

University of Groningen

Pharmacodynamics of Voriconazole in Children

Huurneman, Luc J.; Neely, Michael; Veringa, Anette; Docobo Perez, Fernando; Ramos-Martin, Virginia; Tissing, Willem; Alffenaar, Johannes; Hope, William

Published in:
Antimicrobial Agents and Chemotherapy

DOI:
[10.1128/AAC.03023-15](https://doi.org/10.1128/AAC.03023-15)

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2016

[Link to publication in University of Groningen/UMCG research database](#)

Citation for published version (APA):

Huurneman, L. J., Neely, M., Veringa, A., Docobo Perez, F., Ramos-Martin, V., Tissing, W. J., ... Hope, W. (2016). Pharmacodynamics of Voriconazole in Children: Further Steps along the Path to True Individualized Therapy. *Antimicrobial Agents and Chemotherapy*, 60(4), 2336-2342. DOI: 10.1128/AAC.03023-15

Copyright

Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

Take-down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): <http://www.rug.nl/research/portal>. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.

Pharmacodynamics of Voriconazole in Children: Further Steps along the Path to True Individualized Therapy

Luc J. Huurneman,^a Michael Neely,^b Anette Veringa,^a Fernando Docobo Pérez,^{c,d} Virginia Ramos-Martin,^c Wim J. Tissing,^e Jan-Willem C. Alffenaar,^a William Hope^c

University of Groningen, University Medical Center Groningen, Department of Clinical Pharmacy and Pharmacology, Groningen, the Netherlands^a; Laboratory of Applied Pharmacokinetics and Bioinformatics, The Saban Research Institute, and Division of Pediatric Infectious Diseases, Children's Hospital Los Angeles, University of Southern California, Los Angeles, California, USA^b; Department of Molecular and Clinical Pharmacology, Antimicrobial Pharmacodynamics and Therapeutics, University of Liverpool, Liverpool, United Kingdom^c; Unidad Intercentros de Enfermedades Infecciosas, Microbiología Clínica y Medicina Preventiva, Hospital Universitario Virgen Macarena, Seville, Spain^d; University of Groningen, University Medical Center Groningen, Beatrix Children's Hospital, Department of Pediatric Oncology, Groningen, the Netherlands^e

Voriconazole is the agent of choice for the treatment of invasive aspergillosis in children at least 2 years of age. The galactomannan index is a routinely used diagnostic marker for invasive aspergillosis and can be useful for following the clinical response to antifungal treatment. The aim of this study was to develop a pharmacokinetic-pharmacodynamic (PK-PD) mathematical model that links the pharmacokinetics of voriconazole with the galactomannan readout in children. Twelve children receiving voriconazole for treatment of proven, probable, and possible invasive fungal infections were studied. A previously published population PK model was used as the Bayesian prior. The PK-PD model was used to estimate the average area under the concentration-time curve (AUC) in each patient and the resultant galactomannan-time profile. The relationship between the ratio of the AUC to the concentration of voriconazole that induced half maximal killing (AUC/EC₅₀) and the terminal galactomannan level was determined. The voriconazole concentration-time and galactomannan-time profiles were both highly variable. Despite this variability, the fit of the PK-PD model was good, enabling both the pharmacokinetics and pharmacodynamics to be described in individual children. (AUC/EC₅₀)/15.4 predicted terminal galactomannan ($P = 0.003$), and a ratio of >6 suggested a lower terminal galactomannan level ($P = 0.07$). The construction of linked PK-PD models is the first step in developing control software that enables not only individualized voriconazole dosages but also individualized concentration targets to achieve suppression of galactomannan levels in a timely and optimally precise manner. Controlling galactomannan levels is a first critical step to maximizing clinical response and survival.

