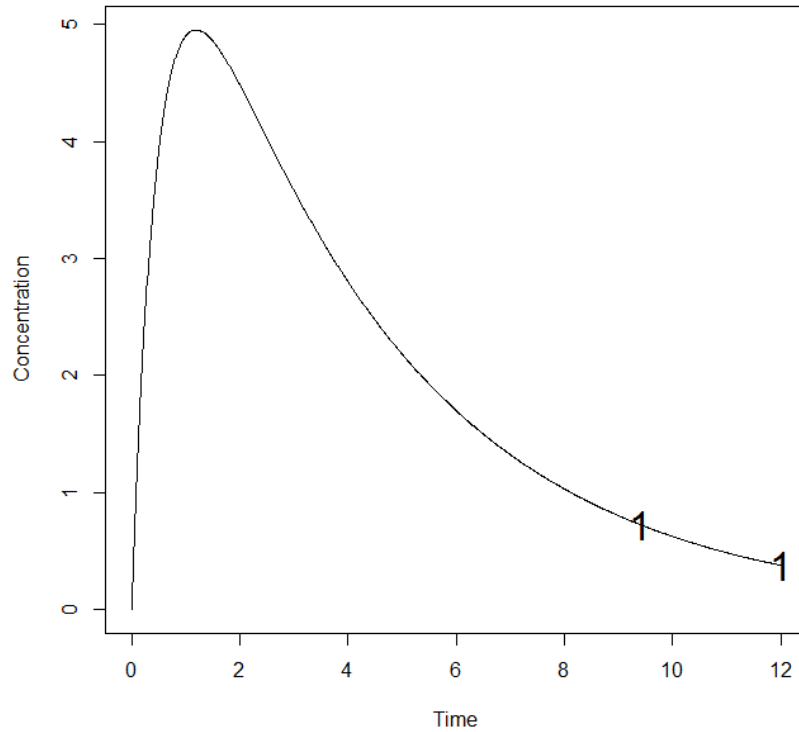
***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
V1  1.0000000 -0.3807142  0.4259198  0.0000000  0.0000000  0.0000000
V2 -0.3807142  1.0000000 -0.6361945  0.0000000  0.0000000  0.0000000
V3  0.4259198 -0.6361945  1.0000000  0.0000000  0.0000000  0.0000000
V4  0.0000000  0.0000000  0.0000000  1.0000000  0.01474447 -0.06375773
V5  0.0000000  0.0000000  0.0000000  0.01474447  1.0000000 -0.19257026
V6  0.0000000  0.0000000  0.0000000 -0.06375773 -0.19257026  1.0000000
V7  0.0000000  0.0000000  0.0000000 -0.01288362 -0.22656742  0.07587018
V8  0.0000000  0.0000000  0.0000000 -0.09204095  0.08428419 -0.20714892

      [,7]      [,8]
V1  0.00000000  0.00000000
V2  0.00000000  0.00000000
V3  0.00000000  0.00000000
V4 -0.01288362 -0.09204095
V5 -0.22656742  0.08428419
V6  0.07587018 -0.20714892
V7  1.00000000 -0.61883269
V8 -0.61883269  1.00000000

Time difference of 0.5510318 secs
sys.self
0

```



1.1.6.2. Best One Group design

INPUT FILE

```
#Name of the project
#-----
project<-"1.1.6.2"

#Previous information for population design (FIM<-"P") only:
#If previous information is available, please specify below the file name;
#otherwise leave it as the default
#-----
previous.FIM<-"priorFIM.txt"

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(1.5,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(100)
```

OUTPUT FILE

PFIM 4.0

Option: 1

Project: 1.1.6.2

Date: Thu Apr 17 10:00:15 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

```
Sample times for response: A
  Protocol subjects doses
1 c=(1.5, 8)      100  100
```

Total number of samples: 200

Associated criterion value: 148.6897

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A

Window 1 : t= 0.33 1 1.5 3 5 8 12

Nb of sampling points to be taken in this window, n[1]= 2

Maximum total number of points in one elementary protocol : 2

Minimum total number of points in one elementary protocol : 2

BEST ONE GROUP PROTOCOL:

Sample times for response: A

```
  times freq Subjects doses
1 c(8, 12)  1      100  100
```

Associated criterion: 214.2912

Previous FIM from file priorFIM.txt

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	V1	V2	V3	V4	V5	V6	V7
[1,]	19.25130	54.34133	-1.802420	0.000000	0.000000	0.000000	0.000000
[2,]	54.34133	5159.92461	73.396138	0.000000	0.000000	0.000000	0.000000
[3,]	-1.80242	73.39614	2.967429	0.000000	0.000000	0.000000	0.000000
[4,]	0.000000	0.000000	0.000000	28.535801	2.132073	21.26297	14.92574
[5,]	0.000000	0.000000	0.000000	2.132073	398.485186	189.64107	117.76653
[6,]	0.000000	0.000000	0.000000	21.262967	189.641070	1798.82796	155.27405
[7,]	0.000000	0.000000	0.000000	14.925744	117.766531	155.27405	777.68829
[8,]	0.000000	0.000000	0.000000	47.680745	147.847133	619.48320	1115.37779

V8

[1,]	0.000000
[2,]	0.000000
[3,]	0.000000
[4,]	47.68074
[5,]	147.84713
[6,]	619.48320
[7,]	1115.37779
[8,]	3804.74812

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	2.00	0.25623615	12.811807 %
k	0.25	0.01887938	7.551752 %
V	15.00	0.79850925	5.323395 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
ka	1.00	0.18957978	18.95798 %
k	0.25	0.05245918	20.98367 %
V	0.10	0.02489123	24.89123 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.04797421	9.594842 %
sig.slopeA	0.15	0.02187599	14.583994 %

***** DETERMINANT *****

4.446667e+18

***** CRITERION *****

214.2912

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

```

FixedEffects VarianceComponents
min      1655.303411      1.536137
max      5161.542600      1655.303411
max/min   3.118185      1077.575115

```

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
V1	1.0000000	-0.4014290	0.4296629	0.0000000	0.0000000	0.0000000
V2	-0.4014290	1.0000000	-0.6630338	0.0000000	0.0000000	0.0000000
V3	0.4296629	-0.6630338	1.0000000	0.0000000	0.0000000	0.0000000
V4	0.0000000	0.0000000	0.0000000	1.0000000	0.0122804	-0.06344937
V5	0.0000000	0.0000000	0.0000000	0.0122804	1.0000000	-0.21184140
V6	0.0000000	0.0000000	0.0000000	-0.06344937	-0.21184140	1.0000000
V7	0.0000000	0.0000000	0.0000000	-0.01245173	-0.18708739	0.06908877
V8	0.0000000	0.0000000	0.0000000	-0.08998746	0.06500474	-0.20304566

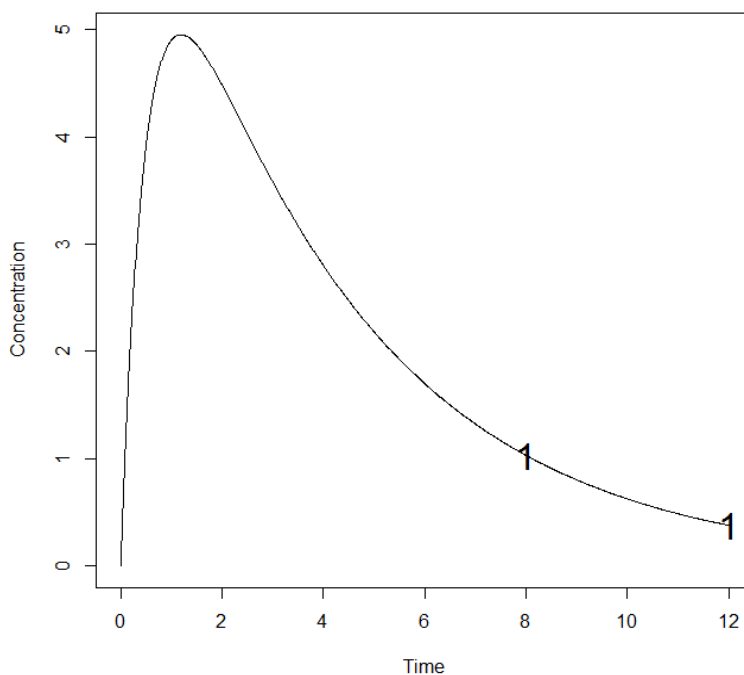
	[,7]	[,8]
V1	0.00000000	0.00000000
V2	0.00000000	0.00000000
V3	0.00000000	0.00000000
V4	-0.01245173	-0.08998746
V5	-0.18708739	0.06500474
V6	0.06908877	-0.20304566
V7	1.00000000	-0.63690284
V8	-0.63690284	1.00000000

Time difference of 0.4670269 secs

sys.self

0.04

Concentration model
Dose = 100



1.2. Individual Fisher Information Matrix (I-FIM)

Optimize the design in 1.1 this time for the Individual Fisher Information Matrix (I-FIM). Use same constraints in 1.1.1 and 1.1.2.

1.2.1. Simplex algorithm

INPUT FILE

```
#Name of the project
#-----
project<-"1.2.1"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"
subjects<-c(1)
#####ONLY FOR OPTIMISATION #####

#Identical sampling times for each response
# (only if you do not have sampling times==NULL)
#-----
identical.times<-F

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----
algo.option<-"SIMP"
```

OUTPUT FILE

```
PFIM 4.0

Option 1

Project: 1.2.1

Date: Fri Apr 04 09:53:14 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

```

Initial design:

Sample times for response: A
                times subjects.prop doses
1 c(0.33, 1.5, 5, 12)                1   100

Total number of samples (nr responses): 4

Associated criterion value: 5.0603

Window of the allowed optimised sampling times:

Upper and lower admissible samples times for the response A : [ 0 : 12 ]

Minimum delay between two sampling times: 0

Optimisation of the proportions of subjects: FALSE

Variance error model response A : ( 0.5 + 0.15 *f)^2

***** OPTIMISED DESIGN *****

Number of iterations: 64
Number of function evaluations: 113
Convergence Achieved

Design:

Sample times for response: A
                times subjects.prop subjects
1 c(0.388, 1.984, 10.109, 10.103)        1     1

Associated optimised criterion: 5.4114

Computation of the Individual Fisher information matrix

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 1.2325747  0.8128637 -0.2303858  0.00000  0.00000
[2,] 0.8128637 225.3364445  2.9541561  0.00000  0.00000
[3,] -0.2303858  2.9541561  0.1250120  0.00000  0.00000
[4,] 0.0000000  0.0000000  0.0000000 14.84573 20.15214
[5,] 0.0000000  0.0000000  0.0000000 20.15214 56.25539

```

```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta   StdError   RSE
ka  2.00  1.34079039  67.03952 %
k   0.25  0.09664122  38.65649 %
V   15.00  5.06159668  33.74398 %

----- Standard deviation of residual error -----

      Sigma  StdError   RSE
sig.interA  0.50  0.3621023  72.42046 %
sig.slopeA  0.15  0.1860159  124.01058 %

***** DETERMINANT *****

4640.201

***** CRITERION *****

5.411372

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

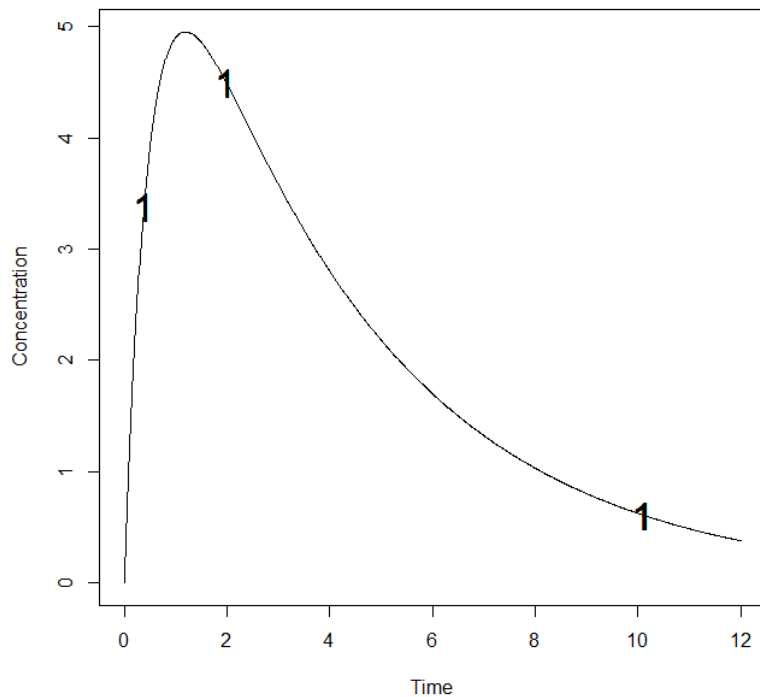
      FixedEffects  VarianceComponents
min          6.657679          0.03753734
max         225.378114          1.27838008
max/min      33.852356          34.05622730

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,]  1.0000000 -0.5581703  0.7400163  0.0000000  0.0000000
[2,] -0.5581703  1.0000000 -0.7236730  0.0000000  0.0000000
[3,]  0.7400163 -0.7236730  1.0000000  0.0000000  0.0000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000 -0.6973299
[5,]  0.0000000  0.0000000  0.0000000 -0.6973299  1.0000000

Time difference of 0.5850329 secs
sys.self
0

```



1.2.2. Fedorov-Wynn algorithm

```
#Name of the project
#-----
project<-"1.2.2"
#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
##### END OF OPTIMISATION ALGORITHM OPTION #####
```

OUTPUT FILE

```
PFIM 4.0

Option: 1

Project: 1.2.2

Date: Tue Jan 28 16:09:34 2014

***** INPUT SUMMARY *****
```

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

Sample times for response: A
Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 1 100

Total number of samples: 4

Associated criterion value: 5.0603

Identical sampling times for each response: FALSE

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

BEST ONE GROUP PROTOCOL:

Sample times for response: A
times freq Subjects doses
1 c(0.33, 1.5, 8, 12) 1 1 100

Associated criterion: 5.2451

Computation of the Individual Fisher information matrix

```

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 1.3081545  0.2980722 -0.2731617  0.000000  0.000000
[2,] 0.2980722 214.9216449  2.7951498  0.000000  0.000000
[3,] -0.2731617  2.7951498  0.1280792  0.000000  0.000000
[4,] 0.0000000  0.0000000  0.0000000 14.60286 20.34989
[5,] 0.0000000  0.0000000  0.0000000 20.34989 57.63562

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta StdError      RSE
ka  2.00 1.4570744 72.85372 %
k   0.25 0.1000394 40.01575 %
V   15.00 5.5016631 36.67775 %

----- Standard deviation of residual error -----

      Sigma StdError      RSE
sig.interA 0.50 0.367167 73.4334 %
sig.slopeA 0.15 0.184815 123.2100 %

***** DETERMINANT *****

3969.882

***** CRITERION *****

5.245121

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

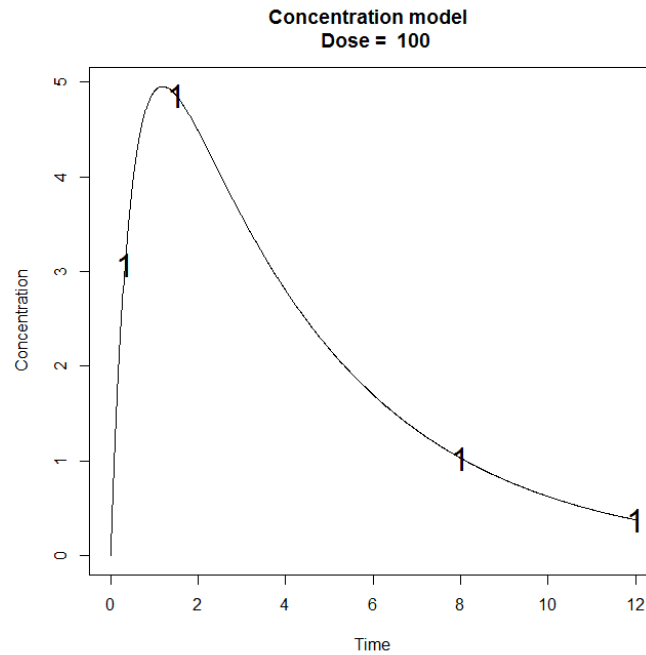
      FixedEffects VarianceComponents
min          6.503832          0.03157995
max          214.958418          6.50383230
max/min      33.051040          205.94814148

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]
[1,] 1.0000000 -0.5923083  0.7998900  0.0000000  0.0000000
[2,] -0.5923083  1.0000000 -0.7313914  0.0000000  0.0000000
[3,] 0.7998900 -0.7313914  1.0000000  0.0000000  0.0000000
[4,] 0.0000000  0.0000000  0.0000000  1.0000000 -0.7014513
[5,] 0.0000000  0.0000000  0.0000000 -0.7014513  1.0000000

Time difference of 0.6250348 secs
sys.self
0.09

```



1.2.3. Fixed Parameter and Fixed Sampling Times

Optimize I-FIM using the Fedorov-Wynn algorithm keeping the parameter k_a fixed (assuming no variability on k_a) and keeping fixed 2 sampling times (0.33, 1.5).

INPUT FILE

```
#Name of the project
#-----
project<-"1.2.3"
#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"I"
#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
#Diagonal Matrix of variance for inter-subject random effects:
#-----
#####ONLY FOR OPTIMISATION #####

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
#####
#FEDOROV-WYNN SPECIFICATION #
#####
```

```
#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c(0.33,1.5)
```

OUTPUT FILE

```
PFIM 4.0

Option: 1

Project: 1.2.3

Date: Tue Apr 22 15:20:04 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Initial design:

Sample times for response: A
      Protocol subjects doses
1 c=(0.33, 1.5, 5, 12)      1 100

Total number of samples: 4

Associated criterion value: 9.1489

Identical sampling times for each response: FALSE

Variance error model response A : ( 0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
      Nb of sampling points to be taken in this window, n[ 1 ]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4
```


BEST ONE GROUP PROTOCOL:

Sample times for response: A
 times freq Subjects doses
1 c(0.33, 1.5, 8, 12) 1 1 100

Associated criterion: 9.5815

Computation of the Individual Fisher information matrix

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]
[1,]	214.92164	2.7951498	0.00000	0.00000
[2,]	2.79515	0.1280792	0.00000	0.00000
[3,]	0.00000	0.0000000	14.60286	20.34989
[4,]	0.00000	0.0000000	20.34989	57.63562

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
k	0.25	0.08060287	32.24115 %
V	15.00	3.30180471	22.01203 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.367167	73.4334 %
sig.slopeA	0.15	0.184815	123.2100 %

***** DETERMINANT *****

8428.321

***** CRITERION *****

9.581539

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	65.734649	0.09171149
max	214.958013	65.73464882
max/min	3.270087	716.75477518

```
***** CORRELATION MATRIX *****
```

```

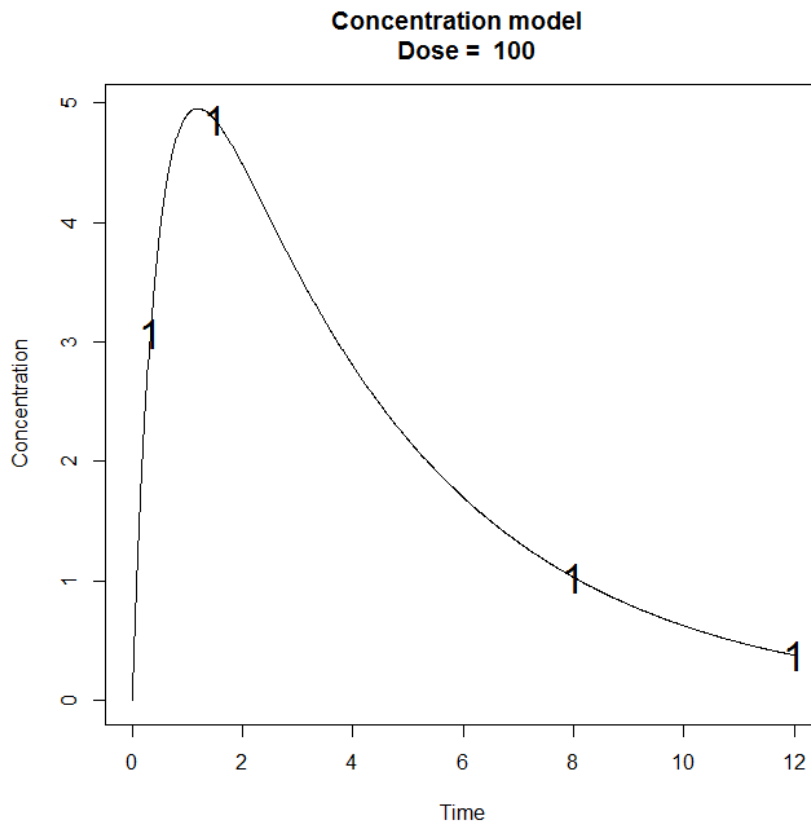
      [,1]      [,2]      [,3]      [,4]
[1,] 1.0000000 -0.5327529 0.0000000 0.0000000
[2,] -0.5327529 1.0000000 0.0000000 0.0000000
[3,] 0.0000000 0.0000000 1.0000000 -0.7014513
[4,] 0.0000000 0.0000000 -0.7014513 1.0000000

```

```

Time difference of 0.2652011 secs
sys.self
0.03

```



1.3. Bayesian Fisher Information Matrix (B-FIM)

Optimize the design in 1.1 this time for the Bayesian Fisher Information Matrix (B-FIM). Use same constraints in 1.1.1 and 1.1.2.

1.3.1. Simplex algorithm

INPUT FILE

```

#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"1.3.1"

```

```

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#      "FW" for the Fedorov-Wynn algorithm
#      "SIMP" for the Simplex algorithm
#-----

algo.option<-"SIMP"

```

OUTPUT FILE

```

PFIM 4.0

Option 1

Project: 1.3.1

Date: Thu Apr 17 15:06:38 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

Initial design:

Sample times for response: A
                times subjects.prop doses
1 c(0.33, 1.5, 5, 12)                1  100

Total number of samples (nr responses): 4

Associated criterion value: 3.5272

Window of the allowed optimised sampling times:

Upper and lower admissible samples times for the response A : [ 0 : 12 ]

```

Minimum delay between two sampling times: 0

Optimisation of the proportions of subjects: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

***** OPTIMISED DESIGN *****

Number of iterations: 70

Number of function evaluations: 122

Convergence Achieved

Design:

Sample times for response: A

	times	subjects.prop	subjects
1 c(0.332, 2.13, 6.537, 6.538)		1	1

Associated optimised criterion: 3.8917

Computation of the Bayesian Fisher information matrix

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]
[1,]	1.5579242	3.378667	-0.1817856
[2,]	3.3786674	402.310191	5.0219864
[3,]	-0.1817856	5.021986	0.1912682

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE	Shrinkage
ka	2.00	0.93681117	46.84056 %	21.94038 %
k	0.25	0.06704337	26.81735 %	28.76681 %
V	15.00	3.23108382	21.54056 %	46.39957 %

***** DETERMINANT *****

58.94244

***** CRITERION *****

3.89173

```
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****
```

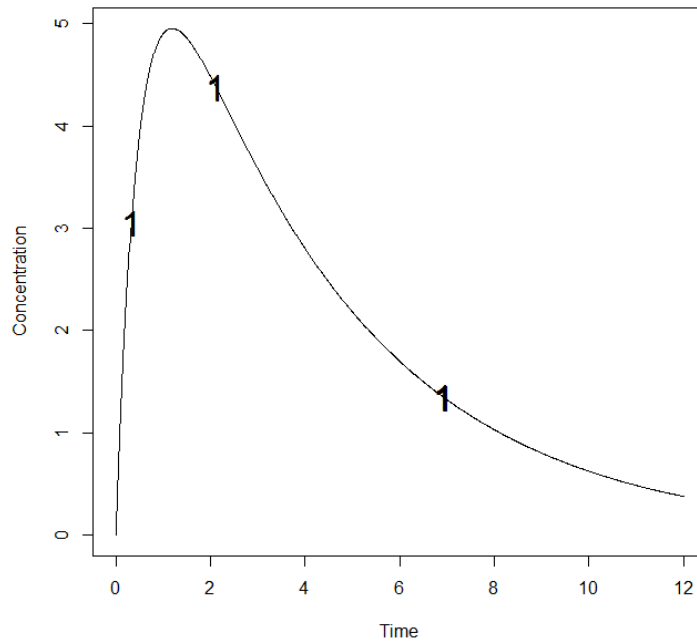
```
FixedEffects VarianceComponents
min      9.363022e-02      NA
max      4.024013e+02      NA
max/min  4.297772e+03      NA
```

```
***** CORRELATION MATRIX *****
```

```
      [,1]      [,2]      [,3]
[1,] 1.0000000 -0.4211662  0.5050159
[2,] -0.4211662  1.0000000 -0.6608620
[3,]  0.5050159 -0.6608620  1.0000000
```

```
Time difference of 0.6450372 secs
```

```
sys.self
0
```



1.3.2. Fedorov-Wynn algorithm

INPUT FILE

```
#Name of the project
#-----
project<-"1.3.2"
#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"
##### OPTIMISATION ALGORITHM OPTION #####
#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----
algo.option<-"FW"
```

OUTPUT FILE

PFIM 4.0

Option: 1

Project: 1.3.2

Date: Tue Jan 28 16:45:40 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

Sample times for response: A
 Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 1 100

Total number of samples: 4

Associated criterion value: 3.5272

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
 Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

BEST ONE GROUP PROTOCOL:

Sample times for response: A
 times freq Subjects doses

```
1 c(0.33, 1.5, 5, 8) 1 1 100
```

```
Associated criterion: 3.8066
```

```
Computation of the Bayesian Fisher information matrix
```

```
***** FISHER INFORMATION MATRIX *****
```

```
      [,1]      [,2]      [,3]
[1,] 1.590507 2.096455 -0.2426030
[2,] 2.096455 354.843266 4.4964361
[3,] -0.242603 4.496436 0.2013882
```

```
***** EXPECTED STANDARD ERRORS *****
```

```
----- Fixed Effects Parameters -----
```

	Beta	StdError	RSE	Shrinkage
ka	2.00	0.9638509	48.19255 %	23.22522 %
k	0.25	0.0688475	27.53900 %	30.33586 %
V	15.00	3.1862487	21.24166 %	45.12080 %

```
***** DETERMINANT *****
```

```
55.15913
```

```
***** CRITERION *****
```

```
3.806617
```

```
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****
```

	FixedEffects	VarianceComponents
min	9.552493e-02	NA
max	3.549127e+02	NA
max/min	3.715393e+03	NA

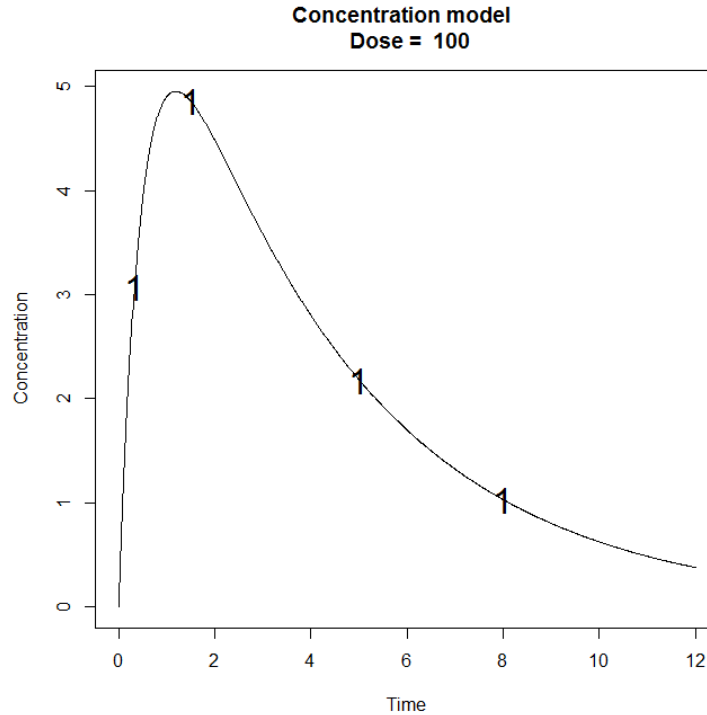
```
***** CORRELATION MATRIX *****
```

```
      [,1]      [,2]      [,3]
[1,] 1.0000000 -0.4133690 0.5638373
[2,] -0.4133690 1.0000000 -0.6330761
[3,] 0.5638373 -0.6330761 1.0000000
```

```
Time difference of 0.7720439 secs
```

```
sys.self
```

```
0.09
```



1.3.3. Fixed Parameter and Fixed Sampling Times

Optimize B-FIM using the Fedorov-Wynn algorithm keeping the parameter k_a fixed (assuming no variability on k_a) and keeping fixed 2 sampling times (0.33, 1.5).

