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Pharmacometric models to inform dose selection and study design

Applied in hemophilia and tuberculosis

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Abstract
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While tuberculosis is a global pandemic, hemophilia is a rare disease which many have not heard of. Due to tuberculosis mainly being a problem in developing countries and hemophilia being a rare disease, they are not as heard of as other diseases such as cancer or metabolic diseases which are on the rise in Western societies. The quality of life for patients suffering from these diseases is notably impaired and novel drugs are warranted to further improve the treatment and management of both diseases. As market incentives are a limiting factor, it is important that the efforts that are taken to develop novel drugs are carried out in an informative manner.

One strategy to incorporate as much information as possible to inform decision making in drug development is to use pharmacometric methods. Such strategies enable simultaneous analysis of different types of data that are generated during drug development programs. In this thesis, the aim was to develop and apply pharmacometric models to facilitate dose selection and study designs in clinical programs that aim at developing new drugs for tuberculosis and hemophilia.

A standardized analysis approach of early clinical trials studying drugs against tuberculosis was presented including power calculations that showed the number of patients needed to detect drug effects. Such efforts are important as showing drug effect in early trials will aid decision making into significantly longer and costlier late trials. The approach was used to analyze a clinical trial studying if the current dose of meropenem can be lowered without negatively impacting drug effects and improving the already poor tolerability of the drug. The study found that lowering the dose may lower activity without any improvement of the tolerability properties. Furthermore, population pharmacokinetic models were developed for two novel hemostatic drugs in development for prophylactic and on-demand treatment of hemophilia. Based on the models, clinical trials in adult and pediatric subjects were supported. One of the trials were performed and it was showed with a model-based analysis that the new drug which is given subcutanously has similar efficacy as current intravenously given standard of care alternatives. Using the developed models, different strategies for designing pharmacokinetic trials in children was also presented. In conclusion, the work performed within this thesis has contributed to the development of new drugs against tuberculosis and hemophilia.

Keywords: drug development, pharmacometrics, pharmacokinetics, pharmacodynamics, non-linear mixed effect models, hemophilia, tuberculosis, dose selection, study design, pediatric extrapolation

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The idea is to go from numbers to information to understanding

Hans Rosling

To everyone that made this possible

List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.

I. Mockeliunas L, **Faraj A**, van Wijk RC, Upton CM, van den Hoogen G, Diacon AH, et al. Standards for model-based early bactericidal activity analysis and sample size determination in tuberculosis drug development. Front Pharmacol. 2023 Apr; 13:14:1150243

II. De Jager, V., Gupte, N., Nunes, S., Barnes, G.L., van Wijk, R.C., Mostert, J., Dorman, S.E., Abulfathi, A.A., Upton, C.M., **Faraj, A.**, Nuermberger, E.L., Lamichhane, G., Svensson, E.M., Simonsson, U.S.H., Diacon, A.H., Dooley, K.E. Early Bactericidal Activity of Meropenem plus Clavulanate (with or without Rifampin) for Tuberculosis: The COMRADE Randomized, Phase 2A Clinical Trial. Am. J. Respir. Crit. Care Med. 2022 May 15;205(10):1228-35

III. **Faraj, A**. et al. Phase-3 Dose Selection of Marzeptacog Alfa (Activated) Informed by Population Pharmacokinetic Modelling: A Novel Hemostatic Drug. CPT Pharmacometrics Syst Pharmacol. 2022 Dec;11(12):1628-37

IV. **Faraj A**, van Wijk RC, Neuman L, Desai S, Blouse GE, Knudsen T, et al. Model-informed pediatric dose selection of marzeptacog alfa (activated): An exposure matching strategy. CPT Pharmacomet Syst Pharmacol. 2023 Jul;12(7):977–87

V. **Faraj A**, Le Moan N, Gorina E, Blouse GE, Knudsen T, Simonsson USH. Model-Informed Support of Dose Selection for Prophylactic Treatment with Dalcinonacog Alfa in Adult and Paediatric Hemophilia B Patients. Adv Ther. 2023 Sep;40(9):3739–50

VI. **Faraj A**, Knudsen T, Nyberg J, Blouse GE, Simonsson USH. Subcutaneous marzeptacog alfa (activated) for on-demand treatment of bleeding events in subjects with hemophilia A or B with inhibitors. Clin Pharmacol Ther. 2024 Jan;0(0)

VII. **Faraj A**, Ayoun Alsoud R, Nyberg J, Simonsson USH. Model-based approaches to prospectively power pediatric pharmacokinetic trials with limiting sample size (manuscript)

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List of Additional Papers

Additional papers not included in the thesis.

I. **Faraj A**, Svensson RJ, Diacon AH, Simonsson USH. Drug effect of clofazimine on persisters explain an unexpected increase in bacterial load from patients. Antimicrob. Agents Chemother. 2020 Apr 21;64(5):01905-19

II. Clewe O, **Faraj A**, Hu Y, Coates ARM, Simonsson USH. A model-based analysis identifies difference in phenotypic resistance between in vitro and in vivo – implications for translational medicine. Journal of Pharmacokinetics and Pharmacodynamics. 2020 Jun;47(5): 421-430

III. **Faraj A**, Clewe O, Svensson RJ, Mukamolova GV, Barer MR, Simonsson USH. Difference in persistent tuberculosis bacteria between in vitro and sputum from patients – implications for translational medicine. Scientific Reports. 2020 Sep 23;10(1):15537

IV. Saskia J, Upton CM, De Jager VR, **Faraj A**, Pahar M, Miranda IDS, Diacon AH, Simonsson USH, Niesler RT. Cough as Non-Invasive Biomarker for Monitoring Tuberculosis Treatment – a Proof of Concept Study. Annals of the American Thoracic Society. 2023 Dec;20(12):1822-1825

V. Krishnan N, Priestman M, Uhía I, Charitakis N, Glegola-Madejska IT, Baer TM, Tranberg A, **Faraj A**, Simonsson USH, Robertson BD. A BCG Skin Challenge Model for Assessing TB Vaccines (manuscript)

Contents

Abbreviations

ABR	annualized bleeding rate
ADA	anti-drug antibody
ADE	age-dependent exponent
APCC	activated prothrombin complex concentrates
AUC	area under the curve
BASE	baseline endogenous coagulation factor level
BDBP	baseline diastolic blood pressure
BLOQ	below limit of quantification
BMI	body mass index
CFU	colony forming units
CL	clearance
C_{max}	maximum concentration
C_{min}	minimum concentration
CV	coefficient of variation
CI	confidence interval
D	duration of zero order absorption
DalcA	dalcinonacog alfa
DS-TB	drug-sensitive tuberculosis
EBA	early bactericidal activity
EMA	European Medicines Agency
EOD	every other day
F	bioavailability
IF	individual bioavailability
FDA	Food and Drug Administration
FIX	factor IX
FVIIa	activated factor VII
FVIII	factor VIII
HA	hemophilia A
HB	hemophilia B
HIV	human immunodeficiency virus
IBV	inter-bleed variability
ICL	individual clearance
I_{dur}	individual duration of zero order absorption
IIV	inter-individual variability
Ik_a	individual absorption rate constant

IPRED	individual prediction
IV	intravenous
MarzAA	marzeptacog alfa (activated)
MBDD	model-based drug development
MCMP	monte-carlo mapped power
MDR-TB	multi-drug resistant tuberculosis
MIDD	model-informed drug development
M&S	modelling and simulation
NCA	non-compartmental analysis
NHANES	National Health and Nutrition Examination Survey
NLMEM	nonlinear mixed effects model
IOV	inter-occasional variability
OD	omne in die (once daily)
OFV	objective function value
pcVPC	prediction-corrected visual predictive check
PD	pharmacodynamics
PIP	pediatric investigation plan
PK	pharmacokinetics
PK-PD	pharmacokinetic-pharmacodynamic
PsN	perl-speaks-nonmem
PSP	pediatric study plan
R&D	research and development
TB	tuberculosis
rFVIIa	recombinant activated factor VII
RSE	relative standard error
RUV	residual unexplained variability
SC	subcutaneous
SCM	stepwise covariate modelling
SD	standard deviation
SE	standard error
SoC	standard of care
SSE	stochastic simulation and estimation
TF	treatment failure
TID	ter in die (three times a day)
T_{max}	time to maximum concentration
TS	treatment success
TTP	time to positivity
V	volume of distribution
VPC	visual predictive check
WHO	World Health Organization
XDR-TB	extensively drug-resistant tuberculosis
x	independent variable
y	dependent variable, observation
ϵ	epsilon

κ	kappa, occasion level random effect
η	eta, individual level random effect
θ	theta, population fixed effect parameter
X	individual characteristics (demographics, covariates, dose)

Introduction

The history of drug development dates back to early days of civilization when plants and minerals were tested in observational studies dominated by trial and error, and later used for healing purposes. With the emergence of chemistry as a discipline in the 1800th century, potent molecules such as morphine were isolated and used for their medicinal characteristics (1). The discovery and isolation of antibiotics in the first half of the 1900 century led to a significant increase in life expectancy and introduced possibility to treat previously deadly infectious diseases. Along the rise of pharmaceutical industry, the first legislation that laid the grounds for modern drug regulation was signed in 1906 by US president Theodore Roosevelt, namely the Pure Food and Drug Act (2) which in 1927 led to the birth of The Food and Drug Administration (FDA). In 1995, the European Medicines Agency (EMA) was formed. These agencies oversee drug development and play an essential role in protecting public health and guiding development of new drugs.

Drug development can be divided into preclinical and clinical development (3). The former includes discovering and identifying a target that plays a role in a disease followed by identification of compounds (also referred to as leads) that may exhibit activity towards the target. Lead compounds are then further studied in *in-vitro* and in animal experiments to assess absorption, distribution, metabolism, elimination, toxicology and potential efficacy. The molecule with most promising results (candidate drug) may be taken forward into significantly costlier (4) clinical development following submission of an Investigational New Drug application which include all data that shows promise of the new drug in humans.

Drug research and development (R&D) has become more expensive and inefficient over the years and is estimated to cost an average of $6.16 billion per new drug (5). In parallel to increase in cost, there are high attrition rates primarily due to safety and inability to show efficacy in late clinical development (6–9). This indicates that information in early clinical development may be utilized more optimally to either proceed the R&D program with more informed designs or to terminate the program earlier and redirect economic incentives elsewhere. These aspects are particularly important for rare diseases such as hemophilia or for infectious diseases as tuberculosis which suffer from relatively low market incentive.

Tuberculosis

A silent killer

In developed countries, tuberculosis (TB) disease may be viewed as a problem of the past, however, although often not spoken of, TB is the infectious disease that lead to most deaths worldwide after COVID (10). TB is vastly spread through coughing and is caused by Mycobacterium tuberculosis, appearing primarily but not exclusively in the lungs. The time of infection and time of developing active disease is often different and symptoms usually appear late in clinical infection (11). A large number of people are infected every year but only 5-15% develop active disease over a lifetime. Subjects with TB experience a general feeling of weakness with excessive coughing and loss in appetite with subsequent weight loss. The active form the disease is diagnosed by chest x-ray or by sputum based mycobacterial methods that aims to grow bacteria on solid (colony forming units [CFU]) or in liquid medium (mycobacterium growth indicator tube-based time-to-positivity [TTP]).

Different classifications of TB exist and they are related to the susceptibility to a combination of drugs. Drug sensitive (DS-TB) is a form of the disease where the strain is not resistant to the main drugs used to treat TB, namely rifampicin and isoniazid. Multidrug-resistant (MDR-TB) is defined by that the strain is resistant to rifampicin and isoniazid. Extensively drug-resistant (XDR-TB) is a rare form of TB and is defined by that the strain is resistant to several drugs on top of rifampicin and isoniazid leading to limited treatment options that are considerably less effective (10). The different forms of the disease are handled with different treatment strategies and the duration of treatment varies. As TB is treated with combination of drugs leading to a mixture of side effects, adherence is also a problem (10).

Treatment recommendations

Sensitive forms of TB (DS-TB) is treated for 6 months with 4 months of rifampicin, isoniazid, pyrazinamide and ethambutol followed by 2 months of rifampicin and isoniazid (12). A newer 4-month regimen exists where patients are treated with rifapentine, moxifloxacin, isoniazid and pyrazinamide for 8 weeks followed by rifapentine, moxifloxacin and isoniazid for 9 weeks (12). The older regimen is still recommended as the first choice by the World Health Organization (WHO) as the certainty of evidence for efficacy is higher (12). For drug resistant TB, a 6-9 month regimen consisting of bedaquiline, pretomanid, linezolid and moxifloxacin is recommended for both MDR- and XDR-TB patients (13). However, these recommendations are not always fully followed and countries have slightly different strategies depending on availability of drugs and local clinical experience. In either strategy, sputum-based quantification of bacteria is done after 2 months to assess clinical response.

Although it is common that a patient is declared culture negative after 2 months, treatment is proceeded to avoid relapse. To tackle further development of resistance and bring more efficacious and safe drugs to patients, development of new drugs in parallel to optimization of the use of current antibiotics is essential. In this thesis, model-based analysis methods for early clinical trials are presented.

Hemophilia

A bleeding disorder

Unlike TB which spreads through the air, hemophilia is classified as a rare hereditary disease and is mostly prevalent in boys and men. Both hemophilia A and B (HA/HB) are X-linked inherited bleeding disorders that occur in approximately 1 out of 5000 or 30000 male births, respectively (14), and lead to deficiency of coagulation factors VIII (FVIII) and IX (FIX), respectively. The deficiency of these coagulation factors disrupts the coagulation cascade by delaying and reducing the formation of thrombin, leading to inadequate clotting. Diagnosis of the disease is usually done based on a blood sample that is analyzed to detect what protein is missing and to what extent.

The disease can be clinically classified into different phenotypes depending on the plasma level activity of the coagulation factor. Severe forms of the disease are defined as having <1 % of normal levels, whereas the moderate and mild forms are defined as having 1-5 % or <5 % and <40 %, respectively (15). Severe phenotypes of the disease often get diagnosed within the first year after birth as the child become more physically active, whereas mild forms might become apparent later in adulthood. Patients suffering from severe hemophilia often experience bleeding events in joints, muscles or soft tissue without any apparent reasons and in addition, the severe phenotype can cause life-threatening intracranial hemorrhages. In contrast, patients with the moderate or mild phenotype rarely experience spontaneous bleeds but are subject to excessive bleeding following physical trauma or surgery (16). Repeated bleeding events in the same joint may cause arthropathy with negative effects on mobility, risk of chronic pain and subsequent impairment of quality of life (17).

Treatment recommendations

HA/HB has historically been treated by replacing the missing coagulation factor with intravenous (IV) infusions, either continuously (prophylaxis) or on-demand. Recombinant variants of FVIII and FIX has led to extended half-life products with potential to reduce the frequency of administration (18). In parallel to the development of recombinant variants with longer half-life, the emergence of easier-to-administer subcutaneous (SC) formulations has

contributed to making management of the disease easier for patients (19). Although replacement treatments have shown efficacy, the developments of inhibitors by the immune system make replacement medicines ineffective. Furthermore, in 22th of November 2022, the first gene therapy for HB was approved by FDA (20) and in 17th of April 2023, the first gene therapy for severe HA was approved by FDA (21). Gene therapies are promising as the treatment is administered once, but come with high price ($3 million per injection for the HB treatment alternative) and the health economic benefit remains to be shown, as more follow-up data is needed on the effect duration. This thesis will not address gene therapies.

Hemophilia A

The most common treatment strategy in modern healthcare for HA have been to replace the missing coagulation factor FVIII. Either standard half-life or extended half-life (recombinant variants) replacement therapies are used, usually dosed per kilogram of bodyweight by infusion (22). Preferably, prophylaxis is the strategy of choice to improve quality of life and bleeding-related sequalae and is then given 2-3 times per week (22). The alternative is to treat bleeds as they occur on-demand, and is then also dosed per kilogram of bodyweight. However, the number of doses needed and the dose level will depend on the severity of the bleeding event and treatment response. In 16th of November, 2017, the FDA approved the first SC prophylactic treatment, emicizumab (Hemlibra®) which is an antibody that mimics the function of FVIII by binding activated FIX and factor X (23). Emicizumab was approved for HA with and without inhibitors to FVIII and is recommended to be given every 1-4 weeks (depends on dose level) as a single SC dose (24). However, subjects that produce anti-drug antibodies (ADAs) to emicizumab or experience breakthrough bleeds when on prophylactic treatment with emicizumab are still in need of IV given by-passing agents. Examples of by-passing agents are wild-type FVIIa (wt-FVIIa) or activated prothrombin complex concentrates (APCCs). For these patients, a new efficacious, safe and easy-to-administer alternative is needed. Marzeptacog alfa (activated [MarzAA]) is a SC given recombinant FVIIa variant (25–27) for acute bleeding events and the clinical development program has been informed by the work in this thesis (28,29).

Hemophilia B

Similar to HA, HB treatment is also dominated by prophylactic replacement therapy given by the IV route and is also dosed per kilogram of bodyweight. Both standard and extended half-life products exist (allowing for weekly to every-two-weekly dosing) (30) and the treatment aim is to elevate FIX levels into milder forms of the disease. In 17th of April, the first SC prophylactic treatment alternative (concizumab [Alhemo®]) for HB with inhibitors was approved in Canada (31), but is yet to be approved in Europe and the US.

However, for subjects that develop ADAs to concizumab or experience breakthrough bleeds, IV given by-passing agents are recommended (22). In addition to SC MarzAA as an alternative to IV by-passing agents for treatment of acute bleeds, the development of another SC prophylactic treatment alternative is addressed in this thesis, namely dalcinonacog alfa (DalcA), a novel recombinant FIX variant.

Drug development pathway

Tuberculosis

Clinical development for new TB therapies usually starts with Phase I trials. Phase I trials are often conducted in healthy volunteers and no mycobacterial information is collected, meaning that the focus is on pharmacokinetics (PK) and safety. Following the phase I trial, early bactericidal activity (EBA) trials are conducted (phase IIa trials) which study the drug in monotherapy or in combination with other drugs over 14 days (n=15 per arm). This is the first time data is collected to measure if the drug has activity against TB in humans (32), and is close to what is referred to as proof-of-concept trials. Activity is determined by solid or liquid culture based on sputum from patients. However, it is common that patients have lesions in the lungs which may act as a bacterial reservoir. Although culture conversion has occurred (negative culture is not uncommon to happen after 8 weeks using SoC), the patient is not cured. This highlights the problems with EBA trials and subsequent phase IIb trials that are conducted over 8 weeks in 50-100 patients/arm (33). EBA trials are not associated with efficacy and therefore should not be translated to the conventional proof-of-concept trial. This has been observed with drugs such as pyrazinamide and clofazimine which show none or limited EBA using traditional statistical analysis methods, but are essential parts of the treatment guidelines for TB (12,13,34,35).

As economic incentives historically have been a limiting factor for TB drug development, EBA trials have been conducted to guide decision making. Although not informative on the efficacy of the drug or combination of drugs, it can still be useful to inform dose-finding phase IIb trials that are significantly longer and costlier and aim to establish the dose level or combination with highest potential for efficacy (36,37). Based on the proportion of patients with culture negativity at 8 weeks, a phase III program can be initiated where the primary efficacy endpoint is relapse free cure. These trials are conducted over months to years and include approx. 500 subjects per arm (33,38). Due to the difficulties that arise in absence of a measurement of clinical benefit in early development, novel trial designs are discussed to mitigate some of the problems (37).

The final endpoint of interesting for novel HA/HB treatments depend on if the treatment is prophylactic or intended to be given on-demand. For novel prophylactic treatments, the primary efficacy endpoint is annualized bleeding rate (ABR), defined as the number of total bleeding events per year per patient (39). As current prophylactic treatments in general perform well with respect to ABR, it is common to aim for non-inferiority to current prophylactic standard or superiority to on-demand treatment. Another efficacy endpoint that aims to study hemostatic effect during on-demand treatment is a predefined 4-point scale for measurement of the response (40,41). The predefined scale describes the improvement of bleeding symptoms (poor, fair, good and excellent) and is usually determined by both the patient and treating clinician. Each bleeding event is treated and dependent on the response at given time points after the first dose of the investigational drug, additional dosing may be performed. Poor and fair improvement is usually considered treatment failure whereas good or excellent improvement is considered treatment success from a clinical point of view. From a conventional statistical point of view, comparison can be made to the current standard-of-care (SoC) in proportion with treatment success at a given timepoint after the bleed (commonly 24 hours). Both ABR and the 4-point clinical outcome scale for on-demand treatment suffer from subjective perception of symptoms and more objective endpoints had been discussed, such as factor activity levels (39), although they are associated with large inter-individual variability (IIV). However, to use factor activity levels as a surrogate endpoint, the relationship between exposure and bleeding outcome need to be scrutinized for the novel product.

A conventional drug development program starts with a first-in-human phase 1 study to investigate the new drugs safety and PK in healthy volunteers. This is followed by phase 2 studies in which proof-of-concept and safety is the focus and finding the dose to be tested in phase 3 is the main goal. Then, phase 3 trials are performed in an inferential matter to determine efficacy and monitor adverse events in a larger sample size and often over longer time. In drug development programs for HA/HB, it is not uncommon that patients are enrolled already in phase I and that efficacy data such as ABR is already accumulated when studying PK and safety (42,43). The rarity of the disease has led to merging of the phases of clinical drug development and led to combination of phases such as phase I/II (42,43) and phase II/III (44) trials. For MarzAA, prophylactic efficacy was demonstrated in phase II (26) but the development program was shifted towards a SC alternative for acute bleeds, as the current SoC is given by the IV route (NovoSeven ®, Feiba ®). Although MarzAA was not been studied in an on-demand setting in a proof-of-concept study, a registrational phase III trial was initiated (planned enrollment of 60 patients) and partially executed in 18 patients (27). For DalcA, the phase I/IIa (45) and the phase IIb (46) trials enrolled in total 17 patients with HB, and the

phase IIb trial aimed to study if the patients factor activity levels can be moved from severe phenotype (<1 % of normal levels) into mild HB (>10 % of normal levels). Although the trial was conducted in a limited sample size and during a short duration of 28 days, it informed on the potential of SC DalcA in elevating FIX levels into mild HB. The current guideline from EMA state that clinical efficacy for FIX replacement products should be studied for 50 exposure days, a design which was followed when demonstrating prophylactic efficacy for SC MarzAA. The rarity of the disease introduces challenges in recruitment, leading to small sample sizes which make development of hemostatic drugs difficult. In particular, this becomes a problem for the pediatric drug development program.

Challenges

The main challenge and aim of drug development after the molecular structure is set and a candidate drug has been selected is dose or regimen selection. As the clinical phases of drug development that aim to find the dose are overlapping and a more varied type of information is collected early for drugs in development against HA/HB, powerful analysis methods that can integrate different data are needed. Novel recombinant coagulation factors exhibit a large IIV in PK and is important to handle during analysis as factor activity levels may act as a surrogate for efficacy (39). In addition, as recruitment in general is a problem, in particular for pediatric programs, extrapolation of adult information to children is essential. Furthermore, as accumulated data come from small sample sizes, it is important to make use of as much evidence as possible during analysis. For drug development against TB, EBA trials can be informative on the design for significantly longer and costlier phase IIb trials. Therefore, it is important to analyze EBA trials in an informative way with respect to detecting EBA at all or differences in EBA between treatment arms.