Voriconazole is an extended-spectrum triazole antifungal agent with activity against *Aspergillus* spp., *Candida* spp., *Cryptococcus neoformans*, *Fusarium* spp., and *Scedosporium apiospermum* (1). Voriconazole is licensed for use in children >12 (United States) or >2 (Europe) years of age with invasive aspergillosis (IA), fluconazole-resistant invasive *Candida* infections, or infections caused by *Scedosporium* spp. and *Fusarium* spp. and in non-neutropenic children with candidemia (2). In all patient age groups, voriconazole is a first-line agent for the treatment of IA (2, 3). The inter- and intraindividual variabilities in drug exposure are high, and some of this variability can be attributed to CYP2C19 genotype (4), impaired liver function (5), age (6), inflammation (7), and CYP2C19/CYP3A-interacting comedication (8). This coupled with a reasonably detailed understanding of the relationship between drug exposure and the probability of both therapeutic response and toxicity has led to a recommendation to use therapeutic drug monitoring (TDM) as an adjunct to routine clinical use of voriconazole (9). We have recently developed software for dosage individualization in both adults and children (10, 11). The underlying algorithms can be used to achieve desired serum drug concentration targets in both adults and children in an optimally precise manner.

In clinical settings, the galactomannan index is increasingly used as a biomarker for the diagnosis of invasive aspergillosis. According to the EORTC/MSG diagnostic criteria for invasive fungal diseases, galactomannan can be used as a microbiological criterion to establish a diagnosis of invasive aspergillosis (12). Galactomannan is a high-molecular-weight polysaccharide cell wall

fungal antigen that is released into the bloodstream during hyphal growth and angioinvasion (13). Routine sequential monitoring of serum galactomannan levels can be used for the early detection of invasive aspergillosis (14). There is now increasing interest in using galactomannan to follow the response to antifungal therapy (15). Such a strategy is supported by the observation that patients with unremittingly high circulating antigen levels tend to have a poor clinical outcome. The availability of a biomarker with both diagnostic and prognostic significance is relatively unique in infectious diseases. An understanding of galactomannan kinetics and its response to antifungal drug concentrations provides the possibility to provide true individualized antifungal therapy.

Here, we developed a linked pharmacokinetic-pharmacodynamic (PK-PD) mathematical model to describe the serum pharmacokinetics of voriconazole and the pharmacodynamics quantified in terms of the circulating galactomannan levels. Since much

Received 17 December 2015 Returned for modification 18 January 2016

Accepted 27 January 2016

Accepted manuscript posted online 1 February 2016

Citation Huurneman LJ, Neely M, Veringa A, Docobo Pérez F, Ramos-Martin V, Tissing WJ, Alffenaar J-WC, Hope W. 2016. Pharmacodynamics of voriconazole in children: further steps along the path to true individualized therapy. *Antimicrob Agents Chemother* 60:2336–2342. doi:10.1128/AAC.03023-15.

Address correspondence to William Hope, william.hope@liverpool.ac.uk.

L.J.H. and M.N. contributed equally to this article.

Copyright © 2016, American Society for Microbiology. All Rights Reserved.

of the pharmacokinetic and pharmacodynamic data were necessarily sparse, we buttressed the pharmacokinetics by using richer data obtained from the early phases of drug development. Such an approach enabled us to ensure robust estimates of the pharmacokinetics, which would have otherwise been extremely difficult or resulted in biased parameter estimates. The development of a linked PK-PD model is a further step in the provision of true individualized therapy where a drug is administered to control a biomarker that is itself intricately linked to therapeutic responses and optimal clinical outcomes.

MATERIALS AND METHODS

Patients. All patients aged <18 years receiving voriconazole, with at least one voriconazole serum concentration and galactomannan level measured, within the 9-year period from January 2005 to March 2014 were eligible for inclusion in this study. The medical, pharmacy, and laboratory records at the University Medical Center Groningen were reviewed. Demographic, microbiological, and clinical data were collected using standardized case report forms. The voriconazole treatment regimen and serum concentrations of voriconazole were also collected. Information that could potentially influence the voriconazole serum concentrations was identified and reviewed for potential inclusion of these serum concentrations into the population PK-PD model. The EORTC/MSG criteria (12) were used to determine the probability of invasive fungal disease for each patient at the start of voriconazole therapy. The Medical Ethical review board of the University Medical Center Groningen (metc 2013-491) waived the requirement to obtain informed consent from individual patients.