INPUT FILE

```
#Name of the project
#-----
project<-"1.3.3"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"B"
#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(T,F,F)
#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0,0.25,0.1))

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#   "FW" for the Fedorov-Wynn algorithm
#   "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c(0.33,1.5)
```


OUTPUT FILE

PFIM 4.0

Option: 1

Project: 1.3.3

Date: Tue Apr 22 15:24:41 2014

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

Sample times for response: A
 Protocol subjects doses
1 c=(0.33, 1.5, 5, 12) 1 100

Total number of samples: 4

Associated criterion value: 6.3628

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
 Nb of sampling points to be taken in this window, n[1]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4

BEST ONE GROUP PROTOCOL:

Sample times for response: A

```
times freq Subjects doses
1 c(0.33, 1.5, 5, 8) 1 1 100
```

Associated criterion: 7.1584

Computation of the Bayesian Fisher information matrix

***** FISHER INFORMATION MATRIX *****

```
      [,1]      [,2]
[1,] 354.843266 4.4964361
[2,]  4.496436 0.2013882
```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE	Shrinkage
k	0.25	0.06269002	25.07601 %	25.15225 %
V	15.00	2.63147786	17.54319 %	30.77634 %

***** DETERMINANT *****

51.2433

***** CRITERION *****

7.158443

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

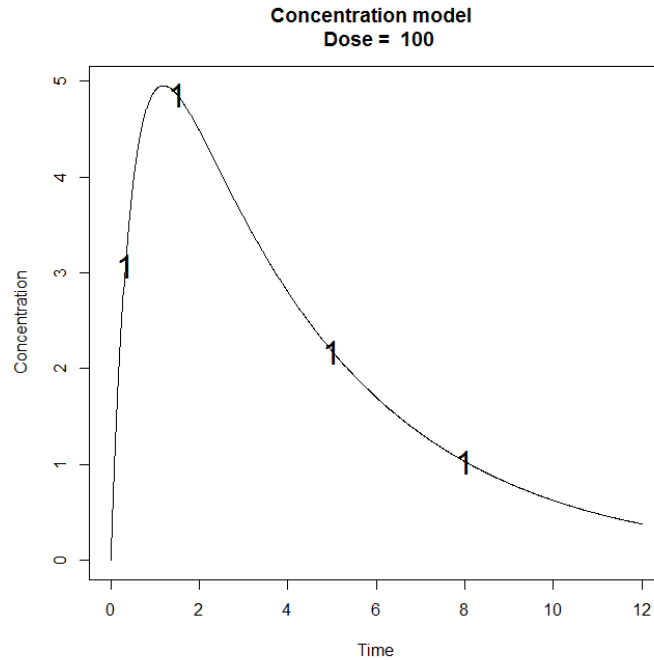
	FixedEffects	VarianceComponents
min	0.1443879	NA
max	354.9002664	NA
max/min	2457.9640320	NA

***** CORRELATION MATRIX *****

```
      [,1]      [,2]
[1,] 1.0000000 -0.5319039
[2,] -0.5319039 1.0000000
```

Time difference of 0.1715999 secs

```
sys.self
  0.03
```



2. Example 2: PK and immediate response PD model using the libraries of PK and PD Models (ODE)

- 100 subjects with a dose of 100
- Allowed sampling times for PK response: 0.5, 2, 30, 49, 180
- Allowed sampling times for PD response: 0.5, 2, 14, 110, 150
- Number of sampling times to be optimized: 3

2.1. Input File (P-FIM)

```
#####
##          INPUT FILE FOR PFIM 4.0          ##
#####

#Name of the project
#-----
project<-"EXAMPLE 2"
#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"
#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"OPT"
##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----

modelform<-"DE"
#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5, 2, 30))
protB<-list(c(14, 110,150))
```

```

#####ONLY FOR OPTIMISATION #####

#Identical sampling times for each response
# (only if you do not have sampling times=NULL)
#-----
identical.times<-F

##### OPTIMISATION ALGORITHM OPTION #####

#Character string for choice of the optimisation algorithm:
#      "FW" for the Fedorov-Wynn algorithm
#      "SIMP" for the Simplex algorithm
#-----

algo.option<-"FW"
#####
#FEDOROV-WYNN SPECIFICATION #
#####

#Number of sampling windows
#-----
nwindA<-1
nwindB<-1

#List of vector of the allowed sampling times for each sampling window
#-----

sampwinA<-list(c(0.5, 2, 30, 49, 180))
sampwinB<-list(c(0.5, 2, 14, 110, 150 ))

#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c()
fixed.timesB<-c()

#List of vector of allowed number of points to be taken from each sampling window
#-----

nsampA<-list(c(3))
nsampB<-list(c(3))

#Maximum total number of sampling times per subject
#-----

nmaxptsA<-3
nmaxptsB<-3

#Minimum total number of sampling times per subject
#-----

nminptsA<-3
nminptsB<-3
##### END OF OPTIMISATION ALGORITHM OPTION #####

```

2.2. Output File

```
PFIM 4.0

Option: 1

Project: EXAMPLE 2

Date: Sun Mar 23 19:53:50 2014

***** INPUT SUMMARY *****

Differential Equations form of the model:

function(t,y,p){
V<-p[1]
Vm<-p[2]
km<-p[3]
Alin<-p[4]
pk<-y[1:1]
pd<-y[2:2]
conc<-y[1]
if(t<=1){
dpk1<-(100/(1*V))+(-Vm)*pk[1]/(km+pk[1])}
else{
dpk1<-(-Vm)*pk[1]/(km+pk[1])}
dpd1<-0
pdIm<-Alin*conc
return(list(c(dpk1,dpd1),c(pk[1],pdIm)))
}

Initial Conditions at time 0 :

0 0

Error tolerance for solving differential equations system: RtolEQ = 1e-08 , AtolEQ
= 1e-08 , Hmax = Inf

Initial design:

Sample times for response: A
      Protocol subjects condinit
1 c=(0.5, 2, 30)      100 c(0, 0)

Sample times for response: B
      Protocol subjects condinit
1 c=(14, 110, 150)   100 c(0, 0)

Total number of samples: 600

Associated criterion value: 1098.507

Identical sampling times for each response: FALSE
```

```

Random effect model: Trand = 2

Variance error model response A : ( 0 + 0.2 *f)^2
Variance error model response B : ( 0.1 + 0 *f)^2

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.5 2 30 49 180
    Nb of sampling points to be taken in this window, n[ 1 ]= 3
Maximum total number of points in one elementary protocol : 3
Minimum total number of points in one elementary protocol : 3

Sampling windows for the response: B
Window 1 : t= 0.5 2 14 110 150
    Nb of sampling points to be taken in this window, n[ 1 ]= 3
Maximum total number of points in one elementary protocol : 3
Minimum total number of points in one elementary protocol : 3

Now evaluating the Fisher Information Matrix for the 100 protocols generated

BEST ONE GROUP PROTOCOL:

Sample times for response: A
    times freq Subjects condinit
1 c(0.5, 49, 180)    1    100 c(0, 0)
Sample times for response: B
    times freq Subjects condinit
1 c(2, 14, 110)    1    100 c(0, 0)

Associated criterion: 2549.118

***** OPTIMISED DESIGN *****

Optimised design:
Sample times for response: A
    times      freq Subjects condinit
1 c(0.5, 49, 180) 0.8449125 84.49125 c(0, 0)
2 c(0.5, 30, 180) 0.1550875 15.50875 c(0, 0)

Sample times for response: B
    times      freq Subjects condinit
1 c(2, 14, 110) 0.8449125 84.49125 c(0, 0)
2 c(2, 14, 110) 0.1550875 15.50875 c(0, 0)

```

Associated optimised criterion: 2549.662

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	2.6759559	10.42278	-56.61471	-0.7251327	0.000000e+00
[2,]	10.4227805	50031.09185	7276.78572	657.1611100	0.000000e+00
[3,]	-56.6147102	7276.78572	18134.47440	-6628.5770930	0.000000e+00
[4,]	-0.7251327	657.16111	-6628.57709	38614.6748550	0.000000e+00
[5,]	0.0000000	0.00000	0.00000	0.0000000	7.931719e+02
[6,]	0.0000000	0.00000	0.00000	0.0000000	5.505482e-01
[7,]	0.0000000	0.00000	0.00000	0.0000000	3.962926e-03
[8,]	0.0000000	0.00000	0.00000	0.0000000	2.159984e+00
[9,]	0.0000000	0.00000	0.00000	0.0000000	9.940642e+00

	[,6]	[,7]	[,8]	[,9]
[1,]	0.0000000	0.000000e+00	0.0000000	0.0000000
[2,]	0.0000000	0.000000e+00	0.0000000	0.0000000
[3,]	0.0000000	0.000000e+00	0.0000000	0.0000000
[4,]	0.0000000	0.000000e+00	0.0000000	0.0000000
[5,]	0.5505482	3.962926e-03	2.1599843	9.940642
[6,]	566.1136216	1.470446e-01	80.1122193	369.810105
[7,]	0.1470446	7.455466e+02	0.5814468	131.816702
[8,]	80.1122193	5.814468e-01	7762.5613244	1446.770561
[9,]	369.8101053	1.318167e+02	1446.7705613	23734.044514

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
V	12.200	0.637550069	5.225820 %
Vm	0.082	0.004646813	5.666846 %
km	0.370	0.008266279	2.234130 %
Alin	0.100	0.005290312	5.290312 %

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE
V	0.25	0.03550730	14.20292 %
Vm	0.25	0.04226093	16.90437 %
Alin	0.25	0.03664206	14.65682 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.slopeA	0.2	0.011419528	5.709764 %
sig.interB	0.1	0.006562738	6.562738 %

***** DETERMINANT *****

4.553482e+30

***** CRITERION *****

2549.662

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

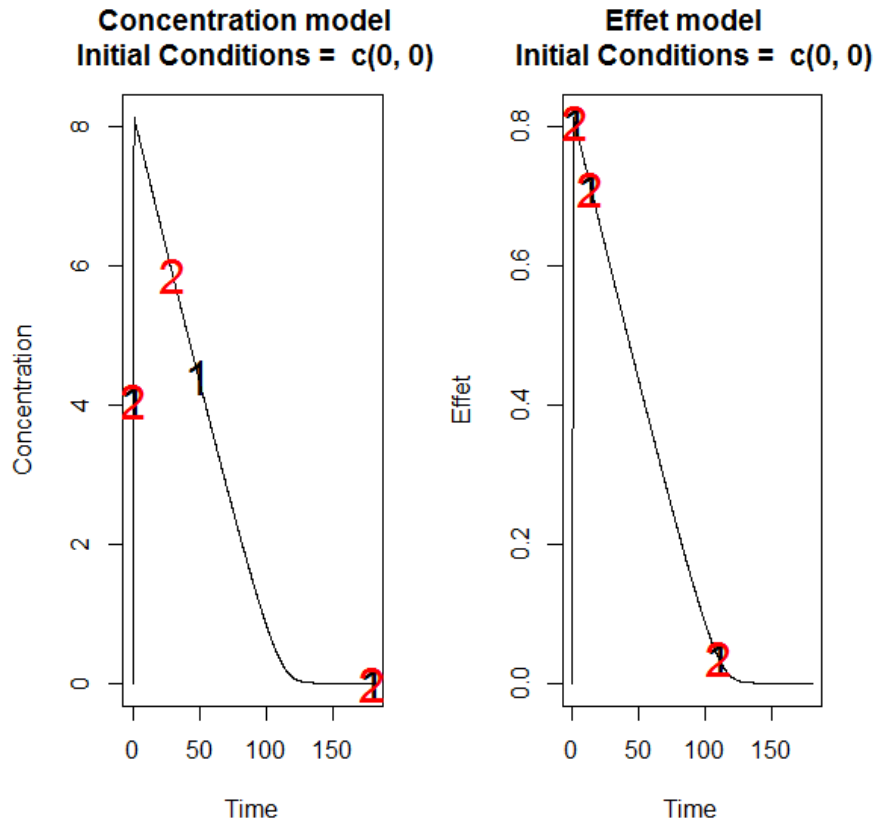
	FixedEffects	VarianceComponents
min	14719.494342	2.46017
max	51662.449933	14719.49434
max/min	3.509798	5983.12052

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	1.00000000	-0.10289582	0.2826168	0.07960564	0.000000e+00
[2,]	-0.10289582	1.00000000	-0.2708936	-0.08784124	0.000000e+00
[3,]	0.28261681	-0.27089364	1.0000000	0.27291243	0.000000e+00
[4,]	0.07960564	-0.08784124	0.2729124	1.0000000	0.000000e+00
[5,]	0.00000000	0.00000000	0.0000000	0.0000000	1.000000e+00
[6,]	0.00000000	0.00000000	0.0000000	0.0000000	-5.760357e-04
[7,]	0.00000000	0.00000000	0.0000000	0.0000000	6.305945e-05
[8,]	0.00000000	0.00000000	0.0000000	0.0000000	-6.134416e-04
[9,]	0.00000000	0.00000000	0.0000000	0.0000000	-2.145170e-03
	[,6]	[,7]	[,8]	[,9]	
[1,]	0.0000000000	0.000000e+00	0.0000000000	0.00000000	
[2,]	0.0000000000	0.000000e+00	0.0000000000	0.00000000	
[3,]	0.0000000000	0.000000e+00	0.0000000000	0.00000000	
[4,]	0.0000000000	0.000000e+00	0.0000000000	0.00000000	
[5,]	-0.0005760357	6.305945e-05	-0.0006134416	-0.00214517	
[6,]	1.0000000000	2.866158e-03	-0.0277520479	-0.09748089	
[7,]	0.0028661579	1.000000e+00	0.0030368822	-0.03161925	
[8,]	-0.0277520479	3.036882e-03	1.0000000000	-0.10337783	
[9,]	-0.0974808856	-3.161925e-02	-0.1033778325	1.00000000	

Time difference of 4.783273 secs

sys.self
0.2



3. Example 3: PK model with inter-occasion variability

- 40 subjects with a dose of 30
- Allowed sampling times: 0.5,1,1.5,2,4,6,8
- Number of sampling times to be optimized: 4

3.1. Input File (P-FIM)

```
#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"EXAMPLE 3"

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"OPT"

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----
```

```

modelform<-"AF"

##### ANALYTICAL MODEL OPTION #####
#####

#Identical dose in each elementary design (Yes=T, No=F)
#-----
dose.identical<-T

# If 'Yes', enter the value of the dose,
# else, enter the vector of the dose values for each elementary design
#-----
dose<-c(30)

#Vector of the times intervals of each expression
#-----
boundA<-list(c(0,Inf))

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-F

##### END ANALYTICAL MODEL OPTION #####

#Name of the fixed effects parameters
#-----
parameters<-c("ka","V","Cl")

#Fixed effects parameters values
#-----
beta<-c(1,3.5,2)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(F,F,F)

#Number of occasions
#-----
n_occ<-2

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0.09,0.09,0.09))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0.0225,0.0225,0.0225))

#Standard deviation of residual error (sig.inter+sig.slope*f)^2:
#-----
sig.interA<-0.1
sig.slopeA<-0

```

```

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5,2,4,8))

#Vector of initial proportions or numbers of subjects for each elementary design
#-----
subjects<-c(40)

#Subjects input: (1) for number of subjects (2) for proportions of subjects
#-----
subjects.input<-1

#If 'proportions of subjects' give the total number of samples
#-----
#Ntot<-40

```

3.2. Output File

```

PFIM 4.0

Option: 1

Project: EXAMPLE 3

Date: Wed Jan 29 10:30:16 2014

***** INPUT SUMMARY *****

Analytical function model:

dose/V * ka/(ka - (Cl/V)) * (exp(-(Cl/V) * t) - exp(-ka * t))

Initial design:

Sample times for response: A
      Protocol subjects doses
1 c=(0.5, 2, 4, 8)      40    30

Total number of samples: 160

Associated criterion value: 1826.068

Identical sampling times for each response: FALSE

Number of occasions: 2

Random effect model: Trand = 2

Variance error model response A : ( 0.1 + 0 *f)^2

Optimization step:

```

Sampling windows for the response: A

Window 1 : t= 0.5 1 1.5 2 4 6 8

Nb of sampling points to be taken in this window, n[1]= 4

Maximum total number of points in one elementary protocol : 4

Minimum total number of points in one elementary protocol : 4

Now evaluating the Fisher Information Matrix for the 35 protocols generated

BEST ONE GROUP PROTOCOL:

Sample times for response: A

	times	freq	Subjects	doses
1	c(0.5, 2, 6, 8)	1	40	30

Associated criterion: 1913.953

***** OPTIMISED DESIGN *****

Optimised design:

Sample times for response: A

	times	freq	Subjects	doses
1	c(0.5, 2, 6, 8)	1	40	30

Associated optimised criterion: 1913.953

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]
[1,]	342.070959	-11.909950	2.294299	0.0000000	0.0000000	0.0000000
[2,]	-11.909950	29.371134	0.624963	0.0000000	0.0000000	0.0000000
[3,]	2.294299	0.624963	98.030214	0.0000000	0.0000000	0.0000000
[4,]	0.0000000	0.0000000	0.0000000	1462.6567613	21.7203074	0.2631903
[5,]	0.0000000	0.0000000	0.0000000	21.7203074	1618.1679873	0.2392295
[6,]	0.0000000	0.0000000	0.0000000	0.2631903	0.2392295	1921.9845543
[7,]	0.0000000	0.0000000	0.0000000	731.3283806	10.8601537	0.1315952
[8,]	0.0000000	0.0000000	0.0000000	10.8601537	809.0839937	0.1196147
[9,]	0.0000000	0.0000000	0.0000000	0.1315952	0.1196147	960.9922772
[10,]	0.0000000	0.0000000	0.0000000	414.3130643	276.7056180	28.1724200
	[,7]	[,8]	[,9]	[,10]		
[1,]	0.0000000e+00	0.0000000e+00	0.0000000e+00	0.000000		
[2,]	0.0000000e+00	0.0000000e+00	0.0000000e+00	0.000000		
[3,]	0.0000000e+00	0.0000000e+00	0.0000000e+00	0.000000		
[4,]	7.313284e+02	1.086015e+01	1.315952e-01	414.31306		
[5,]	1.086015e+01	8.090840e+02	1.196147e-01	276.70562		
[6,]	1.315952e-01	1.196147e-01	9.609923e+02	28.17242		
[7,]	1.252260e+04	4.388584e+03	3.991645e+01	2608.00290		

```
[8,] 4.388584e+03 1.961824e+04 6.185025e+01 1889.93103
[9,] 3.991645e+01 6.185025e+01 3.560094e+04 926.11512
[10,] 2.608003e+03 1.889931e+03 9.261151e+02 20551.58326
```

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE	
ka	1.0	0.05445931	5.445931	%
V	3.5	0.18585115	5.310033	%
Cl	2.0	0.10101646	5.050823	%

----- Variance of Inter-Subject Random Effects -----

	omega ²	StdError	RSE	
ka	0.09	0.02660961	29.56624	%
V	0.09	0.02516549	27.96165	%
Cl	0.09	0.02296550	25.51722	%

----- Variance of Inter-Occasion Random Effects -----

	gamma ²	StdError	RSE	
ka	0.0225	0.009552479	42.45546	%
V	0.0225	0.007539081	33.50703	%
Cl	0.0225	0.005339183	23.72970	%

----- Standard deviation of residual error -----

	Sigma	StdError	RSE	
sig.interA	0.1	0.007098313	7.098313	%

***** DETERMINANT *****

6.596486e+32

***** CRITERION *****

1913.953

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	18402.235009	28.91084
max	35690.090280	18402.23501
max/min	1.939443	636.51680

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	1.00000000	0.11898386	-0.01401299	0.0000000000	0.0000000000

```

[2,] 0.11898386 1.00000000 -0.01323044 0.000000000 0.000000000
[3,] -0.01401299 -0.01323044 1.00000000 0.000000000 0.000000000
[4,] 0.00000000 0.00000000 0.00000000 1.000000000 -0.0192963816
[5,] 0.00000000 0.00000000 0.00000000 -0.0192963816 1.000000000
[6,] 0.00000000 0.00000000 0.00000000 -0.0001505132 -0.0001422569
[7,] 0.00000000 0.00000000 0.00000000 -0.1695130986 0.0478699472
[8,] 0.00000000 0.00000000 0.00000000 0.0533692914 -0.1475617586
[9,] 0.00000000 0.00000000 0.00000000 0.0019716819 0.0016536078
[10,] 0.00000000 0.00000000 0.00000000 -0.0509821501 -0.0400931222
      [,6]      [,7]      [,8]      [,9]      [,10]
[1,] 0.000000000 0.000000000 0.000000000 0.000000000 0.000000000
[2,] 0.000000000 0.000000000 0.000000000 0.000000000 0.000000000
[3,] 0.000000000 0.000000000 0.000000000 0.000000000 0.000000000
[4,] -0.0001505132 -0.1695130986 0.0533692914 0.001971682 -0.0509821501
[5,] -0.0001422569 0.0478699472 -0.1475617586 0.001653608 -0.0400931222
[6,] 1.000000000 0.0002223017 0.0002468429 -0.116089527 -0.0005440808
[7,] 0.0002223017 1.000000000 -0.2776209058 0.003259840 -0.1332452332
[8,] 0.0002468429 -0.2776209058 1.000000000 -0.000289734 -0.0472408862
[9,] -0.1160895270 0.0032598400 -0.0002897340 1.000000000 -0.0341367622
[10,] -0.0005440808 -0.1332452332 -0.0472408862 -0.034136762 1.000000000

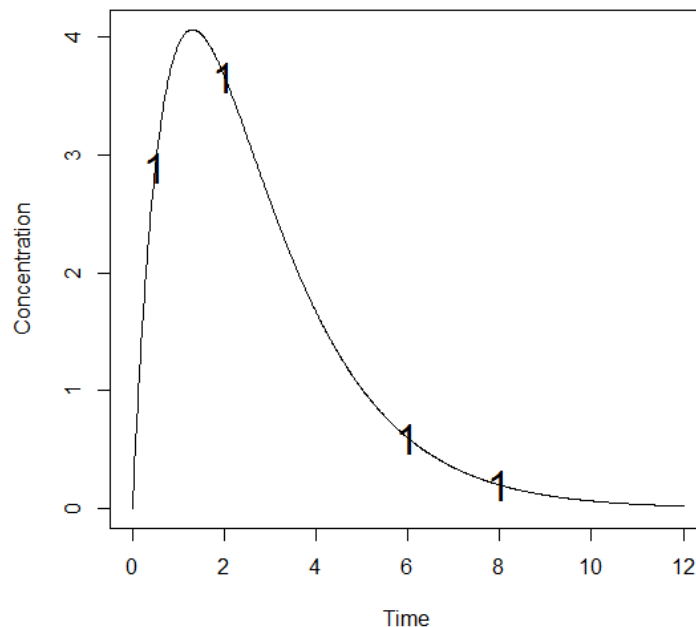
```

```

Time difference of 1.110063 secs
sys.self
0.05

```

Concentration model
Dose = 30



4. Example 4: PK model with inter-occasion variability and covariate effects (Equivalence test)

- 40 subjects with a dose of 30
- Allowed sampling times: 0.5, 2, 4, 6, 8

- Number of sampling times to be optimized: 3

4.1. Input File (P-FIM)

```
#####
##                               INPUT FILE FOR PFIM 4.0                               ##
#####

#Name of the project
#-----
project<-"EXAMPLE 4"

#Name of the file containing the PK or PD model
#-----
file.model<-"model.r"

#Name of the output file for the results and for the Fisher information matrix
#-----
output<-"Stdout.r";
outputFIM<-"";

#FIM: Population (P) or Individual (I) or Bayesian (B) Fisher information matrix
#-----
FIM<-"P"

#RUN: Evaluation (EVAL) or Optimisation (OPT)
#-----
run<-"OPT"

#To display only graphs of models and/or sensitivity functions before evaluating
the Fisher Information matrix
graph.only<-F

#Block diagonal Fisher information matrix (option<-1) or complete Fisher
information matrix (option<-2)
#-----
option<-1

#Number of responses
#-----
nr<-1

##### MODEL OPTION #####

#Model form: Differential equations (DE) or analytical form (AF)
#-----
modelform<-"AF"

##### ANALYTICAL MODEL OPTION #####
#####

#Identical dose in each elementary design (Yes=T, No=F)
#-----
dose.identical<-T

# If 'Yes', enter the value of the dose,
# else, enter the vector of the dose values for each elementary design
#-----
```

```

dose<-c(30)

#Vector of the times intervals of each expression
#-----
boundA<-list(c(0,Inf))

#Numerical derivatives (Yes=T, No=F)
#If 'Yes', specify the model function "form" in the model file
#If 'No', specify the object "form" which is a vector of expressions in the model
file
#-----
NUM<-T

##### END ANALYTICAL MODEL OPTION #####

#Name of the fixed effects parameters
#-----
parameters<-c("ka","V","Cl")

#Fixed effects parameters values
#-----
beta<-c(1,3.5,2)

#Some parameters may not be estimated (not estimated = T, estimated = F)
#-----
beta.fixed<-c(F,F,F)

#Number of occasions
#-----
n_occ<-2

#Random effect model (1) = additive (2) = exponential
#-----
Trand<-2;

#Diagonal Matrix of variance for inter-subject random effects:
#-----
omega<-diag(c(0.09,0.09,0.09))

#Diagonal Matrix of variance for inter-occasion random effects:
#-----
gamma<-diag(c(0.0225,0.0225,0.0225))

#List of the vectors of sampling times for each elementary design
#You can specify that a group has no sampling time by writing NULL
#(ONLY if you have several response)
#-----
protA<-list(c(0.5,2,4))
#####
#FEDOROV-WYNN SPECIFICATION #
#####

#Number of sampling windows
#-----
nwindA<-1
nwindB<-1

```



```

#List of vector of the allowed sampling times for each sampling window
#-----

sampwinA<-list(c(0.5,2,4,6,8))
#sampwinB<-list(c(0, 0.5, 1, 2, 6, 9, 12, 24, 36, 48, 72, 96, 120))

#Fixed times (times which will be in all evaluated protocols, corresponding to
fixed constraints)
#-----
fixed.timesA<-c()
#fixed.timesB<-c()

#List of vector of allowed number of points to be taken from each sampling window
#-----

nsampA<-list(c(3))
#nsampB<-list(c(3))

#Maximum total number of sampling times per subject
#-----

nmaxptsA<-3
#nmaxptsB<-3

#Minimum total number of sampling times per subject
#-----

nminptsA<-3
#nminptsB<-3
##### END OF OPTIMISATION ALGORITHM OPTION #####

```

4.2. Output File

```

PFIM 4.0

Option: 1

Project: EXAMPLE 4

Date: Wed Jan 29 10:52:55 2014

***** INPUT SUMMARY *****

Analytical function model:

function(t,p,X){
ka<-p[1]
V<-p[2]
Cl<-p[3]
y<-X/V * ka/(ka - (Cl/V)) * (exp(-(Cl/V) * t) - exp(-ka * t))
return(y)
}

Initial design:

```

```

Sample times for response: A
  Protocol subjects doses
1 c=(0.5, 2, 4)      40   30

Total number of samples: 120

Associated criterion value: 1115.614

Identical sampling times for each response: FALSE

Number of occasions: 2

Random effect model: Trand = 2

Variance error model response A : ( 0.1 + 0 *f)^2

Covariate model :

  NB: Covariates are additive on log parameters

  Covariates not changing with occasion

  Covariate 1 : Sex ( V )
  Categories References Proportions
(1)           M           *         0.5
(2)           F           *         0.5

  Covariates changing with occasion

  Covariate 1 : Treat ( C1 )
  Categories References
(1)           A           *
(2)           B

  Sequences Proportions
(1)           A B         0.5
(2)           B A         0.5

Optimization step:

Sampling windows for the response: A
Window 1 : t= 0.5 2 4 6 8
  Nb of sampling points to be taken in this window, n[ 1 ]= 3
Maximum total number of points in one elementary protocol : 3
Minimum total number of points in one elementary protocol : 3

Now evaluating the Fisher Information Matrix for the 10 protocols generated

```

BEST ONE GROUP PROTOCOL:

Sample times for response: A
times freq Subjects doses
1 c(0.5, 2, 6) 1 40 30

Associated criterion: 1189.867

***** OPTIMISED DESIGN *****

Optimised design:
Sample times for response: A
times freq Subjects doses
1 c(0.5, 2, 6) 1 40 30

Associated optimised criterion: 1189.867

Computation of the Population Fisher information matrix: option = 1

***** FISHER INFORMATION MATRIX *****

		s2					
		329.525019	-14.8691158	3.6912288	-22.229595	3.834115	0.0000000
		-14.869116	28.6258030	0.9910258	51.227048	1.140631	0.0000000
		3.691229	0.9910258	97.7403300	1.572171	97.536153	0.0000000
s2		-22.229595	51.2270485	1.5721714	179.294670	1.950838	0.0000000
		3.834115	1.1406306	97.5361532	1.950838	921.108493	0.0000000
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	1357.8361093
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	34.5732189
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	0.6847072
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	693.0899074
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	34.3142049
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	24.8299228
		0.0000000	0.0000000	0.0000000	0.0000000	0.0000000	471.2181021
		0.0000000	0.0000000	0.000000e+00	0.000000e+00	0.000000	0.000000
		0.0000000	0.0000000	0.000000e+00	0.000000e+00	0.000000	0.000000
		0.0000000	0.0000000	0.000000e+00	0.000000e+00	0.000000	0.000000
s2		0.0000000	0.0000000	0.000000e+00	0.000000e+00	0.000000	0.000000
		0.0000000	0.0000000	0.000000e+00	0.000000e+00	0.000000	0.000000
		34.5732189	0.6847072	6.930899e+02	3.431420e+01	24.82992	471.21810
		1537.8685241	0.6068128	2.694200e+01	7.729341e+02	26.16533	318.44875
		0.6068128	1910.6360130	3.082288e-01	9.221043e-01	954.18480	37.92596
		26.9419991	0.3082288	1.123026e+04	5.252065e+03	167.39074	2376.07691
		772.9340538	0.9221043	5.252065e+03	1.996723e+04	197.52750	2192.63445
		26.1653310	954.1848020	1.673907e+02	1.975275e+02	35807.24708	1526.11630
		318.4487533	37.9259602	2.376077e+03	2.192634e+03	1526.11630	5510.31204

***** EXPECTED STANDARD ERRORS *****

```

----- Fixed Effects Parameters -----
          Beta      StdError      RSE
ka          1.00000000  0.05577975  5.577975 %
V           3.50000000  0.26955025  7.701436 %
Cl          2.00000000  0.10700134  5.350067 %
beta_V_Sex_2  0.18232156  0.10686697  58.614553 %
beta_Cl_Treat_B 0.09531018  0.03484141  36.555810 %

----- Variance of Inter-Subject Random Effects -----
          omega^2      StdError      RSE
ka      0.09  0.02788743  30.98604 %
V       0.09  0.02590788  28.78653 %
Cl      0.09  0.02303141  25.59046 %

----- Variance of Inter-Occasion Random Effects -----
          gamma^2      StdError      RSE
ka  0.0225  0.010554907  46.91070 %
V   0.0225  0.007702135  34.23171 %
Cl  0.0225  0.005353992  23.79552 %

----- Standard deviation of residual error -----
          Sigma      StdError      RSE
sig.interA  0.1  0.01451322  14.51322 %

***** DETERMINANT *****
8.053415e+36

***** CRITERION *****
1189.867

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

          FixedEffects VarianceComponents
min          1883.79090          12.66685
max          35923.72080          1883.79090
max/min          19.06991          148.71823

***** CORRELATION MATRIX *****

                                     s2
1.0000000000  0.1263435890 -0.022352927 -0.026138657 -0.0004354741
0.1263435890  1.0000000000 -0.016440418 -0.712334240 -0.0004483848
-0.0223529273 -0.0164404185  1.000000000  0.002774252 -0.3249099148
s2 -0.0261386565 -0.7123342396  0.002774252  1.000000000 -0.0003998150
-0.0004354741 -0.0004483848 -0.324909915 -0.000399815  1.0000000000
0.0000000000  0.0000000000  0.000000000  0.000000000  0.0000000000
0.0000000000  0.0000000000  0.000000000  0.000000000  0.0000000000
0.0000000000  0.0000000000  0.000000000  0.000000000  0.0000000000

```

```

0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000

0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
s2 0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
0.0000000000 0.0000000000 0.0000000000 0.0000000000 0.0000000000
1.0000000000 -0.0194471821 -0.0002298133 -0.1511036995 0.0781512752
-0.0194471821 1.0000000000 -0.0000652991 0.0711693764 -0.1362821286
-0.0002298133 -0.0000652991 1.0000000000 0.0005569804 0.0003365315
-0.1511036995 0.0711693764 0.0005569804 1.0000000000 -0.3212630803
0.0781512752 -0.1362821286 0.0003365315 -0.3212630803 1.0000000000
0.0125884017 0.0078633772 -0.1147072179 0.0201752344 0.0078965258
-0.1327148141 -0.0943115113 0.0005960777 -0.2303374371 -0.1100174414

0.0000000000 0.0000000000
0.0000000000 0.0000000000
0.0000000000 0.0000000000
s2 0.0000000000 0.0000000000
0.0000000000 0.0000000000
0.012588402 -0.1327148141
0.007863377 -0.0943115113
-0.114707218 0.0005960777
0.020175234 -0.2303374371
0.007896526 -0.1100174414
1.000000000 -0.1117917937
-0.111791794 1.0000000000

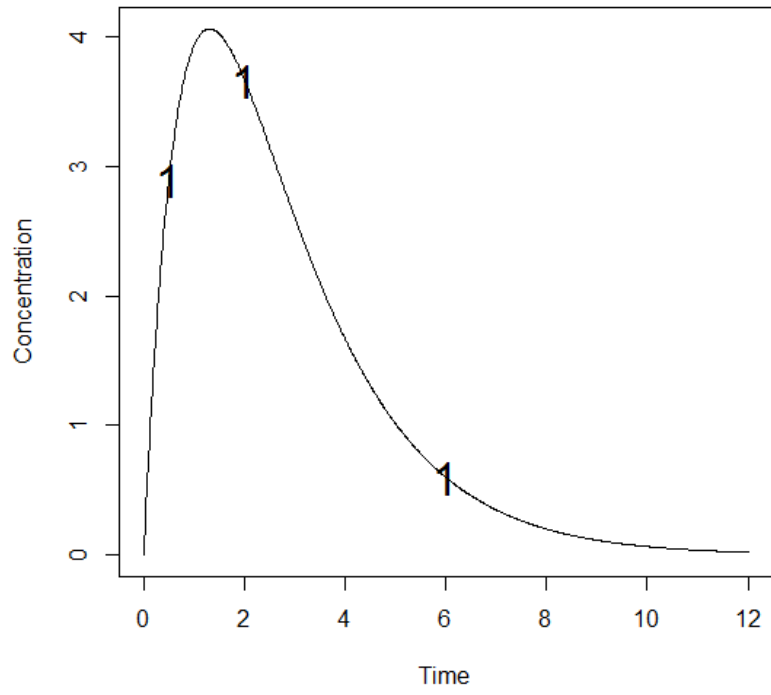
```

```

Time difference of 0.5230298 secs
sys.self      0.06

```

Concentration model
Dose = 30



1.2 PFIM Interface 4.0. User guide documentation



PFIM Interface 4.0

PFIM Group

IAME UMR1137, INSERM and Université Paris Diderot, Paris, France

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www.pfim.biostat.fr

User guide

Written by Giulia Lestini, Cyrielle Dumont and Thu Thuy Nguyen

PFIM Interface 4.0 is free library of functions.
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1. Introduction

Model based optimal design approaches are increasingly performed in population pharmacokinetic/pharmacodynamics (PKPD) [1], which consist in determining a balance between the number of subjects and the number of samples per subject, as well as the allocation of times and doses, according to experimental conditions. A good choice of design is crucial for an efficient estimation of model parameters, especially when the studies are conducted in patients where only a few samples can be taken per subject. These approaches rely on the Fisher information matrix (FIM) for nonlinear mixed effect models (NLMEM), available in several software tools [2] and are a good alternative to clinical trial simulation. They require a *priori* knowledge of the model and its parameters, which can usually be obtained from previous experiments.