Model-informed drug development

Model-informed drug development (MIDD), or initially referred to as model-based drug development (MBDD) have been described in the literature as development and application of pharmaco-statistical models of drug efficacy and safety data from preclinical and clinical phases to improve drug development and decision-making (47). The importance of integrating different data arising in drug development was however pointed out earlier than MIDD/MBDD was coined as a concept (48). Also, in 1994, Lewis Sheiner described a *Learning versus Confirming* paradigm to improve how we perform clinical drug development (49). The paradigm described that each phase of the development program should focus on being either *learning* or *confirming* oriented, and that model-based methods are particularly important when focus is on *learning*.

Today. MIDD and application of modelling and simulation is being encouraged and supported by regulators (50), in particular for rare diseases which suffer from small sample sizes (51). MIDD covers many types of approaches such as more mechanistic "bottom-up" methods being physiologically based pharmacokinetic modelling and quantitative systems pharmacology, however, the work in this thesis utilize population PK and pharmacodynamic (PD) methods which are referred to a "top-down" approaches, also called pharmacometric models.

Pharmacometrics

Pharmacometrics has been described as "*the science of developing and applying mathematical and statistical methods to characterize, understand and predict a drugs PK, PD and biomarker outcomes behavior*" (52). The PK of a drug can be viewed as what the body does to the drug (absorption, distribution, metabolism, excretion) whereas the PD can be viewed as what the drug does to the body. Pharmacometrics models can be built to describe both components independently of each other, but also ultimately how drug PK relates to the PD (PKPD). In addition, these models can incorporate several endpoints across different studies longitudinally, which may lead to a higher statistical power when making inference (53–55) on drug effects and when studying potential covariate effects. The built models can subsequently be used for clinical trial simulations for interpolation and extrapolation outside the observed data to inform future study designs and dose selections. As drug PK and PD are associated with variability between individuals and within individuals, mixed effects models that can quantify the magnitude of variability in parameters of interest are informative.

Nonlinear mixed effects models

Pharmacometrics models are primarily but not exclusively non-linear mixed effect models (NLMEM). These models enable description of the overall trend of a dependent variable (such as concentrations of a drug, biomarker concentrations, or a clinical endpoint) using fixed effects parameters (also referred to as typical parameters) and variability in data using random effects to describe distributions of parameters. The random effects are stochastic model components which often are estimated on a variance scale and can consist of several levels. A common division of random effects is i) variability in parameter estimates between individuals (IIV); ii) variability in parameter estimates between occasions within the same individual (IOV); and iii) residual unexplained variability (RUV) which includes remaining errors of measurements arising from analytical assay errors or model misspecification. Mixed effects modelling is beneficial when analyzing longitudinal data and is suitable to analyze rich, unbalanced or sparse data as all data is analyzed simultaneously

and information is "borrowed" between individuals. The mathematical definition of a continuous data NLMEM (such as a population PK model) is:

$$y_{i,j} = f\left(t_{i,j}, \theta, \mathrm{X}_i, \eta_i, \kappa_{i,m}\right) + \epsilon_{i,j} \quad (1)$$

where the dependent variable y at the j^{th} observation for individual i is modelled by a function f which defines the individual parameters. f is described by independent variable t, a vector of structural population (or typical) parameters θ, subject η_i or occasion m $\kappa_{i,m}$ specific random effect parameters and subject specific individual characteristics X_i (for instance covariates and/or dose). $\epsilon_{i,j}$ describe the RUV of the model and is the model component that describe the difference between the individual predictions and observed values. In the equation above, the RUV is additive, normally distributed with mean 0 and variance σ^2. However, RUV models can vary but the most commonly applied ones are additive, proportional (to *f*) or a combination of both. A common distribution to describe individual parameters is the log-normal distribution as it ensures positivity of the parameter in question. Such a parameterization can be defined as:

$$\theta_i = \theta \times e^{\eta_i} \quad (2)$$

where θ_i is the log-normally distributed individual parameter arising from the population parameter θ, and normally distributed random effect parameter η_i with mean 0 and variance ω^2. Other implementations of the random effects can be applied which result in a different distribution of the individual parameter (56). In addition to applying mixed effects models to describe continuous type of data, they can be used with the same benefits to analyze discrete-type data. An example of such a model is presented in Paper VI.

Pediatric drug development

Historically, many drugs were not adequately studied in pediatric subjects which led to uncertainty about the new drugs safety and efficacy in this vulnerable population. Today, regulators require submission of a pediatric study plan (PSP [FDA]) (57) or a pediatric investigation plan (PIP [EMA]) (58) after the end of a phase II meeting or before the first-in-human trial. The documentation should address the therapeutic need in this population and if a need is identified, a strategy outlined to ensure development of the drug to be used in pediatric subjects. The plan should outline details about study design, endpoints of interest and details on statistical analysis of planned studies.

Most drugs are first studied in adults. Based on adult evidence generated from early clinical development, model-based methods can aid in informing the first-in-pediatric clinical trial (59,60). From a PK point of view, population

PK models coupled with allometry can be applied to scale PK parameters estimated in adults to expected parameters in children (61). The following exemplifies allometric scaling for CL:

$$CL_i = CL_{std} \times \left(\frac{BW_i}{BW_{std}}\right)^b \quad (3)$$

where CL_i is the individual CL in individual i and CL_{std} is the standardized CL in an individual with bodyweight BW_{std}, often set to 70 kg. BW_i is the bodyweight of individual i and b is the exponent used for scaling. Different scaling methods have been presented in the literature (62–66). The most common method in particular for small molecules is scaling using bodyweight with fixed exponents for clearance (CL) and volume of distribution (V) terms. CL terms are usually scaled with an exponent of 0.75 whereas V terms are scaled with an exponent of 1. When scaling parameters below 2 years of age, the general recommendation is to account for maturation effects in metabolic pathways (67). However, in absence of data on ontogeny effects, different methods has been described in the literature to improve the prediction in younger children (63,67). For coagulation factors FVIII and FIX, allometric scaling using fixed exponents has been described to provide adequate description of PK down to 5 years of age (68). Although these methods are helpful and commonly applied in model-based frameworks, they are not intended to replace clinical trials but to inform dosing when initiating a clinical trial involving pediatric subjects.

It is not uncommon that safety and efficacy can be informed in pediatrics when performing extrapolation of PK information from adults. The planned adult exposures that balance safety and efficacy can be leveraged with some key assumptions. If the underlying disease mechanism and the exposure-response relationship can be assumed to be sufficiently similar, a full extrapolation approach, i.e. an exposure matching approach can be targeted (69). In such an approach, the extrapolated PK parameters in different age groups (or bodyweight groups) of children can be used to simulate different doses to inform the pediatric program. Instead of a dedicated PK trial in children, a run-in phase can be performed where the extrapolations are confirmed or updated, before initiation of efficacy studies in children. As data become available in children, the assumptions can be reevaluated and the pediatric exposure-response relationship can be established or confirmed. The application of pharmacometrics approaches are particularly helpful for rare diseases that suffer from difficulties in recruitment and setting an informed first-in-pediatric dose is essential.

Aims

The overall aim of this thesis was to develop and apply pharmacometric approaches to support dose selection and to inform study designs in clinical development for drugs against hemophilia and tuberculosis.

The specific aims were:

- To present a standardized model-based approach including power calculations to determine sample size needed to detect activity and differentiate between treatment arms activity for drugs in development for treatment of tuberculosis

- To characterize the early bactericidal activity of meropenem plus clauvulanate with or without rifampicin against tuberculosis including evaluation of covariate relationships

- To integrate preclinical and clinical pharmacokinetic and potency data of a novel hemostatic drug and characterize the population pharmacokinetics to support dose selection in a pivotal phase III trials against acute bleeds in adult and pediatric HA/HB patients with inhibitors

- To characterize the population pharmacokinetics of a novel subcutaneous prophylactic treatment for HB based on sparse clinical data to support dose selection in adult and pediatric subjects

- To characterize clinical response following treatment of acute bleeding episodes and assess difference between a novel subcutaneous treatment versus intravenous standard-of-care using a continuous-time Markov model

- To evaluate different model-based approaches to prospectively power pediatric pharmacokinetic trials in hemophilia

Methods

Study designs and data

The analyzed data in this thesis was a combination of clinical trial data and simulated data. Based on observed data, models were developed (Papers II, III, V-VI) that were used for simulation (Papers I-VII). An overview of all papers is presented in Table 1.

Table 1. Purpose, task, disease and model type for all papers included in this thesis.

Paper	Purpose	Usage	Disease	Phase	Model
I	Development of a model-based framework for analysis of EBA trials	S	TB	IIa	PD, PKPD
II	Inferential application investigating EBA of meropenem	M&S	TB	IIa	PD
III	Dose selection in a pivotal trial in absence of proof-of-concept hemostatic data	M&S	HA/HB	I-III	PK
IV	Pediatric dose selection through exposure-matching based on adult information	S	HA/HB	III	PK
V	Adult and pediatric dose selection based on FIX target guidelines	M&S	HA/HB	I-III	PK
VI	Inferential investigation of clinical efficacy of a SC administered on-demand treatment for acute bleeds	M&S	HA/HB	III	PKPD
VII	Optimization of study design and investigation of pediatric PK trial analysis approaches	M&S	HA/HB	I-III	PK

M&S; modelling and simulation, M; modeling, S; simulation, HA/HB; hemophilia A/hemophilia B, TB; tuberculosis, PK; pharmacokinetic, PD; pharmacodynamic, PKPD; pharmacokinetic-pharmacodynamic

The study design and data information are detailed hereafter for all papers handling clinical trial data. For projects that were simulation studies, the corresponding information are detailed in the clinical trial simulation section.

Meropenem early bactericidal activity data (Paper II)

Individuals aged between 18 to 65 years with confirmed pulmonary TB were enrolled in this single site phase IIa study (NCT03174184) to study the PK, adverse events and EBA during 14 days of treatment with i) meropenem 2 g thrice daily (TID) plus oral rifampicin 20 mg/kg once daily (OD [arm C]); ii) meropenem 2 g TID (arm D); iii) meropenem 1 g TID (arm E); iv) meropenem 3 g OD (arm F). Patients in C-E received oral amoxicillin/clavulanate 500 mg/125 mg TID whereas arm F was dosed 875 mg/125 mg OD with each meropenem infusion. The sample size per arm was 15 subjects. Adverse events were collected daily by the study investigators and blood samples were collected at screening and on days 7, 14 and 28 for laboratory safety assessment. Mycobacterial assessments were sputum based and done on days 1, 2, 3, 4, 6, 8, 10, 12 and 14. Sputum samples were treated and inoculated on 7H11S agar plates to determine CFU counts on solid culture. In addition, samples were incubated in the BD Bactec MGIT 960 Mycobacteria Culture system and TTP was recorded in hours. Blood samples were collected for PK assessment on the 13th day of study conduct pre-dose and on 0.5, 1, 1.5, 2, 3, 4, 6- and 8-hours post-dose. For the participants that received rifampicin, two extra samples at 12- and 24-hours post-dose was collected. For analytical assay details, readers are referred to the methods section of Paper II.

Marzeptacog alfa (activated) pharmacokinetic data (Paper III)

MarzAA concentration versus time data (n=46 adult males providing a total of 1225 observations) was obtained from three previously conducted trials (Table 2). In NCT01439971, a single IV dose (0.5, 4.5, 9, 18 or 30 µg/kg) was given. Blood samples for PK assessment were taken pre-dose, and at 1, 3, 6, 9, 12, 24 and 48 hours after the dose. Only one subject received the 0.5 µg/kg dose level and was therefore excluded from the analysis. In NCT04072237, a single IV dose was followed by ascending SC doses. The dose levels were 18 (IV), and 30, 45, 60, 60 (2 x 30 at two anatomical sites), 90 and 120 µg/kg. In a separate stage, 60 µg/kg two times 3 hours apart and in a separate stage, 60 µg/kg three times 3 hours apart was given. Blood samples for PK assessment was obtained at pre-dose and at 5 minutes, 1, 2, 6, 9, 12, 24 hours after the IV dose. For the SC doses, samples were obtained at pre-dose and then at 0.5, 1, 1.5, 2, 6, 9, 24 and 48 hours after the dose. For the last two stages, the sampling schedule was pre-dose and then at 2, 3, 4, 5, 6, 7, 8, 9, 12, 24, 48 and 72 hours after dose. Patients in NCT03407861 received a single IV dose of 18 µg/kg and sampling taken at 5 minutes, 0.5, 1, 3, 6, 9,

12- and 24-hours post-dose. Each subject was subsequently dosed 30 µg/kg SC after a wash-out period of 24 hours after the last sample taken after the IV dose. For the SC doses, samples were taken pre-dose and at 3, 5, 7, 9, 12, 24, 30- and 48-hours post-dose. As a second part of the study, SC MarzAA was given OD (30 µg/kg) for 50 days and samples were taken at pre-dose followed by 7 hours on days 1, 3, 5, 7, 14, 21, 28, 50 and at the end of the study. As part of the protocol, if an individual experienced a bleed during the 50 days, the dose was escalated to 60 µg/kg OD and followed for an additional 50 days. The number of observations per dose level, trial and the number of subjects per dose group can be found in Table 2.

Table 2. Number of observations per dose level and trial and subjects per dose group

Dose level (µg/kg)	4.5[a]	9[a]	18[a]	30[a, b, c]	45[b]	60[b, c]	90[b]	120[b]	Total
NCT01439971	72	72	72	72	-	-	-	-	288
NCT04072237	-	-	63	74	68	344	67	67	683
NCT03407651	-	-	81	150	-	23	-	-	254
Total observations	72	72	216	296	68	367	67	67	1225
Total subjects	6	6	26	23	8	12	8	8	46

[a]Intravenous dose levels
[b]Subcutaneous dose levels
[c]Multiple subcutaneous dosing

A total of 19 observations were reported as below the limit of quantification (BLOQ), which equaled 1.6 % of all observations and was therefore removed from the analysis together with an additional three samples that had missing timing information or due to be during unscheduled visits. Median and 10-90th percentile of bodyweights were 75 and 60-94 kg, respectively. The corresponding values for age was 32 and 23-52 years, respectively. For details on the analytical assay, readers are referred to the Methods section of Paper III.

Dalcinonacog alfa pharmacokinetic data (Paper V)

DalcA concentration versus time data (21 males providing 303 observations) was obtained from two separate clinical trials in severe HB patients. The phase I/II dose escalation trial (NCT03186677) was conducted in five cohorts. Cohort 1 was dosed a single IV dose of 70 IU/kg whereas cohorts 2-3 were dosed IV doses of 70 IU/kg with subsequent SC dosing (70 and 140 IU/kg, respectively) 4 days after the IV dose. In cohort 4, daily SC dosing was performed for 6 days (140 IU/kg) whereas patients in cohort 5 was dosed a single IV dose of 70 IU/kg followed by 8 days of SC dosing of 140 IU/kg. For each IV dose, samples were taken for PK assessment pre-dose and 0.25, 0.5, 1, 3, 6, 9, 24, 48 and 72 hours after the dose. For the SC doses, samples were taken pre-dose and at 1, 2, 4, 6, 8, 10, 12, 24, 48 and 72 hours after the dose. In cohort 5, samples were drawn pre-dose and 6 hours post the first injection, after the 6th injection and at 24 hours after the dose for doses given after the 6th injection. The second trial (a phase IIb trial [NCT03995784]) are referred to as cohort 6. In cohort 6, a single IV dose of 50 IU/kg was given followed by 28 days of 100 IU/kg SC OD dosing. A single individual received a 150 IU/kg IV dose on the first day and another individual received a lower SC dose (50 IU/kg) during the first 2 injections. PK assessment was performed based on samples taken pre-dose on days 2, 3, 7, 14, 21 and 28 and washout sampling was performed at 24, 48, 72, 96 and 120 hours after the last dose and at the end of the study.

Only 2.9 % of the observations were reported as BLOQ. In the elimination phase, the first BLOQ sample was set to the lower limit of quantification divided by 2 whereas the rest of the samples were omitted. In the absorption phase, the opposite was applied. The median and 10-90th percentile bodyweight was 67 and 61-81 kg and the corresponding values for age was 43 and 26-57 years, respectively. For details on the analytical assay, readers are referred to the Methods section of Paper V.

Marzeptacog alfa (activated) clinical outcome data (Paper VI)

Clinical outcome efficacy data and non-bleeding PK data were received from a registrational phase III clinical trial (NCT04489537) in 15 patients. Each subject was randomized to sequence A or B. In sequence A, the subjects were treated on-demand for each bleeding event with 60 µg/kg SC MarzAA in 3-hourly intervals. For each bleeding event in each subject, the clinical investigator measured improvement of the bleed with a 4-point scale corresponding to poor, fair, good or excellent improvement in signs of bleed. The efficacy measurements were performed at 1, 3, 6, 9, 12, 24 and 48 hours after the first dose. Additional dosing was motivated by poor or fair response at 3 and 6 hours after the first dose, allowing for up to 3 doses in total. This was followed by treatment with IV SoC (Feiba, Coagil VII or NovoSeven), allowing for 1-

3 doses according to the dosing recommendations. In sequence B, the subjects followed the reverse order in which a SoC treatment period was followed by SC MarzAA treatment period.

In total, 66 evaluable bleeding events were studied for drug effect evaluation of SoC and SC MarzAA. On average, subjects experienced 5 bleeds with a 90 % inter-percentile range of 1-8 bleeds. The efficacy measurements (poor, fair, good or excellent improvement) were 222 and 265 for SC MarzAA and IV SoC, respectively. The median and (minimum-maximum) of time to cessation of bleeding, time from bleed recognition to initial dosing and number of doses per bleed was 9.0 (1.0-36.0) and 6.0 (0.8-61.0) hours, 0.75 (0.2-10.3) and 1.0 (0.0-59.0) hours and 2.0 (1.0-5.0) and 1.0 (1.0-4.0) doses, for MarzAA and SoC, respectively. Only one subject that was treated with MarzAA developed low-titer cross-reactive non-neutralizing anti-drug antibodies (ADAs), but was resolved at end of study. One subject in the MarzAA and one subject in the SoC group needed rescue therapy. Rescue therapy was defined as additional doses after 24 hours post the initial dose due to inadequate hemostasis. No injection site reactions, drug-related adverse events or thrombotic events were reported. As only 4 bleeding PK observations were available for SC MarzAA, non-bleeding PK information was used from this study.

Ethics

All clinical trials that were analyzed in this thesis were conducted in accordance with the Declaration of Helsinki, US Code of Federal Regulations and International Conference on Harmonization Guidelines on Good Clinical Practice and each study was approved by local institutional review boards or independent ethics boards at each study site. All participants provided written informed consent prior to initiation of each study. All participants had the right to drop out from each of the studies upon request. Papers I and IV were simulation studies and as such, ethical permission was not needed. In Paper VII, clinical trial data were re-used from Paper III and V but was covered by the same ethical clearance.

Model development

All papers of this thesis that included model development are covered in this section. The methodology of papers that were pure simulation studies will be detailed in the clinical trial simulations section.

Early bactericidal activity model development (Paper I-II)

In Paper I, only a TTP model was developed and was a simulation study that will be further detailed a coming section. In Paper II, separate EBA models were developed for CFU and TTP data. For each endpoint, mono- and bi-exponential models were fitted. Below follows an example for CFU data using a mono-exponential model:

$$log_{10}CFU = log_{10}\left(e^{A_{mono}} \times e^{-\alpha \times t}\right) \quad (4)$$

and a bi-exponential model:

$$log_{10}CFU = log_{10}\left(e^{A_{bi}} \times e^{-\alpha \times t} + e^{B_{bi}} \times e^{-\beta \times t}\right) \quad (5)$$

where in each model, one or two intercepts are defined by A and/or B and one or two slopes defined by α and/or β. CFU data were modelled using negative slope(s) whereas TTP data were modelled with positive slope(s). If clear model misspecifications from these structural models were detected, another or additional structural elements were considered. IIV was tested in relevant parameters using an exponential parameterization. If distributions of random effects were non-normal, transformations were considered. Negative data (labelled as no growth by the lab) for CFU was set to 0 (left censoring) and included in the analysis dataset. The corresponding data for TTP (labelled as negative at day 42) was set to 1008 hours (42 days) and considered right censored. The RUV models consisted of a combined structure of common and replicate specific error term, for each of the two technical replicates for each timepoint. CFU data was modelled using log-transformation and an additive error on the log scale was used. For TTP, additive, proportional and a combined RUV model was evaluated.

The best structural model was taken forward to the covariate analysis. The covariate analysis was informed by scientific plausibility in agreement with the clinical investigators. For intercept parameters (baseline bacterial load), sex, age, weight, height, body mass index (BMI), ethnicity, human immunodeficiency virus (HIV), category of cavity and extent of disease were tested. For slopes (bacterial decline), sex, age, weight, height, BMI, ethnicity, HIV, category of cavity, extent of disease and mean observed baseline bacterial load were tested. The covariates were tested through the stepwise-covariate-modelling tool (SCM) (70) using a significance level of 0.05 for inclusion of a covariate and 0.01 for the backward elimination step. Continuous covariates were implemented with a power function and categorical covariates were implemented as a fractional difference from the most common category. Other parameterizations were considered if indicated by the graphical exploration. The best covariate model was taken forward to evaluate regimen effects.

Evaluation of regimen effects were performed in a stepwise manner. Initially, each regimen was tested against no change from baseline to ensure statistically significant EBA. Arms with a statistically significant EBA were taken forward into a univariate search where a separate slope was tested for each arm. The univariate model with most improvement in fit from having a separate slope estimated was taken forward and in the next step, the second most significant arm specific slope was tested for. This procedure was repeated until not additional significant difference in slopes could be detected, using a significance level of 5 %.

Pharmacokinetic and safety data analysis

Paper II was an analysis of a clinical trial that collected PK and safety information in addition to EBA data. The PK information was analyzed at steady state using non-compartmental analysis (NCA) followed by descriptive statistics (median and range) per arm, but only for meropenem. The NCA analysis was performed using Phoenix WinNonlin version 8.1. Safety data was analyzed by summarizing adverse events of grade 2 or higher in a tabular format (number of events and proportions) per arm. Neither the PK or the safety data was analyzed using model-based pharmacometric methods.

Pharmacokinetic model development (Paper III and V)

In general, one-, two- and three-compartmental models were explored. For both MarzAA and DalcA, patients were expected to have endogenous levels of FVIIa and FIX. The observations were not baseline corrected, and as such, baseline levels were modelled using a separate parameter according to:

$$IPRED = BASE + \frac{A}{V_c} \quad (6)$$

in which IPRED is the individual prediction of the concentration, BASE is the baseline levels of endogenous FVIIa or FIX, respectively, A is the amount in the central compartment and V_c is the estimated volume of distribution in the central compartment. For both MarzAA and DalcA, allometry was included on att CL and V terms with fixed exponents of 0.75 and 1, respectively.