TDM. All patients at the University Medical Center Groningen who were treated with voriconazole underwent therapeutic drug monitoring (TDM). The first sample was typically taken after 2 days, and results were reported the same day. The therapeutic trough concentration targets were >1 mg/liter and <6 mg/liter. Concentrations outside these values prompted a change in dosage. There was no algorithm for dosage adjustment, but typically the dose of voriconazole was increased or decreased by 30 to 50% and concentrations were remeasured after several days. Galactomannan was not used to make decisions about dosage adjustment.

Voriconazole assay. The voriconazole serum concentrations were determined using a validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method (16). All measurements were performed on a Thermo Fisher (San Jose, CA, USA) triple-quadrupole LC-MS/MS with a Finnigan Surveyor LC pump and a Finnigan Surveyor autosampler, which was set at a temperature of 20°C. The Finnigan TSQ Quantum Discovery mass selective detector was operating in electrospray positive-ionization mode and performed selected reaction monitoring. The ion source spray voltage was set at 3,500 V, the sheath and auxiliary gas pressures at 35 and 5 arbitrary units, respectively, and the capillary temperature at 350°C. Cyanoimipramine was used as internal standard. Analyses were performed on a 50-mm by 2.1-mm C₁₈ 5-μm analytic column (HYPURITY Aquastar; Interscience Breda, The Netherlands). The column temperature was set at 20°C. The mobile phase consisted of an aqueous buffer (containing ammonium acetate [10 g/liter water], acetic acid [35 mg/liter water], and trifluoroacetic anhydride [2 ml/liter water]), water, and acetonitrile. Chromatographic separation was performed using a gradient with a flow of 0.3 ml/min and a run time of 3.6 min. Sample preparation was performed by protein precipitation and found to be suitable, resulting in linear calibration curves in the range of 0.1 to 10 mg/liter. The peak height ratios of voriconazole and the internal standard were used to calculate concentrations. This method was validated in accordance with the Guidance for Industry Bioanalytical Method Validation of the Food and Drug Administration. The validation showed an overall bias ranging from 0.1 to 2.3%, a within-run coefficient of variation (CV) ranging from 1.9 to 7.8%, and a between-run CV ranging from 0.0 to 3.1%.

Galactomannan assay. The samples for the determination of the galactomannan index were measured using the Platelia *Aspergillus* enzyme

immunoassay (EIA) kit (Bio-Rad Laboratories) as described by the manufacturer. A cutoff value for positivity in serum of > 0.5 was used.

PK-PD modeling. The pharmacokinetic (i.e., serum voriconazole concentrations) and pharmacodynamic (i.e., galactomannan values) data from the 12 children were necessarily sparse, as they were collected as part of routine clinical care rather than as part of a prospective clinical trial. In addition, these sparse pharmacokinetic and pharmacodynamic data were not necessarily optimally informative. Fitting any pharmacokinetic model to a limited, sparse, and nonoptimally informative data set either is not possible or will lead to biased parameter estimates. We circumvented this problem using a two-step process. In the first step, each of the 12 new patients had their pharmacokinetics estimated using a previously described population PK model in which the PK model served as the Bayesian prior (11). In the second step, the Bayesian posterior estimates for each patient's PK parameters were fixed, and the pharmacodynamic parameters were then estimated by fitting the pharmacodynamic component of the model to each patient's galactomannan data. The population program Pmetrics was used for all modeling (17).