PFIM (www.pfim.biostat.fr), developed in our group since 2001 [3,4], is the first tool for design evaluation and optimisation that has been developed in R. Two versions are available: a R script version and a graphical user interface version. PFIM Interface 4.0 is an extension of the graphical user version PFIM Interface 3.1 and includes several new features based on the R script program of PFIM 4.0 [5].

In this new version, for population designs, optimisation can be performed with fixed parameters or fixed sampling times. The Fisher information matrix obtained after evaluation or optimisation can be saved in a file. Additional features for Bayesian designs are now available. The Bayesian Fisher information matrix has been implemented. Design for maximum a *posteriori* estimation of individual parameters can be evaluated or optimised and the predicted shrinkage is also reported [6]. A new way has been added to specify user-defined models through an R function. It is now possible to visualise the graphs of the model and the sensitivity functions without performing evaluation or optimisation.

This documentation describes the methodology implemented in PFIM Interface 4.0 in Section 2. Section 3 describes how to specify models, either by using the PKPD library or the user-defined model option. Section 4 explains how to install and use PFIM Interface 4.0. Lastly, Section 5 presents in detail an evaluation and an optimisation output of PFIM Interface 4.0.

2. Methodology

2.1 Design

The elementary design ξ_i of individual i ($i = 1, \dots, N$) is defined by the number n_i of samples and their allocation in time (t_{i1}, \dots, t_{ini}) .

For N individuals, the population design is composed of the N elementary designs such as $\mathcal{E} = \{\xi_1, \dots, \xi_N\}$. Usually, population designs are composed of a limited number Q of groups of individuals with identical design ξ_q within each group, performed in a number N_q of individuals. The population design can thus be written as $\mathcal{E} = \{[\xi_1, N_1]; \dots; [\xi_Q, N_Q]\}$.

Individual and Bayesian designs include only one elementary design.

2.2 Nonlinear mixed effects models

A nonlinear mixed effects model, or a population model, is defined as follows. The vector of observations Y_i for the individual i ($i = 1, \dots, N$) is defined as

$$Y_i = f(\theta_i, \xi_i) + \varepsilon_i,$$

where the function f defines the nonlinear structural model, θ_i is the vector of the p -individual parameters for individual i , ξ_i is the elementary design of individual i and ε_i is the vector of residual error.

The vector of individual parameters θ_i depends on μ , the p -vector of the fixed effects parameters and on b_i , the p -vector of the random effects for individual i . The relation between θ_i and (μ, b_i) can be additive for a normal distribution of parameters, that is

$$\theta_i = \mu + b_i,$$

or exponential for a lognormal distribution of parameters so that

$$\theta_i = \mu \times \exp(b_i).$$

It is assumed that $b_i \sim N(0, \Omega)$ with Ω defined as a $p \times p$ diagonal variance-covariance matrix, for which, each diagonal element ω_j , $j = 1, \dots, p$, represents the inter-individual variability of the j^{th} component of the vector b_i .

It is also supposed that $\varepsilon_i \sim N(0, \Sigma_i)$, where Σ_i is a $n_i \times n_i$ -diagonal matrix such that

$$\Sigma_i(\mu, b_i, \sigma_{inter}, \sigma_{slope}, \xi_i) = \text{diag}(\sigma_{inter} + \sigma_{slope} \times f(\theta_i, \xi_i))^2.$$

The terms σ_{inter} and σ_{slope} are the additive and proportional parts of the error model, respectively. Conditionnally on the value of b_i , it is assumed that the ε_i errors are independently distributed.

In the case of K multiple responses, the vector of observations Y_i can then be composed of K vectors for the different responses:

$$Y_i = [y_{i1}^T, y_{i2}^T, \dots, y_{iK}^T]^T,$$

where y_{ik} , $k=1, \dots, K$, is the vector of n_{ik} observations for the k^{th} response. Each of these responses is associated with a known function f_k , which can be grouped in a vector of multiple response model F , such as

$$F(\theta_i, \xi_i) = [f_1(\theta_i, \xi_{i1})^T, f_2(\theta_i, \xi_{i2})^T, \dots, f_K(\theta_i, \xi_{iK})^T]^T,$$

where ξ_i is composed of K sub-designs such that $\xi_i = (\xi_{i1}, \xi_{i2}, \dots, \xi_{iK})$. The sub-design ξ_{ik} is then defined by $(t_{ik1}, t_{ik2}, \dots, t_{ikn_{ik}})$, with n_{ik} sampling times for the observations of the k^{th} response, so that $n_i = \sum_{k=1}^K n_{ik}$. Each response can have its error model and ε_i is then the vector composed of the K vectors of residual errors ε_{ik} , $k=1, \dots, K$, associated with the K responses.

2.3 Fisher information matrix

2.3.1 Population Fisher information matrix

The population Fisher information matrix $M_F(\Psi, \xi)$ for multiple response models, for an individual with an elementary design ξ , with the vector of population parameters Ψ , is given as:

$$M_F(\Psi, \xi) \equiv \frac{1}{2} \begin{pmatrix} A(E, V) & C(E, V) \\ C^T(E, V) & B(E, V) \end{pmatrix}$$

with E and V the approximated marginal expectation and the variance of the observations of the individual. The vector of population parameter Ψ is defined by $\Psi^T = (\mu^T, \lambda^T)$ with μ the p -vector of the fixed effects and λ the vector of the variance terms. M_F is given as a block matrix (more details are given in [7-9]) with:

$$(A(E, V))_{ml} = 2 \frac{\partial E^T}{\partial \mu_m} V^{-1} \frac{\partial E}{\partial \mu_l} + \text{tr} \left(\frac{\partial V}{\partial \mu_l} V^{-1} \frac{\partial V}{\partial \mu_m} V^{-1} \right) \text{ with } m \text{ and } l = 1, \dots, p$$

$$(B(E, V))_{ml} = \text{tr} \left(\frac{\partial V}{\partial \lambda_m} V^{-1} \frac{\partial V}{\partial \lambda_l} V^{-1} \right) \text{ with } m \text{ and } l = 1, \dots, \dim(\lambda)$$

$$(C(E, V))_{ml} = \text{tr} \left(\frac{\partial V}{\partial \lambda_l} V^{-1} \frac{\partial V}{\partial \mu_m} V^{-1} \right) \text{ with } l = 1, \dots, \dim(\lambda) \text{ and } m = 1, \dots, p$$

If the dependence of V in μ is neglected so that $\frac{\partial V}{\partial \mu} = 0$, the population

Fisher information matrix is a block diagonal matrix that is to say the block C of the matrix is supposed to be 0. Also, the block A is simplified and expressed as:

$$(A(E, V))_{ml} = 2 \frac{\partial E^T}{\partial \mu_m} V^{-1} \frac{\partial E}{\partial \mu_l} \text{ with } m \text{ and } l = 1, \dots, p$$

Since PFIM Interface 3.1, the user can choose to compute either a full or a block diagonal matrix for population designs. However, based on publications showing the better performance of the block diagonal expression compared to the full one with linearisation [2], the default option in PFIM is the block diagonal information matrix.

Prediction of standard errors

According to the inequality of Cramer-Rao, the inverse of M_F is the lower bound of the variance-covariance matrix of any unbiased estimate of the parameters. From the square roots of the diagonal elements of the inverse of M_F , the predicted standard errors (SE) for estimated parameters can be calculated.

2.3.2 Bayesian Fisher information matrix

New feature: The new version 4.0 of PFIM Interface enables design evaluation and optimisation for maximum a posteriori estimation of individual parameters based on the Bayesian Fisher information matrix [6].

We are interested in the precision estimation of individual parameters for a subject i , associated to the vector of observation y (index i being omitted). These individual parameters can be estimated by maximum a posteriori (MAP). As μ is known, estimating θ is similar to estimating η . More precisely, the MAP estimate of η is given by

$$\hat{\eta} = \operatorname{argmax} (p(\eta | y)) = \operatorname{argmax} \left(\frac{p(y | \eta) p(\eta)}{p(y)} \right) = \operatorname{argmax} (\log (p(y | \eta)) + \log (p(\eta)))$$

where p is the probability density. The Bayesian Fisher information matrix, taking into account the a priori distribution of the random effects, is expressed as

$$\begin{aligned} M_{BF}(\xi) &= -E_{\eta} \left(\frac{\partial^2 \log(p(\eta | y))}{\partial \eta \partial \eta^T} \right) = -E_{\eta} \left(E_{y | \eta} \left(\frac{\partial^2 \log(p(y | \eta))}{\partial \eta \partial \eta^T} \right) \right) - E_{\eta} \left(\frac{\partial^2 \log(p(\eta))}{\partial \eta \partial \eta^T} \right) \\ &= E_{\eta} (M_{IF}(\mathcal{G}(\mu, \eta), \xi)) - \Omega^{-1} \end{aligned}$$

where $M_{IF}(\theta, \xi) = -E_y \left(\frac{\partial^2 \log(p(y | \theta))}{\partial \theta \partial \theta^T} \right)$, expression of the individual Fisher

information matrix in classical nonlinear regression models. The expectation $E_{\eta} (M_{IF}(\mathcal{G}(\mu, \eta), \xi))$ can be obtained by first order approximation of the model around the expectation of random effects (i.e., 0).

The shrinkage (Sh) is quantified from the ratio of the estimation variance predicted by M_{BF}^{-1} and the a priori variance, and can be calculated as the diagonal elements of the matrix $I - W(\xi) = M_{BF}(\xi)^{-1} \Omega^{-1}$ (see [6] for more details).



When a parameter has an a priori variance equal to 0, it will be considered as fixed to the mean value and no predicted shrinkage will be computed.

2.4 Design evaluation

Population, individual and Bayesian design evaluation is based on the computation of the population, individual and Bayesian Fisher information matrix, respectively. During this process, the expected standard errors on the population or individual parameters with the design are evaluated. The user can choose to fix one or several parameters in the model that will not be computed in the Fisher information matrix.

Eigenvalues and conditional number are given by default. When considering design for Bayesian estimation of individual parameters, the shrinkages are also reported.

The computed Fisher information matrix can be saved in a file if requested.

2.5 Design optimisation

PFIM Interface 4.0 allows to optimise exact or a statistical designs. In the case of an exact optimisation, the group structure of the design is fixed: the number of elementary designs, the number of samples per elementary design and the number of subjects per elementary design are given and the design variables to optimise are only the sampling times. In the case of statistical optimisation, the sampling times (number and allocation) and the proportions of subjects in each elementary design are optimised.

PFIM Interface 4.0 optimises population design using the D-optimal criterion, *i.e.* maximising the determinant of the population Fisher information matrix, or, similarly, minimising its inverse.

The Fedorov-Wynn algorithm and the Simplex algorithm are available to design optimisation. Compared to the Simplex algorithm, the Fedorov-Wynn algorithm better affords high design variables optimisation. Moreover, it considers only pre-specified sampling times, avoiding, clinically unfeasible sampling times. The drawback is the huge number of elementary designs to be created (with corresponding huge number of Fisher information matrices to compute) when the set of allowed sampling times is very large.

2.5.1 Simplex algorithm

The Simplex algorithm optimises statistical or exact designs in constrained intervals, given a total number of samples.

An initial population design needs to be supplied to start the optimisation. The maximum number of elementary designs and the number of sampling times per elementary design are fixed, the sampling times and the proportions of subjects in each elementary design are then optimised. From this initial design, initial vertices for the simplex algorithm are derived, reducing successively each component by 20% (a default value which can be changed) from the original component.

PFIM Interface 4.0 uses the Splus function "fun.amoeba" from Daniel Heitjan (revised 12/94), which is a translation from the Numerical Recipes for Nelder and Mead Simplex function [10].

2.5.2 Fedorov-Wynn algorithm

The Fedorov-Wynn algorithm is specifically dedicated to design optimisation problems and has the property to converge towards the D-optimal design [11-

13]. It optimises statistical designs for a given total number of samples. The sampling times are chosen among a given finite set of times. Minimum and maximum numbers of samples per subject are specified. To start the algorithm, an initial population design is then required. The Fedorov-Wynn algorithm is programmed in a C code and is linked to PFIM Interface 4.0 through a dynamic library, called libFED.dll and libFED64.dll for R 32-bit and 64-bit respectively. Moreover, PFIM Interface uses the function `combn` in the R package "combinat".

New feature: The best one group protocol, which maximises the determinant of the elementary Fisher information matrix of all elementary protocols chosen among the predefined set of samples, is given by default when running Fedorov-Wynn algorithm (before calling the dynamic library). This is the optimal protocol for individual design and Bayesian design. Moreover, in PFIM Interface 4.0, optimisation with Fedorov-Wynn algorithm can be performed assuming that some sampling times are fixed.

3. Models

Models in PFIM Interface 4.0 can be specified either through their analytical form or as a solution of system of differential equations. PFIM Interface provides libraries of models (see Section 3.1), and users may also define their own model analytically or using a system of differential equations (see Section 3.2).

The PFIM Interface 4.0 library implements R expressions or differential equation systems for PKPD models. The PK model library includes one, two and three compartment models with linear elimination and with Michaelis-Menten elimination. The PD model library supports immediate response models (alone or linked to a pharmacokinetic model) and the turnover response models (linked to pharmacokinetic model). These libraries have been derived from the PKPD library developed by Bertrand and Mentré for the MONOLIX software, and all analytical expressions are in that document [14]. A documentation of PKPD models for PFIM Interface is available when downloading PFIM Interface 4.0. Presently, there is no model with lag time in the library.

New feature: In the previous versions of PFIM Interface, a user-defined model given in analytical form needed to be specified through an R expression. An alternative way to write the model is now available, through an R function with a specific format (see section 3.2.3).

3.1 Library of models

3.1.1 Library of pharmacokinetic models

Two types of PK models can be used in PFIM Interface 4.0, PK models with a first order linear elimination or PK models with a Michaelis-Menten elimination. The PK models with a linear elimination are written using an analytical form through an R expression whereas the PK models with a Michaelis-Menten elimination are written using a differential equation system.

The following sections show the list of models for each type of PK model in separate tables. These tables display all the information in order to use the model function chosen. The model is described by:

- a **name**
- the type of **input**
- the type of **elimination**
- the **number of compartments**
- the parameters used (**parameterisation**)
- the type of **administration** (**sd** : single dose, **md**: multiple dose, **ss**: steady state) depending on administration type, additional variables may be required. They are specified in the arguments (**N**: number of doses, **tau**: interval between two doses, **TInf**: duration of the infusion, **dose**: dose)

For models with infusion, the user has to specify the duration of infusion (TInf) in the needed variables. The rate of infusion is computed automatically in the function model by the expression: dose/TInf. The dose has to be specified in the tab design (see section 4.4).

For example, if one uses after a multiple dose administration, the first order oral absorption with one compartment model (**oral1_1cpt_kaVCl_md**) from the library which has three parameters (**ka**, **Cl** and **V**) and two needed

variables (**N**, **tau**), the number of doses (**N**) and the interval between two doses (**tau**) have to be specified.

Pharmacokinetic models with a linear elimination

The library of PK models with linear elimination is composed of one, two and three compartment models for the three types of input (bolus, infusion and first order oral absorption) and the three types of administration (single dose, multiple dose, steady state).

The list of these PK models is given in Table 1.

Table 1. Pharmacokinetic models with first order linear elimination included in the library of models

Name	Input	Cpt	Elimination	Parameterisation	Administration	Arguments
bolus_1cpt_Vk	IV-bolus	1	1st order	V, k	sd	-
					md	N, tau
					ss	tau
bolus_1cpt_VC1	IV-bolus	1	1st order	V, C1	sd	-
					md	N, tau
					ss	tau
infusion_1cpt_Vk	IV-infusion	1	1st order	V, k	sd	TInf
					md	TInf, N, tau
					ss	TInf, tau
infusion_1cpt_VC1	IV-infusion	1	1st order	V, C1	sd	TInf
					md	TInf, N, tau
					ss	TInf, tau
orall_1cpt_kaVk	1st order	1	1st order	ka, V, k	sd	-
					md	N, tau
					ss	tau
orall_1cpt_kaVC1	1st order	1	1st order	ka, V, C1	sd	-
					md	N, tau
					ss	tau
bolus_2cpt_Vkk12k21	IV-bolus	2	1st order	V, k, k12, k21	sd	-
					md	N, tau
					ss	tau
bolus_2cpt_C1V1QV2	IV-bolus	2	1st order	C1, V1, Q, V2	sd	-
					md	N, tau
					ss	tau
infusion_2cpt_Vkk12k21	IV-infusion	2	1st order	V, k, k12, k21	sd	TInf
					md	TInf, N, tau
					ss	TInf, tau

infusion_2cpt_C1V1QV2	IV-infusion	2	1st order	C1, V1, Q, V2	sd md ss	TInf TInf, N, tau TInf, tau
orall_2cpt_kaVkk12k21	1st order	2	1st order	ka, V, k, k12, k21	sd md ss	- N, tau tau
orall_2cpt_kaC1V1QV2	1st order	2	1st order	ka, C1, V1, Q, V2	sd md ss	- N, tau tau
bolus_3cpt_Vkk12k21k13k31	IV-bolus	3	1st order	V, k, k12, k21, k13, k31	sd md ss	- N, tau tau
bolus_3cpt_C1V1Q1V2Q2V3	IV-bolus	3	1st order	C1, V1, Q1, V2, Q2, V3	sd md ss	- N, tau tau
infusion_3cpt_Vkk12k21k13k31	IV-infusion	3	1st order	V, k, k12, k21, k13, k31	sd md ss	TInf TInf, N, tau TInf, tau
infusion_3cpt_C1V1Q1V2Q2V3	IV-infusion	3	1st order	C1, V1, Q1, V2, Q2, V3	sd md ss	TInf TInf, N, tau TInf, tau
orall_3cpt_kaVkk12k21k13k31	1st order	3	1st order	ka, V, k, k12, k21, k13, k31	sd md ss	- N, tau tau
orall_3cpt_kaC1V1Q1V2Q2V3	1st order	3	1st order	ka, C1, V1, Q1, V2, Q2, V3	sd md ss	- N, tau tau

Pharmacokinetic models with a Michaelis-Menten elimination

One, two and three compartment models are implemented for the three types of input. For bolus input, only single dose models are implemented. For infusion and first order absorption input, single dose and multiple dose are implemented. There is no steady-state form for PK models with Michaelis-Menten elimination (in this case the user can use a multiple dose model with enough doses to reach SS). The list of these PK models is given in Table 2.



For models with a bolus input, the dose has to be specified in the tab of the ODE variables (see section 4.3) as the initial condition of the differential equation system. For models with infusion or first order absorption input, dose has to be specified as an argument and NOT IN THE INITIAL CONDITION OF THE MODEL IN THE ODE VARIABLE TAB.



As the dose is an argument, it is not possible to specify different doses per group for models with infusion or first order absorption input. All groups of the design considered have the same dose. Otherwise, the user should use the user defined model option.

Table 2. Pharmacokinetic models with Michaelis-Menten elimination included in the library of models

Name	Input	Cpt	Elimination	Parameterisation	Administration	Arguments
bolus_1cpt_VVmkm	IV-bolus	1	Michaelis-Menten	V, Vm, km	sd	-
infusion_1cpt_VVmkm	IV-infusion	1	Michaelis-Menten	V, Vm, km	sd md	doseMM, TInf doseMM, TInf, tau
orall_1cpt_kaVVmkm	1st order	1	Michaelis-Menten	ka, V, Vm, km	sd md	doseMM doseMM, tau
bolus_2cpt_Vk12k21Vmkm	IV-bolus	2	Michaelis-Menten	V, k12, k21, Vm, km	sd	-
bolus_2cpt_V1QV2Vmkm	IV-bolus	2	Michaelis-Menten	V1, Q, V2, Vm, km	sd	-
infusion_2cpt_Vk12k21Vmkm	IV-infusion	2	Michaelis-Menten	V, k12, k21, Vm, km	sd md	doseMM, TInf doseMM, TInf, tau
infusion_2cpt_V1QV2Vmkm	IV-infusion	2	Michaelis-Menten	V1, Q, V2, Vm, km	sd md	doseMM, TInf doseMM, TInf, tau
orall_2cpt_kaVk12k21Vmkm	1st order	2	Michaelis-Menten	ka, V, k12, k21, Vm, km	sd md	doseMM doseMM, tau
orall_2cpt_kaV1QV2Vmkm	1st order	2	Michaelis-Menten	ka, V1, Q, V2, Vm, km	sd md	doseMM doseMM, tau
bolus_3cpt_Vk12k21k31k13Vmkm	IV-bolus	3	Michaelis-Menten	V, k12, k21, k13, k31, Vm, km	sd	-
bolus_3cpt_V1Q1V2Q2V3Vmkm	IV-bolus	3	Michaelis-Menten	V1, Q1, V2, Q2, V3, Vm, km	sd	-
infusion_3cpt_Vk12k21k13k31Vmkm	IV-infusion	3	Michaelis-Menten	V, k12, k21, k13, k31, Vm, km	sd md	doseMM, TInf doseMM, TInf, tau
infusion_3cpt_V1Q1V2Q2V3Vmkm	IV-infusion	3	Michaelis-Menten	V1, Q1, V2, Q2, V3, Vm, km	sd md	doseMM, TInf doseMM, TInf, tau
orall_3cpt_kak12k21k13k31Vmkm	1st order	3	Michaelis-Menten	ka, k12, k21, k13, k31, Vm, km	sd md	doseMM doseMM, tau
orall_3cpt_kaV1Q1V2Q2V3Vmkm	1st order	3	Michaelis-Menten	ka, V1, Q1, V2, Q2, V3, Vm, km	sd md	doseMM doseMM, tau

3.1.2 Library of pharmacodynamic models

The library of PD models supports immediate response models (either as a function of observed concentrations, or linked to a pharmacokinetic model) and turnover response models (linked to pharmacokinetic models). The following tables present these models, giving the following elements for each drug model:

- the **name** of the model function in the library
- the parameters used (**parameterisation**)

Immediate response pharmacodynamic models alone

Linear, quadratic, logarithmic, Emax, sigmoid Emax, I_{max}, sigmoid I_{max} models with null or constant baseline are available. The list of these models is given in Table 3.

These models are written in closed form and can be used in the case of a model with one response (PD evaluation or optimisation).

For these models, the design variables are the concentrations or the doses instead of the sampling times.

For example, if one uses a linear drug action model with a constant baseline (**immed_lin_const**) from the library, the model uses two parameters (**Alin, S0**).

Pharmacodynamic models linked to pharmacokinetic model

In this section, we consider models with two responses, with one response for the PK and the other one for the PD. We thus optimise sampling times for both responses using a PK/PD model. Using the libraries of models, we have four cases to compose the PK/PD model depending on the form for each submodel: either with an analytical form (AF) or a differential equation system (ODE).

Therefore, there are four cases of PK/PD models in PFIM library:

1. PK model with linear elimination (AF) and immediate response PD model (AF)
2. PK model with linear elimination (AF) and turnover response PD model (ODE)
3. PK model with Michaelis-Menten elimination (ODE) and immediate response PD model (AF)
4. PK model with Michaelis-Menten elimination and turnover response PD model (ODE)

Table 3. Immediate response pharmacodynamic models included in the PD library for PD alone and for PK/PD model

Drug action models	Baseline			
	Null baseline		Constant baseline	
	Name	Parameterisation	Name	Parameterisation
Linear	immed_lin_null	Alin	immed_lin_const	Alin, S0
Quadratic	immed_quad_null	Alin, Aquad	immed_quad_const	Alin, Aquad, S0
Logarithmic	immed_log_null	Alog	immed_log_const	Alog, S0
E _{max}	immed_E_{max}_null	E _{max} , C50	immed_E_{max}_const	E _{max} , C50, S0
Sigmoid E _{max}	immed_gammaE_{max}_null	E _{max} , C50, gamma	immed_gammaE_{max}_const	E _{max} , C50, gamma, S0
I _{max}	immed_I_{max}_null	I _{max} , C50	immed_I_{max}_const	I _{max} , C50, S0
Sigmoid I _{max}	immed_gammaI_{max}_null	I _{max} , C50, gamma	immed_gammaI_{max}_const	I _{max} , C50, gamma, S0

To use PFIM Interface for design evaluation and optimisation for a PK/PD model, the two models must be in the same format.

In the first case, immediate response pharmacodynamic models are written with an analytical form and thus they can be associated to pharmacokinetic models with first order linear elimination (Table 1) which are also written with analytical forms. In this case, the user has to complete the tab using analytical form options.

However, for the three other cases, the PK response and the PD response are written either with different forms or both with a differential equation system (Case 4). That is why, PFIM Interface 4.0 calls a specific function in order to create a system of differential equations describing the corresponding PK/PD model.

For these cases, the user has thus to complete the tab of the ODE variables (section 4.3.3).

The list of the immediate response PD models is thus given in Table 3 plus those of Table 4. The list of the turnover response PD models is given in Table 5.

For the second case where a PK model with linear elimination is associated to a turnover PD response model, the PK model is written with a differential equations system. Consequently, only some PK models from the Table 1 are implemented:

- for bolus input, only single dose models;
- for infusion input, single dose and multiple dose
- for first order absorption input, single dose and multiple dose\\



For models with a bolus input, the dose has to be specified in the tab of the ODE variables (section 4.3.3) as the initial condition of the differential equation system. For models with infusion or first order absorption input, dose has to be specified as an argument. Consequently, it is not possible to specify different doses per group for models with infusion or first order absorption input. All groups of the design considered have the same dose. Otherwise, the user should use the user defined model option.

Table 4. Immediate response pharmacodynamic models linked to a pharmacokinetic model included in the library*

Drug action models	Baseline/disease models					
	Linear progression		Exponential increase		Exponential decrease	
	Name	Param.	Name	Param.	Name	Param.
Linear	immed_lin_lin	Alin, S0, kprog	immed_lin_exp	Alin, S0, kprog	immed_lin_dexp	Alin, S0, kprog
Quadratic	immed_quad_lin	Alin, Aquad, S0, kprog	immed_quad_exp	Alin, Aquad, S0, kprog	immed_quad_dexp	Alin, Aquad, S0, kprog
Logarithmic	immed_log_lin	Alog, S0, kprog	immed_log_exp	Alog, S0, kprog	immed_log_dexp	Alog, S0, kprog
E _{max}	immed_E_{max}_lin	E _{max} , C50, S0, kprog	immed_E_{max}_exp	E _{max} , C50, S0, kprog	immed_E_{max}_dexp	E _{max} , C50, S0, kprog
Sigmoid E _{max}	immed_gammaE_{max}_lin	E _{max} , C50, gamma, S0, kprog	immed_gammaE_{max}_exp	E _{max} , C50, gamma, S0, kprog	immed_gammaE_{max}_dexp	E _{max} , C50, gamma, S0, kprog
I _{max}	immed_I_{max}_lin	I _{max} , C50, S0, kprog	immed_I_{max}_exp	I _{max} , C50, S0, kprog	immed_I_{max}_dexp	I _{max} , C50, S0, kprog
Sigmoid I _{max}	immed_gammaI_{max}_lin	I _{max} , C50, gamma, S0, kprog	immed_gammaI_{max}_exp	I _{max} , C50, gamma, S0, kprog	immed_gammaI_{max}_dexp	I _{max} , C50, gamma, S0, kprog

* In addition to those in Table 3.

Table 5. Turnover response pharmacodynamic models linked to a pharmacokinetic model included in the library

Types of response	Models with impact on the			
	Input		Output	
	Name	Parameterisation	Name	Parameterisation
Emax	turn_input_Emax	Rin,kout,Emax,C50	turn_output_Emax	Rin,kout,Emax,C50
Sigmoid Emax	turn_input_gammaEmax	Rin,kout,Emax,C50,gamma	turn_output_gammaEmax	Rin,kout,Emax,C50,gamma
Imax	turn_input_Imax	Rin,kout,Imax,C50	turn_output_Imax	Rin,kout,Imax,C50
Sigmoid Imax	turn_input_gammaImax	Rin,kout,Imax,C50,gamma	turn_output_gammaImax	Rin,kout,Imax,C50,gamma
Full Imax^a	turn_input_Imaxfull	Rin,kout,C50	turn_output_Imaxfull	Rin,kout,C50
Sigmoid full Imax^a	turn_input_gammaImaxfull	Rin,kout,C50,gamma	turn_output_gammaImaxfull	Rin,kout,C50,gamma

^a Full Imax means Imax is fixed equal to 1

3.2 User-defined models

Users can also define their own model, analytically (as an R expression or an R function) or using a system of differential equations. A file has to be created according to each model form (see Section 4.3.2).

3.2.1 Analytical form defined through an R expression

Description

The model file must start with the three following red lines, without any space between each line; moreover they have to start with the key symbol "#\$" to be understood by PFIM.

```
#$Model definition  
#$ka, V, Cl  
#$  
formA <-expression()  
form<-c(formA)  
tf<-list(Inf)
```

The first line cannot be changed. The second line indicates the names of the parameters of the model to be estimated. The third line specified the argument(s) when a function is specified on the fourth line.

The user must start to specify the model from the fourth line. Here only "#\$" is written on the third line (no additional argument to be specified since the model equation is given directly using the R function "expression" on the fourth line.