Marzeptacog alfa (activated)

The structural model was developed using only IV data to first capture the PK independent of absorption processes. After choosing the structural model, IIV was tested in all relevant parameters followed by testing for covariance between IIVs. Michaelis-menten elimination and nonlinear volume of distribution was tested for. The best model was taken forward and all parameters were fixed in the next step where data following SC administration was included. Different absorption models such as first-order, zero-order and sequential

zero-order followed by first-order models were evaluated, followed by IIV testing in all absorption parameters. As SC data was obtained from several occasions of dosing within an individual, IOV was tested for in all parameters. Subsequently, all parameters were unfixed and re-estimation of all parameters were done given all the data. The developed base model was taken into the covariate search using SCM, with a significance criterion of 0.05 for the forward inclusion and 0.01 for the backward exclusion test. Age was tested on all parameters whereas bodyweight was tested on all parameters but CL and V terms as they already included bodyweight through allometry. In addition to statistical significance, a covariate was only kept in the model if the effect on the maximum concentration (C_{max}) was over 30 % for the 10^{th} and 90^{th} percentile of the covariate. Different RUV models (proportional and combined additive and proportional) were tested for the base model and before concluding the final model.

Dalcinonacog alfa

Before the structural model optimization, a test was performed in which a simple one-compartment model with IIV in CL (and first-order absorption when SC data was included) was estimated on IV, SC and IV plus SC data simultaneously. Parameter estimates were compared and it was concluded that IV and SC data can be modelled simultaneously without biasing CL and V estimates. The chosen structural model was taken through an IIV optimization step followed by testing of nonlinear elimination and lastly, testing of IOV. All identified stochastic parameters were re-evaluated in a backward deletion step after updating of initial estimates. The established base model was moved into the covariate model building step using SCM with the same criterion as applied for MarzAA. Trial and age were tested on all parameters whereas bodyweight was tested on all parameters but CL and V terms.

Clinical bleed outcome model development (Paper VI)

The outcome data was treated as an ordinal categorical variable. A four-state continuous time Markov model (71–73) as well as a simplified two-state model were developed. For the four-state model, each state represented the probability of each possible efficacy measurement (poor, fair, good and excellent), whereas in the two-state model poor and fair was pooled to treatment failure (TF) meanwhile good and excellent responses were pooled and defined as treatment success (TS). At the time of start of each bleeding event, the probability in the poor state for the four-state model and the probability in the treatment failure state for the two-state model was set to 1 whereas all other states were set to 0. A set of differential equations using first-order transfer rates was used to describe transfer of the probability between the states. When an observation was made, the probability was reset in all compartments but

the compartment of the observed state which was set to 1 (100 % probability). The four-state model differential equation system were defined as follows:

$$\frac{dPr(P)}{dt} = k_{FP} \cdot \Pr(F) - k_{PF} \cdot \Pr(P) \quad (7)$$

$$\frac{dPr(F)}{dt} = k_{PF} \cdot \Pr(P) + k_{GF} \cdot \Pr(G) - k_{FP} \cdot \Pr(F) - k_{FG} \cdot \Pr(F) \quad (8)$$

$$\frac{dPr(G)}{dt} = k_{FG} \cdot \Pr(F) + k_{EG} \cdot \Pr(E) - k_{GE} \cdot \Pr(G) - k_{GF} \cdot \Pr(G) \quad (9)$$

$$\frac{dPr(E)}{dt} = k_{GE} \cdot \Pr(G) - k_{EG} \cdot \Pr(E) \quad (10)$$

in which $Pr(P)$, $Pr(F)$, $Pr(G)$ and $Pr(E)$ describe the probability of poor, fair, good and excellent improvement at every timepoint. The transfer rates are defined with subscripts that represent current category (first letter) and the direction of movement (second letter). This structure was used as a starting point from which additional models were tested for, such as direct movement from poor to good, or sharing of transfer rates. The simplified two-state alternative modelling approach was defined as following:

$$\frac{dPr(TF)}{dt} = k_{TSTF} \cdot \Pr(TS) - k_{TFTS} \cdot \Pr(TF) \quad (11)$$

$$\frac{dPr(TS)}{dt} = k_{TFTS} \cdot \Pr(TF) - k_{TSTF} \cdot \Pr(TS) \quad (12)$$

in which $Pr(TF)$ and $Pr(TS)$ represented the probability of observing TF or TS. All bleeding events were assumed to start in TF and two transfer rates were estimated between the states. IIV and inter-bleed-variability (IBV) was evaluated in all transfer rates.

Covariate and exposure-response evaluation

The chosen base models were taken forward into the covariate search using the same SCM procedure and statistical criterions as previously described. Tested covariates were severity of bleed (each bleed was classified to be mild, moderate or severe), location of bleed (knee, ankle, elbow and other [buttock, forearm, foot, hip, hand, leg, eye, chest, ear, mouth, nose, neck, shoulder, stool, throat, vaginal or wrist]), heart rate, systolic and baseline diastolic blood pressure (BDBP), respiratory rate, age, weight, and treatment group (MarzAA/SoC). The best covariate model for both modelling approaches was taken forward for exposure-response evaluation for SC MarzAA.

Using the population PK model developed in Paper III, PK metrics were derived through Bayesian *post hoc* estimation based on non-bleeding PK information. Single dose area under the curve over 48 hours ($AUC_{0\text{-}48h}$), maximum concentration over 48 hours ($Cmax_{0\text{-}48h}$), minimum concentration over 48 hours ($Cmin_{0\text{-}48h}$), time until maximum concentration over 48 hours ($Tmax_{48h}$), individual clearance estimates (ICL), individual bioavailability estimates (IF),

individual absorption rate (Ik_a) and individual duration of zero-order release of the drug (I_{dur}). Continuous covariates were tested through linear and piece-wise linear functions and categorical covariates were tested through linear-proportional inclusion (one additional parameter per category). Exponential and power relationships between covariates/exposure metrics and transfer rates were not considered as the relationships were tested on a logit scale. The included covariates were subsequently evaluated for their effect size on a typical level for all possible combination of covariate effects to visualize how the covariate effects impact the probability of treatment success using both the four-state and two-state modelling approach. A lower and higher value relative to the median was tested for continuous covariates. If the continuous covariate was described by a piece-wise linear model, relevant values in relation to when the slopes changed was tested. For BDBP, 70 and 90 mmHg were used to study the effect of low and high BDBP in relation to 80 mmHg which was considered a normal BDBP. For the effect of bleeding location on the probability of treatment success, the difference between elbow/ankle joints versus knees and other joints were studied. In the two-state model, the minimum and maximum bodyweight in the data was used whereas for BDBP, the same approach as for the four-state model was implemented.

Model selection

From a model building point of view, model selection is a complex process that involves consideration of many aspects. To ensure adequacy in clinical trial simulations and validity of inference, a range of graphical and numerical criteria were applied in parallel to considering scientific plausibility. Different diagnostic plots come with different strengths and weaknesses (74) which have been considered in each project in accordance with regulatory guidelines on model diagnostics (75). The likelihood ratio test has been applied to compare nested models in all projects, where a difference in objective function value (OFV) of 3.84 was considered to correspond to a statistical significance level of $p<0.05$ for one degree of freedom. Graphical inspection of different goodness-of-fit plots based on model predictions and observations, residual plots and simulation-based plots such as the visual predictive check (VPC) and the prediction-corrected visual predictive check (pcVPC) (76) have been used in all projects. In addition, estimated parameter precision was considered for model selection. The relative weight put on these criterions were situation dependent.

Clinical trial simulations

A number of clinical trial simulation were performed in this thesis. In Paper I, the model developed in Paper II and another model based on unpublished data

(NCT04629378) were used to visualize the standardized analysis approach of phase IIa EBA trials and in subsequent sample size determinations. In Paper III and IV, the developed population PK model for MarzAA was used to perform clinical trial simulations to inform dose selection in adults and pediatric HA/HB subjects (on-demand treatment setting), respectively. In Paper V, the developed population PK model for DalcA was used to inform dose selection in adult and pediatric HB subjects (prophylaxis setting). In Paper VII, both the MarzAA and DalcA models were used in model-based optimization of pediatric sampling schedule for MarzAA and DalcA with subsequent evaluation of different model-based approaches to prospectively estimate important PK parameters (CL and V) in pediatric subjects.

For all clinical trial simulations that were performed in pediatric subjects, individual age and bodyweight combinations were samples from the National Health and Nutrition Examination Survery (NHANES) database (77). To avoid outlying bodyweights, values outside of the 97.5^{th} and 2.5^{th} percentile of bodyweight per age in months were omitted. The NHANES database were also used for adult simulations for DalcA, whereas adult simulations for MarzAA were based on a truncated normal distribution of bodyweights observed in the MarzAA PK dataset.

Model-based early bactericidal activity evaluation and sample size determination (Paper I)

Standardized early bactericidal activity analysis approach

For the visualization of the standardized analysis of EBA trials in Paper I, a similar model as developed in Paper II were reused together with another model that is yet not published but was developed on data from NCT04629378. The structural model and parameters are reported in Table S1 and equation 3 of Paper I, respectively. Four covariates were sampled, being age, sex, cavity extent at baseline (no cavity, cavity <4 cm and cavities ≥ 4 cm on baseline TTP) and meropenem area under the curve from time 0 to infinity ($AUC_{0\text{-}inf}$). $AUC_{0\text{-}inf}$ values were generated from a normal distribution with mean 640 h mg/l and standard deviation of 86 mg/l (unpublished data). Age was sampled from a truncated normal distribution with mean 33 years, a lower boundary of 18 years and an upper boundary of 60 years with a standard deviation of 13 years. The simulation model was used to simulate TTP values in two arms A and B and $AUC_{0\text{-}inf}$ was included on the TTP slope and cavity extent was included on the intercept as per Table S1 in Paper I. For realistic TTP value simulations, 2 % of the samples were set randomly as negative at day 42, missing and contaminated, respectively. A proportional RUV was applied, with a common component between samples and a component that was replicate-specific.

Using the simulated data, the data exploration was performed to identify extreme TTP values. This procedure should be performed in close collaboration with clinicians, microbiologists and data management teams to ensure data quality. Replicate versus replicate (one replicate on the x-axis and the other replicate on the y-axis) and biomarker versus biomarker (e.g. if CFU and TTP data is analyzed) plots was constructed to identify outlying replicates and observations. Population and individual plots of the biomarker over time (also stratified by covariates) can help confirming outliers identified in the previous plots and inform on what structural model to use and potential covariates. A thorough descriptive statistical summary of all data should then be performed including summary of censored observations.

After establishing the analysis dataset, base model development is performed as presented in the previous section on model development of EBA models for meropenem. The example for meropenem is shown for CFU data but applies to TTP data as well, but with the slopes being positive. The chosen base model is then moved into the covariate search where the goal is to explain previously unexplained variability and identify important covariate effects. Nowadays, most EBA trials have relatively similar covariates being collected. A representative set is described in the previous section for covariate model development of meropenem, including how they are used in the covariate search. The covariate model is then used to test if each arm shows EBA i.e. testing if a model with estimated slope is statistically significantly better ($p<0.05$) than a model with slope fixed to 0. For arms with no EBA, the biomarker slope is fixed to 0 in further model development. The resulting model is taken forward into the PKPD search which can be seen as a step that is similar to the covariate search but using exposure metrics such as AUC or C_{max} to explain variability in the slopes.

Finally, the arm comparison in EBA is performed, as previously described for meropenem. Prior to the univariate stepwise testing, all IIVs but on intercept is set to 0. After all treatment arm specific slopes have been identified, IIVs are re-evaluated in the model.

Sample size calculations to detect early bactericidal activity

For all sample size calculations, Monte-Carlo Mapped Power (MCMP) was utilized (78) with at a significance level of 5 %. The model used for simulation of two arms (n=15 per arm) was a mono-exponential model with separate slopes for each arm and cavity extent on intercept. IIV was included in intercept and on the slope. The reduced model (representing the null hypothesis of no EBA) was a model where the TTP slope is fixed to 0. To study how different slopes and IIV in slope impact the power to detect EBA, the following scenarios were tested (Table 3).

Table 3. Scenarios for calculation of power to detect EBA given different sample sizes using the standardized model-based analysis approach. The different slope values were re-calculated to EBA over 14 days ($TTP\text{-}EBA_{0\text{-}14}$) for communication purposes.

TTP-EBA model	$TTP\text{-}EBA_{0\text{-}14}$ [$SLOPE_{TTP}$]	IIV_{SLOPE}	Scenarios
$BASE_{TTP}$ Cavities ≥ 4 cm = 108 hours[a] Cavities < 4 cm = 131 hours[a] No cavities = 160 hours[a] IIV_{base} = 6%[a] RUV_{common} = 13%[a] $RUV_{replicate}$ = 4%[a]	152 hours [0.0628 hours/day][b]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 152 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 152 hours, IIV_{SLOPE} 104%
	30 hours [0.0174 hours/day][a]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 30 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 30 hours, IIV_{SLOPE} 104%
	25 hours [0.0148 hours/day]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 25 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 25 hours, IIV_{SLOPE} 104%
	20 hours [0.0122]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 20 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 20 hours, IIV_{SLOPE} 104%
	16 hours [0.0098 hours/day]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 16 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 16 hours, IIV_{SLOPE} 104%
	11 hours [0.0069 hours/day]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 11 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 11 hours, IIV_{SLOPE} 104%
	7 hours [0.0044 hours/day]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 7 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 7 hours, IIV_{SLOPE} 104%
	3 hours [0.0017 hours/day]	22%[b]	$TTP\text{-}EBA_{0\text{-}14}$ 3 hours, IIV_{SLOPE} 22%
		104%[a]	$TTP\text{-}EBA_{0\text{-}14}$ 3 hours, IIV_{SLOPE} 104%

$BASE_{TTP}$, IIV_{base}, RUV_{common} and $RUV_{replicate}$ were the same in all scenarios. EBA was expressed as early bactericidal activity based on difference in time-to-positivity between 0 and 14 days ($TTP\text{-}EBA_{0\text{-}14}$). IIV was expressed as a coefficient of variation (CV). A proportional error model was used consisting of a common error (RUV_{common}) and replicate-specific ($RUV_{replicate}$) error for each replicate. [a]Parameter estimate from De Jager et al. 2022. [b]Parameter estimate from unpublished data (NCT04629378).

Sample size calculations to detect treatment arm difference

As in the power calculations for detecting EBA, the MCMP methods was applied for these power calculations as well. Different combination of TTP slopes, difference in TTP slope between the two arms (expressed as percentage higher slope in one arm relative to the other) and IIV magnitude in slope was evaluated for the impact on the sample size calculations. Each scenario is detailed in Paper I. The null hypothesis in these calculations were that there is no difference in EBA between the treatment arms, whereas the alternative hypothesis represented a difference in EBA between the treatment arms.

Marzeptacog alfa (activated) dose selection in adults and pediatrics (Paper III and IV)

For MarzAA, the target range of the simulations were defined as 24-120 ng/ml which was considered equivalent to 6-30 IU/ml wt-FVIIa under the assumption that MarzAA is at least five times as potent than wt-FVIIa. This assumption was derived in accordance with the sponsor data from preclinical experiments that showed 5-10 times higher potency of MarzAA when compared to NovoSeven®. The picked dose regimen for MarzAA in adults was used in an exposure-matching strategy in pediatric subjects, where the aim was to match the exposures arising from the chosen dose level in adults.

Dose selection in adult subjects

MarzAA concentration versus time profiles were simulated in 1000 adults with rich sampling, following 60, 90 and 120 µg/kg given once, twice or thrice. If more than one dose was given, they were given in 3 hourly intervals. For 60 µg/kg, a 6 hourly interval was also studied. Median and 80 % prediction intervals were derived for area under the concentration time curve over 24 hours ($AUC_{0\text{-}24h}$), maximum concentration over 24 hours (C_{max24h}), minimal concentration over 24 hours (C_{min24h}) and time to maximal concentration over 24 hours (T_{max24h}). In addition, the median and 80 % prediction interval of percentage of the population above the target at different timepoints and the time required to reach therapeutic targets were derived.

Dose selection in pediatric subjects

Different dose levels and dosing frequencies were used in the clinical trial simulations to inform dose selection in pediatrics. Initially, the regimens were simulated in large populations (n=1000) per age groups 11, 6, 4, 2, and 0.5-year-old children (Table 4):

Table 4. Dose levels, frequency of administration, number of children and age groups used in all clinical trial simulations for prediction of $AUC_{0\text{-}24h}$ and $C_{max(0\text{-}24h)}$.

Dose (µg/kg)	Dosing interval	Age groups (years)[a]
60	Single dose, two doses 3 hours apart or three doses 3 hours apart	11, 6, 4, 2, 0.5
60	Single dose	<2, 2 to <6 and 6-11[b]
75	Single dose, two doses 3 hours apart or three doses 3 hours apart	11, 6, 4, 2, 0.5
90	Single dose, two doses 3 hours apart or three doses 3 hours apart	11, 6, 4, 2, 0.5
105	Single dose, two doses 3 hours apart or three doses 3 hours apart	11, 6, 4, 2, 0.5
120	Single dose, two doses 3 hours apart or three doses 3 hours apart	11, 6, 4, 2, 0.5

$AUC_{0\text{-}24h}$ is the area under the concentration curve over 24 hours. $C_{max(0\text{-}24h)}$) is the maximum concentrations over 24 hours.

[a]) n=1000 per each age group.

[b]) 2, 10 and 12 pediatric patients in age subgroups <2, 2 to <6 and 6 to 11.

As part of a sensitivity analysis, a putative doubling of absorption rate compared to the rate estimated in adults were implemented to study the effect on dose selection. The doubling in absorption rate constant was studied for the 60, 90 and 120 µg/kg dose levels. In addition, as no information was available on maturational development functions for elimination of FVIIa, a sensitivity analysis on the exponents was performed where the age-dependent (ADE) allometric model was used for CL (63). The ADE model applied an exponent of 0.75 for the 11 and 6-year-old groups, whereas an exponent of 0.9 was used for the 4-year-old group. For younger children in the 2, 1 and 0.5-year-old groups, an exponent of 1 was used.

Based on the chosen dose level in large populations, the probability of trial success was studied. One thousand clinical trials with 24 pediatric subjects (2, 10 and 12 subjects in age groups below 2, 2 to less than 6 and 6 to 11 years of age, respectively) in each trial was simulated following a single dose. The sampling schedule was set as planned for the pediatric program (pre-dose, 2, 4, 8, 12 and 24 hours post dose). Each simulated subject $AUC_{0\text{-}24h}$ and C_{max24h} were compared with the adult prediction interval resulting from the same dose regimen. When deciding if a trial was successful or not, different cutoffs for the definition of successful was tested corresponding to 20, 21 or 22 out of 24 children to be within the adult prediction interval.

Dalcinonacog alfa dose selection in adults and pediatrics (Paper V)

For DalcA, the target in both adults and pediatric subjects was set to the threshold that put severe HB patients into mild HB (0.1 IU/ml) (15,79), independent of age. Different dose levels given as OD (50, 75, 90, 100 IU/kg) and every-other day (EOD [125, 150 IU/kg]) were simulated in 1000 adult subjects over 40 days with rich sampling. Based on the simulated concentration profiles, the percentage of the population above the target was derived at steady state based on trough values ($C_{min,ss}$). Based on the same simulation, the median and 90 % prediction interval was derived for $C_{min,ss}$. Furthermore, the median and 90 % prediction interval for time-to-reach at or above the target was performed for all OD regimens.

The clinical trial simulations in pediatric subjects were performed in 11, 6, 4 and 2-year-old groups with 1000 subjects per age group. A similar setup was used as for adults, but only using OD regimens of 100, 125 and 150 IU/kg followed by construction of median and 90 % prediction intervals for $C_{min,ss}$ in each dose level and age group. A potential dose escalation to 175 and 200 IU/kg was also studied for the subjects that did not reach target steady state levels of FIX.

Approaches for prospectively powering pharmacokinetic trials in pediatric subjects (Paper VII)

Before exploring the different approaches for powering a pediatric PK study, the sampling schedule was optimized for both MarzAA and DalcA using simplified models due to a limited sample size. The simplified models were derived from the final population PK models of MarzAA and DalcA which were built on adult data (see Paper III and V). Using the chosen sampling schedules for both drugs in pediatric subjects, power calculations were performed which investigated different analysis methods using a fixed sample size.

Study designs

The sample size was set to 24 subjects for both drugs. For MarzAA, 2 subjects were included below 2 years of age, 10 subjects were included between 2 and 6 years of age and 12 subjects were included between 6 to 12 years of age. For DalcA, 12 subjects were included between 2 to 6 years of age and 12 subjects were included between 6 to 12 years of age. The allocation of sample size in each age group were set as planned for in the pediatric drug development programs for both drugs. MarzAA was given as a single SC dose of 60 µg/kg whereas DalcA was given as a single SC dose of 150 IU/kg or 125 IU/kg for subjects below and equal to or above 6 years of age, respectively (see Paper III and V).

Simplification of adult population PK models

Using the final adult population PK models, pediatric PK data was simulated using the design as previously described. The models were then reduced stepwise and re-estimated. For MarzAA, the sampling schedule were set to the planned design in the phase III study (27) (pre-dose and 2, 4, 8 and 24 hours after the dose). For DalcA, the EMA guidelines for investigation of FIX products (80) were used with sampling pre-dose and 1, 10, 24 and 48 hours post-dose. The guidelines are however focusing on infusion treatments as FIX products have historically been dominated by infusion, thus allowing for adjustment of the sampling timepoints for products that have a longer half-life. Therefore, an additional sample at 96 hours after the dose was included.

The simplified models were chosen based on predictive performance as guided by the VPCs and by model parameter uncertainty. The chosen simplified models were taken forward to the sampling design optimization step.

Sampling design optimization

For MarzAA, 3 candidate sampling schedules in pediatric subjects were considered. These were i) pre-dose, 2, 4, 8- and 24-hours post-dose, ii) pre-dose, 3, 6, 24- and 30-hours post-dose and iii) pre-dose, 2, 6, 24 and 28hours post-dose. For DalcA, 4 sampling schedules were considered being i) pre-dose, 3, 24, 48, 96 hours post-dose, ii) pre-dose, 1, 24, 48, 96 hours post-dose, iii) pre-dose, 3, 10, 24, 96 hours post-dose and iv) pre-dose, 1, 10, 24, 96 hours post-dose. One thousand datasets were simulated using the adult models followed by re-estimation of the simplified models using stochastic simulations and re-estimation procedure (SSE). Relative standard error and relative absolute bias for each sampling schedule were derived according to:

$$RSE\ (\%) = 100 \cdot \frac{SD(estimates)}{true} \quad (13)$$

$$Relative\ absolute\ bias\ (\%) = 100 \cdot \frac{1}{N}\sum_{i=1}^{N}\left|\frac{estimate_i - true}{true}\right| \quad (14)$$

in which RSE is the relative standard error in each parameter, SD is the standard deviation, $estimate_{\ i}$ are the parameter estimates from each ith simulated trial, $true$ is the adult model estimate, N is the number of simulated trials (N=1000). The chosen sampling design was taken forward to study the different pre-specified approaches to prospectively power the pediatric trial.