The structural pharmacokinetic mathematical model consisted of three differential equations that described the rate of change of the amount of voriconazole within each compartment. A fourth equation described the rate of change of galactomannan in the serum. These four inhomogeneous ordinary differential equations are as follows:

$$\frac{dX_1}{dt} = -K_a \cdot X_1 \quad (1)$$

$$\frac{dX_2}{dt} = K_a \cdot X_1 + \text{RateIV} - \frac{V_{\max}}{K_m \cdot V + X_2} \cdot X_2 - K_{cp} \cdot X_2 + K_{pc} \cdot X_3 \quad (2)$$

$$\frac{dX_3}{dt} = K_{cp} \cdot X_2 - K_{pc} \cdot X_3 \quad (3)$$

$$\frac{dX_4}{dt} = \text{KGM}_{\text{prod}} \cdot \left[1 - \left(\frac{x_4}{\text{POP}_{\text{max}}} \right) \right] \cdot \left(1 - \frac{\frac{X_2^H}{V}}{\text{EC}_{50}^H + \frac{X_2^H}{V}} \right) \cdot X_4 \quad (4a)$$

$$- \text{KGM}_{\text{elim}} \cdot X_4 \quad (4b)$$

where X_1 , X_2 , and X_3 are the amounts (in milligrams) in the gut, central compartment, and peripheral compartment, respectively, and dX_n/dt is the instantaneous rate of change in the amount of drug in compartment $n = 1, 2, \text{ or } 3$, K_a is the first-order rate constant of drug absorption after an oral bolus dose from the gut compartment (compartment 1) to the central serum compartment (compartment 2), RateIV is the rate of intravenous voriconazole infusion; V_{\max} is the maximum rate of the enzyme activity in metabolism of voriconazole (mg/h) and was allometrically scaled for body weight (kg) using the equation $V_{\max} = V_{\max 0} \cdot \text{kg}^{0.75}$, K_m is the concentration of voriconazole in the central compartment at which voriconazole clearance is half-maximal, V is the volume of the central compartment (liters) and was also allometrically scaled as $V = V_0 \cdot \text{weight}$, K_{cp} and K_{pc} are the first-order rate constants connecting the central compartment and peripheral compartment (compartment 3), X_4 is the concentration of galactomannan in the serum, KGM_{prod} is the maximal rate of production of galactomannan in the central compartment, POP_{max} is the maximal achievable galactomannan value, KGM_{elim} is the maximal rate of elimination of galactomannan from the central compartment, H controls the steepness of the relationship between drug concentration and reduction in galactomannan production in the central compartment, and EC_{50} is the concentration of voriconazole at which half-maximal reduction in galactomannan production is achieved. The oral bioavailability of voriconazole, F , was included because patients received voriconazole both orally and intravenously. In Pmetrics, F is a multiplier on oral doses, and it is not included within the differential equations.

Equations 1, 2, and 3 describe the rates of change of voriconazole in the gut, central serum, and peripheral tissue kinetic compartments, respectively. Equation 4 describes the rate of change of galactomannan in the central serum kinetic compartment, and we divide this equation into two separate terms for clarity. First, equation 4a describes the production of galactomannan, which is limited by a maximum value and also by voriconazole concentrations in a sigmoidal function, such that when concentrations are infinite, production is zero; second, equation 4b describes the sum of all physiologic galactomannan elimination mechanisms. A baseline galactomannan value within compartment 4 on the day of the first voriconazole dose was also estimated within Pmetrics, with a possible range of 0.1 to 12, reflective of clinically observed extremes. The fit of the mathematical model to the data was assessed using visual inspection and linear regression of the observed versus predicted values both before and after the Bayesian step.

Exposure-response relationships. The relationship between a traditional pharmacodynamic measures of drug exposure, such as the ratio of the area under the concentration-time curve (AUC) to the MIC, and a therapeutic response is often not possible to determine for invasive aspergillosis because the invading pathogen is usually not recovered. Therefore, we used a new concept in these analyses. The AUC/EC_{50} is the ratio of the voriconazole daily AUC to the EC_{50} , which is the posterior Bayesian estimate of the (*in vivo*) concentration of voriconazole required to induce half-maximal reduction in galactomannan levels in each individual patient. Thus, the EC_{50} is analogous to the more traditional *in vitro* estimate of drug potency, which is the MIC, but instead reflects an *in vivo* estimate of potency that can be derived from the change in galactomannan and voriconazole drug concentrations. The average daily AUC was calculated by estimating the total fitted AUC for each patient and dividing by the number of 24-hour treatment intervals. The average AUC circumvents the problem of defining which AUC is important for treatment effect (e.g., the AUC following the first dose or after a week of dosing). The relationship between the AUC/EC_{50} ratio and the final galactomannan level or survival was explored.