In case of analytical form, the model for each response should be written assigned in an object called 'form i ' where i is the letter of the alphabet A,B,C,... The "form i " for all the responses are then grouped in a vector called "form":

```
form<-c(formA, formB, formC, ...)
```

If the model for a response is defined over intervals by different expressions, each response should be written as a vector of expressions. Each expression can be defined in an object 'form I ', where $I = 1, 2, 3, \dots$. For example, if the user wants to give three expressions for the first response, he can write as follows:

```
formA<-c(form1, form2, form3)
```

formA can be a model of the PFIM libraries or defined by the user.

User also needs to define an object "tf" which indicates the time until when to use the model for the expression form1. "tf" has to be a list of objects corresponding to "tf" for each response. The length of "tf" must be equal to the number of responses. In case of one response model, using one expression defined from 0 to Infinity:

```
tf<-c(Inf)
```

Example 1: Single response model at steady-state in analytical form using an R expression

```
#$Model definition
#$k,V
#$
formA <-expression(1/(V)/(1-exp(-k*24))*(exp(-k*t)))
form<-c(formA)
tf<-list(Inf)
```

The analytical expression in this example describes a one compartment model after IV bolus at steady-state, with a dose interval equal to 24h (bolus_lcpt_Vk). In this case, if the dose is not equal to 1, the user has to specify the variable "dose" in the expression and the value of the dose in the design tab (see section 4.4). However, if the user defines his model, he can also specified the value of the dose in the analytical expression by replacing 1 by 500 for instance for a dose equal to 500. In this case, the user has to put in the design tab the dose equal to 1. If the dose is defined by the user in the analytical expression of the model, the options of design with multiple groups with different doses can no longer be used. The "tf" object indicates the time until when to use the specified expression for the model (here, time Infinity).

Example 2: PK model with a linear elimination and immediate response PD model in analytical form using an R expression

```
#$Model definition
#$ka,V,C1,Imax,C50,S0
#$
formA<-expression(dose/V*ka(ka-(C1/V))*(exp(-(C1/V)*t)-exp(-ka * t)))
formB<-paste("-Imax*",formA,"/(C50+",formA,")+S0")
formB<-parse(text=formB)
tf<-list(Inf,Inf)
form<-c(formA,formB)
```

These analytical expressions describe a PK/PD model. The PK model is a one compartment model with a first order absorption and elimination (**formA**) and the PD model (**formB**) is an immediate response model with a constant baseline. In this case, the user has to specify the dose in the design tab (see section 4.4).

Note that to write formB, we use the R function *paste* which converts its arguments to character strings and concatenate them. The R function *parse* is used in order to obtain an expression of the model.

The "tf" object indicates the time until when to use the model for the PK (here, time Infinity) and until when to use the PD model (here, time Infinity). Here, "tf" is thus a list of two elements.

"form" is the vector of the models for all responses, and the second object is "tf".

Example 3: PK model after multiple dose administration using an analytical form, based on functions from the library of models

It is also possible to use the functions of the library of models to create new models. This is illustrated in the following example.

```
##$Model definition
##$ka,V,k
##$
form1<-oral_1cpt_kaVk() [[1]]
form2<-oral_1cpt_kaVk_md(N=5,tau=12) [[1]]
formA<-c(form1,form2)
tf<-list(c(12,Inf))
form<-formA
```

In this illustration, the user creates a model combining two analytical expressions for a one compartment oral absorption: the first expression corresponds to the model after the first administration (form1) and the second expression corresponds to the model after the fifth administration (form2). Use of predefined functions of the library of models implies the use of "[[1]]" at the end of the call of the function to select the part of the function corresponding to the expression of the model.

The "tf" object indicates the time until when to use the model for the first administration (here, time 12) and until when to use the model for the fifth administration (here, time Infinity). Then, the user defines two objects: the first object is "form", the vector of the models, and the second one is "tf".

This case is useful for evaluation and optimisation of a design including sampling times after the first and the fifth administration. If the user defines the model by using the library of model, he has to specify the dose in the design tab (see section 4.4).

Example 4: PK model with a linear elimination and immediate response PD model in analytical form, based on functions from the library of models

This example illustrates how to write a PK/PD model using functions implemented the PKPD library.

```
##$Model definition
##$ka,V,Cl,Imax,C50,S0
##$
formA<-orall_1cpt_kaVCl() [[1]]
formB<-immed_Imax_const(formA) [[1]]
tf<-list(Inf,Inf)
form<-c(formA,formB)
```

3.2.2 Analytical form defined through an R function

Description

The R function for a PFIM Infertace 4.0 model should take the following form:

```
#$Model definition
#$ ka,k,V
#$
form<-function(t,p,X) {
. . .
}
```

The four lines in red have to be written, without any space between each line; moreover the three first lines have to start with the key symbol "\$" to be understood by PFIM. The first line cannot be changed. The second line indicates the names of the parameters of the model to be estimated. The third line specified the additional argument(s) of the function. Here there is no additional argument except t, y and p which are default arguments. Therefore only "\$" is written on the third line. Last, the fourth line indicates the name of the function (form) which **must remain unchanged**. The 3 arguments of the function are:

- a vector of times t
- a vector of parameters p
- a scalar X which represents **the dose**

The function returns a vector of predictions of each time point in t, computed using the dose X and the parameters p.

Example 5: PK model after single dose administration using an analytical form with user-defined R function

```
#$Model definition
#$ ka,k,V
#$
form<-function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

In this example, the user creates a function of a one response model describing a one compartment oral absorption.

Example 6: PK model after multiple dose administration using an analytical form with user-defined R function

```
##$Model definition
##$ ka,V, Cl
##$
form<-function(t,p,X){
ka<-p[1]
V<-p[2]
Cl<-p[3]

N<-5
tau<-12

y<-0
for (n in 1:N)
{
indic<-t>=(n-1)*tau
yn<-indic*(X/V*ka/(ka-Cl/V)*(exp(-Cl/V*(t - (n - 1) * tau))-exp(-ka*(t -
(n - 1) * tau))))
y<-y+yn
}
return(y)
}
```

In this illustration, the user creates a function of one response model describing a one compartment oral absorption after five administration doses with a between dose interval equal to twelve hours. The number of doses and the between dose interval are defined within the function.

3.2.3 Models defined through a differential equation system

Description

Model defined as a solution of a differential equation system must be called "formED". It can be given by the users who need to write an R function in a format suitable for the solver package deSolve, using the following form:

```
##$Model definition ODE
##$ka,km,Vm,V
##$
formED<-function(t,y,p)
{
...
}
```

The four lines in red have to be written, without any space between each line; moreover, the three first lines have to start with the key symbols "\$". The first line of this Block, '\$Model definition ODE' cannot be changed. The second line indicates the names of the parameters of the model to be estimated. The third line specified the additional argument(s) of the function. Here there is no additional argument, except t, y and p which are default arguments. Therefore only "\$" is written on the third line. Last, the fourth line indicates the name of the function (formED) which **must remain unchanged**. The 3 arguments of the function are:

- a vector of time t
- the current estimate of the variables in the ode system y
- a vector of parameters p

Within the function, the user has to define the name of the parameters in vector p and the differential equation system.

The function returns a list with 2 elements:

- the first element is a vector giving the values of the derivatives for each equation in the differential equation system, computed for each time point in t using the parameters p
- the second element is a vector of predictions computed for each time point in t using the parameters p; in PFIM, this vector contains the response(s) we are observing

The initial values of the system have to be specified in the ODE tab presented in the section 4.3.3.

The implementation of differential equations system requires the use of the lsoda function included in the library "deSolve" (R. Thomas Petzoldt) and of the fdHess function included in the library "nlme" developed by Jose Pinheiro and Douglas Bates.

The lsoda function uses a function of the same name written in Fortran by Linda R. Petzold and Alan C. Hindmarsh. This function solves system of differential equations using the Adams method, a predictor - corrector method for non-stiff systems; it uses the Backward Differentiation Formula (BDF) for stiff systems. The fdHess is used for numerical derivation. It evaluates an approximate gradient of a scalar function using finite differences.

Example 7: Single response PK model using a differential equation system created by the user

```

#$Model definition ODE
#$ka,km,Vm,V
#$
formED<-function(t,y,p)
  {
    ka<-p[1]
    km<-p[2]
    Vm<-p[3]
    V<-p[4]

    yd1<--ka*y[1]
    yd2<-+ka*y[1]- V * (Vm * y[2]/(V * km + y[2]))

    list(c(yd1,yd2),c(y[[2]]/V))
  }

```

This example describes a one compartment model first order absorption and Michaelis-Menten elimination.

The first four lines in the body of the function assign model parameters from the vector p. The next two lines describe the derivatives of the system (yd1 and yd2). More specifically, each derivative represent the drug concentration in the specific compartment at the instant t, and its elements can be either positive or negative. The notation ydX denotes the

derivative of the variable in compartment X while the notation $y[X]$ denotes the quantity in the same compartment (see documentation for the deSolve package for details). The last line defines the elements returned by the function:

- the first item is mandatory for the deSolve package, and should always consist of a vector with the derivatives of the system (here, the two elements $yd1$ and $yd2$)
- the second item defines the response, here the concentration in the second (central) compartment which is defined by the quantity in this compartment ($y[2]$) divided by the volume of distribution V . Several responses can be given.

Example 8: Multiple response PK model using a differential equation system created by the user

```
##$Model definition ODE
##$ka,cl,V,Clm,R
##$
formED<-function(t,y,p)
{
  ka<-p[1]
  cl<-p[2]
  V<-p[3]
  clm<-p[4]
  R<-p[5]
  yd1<-ka*y[1]
  yd2<-ka*y[1]-cl/V*y[2]-R*y[2]
  yd3<-R*y[2]-clm*y[3]

  list(c(yd1,yd2,yd3),c(y[2]/V,y[3]))
}
```

This example describes a two response model using a differential equation system. In this case, the second argument of the list is composed of two objects corresponding to: the first measure of interest is the concentration in the compartment 2 scaled by the volume and the second measure of interest is the concentration in the compartment 3, respectively.

Example 9: PK model after multiple dose administration using a differential equation system created by the user

```
#$Model definition ODE
#$ka,V, Cl
#$
formED<-function(t,y,p)
{
  ka<-p[1]
  V<-p[2]
  Cl <-p[3]

  tau<-12
  input_orall<-function(ka,V,dose,n,tau,t){
    if(n==0){return(dose*ka/V*exp(-ka*t))}
    else{return(dose*ka/V*exp(-ka*(t-
n*tau))+input_orall(ka,V,dose,n-1,tau,t))}
  }
  n<-t/%tau
  input<-input_orall(ka,V,dose,n,tau,t)