Analysis approaches and power calculations

Using the sampling schedules derived in the previous step, 1000 pediatric trials were simulated to be used for re-estimation. Eight different approaches were defined to prospectively calculate the power to estimate CL and V in pediatrics (Table 5). The approaches could be either based on standalone analysis of pediatric data (only pediatric data used in the re-estimation step) or

pooled analysis in which re-estimation is done on pooled simulated pediatric and observed adult data. Approaches i-iii was performed using both standalone and pooled analysis whereas iv-viii only applied to standalone analysis of the data.

Table 5. Approaches i-viii outlines analysis methods used in the power calculations.

Approach	Description
i	Analysis using the adult model
ii	Analysis using a simplified adult model
iii	Analysis with the adult model with fixed parameters except the clearance and volume of distribution parameters
iv	Analysis using the adult model with the estimation of the deviation of the pediatric PK parameters from the adult ones
v	Analysis using the adult model with informative Bayesian priors
vi	Analysis using the adult model with uninformative (inflated 2x) Bayesian priors
vii	Analysis using the adult model with uninformative (inflated 5x) Bayesian priors
viii	Analysis using the adult model with uninformative (inflated 10x) Bayesian priors

Approaches i-iii involved both standalone analysis of pediatric data and pooled analysis of pediatric and adult data whereas for the rest of the approaches, only pediatric data was used for analysis.

For a detailed description of each approach, readers are referred to the methods section in Paper VII. In the re-estimation step, CL and V were re-parameterized according to the FDA recommendation (81) as:

$$P = \exp(\theta) \cdot \left(\frac{WT}{70}\right)^{b} \tag{15}$$

in which P is the PK parameter, θ is the typical value of the parameter, WT is the body weight in kg, and b is the allometric exponent. The standard errors (SE) from each re-estimation were used in the power calculations. The estimation SE was calculated according to:

$$SE_{LP} = \sqrt{\sigma_1^2 + \left[\log\left(\frac{WT}{70}\right)\right]^2 \sigma_2^2 + 2 \cdot \sigma_{12} \cdot \log\left(\frac{WT}{70}\right)} \tag{16}$$

where SE_{LP} is the estimation standard error of the logarithm of CL, σ are the elements of the variance-covariance matrix (σ_1 is the SE of CL, σ_2 is the SE of the exponent and σ_{12} is the covariance) and WT is body weight in kg. The exponents were fixed and not estimated in this work. Therefore, σ_2 and σ_{12} were set to 0 and the equation was simplified to:

$$SE_{LP} = \sqrt{\sigma_1^2} \quad (17)$$

Using the SE, the 95 % confidence interval (CI) of LP was calculated as $LP \pm t(0.975, df) \cdot SE_{LP}$ where LP is the logarithm of the parameter of interest, $t(.)$ is the t-value corresponding to 97.5th percentile of a Student t-distribution and df is degrees of freedom, i.e. the total number of pediatric patients minus the number of estimated parameters in the population PK model. Taking the exponentiation of the lower and upper CI bounds resulted in the 95 % CI of the parameter of interest. To fulfill the criteria, the 95 % CI of the CL and V parameters (alone and/or in combination) should fall within 60 % and 140 % of the true model parameters estimates. The number of trials meeting the criteria was divided by the total number of simulated trials to obtain the power. All power calculations were performed on all age groups (n=24 in total) simultaneously due to a limited sample size per age group.

Software

Model parameter estimation and simulations have in all projects been conducted in NONMEM (Icon Development Solutions, Elliot City, MD, United States) (82). Data handling, visualization and post-processing has been performed using different packages in R (R Foundation for Statistical Computing, Vienna, Austria) (83). Perl-Speaks-NONMEM toolkit (84) were used to facilitate usage of NONMEM. Run records were created using Pirana (85). Computations have mainly been performed on a cluster provided by the Swedish National Infrastructure for Computing (SNIC) at UPPMAX. For version numbers and details, readers are referred to the methods section of each paper.

Results and Discussion

Model-based analysis of early bactericidal activity trials (Papers I-II)

In Paper I which was a simulation study, a workflow of analyzing EBA data was presented along sample size calculations to highlight important factors that influence detection of EBA and detecting difference in EBA between treatment arms. In Paper II, the analysis approach outlined in Paper I was applied on observed data. Although the work in Paper II was performed before Paper I, they are presented in the chosen order to clarify the standardized analysis approach followed by a real-world applied example.

A standardized early bactericidal activity determination framework

The final outlined workflow consisted of data exploratory analysis, base model development, covariate model development, EBA detection, PKPD modelling, treatment arm comparison and EBA reporting. Following this workflow, the base model was described with a mono-exponential model with IIV on TTP slope and intercept. A proportional RUV with common and replicate specific component was used. The covariate model included cavity extent on the intercept (separate intercept estimated for each cavity category). Less cavities were associated with higher TTP at baseline ($p<0.01$). Both arms A and B showed EBA after the covariate model development ($p<0.05$). The PKPD assessment resulted in a statistically significant positive PKPD relationship between AUC and TTP slope ($\Delta OFV = -45$) following an exponential relationship. In addition, the unexplained IIV in TTP slope was reduced from 50 % to 21 %. Based on the PKPD model, a treatment arm comparison test was performed and it was found that arm B had a statistically significantly higher EBA (3.7x) when compared to arm A ($p<0.05$).

The sample size calculations for detecting EBA was performed using different slopes and magnitude of IIV in the slope (Figure 1). Regardless of IIV in the slope, a TTP-$EBA_{0\text{-}14}$ of equal or greater than 11 hours was needed to achieve 80 % power of detecting EBA using a significance level of 5 % and a sample size of 13 individuals. This corresponds to a slope of 0.0069 hours/day and

allows for 2 individuals dropping out from the 15 enrolled individuals per arm. For a scenario where IIV is low (22 %), the higher the slope, the smaller the sample size required for an 80 % power to detect EBA. If TTP-$EBA_{0\text{-}14}$ was 7 hours, 18 and 31 individuals were needed per arm to reach 80 % power for a high and low IIV scenario, respectively. The trend of a higher sample size required for 80 % power with low IIV was observed for any TTP-$EBA_{0\text{-}14}$ equal or less than 30 hours.

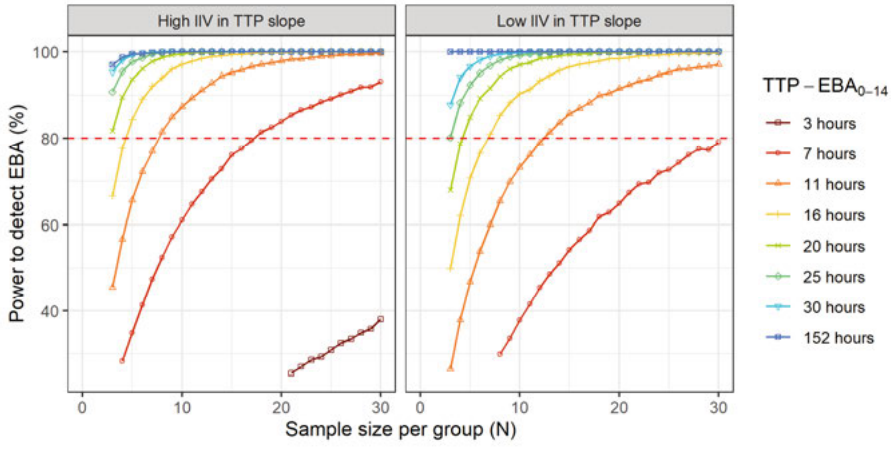

Figure 1. Power to detect early bactericidal activity based on different sample sizes for different changes in time-to-positivity between day 0 and 14 and a high and low inter-individual variability scenario (coefficient of variation of 104 % or 22 %, respectively).

An explanation for these results was found in the *post hoc* individual slope parameters which revealed that the distribution was relatively skewed when a low slope was combined with a high IIV, compared with low slope with low IIV. For the scenario of high slope (e.g. a TTP-$EBA_{0\text{-}14}$ of 152 hours), the opposite was observed in which a lower IIV favored detection of EBA and therefore led to a higher power.

The sample size calculations for detecting a difference in EBA between two treatment arms covered several combinations of TTP slope, IIV in the slope and an expected effect difference in slope between the arms. An overview of the results for all scenarios are shown in Table 6. For a scenario with relatively low EBA (TTP-$EBA_{0\text{-}14}$ of 30 hours), high IIV in the slope, and an effect difference of at least 175 %, 15 individuals per arm would be sufficient to detect the difference in effect between the arms. For effect differences below 175 %, more participants would be needed. In a scenario with high EBA (TTP-$EBA_{0\text{-}14}$ of 152 hours) and high IIV, 15 participants would not be enough to reach 80 % power to detect a difference for the studied effect differences between the two arms (>125 individuals needed for 25 % difference).

Table 6. Sample size needed to detect an effect difference with 80 % power at a significance level of 5 % between two treatment arms given different slopes, effect sizes and levels of variability.

ΔEffect (%)*	N per treatment group			
	TTP-EBA$_{0\text{-}14}$ of 30 hours[a]		TTP-EBA$_{0\text{-}14}$ of 152 hours[b]	
	Low IIV in TTP slope[c]	High IIV in TTP slope[d]	Low IIV in TTP slope[c]	High IIV in TTP slope[d]
+25	64	>125	21	>125
+50	20	90	7	87
+75	10	48	3	50
+100	6	31	3	37
+125	5	23	3	31
+150	4	18	3	28
+175	3	15	3	26
+200	3	13	3	26

*An increase in TTP-EBA$_{0\text{-}14}$
[a]TTP-EBA$_{0\text{-}14}$ similar to treatment with 2 g meropenem TID with 500 mg amoxicillin and 125 mg clavulanate with each dose of meropenem (De Jager et al. 2022)
[b]TTP-EBA$_{0\text{-}14}$ corresponding to treatment with 6 g meropenem OD with 2 × 1000 mg amoxicillin and 62.5 mg clavulanate plus 400 mg bedaquiline OD during the 14 days of treatment (unpublished data, NCT04629378)
[c]Low IIV in TTP slope defined as 22% coefficient of variation (unpublished data, NCT04629378)
[d]High IIV in TTP slope defined as 104% coefficient of variation (De Jager et al. 2022)

For an effect difference of 200 %, at least 26 participants would be needed per arm. In the case of low EBA and low IIV in the TTP slope, at least 20 individuals would be needed per arm for detection of effect differences of 50 % or below. For effect differences of 75 % or higher between the arms, 15 participants per arm would be sufficient for 80 % power to detect the effect difference between the two arms. In all of the power calculations for detecting effect differences, the highest power was found for low IIV in the TTP slope paired with a TTP-EBA$_{0\text{-}14}$ of 152 hours. This translates to that it is easiest to detect effect differences when the effect difference is large and the unknown random variability is low. Only one scenario where the effect difference was 25 % needed 21 individuals per arm, whereas all other studied scenarios reached 80 % power with 15 individuals per arm. To further visualize how IIV in TTP slope will impact detection of an effect difference, a fixed effect difference of 50 % was studied together with different levels of IIV and two values of TTP slope. The higher TTP slope scenarios usually resulted in a higher

power whereas increasing the IIV lowered the power. A detailed description of the results can be found in Paper I.

Early bactericidal activity of meropenem

The standardized analysis framework was applied in Paper II. No deaths or grade-4 adverse events occurred in the trial. Gastrointestinal and hepatobiliary adverse events (grade-2 or higher) were the most common of all adverse events. Twelve individuals withdrew from the trial early and due to poor tolerability over 14 days of treatment. The PK of meropenem appeared to be linear and was not found to be impacted by rifampicin administration. The half-life of meropenem was approximately 1 hours and giving meropenem as a single dose of 3 g or 1 g TID produced similar $AUC_{0\text{-}24hours}$. Baseline demographics are detailed in Paper II.

After the numerical and graphical exploration procedure, 2 CFU and 11 TTP observations were excluded from the model-based EBA analysis. The final CFU model was a mono-exponential model with IIV in baseline and slope. Presence of cavitary disease was found to be a statistically significant covariate on baseline CFU (intercept parameter) in which presence of cavities led to a higher baseline bacterial load. All arms were found to exhibit EBA. The treatment arm difference procedure found that arm C and D had statistically significantly different slopes each (0.202 and 0.115 log10 CFU/mL/day) whereas arms E and F shared a common slope (0.053 log10 CFU/mL/day). The RUV model was additive on a log scale. The final TTP model was also a mono-exponential model with IIV on baseline and slope, with presence of cavity was associated with lower TTP values. In addition to the mono-exponential structural model, a time-dependent function was applied for arm C, and the T_{max} parameter was also found to exhibit IIV. All arms were found to have EBA but only the rifampicin containing arm (C) was described by a separate EBA model in the treatment arm comparison. The RUV model was a proportional model. In general, the models described the data well (Figure 2). A daily meropenem dose of 6 g produced a higher EBA than 3 g (Figure 3), but was only detectable as measure by CFU and not by TTP. The rifampicin containing arm had greater EBA given both CFU and TTP data (Figure 3). All final parameter estimates are shown in Table 7.

Table 7. Final parameter estimates for the colony forming unit and time-to-positivity EBA models.

Parameter	Estimate	RSE (%)	Shrinkage (%)
CFU model			
Baseline (incl. covariate relationship)			
3. ≥ 4 cm (log10 CFU/mL)	6.2973	3	
2. < 4 cm (%)	-7.09	74	
1. Absent (%)	-17.2	19	
Slope C (M+R) (log10 CFU/mL/day)	0.202	13	
Slope D (M) (log10 CFU/mL/day)	0.1147	19	
Slope E (M1x3), F (M3x1) (log10 CFU/mL/day)	0.053	19	
$\omega^2_{baseline}$ (%CV)	17.1	32	3
ω^2_{slope} (%CV)	47.6	52	30
σ^2_{common} (%CV)	59.5	12	4
$\sigma^2_{replicate}$ (%CV)	19.8	15	4
TTP model			
Baseline (incl. covariate relationship)			
3. ≥ 4 cm (h)	107.7701	1	
2. < 4 cm (%)	4.16	45	
1. Absent (%)	8.43	31	
T_{max} C (M+R) (-)	0.862	33	
T_{50} C (M+R) (day)	4.12	16	
Slope (h/day)	0.0174	17	
$\omega^2_{baseline}$ (%CV)	5.75	23	2
ω^2_{slope} (%CV)	104	32	22
ω^2_{Tmax} (%CV)	113	53	56
σ^2_{common} (%CV)	12.5	15	4
$\sigma^2_{replicate}$ (%CV)	3.74	20	5

*CFU = colony forming unit; CV = coefficient of variation; RSE = relative standard error; σ^2 = variance of the residual unexplained variability, ω^2 = variance of inter-individual variability. Category 3 (cavities ≥ 4 cm) was used as reference for the covariate effect on the intercept and is presented on absolute scale whereas other categories are presented on percentage change scale.

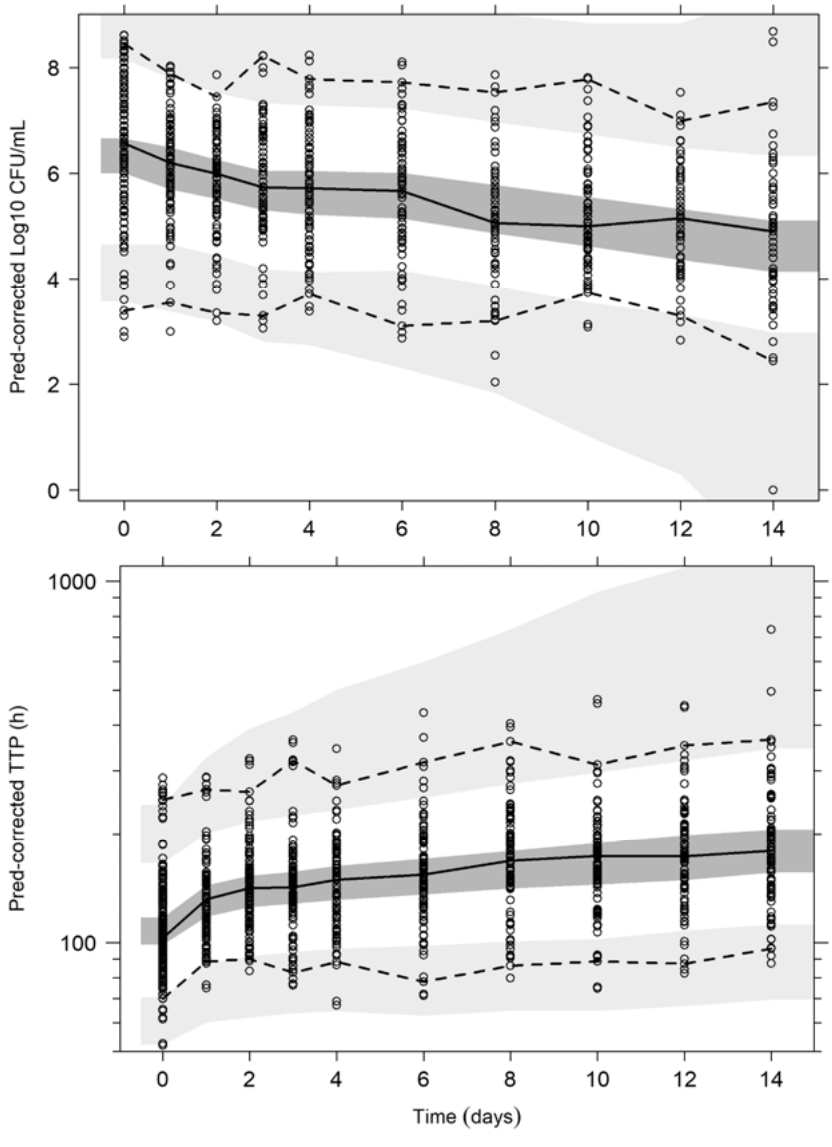

Figure 2. Prediction corrected visual predictive check of the final CFU and TTP models. Observations are shown as median (solid) and the 2.5th and 97.5th percentiles (dashed) lines respectively. Dark and light shaded areas show the model based 95% confidence interval of the median and 2.5th and 97.5th percentiles, respectively.

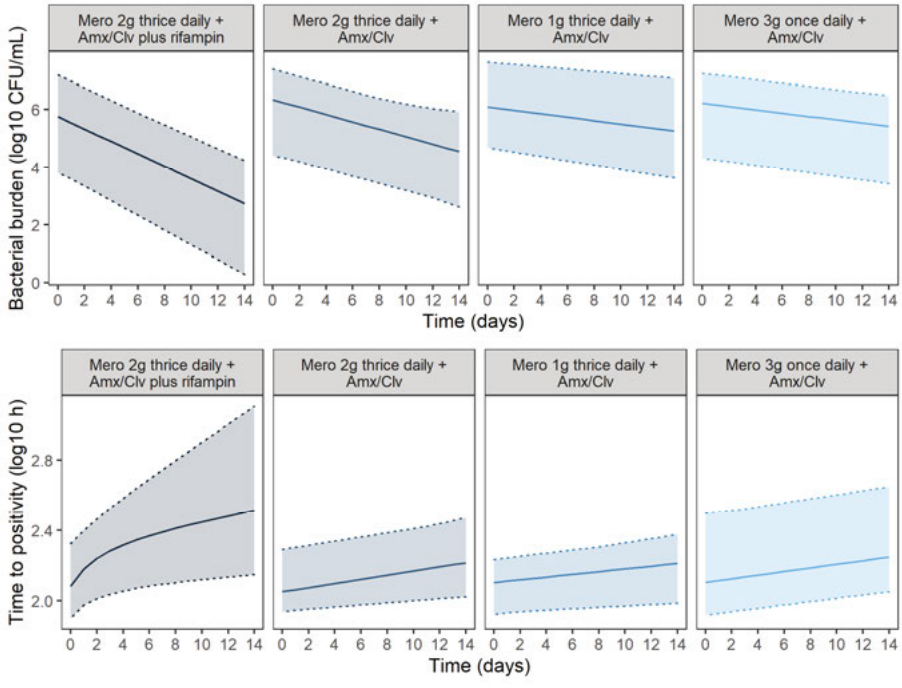

Figure 3. Mycobacterial counts based on colony forming units and time-to-positivity over 14 days. The solid lines are medians and the shaded areas are 95 % prediction intervals based on individual patient parameter estimates. Amx/Clv; amoxicillin/clavulanate, Mero; meropenem, QD; once daily, TID; every 8 hours.

Although the utility of phase IIa trials can be questioned due to measuring activity of a drug and not efficacy, it is the first instance of studying the novel drugs or combinations PK and PD in patients. As phase IIa trials aid decision making into significantly longer and costlier phase IIb trials, it is essential to analyze the data generated in an informative way. The presented approach in Paper I was inspired by the analysis plan written before analyzing the data in Paper II. However, in this thesis, they are presented in a reversed order to first give an overview of how one can analyze phase IIa trials for anti-TB drugs, followed by an applied real-world example. For example, in Paper II, exposure was not incorporated in the analysis whereas this component was changed in the analysis plan of a subsequent trial and also an important component of Paper I. Efforts have shown that the power to detect EBA can be further optimized using more mechanistic PKPD modelling (55), but was not the focus of Paper I-II. Instead, a more empirical mixed effects approach was presented where important covariates and exposure metrics can be corrected for when analyzing activity data from phase IIa trials. The focus was on finding the structural model that describe the longitudinal totality of data, assessing random variability in relevant parameters and then try to describe that variability with covariates. After that is done, the testing for EBA was done followed by

PKPD modelling in which exposure metrics are allowed to explain variability in the slope. This approach is conservative as it first let covariates describe variability, followed by exposure levels. This approach also come with drawbacks as demographic factors can impact PK and that the variability explained by covariates actually is explained by differences in PK. Although the PKPD modelling step is important, it was not performed in the applied example prior to testing for treatment arm differences. In Paper I, the hypothetical model development results showed that after implementation of AUC, IIV in the slope decreased from 50 % to 21 %. These results should be interpreted together with the power calculations in Paper I which showed that the power to detect treatment arm differences increase when IIV is reduced, which could have been a result of doing PKPD modelling in Paper II. From a *learning and confirming* point of view, confirmation of EBA and difference between two treatment arms is important to inform further development. However, given that EBA trials are rather uninformative on the efficacy of drugs or combinations of drugs, EBA trials can be seen as a learning phase for more efficacy predictive, and perhaps more proof-of-concept alike phase IIb trials. Given this paradigm of thinking, a question arises if it is possible to perform novel trial designs that go beyond 14 days EBA setting, but is shorter than the phase IIb trials (37). From an ethical point of view, the phase IIa trials have been dominated by studying drugs in monotherapy which can lead to development of resistance. However, as it is more and more common to study combinations, alternative designs have been suggested (37). Such designs coupled with interim analysis may help in advancing clinical drug development for anti-TB drugs and introduce more informative proof-of-concept data prior to studying efficacy in longer trials. In addition to informative designs, novel biomarkers are needed. The current standards rely on harvesting bacteria found in sputum. As treatment proceeds, many patients have difficulties producing sputum. Furthermore, what is found in sputum may not be reflective of bacteria found inside lesions and although mycobacterial tests indicate culture negativity, patients can relapse. Many new non-sputum based biomarkers are under investigation but remains to be validated in controlled clinical trials (86,87).