RESULTS

Demographics. The demographic data for the study population are summarized in Table 1. Fifty percent of patients had either acute myelogenous leukemia (AML) or acute lymphoblastic leukemia (ALL). The total mortality rate of the patient population was distressingly high: 10 (83.3%) of the 12 children died. For 4 of the 12 patients, *Aspergillus* spp. were recovered, and three of these four patients died from invasive aspergillosis. In the remaining patients, a diagnosis of probable invasive aspergillosis was established using galactomannan.

TDM data for voriconazole and galactomannan. There were a total of 261 and 33 measurements available for voriconazole concentrations and galactomannan levels from the 12 children, respectively. The concentration-time profiles for these respective readouts are shown in Fig. 1.

Population PK-PD model. The fit of the population PK-PD model to the data was acceptable despite the extreme pharmacokinetic and pharmacodynamic variability that is evident in Fig. 1. The Bayesian posterior estimates for the pharmacokinetics and pharmacodynamics are shown in Fig. 2 (left and right panels, respectively). The pharmacodynamics (i.e., galactomannan) were not well described using either the mean or median values for the parameters from the population model (data not shown). There simply was not a single set of parameter values that could be identified that was adequate to describe the time course of galactomannan in every patient. In contrast, however, the time course of galactomannan in each individual patient was readily described with a high degree of precision using the Bayesian posterior esti-

TABLE 1 Patients demographics and characteristics

Parameter	Value ^a
Demographics	
Male gender	8 (58.3)
Age (yr)	6 (5–10)
Wt (kg)	22.4 (18.8–33)
Underlying disease	
ALL	5 (41.7)
AML	1 (8.3)
Neuroblastoma	3 (25)
Other malignancies ^b	3 (25)
EORTC classification^c	
Proven	2 (16.7)
Probable	6 (50)
Possible	4 (33.3)
Voriconazole administration	
Intravenous (mg/kg/dose)	6.1 (4.7–6.9)
Oral (mg/kg/dose)	10.2 (6.8–10.8)
Voriconazole trough concn (mg/liter)	
	4.1 (1.6–6.1)
Galactomannan index	
	1.3 (0.5–8.1)

^a Values are number (percentage) of patients or median (interquartile range).

^b Other malignancies included non-Hodgkin lymphoma, immunodeficiency, osteosarcoma, Hodgkin lymphoma, and rhabdomyosarcoma.

^c Based on the opinion of the attending physician of the Children's Oncology ward at the beginning of the admission. Patients were classified as having possible invasive aspergillosis if the galactomannan was initially negative when voriconazole was commenced but later became positive, at which time the diagnostic classification was upgraded to probable.

mates for each patient. The heterogeneity of the different trajectories of galactomannan in individual patients is evident in Fig. 1 and 3. Of note, the initial condition (i.e., the galactomannan level at the commencement of treatment) was strikingly different between patients, potentially reflecting differences in underlying fungal burden. Furthermore, the time course of galactomannan in response to voriconazole therapy was also highly variable and ranged from a prompt decrease through to persistent antigenemia with no apparent therapeutic response.

Relationship between AUC/EC_{50} ratio and terminal galactomannan level or survival. The relationship between the AUC/EC_{50} ratio and the terminal galactomannan level is shown in Fig. 4. Using a simple nonlinear relationship, terminal galactomannan was strongly predicted by $(AUC/EC_{50})/15.4$ ($P = 0.003$). As a possible breakpoint, patients with an AUC/EC_{50} ratio of >6 tended to have a more consistently lower terminal galactomannan level ($P = 0.07$). In contrast, the AUC/EC_{50} ratio was not associated with survival. The mean in those who died was 6.1, and that in those who survived was 7.6 ($P = 0.76$).