  dy<--Cl/V*y[1]+input

  list(c(dy),c(y[1]))
}
```

In this illustration, the user creates a function of one response model describing a one compartment oral absorption after multiple dose administration with a between dose interval between two doses equal to twelve hours. The number of doses and the between dose interval are defined within the function.

4. Installation

The R 2.6 or higher and available in Windows operating system (32bits or 64bits) needs to be installed. Depending on the use of PFIM Interface 4.0, additional packages available in the R library are needed:

to use a differential equation system to describe the model: "deSolve" and "nlme" packages

to use the Federov-Wynn algorithm: "combinat" package.

An additional package "numDeriv" is needed for the computation of the full Fisher information matrix and for numerical derivatives of models written as standard R functions

The easiest way to install packages is directly from the web. To install the packages deSolve, nlme, combinat and numDeriv, start R and choose the Packages item from the menu. Choose Install package(s) from CRAN to install from the web (you will see a list of all available packages pop up -- choose deSolve, nlme, combinat and numDeriv).

4.1 Windows installation

To install the **Windows version** of PFIM Interface 4.0, download the application '<http://www.pfim.biostat.fr/download/PFIM-4.0-windows-installer.exe>' available on the webpage www.pfim.biostat.fr. Then simply double click on this application and click on the button to execute the program.

To complete the installation of PFIM interface 4.0, follow the different steps detailed below.

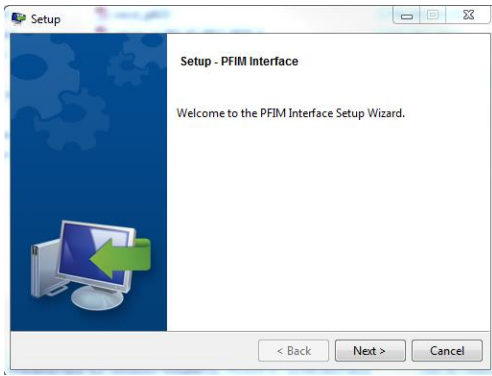


Figure 1: Click the button "Next" to continue the procedure

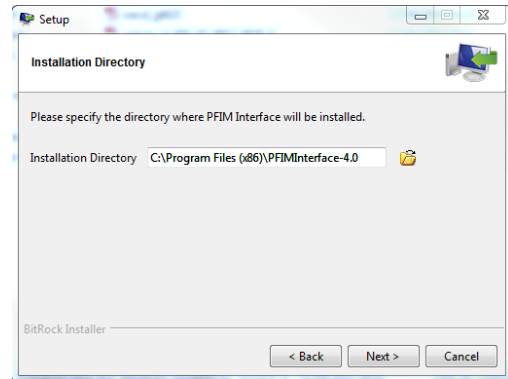


Figure 2: Indicate the path to install the directory files and click the button next

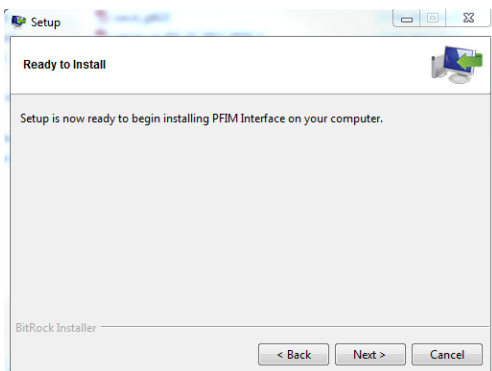


Figure 3: Click the button « Next » To begin the installation

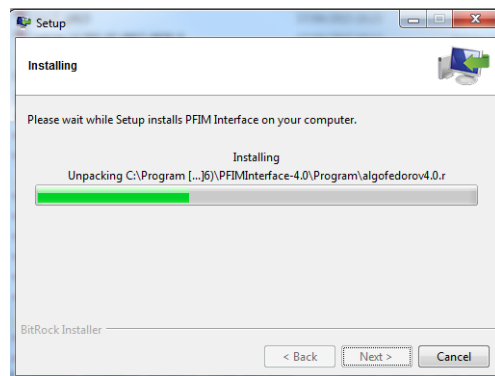


Figure 4: The setup is proceeding

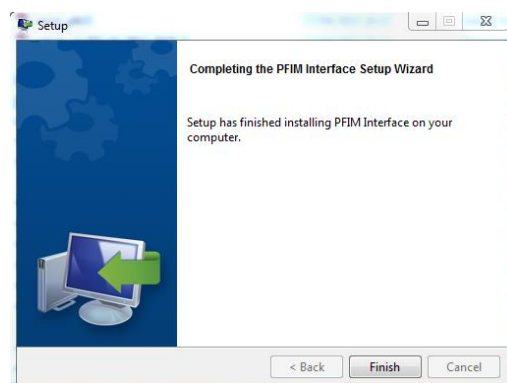


Figure 5: To complete the installation click on the button "Finish". Installation is successful

5. Use

5.1 Main user Interface

Figure 6 shows the screen that appears when the user starts the program PFIM Interface 4.0.

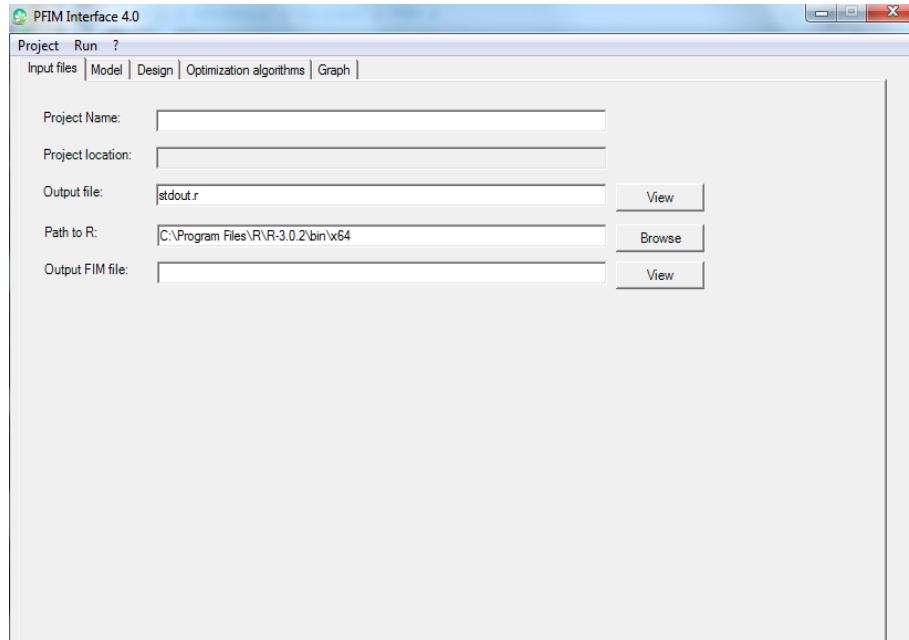


Figure 6: PFIM Interface 4.0: initial screen.

The user can either create a new project directory (File/New project) or load an existing one (File/ Open project) as it is shown in Figure 7.

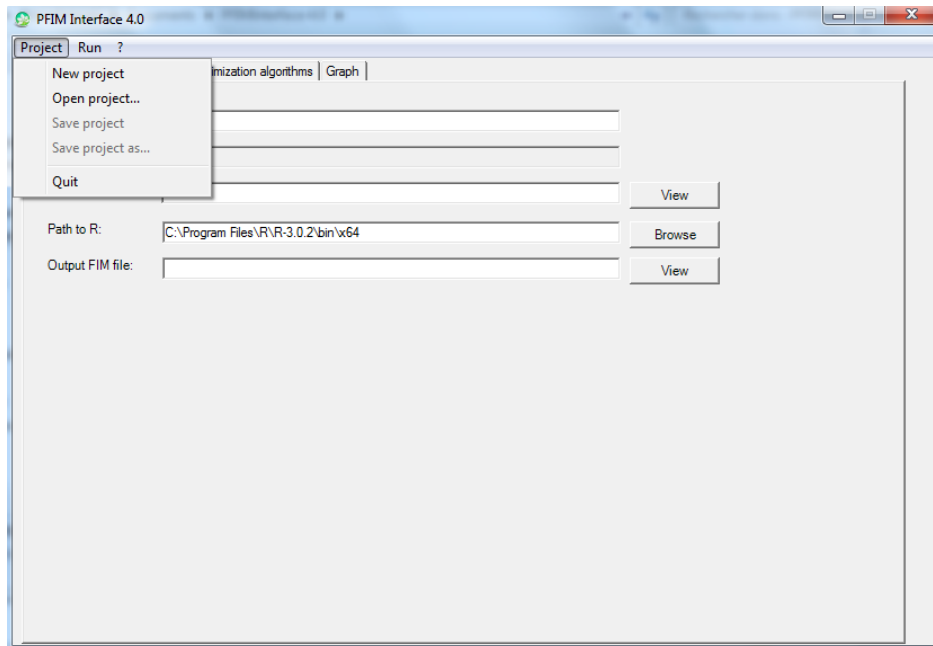


Figure 7: Load or create a new project.

5.1.1 Creation of a new project

In order to create a new project, the path of the directory for the "New project" must be specified as shown in Figure 8.

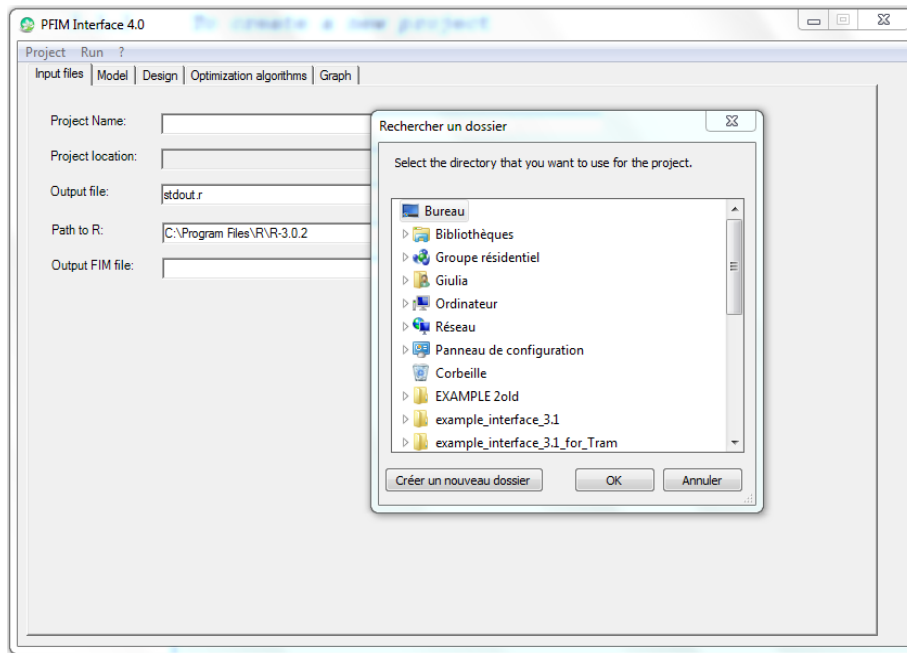


Figure 8: Choose directory for the new project

By default, the names of the new project and of the output file (Figure 9) are "My Project" and Stdout.r, respectively, but these names can be modified.

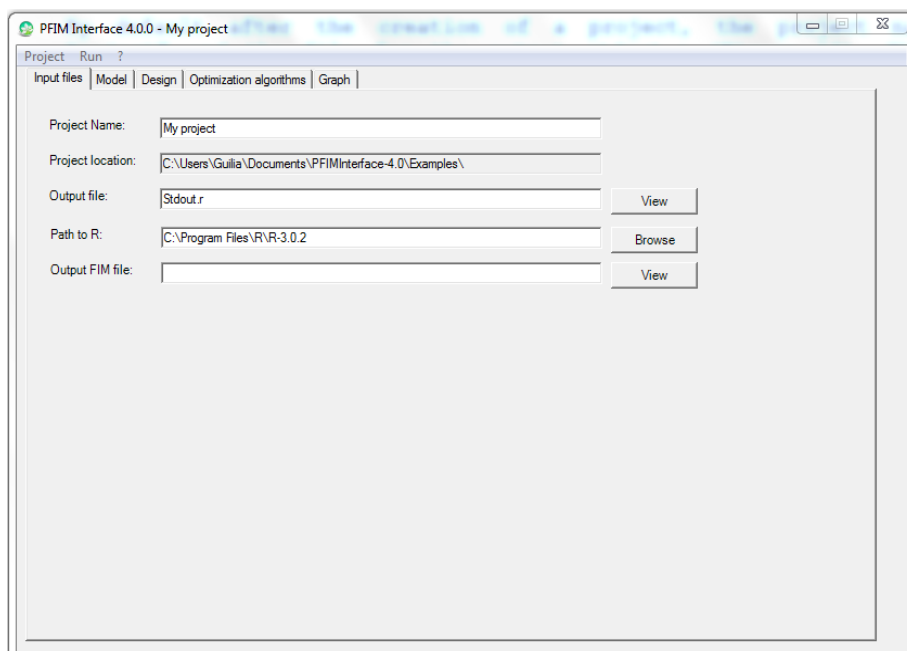


Figure 9: Default tab after the creation of a project.

5.1.2 Loading existing project

In order to load an existing project, it is either possible to use the Menu of PFIM Interface 4.0 as shown in Figure 10, or by clicking on the stdin.pfim file stored in the directory of the project selected.

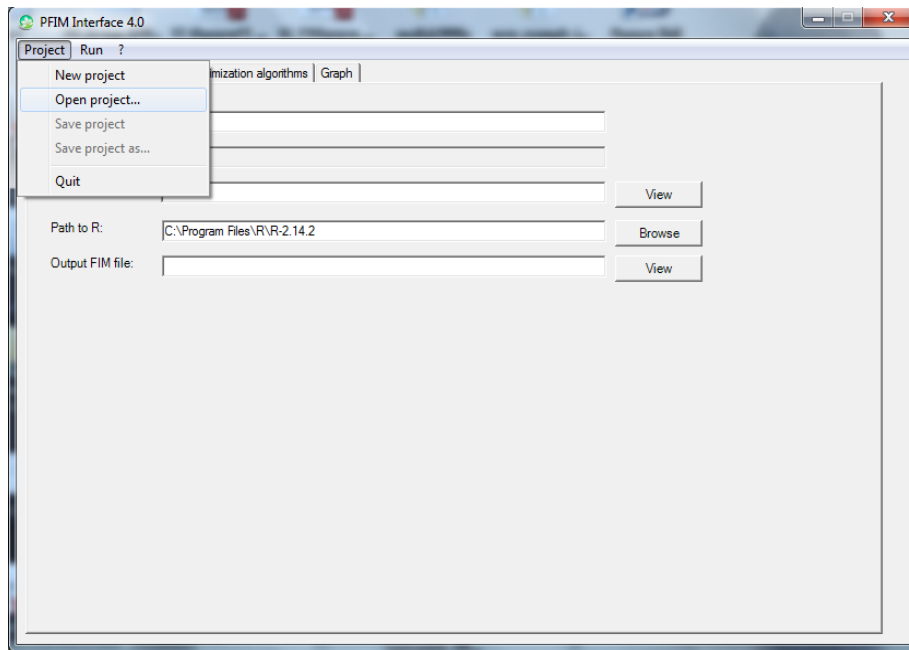


Figure 10 Loading existing project tab

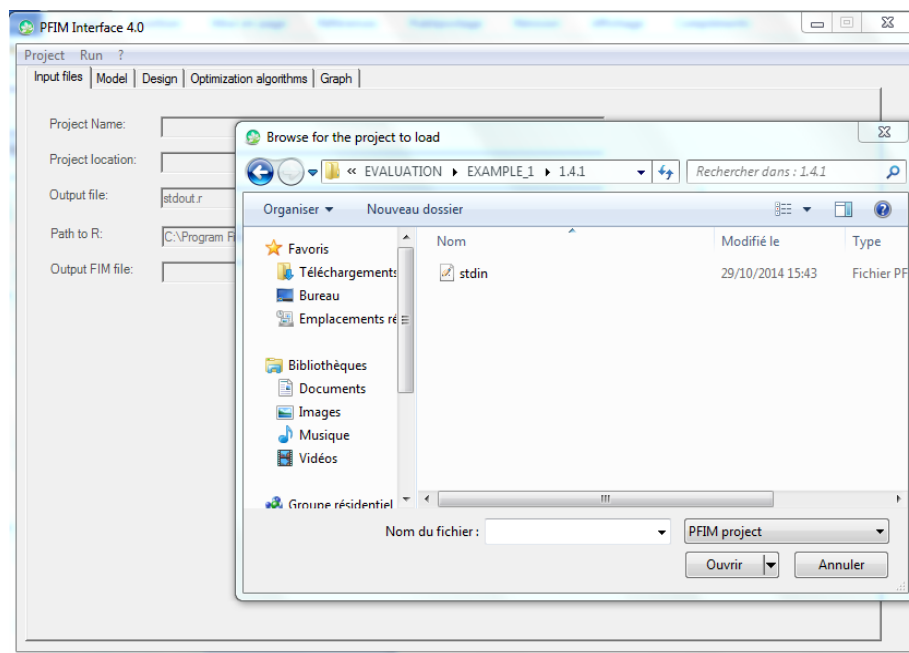


Figure 11 Browsing project to be loaded tab

5.2 Input tab

The user should enter a name for the project and specify the name for the output file where the results will be stored. The Project location is created automatically once selected the folder where the new project can be stored. The user can choose the R version to be used by specifying the path in the "Path to R" section. It is also possible to require the saving of

the Fisher Information Matrix in a text file by specifying the name of the text file ("filename.txt") in the "Output FIM file" section.

5.3 Model tab

This tab is for model specification. A model can be either selected from the PFIM library or it can be written in a user defined form, using either analytical form or a system of differential equations (ODE). Below some figures showing how specify a model in the PFIM Interface 4.0. In the "Models" section of this documentation, model writings and examples are reported in more details.

5.3.1 Model from the library

After having opened a project or created a new project in PFIM Interface 4.0 it is possible to select the preferred model from the library of models using the scroll bar in the model tab (Figure 12) and choose the regimen (Single dose, Multiple doses, Steady State). For the first example we selected a PK one compartment first order absorption model, with parameters k_a , V , k , and a single dose regimen.

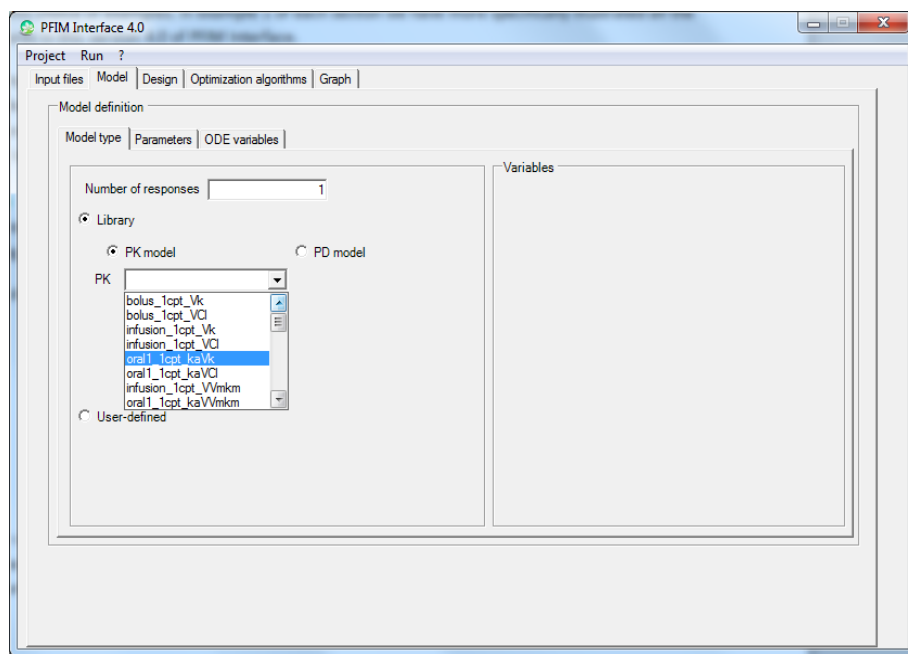


Figure 12: Model-selection from the model library

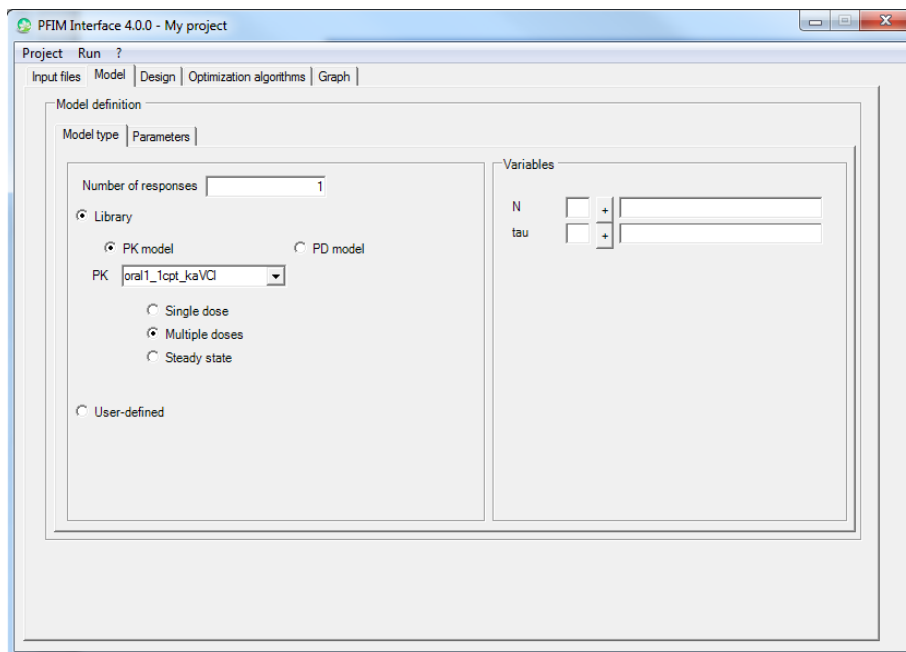


Figure 13: Specification of multiple doses administration type and specification of the needed variables N and tau

It is also possible to define a PKPD model through the model library as shown is Figure 14. Please mind to specify the two responses in the number of responses section.

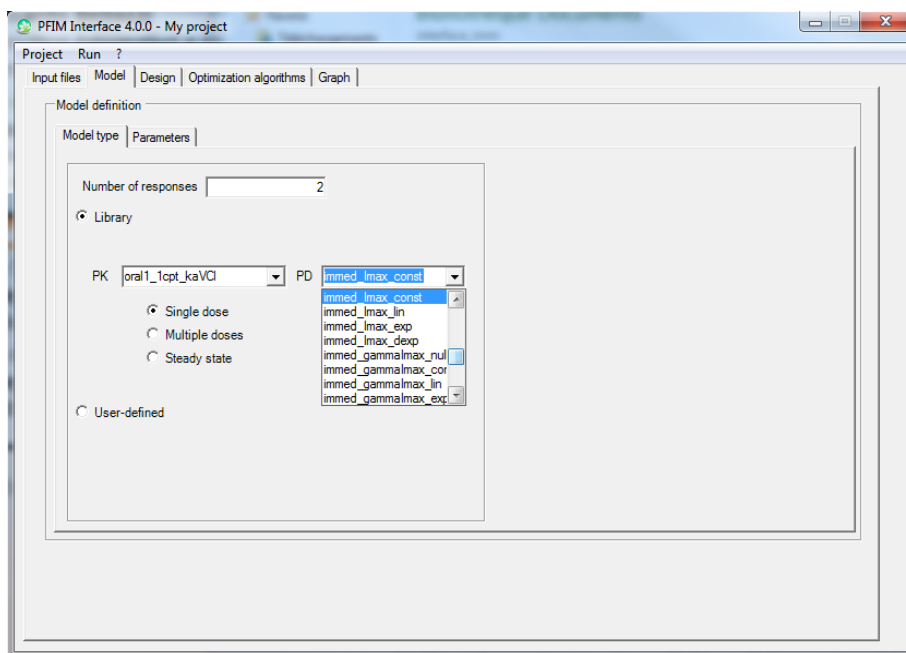


Figure 14: PKPD model selection from the model library. The PK is a one compartment first order oral absorption model, with function in the library parameterized in k_a , V and k . The PD is an I_{max} model with constant baseline parameterized in I_{max} , C_{50} and S_0 .

5.3.2 User-defined model

The user can create either an analytical model or an ODE system (Figure 15). If a user defined model has already been specified in this project, it can also be edited and modified.

See "Models" section for details on how to write its own model.

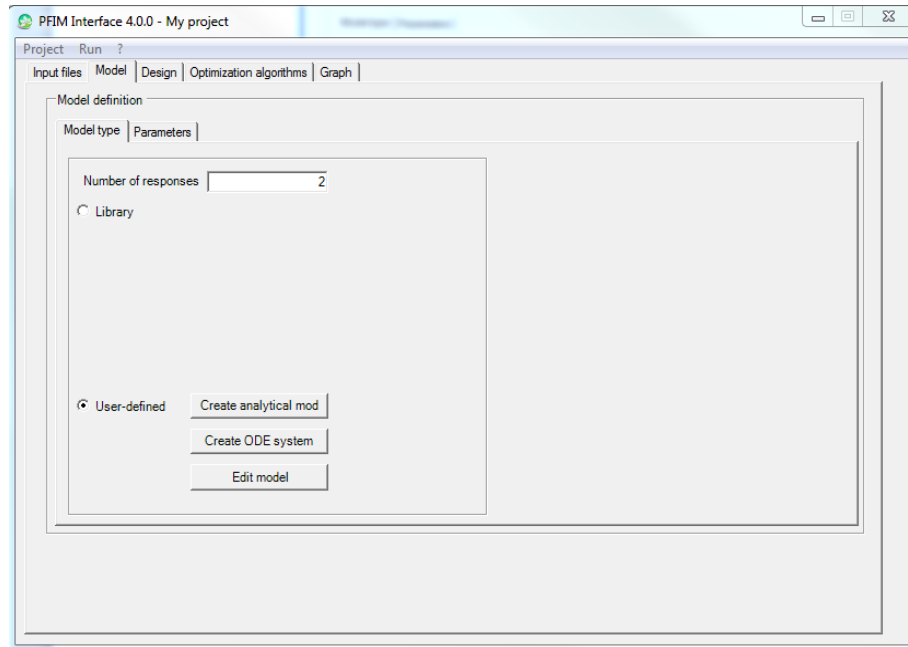


Figure 15: Definition of the model by the user here for a model with two responses

5.3.3 Parameters of the model

Once the model is specified, one can click on the "Parameters" section (Figure 16), where values of the mean and the variance of the population parameters have to be defined.

If the between-subject variance of a parameter is assumed to be zero, this should be specified under the variance column of the related parameter. PFIM would then remove the corresponding row and column in the Fisher information matrix. In the parameter tab it is also possible to indicate whether some parameter should be kept fixed in the evaluation or optimisation of FIM. In that case, the variance of the correspondent fixed parameter will be automatically set to 0.

In the same tab it is possible to choose between either additive or exponential model for the between-subject variance.

Values of the standard deviation of the residual error should be specified. The residual error is additive with a general model for variance: $\text{var}(\varepsilon) = (\sigma_{\text{inter}} + \sigma_{\text{slope}} * f)^2$, where f is the structural model. This variance error model includes the constant variance model ($\sigma_{\text{slope}} = 0$) or the constant coefficient of variation model ($\sigma_{\text{inter}} = 0$) as special cases. The parameters σ_{inter} and σ_{slope} are included in the population parameters to be estimated.

Regarding a multiple response model, the user has to complete the different values for the standard deviation of the residual error for all responses one by one using the list box (Figure 17).

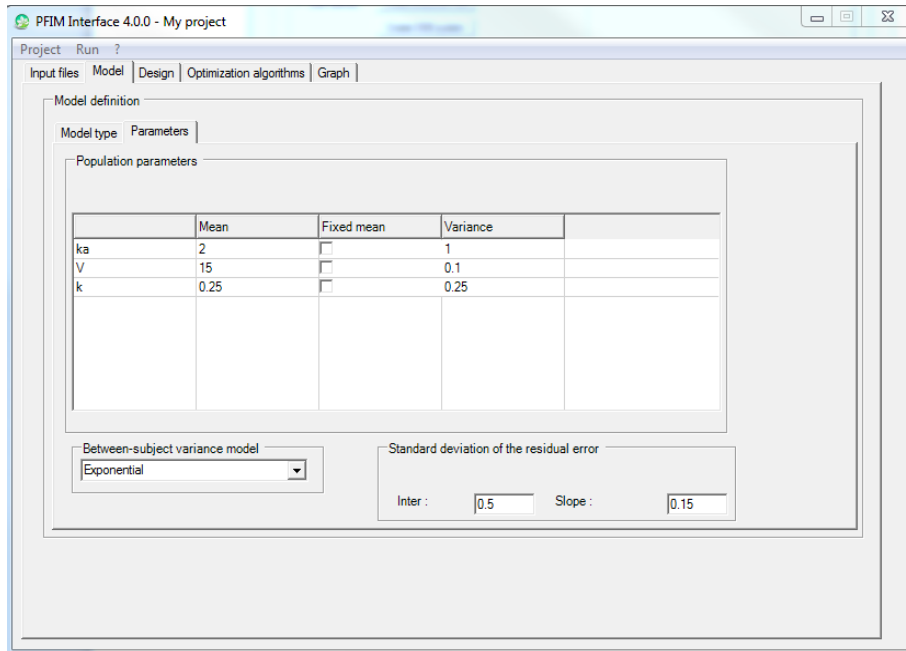


Figure 16: Parameters section tab for a single response model

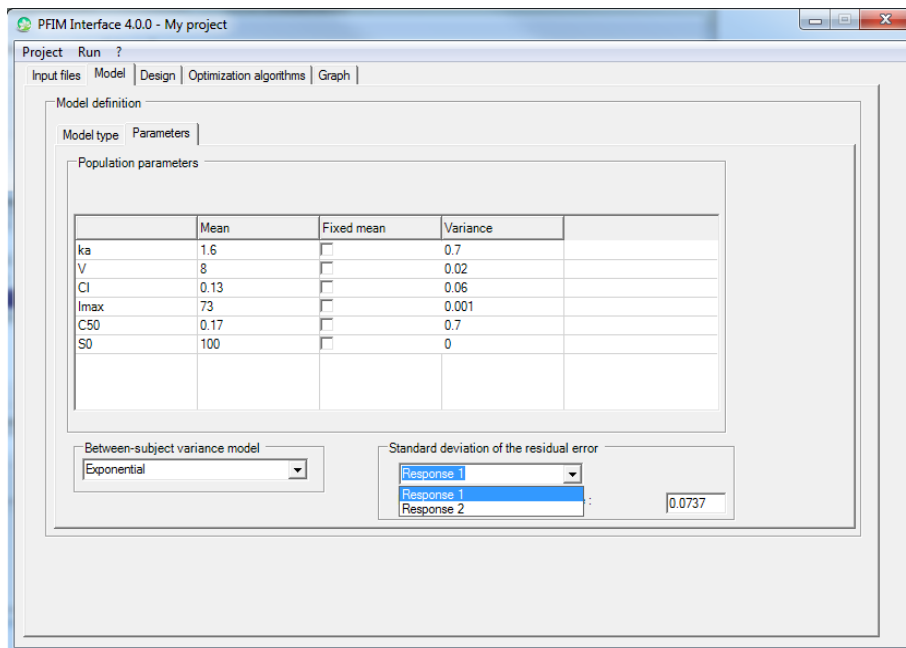


Figure 17: Parameters section tab for a multiple response model with the list box to choose the parameters of the residual error for each response

In case of an ODE system, the ODE variables section has to be filled as in Figure 18.

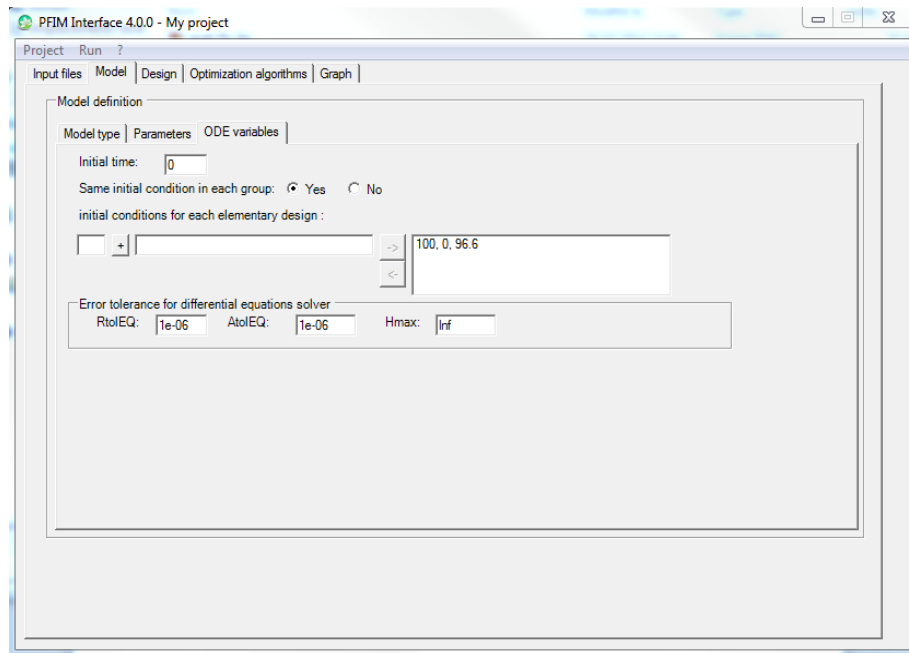


Figure 18: Tab of the ODE variables

In this case, the time for the initialisation of the system must be given (usually 0) and the initial values in each compartment at this time have to be given as a vector in the "initial conditions for each elementary design". The size of this vector has to be equal to the number of equations of the system. **For the models coming from the libraries, the number of equations of the system is equal to the number of compartments of the model.** Several vectors can be specified in case of different groups of subjects with different initial conditions.

To use this tab, an example is presented below illustrated in Figure 19 using a system with two equations.

If there are two elementary design with different initial conditions defined as (13.8; 0) and (15; 0) respectively for the first and the second elementary design. The size of the vectors for the initial conditions is equal to 2 due to the two equation system.

To input the initial condition for the first elementary design, the user has to follow the next instructions with the illustration on Figure 19.

- Put the value 13.8 in the white box **1**.
- To validate the value click on the button **2**, the value is in the box **3**.
- Repeat these two previous steps for the value 0.
- Click on the button **4**, the first elementary design is specified and validated in the box **5**.

In order to validate the initial conditions for the second elementary design defined here 15 and 0, the user has to repeat the previous steps. The screen showed on Figure 19 should be obtained. The first line and the second line in the box **5** correspond respectively to the initial conditions for the first and the second elementary design.

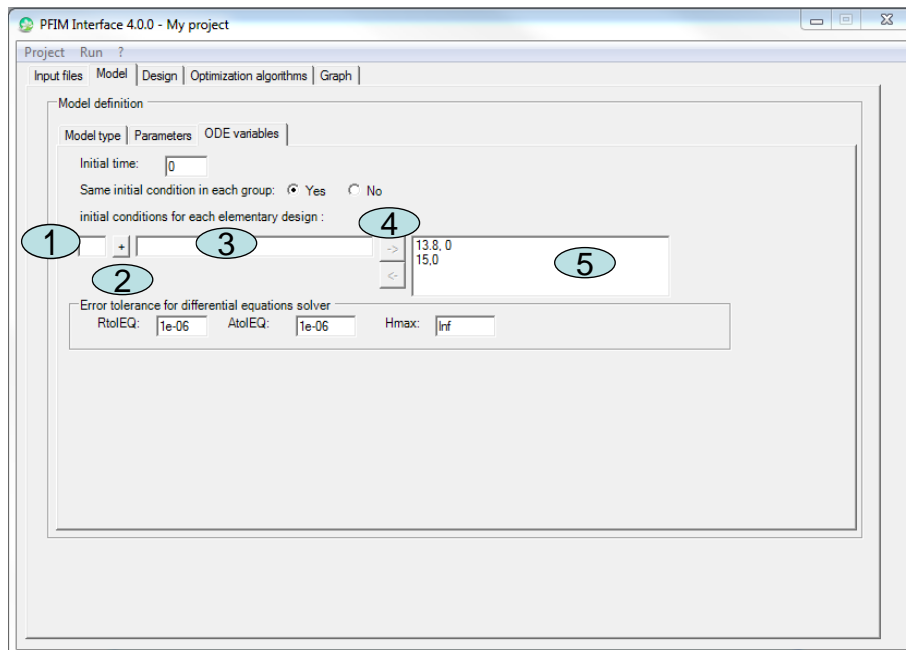


Figure 19: Tab of the ODE variables: illustration to enter the initial conditions

Finally, parameters of "Error tolerance for the differential equation solver" are set by default but can be changed by the user. Those parameters are the following:

- RtolEQ: relative error tolerance, either a scalar or an array as long as 'y'. See details in help for lsoda function. Default value is 1e-06.
- AtolEQ: absolute error tolerance, either a scalar or an array as long as 'y'. See details in help for lsoda function*. Default value is 1e-06
- Hmax: an optional maximum value of the integration stepsize. A maximum value may be necessary for non-autonomous models (with external inputs), otherwise the simulation possibly ignores short external events. Default value is Inf.

*Copied from help for lsoda:

"The input parameters 'rtol', and 'atol' determine the error control performed by the solver. The solver will control the vector *e* of estimated local errors in *y*, according to an inequality of the form $\max\text{-norm of } (*e*/*ewt*) \leq 1$, where *ewt* is a vector of positive error weights. The values of 'rtol' and 'atol' should all be non-negative. The form of *ewt* is:

$$*RtolEQ* * abs(*y*) + *AtolEQ*$$

where multiplication of two vectors is element-by-element. If the request for precision exceeds the capabilities of the machine, the Fortran subroutine lsoda will return an error code; under some circumstances, the R function 'lsoda' will attempt a reasonable reduction of precision in order to get an answer. It will write a warning if it does so."

5.4 Design tab

In this tab, the user specifies the characteristics of the population design to be evaluated or optimised.

In the Fisher Information Matrix section, the type of Fisher information matrix to be evaluated or optimised can be selected. Three possible Fisher information matrices are implemented in PFIM Interface 4.0.: Population

(also available in previous versions of PFIM Interface), Individual and Bayesian.

In the Dose regimen section, the user specifies if the dose is the same or not for all the involved groups in the population design. If the dose is the same, its value should be specified; if not, the values for each group can be defined (Figure 20).

NB: if the model used is a user-defined model in which the dose was defined, the default value of a dose of 1 should be kept here.

NB2: for models of the library after infusion, total dose should be given and the rate of infusion will be computed using the needed variable T_{inf}.

In the initial population design section, the user enters, for each group, the corresponding elementary designs. The value for the number of groups in the population design is then computed automatically.

The user specifies also if the subjects in each elementary design are given as numbers or as proportions and enter the values.

Figure 20 shows an example of Design tab for a single response model, for which the population design is composed of 2 groups of 30 and 90 subjects respectively, both with dose of 100 and 200, respectively, with the sampling times (0.5, 1, 4, 12) and (0.5, 2, 6) respectively.

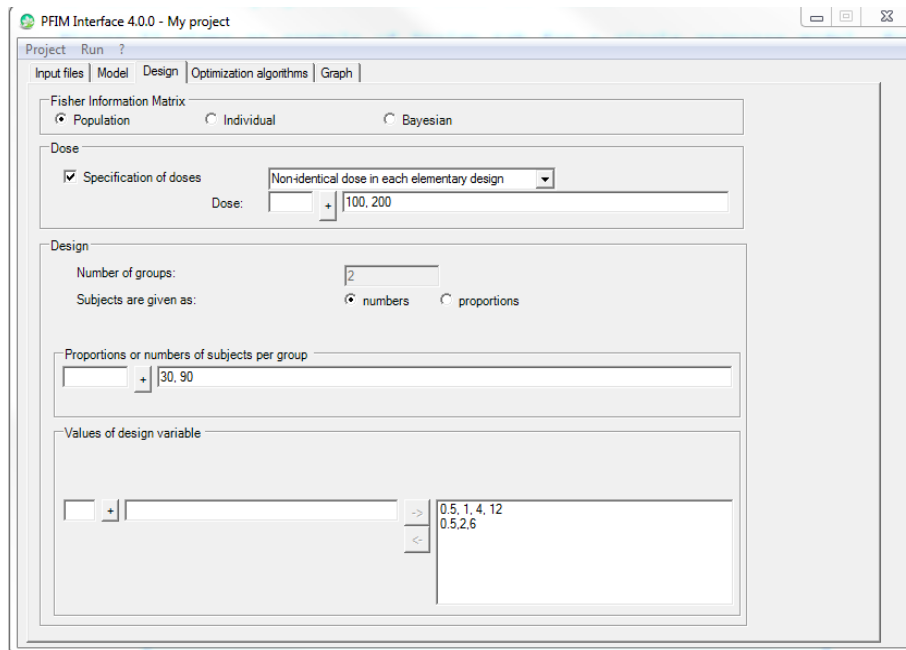


Figure 20: Design tab example for a single response model

Figure 21 provides with an example of Design tab for a two response model with the same sampling times for both responses. Indeed the button "identical design for all responses" is selected.

The population design is composed of one group of 32 subjects with a dose of 100 and with the same sampling times for both responses (0.5, 1, 4, 12).

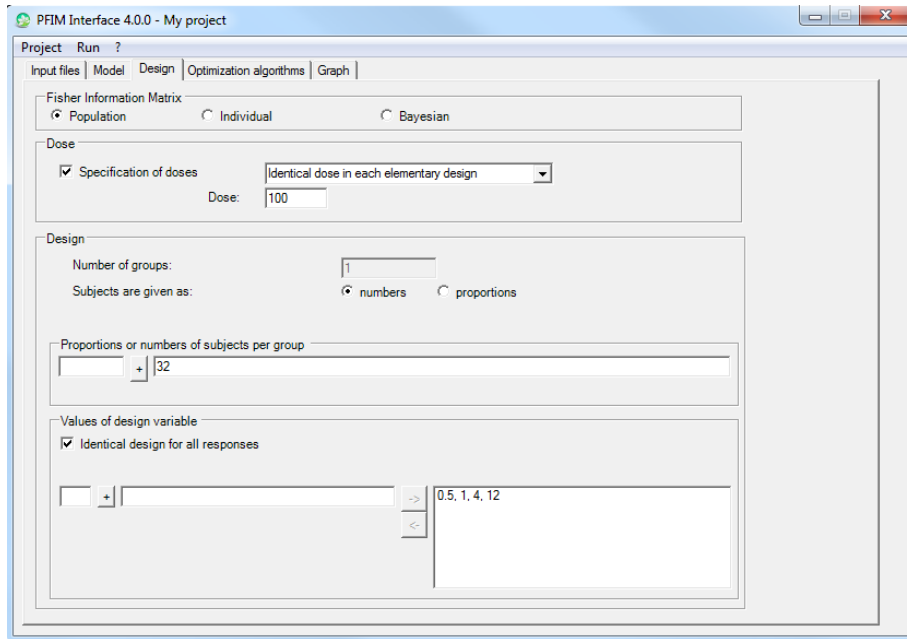


Figure 21: Design tab example for a two response model with the same sampling times for both responses

Figure 22 and Figure 23 give an example of Design tab for a two response model with different sampling times for each response. The button "identical design for all responses" has to be unselected.

The population is composed of one group of 32 subjects with a dose of 100 and with the sampling times for the first response (0.5, 1, 4, 12) (Figure 22) and for the second response: (0, 0.5, 12, 48, 120) (Figure 23).

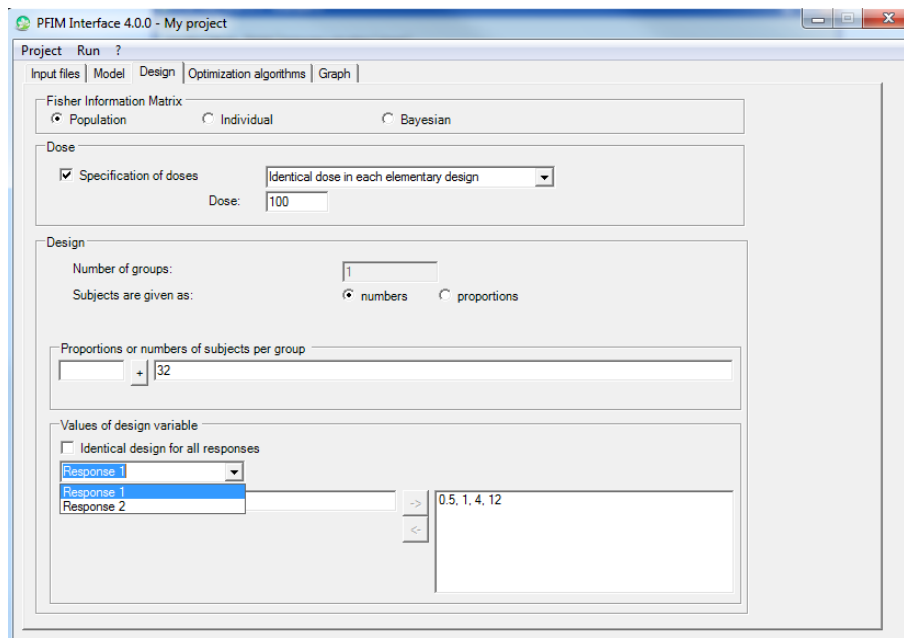


Figure 22: Design tab example for a two response model with different sampling times for each response: choice of the design for the first response

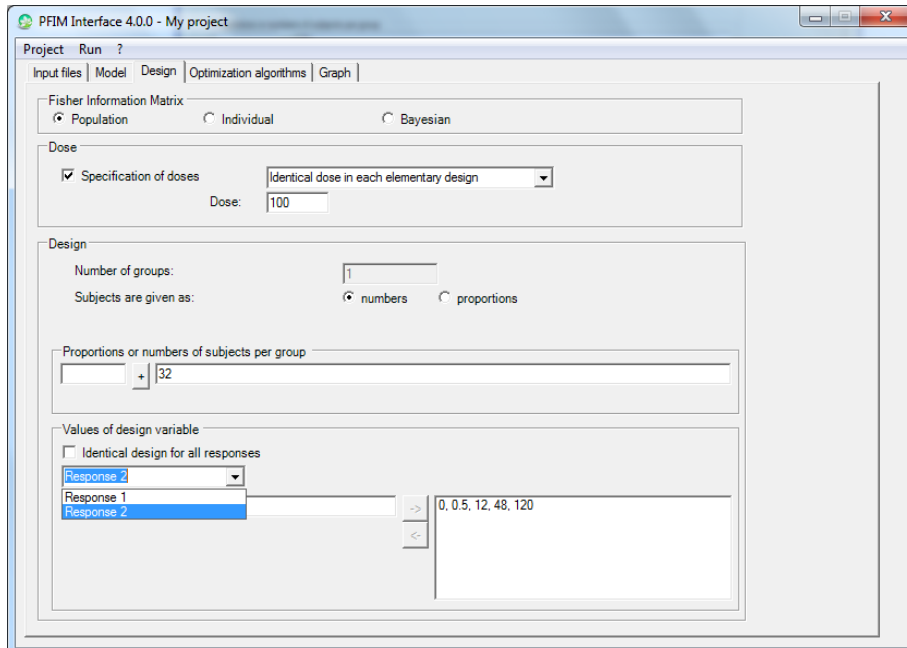


Figure 23: Design tab example for a two response model with different sampling times for each response: choice of the design for the second response

In all these examples (Figure 20 - Figure 23) the requested Fisher information matrix to be evaluated or optimized is the Population one.

5.5 Design evaluation step tab

At this step, evaluation of the population design entered in the "Design tab" can be performed by clicking on the 'Run' button on the windows toolbar, following with a click on the 'Evaluation' (see Figure 24). See Section 6 for the output. Evaluation can be performed either using the expression of the block diagonal Fisher information matrix (Figure 24) or the full expression of the Fisher information matrix (Figure 25).

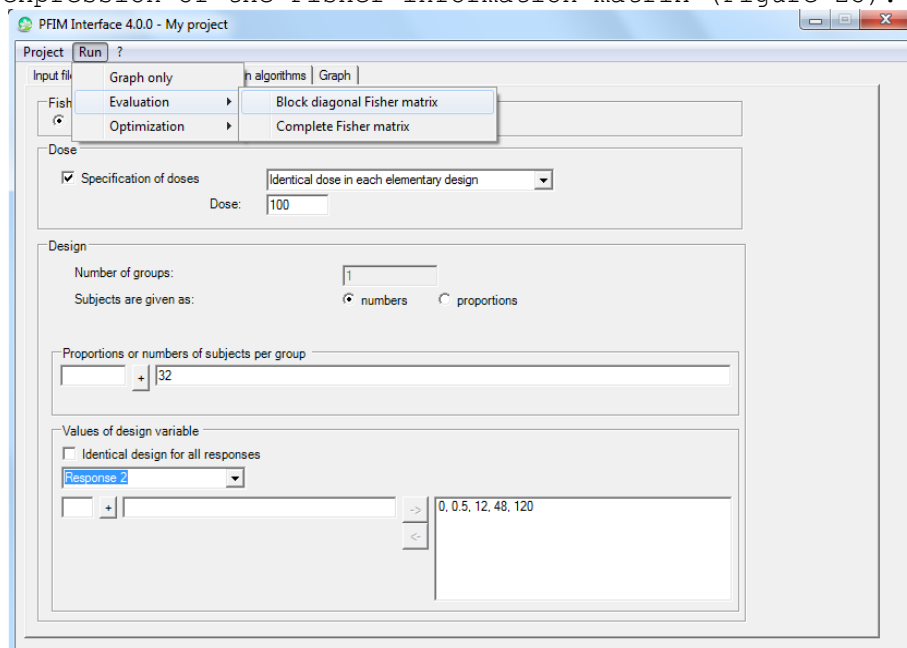


Figure 24: How to perform evaluation with the "run" button using the block diagonal expression of the Fisher information matrix

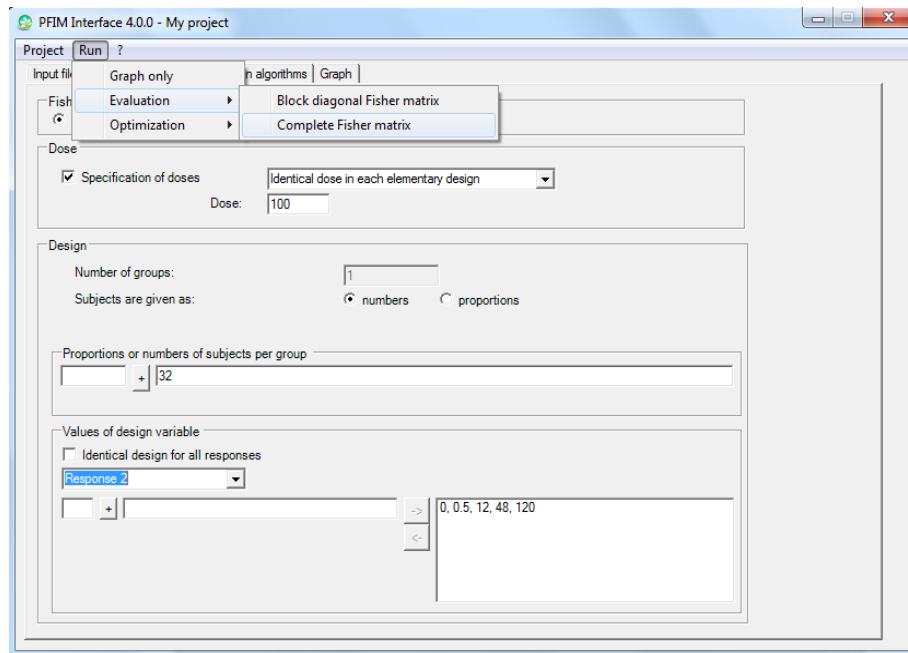


Figure 25: How to perform evaluation with the "run" button using the full expression of the Fisher information matrix

5.6 Optimisation algorithms tab

This tab must be filled in to optimise a design. The optimisation can be performed with either the Simplex or the Fedorov-Wynn algorithm. Corresponding section of the chosen algorithm must be filled in. In the case of multiple response models, the selection of the button "**identical design for all responses**" on the Design tab allows to optimise the design with the same sampling times for all responses.

5.6.1 Simplex algorithm

The first option to be chosen is whether optimising or not the proportions or number of subjects. Then, a value for the minimum delay between two sampling times can be entered. By default, this delay is set to 0. It is then possible to specify whether the iteration step should be printed or not in the R command window.

If different optimal sampling times for each response are required, the allowed intervals of sampling times for the optimisation must be provided for each response (Figure 26).

Parameters for the Simplex algorithm are set by default but they can be changed by the user. These are:

- the parameter for the initial simplex building gives the percentage of change from the initial design to create the initial vertices of the Simplex algorithm. Default is 20%
- the maximum iteration number of the Simplex algorithm which is set by default to 5000
- the relative convergence criterion of the Simplex algorithm set by default to $1e-6$.

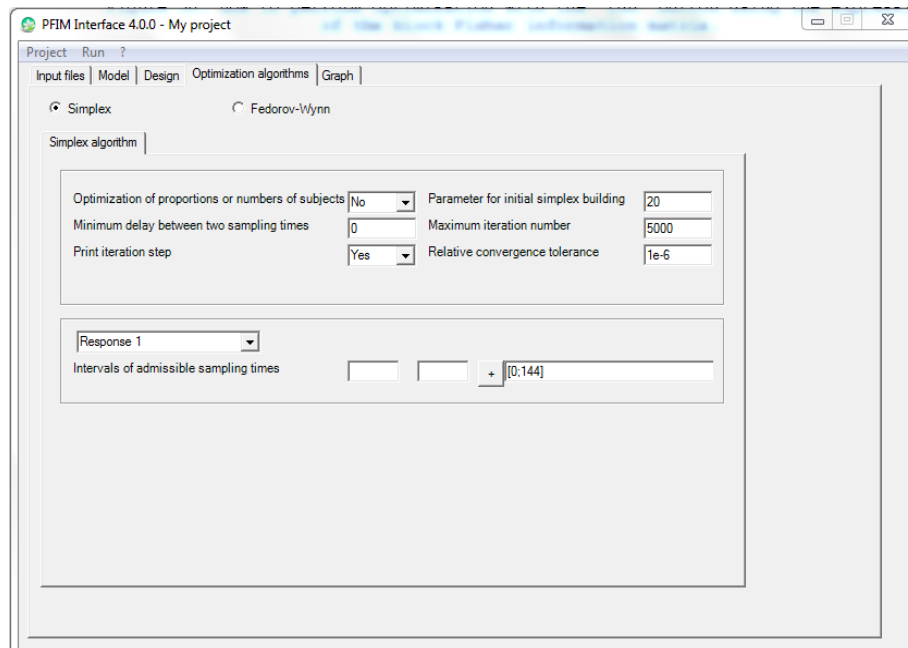


Figure 26: Optimisation algorithms tab for the Simplex algorithm

5.6.2 Fedorov-Wynn algorithm

Allowed sampling times can be specified in the Fedorov-Wynn algorithm corresponding tab. Several set of allowed sampling times may be provided, each set being called the sampling interval.

Then, the user must specify the number of sampling times to be taken from each sampling interval. More flexibility can be given by specifying for each sampling interval several numbers: the Fedorov-Wynn algorithm will then select the best ones.

The number of sampling intervals and the total number of samples per subject are then computed automatically.

The Federov-Wynn algorithm always optimises the number of groups and the proportions of subjects per group.

Please note that the initial population design given in the "Design" tab must correspond to the constraint specified in this Fedorov-Wynn algorithm: the sampling times must be included in the sampling interval and the number of sampling times from each interval must be concordant with the allowed numbers.

An example of the Fedorov-Wynn section is given in Figure 27 for a single response model. In this example, two sampling intervals are specified with the allowed sampling times (0.5, 1, 2, 3, 4, 6, 8, 10, 12) and (48.5, 49, 50, 51, 52, 56, 58, 60) respectively. The user allowed optimization of a design with either two or three sampling times in each interval. The minimal total number of allowed sampling times per subject is then 4 and the maximum 6.

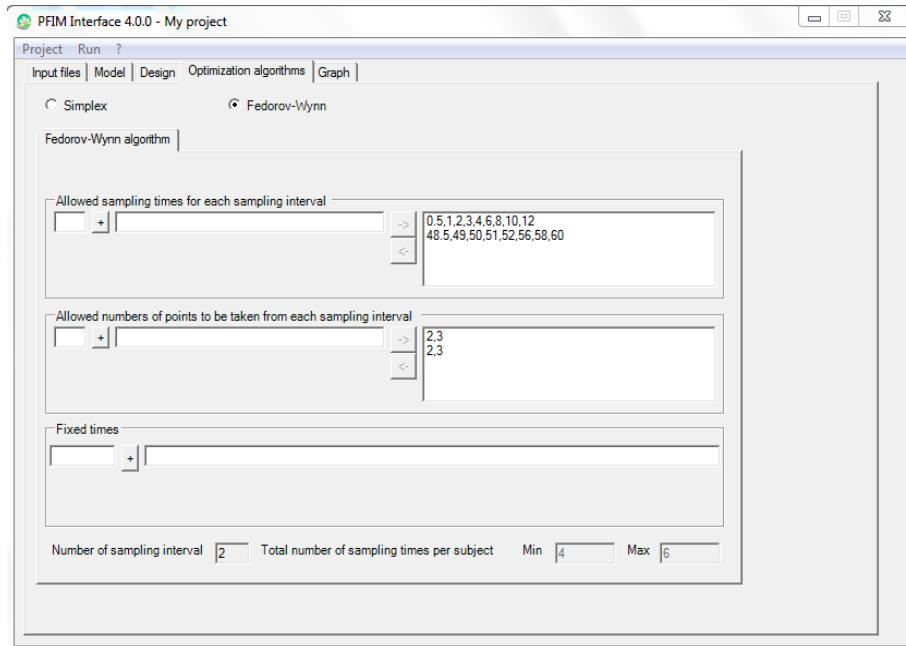


Figure 27: Example of the Optimisation algorithms tab for the Fedorov-Wynn algorithm for a single response model

An example of the Fedorov-Wynn section is given in Figure 28 and Figure 29 for a two response model. In this example, one sampling interval is specified with the allowed sampling times (0, 0.5, 1, 6, 12, 24, 48, 72, 96, 120, 144) for the first response (see Figure 28) and for the second response (0, 24, 36, 48, 72, 96, 120, 144) (see Figure 29). The user allowed optimization of a design with five sampling times in this interval for each response. The minimal total number of allowed sampling times per subject is then 4 and the maximum 4.

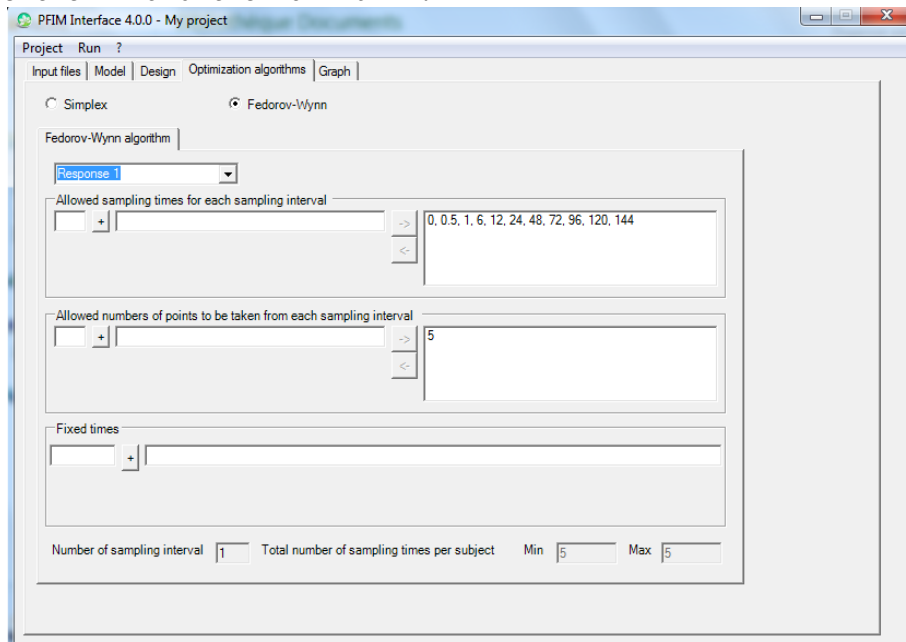


Figure 28: Example of the Optimisation algorithms tab for the Fedorov-Wynn algorithm for a two response model: choice for the first response

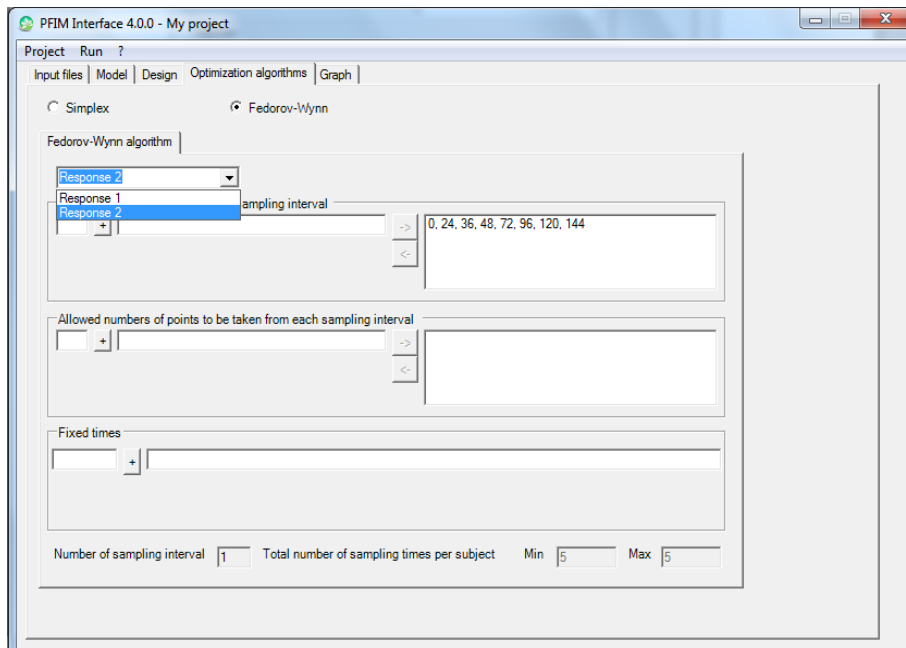


Figure 29: Example of the Optimisation algorithms tab for the Fedorov-Wynn algorithm for a two response model: choice for the second response

5.7 Design optimisation step tab

At this step, optimisation can be performed by clicking on the 'Run' button on the windows toolbar and then choosing 'Optimization'.

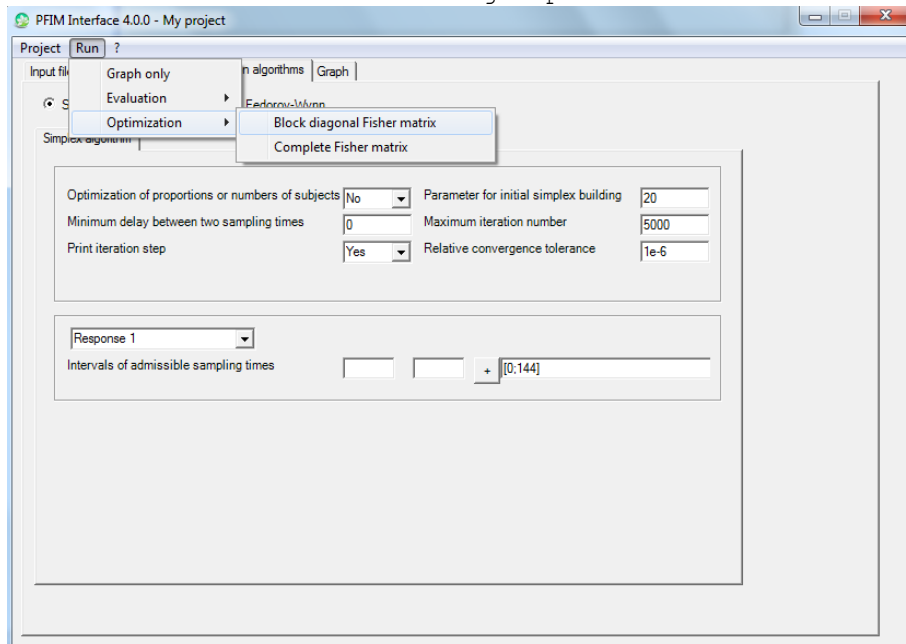


Figure 30: How to perform optimisation with the 'run' button using the expression of the block Fisher information matrix

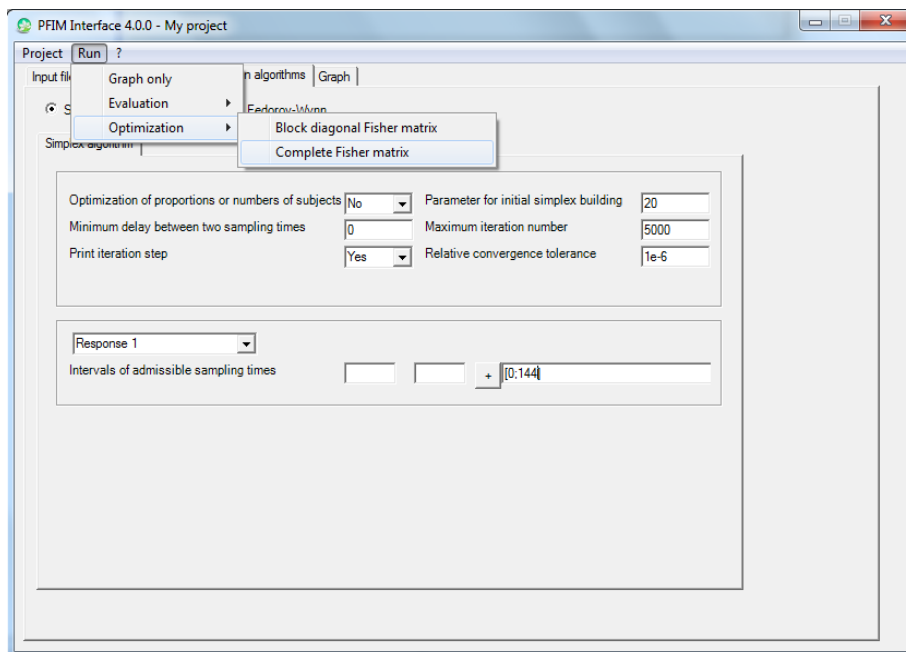


Figure 31: How to perform optimisation with the 'run' button using the expression of the complete Fisher information matrix

5.8 Graph tab

Graph of either the model or sensitivity function or both can be requested by selecting corresponding button(s) (Figure 32). Intervals for the times (X axis) have to be specified. Intervals for the Y axis are set by defaults to the range of the concentrations but can be changed.

It is possible to plot a graph with a log scale for X and/or Y axis by selecting respectively the 'Log X axis' button and/or the 'Log Y axis' button.

Format of the graph can be either jpeg or pdf.

Regarding multiple response models, the user can choose to have a graph for each response with different scales and different labels. To do that, the user has to unselect the button entitled "Identical lower and upper sampling times for each response".

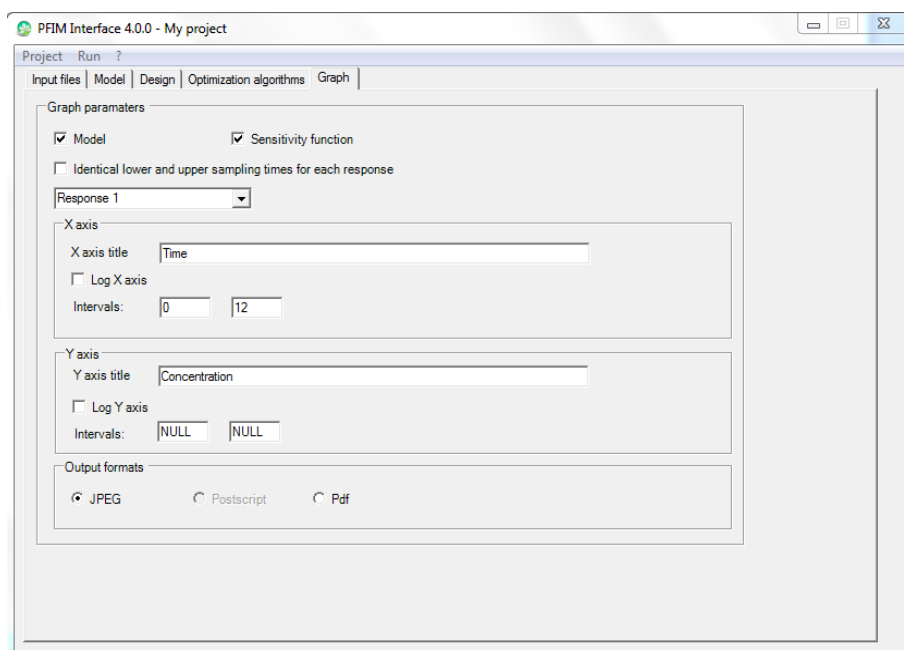


Figure 32: Graph tab

Please note that graphs of the model or sensitivity function may also be obtained before performing any design evaluation or optimisation by clicking the 'Run' button on the windows toolbar and then choosing "Graph only" (Figure 33)

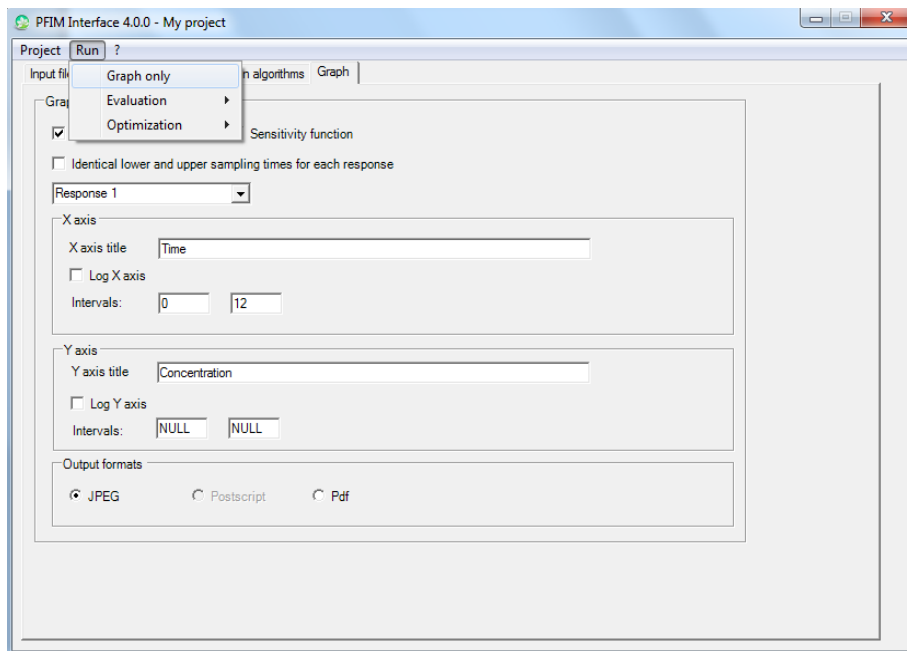


Figure 33: Run graph only

6. Results

PFIM Interface 4.0 opens an R command window to run the evaluation or the optimisation (Figure 34). At the end, an output file (named by default Stdout.r or with the name specified in the input files tab) is created in the directory of the project. It can be viewed by clicking on the button "View output file" in the output R command window.

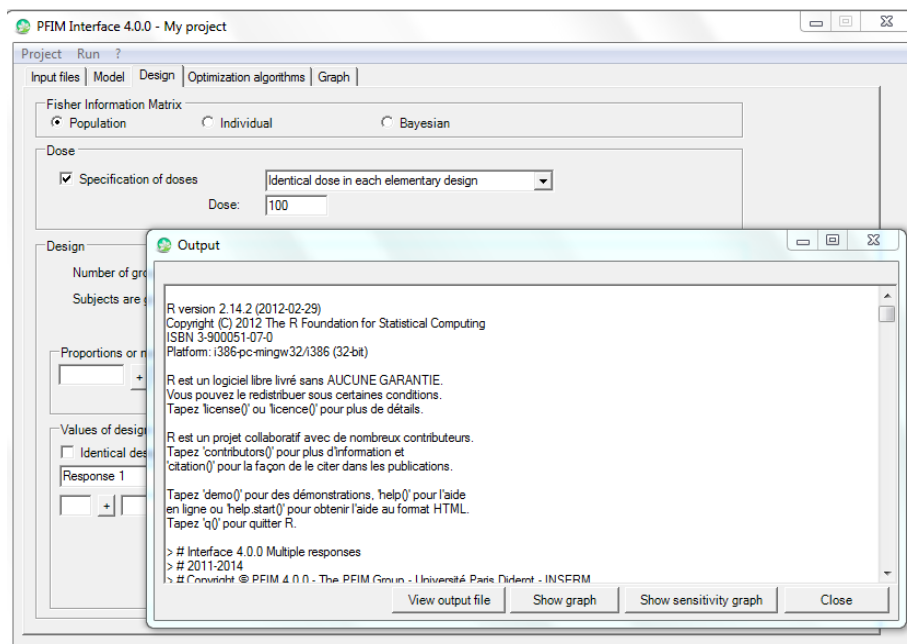


Figure 34: Tab of the results with buttons to see the output file and the graph

Regarding optimisation step with the Fedorov-Wynn algorithm, in addition to the R command windows PFIM Interface 4.0 opens a warning window (Figure 35)

but as it does not prevent the PFIM Interface to run it can just be ignored. It will be removed for the next version of PFIM.

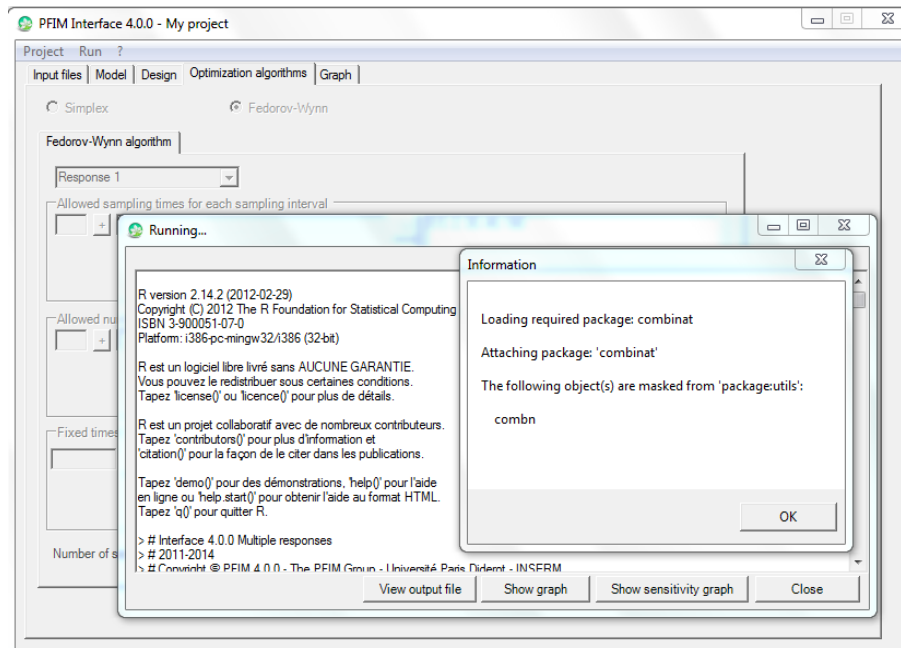


Figure 35: Tab of the results with buttons to see the output file and the graph and a warning window

If any graph was requested, a file called 'Rplots' is also created in the project directory and can be viewed by clicking on the "Show graph" or "Show sensitivity graph" button in the same output R command window. The results are also written in the output file named by default stdout.r. According design evaluation or design optimisation, the following sections are going to describe the different elements of the output file.

6.1 Evaluation output file and objects

6.1.1 Single response model

Figure 36 represents the output file from the design evaluation as in the Example 1 - described in the "Examples" section.

The user can read on Figure 36:

- ① The name of the function used: PFIM Interface 4.0.
- ② The name of the project and the date.
- ③ A summary of the input: model, variance error model, residual between-subject variance model, initial population design, initial numbers or proportions of subjects and doses, initial conditions values, errors tolerances for the solver of differential equations system if used and the expression of the Fisher information matrix used (block or full).
- ④ The population Fisher information matrix, a dim*dim symmetric matrix where dim is the total number of population parameters to be estimated.
- ⑤ The value of each population parameter with the expected standard error on each parameter and the corresponding coefficient of variation.

6 The value of the determinant of the Fisher information matrix and the value of the criterion (determinant^(1/dim)) where dim is the total number of population parameters.

7 The eigenvalues of the Fisher information matrix and the correlation matrix.

PFIM Interface 4.0

Project: Example 1.1_ExpressionMod_1.2.1

Date: Tue May 12 08:16:29 2015

***** INPUT SUMMARY *****

Analytical function models :

dose/V * ka/(ka - k) * (exp(-k * t) - exp(-ka * t))

Design:

Sample times for response: A

times subjects doses
1 c(0.33, 1.5, 5, 12) 200 100

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Computation of the Population Fisher information matrix: option = 1

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]	[,6]	[,7]
[1,]	38.467601	82.866694	-3.770151	0.000000	0.000000	0.000000	0.000000
[2,]	82.8666939	8310.88576	77.977971	0.000000	0.000000	0.000000	0.000000
[3,]	-3.770151	77.97797	4.938229	0.000000	0.000000	0.000000	0.000000
[4,]	0.000000	0.000000	0.000000	59.190253	4.291831	31.98158	28.15946
[5,]	0.000000	0.000000	0.000000	4.291831	674.519747	213.76982	193.67427
[6,]	0.000000	0.000000	0.000000	31.981582	213.769824	3086.36693	295.74230
[7,]	0.000000	0.000000	0.000000	28.159462	193.674273	295.74230	1208.60606
[8,]	0.000000	0.000000	0.000000	85.786235	226.638153	1167.39328	1544.00257

[,8]

[1,]	0.000000
[2,]	0.000000
[3,]	0.000000
[4,]	85.78624
[5,]	226.63815
[6,]	1167.39328
[7,]	1544.00257
[8,]	4118.40001

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
ka	2.00	0.17480765	8.740383 %
k	0.25	0.01239415	4.957658 %
V	15.00	0.52291110	3.486074 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
ka	1.00	0.13203572	13.20357 %
k	0.25	0.03977275	15.90910 %
V	0.10	0.01933249	19.33249 %

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.interA	0.50	0.04077340	8.154681 %
sig.slopeA	0.15	0.02293716	15.291443 %

1

2

3

4

5