Model-based dose selection of marzeptacog alfa (activated) in adult and pediatric subjects (Paper III-IV)

Final population PK model

The final population PK model described both the IV and SC data well (Figure 4). It consisted of a two-compartment disposition model with zero-order input to a depot compartment followed by first-order absorption into the central compartment and first-order elimination. IIV was supported for CL, V, baseline FVIIa, F and k_a, with a covariance term for the IIV in CL and V. IOV was

supported for the duration of zero-order input to the depot compartment (D), F and V.

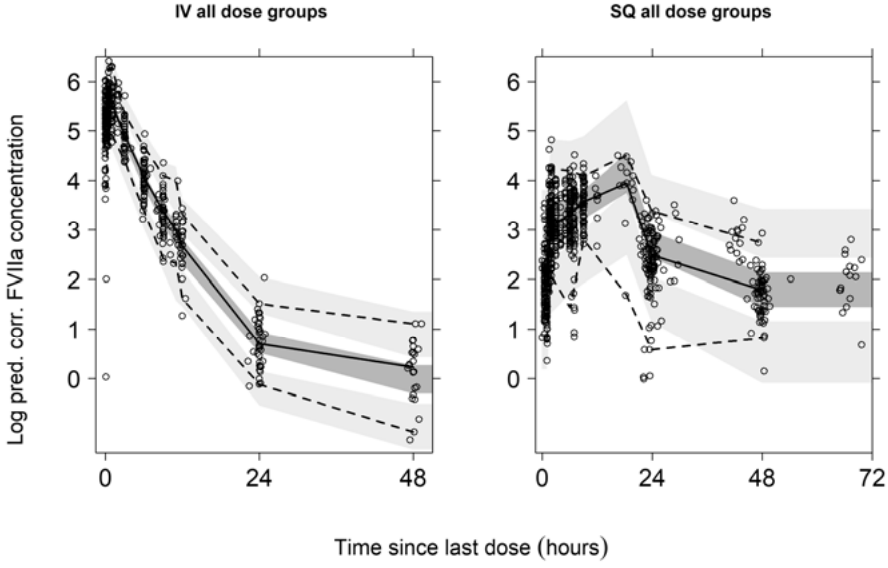

Figure 4. Prediction-corrected visual predictive check of the final population PK model on log scale following intravenous and subcutaneous dosing. The dashed lines are the 97.5th (top) and 2.5th (bottom) and the solid line is the 50th percentile of the data. The shaded areas are the 95% confidence intervals of each percentile (light grey for 97.5th and 2.5th and dark grey for 50th) based on the simulated data. Open circles are the prediction corrected observations.

The parameter estimates were in general determined with adequate precision (see Paper III). Non-linear elimination was estimated through Michaelis-Menten kinetics but led to high uncertainty in elimination parameters. A possible explanation is that most observations included in the dataset were well below the estimated concentration at half of maximum rate of elimination, in an area approaching linear elimination. Allowing for estimation of allometric exponents led to no change in OFV and high parameter uncertainty, which likely was due to the relatively narrow bodyweight range in the dataset.

Dose selection in adult subjects

The clinical trial simulations in large populations (n=1000) per dose regimen in adults are summarized using percentage of population above the target (Figure 5) and with summary statistics of secondary PK parameters of interest (Paper III). The percentage of the population above the target after a single dose of 60, 90 or 120 µg/kg was 51 %, 74 % and 86 % after 3 hours and 70 %, 87 % and 94 % after 6 hours, respectively. As this project aimed to inform the dose selection for a registrational phase III trial (Paper VI), the simulated

dose regimens were performed based on the design of the phase III trial. The protocol of the phase III trial allowed for additional dosing at 3 hours if hemostasis was not reached. The clinical trial simulations of a second dose 3 hours after the first dose result in 90 %, 96 % and 98 % being above the target 6 hours after the first dose following 60, 90 or 120 µg/kg. The protocol of the phase III trial allowed for a third and last dose if hemostasis was not reached at 6 hours after the first dose. If a third dose was given at 6 hours after the first dose, 98%, 100 % and 100 % was found to be above target at 9 hours after the first dose and 96 %, 99 % and 100 % being above the target at 24 hours after the first dose, for 60, 90 or 120 µg/kg.

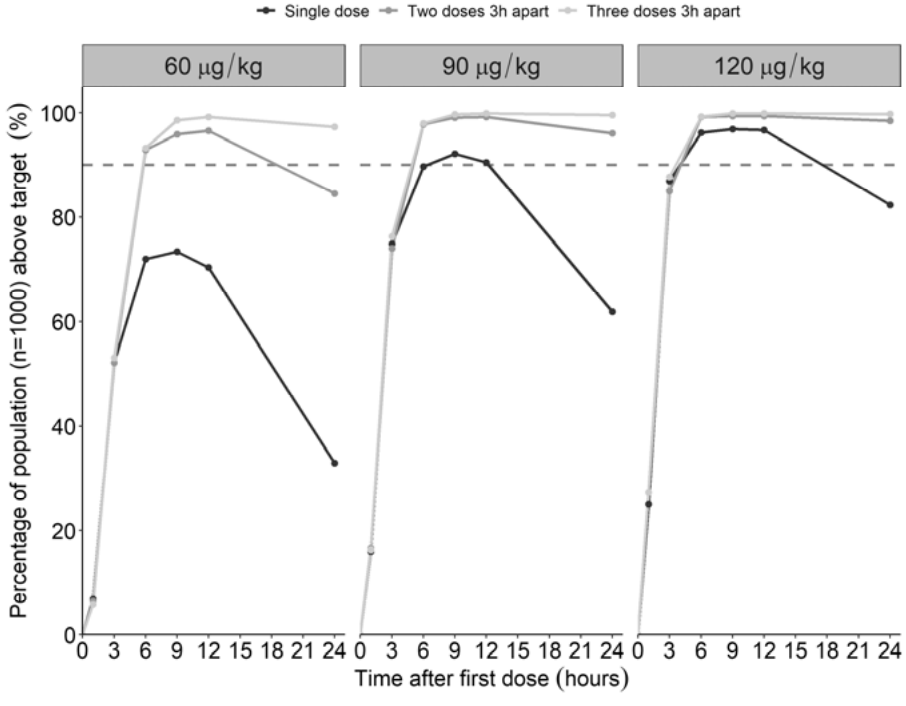

Figure 5. Percentage of the population (n=1000) above the target following 60, 90 or 120 µg/kg given as a single dose (black line), two doses 3 hours apart (dark gray) or three doses (light gray) 3 hours apart. The dashed line indicates 90 % of the population being above the target.

All dose regimens produced median C_{max24h} levels above the target. The median and 80 % prediction interval of the time-to-above the target were 2.2 (1.1-5.0), 1.6 (0.9-3.5) and 1.3 (0.7-2.7) hours after a single dose of 60, 90 or 120 µg/kg.

Selecting the dose was a complex task that was dependent on many different factors. SC MarzAA has already been shown to be efficacious in a prophylaxis

setting using a dose of 30 μg/kg given OD. The design of that trial allowed for dose escalation to 60 μg/kg given OD (26) if a bleeding occurred on 30 μg/kg OD. The only patients that were dose escalated did not experience any bleeds in the following 50 days after dose escalation (26). Furthermore, recombinant biologics are in general expensive medicines and the dose will also affect injection volume. As both of these factors need to be kept down to reduce economic burden and issues related to administration of the drug, it is of interest to pick the lowest possible efficacious dose. This strategy together with the possibility to re-dose in case of failure to hemostasis assure cost-effectiveness, which is an important aspect after drug approval from a marketing point-of-view. Taken together, dose selection was supported by population PK modelling in absence of an exposure-response relationship, but by using PK and preclinical potency information accumulated throughout development.

Dose selection in pediatric subjects

Based on the levels arising from the chosen dose level in adults, an exposure-matching strategy towards adult subjects were performed in pediatric subjects. The clinical trial simulations in large populations of different age groups were summarized by calculating median and 95 % prediction intervals for the different dose regimens. The main metrics of interest used for exposure-matching were $C_{max0\text{-}24h}$ and $AUC_{0\text{-}24h}$ and an overview of the results are presented in Figure 6 and Figure 7. In general, for all dosing frequencies, reducing the dose from 120 to 60 μg/kg had a more pronounced effect on the upper end of the prediction interval than the lower end for both exposure metrics. The 60 μg/kg dose level resulted in the lower bound of the prediction interval of $AUC_{0\text{-}24h}$ to be slightly below the lower bound of the adult matching range across all dosing intervals and age groups. The 75 μg/kg dose level was similar for the 2, 1 and 0.5-year-old groups, whereas the older children had levels above the lower end of the adult target range. The 60 μg/kg dose level did not exceed the upper end of the adult interval in any dose frequency or age group. All other dose levels notably exceeded the adult matching range in most age groups. Therapeutic proteins can have higher absorption in children than in adults, which has been found for SC tocilizumab and abatacept and intramuscularly administered palivizumab (88,89). The hypothetical doubling of the absorption rate constant in the pediatric clinical trial simulations in general increased both exposure metrics with a larger effect on $C_{max0\text{-}24h}$ (Figure 8), which exceeded the adult matching range across all dose levels and age groups. The most common adverse event following IV administered SoC is thrombotic events, which potentially can be mitigated using SC formulations due to a smoother PK profile (under the assumption that C_{max} is the driver). Levels up to 30 IU/ml has been observed safely in patients, which correspond to 120 ng/ml of MarzAA assuming a 5-fold difference in potency. The rather empirically set, and perhaps extreme scenario of doubling the absorption rate

still produced $C_{max0\text{-}24h}$ below 120 ng/ml in a vast majority of the virtual subjects. It should be noted that this sensitivity analysis is hypothetical and potential differences in absorption rate remain to be studied in a clinical setting.

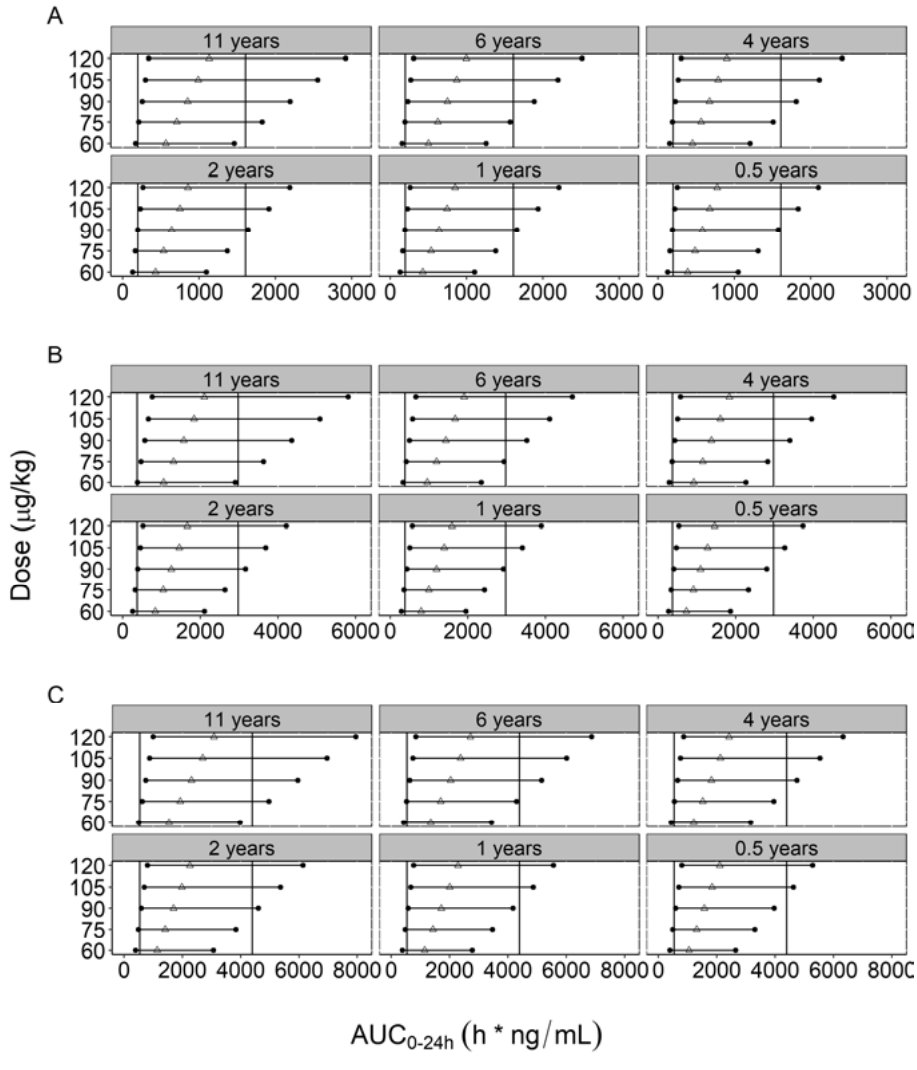

Figure 6. Median (triangle) and 95 % prediction interval (horizontal lines) of area under the curve during 24 hours following a single (A), two doses given 3 hours apart (B) and three doses given 3 hours apart (C) for dose levels 60, 75, 90, 105 and 120 µg/kg. The vertical solid lines indicate the adult matching range (95 % prediction interval following 60 µg/kg of the same dosing schedules.

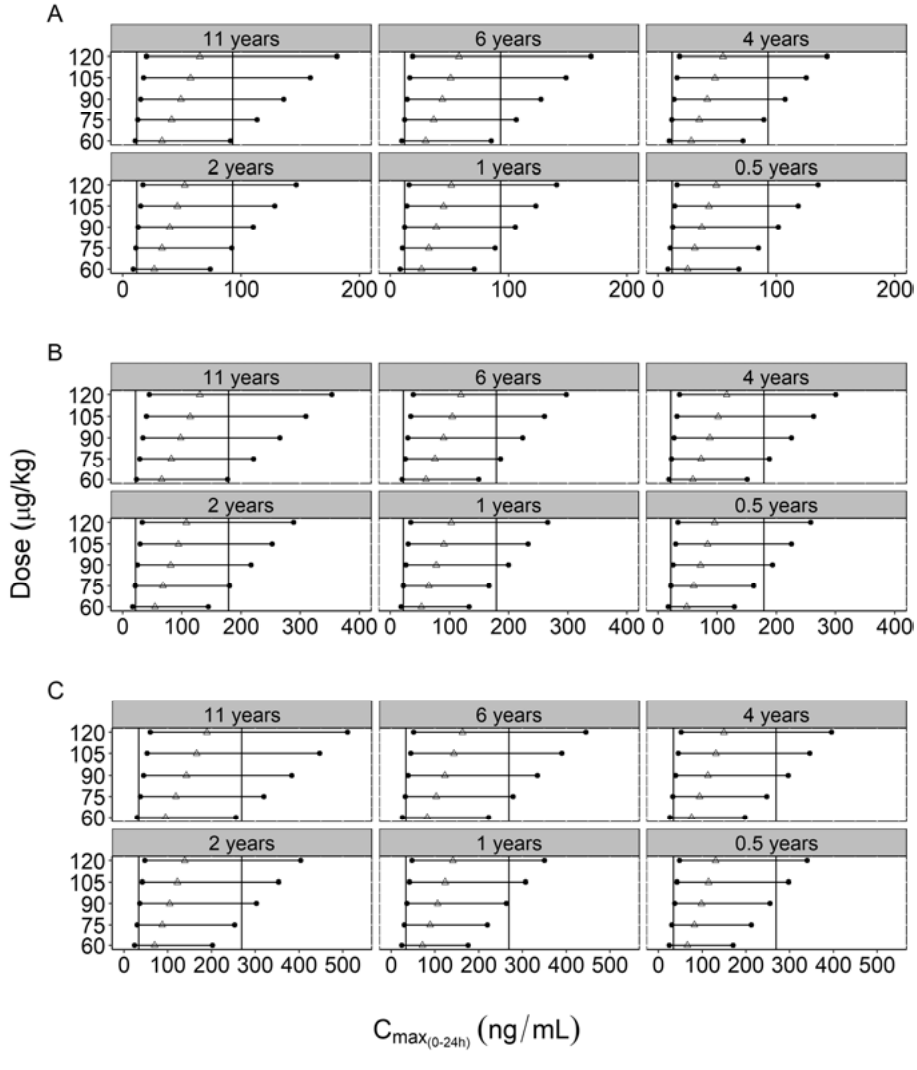

Figure 7. Median (triangle) and 95 % prediction interval (horizontal lines) of maximum concentration during 24 hours following a single (A), two doses given 3 hours apart (B) and three doses given 3 hours apart (C) for dose levels 60, 75, 90, 105 and 120 µg/kg. The vertical solid lines indicate the adult matching range (95 % prediction interval following 60 µg/kg of the same dosing schedules.

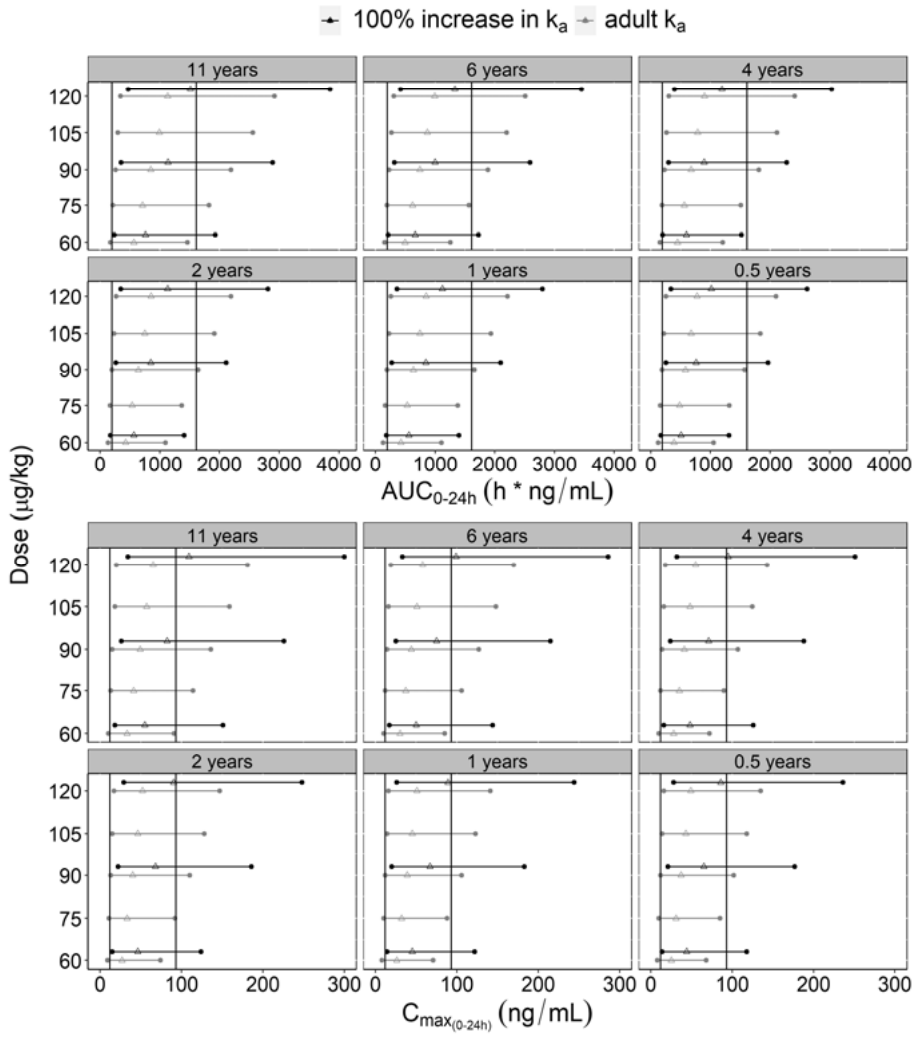

Figure 8. Median (triangle) and 95 % prediction interval (horizontal lines) of maximum concentration and area under the curve during 24 hours following a single dose of 60, 75, 90, 105 and 120 µg/kg. The vertical solid lines indicate the adult matching range (95 % prediction interval following a single 60 µg/kg dose).

Another important factor influencing extrapolation of PK from adults to children is the choice of allometric exponents. For CL, an exponent of 0.75 implies an increase in CL with bodyweight, but a decrease in CL normalized by bodyweight, with increasing bodyweight values. The increase in bodyweight normalized CL in children have been observed for rFVIIa (90). In general, allometry with fixed exponents has been shown to be a useful tool for initial dose selection in children above 2 years of age for small molecules, and also down to 5 years for FVIII and FIX (64,68). Below 2 years of age, accounting

for maturational factors are recommended. As no information was available on maturational effects for FVIIa, a second sensitivity analysis was performed using the ADE method (68,91) aiming to reduce the risk of overdosing children below 2 years of age as a result of overestimating CL. Although the ADE method has been shown to be useful for small molecules (63,67) and biologics (91), it is a simplification and the exponents might be drug specific. When applied to inform first-in-pediatric dose selection, the assumptions should be validated and the simulations revisited once data in children is available. The clinical trial simulations in large populations of children of different age groups suggested that 60 µg/kg is a promising dose regimen to match levels arising from the adult regimen.

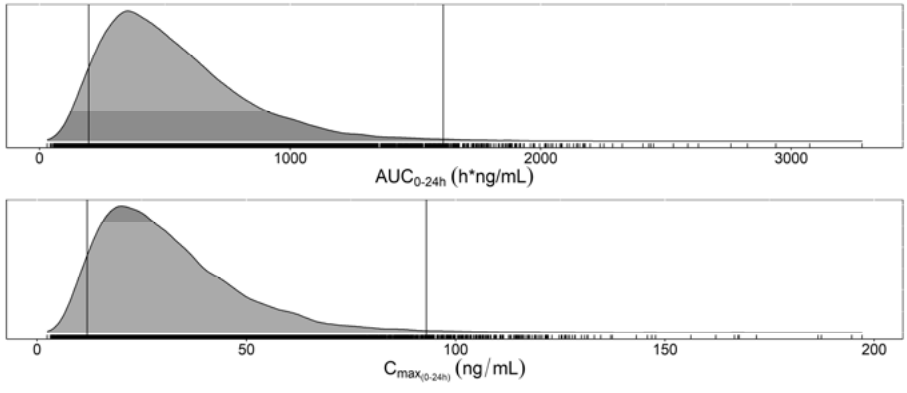

Figure 9. Distributions of $AUC_{0\text{-}24h}$ and $C_{max0\text{-}24h}$ across all trials with 24 pediatric subjects in each trial (n=24000). The vertical lines illustrate the adult 95 % prediction interval arising from a single 60 µg/kg dose.