DISCUSSION

Much has been written about the use of therapeutic drug monitoring as an indispensable adjunct to the use of voriconazole for the treatment of invasive aspergillosis and other invasive fungal diseases (9). There is a strong and growing evidence base to support such an assertion. Patients with serum concentrations of <1 mg/liter appear to have poorer clinical outcomes and higher mortality than patients with concentrations of >1 mg/liter (18). Similarly, patients with trough concentrations of >5 to 6 mg/liter

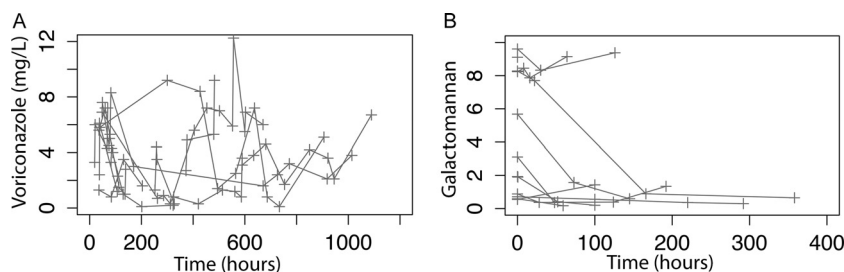


FIG 1 Voriconazole concentration-time profiles (A) and galactomannan-time profiles (B) for 12 pediatric patients who had concomitantly collected galactomannan and serum voriconazole concentration data. The samplings of voriconazole and galactomannan were not linked; hence, voriconazole serum concentration data were available after galactomannan sampling had stopped.

have a higher probability of having hepatotoxicity and confusion (18, 19). The case for routine TDM is further enhanced by the extreme pharmacokinetic variability that is characteristic of voriconazole and clearly evident in this study. The question raised by these analyses is whether TDM and dosage adjustment to achieve desired serum drug concentrations constitute the ultimate solution for using voriconazole and whether they constitute “true individualized therapy.”

The current strategy for TDM of voriconazole (or any other antimicrobial) is quite inconsistent with respect to individualization. The case for quantifying and controlling individual pharmacokinetic variability through dose modification has been made time and again by many people (including us). We have gone as far as use the information stored within population pharmacokinetic models to construct software that can be used for voriconazole dosage individualization in adults and children (10, 11). Importantly, the use of such software demands that the clinician define a drug concentration target that is deemed to have a high probability of therapeutic success and a low probability of toxicity. All the therapeutic targets that are used and cited in various guidelines are derived from large populations of patients, which are in effect “average” values. Such an approach is counter to all notions of individualized therapy and in fact is “one-size-fits-all” target selection. A significant advance that is enabled by the use of biomarkers such as galactomannan is the prospect of achieving true individualized target concentrations based on measured pharmacodynamics. Some patients will need more drug exposure, while others will need less. A different way of expressing this idea is that

both the pharmacokinetics and pharmacodynamics are different from patient to patient but need to be optimized for an individual. *A priori* the trajectory of the voriconazole concentration-time profile or the galactomannan in an individual patient is unknowable. Variability in both pharmacokinetics and pharmacodynamics contributes to both good and poor clinical outcomes, and the achievement of optimal clinical outcomes requires control of both.

Figure 3 is particularly illustrative of the many challenges facing clinicians who are treating children with invasive fungal diseases. First, the pharmacokinetics of voriconazole are highly variable, as previously described by us and many others. Second, and perhaps more important, is the observation that the pharmacodynamics are also highly variable. There is no way of predicting which path (galactomannan trajectory) an individual patient will follow once voriconazole is started. Results from phase II and III clinical studies (20, 21) suggest that on average a satisfactory clinical response will be obtained when a fixed dosing strategy is used, but that does not provide any guarantee that the patient has been dosed such that the likelihood of a response is above average. Galactomannan provides a real-time indication of the patient’s individual response to voriconazole and whether a therapeutic response is being achieved or not. It is possible to react to changes in galactomannan directly rather than just to the voriconazole concentration. Consider the differences in galactomannan responses between patient 177 and patient 180 in Fig. 3. Both patients achieve comparable voriconazole serum concentrations in the first days of therapy, but their pharmacodynamics are com-