```

***** DETERMINANT *****
2.930397e+20
***** CRITERION *****
361.7144
***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min      2490.782416      3.598669
max      8312.446928      702.614507
max/min   3.337283      195.242884
***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]      [,4]      [,5]      [,6]
[1,] 1.0000000 -0.2836926  0.3614018  0.0000000000  0.0000000000  0.0000000000
[2,] -0.2836926  1.0000000 -0.4466787  0.0000000000  0.0000000000  0.0000000000
[3,]  0.3614018 -0.4466787  1.0000000  0.0000000000  0.0000000000  0.0000000000
[4,]  0.0000000  0.0000000  0.0000000  1.0000000000  0.0008945383 -0.01707708
[5,]  0.0000000  0.0000000  0.0000000  0.0008945383  1.0000000000 -0.13147266
[6,]  0.0000000  0.0000000  0.0000000 -0.0170770841 -0.1314726648  1.0000000000
[7,]  0.0000000  0.0000000  0.0000000  0.0186040109 -0.1824110325  0.12863647
[8,]  0.0000000  0.0000000  0.0000000 -0.1283097535  0.0574454432 -0.31004589

      [,7]      [,8]
[1,] 0.00000000  0.00000000
[2,] 0.00000000  0.00000000
[3,] 0.00000000  0.00000000
[4,] 0.01860401 -0.12830975
[5,] -0.18241103  0.05744544
[6,]  0.12863647 -0.31004589
[7,]  1.00000000 -0.68199073
[8,] -0.68199073  1.00000000

```

6

7

Figure 36: Example of design evaluation output file for a single response model

Moreover, several R objects are returned in the R command window:

```

dose
prot: design evaluated for each response
subjects: number of subjects for each group
mfisher: the population Fisher information matrix
determinant: the determinant of the population Fisher information matrix
crit: the value of the criterion
p: the vector
se: the vector of the expected standard errors for each parameter
cv: the corresponding coefficient of variation, expressed in percent.
EigenValues: the eigenvalues of the Fisher information matrix
corr.matrix: the correlation matrix

```

6.1.2 Multiple response model

Figure 37 represents the output file from the design evaluation as in the Example 2 described in the "Examples" section.

The user can read on the Figure 37:

- ① The name of the function used: PFIM Interface 4.0.
- ② The name of the project and the date.
- ③ A summary of the input: model(s), sampling times in the elementary designs for each model(s), doses or initial conditions and subjects corresponding to those designs, residual variance error model for each model(s), residual between-subject variance model, initial population design, errors tolerances for the solver of differential equations system if used and the expression of the Fisher information used (block or full).

The figure shows a two responses model (differential equations form) with a group described by 5 sampling times for both first and second responses for 100 subjects. The dose is equal to 100.

④ The population Fisher information matrix, a dim*dim symmetric matrix where dim is the total number of population parameters to be estimated.

⑤ The value of each population parameter with the expected standard error on each parameter and the corresponding coefficient of variation.

⑥ The value of the determinant of the Fisher information matrix and the value of the criterion (determinant^(1/dim)) where dim is the total number of population parameters.

⑦ The eigenvalues of the Fisher information matrix and the correlation matrix.

PFIM Interface 4.0

Project: Example 2

Date: Tue May 12 08:24:26 2015

①

} ②

***** INPUT SUMMARY *****

Differential Equations form of the model:

```
function (t, y, p)
{
  V <- p[1]
  Vm <- p[2]
  km <- p[3]
  Alin <- p[4]
  pk <- y[1:1]
  pd <- y[2:2]
  conc <- y[1]
  if (t <= 1) {
    dpk1 <- (100/(1 * V)) + (-Vm) * pk[1]/(km + pk[1])
  }
  else {
    dpk1 <- (-Vm) * pk[1]/(km + pk[1])
  }
  dpd1 <- 0
  pdIm <- Alin * conc
  return(list(c(dpk1, dpd1), c(pk[1], pdIm)))
}
```

Design:

Sample times for response: A
 times subjects
 1 c(0.5, 2, 30, 49, 180) 100

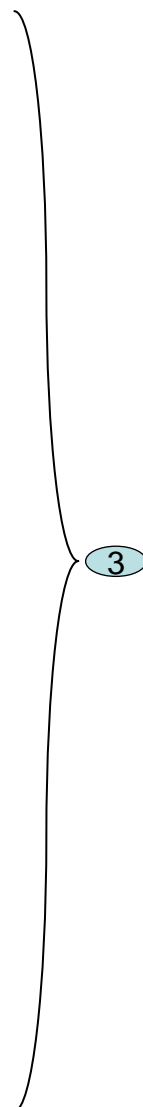
Sample times for response: B
 times subjects
 1 c(0.5, 2, 14, 110, 150) 100

Initial Conditions at time 0 :
 0 0

Random effect model: Trand = 2

Variance error model response A : (0 + 0.2 *f)^2
 Variance error model response B : (0.1 + 0 *f)^2

Error tolerance for solving differential equations system:
 RtolEQ = 1e-08 , AtolEQ = 1e-08 , Hmax = Inf
 Computation of the Population Fisher information matrix: option = 1
 FIM saved in FIM.txt



***** FISHER INFORMATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	2.6770341	9.444202	-57.39048	-0.7254162	0.000000e+00
[2,]	9.4442023	50919.213161	7982.73905	657.4076389	0.000000e+00
[3,]	-57.3904833	7982.739050	33057.82792	-6714.1078293	0.000000e+00
[4,]	-0.7254162	657.407639	-6714.10783	38772.8151987	0.000000e+00
[5,]	0.0000000	0.000000	0.00000	0.0000000	7.938111e+02
[6,]	0.0000000	0.000000	0.00000	0.0000000	4.463216e-01
[7,]	0.0000000	0.000000	0.00000	0.0000000	3.916193e-03
[8,]	0.0000000	0.000000	0.00000	0.0000000	2.546794e+00
[9,]	0.0000000	0.000000	0.00000	0.0000000	8.097541e+00

	[,6]	[,7]	[,8]	[,9]
[1,]	0.0000000	0.000000e+00	0.000000e+00	0.000000
[2,]	0.0000000	0.000000e+00	0.000000e+00	0.000000
[3,]	0.0000000	0.000000e+00	0.000000e+00	0.000000
[4,]	0.0000000	0.000000e+00	0.000000e+00	0.000000
[5,]	0.4463216	3.916193e-03	2.546794e+00	8.097541
[6,]	586.1230244	1.453005e-01	9.449987e+01	301.234913
[7,]	0.1453005	7.516656e+02	8.332548e-01	116.540931
[8,]	94.4998735	8.332548e-01	1.725786e+04	1714.876645
[9,]	301.2349133	1.165409e+02	1.714877e+03	64572.948196

4

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

	Beta	StdError	RSE
V	12.200	0.624822332	5.121495 %
Vm	0.082	0.004534793	5.530235 %
km	0.370	0.005841524	1.578790 %
Alin	0.100	0.005182493	5.182493 %

----- Variance of Inter-Subject Random Effects -----

	omega2	StdError	RSE
V	0.25	0.03549293	14.19717 %
Vm	0.25	0.04137028	16.54811 %
Alin	0.25	0.03647948	14.59179 %

5

----- Standard deviation of residual error -----

	Sigma	StdError	RSE
sig.slopeA	0.2	0.007625037	3.812519 %
sig.interB	0.1	0.003945487	3.945487 %

***** DETERMINANT *****

5.994606e+31

***** CRITERION *****

3395.176

6

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

	FixedEffects	VarianceComponents
min	26521.376944	2.561446
max	64636.681018	17196.209993
max/min	2.437154	6713.478513

***** CORRELATION MATRIX *****

	[,1]	[,2]	[,3]	[,4]	[,5]
[1,]	1.00000000	-0.06785010	0.2062542	0.04352023	0.000000e+00
[2,]	-0.06785010	1.00000000	-0.2101514	-0.05600795	0.000000e+00
[3,]	0.20625422	-0.21015142	1.00000000	0.19876898	0.000000e+00
[4,]	0.04352023	-0.05600795	0.1987690	1.00000000	0.000000e+00
[5,]	0.00000000	0.00000000	0.00000000	0.00000000	1.000000e+00
[6,]	0.00000000	0.00000000	0.00000000	0.00000000	-5.826757e-04
[7,]	0.00000000	0.00000000	0.00000000	0.00000000	1.311462e-05
[8,]	0.00000000	0.00000000	0.00000000	0.00000000	-6.146867e-04
[9,]	0.00000000	0.00000000	0.00000000	0.00000000	-1.068259e-03

	[,6]	[,7]	[,8]	[,9]
[1,]	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[2,]	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[3,]	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[4,]	0.0000000000	0.000000e+00	0.0000000000	0.0000000000
[5,]	-0.0005826757	1.311462e-05	-0.0006146867	-0.001068259
[6,]	1.0000000000	5.840399e-04	-0.0272653545	-0.047524772
[7,]	0.0005840399	1.000000e+00	0.0006127203	-0.016746938
[8,]	-0.0272653545	6.127203e-04	1.0000000000	-0.050000216
[9,]	-0.0475247719	-1.674694e-02	-0.0500002164	1.0000000000

7

Figure 37: Example of design evaluation output file for a two response model

Moreover, the PFIM() function returns the following R objects:

dose
prot: design evaluated for each response
subjects: number of subjects for each group
mfisher: the population Fisher information matrix
determinant: the determinant of the population Fisher information matrix
crit: the value of the criterion
p: the vector
se: the vector of the expected standard errors for each parameter
cv: the corresponding coefficient of variation, expressed in percent.
EigenValues: the eigenvalues of the Fisher information matrix
corr.matrix: the correlation matrix

6.2 Optimisation output file and objects

Figure 38 represents the output file corresponding to the optimal Bayesian design described in the Examples section in the paragraph 1.3.

The user can read on the Figure 38:

- ① The name of the function used: PFIM Interface 4.0
- ② The name of the project and the date.
- ③_a A summary of the input: structural model, between-subject and error variance model, initial design, initial numbers or proportions of subjects and doses, total number of allowed samples, criterion associated to the initial design.
- ③_b Sampling times specifications (according to the algorithm used) within which the optimal samples will be chosen and error tolerances for the solver of differential equations system if used.
- ④ The optimised design and the associated criterion.
For the simplex algorithm, the number of iterations performed and the number of function evaluations, the status of the convergence (false or achieved) are reported
For the Fedorov-Wynn algorithm for optimal population design, the optimal group structure with the proportion of subjects and the equivalence in number are then reported. The best one group protocol is also always reported with associated criterion.
When optimising a Bayesian or an individual design, the resulted design correspond to the best one group protocol.
- ⑤ The population or individual or Bayesian Fisher information matrix, a $\text{dim} \times \text{dim}$ symmetric matrix where dim is the total number of population parameters to be estimated, the number of individual parameters + the number of the error model parameters or only the number of individual parameters respectively. The name of the file where is possibly saved the Fisher information matrix is given.
- ⑥ The value of each parameter with the expected standard error (StdError) and relative standard error (RSE). In case of Bayesian design, the associated shrinkages values are also reported.
- ⑦ The value of the determinant of the Fisher information matrix and the value of the criterion ($\text{determinant}^{(1/\text{dim})}$) where dim is defined in ⑤
- ⑧ The eigenvalues of the Fisher information matrix and the correlation matrix.

PFIM Interface 4.0

Project: Example Optimisation

Date: Thu Jul 31 09:22:17 2014

1

2

***** INPUT SUMMARY *****

Analytical function model:

```
function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}
```

Initial design:

```
Sample times for response: A
          Protocol subjects doses
1 c=(0.33, 1.5, 5, 12)      1  100
```

Total number of samples: 4

Associated criterion value: 3.5272

Identical sampling times for each response: FALSE

Random effect model: Trand = 2

Variance error model response A : (0.5 + 0.15 *f)^2

Optimization step:

```
Sampling windows for the response: A
Window 1 : t= 0.33 1 1.5 3 5 8 12
          Nb of sampling points to be taken in this window, n[ 1 ]= 4
Maximum total number of points in one elementary protocol : 4
Minimum total number of points in one elementary protocol : 4
```

BEST ONE GROUP PROTOCOL:

```
Sample times for response: A
          times freq Subjects doses
1 c(0.33, 1.5, 5, 8)      1      1  100
```

Associated criterion: 3.8066

3a

3b

4

```

Computation of the Bayesian Fisher information matrix

FIM saved in FIM.txt

***** FISHER INFORMATION MATRIX *****

      [,1]      [,2]      [,3]
[1,] 1.590507  2.096455 -0.2426030
[2,] 2.096455 354.843266  4.4964361
[3,] -0.242603 4.496436  0.2013882

***** EXPECTED STANDARD ERRORS *****

----- Fixed Effects Parameters -----

      Beta StdError      RSE  Shrinkage
ka  2.00 0.9638509 48.19255 % 23.22522 %
k   0.25 0.0688475 27.53900 % 30.33586 %
V   15.00 3.1862487 21.24166 % 45.12080 %

***** DETERMINANT *****

55.15913

***** CRITERION *****

3.806617

***** EIGENVALUES OF THE FISHER INFORMATION MATRIX *****

      FixedEffects VarianceComponents
min      9.552493e-02      NA
max      3.549127e+02      NA
max/min  3.715393e+03      NA

***** CORRELATION MATRIX *****

      [,1]      [,2]      [,3]
[1,] 1.0000000 -0.4133690  0.5638373
[2,] -0.4133690  1.0000000 -0.6330761
[3,]  0.5638373 -0.6330761  1.0000000

```

5

6

7

8

Figure 38: Example of design optimisation output file

Moreover, the PFIM() function returns the following R objects:

- mfisher:** the population or individual or Bayesian Fisher information matrix corresponding to the optimised protocole
- determinant:** the determinant of the Fisher information matrix
- crit:** the value of the criterion
- se:** the vector of the expected standard errors for each parameter
- cv:** the corresponding coefficient of variation, expressed in percent (relative standard error)
- sh:** the shrinkage values for each parameter in case of Bayesian design
- EigenValues:** the eigenvalues of the Fisher information matrix
- corr.matrix:** the correlation matrix

7. Examples

This section contains a series of examples for EVALUATION (first section) and OPTIMISATION (second section) of design in pharmacokinetics (PK) and pharmacodynamics (PD).

We have tried to illustrate all the features of PFIM Interface 4.0, in this choice of examples. In Example_1 of each section we have more specifically illustrated all the new features in this version 4.0 of PFIM Interface. Furthermore, examples available from the previous version of PFIM Interface, version 3.1, were also implemented in PFIM Interface 4.0. All the input, model and output codes used for these examples are available when PFIM is downloaded, in the directory "Examples" stored in Documents in the directory "PFIM Interface 4.0". They are detailed below.

Evaluation

1. Example 1: PK Model

The purpose is to evaluate a design for a one compartment first order absorption PK model with parameters k_a , V and k after single dose administration.

Random effects are exponentially modelled.

	Mean	Fixed mean	Variance
k_a	2	<input type="checkbox"/>	1
k	0.25	<input type="checkbox"/>	0.25
V	15	<input type="checkbox"/>	0.1
σ_{inter}		0.5	
σ_{slope}		0.15	

Please note: as we don't fix here any parameter, the "Fixed mean" column is kept as default that is without any of the squares checked

1.1 Model Files

Four possible and exchangeable ways for defining the model are available in PFIM. These are described in section 3 of this documentation. Below are reported the figures of the 4 possible models (Figure 39-42).

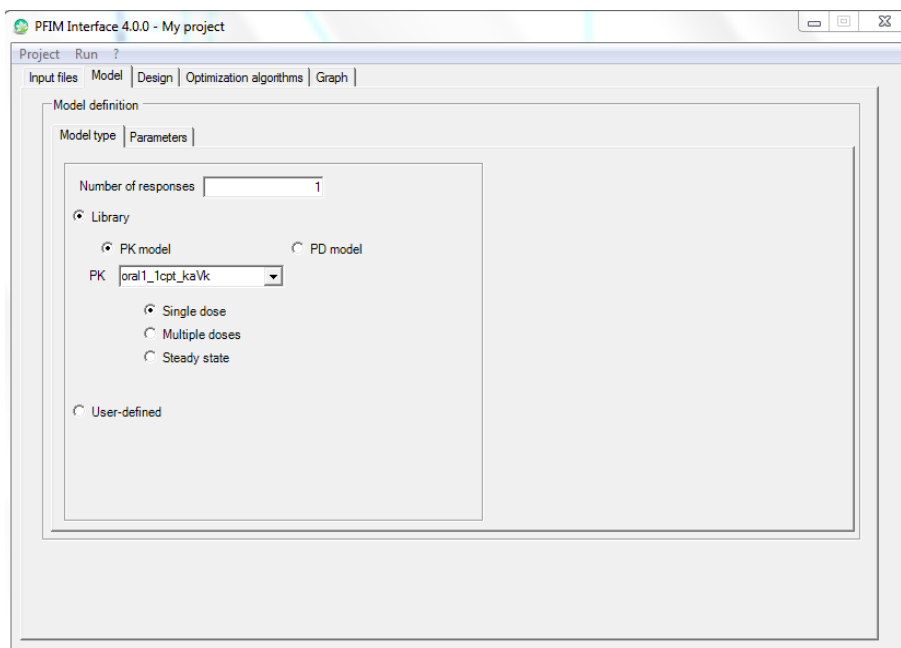


Figure 39 : Model defined from the library

```

model - Bloc-notes
Fichier Edition Format Affichage ?
#$Model definition
#$ka,k,V
#$
formA <-expression(dose/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
form<-c(formA)
tf<-list(Inf)

```

Figure 40 : User defined model through analytical expression

```

model - Bloc-notes
Fichier Edition Format Affichage ?
#$Model definition
#$ ka,k,V
#$
form<-function(t,p,X){
ka<-p[1]
k<-p[2]
V<-p[3]
y<-(X/V*ka/(ka-k)*(exp(-k*t)-exp(-ka*t)))
return(y)
}

```

Figure 41 : User defined model through analytical function