To further support the decision-making process, the probability of trial success (defined as matching adult levels) was studied using a dose level of 60 µg/kg for all age groups. With a criterion of 20 children being within the adult matching range per trial, the probability of trial success was 99 % and 97 % based on $AUC_{0\text{-}24h}$ and $C_{max0\text{-}24h}$. Corresponding probabilities when requiring 21 or 22 children to be within the target was 98 % and 89 % using $AUC_{0\text{-}24h}$, and 91 % and 74 % based on $C_{max0\text{-}24h}$. The distributions of $AUC_{0\text{-}24h}$ and $C_{max0\text{-}24h}$ across all 1000 clinical trials and children are shown in Figure 9, indicating a majority of values to be within the target range.

As recruitment is challenging for rare diseases, it is important to make informed decisions for first-in-pediatric dose selection. The model-informed clinical trial simulations supported the planning of the adult and pediatric trials that aimed to study SC MarzAA for acute bleeding events in absence of early efficacy data.

Model-based dose selection of dalcinonacog alfa in adult and pediatric subjects (Paper V)

Final population PK model

A two-compartmental disposition model with first-order absorption and elimination described the data well. IIV was included on CL, BASE and k_a and the residual error model was proportional. A covariate effect describing a lower F in the trial conducted in South Africa compared to the trial conducted in South Korea was statistically significant ($p<0.01$). However, due to high uncertainty and no impact on the descriptive performance of the model, it was not included in the model. Such covariate effects may be explained by ethnicity. However, the dataset did not include ethnicity information. The final model described the IV and SC data well (Figure 10) and the parameters were estimated with sufficient precision given the small sample size (Paper V). Therefore, the model was considered suitable for clinical trial simulations to inform dose selection in adult and pediatric HB subjects.

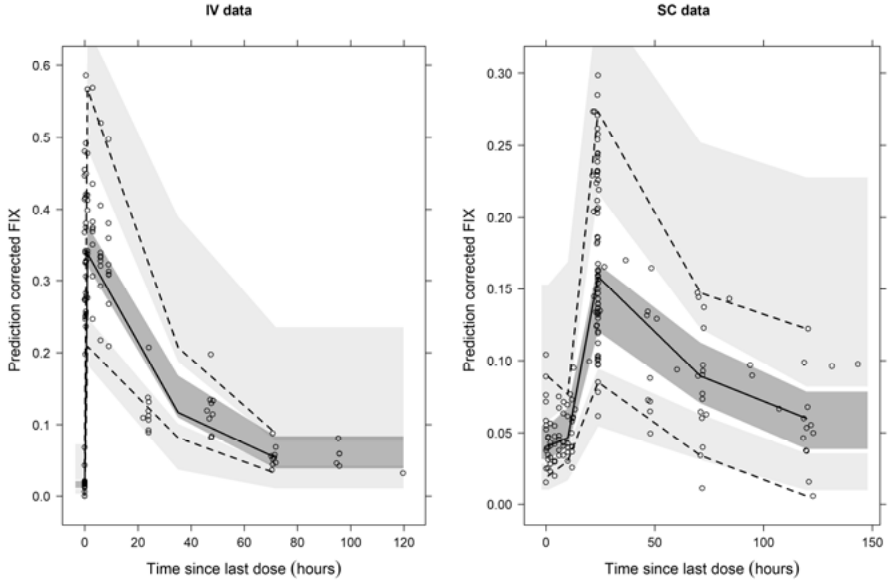

Figure 10. Prediction corrected visual predictive check for intravenous and subcutaneous data. Solid and dashed lines are the median, 2.5th and 97.5th percentiles of prediction corrected data. Shaded areas are from top to bottom the 95 % confidence intervals of the 97.5th (light grey), median (dark grey) and 2.5th percentiles of the simulated data. Open circles are prediction corrected observations.

Dose selection in adult subjects

The exposure metric of interest for prophylactic treatment with DalcA was $C_{min,ss}$, and the target was set to put patients into mild hemophilia (levels equal

to or above 0.1 IU/ml [10 % FIX]). A vast majority of subjects were found to reach levels at or above the target at steady state, with a dose of 100 IU/kg given OD (Figure 11). Dosing less frequently such as EOD was not sufficient to achieve the target at dose levels of 125 IU/kg or 150 IU/kg. However, if a lower target than 0.1 IU/ml is set in clinical praxis, EOD dosing might still be relevant. When a patient initiates prophylactic treatment, another metric of interest is the time it takes until target levels are reached. This may be seen as a time in which the patient is relatively less protected and the risk for spontaneous bleeding events are elevated. Across all the simulated dose levels given OD, the median and 90 % prediction interval was 6.3 (2.1-13.4), 4.4 (5.0-9.5), 3.6 (2.0-8.3) and 3.2 (1.6-7.1) days for 50, 75, 90 and 100 IU/kg, respectively.

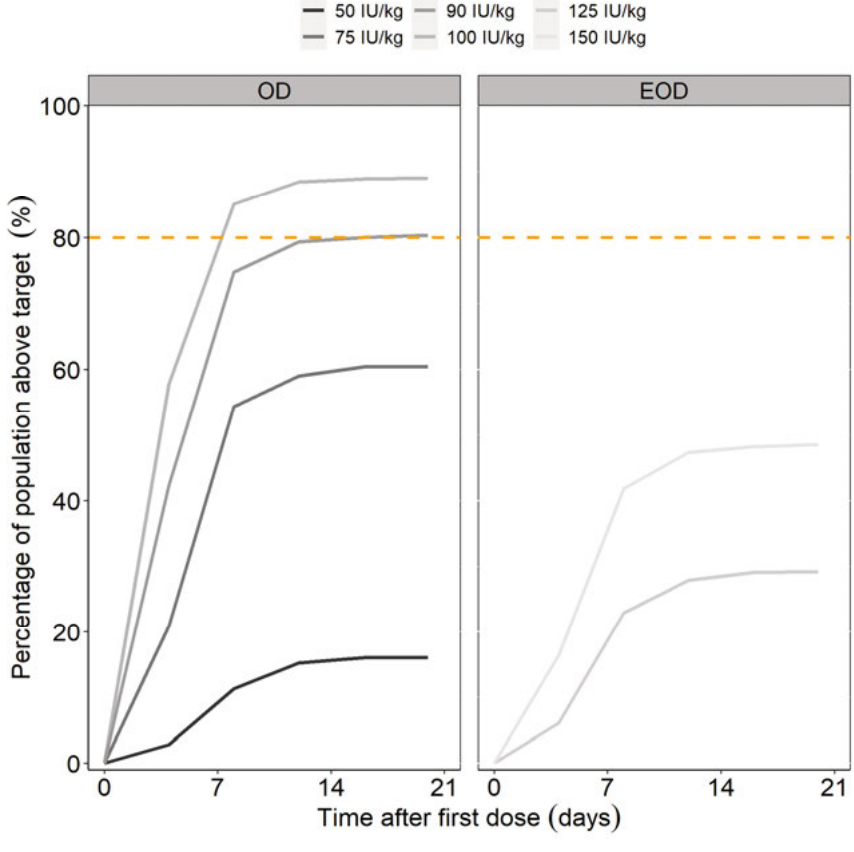

Figure 11. The percentage of the population above target versus time based on C_{min}. The solid lines indicate different dose levels being given either once daily (OD) or every other day (EOD). The dashed line indicates 80 % of the population being above the target.

Dose selection in pediatric subjects

As HB affect both adults and children with negative impact on quality of life, it is important to inform the first-in-pediatric dose based on available adult data. In this work, the model built on adult data including allometric scaling was used for pediatric extrapolation. In general, using allometric scaling with fixed exponents of 0.75 and 1 for CL and V terms has been shown to perform reasonably well with respect to first-in-pediatric dose selection for coagulation factors (68). In contrast to the pediatric extrapolations for MarzAA which had a target set based on potency bridging to wt-FVIIa, the target for DalcA was set to change the hemophilia phenotype in children to mild according to the clinical guidelines (79). The clinical trial simulations showed that 125 IU/kg given OD would lead to a majority of the virtual subjects meeting the target down to and including 6 years of age (Figure 12). For subjects below 6 down to and including 2 years, a dose of 150 IU/kg would be more appropriate.

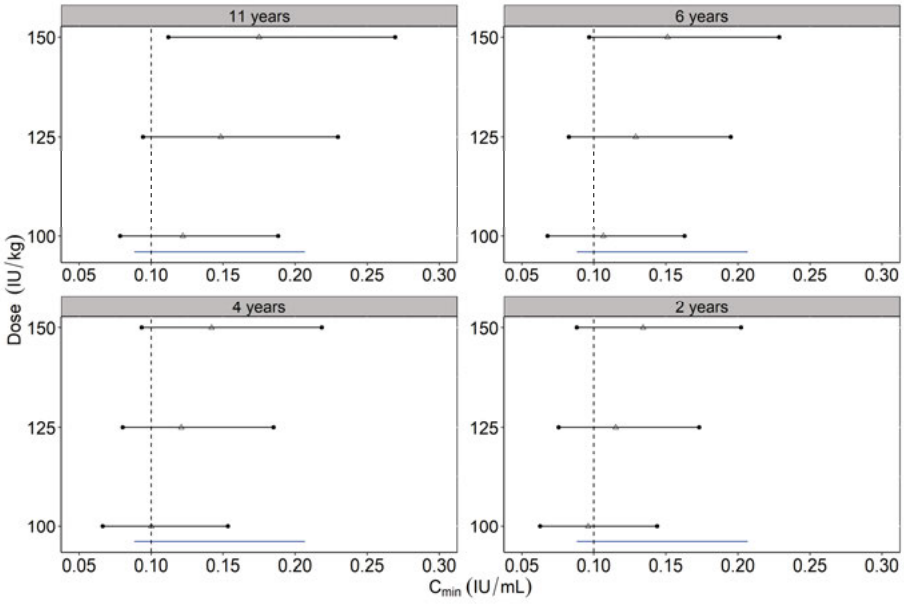

Figure 12. Median (triangle) and 90 % prediction interval (black horizontal bars) of minimum concentration values at steady state for different dose levels and age groups. The blue line is the 90 % prediction interval based on 100 IU/kg given OD in adults.

The median and 90 % prediction interval of time-to-target were 2.7 (1.3-6.2), 3.1 (1.3-7.2), 3.2 (1.4-8.2) and 3.3 (1.5-7.3) days following 125 IU/kg for age groups 11, 6, 4 and 2 years. When dosing 150 IU/kg OD, the corresponding values were 2.2 (1.2–4.4), 2.3 (1.2–5.2), 2.4 (1.2–6.2) and 2.5 (1.3–7.2) days. For the children in the 11 and 6-year old groups that did not meet the target with 125 IU/kg given OD, an escalation to 175 IU/kg OD would put at least

80 % of those subjects above the target. Similarly, for the children in the 4 and 2-year old groups that did not meet the target using 150 IU/kg OD, a dose escalation to 200 IU/kg would be sufficient. Simulations investigating dose escalations are in particular helpful for adaptive trial designs that allow for adjusting the dose based on interim data. Another aspect that may be informed based on interim data is the PK down to 2 years of age, which potentially can inform on dose selection below 2 years of age. The clinical trial simulations indicated that using a one-dose-fits-all approach would probably not be sufficient to change the hemophilia phenotype to mild in all age groups, which is in accordance with other FIX products (92). This work did not include any PD or clinical outcome data informing on bleeding rate such ABR. Although the FIX levels are informative and may be predictive of lowering ABR, the relationship between FIX levels and ABR need to be established prior to using FIX levels as a surrogate for efficacy. Despite analyzing limited data from a small sample size, this modelling work provided information on what dose levels that are likely to be efficacious in a pivotal setting.

Model-based analysis of on-demand clinical outcome data (Paper VI)

Clinical bleed outcome model

The final model was a four-state Markov model (Figure 13) with one parameter describing worsening of the scores ($k_{FP\text{-}GF\text{-}EG}$) whereas two separate parameters described improvement of the scores. A shared transfer rate was estimated describing transfer from poor to fair and fair to good improvement of the scores ($k_{PF\text{-}FG}$), whereas a separate rate was estimated for moving from good to excellent improvement (k_{GE}). The typical rates estimated were 0.01, 0.17 and 0.2 day^{-1} for $k_{FP\text{-}GF\text{-}EG}$, $k_{PF\text{-}FG}$ and k_{GE} and IIV was estimated to a CV of 11 % and 21 % on $k_{PF\text{-}FG}$ and k_{GE}, respectively. The covariate search supported that age increased $k_{PF\text{-}FG}$ for ages above 33 years, translating to a higher rate of improvement in older patients. It is however difficult to determine if age is the driver, or if changes with age such as physical activity is the driver. Bleeding events in knees or other joints were found to have a higher $k_{PF\text{-}FG}$ compared to bleeds in elbow or ankle joints. This covariate effect may be arising from that different joints are differently perfused and are under different type of stress. In addition, delivery of drug to the sites may vary between the bleeding event locations. Finally, BDBP was found to decrease k_{GE} with increasing BDBP values, indicating that patients with higher BDBP have a lower improvement rate to excellent from good. However, as treatment success often is determined as good or excellent improvement, the impact of a lower k_{GE} on treatment success is not obvious. Hemophilia patients has been shown to have higher blood pressure after adjusting for age and other risk

factors (93,94), but the classification of low, normal or high blood pressure is dependent on both systolic and diastolic pressure. In this work, no systolic pressure covariate effect was found. The covariate model was taken forward to the exposure-response search and a statistically significant ($p<0.01$) exposure-response relationship was found between I_{dur} and $k_{FP\text{-}GF\text{-}EG}$, but was determined with low parameter precision and therefore not included in the final model. Furthermore, none of the transfer rates were found to be statistically significantly different between bleeds that were treated with IV SoC or SC MarzAA. The four-state Markov model respected the natural order of the outcome data without simplification and enabled maximized usage of the clinical trial data and therefore chosen as the final model. However, as the data was limited and sample size was small, an alternative two-state model was also investigated.

The alternative two-state model (Figure 13) had one parameter describing transfer from treatment failure to treatment success (k_{TFTS}) and one for the opposite direction (k_{TSTF}), estimated to 0.57 and 0.004 day^{-1}, respectively. The covariate search resulted in identification of bodyweight and BDBP as statistically significant covariates ($p<0.01$). The k_{TFTS} parameter was found to increase with higher bodyweight values, suggesting that higher bodyweight benefits moving from TF to TS. In addition, the same parameter was impacted by BDBP in which BDBP below and equal to 80 mmHg was found to increase the parameter whereas higher values above 80 mmHg was found to worsen improvement (i.e. decrease the k_{TFTS} rate). The exposure-response modelling step identified $Tmax_{48h}$ as a statistically significant ($p<0.01$) predictor decreasing k_{TFTS}, but was not kept in the model due to low parameter precision. As for the final four-state model, no differences were found in parameter estimates between bleeding events treated with IV SoC or SC MarzAA.

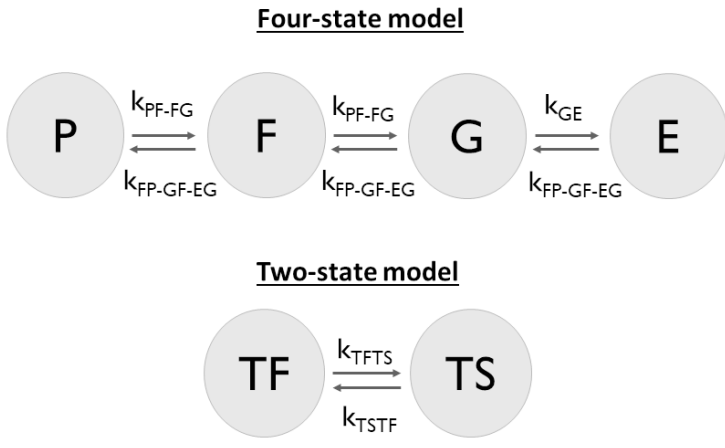

Figure 13. The final four-state Markov model and the alternative two-state model. Poor (P), fair (F), good (G) excellent (E) illustrate possible outcomes in the four-state model whereas treatment failure (TF) and treatment success (TS) illustrate possible outcomes in the two-state model. k_{PF-FG} is a first-order rate describing transfer from P to F and from F to G improvement. k_{GE} is a first order rate describing transfer from G to E improvement. $k_{FP-GF-EG}$ is a first order rate describing worsening of the score in the four-state model. k_{TFTS} is a first order rate describing transfer from TF to TS whereas k_{TSTF} describes the opposite transfer.

Both models described the data well (Figure 14) and parameters were estimated with sufficient certainty given the small sample size (Paper VI). The clinical trial simulations in Paper III showed that if a second 60 µg/kg dose is to be needed due to poor response at 3 hours after the first dose, more than 80 % of the adult subjects were predicted to be above the set FVIIa level target at 24 hours after the first dose.

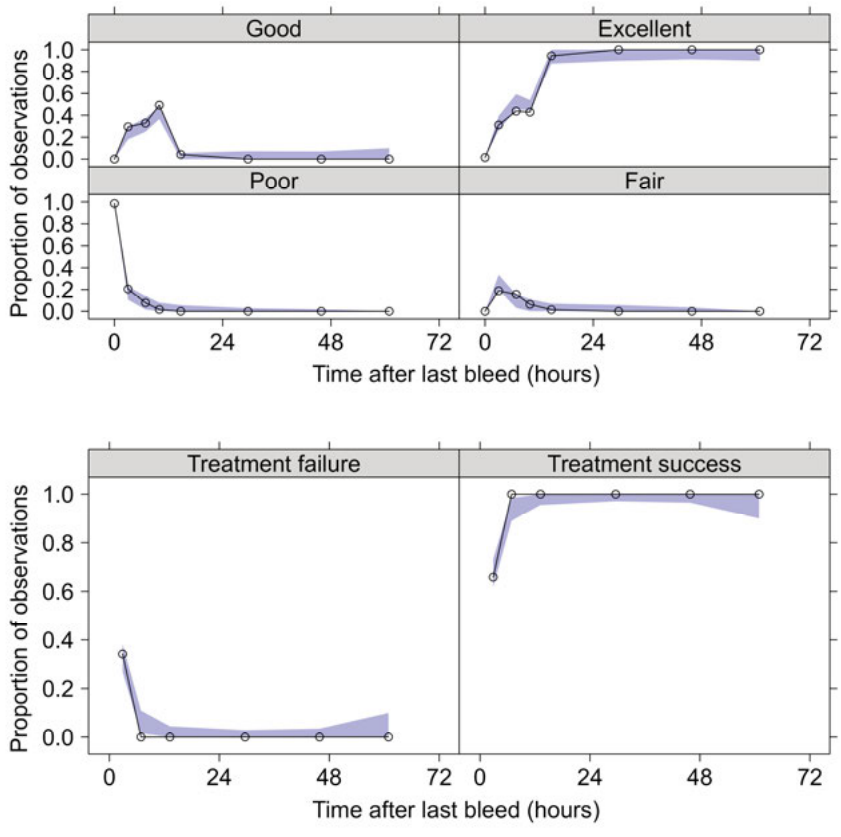

Figure 14. Prediction corrected visual predictive check of proportion of total observations in each outcome category versus time after last bleed based on the final four-state model (upper) and the alternative two state model (lower). The circles are the prediction corrected observed proportions and the blue shaded areas are the 95 % prediction intervals based on 500 simulations.

In light of those results, 2 SC MarzAA doses were needed to treat a bleeding event on a median level. However, the range was 1-5 doses which indicates a high variability in response, potentially due to the high variability in PK of MarzAA (28). However, the potency bridging strategy should be further investigated when data from a larger sample size is available, and the exposure-response relationship can be further studied. In this work, most PK data was from the non-bleeding state. There might be differences in non-bleeding PK and bleeding PK of MarzAA, on top of the already high IOV detected in Paper III. Independently of using the original 4-point scale or a binarized form, no statistically significant difference between SC MarzAA and IV SoC was found in the transfer rates, indicating that SC MarzAA has potential for treatment of acute bleeding events.

Covariate effect impact on bleeding improvement

In general, bleeding events are recommended to be treated as fast as possible to prevent bleeding associated joint damage (95) which can impact mobility and quality of life negatively. Different combinations of covariate effect values were studied to visualize the covariate effect impact on bleeding improvement (Figure 15 for the four-state model and Paper VI for the two-state model). A trend was observed in which the covariate effects had minor impact on the probability of treatment success (good or excellent improvement) at later timepoints (12 or 24 hours post first dose) as most combinations of covariates resulted in a high proportion of treatment success. However, at early timepoints (3 hours post first dose), some combinations notably lowered treatment success (Figure 15).

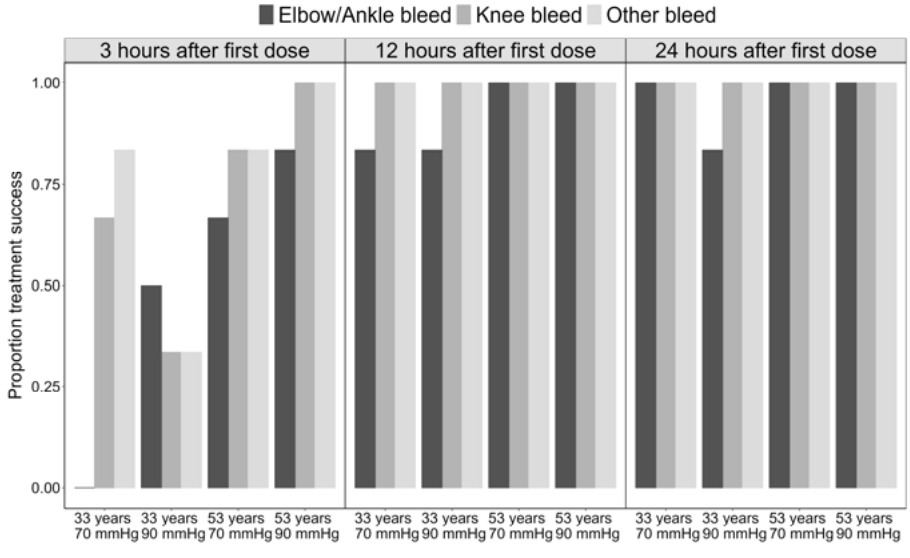

Figure 15. Prediction of proportion of successful treatment (good or excellent response) based on the final four-state model for different subpopulations of patients for different ages, baseline diastolic blood pressure and bleeding location at different time-points after the first dose.

A typical 53-year-old individual with BDBP of 70 mmHg was predicted to have better response compared to a 33-year-old with the same BDBP, if the bleed location was knees (67 % versus 83 %) or elbow/ankles (67 % versus 0 %) compared to bleeds in other joints (83 % for both age groups). Regardless of location of the bleed, a BDBP of 90 mmHg was associated with better response compared to 70 mmHg in a 53-year-old typical subject. In a typical 33-year-old subject, a higher BDBP of 90 mmHg led to worsened response compared to 70 mmHg if the bleed was located in knees (33 % versus 67 %) or other locations (33 % versus 83%). Interestingly, if the bleed location was

in elbows/ankles, a 33-year-old typical subject with a higher BDBP of 90 mmHg had a better response (50 % versus 0 %) compared to having BDBP of 70 mmHg. The alternative two-state model resulted in a similar pattern, being that the covariate effect had most impact on early response (Paper VI). One combination of covariate values (bodyweight of 51 kg and BDBP of 90 mmHg) led to poor treatment response regardless of time after the first dose (Paper VI). The results indicate that some combinations of covariates may lead to an increased number of doses needed to treat an acute bleeding event, but should be further studied with a larger sample size. The difference in detected covariate effects between the two modelling approaches is expected as the base model that the covariates are tested in are different. However, as the more elaborate four-state model respect the nature of data without transformation, it may be more reliable.