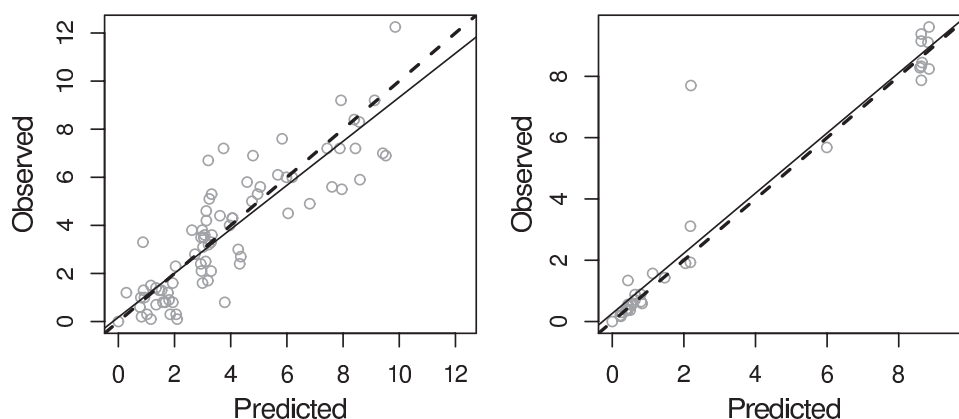


FIG 2 Observed versus predicted values after the Bayesian step for voriconazole serum concentrations (left panel) and for galactomannan (right panel). The solid lines are the linear regressions of the observed-predicted concentrations.