```

model - Bloc-notes
Fichier Edition Format Affichage ?
#$Model definition ODE
#$ka,k,V
#$
formED<-function(t,y,p)
{
ka<-p[1]
k<-p[2]
V <-p[3]
yd1<-ka*y[2]-k*y[1]
yd2<--ka*y[2]
list(c(yd1,yd2),c(y[1]/V))
}

```

Figure 42 : User defined model through differential equations

Graphs pre-evaluation

Graphs of the simulated model and sensitivity function with respect to parameters were obtained by running "Graph only" (see the "Use" section paragraph 6.8 "Graph tab").

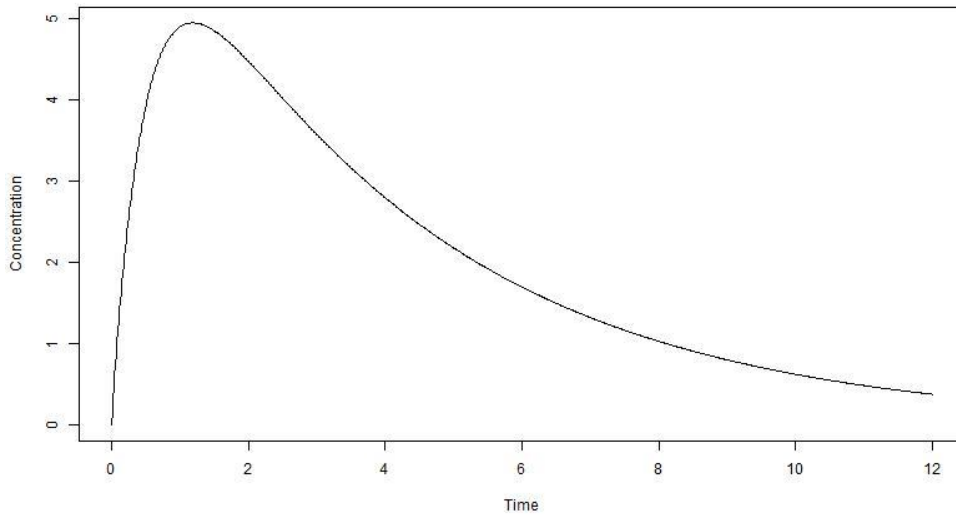


Figure 43 : Simulated model

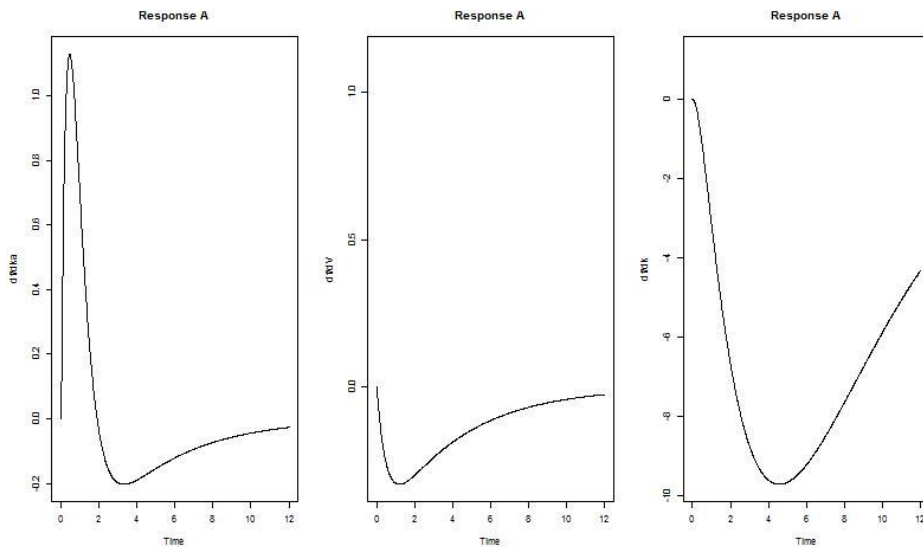


Figure 44 : Sensitivity function with respect to parameters

Population Fisher Information Matrix (P-FIM)

Evaluation of the Population Fisher information matrix (P-FIM) for several designs, all patients have a dose of 100.

1.1.1 One group with Elementary Design ξ_1

200 subjects who have the same elementary design composed of 4 sampling times:

$$\xi_1 = (0.33, 1.5, 5, 12)$$

For this first example, we report the PFIM files (stdin.R, model.r and Stdout.r) for each possible model form (see paragraph 1.1 of this section of Examples). We therefore repeated the example four times, using four input files. Those PFIM files are stored in the directory of Examples, under EVALUATION and EXAMPLE_1 in 4 different folders that are called:

"Exemple 1.1._ModLibrary_1.2.1; 1.1_ExpressionMod_1.2.1; 1.1_ModeOde_1.2.1; 1.1_UserDefMod_1.2.1.

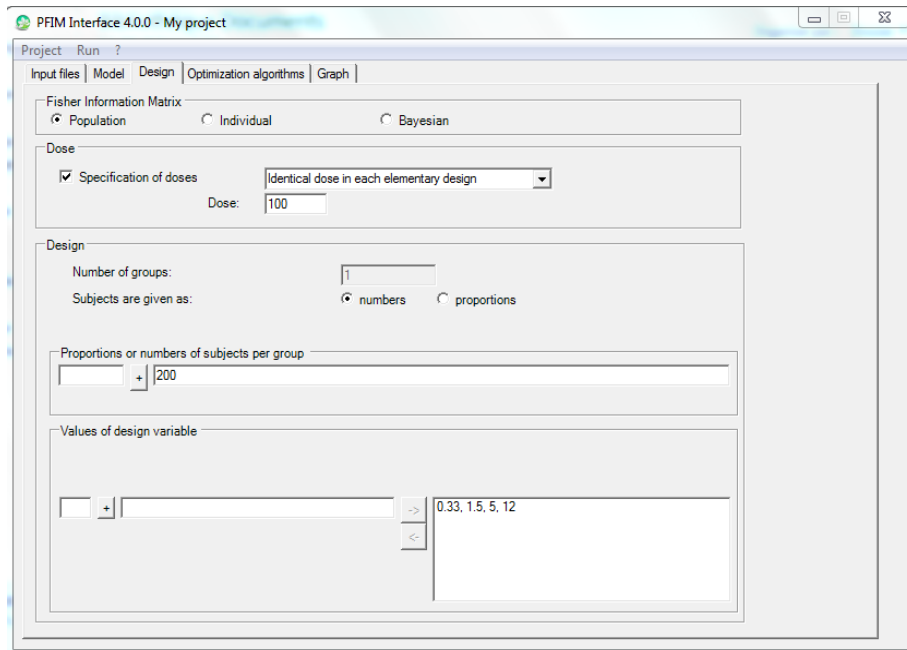


Figure 45 : Design tab for examples of section 1.1.1

1.1.2 One group with Elementary Design ξ_2

200 subjects who have the same elementary design composed of 3 sampling times:

$$\xi_2 = (1, 3, 8)$$

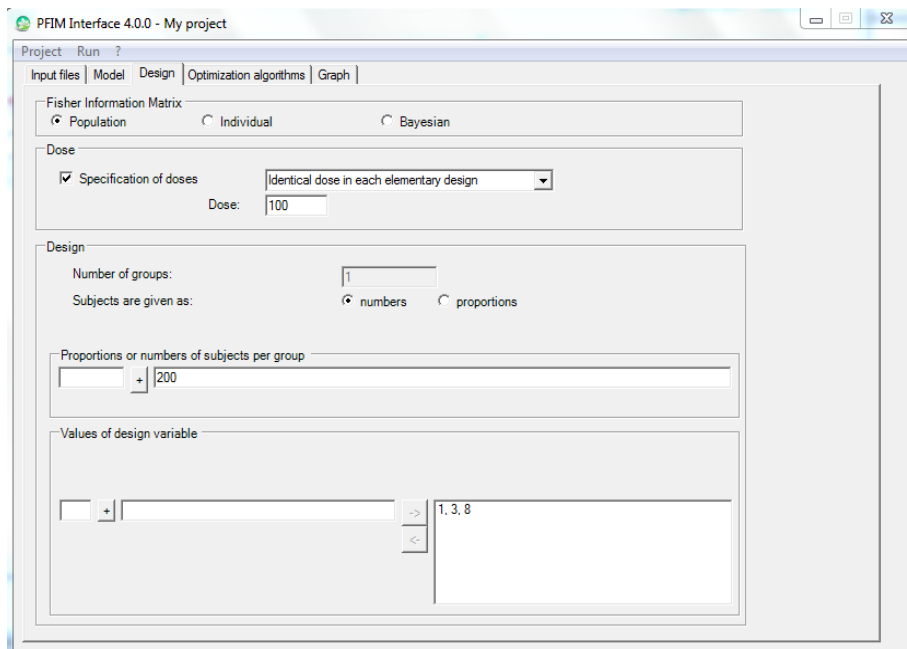


Figure 46: Design tab for example 1.1.2

1.1.3 Two Groups Design

Two group population design of 400 subjects: 200 with elementary designs ξ_1 and 200 with elementary design ξ_2 .

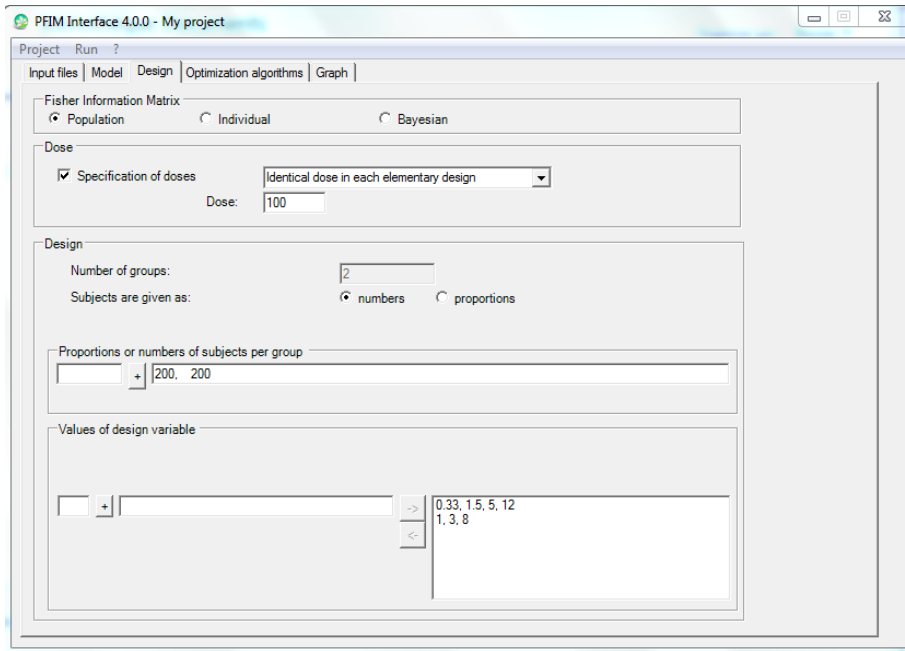


Figure 47: Design tab for example 1.1.3

1.2 Individual Fisher Information Matrix (I-FIM)

Evaluation of the Individual Fisher Information Matrix for the elementary design ξ_1 and then ξ_2 .

1.2.1 Elementary Design ξ_1

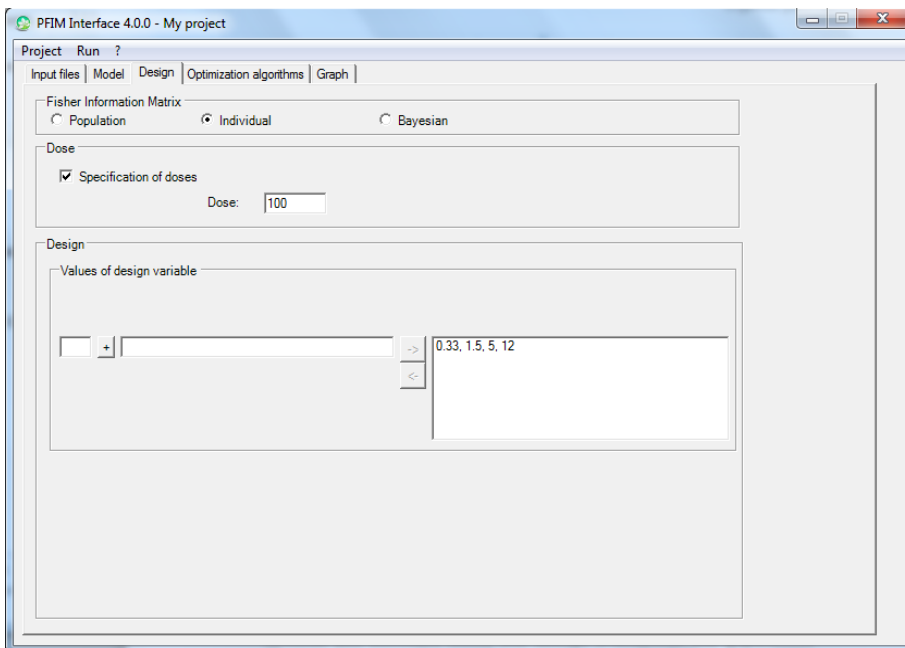


Figure 48 : Design tab for example 1.2.1

1.2.2 Elementary Design ξ_2

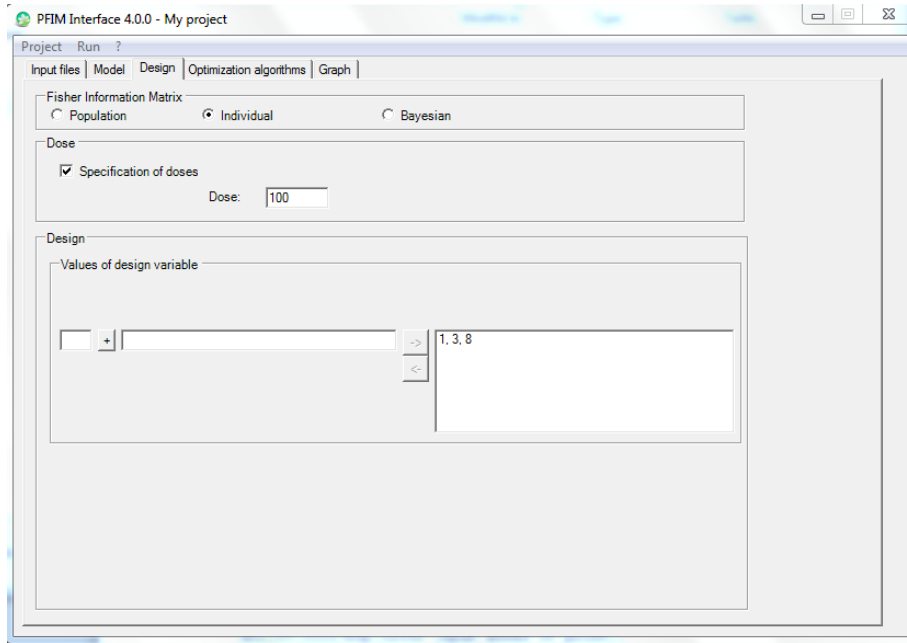


Figure 49: Design tab for example 1.2.2

1.3 Bayesian Fisher Information Matrix (B-FIM)

Evaluation of the Bayesian Fisher Information Matrix for the elementary design ξ_1 and then ξ_2 .

1.3.1 Elementary Design ξ_1

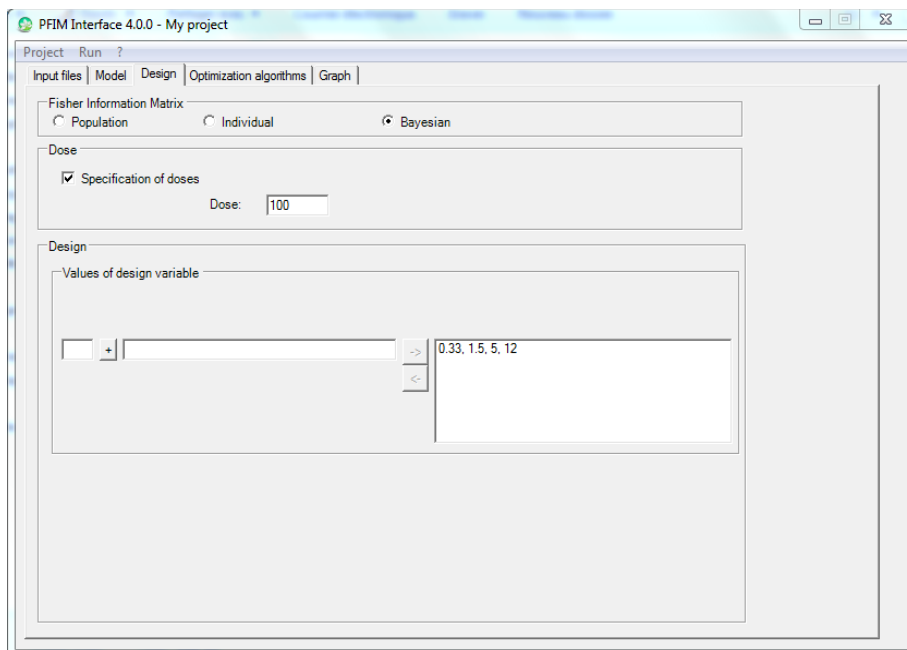


Figure 50: Design tab for example 1.3.1

1.3.2 Elementary Design ξ_2

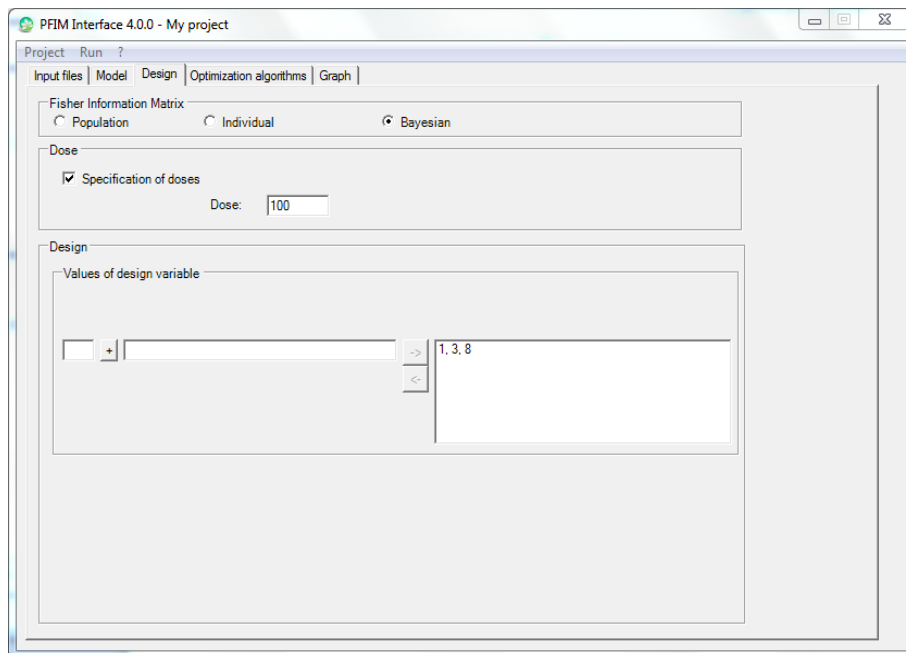


Figure 51: Design tab for example 1.3.2

Comment on results

Comparing examples in 1.2 and 1.3 it is noticeable that the evaluation of B-FIM leads to smaller RSE particularly for the parameter k_a , which in the evaluation of I-FIM is very high, above all for the second elementary design, reaching the value of 138 %.

Comparing the two elementary designs, for both I-FIM and B-FIM, results are overall better with ξ_1 than with ξ_2 , as the variability on k_a is kept lower and the criterion is slightly higher. For B-FIM, shrinkage values are provided and they show that with ξ_1 more information is obtained than with ξ_2 .

1.4 Evaluation of FIM with Fixed Parameter

Another new feature of v4.0 is to assume that a parameter is known (fixed) and not estimated.

Here we evaluated Population, Individual and Bayesian Fisher Information Matrix for the design ξ_1 assuming that the parameter k_a is fixed (and has no variability). Those examples are stored in the directory of EXAMPLE→EVALUATION→EXAMPLE1, and the names of their folder are: 1.4.1, 1.4.2, 1.4.3 for P-FIM, I-FIM or B-FIM evaluation, respectively.

Comment on results

Comparing the output of example 1.4.1 with the one in section 1.1.1, we can see that the standard errors are slightly reduced in this example.

As for example 1.4.1, fixing k_a resulted in slight improvements of the Relative Standard Errors in 1.4.2 with respect to the results in section 1.2.1

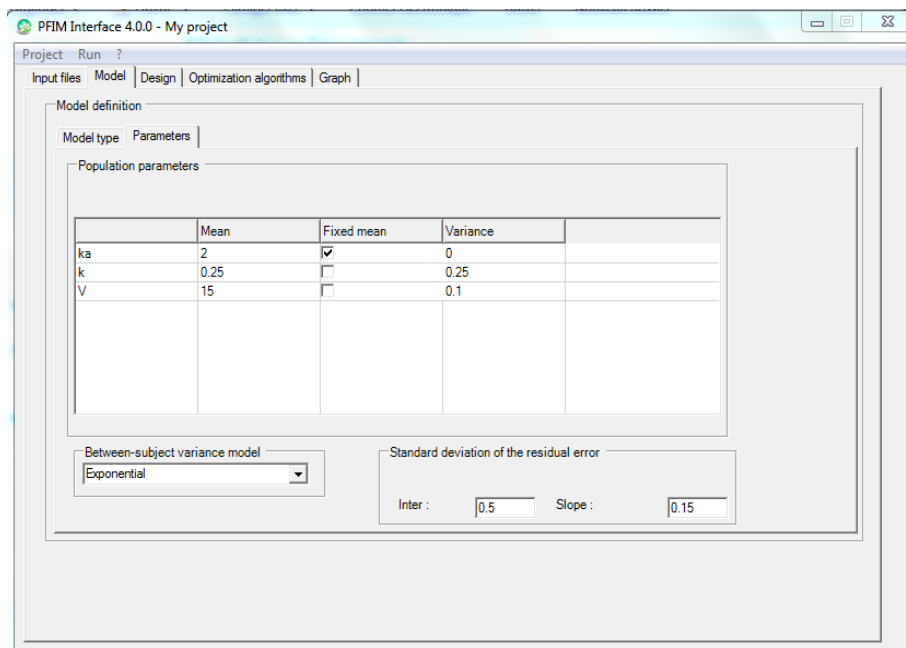


Figure 52: Parameters tab when fixing parameter k_a

2. Example 2: PK and immediate response PD model using the libraries of PK and PD Models (ODE)

- 100 subjects with a dose of 100

The aim is to evaluate, in a population approach, the following one group designs:

- sampling times for PK response: 0.5, 2, 30, 49, 180
- sampling times for PD response: 0.5, 2, 14, 110, 150

for a PKPD model, where the PK is one compartment infusion input with Michaelis-Menten elimination after a single dose administration with parameters V , V_m and k_m and the PD is an immediate response model with a linear drug action and without baseline, where the parameter is A_{lin} .

		Mean	Fixed mean	Variance
	V	12.2	<input type="checkbox"/>	0.25
	V_m	0.082	<input type="checkbox"/>	0.25
	k_m	0.37	<input type="checkbox"/>	0
	A_{lin}	0.1	<input type="checkbox"/>	0.25
Resp A	σ_{inter}		0	
	σ_{slope}		0.2	
Resp B	σ_{inter}		0.1	
	σ_{slope}		0	

Optimisation

1. Example 1: PK Model

We illustrate optimisation algorithms with the same examples used in the Evaluation section and with an additional example (Example 3) that shows a case of repeated dose regimen. As for Evaluation, Example 1 is more detailed with all the new features. For Example 2 and 3 optimisation was

performed only with Federow-Wynn (FW) algorithm. For Example 1 and 2 we therefore show only the parts concerning optimisation.

1.1 Population Fisher Information Matrix (P-FIM)

The aim is to optimise a design for 200 subjects with a dose of 100.

1.1.1 Simplex algorithm

- Initial sampling times vector: $\xi_1 = (0.33, 1.5, 5, 12)$
- Time interval for the optimisation: $(0, 12)$

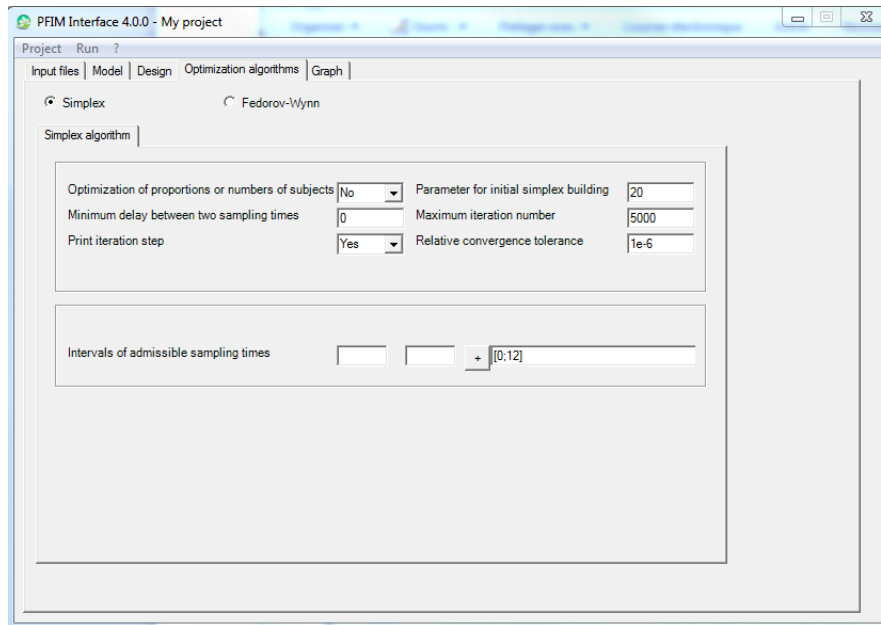


Figure 53: Simplex algorithm specifications

Comment on results

The criterion associated with the initial times (ξ_1) was 361.7. When optimizing with Simplex algorithm, the criterion associated to the optimal times $(0.325, 1.632, 4.9, 12)$ improved of about 1 point, (362.4).

1.1.2 Fedorov-Wynn algorithm

- Allowed sampling times: 0.33, 1, 1.5, 3, 5, 8, 12
- Maximum total number of points in one elementary protocol: 4

Compare the result with the one obtained with Simplex algorithm

Optimal times were found for three groups of approximately 71, 123 and 6 subjects, respectively. These are: $(0.33, 1, 1.5, 8)$; $(0.33, 1.5, 8, 12)$ and $(0.33, 1.5, 3, 12)$ for the first, second and third group, respectively. For this example, the optimisation with Fedorov-Wynn (FW) algorithm led to a criterion of 371.3, higher than the one obtained with the Simplex algorithm (362.4) in point 1.1.1. Relative standard errors are acceptable in both cases (below 20%).

1.1.3 Fixed parameters

The aim is to optimise the design in 1.1 keeping the parameter k_a fixed (assuming no variability on k_a) using the constraints as in 1.1.1 and 1.1.2. Corresponding examples are stored in the directory of PFIM interface 4.0 (Examples \rightarrow OPTIMISATION \rightarrow EXAMPLE_1) in the folder 1.1.3.1 and 1.1.3.2 for the examples optimisation with Simplex or with FW algorithm, respectively.

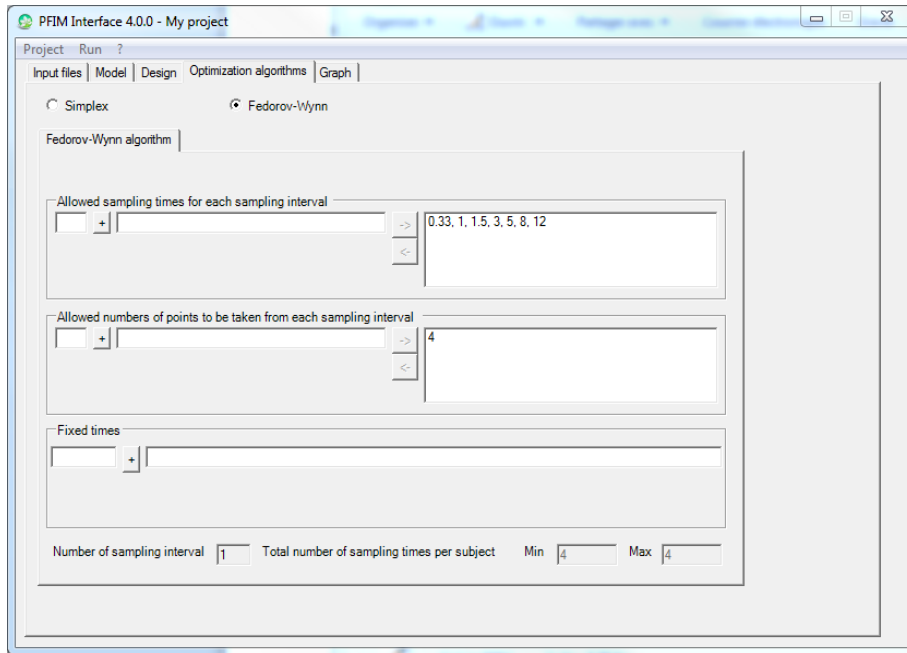


Figure 54: Fedorov-Wynn algorithm specifications

1.1.4 Fixed sampling times

Optimize the design in 1.1 with the Fedorov-Wynn algorithm keeping fixed two sampling times (0.33,1.5), using the constrains as in 1.1.1 and 1.1.2. (In PFIM Interface 4.0 it is possible to fix times only with the Fedorov-Wynn algorithm).

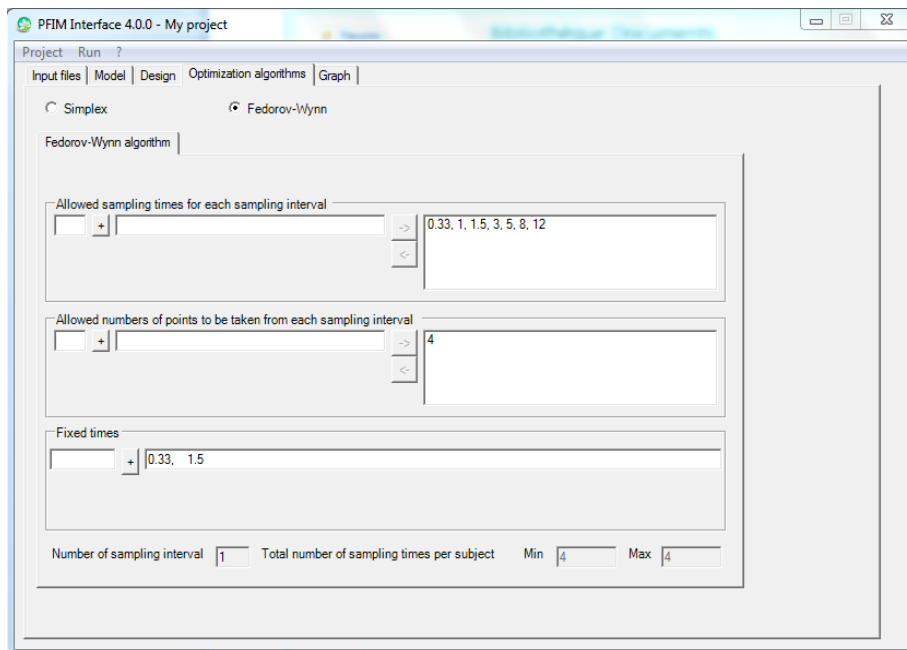


Figure 55: Fedorov-Wynn algorithm specifications: fixed sampling times

1.1.5 Fixed Parameter and Fixed Sampling Times

Optimize P-FIM using the Fedorov-Wynn algorithm keeping the parameter k_a fixed (assuming no variability on k_a) and keeping fixed 2 sampling times (0.33, 1.5). Essentially this example is the combination of examples 1.1.3.2 and 1.4.1.

1.2 Individual Fisher Information Matrix (I-FIM)

Optimize the design in 1.1 this time for the Individual Fisher Information Matrix (I-FIM). Use same constraints in 1.1.1 and 1.1.2. The optimisations performed for P-FIM are repeated in case of I-FIM. Examples 1.2.1 and 1.2.2 show the optimisation of I-FIM with the Simplex algorithm and Fedorov-Wynn algorithm, respectively; in Example 1.2.3 the optimisation is performed with the Fedorov-Wynn algorithm, fixing parameter k_a and two sampling times (0.33, 1.5).

1.3 Bayesian Fisher Information Matrix (B-FIM)

Optimize the design in 1.1 this time for the Bayesian Fisher Information Matrix (I-FIM). Use same constraints in 1.1.1 and 1.1.2. The optimisations performed for P-FIM are repeated in case of B-FIM. Examples 1.3.1 and 1.3.2 show the optimisation of B-FIM with the Simplex algorithm and Fedorov-Wynn algorithm, respectively; in Example 1.3.3 the optimisation is performed with the Fedorov-Wynn algorithm, fixing parameter k_a and two sampling times (0.33, 1.5).

2. Example 2: PK and immediate response PD model using the libraries of PK and PD Models (ODE)

- 100 subjects with a dose of 100
- Allowed sampling times for PK response: 0.5, 2, 30, 49, 180
- Allowed sampling times for PD response: 0.5, 2, 14, 110, 150
- Number of sampling times to be optimized: 3

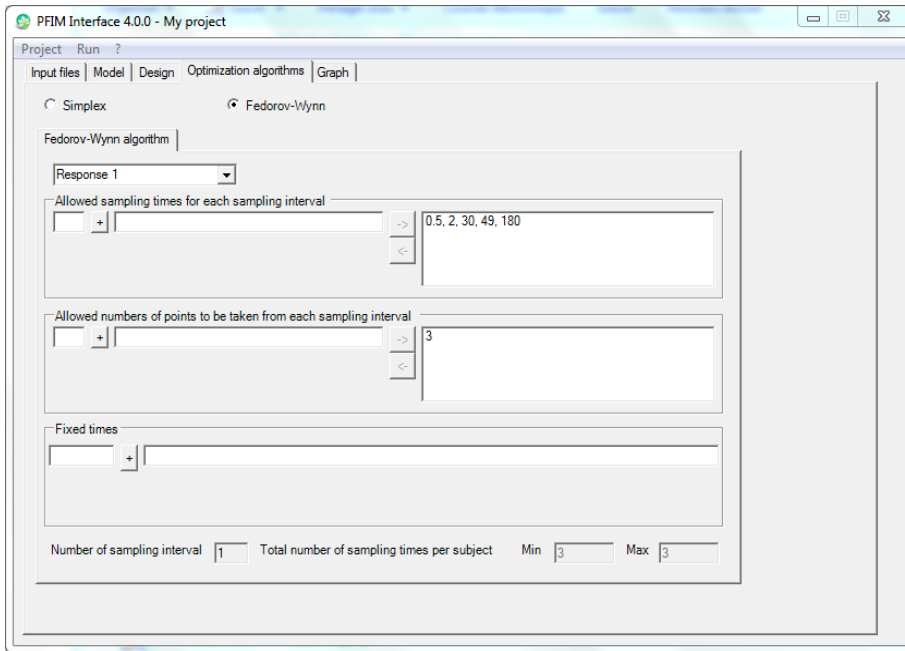


Figure 56: Fedorov-Wynn algorithm specifications for the first response

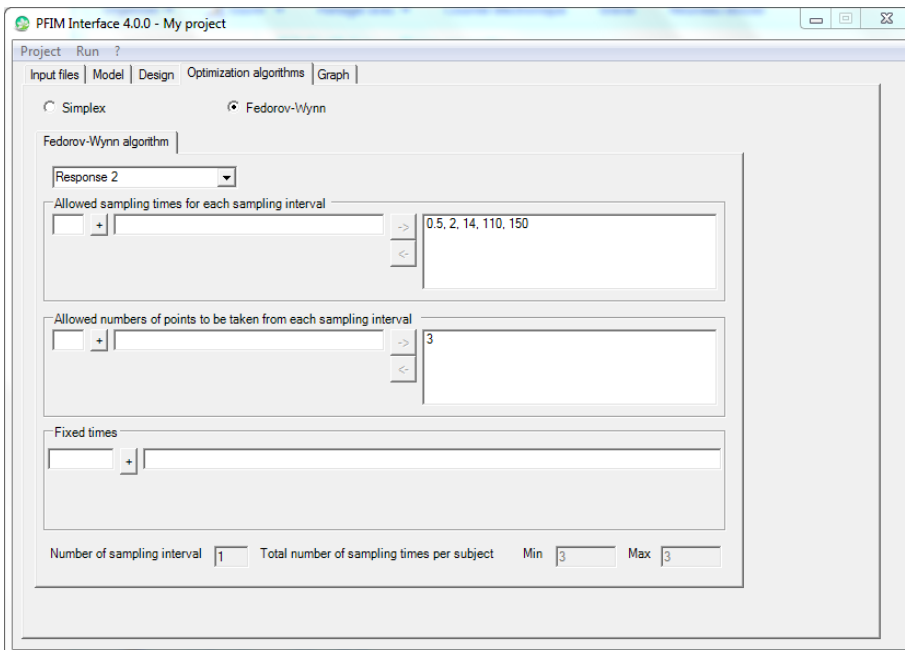


Figure 57: Fedorov-Wynn algorithm specifications for the second response

3. Example 3: Repeated doses

This example deals with the optimisation of a design using the Fedorov-Wynn algorithm and with a model defined by the user using the functions of the library of models.

Repeated doses of 100 with oral absorption every 12 hours are considered. The model is a one compartment model first order absorption, parameterized with rate constant of absorption (k_a), volume (V) and clearance (Cl). The mean and the variance of those parameters are given in the table below. The random effects are exponentially modelled and the variance error model is proportional.

	Mean	Fixed mean	Variance
k_a	0.7	<input type="checkbox"/>	0.25
Cl	0.5	<input type="checkbox"/>	0.25
V	5	<input type="checkbox"/>	0.25
σ_{inter}		0	
σ_{slope}		0.2	

The purpose is to optimise a design with 90 subjects, with sampling times after the first and the fifth doses, using the Fedorov-Wynn algorithm. After the first dose, 2 or 3 samples per subject are allowed in the following set (0.5, 1, 2, 3, 4, 6, 8, 10, 12). After the fifth dose, 2 or 3 samples per subject are also allowed in the following set: (48.5, 49, 50, 51, 52, 56, 58, 60).

The initial population design used to run the Fedorov-Wynn algorithm is composed of four sampling times: (0.5, 12, 50, 60) to be performed in 90 subjects.

4. Examples from PFIM Interface 3.1

4.1 Single response model

4.1.1 Evaluation

4.1.1.1 Example A

This example deals with the evaluation of a population design using the library of model.

The purpose is to evaluate a design using a one compartment model after a single bolus administration. The parameters and their values are given in the table. The random effects are modelled exponentially. The variance error model is a combined error model.

The design to be evaluated is composed of two groups: one group of 30 subjects with a dose of 100 and sampling times at (0.5, 2, 3, 10) and one group of 90 subjects with a dose of 200 and sampling times at (1, 4, 12).

	Mean	Fixed mean	Variance
V	10	<input type="checkbox"/>	0.25
k	0.2	<input type="checkbox"/>	0.25
σ_{inter}	0.5		
σ_{slope}	0.15		

4.1.1.2 Example B

This example deals with the evaluation of a design using a differential equation system. The model is a one compartment model first order absorption and Michaelis-Menten elimination. The parameters and their values are given in the table below.

The design to be evaluated is 0.5, 2, 16, 23.5 with a dose of 13.8 performed in 30 subjects.

The variance error model is proportional and the modelling of the random effects is exponential.

	Mean	Fixed mean	Variance
ka	2.72	<input type="checkbox"/>	0.25
V	12.2	<input type="checkbox"/>	0.25
Vm	1.004	<input type="checkbox"/>	0.25
km	0.37	<input type="checkbox"/>	0.25
σ_{slope}	0.2		

4.1.2 Optimisation



Be careful, remember that, with the Fedorov-Wynn algorithm, the sampling times of the initial population design should be included in the allowed sampling times, so as the number of allowed samples per group.

4.1.2.1 Example C

This example deals with the optimisation of a design using the Fedorov-Wynn algorithm and with a model defined by the user using the functions of the library of models.

Repeated doses of 100 with oral absorption every 12 hours are considered. The model is a one compartment model first order absorption, parameterized with rate constant of absorption (k_a), volume (V) and clearance (Cl). The mean and the variance of those parameters are given in the table below. The random effects are exponentially modelled and the variance error model is proportional.

	Mean	Fixed mean	Variance
k_a	0.7	<input type="checkbox"/>	0.25
Cl	0.5	<input type="checkbox"/>	0.25
V	5	<input type="checkbox"/>	0.25
σ_{slope}	0.2		

The purpose is to optimise a design with 90 subjects, with sampling times after the first and the fifth doses, using the Fedorov-Wynn algorithm. After the first dose, 2 or 3 samples per subject are allowed in the following set (0.5, 1, 2, 3, 4, 6, 8, 10, 12). After the fifth dose, 2 or 3 samples per subject are also allowed in the following set: (48.5, 49, 50, 51, 52, 56, 58, 60).

The initial population design used to run the Fedorov-Wynn algorithm is composed of four sampling times: (0.5, 12, 50, 60) to be performed in 90 subjects.

4.1.2.2 Example D

This example deals with the optimisation of a design using the Fedorov-Wynn algorithm. The model is described by a two compartment model after infusion administration, parameterized in volume (V), rate constant of elimination (k), and inter-compartmental parameters k_{12} and k_{21} . The total dose is equal to 550 and the duration of infusion is 0.0625. The random effects are modelled exponentially. The variance error model is proportional.

The mean, the variance of the parameters and the parameters of the variance error model are given in the tab bellow.

	Mean	Fixed mean	Variance
V	3.08	<input type="checkbox"/>	0.1
k	0.0808	<input type="checkbox"/>	0.2
k_{12}	0.175	<input type="checkbox"/>	0.3
k_{21}	0.116	<input type="checkbox"/>	0.1
σ_{slope}	0.25		

A set of eleven allowed sampling times is given: (0.0625, 1, 2, 3, 4, 6, 7, 10, 14, 18, 21).

90 subjects can be involved with either 3 or 4 samples per subject.

An initial design is proposed, with 4 samples per subject, the same into the 90 subjects: (0.0625, 7, 14, 21).

4.1.2.3 Example E

This example deals with the optimisation of a design using the Simplex algorithm and with a model defined by the user using the functions of the library of models.

Ten repeated doses of 2.5 with oral absorption every 24 hours are considered. The model is a two compartment model first order absorption, parameterized with rate constant of absorption (k_a), volume for the first compartment (V_1), clearance (Cl), volume (V_2) and the intercompartmental clearance (Q). The between subject variance model is exponentially and the variance model is additive.

Values of the parameters are given in the tab bellow:

	Mean	Fixed mean	Variance
k_a	1.5	<input type="checkbox"/>	0.502
Cl	0.345	<input type="checkbox"/>	0.059
V_1	8	<input type="checkbox"/>	0.018
Q	0.145	<input type="checkbox"/>	0
V_2	18	<input type="checkbox"/>	1.9
σ_{inter}	0.08		

The aim is to optimise a design with 250 subjects, with sampling times between the first dose and the tenth doses and also five days after the last one, using the Simplex algorithm. Thus, the admissible sampling times are between 0 and 360 hours.

The initial population design used to run the Simplex algorithm is composed of 6 sampling times: (1, 24, 96, 180, 250, 300) to be performed into 250 subjects.

4.2 Multiple response model

4.2.1 Evaluation

4.2.1.1 Example F

This example deals with the evaluation of a joint modelling of a drug concentration and its effect (two responses): a one compartment model with a first order absorption and elimination for the drug concentration is used and an immediate response model with a constant baseline for the effect. The model is described using the libraries of models. The between subject variance model is exponentially and the variance model is combined for the first response and additive for the second response.

Values of the parameters are given in the tab bellow:

	Mean	Fixed mean	Variance
k_a	1.6	<input type="checkbox"/>	0.70
V	8	<input type="checkbox"/>	0.02
Cl	0.13	<input type="checkbox"/>	0.06
$Imax$	0.73	<input type="checkbox"/>	0.001
C_{50}	0.17	<input type="checkbox"/>	0.7
S_0	100	<input type="checkbox"/>	0
σ_{inter} (first response)	0.6		
σ_{slope}	0.07		
σ_{inter} (second response)	8		

The aim is to evaluate a design with one group with sampling times at 0.5, 1, 2, 3, 6, 9, 12, 24, 36, 48, 72, 96, 120 hours for the first response and 0, 24, 36, 48, 72, 96, 120, 144 hours for the second response with 32 subjects. The total dose is equal to 100.

4.2.1.2 Example G

This example deals with the evaluation of a design for a joint model for a drug and its metabolite. The first response is described by a one compartmental model with first order absorption and the second response is described by a one compartment with a first order metabolic rate constant. Because of structural identifiability problem in absence of urinary data, we fix the volume of distribution (V_m) of the metabolite equal to 1, and thus estimate Cl_m and k_m . The model is described using a differential equation system. The length of vector in the "initial conditions for each elementary design" is equal to 3. The first element of this vector is the dose equal to 300 in this example. The between subject variance model is exponentially and the variance model is combined for the first response and proportional for the second response.

Values of the parameters are given in the tab below:

	Mean	Fixed mean	Variance
ka	2.86	<input type="checkbox"/>	0.7
V	300	<input type="checkbox"/>	0.02
Cl	160	<input type="checkbox"/>	0.06
Cl_m	0.16	<input type="checkbox"/>	0.17
k_m	0.03	<input type="checkbox"/>	0
σ_{inter} (first response)		0.003	
σ_{slope}		0.28	
σ_{inter} (second response)		0.13	

The design to be evaluated is composed of 1 group of 80 subjects with sampling times at (1, 3, 6, 12) for the first response and sampling times at (1, 6, 11, 12) for the second response.

4.2.1.3 Example H

This example deals with the evaluation of a design study for a PK/PD model (two responses). The PK model is a one compartmental model with first order absorption and elimination. The drug effect (PD model) is described by a turnover model with inhibition of the input.

This PK/PD model is described using the libraries of models. In this example, we are in the case where we have a PK model with linear elimination (written using an analytical form) and a turnover response PD model (written using a differential equation system). Thus, the user has to complete the tab of the ODE variables because PFIM Interface 3.1 calls a specific function in order to create a system of differential equation system describing the corresponding PK/PD model. The between subject variance model is exponentially. The variance model is combined for the PK model and additive for the second response.

Values of the parameters are given in the tab bellow:

	Mean	Fixed mean	Variance
ka	1.6	<input type="checkbox"/>	0.70
V	8	<input type="checkbox"/>	0.02
Cl	0.13	<input type="checkbox"/>	0.06
Rin	5.4	<input type="checkbox"/>	0.2
kout	0.06	<input type="checkbox"/>	0.02
Imax	1	<input type="checkbox"/>	0
C50	1.2	<input type="checkbox"/>	0.01
<hr/>			
σ_{inter} (first response)		0.6	
σ_{slope}		0.07	
σ_{inter} (second response)		8	

The design to be evaluated is composed of one group of 32 subjects with sampling times at 0.5, 1, 2, 3, 6, 9, 12, 24, 36, 48, 72, 96, 120 for the PK model and 0, 24, 36, 48, 72, 96, 120, 144 for the PD model. The dose is equal to 100.

4.2.2 Optimisation

4.2.2.1 Example I

This example deals with the optimisation of a design using the Simplex algorithm for a joint modelling of a drug concentration and its effect: a one compartment model with a first order absorption and elimination for the drug concentration is used and an immediate response model with a constant baseline for the effect. The model is described using analytical forms with the 'user defined model' option.

This model has been used for design evaluation in the section 5.2.1.1 using the library of models.

Values of the parameters are given in the tab bellow:

	Mean	Fixed mean	Variance
ka	1.6	<input type="checkbox"/>	0.70
V	8	<input type="checkbox"/>	0.02
Cl	0.13	<input type="checkbox"/>	0.06
Imax	0.73	<input type="checkbox"/>	0.001
C50	0.17	<input type="checkbox"/>	0.7
S0	100	<input type="checkbox"/>	0
<hr/>			
σ_{inter} (first response)		0.6	
σ_{slope}		0.07	
σ_{inter} (second response)		8	

The aim is to optimise a design with one group of 32 subjects with 5 sampling times between 10 min and 120 hours for the drug concentration measurements and 5 sampling times between 0 and 144 hours for the effect measurements.

4.2.2.2 Example J

This example deals with the optimisation of a design using the Fedorov-Wynn algorithm for a joint model for a drug and its metabolite (two responses). The first response is described by a one compartmental model with first order absorption and the second response is described by a one compartment with a first order metabolic rate constant. The model is described using a differential equation system. The between subject variance model is exponentially and the variance model is combined for the first response and proportional for the second response.

This model has been used for design evaluation in the section 5.2.1.2.

Values of the parameters are given in the tab bellow:

	Mean	Fixed mean	Variance
ka	2.8	<input type="checkbox"/>	0.70
V	300	<input type="checkbox"/>	0.02
Cl	160	<input type="checkbox"/>	0.06
Clm	0.16	<input type="checkbox"/>	0.001
R	0.03	<input type="checkbox"/>	0.7
<hr/>			
σ_{inter} (first response)		0.03	
σ_{slope}		0.28	
σ_{slope} (second response)		0.13	

The aim is to optimise a design (same sampling times for both responses) with 4 sampling times for a total number of samples equal to 400 using the following allowed sampling times: 0.0625, 1, 3, 6, 11, 12, 14 and 15hours.



Be careful, remember that, with the Fedorov-Wynn algorithm, the sampling times of the initial population design should be included in the allowed sampling times, so as the number of allowed samples per group.

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