Model-based approaches analysis of pediatric pharmacokinetic trials for hemostatic drugs (Paper VII)

Simplification of adult population PK models

Both the MarzAA and DalcA simplified models were one-compartmental disposition models. A sequential zero-order followed by first-order absorption described the simulated pediatric MarzAA data well whereas a first-order model was used for DalcA. IIV was supported on CL, V and baseline FVIIa for MarzAA whereas the final simplified DalcA model had IIV on CL, baseline FIX and k_a. Allometric scaling with fixed exponents of 0.75 and 1 for CL and V terms was included in both models. Both models were considered to describe the data (Paper VII) and was therefore taken forward to the sampling optimization step.

Sampling design optimization

For MarzAA, the evaluated sampling schedules all provided similar RSE (ranged 11-12 % for CL and 33-36 % for V) and relative absolute bias (range 9-10 % for CL and 29-30 % for V). The sampling design as planned for the phase III study (including adult subjects) with sampling at pre-dose and 2, 4, 8 and 24 hours after the dose provided slightly better results and was chosen. As simplicity of the protocol is beneficial from a trial execution point of view, using the same sampling schedule in both adult and children can be an advantage. The different sampling schedules also provided similar results for DalcA, with RSEs ranging from 12-14 % for CL and 47-53 % for V. Relative absolute bias ranged 11-12 % for CL and 41-45 % for V. The sampling schedule of pre-dose followed by 3, 24, 48 and 96 hours after the dose provided the

best results and chosen for DalcA. For both drugs, it was found that V in general was more difficult to estimate than CL.

Analysis approaches and power calculations

The power to estimate CL and V alone or in combination are reported for each approach in Figure 16 for MarzAA and in Figure 17 for DalcA. In general, the results showed that it is harder to estimate V than CL, or V together with CL as compared to estimating CL alone, which indicate that the sampling schedule chosen in the previous step is more informative for CL compared to V. When applying approaches i-iii, pooling pediatric and adult data prior to analysis improved the results compared to standalone analysis. This was particularly evident for the power to estimate V alone (76, 100, 100 % versus 3, 7 and 52 % for MarzAA and 54, 97, 100 % versus 2, 8 and 0 %, for DalcA). The same trend was observed for when the power calculations were conditioned on CL and V simultaneously, resulting in 73, 99, 100 % when pooling versus 3, 5, 52% with standalone analysis for MarzAA, and 47, 89, 100 % versus 0, 8, 0 % for DalcA. Approaches ii and iii performed better than i when pooling data and powering for CL and V simultaneously (99, 100 % versus 73 % for MarzAA and 89, 100 % versus 47 % for DalcA). When performing standalone data analysis, iii performed better than i and ii when powering for CL alone (96 % versus 64 and 53 % for MarzAA and 99 % versus 71 and 62 % for DalcA). For MarzAA, the same trend was observed when powering CL and V simultaneously or when powering for V alone. However, for DalcA, approaches i-iii was all insufficient for estimating the parameters simultaneously (<10 % power) when performing standalone analysis. Approach iv used a standalone analysis approach and was similar to iii, but in general performed worse than iii for MarzAA. However, for DalcA, it performed better (43 % versus 0 % and 41 % versus 0 %), when powering for V alone or in combination with CL. Approach i did not perform well when performing standalone analysis, likely as a result of sparse data informing a complex adult model leading to identifiability issues. Approaches ii and iii failed to estimate CL and V simultaneously with sufficient certainty with standalone analysis of pediatric data, a result that may be due to the limited sample size and sparse design. Interestingly, approach iii performed better than iv for MarzAA, whereas the opposite was observed for DalcA. The main difference between approach iii and iv is that in iii, adult and/or pediatric parameters are estimated whereas in iv, all adult parameters are fixed and an additional deviation from adult parameter is estimated on top of allometry. The deviation parameter on top of allometry in iv might be estimated to values close to 0 which in general can lead to numerical difficulties when performing estimation. Both iii and iv rely on an assumption that all other parameters than CL and V are the same in adults and children, which can be confirmed when data in children is available.

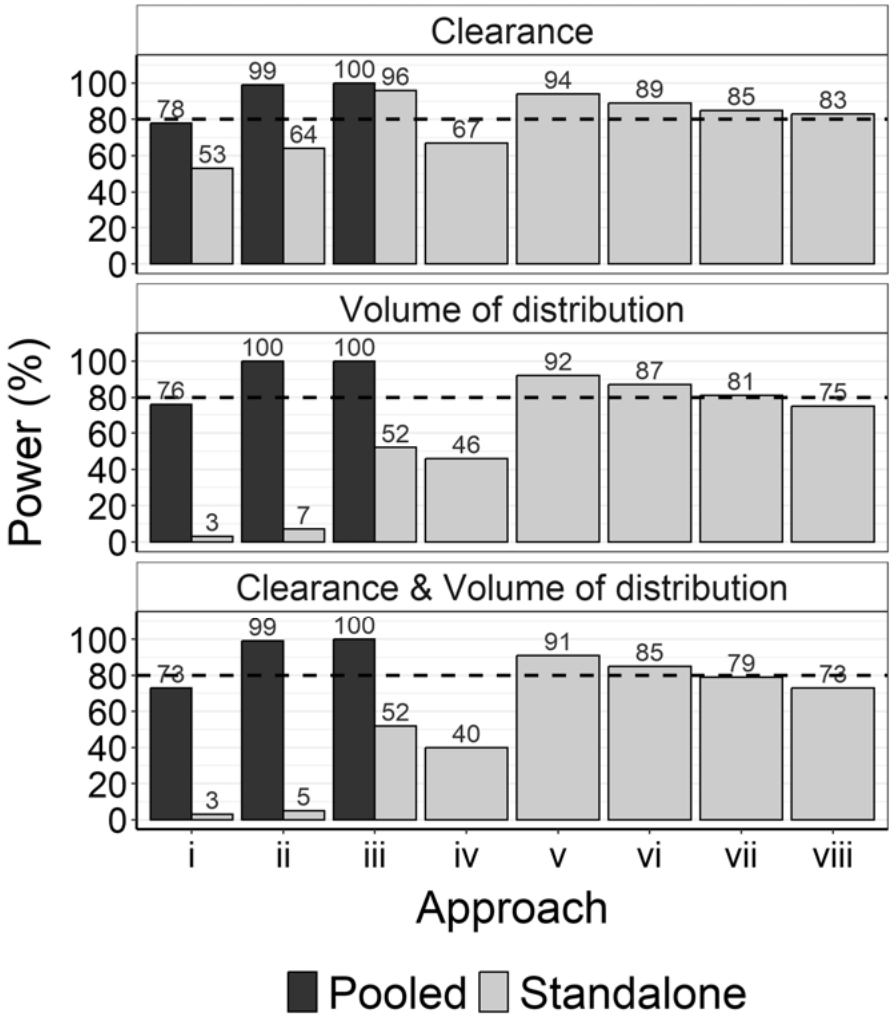

Figure 16. Power to estimate CL, V or both parameters to fulfill the FDA criterion for MarzAA using all analysis approaches. Pooled analysis of adult and pediatric data is indicated by black bars whereas standalone analysis of pediatric data is indicated by grey bars. The dashed horizontal line marks 80 % power.

The Bayesian prior approaches in general performed well when estimating CL and V either alone or in combination when the priors were informative. With decreasing informativeness (vi-viii), the power decreased. For instance, when powering for CL of MarzAA, all approaches led to >80 % power, whereas powering for V led to power <80 % using approach viii. When powering for CL and V together, vii and viii resulted in power slightly lower than 80 % whereas the approaches with higher informativeness in the priors (v and vi) resulted in >80 % power. A similar trend was observed for DalcA when powering for V alone or with CL. Approaches vii and viii resulted in 68 % and 21

% power when powering for both parameters simultaneously, whereas v and vi resulted in 91 % and 84 %. Powering for CL alone resulted in sufficient power using all methods and powering for V alone led to sufficient power for all approaches but viii.

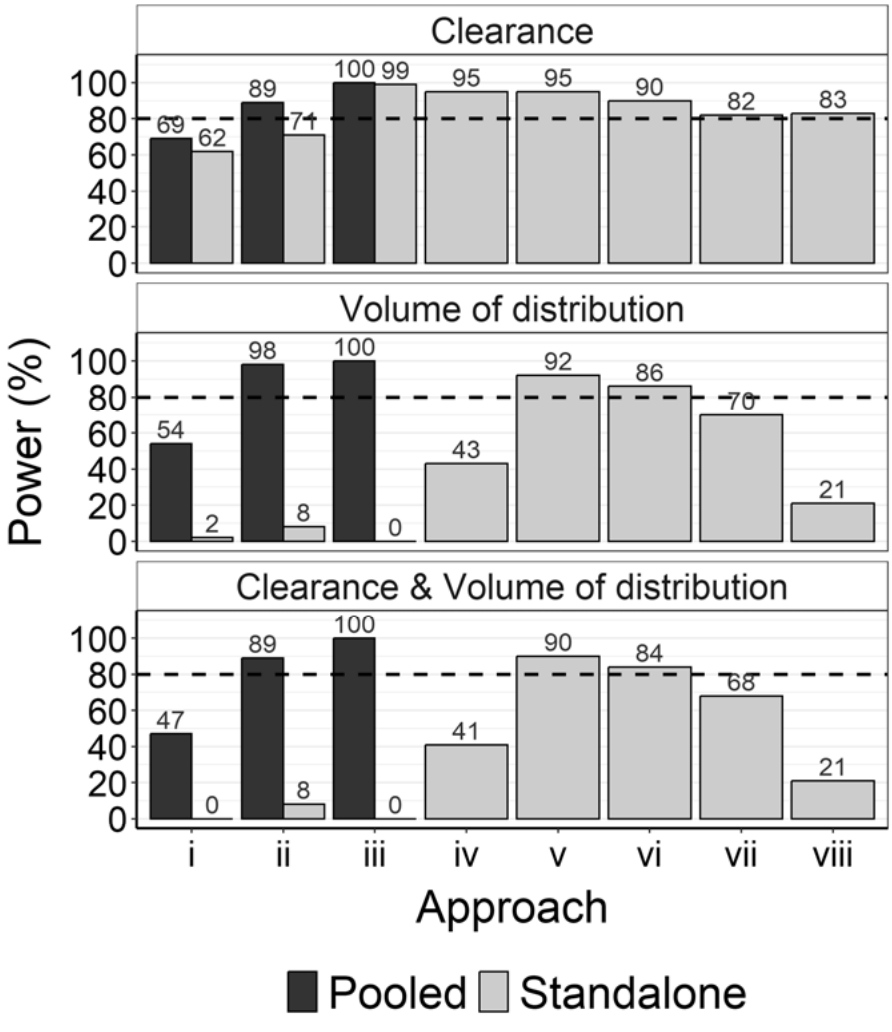

Figure 17. Power to estimate CL, V or both parameters to fulfill the FDA criterion for DalcA using all analysis approaches. Pooled analysis of adult and pediatric data is indicated by black bars whereas standalone analysis of pediatric data is indicated by grey bars. The dashed horizontal line marks 80 % power.

In a case of standalone analysis of pediatric data, the Bayesian priors' methods were robust to estimate CL and V. DalcA was in general more sensitive to reducing the informativeness of the priors. It is important, yet difficult to determine what a good level of informativeness is. When performing analysis

for drugs with complex PK and high variability, a trade-off has to be made between level of confidence in the priors and confidence on the study design to be sufficient for characterization of the PK in children. As blood sampling for PK assessment is sparser in children, it is important to have informative analysis methods. Bayesian priors has previously been reported in the literature (96) to provide stabilization of parameter estimation when analyzing different types of sparse data (97–100). In addition, a problem with pooling relatively rich adult data with sparser data from children is that the adult data may drive parameter estimation, a problem which can be handled using priors. However, issues arise if the priors are not reliable, or if PK is notably different in children compared to adults. It is therefore important to confirm the priors when data in children is available. If adult and pediatric data can be pooled, simple approaches such as iii can be sufficient to meet the precision criterion suggested by FDA (81). As approach ii is dependent on formalizing a simplified model, it is not as time efficient.

Different drug development programs in different disease areas govern the relative importance of accurate estimation of CL and V. In a case which the drug is to be taken over long time (DalcA prophylactic treatment), steady state PK is important and CL might be focused on. But for an on-demand treatment setting such as the use-case of MarzAA, both parameters might be of equal interest. The use of modelling and simulations has been encouraged from a regulatory point of view and in this work, such methods were applied to optimize the sampling schedule and to demonstrate different approaches for powering a PK trial in children with a fixed sample size. Although these results are specific for two hemostatic drugs, they may be generalizable to other drugs but yet to be confirmed.

Conclusions and perspectives

In contrast to TB which is classified as a global pandemic, HA/HB represent rare inherited diseases. Due to TB being prevalent in developing countries and HA/HB being a rare disease, these conditions receive less attention compared to other diseases such as cancer or metabolic disorders which are on the rise in Western societies. Nevertheless, both TB and HA/HB can significantly reduce quality of life with subsequent negative impact on society. Novel treatments that are shorter and efficacious are needed for TB and alternative formulations that can improve management of HA/HB are welcomed. Due to relatively low market incentives and difficulties in recruitment, respectively, it is essential that drug development initiatives targeting these diseases are executed in a methodologically rigorous manner. Moreover, the resulting information in early clinical trials must be leveraged for informing dose selection and study design determination in later more deterministic clinical trials. The projects in this thesis aimed to develop and apply pharmacometric approaches to support dose selection and to inform study designs in clinical TB and HA/HB drug development programs.

In Papers I and II, a standardized model-based analysis approach was presented including power calculations which shed light on number of patients needed to detect EBA or to differentiate EBA between two treatment arms. Using the framework, data from a phase IIa clinical trial was analyzed to evaluate the EBA of meropenem. It was found that lowering the WHO recommended dose regimen that suffer from poor tolerability may impair activity. In addition, poor tolerability remained which has led to questioning the clinical utility of meropenem for management of TB. The same analysis framework has been successfully applied to several EBA trials that are planned to be reported in peer-reviewed scientific journals.

In Papers III-V, sparse PK information from several trials were analyzed to inform dose selection in pivotal trials for novel hemostatic on-demand and prophylactic SC treatment alternatives for HA/HB. It was shown how preclinical potency information can be leveraged in conjunction with clinical information on desirable levels of FVIIa and FIX to inform dose selection in adults and pediatric subjects. Pediatric extrapolation strategies were presented to increase the certainty for first-in-pediatric dose selection using an exposure-matching strategy. These projects showed that model-based methods can be applied to leverage accumulated data in early clinical development to inform

late, often longer and costlier clinical trials. Specifically, a stratified dosing approach was developed in Paper V for adults down to 2 years of age to change the HB phenotype to mild and reduce risk for spontaneous bleeding events. For MarzAA, the work in this thesis facilitated the dose selection of 60 µg/kg given in three hourly intervals in a pivotal phase III trial in adults for treatment of acute bleeding events. Although the clinical trial was terminated earlier than planned due to other reasons than efficacy or safety (Paper VI), the resulting efficacy data following the dose supported by Paper III showed promising results. A model-based Markov analysis approach was applied in Paper VI which concluded that the novel SC alternative is likely to match the efficacy of current IV alternatives without compromising safety. In addition, it was concluded that bleeding location, age and BDBP may be clinically relevant factors that impact early bleed resolution, an important aspect as failure to reach hemostasis early increase risk of bleeding related sequalae.

The last part of this thesis (Paper VII) used the models developed in Papers III and V to optimize the pediatric sampling schedule for MarzAA and DalcA, to characterize the PK in children with sufficient certainty. Several model-based approaches were presented for prospectively powering trials aiming to study PK in children. Bayesian prior methods were concluded to perform well, but alternative simpler methods were suggested when pooling of data can be made.

To conclude, the work performed and presented within this thesis has contributed to the development of drugs against TB and HA/HB. Model-informed drug development strategies has been developed to quantify drug activity and differentiation of activity between treatment arms for anti-tubercular drugs and to support dose selection, study design, and efficacy evaluation for novel hemostatic drugs throughout all clinical development phases in adults and pediatric subjects.

Populärvetenskaplig sammanfattning

Tuberkulos och hemofili är två sjukdomar som påverkar människor över hela världen med betydande konsekvenser för individen och samhället. Till skillnad från tuberkulos som sprids via luften och lungorna, är hemofili en genetisk sjukdom. Varje år insjuknar miljontals människor tuberkulos och ett fåtal diagnosticeras med hemofili. Ingen av dessa sjukdomar är särskilt frekvent omtalade i vår närhet då tuberkulos främst drabbar människor i utvecklingsländer medan hemofili är en sällsynt sjukdom. Den allra vanligaste formen av tuberkulos drabbar lungorna medan hemofili leder till en reducerad förmåga att stoppa blödningar. Tuberkulospatienter insjuknar med hosta, svaghetskänsla och nedsatt aptit vilket kan leda till dödlig följd vid utebliven behandling. Hemofilipatienter har en nedsatt förmåga att koagulera blod, en livsviktig process som skyddar kroppen från att förblöda vid trauma. Tuberkulos behandlas idag i de flesta fallen effektivt, men kräver flera månaders lång behandling med många läkemedel som orsakar biverkningar. Till skillnad från tuberkulos som oftast går att bota, har klassisk hemofilibehandling utgjorts av livslång droppersättning av koagulationsfaktorer direkt till blodet för att återställa koaguleringsförmågan.

Läkemedelsutvecklingsprocessen sker i noggrant myndighetsreglerade steg. Efter att det nya läkemedlets molekyl har identifierats och valts för att tas vidare i klinisk prövning baserat på lovande resultat i prekliniska försök så är huvudutmaningen att välja dos i människa. Att hitta dosen som maximerar sannolikheten för en gynnsam effekt och minimerar risken för biverkningar är huvudmålet i början av klinisk läkemedelsutveckling. Ett kraftfullt verktyg för att möta utmaningen är att applicera farmakometrisk metodologi. Farmakometri är vetenskapen som går ut på att utveckla och använda matematiska modeller som beskriver ett läkemedels upptag, fördelning och utsöndring från kroppen, även känt som farmakokinetik, och läkemedlets påverkan eller effekt på kroppen, även känt som farmakodynamik. När sambanden är fastslagna går det att utföra virtuella kliniska studier innan läkemedlet ges till fler patienter. Fördelen blir att man kan undersöka hur olika doser och doseringsstrategier kan påverka effekten eller biverkningen av intresse. Vidare möjliggör även de virtuella studierna planering för studier i barn, en målgrupp som historiskt på grund av etiska anledningar inkluderats i lägre grad inom klinisk läkemedelsprövning. De virtuella kliniska studierna informerar om hur man bör utföra nästa studie för att kunna påvisa en effekt, eller kanske ännu viktigare, att inte

utföra nästa studie överhuvudtaget. De flesta läkemedelsprojekten når aldrig patienter och om projekt stängs ner i tidigare skede kan man fokusera på andra läkemedel i utveckling. Det är en viktig aspekt för läkemedelsutveckling mot tuberkulos och hemofili, då ekonomiska incitament för utveckling av nya läkemedel mot sjukdomarna i fråga har varit relativt svaga. Detta på grund av att konsumenten av läkemedlet antingen inte förväntats ha råd med behandlingen, eller varit få i antal.

Målet med den här avhandlingen var att utveckla och applicera farmakometriska modeller för att underlätta beslutsfattande i utveckling av nya läkemedel för tuberkulos och hemofili. I Artikel I utvecklades en metod för att effektivt analysera kliniska studier för tuberkulosläkemedel. Metoden användes sedan i Artikel II för att analysera en klinisk studie som undersökte om meropenem, som i sin nuvarande godkända dos nivå har dålig biverkningsprofil, också ger effekt i lägre dos med förhoppning om färre biverkningar. Arbetet konkluderade att en sänkning av dosen kan påverka effekten negativt över 14 dagar. Analysmetoden som presenterades har blivit en standard för att analysera liknande studier. I Artiklarna III-V utvecklades modeller baserat på farmakokinetiska data från två nya hemofililäkemedel för förebyggande behandling men även behandling av en akut blödning. Modellerna användes för att utföra virtuella kliniska studier i vuxna och barn, vilket hjälpte att utforma två nya studier. En av studierna utfördes inom tidsramen för avhandlingen och analyserades i Artikel VI, där en ny subkutan behandling för akuta blödningar jämfördes med dagens dropp-baserade standard. Fördelen med det nya läkemedlet är att patienter inte behöver söka sig till en klinik vid symptom, utan själva kan ta läkemedlet likt hur diabetespatienter idag administrerar insulin. En matematisk Markov modell byggdes och slutsatsen från analysen var att det inte finns någon statistisk signifikant skillnad i effekt mellan det nya läkemedlet och dagens standard. Vidare upptäcktes patientkaraktäristika som kan leda till fördröjd effekt, vilket är viktigt då en fördröjd behandling av blödningar kan motivera flera doser eller en högre dos. Det sista arbetet i Artikel VII återanvände modellerna som byggdes i Artikel III och V för att utveckla och jämföra metoder som syftar till att planera farmakokinetiska studier i barn. Syftet med studierna är ofta att undersöka farmakokinetiken och att innan man utför studien måste visa att studien har en hög sannolikhet att informera om farmakokinetiken i barn. Artikel VII konkluderade att med farmakometriska metoder kan undersöka viktiga farmakokinetiska processer trots få rekryterade barn, vilket är viktigt för sällsynta sjukdomar som hemofili.

Sammanfattningsvis har projekten inom den här avhandlingen bidragit till utveckling av läkemedel mot tuberkulos och hemofili. Farmakometriska modeller och strategier har utvecklats för kvantifiering av läkemedelseffekt, val av dos-styrka samt beslutsfattning i studieupplägg för nya tuberkulos- och hemofili-läkemedel i vuxna och barn.

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References

1. Brook K, Bennett J, Desai SP. The Chemical History of Morphine: An 8000-year Journey, from Resin to de-novo Synthesis. J Anesth Hist. 2017 Apr;3(2):50–5.
2. Commissioner O of the. The History of Drug Regulation. FDA [Internet]. 2023 Mar 30 [cited 2023 Oct 20]; Available from: https://www.fda.gov/about-fda/histories-fda-regulated-products/history-drug-regulation
3. Commissioner O of the. FDA. FDA; 2020 [cited 2023 Oct 20]. The Drug Development Process. Available from: https://www.fda.gov/patients/learn-about-drug-and-device-approvals/drug-development-process
4. DiMasi JA, Grabowski HG, Hansen RW. Innovation in the pharmaceutical industry: New estimates of R&D costs. Journal of Health Economics. 2016 May 1;47:20–33.
5. Schuhmacher A, Hinder M, von Stegmann und Stein A, Hartl D, Gassmann O. Analysis of pharma R&D productivity – a new perspective needed. Drug Discovery Today. 2023 Oct 1;28(10):103726.
6. Bunnage ME. Getting pharmaceutical R&D back on target. Nat Chem Biol. 2011 Jun;7(6):335–9.
7. Sun D, Gao W, Hu H, Zhou S. Why 90% of clinical drug development fails and how to improve it? Acta Pharm Sin B. 2022 Jul;12(7):3049–62.
8. Kola I, Landis J. Can the pharmaceutical industry reduce attrition rates? Nat Rev Drug Discov. 2004 Aug;3(8):711–6.
9. Hay M, Thomas DW, Craighead JL, Economides C, Rosenthal J. Clinical development success rates for investigational drugs. Nat Biotechnol. 2014 Jan;32(1):40–51.
10. Global Tuberculosis Report 2022 [Internet]. [cited 2023 Dec 8]. Available from: https://www.who.int/teams/global-tuberculosis-programme/tb-reports/global-tuberculosis-report-2022
11. Vynnycky E, Fine PE. Lifetime risks, incubation period, and serial interval of tuberculosis. Am J Epidemiol. 2000 Aug 1;152(3):247–63.
12. WHO consolidated guidelines on tuberculosis: module 4: treatment: drug-susceptible tuberculosis treatment [Internet]. [cited 2023 Nov 8]. Available from: https://www.who.int/publications-detail-redirect/9789240048126
13. WHO consolidated guidelines on tuberculosis. Module 4: treatment - drug-resistant tuberculosis treatment, 2022 update [Internet]. [cited 2023 Nov 8]. Available from: https://www.who.int/publications-detail-redirect/9789240063129
14. Mannucci PM, Tuddenham EG. The hemophilias--from royal genes to gene therapy. N Engl J Med. 2001 Jun 7;344(23):1773–9.
15. Blanchette VS, Key NS, Ljung LR, Manco-Johnson MJ, van den Berg HM, Srivastava A, et al. Definitions in hemophilia: communication from the SSC of the ISTH. J Thromb Haemost. 2014 Nov;12(11):1935–9.

16. Castaman G, Matino D. Hemophilia A and B: molecular and clinical similarities and differences. Haematologica. 2019 Sep;104(9):1702–9.
17. Jansen NWD, Roosendaal G, Lafeber FPJG. Understanding haemophilic arthropathy: an exploration of current open issues. Br J Haematol. 2008 Dec;143(5):632–40.
18. Okaygoun D, Oliveira DD, Soman S, Williams R. Advances in the management of haemophilia: emerging treatments and their mechanisms. Journal of Biomedical Science. 2021 Sep 14;28(1):64.
19. Stoner KL, Harder H, Fallowfield LJ, Jenkins VA. Intravenous versus Subcutaneous Drug Administration. Which Do Patients Prefer? A Systematic Review. Patient. 2014 Jul 12;
20. Commissioner O of the. FDA. FDA; 2022 [cited 2022 Nov 28]. FDA Approves First Gene Therapy to Treat Adults with Hemophilia B. Available from: https://www.fda.gov/news-events/press-announcements/fda-approves-first-gene-therapy-treat-adults-hemophilia-b
21. Commissioner O of the. FDA. FDA; 2023 [cited 2023 Dec 5]. FDA Approves First Gene Therapy for Adults with Severe Hemophilia A. Available from: https://www.fda.gov/news-events/press-announcements/fda-approves-first-gene-therapy-adults-severe-hemophilia
22. Srivastava A, Brewer AK, Mauser-Bunschoten EP, Key NS, Kitchen S, Llinas A, et al. Guidelines for the management of hemophilia. Haemophilia. 2013 Jan;19(1):e1-47.
23. Research C for DE and. FDA approves emicizumab-kxwh for prevention and reduction of bleeding in patients with hemophilia A with factor VIII inhibitors. FDA [Internet]. 2019 Feb 9 [cited 2023 Dec 5]; Available from: https://www.fda.gov/drugs/resources-information-approved-drugs/fda-approves-emicizumab-kxwh-prevention-and-reduction-bleeding-patients-hemophilia-factor-viii
24. Oldenburg J, Mahlangu JN, Kim B, Schmitt C, Callaghan MU, Young G, et al. Emicizumab Prophylaxis in Hemophilia A with Inhibitors. New England Journal of Medicine. 2017 Aug 31;377(9):809–18.
25. Gruppo RA, Malan D, Kapocsi J, Nemes L, Hay CRM, Boggio L, et al. Phase 1, single-dose escalating study of marzeptacog alfa (activated), a recombinant factor VIIa variant, in patients with severe hemophilia. Journal of Thrombosis and Haemostasis. 2018;16(10):1984–93.
26. Mahlangu J, Levy H, Kosinova MV, Khachatryan H, Korczowski B, Makhaldiani L, et al. Subcutaneous engineered factor VIIa marzeptacog alfa (activated) in hemophilia with inhibitors: Phase 2 trial of pharmacokinetics, pharmacodynamics, efficacy, and safety. Res Pract Thromb Haemost. 2021 Aug 17;5(6):e12576.
27. Crimson 1: A Phase 3 study to evaluate the efficacy and safety of subcutaneous marzeptocog alfa (activated) for on-demand treatment of bleed events in subjects with hemophilia A or B, with inhibitors [Internet]. ISTH Congress Abstracts. [cited 2022 Jul 28]. Available from: https://abstracts.isth.org/abstract/crimson-1-a-phase-3-study-to-evaluate-the-efficacy-and-safety-of-subcutaneous-marzeptocog-alfa-activated-for-on-demand-treatment-of-bleed-events-in-subjects-with-hemophilia-a-or-b-with-inhibitors/
28. Faraj A, Knudsen T, Desai S, Neuman L, Blouse GE, Simonsson USH. Phase III dose selection of marzeptacog alfa (activated) informed by population pharmacokinetic modeling: A novel hemostatic drug. CPT Pharmacometrics Syst Pharmacol. 2022 Dec;11(12):1628–37.

29. Faraj A, van Wijk RC, Neuman L, Desai S, Blouse GE, Knudsen T, et al. Model-informed pediatric dose selection of marzeptacog alfa (activated): An exposure matching strategy. CPT Pharmacometrics Syst Pharmacol. 2023 Jul;12(7):977–87.
30. Mannucci PM. Hemophilia therapy: the future has begun. Haematologica. 2020 Mar 1;105(3):545–53.
31. Summary Basis of Decision for Alhemo [Internet]. [cited 2023 Dec 4]. Available from: https://dhpp.hpfb-dgpsa.ca/review-documents/resource/SBD1685635290781
32. Jindani A, Aber VR, Edwards EA, Mitchison DA. The early bactericidal activity of drugs in patients with pulmonary tuberculosis. Am Rev Respir Dis. 1980 Jun;121(6):939–49.
33. Research C for DE and. Pulmonary Tuberculosis: Developing Drugs for Treatment [Internet]. FDA; 2022 [cited 2023 Dec 11]. Available from: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/pulmonary-tuberculosis-developing-drugs-treatment
34. Diacon AH, Dawson R, von Groote-Bidlingmaier F, Symons G, Venter A, Donald PR, et al. Bactericidal activity of pyrazinamide and clofazimine alone and in combinations with pretomanid and bedaquiline. Am J Respir Crit Care Med. 2015 Apr 15;191(8):943–53.
35. Faraj A, Svensson RJ, Diacon AH, Simonsson USH. Drug Effect of Clofazimine on Persisters Explains an Unexpected Increase in Bacterial Load in Patients. Antimicrob Agents Chemother. 2020 Apr 21;64(5):e01905-19.
36. Mockeliunas L, Faraj A, van Wijk RC, Upton CM, van den Hoogen G, Diacon AH, et al. Standards for model-based early bactericidal activity analysis and sample size determination in tuberculosis drug development. Front Pharmacol. 2023;14:1150243.
37. Davies G, Boeree M, Hermann D, Hoelscher M. Accelerating the transition of new tuberculosis drug combinations from Phase II to Phase III trials: New technologies and innovative designs. PLoS Med. 2019 Jul 9;16(7):e1002851.
38. EMA. European Medicines Agency. 2018 [cited 2022 Feb 17]. Addendum to the note for guidance on evaluation of medicinal products indicated treatment bacterial infections specifically address clinical development new agents treat disease due Mycobacterium Tuberculosis. Available from: https://www.ema.europa.eu/en/addendum-note-guidance-evaluation-medicinal-products-indicated-treatment-bacterial-infections
39. Research C for BE and. U.S. Food and Drug Administration. FDA; 2020 [cited 2022 Feb 17]. Human Gene Therapy for Hemophilia. Available from: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/human-gene-therapy-hemophilia
40. EMA. European Medicines Agency. 2018 [cited 2023 Nov 21]. Clinical investigation of recombinant and human plasma-derived factor VIII products - Scientific guideline. Available from: https://www.ema.europa.eu/en/clinical-investigation-recombinant-human-plasma-derived-factor-viii-products-scientific-guideline
41. Lentz SR, Misgav M, Ozelo M, Šalek SZ, Veljkovic D, Recht M, et al. Results from a large multinational clinical trial (guardian™1) using prophylactic treatment with turoctocog alfa in adolescent and adult patients with severe haemophilia A: safety and efficacy. Haemophilia. 2013;19(5):691–7.

42. Shima M, Hanabusa H, Taki M, Matsushita T, Sato T, Fukutake K, et al. Long-term safety and efficacy of emicizumab in a phase 1/2 study in patients with hemophilia A with or without inhibitors. Blood Adv. 2017 Sep 27;1(22):1891–9.
43. Neuman L, Desai S, Del Greco F, Knudsen T, Levy H. Phase 1/2 Study to Evaluate the Pharmacokinetics, Pharmacodynamics, Safety, and Efficacy of Marzeptacog Alfa (activated) in the Treatment of Episodic Bleeding in Subjects with Inherited Bleeding Disorders. Blood. 2020 Nov 5;136(Supplement 1):32–3.
44. Levy H, Makhaldiani L, Iosava G, Mahlangu J, Kosinova MV, Khacchatryan H, et al. Phase 2/3 Trial of Subcutaneously Administered Marzeptacog Alfa (activated) an Engineered FVIIa in Hemophilia with Inhibitors - Pharmacokinetics, Pharmacodynamics, Safety and Efficacy. Blood. 2018 Nov 29;132(Supplement 1):1198.
45. You CW, Hong SB, Kim S, Shin HJ, Kim JS, Han JW, et al. Safety, pharmacokinetics, and pharmacodynamics of a next-generation subcutaneously administered coagulation factor IX variant, dalcinonacog alfa, in previously treated hemophilia B patients. Journal of Thrombosis and Haemostasis. 2021;19(4):967–75.
46. Mahlangu J, Levy H, Lee M, Del Greco F. Efficacy and safety of subcutaneous prophylaxis with dalcinonacog alfa in adults with haemophilia B. Haemophilia. 2021 Jul;27(4):574–80.
47. Lalonde RL, Kowalski KG, Hutmacher MM, Ewy W, Nichols DJ, Milligan PA, et al. Model-based drug development. Clin Pharmacol Ther. 2007 Jul;82(1):21–32.
48. Peck CC, Barr WH, Benet LZ, Collins J, Desjardins RE, Furst DE, et al. Opportunities for integration of pharmacokinetics, pharmacodynamics, and toxicokinetics in rational drug development. J Clin Pharmacol. 1994 Feb;34(2):111–9.
49. Sheiner LB. Learning versus confirming in clinical drug development. Clinical Pharmacology & Therapeutics. 1997;61(3):275–91.
50. Madabushi R, Seo P, Zhao L, Tegenge M, Zhu H. Review: Role of Model-Informed Drug Development Approaches in the Lifecycle of Drug Development and Regulatory Decision-Making. Pharm Res. 2022;39(8):1669–80.
51. Li RJ, Ma L, Li F, Li L, Bi Y, Yuan Y, et al. Model-Informed Approach Supporting Drug Development and Regulatory Evaluation for Rare Diseases. J Clin Pharmacol. 2022 Dec;62 Suppl 2:S27–37.
52. Wiley.com [Internet]. [cited 2023 Dec 13]. Pharmacometrics: The Science of Quantitative Pharmacology | Wiley. Available from: https://www.wiley.com/en-us/Pharmacometrics%3A+The+Science+of+Quantitative+Pharmacology-p-9781118679517
53. Rekić D, Röshammar D, Simonsson USH. Model based design and analysis of phase II HIV-1 trials. J Pharmacokinet Pharmacodyn. 2013 Aug;40(4):487–96.
54. Karlsson KE, Vong C, Bergstrand M, Jonsson EN, Karlsson MO. Comparisons of Analysis Methods for Proof-of-Concept Trials. CPT Pharmacometrics Syst Pharmacol. 2013 Jan 16;2(1):e23.
55. Svensson RJ, Gillespie SH, Simonsson USH. Improved power for TB Phase IIa trials using a model-based pharmacokinetic-pharmacodynamic approach compared with commonly used analysis methods. J Antimicrob Chemother. 2017 Aug 1;72(8):2311–9.

56. Petersson KJF, Hanze E, Savic RM, Karlsson MO. Semiparametric distributions with estimated shape parameters. Pharm Res. 2009 Sep;26(9):2174–85.
57. Research C for DE and. U.S. Food and Drug Administration. FDA; 2020 [cited 2022 Aug 30]. Pediatric Study Plans: Content of and Process for Submitting Initial Pediatric Study Plans and Amended Initial Pediatric Study Plans. Available from: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/pediatric-study-plans-content-and-process-submitting-initial-pediatric-study-plans-and-amended
58. EMA. European Medicines Agency. 2018 [cited 2022 Aug 30]. Paediatric investigation plans: questions and answers. Available from: https://www.ema.europa.eu/en/human-regulatory/research-development/paediatric-medicines/paediatric-investigation-plans/paediatric-investigation-plans-questions-answers
59. Green FG, Park K, Burckart GJ. Methods Used for Pediatric Dose Selection in Drug Development Programs Submitted to the US FDA 2012-2020. J Clin Pharmacol. 2021 Jun;61 Suppl 1:S28–35.
60. Manolis E, Musuamba FT, Karlsson KE. The European Medicines Agency Experience With Pediatric Dose Selection. The Journal of Clinical Pharmacology. 2021;61(S1):S22–7.
61. Anderson BJ, Holford NHG. Mechanism-based concepts of size and maturity in pharmacokinetics. Annu Rev Pharmacol Toxicol. 2008;48:303–32.
62. Calvier EAM, Krekels EHJ, Välitalo PAJ, Rostami-Hodjegan A, Tibboel D, Danhof M, et al. Allometric Scaling of Clearance in Paediatric Patients: When Does the Magic of 0.75 Fade? Clin Pharmacokinet. 2017 Mar;56(3):273–85.
63. Tegenge MA, Mahmood I. Age- and Bodyweight-dependent Allometric Exponent Model for Scaling Clearance and Maintenance Dose of Theophylline From Neonates to Adults. Therapeutic Drug Monitoring. 2018 Oct;40(5):635–41.
64. Mahmood I. Prediction of drug clearance in children: a review of different methodologies. Expert Opin Drug Metab Toxicol. 2015 Apr;11(4):573–87.
65. Liu T, Ghafoori P, Gobburu JVS. Allometry Is a Reasonable Choice in Pediatric Drug Development. The Journal of Clinical Pharmacology. 2017;57(4):469–75.
66. Mahmood I. Allometric extrapolation of factors VII, VIII and IX clearance in children from adults. Journal of Thrombosis and Haemostasis. 2012;10(8):1609–13.
67. Mahmood I. Prediction of Drug Clearance in Premature and Mature Neonates, Infants, and Children ≤2 Years of Age: A Comparison of the Predictive Performance of 4 Allometric Models. The Journal of Clinical Pharmacology. 2016;56(6):733–9.
68. Mahmood I. Initiation of Pediatric Clinical Trials for Coagulation Factors: Application of Pharmacokinetics and Allometry to First-in-Pediatric Dose Selection. J Clin Pharmacol. 2019 Jun;59(6):829–34.
69. General Clinical Pharmacology Considerations for Pediatric Studies for Drugs and Biological Products. :28.
70. Khandelwal A, Harling K, Jonsson EN, Hooker AC, Karlsson MO. A fast method for testing covariates in population PK/PD Models. AAPS J. 2011 Sep;13(3):464–72.
71. Markov AA. An Example of Statistical Analysis of the Text of Eugene Onegin Illustrating the Association of Trials into a Chain. Bulletin de l'Acadamie Imperiale des Sciences de St Petersburg. 1913;7:153–62.

72. Bergstrand M, Söderlind E, Eriksson UG, Weitschies W, Karlsson MO. A semi-mechanistic modeling strategy for characterization of regional absorption properties and prospective prediction of plasma concentrations following administration of new modified release formulations. Pharm Res. 2012 Feb;29(2):574–84.
73. Schindler E, Karlsson MO. A Minimal Continuous-Time Markov Pharmacometric Model. AAPS J. 2017 Sep 1;19(5):1424–35.
74. Karlsson MO, Savic RM. Diagnosing Model Diagnostics. Clinical Pharmacology & Therapeutics. 2007;82(1):17–20.
75. Research C for DE and. U.S. Food and Drug Administration. FDA; 2022 [cited 2022 Aug 2]. Population Pharmacokinetics. Available from: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/population-pharmacokinetics
76. Bergstrand M, Hooker AC, Wallin JE, Karlsson MO. Prediction-corrected visual predictive checks for diagnosing nonlinear mixed-effects models. AAPS J. 2011 Jun;13(2):143–51.
77. NHANES - National Health and Nutrition Examination Survey Homepage [Internet]. 2022 [cited 2022 Feb 14]. Available from: https://www.cdc.gov/nchs/nhanes/index.htm
78. Vong C, Bergstrand M, Nyberg J, Karlsson MO. Rapid sample size calculations for a defined likelihood ratio test-based power in mixed-effects models. AAPS J. 2012 Jun;14(2):176–86.
79. White GC, Rosendaal F, Aledort LM, Lusher JM, Rothschild C, Ingerslev J, et al. Definitions in hemophilia. Recommendation of the scientific subcommittee on factor VIII and factor IX of the scientific and standardization committee of the International Society on Thrombosis and Haemostasis. Thromb Haemost. 2001 Mar;85(3):560.
80. Guideline on clinical investigation of recombinant and human plasma-derived factor IX products.
81. Wang Y, Jadhav PR, Lala M, Gobburu JV. Clarification on Precision Criteria to Derive Sample Size When Designing Pediatric Pharmacokinetic Studies. The Journal of Clinical Pharmacology. 2012;52(10):1601–6.
82. Beal SL, Sheiner LB, Boeckmann AJ, Bauer RJ. 1989-2011. NONMEM Users Guides. Icon Development Solutions, Ellicott City, Maryland, USA.
83. R: The R Project for Statistical Computing [Internet]. [cited 2022 Jul 29]. Available from: https://www.r-project.org/
84. PsN :: Home [Internet]. [cited 2021 Feb 16]. Available from: https://uupharmacometrics.github.io/PsN/
85. Keizer RJ, Karlsson MO, Hooker A. Modeling and Simulation Workbench for NONMEM: Tutorial on Pirana, PsN, and Xpose. CPT Pharmacometrics Syst Pharmacol. 2013 Jun 26;2:e50.
86. Goletti D, Lee MR, Wang JY, Walter N, Ottenhoff THM. Update on tuberculosis biomarkers: From correlates of risk, to correlates of active disease and of cure from disease. Respirology. 2018;23(5):455–66.
87. Janssen S, Upton CM, De Jager VR, Faraj A, Pahar M, Miranda IDS, et al. Cough as Noninvasive Biomarker for Monitoring Tuberculosis Treatment: A Proof-of-Concept Study. Ann Am Thorac Soc. 2023 Dec;20(12):1822–5.
88. Robbie GJ, Zhao L, Mondick J, Losonsky G, Roskos LK. Population pharmacokinetics of palivizumab, a humanized anti-respiratory syncytial virus monoclonal antibody, in adults and children. Antimicrob Agents Chemother. 2012 Sep;56(9):4927–36.

89. Temrikar ZH, Suryawanshi S, Meibohm B. Pharmacokinetics and Clinical Pharmacology of Monoclonal Antibodies in Pediatric Patients. Paediatr Drugs. 2020 Apr;22(2):199–216.
90. Villar A, Aronis S, Morfini M, Santagostino E, Auerswald G, Thomsen HF, et al. Pharmacokinetics of activated recombinant coagulation factor VII (NovoSeven®) in children vs. adults with haemophilia A. Haemophilia. 2004;10(4):352–9.
91. Mahmood I. Prediction of Clearance of Monoclonal and Polyclonal Antibodies and Non-Antibody Proteins in Children: Application of Allometric Scaling. Antibodies (Basel). 2020 Aug 5;9(3):40.
92. Research C for BE and. BeneFIX. FDA [Internet]. 2022 Feb 14 [cited 2022 Sep 8]; Available from: https://www.fda.gov/vaccines-blood-biologics/approved-blood-products/benefix
93. Barnes RFW, Cramer TJ, Sait AS, Kruse-Jarres R, Quon DVK, von Drygalski A. The Hypertension of Hemophilia Is Not Explained by the Usual Cardiovascular Risk Factors: Results of a Cohort Study. Int J Hypertens. 2016;2016:2014201.
94. Sartori MT, Bilora F, Zanon E, Varvarikis C, Saggiorato G, Campagnolo E, et al. Endothelial dysfunction in haemophilia patients. Haemophilia. 2008 Sep;14(5):1055–62.
95. Carcao M, Goudemand J. INHIBITORS IN HEMOPHILIA: A PRIMER. :24.
96. Chan Kwong AHXP, Calvier EAM, Fabre D, Gattacceca F, Khier S. Prior information for population pharmacokinetic and pharmacokinetic/pharmacodynamic analysis: overview and guidance with a focus on the NONMEM PRIOR subroutine. J Pharmacokinet Pharmacodyn. 2020 Oct;47(5):431–46.
97. Mondick J, Johnston C. A Modeling and Simulation Primer for the Design of Pediatric Pharmacokinetic Studies.
98. Knøsgaard KR, Foster DJR, Kreilgaard M, Sverrisdóttir E, Upton RN, van den Anker JN. Pharmacokinetic models of morphine and its metabolites in neonates:: Systematic comparisons of models from the literature, and development of a new meta-model. Eur J Pharm Sci. 2016 Sep 20;92:117–30.
99. Knebel W, Gastonguay MR, Malhotra B, El-Tahtawy A, Jen F, Gandelman K. Population Pharmacokinetics of Atorvastatin and Its Active Metabolites in Children and Adolescents With Heterozygous Familial Hypercholesterolemia: Selective Use of Informative Prior Distributions from Adults. The Journal of Clinical Pharmacology. 2013;53(5):505–16.
100. Lohy Das J, Rulisa S, de Vries PJ, Mens PF, Kaligirwa N, Agaba S, et al. Population Pharmacokinetics of Artemether, Dihydroartemisinin, and Lumefantrine in Rwandese Pregnant Women Treated for Uncomplicated Plasmodium falciparum Malaria. Antimicrob Agents Chemother. 2018 Oct;62(10):e00518-18.

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