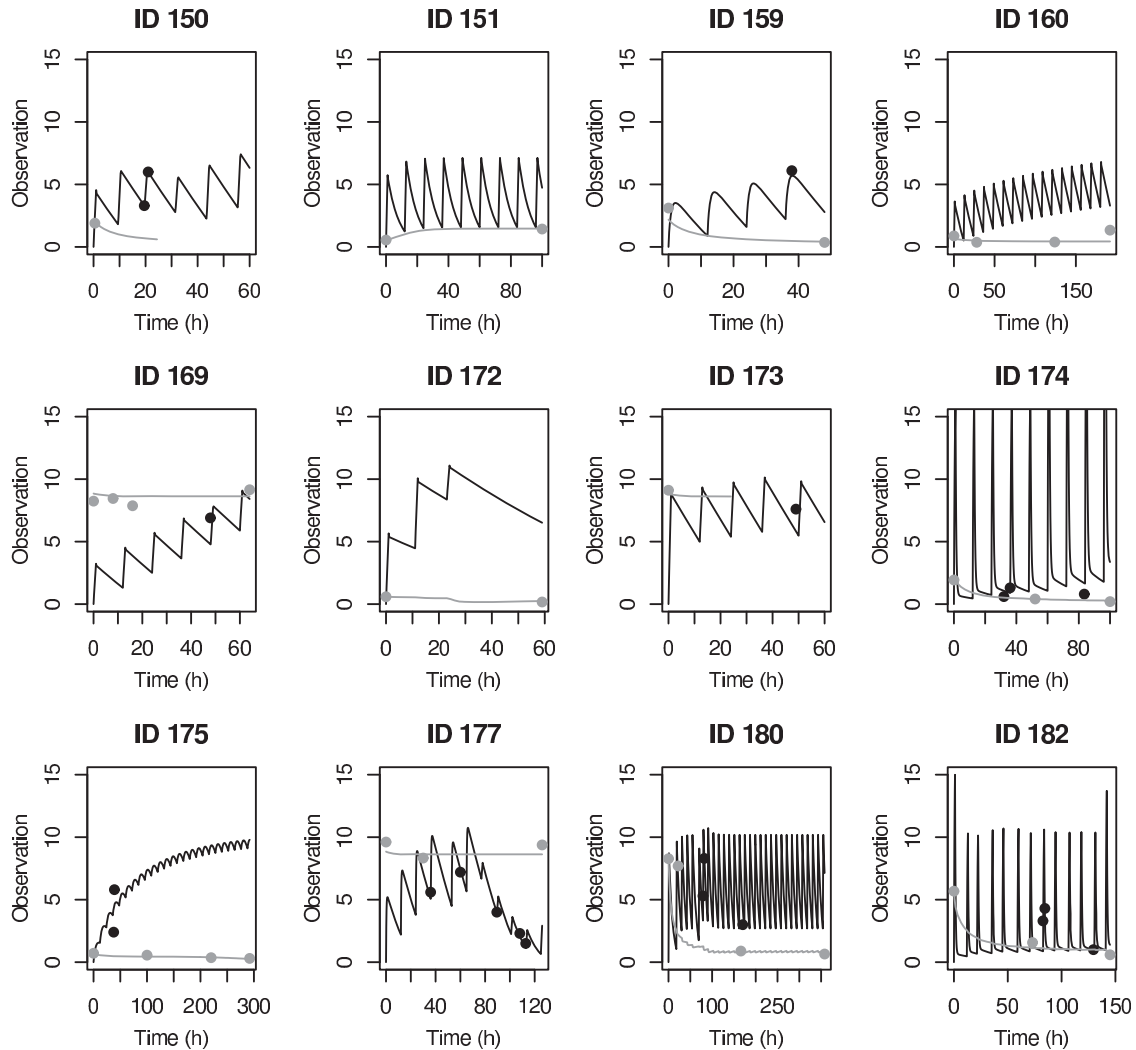


FIG 3 Serum voriconazole concentration-time profiles (solid black lines) and serum galactomannan-time profiles (gray lines) from the 12 children with concomitant PK and PD data. The raw data for voriconazole (black circles) and galactomannan (gray circles) are shown. In each case, the model fit is from the Bayesian posterior estimate. Only patients 159 and 180 survived.

pletely different for reasons that may not be immediately obvious. Patient 177 appears not to be responding to voriconazole and should have the dose increased, the drug changed to an alternative agent, or a second antifungal agent added. Instead, the dosage was reduced, probably because the upper TDM target was exceeded (again, this value is derived from a population of patients). However, the population value may not have been appropriate for that patient. This suboptimal regimen resulted in sustained galactomannan antigenemia, and the patient ultimately died. In contrast, patient 180 achieved a sustained response in their galactomannan trajectory (despite having voriconazole concentrations ordinarily considered to be associated with a higher probability of toxicity) and ultimately survived.

We do not claim that this is an ideal data set. The data are sparse and were not collected at optimally informative times. Fitting was difficult and required a preexisting population PK model that could be used as a Bayesian prior. Despite some limitations, it is remarkable that the PK-PD mathematical model fits any of the data, given that they were collected in routine clinical settings.

Importantly, however, there is an acceptable fit of the model to the data only after the Bayesian step. In this regard, fitting models to galactomannan data is similar to fitting mathematical models to drug resistance data, where population fits are often notoriously bad. The reason for this is obvious following a brief inspection of the raw data in Fig. 1B. The galactomannan data are nonmonotonic. Some profiles rise unexpectedly, while others fall. Such heterogeneity in response makes it nearly impossible to derive a single set of parameter values that account for all the data in a reasonably unbiased yet satisfactorily precise manner. We could have performed Monte Carlo simulation on the Bayesian posterior estimates to explore the impact of both pharmacokinetic and pharmacodynamic variability on the therapeutic outcome, but we ultimately decided that this would have produced unreliable results given the paucity of data (some patients have only one or two observations). However, this could easily be done in the future with larger and more comprehensive data sets.

The AUC/EC₅₀ ratio is a pharmacodynamic index that may be helpful in future studies of invasive aspergillosis. While the

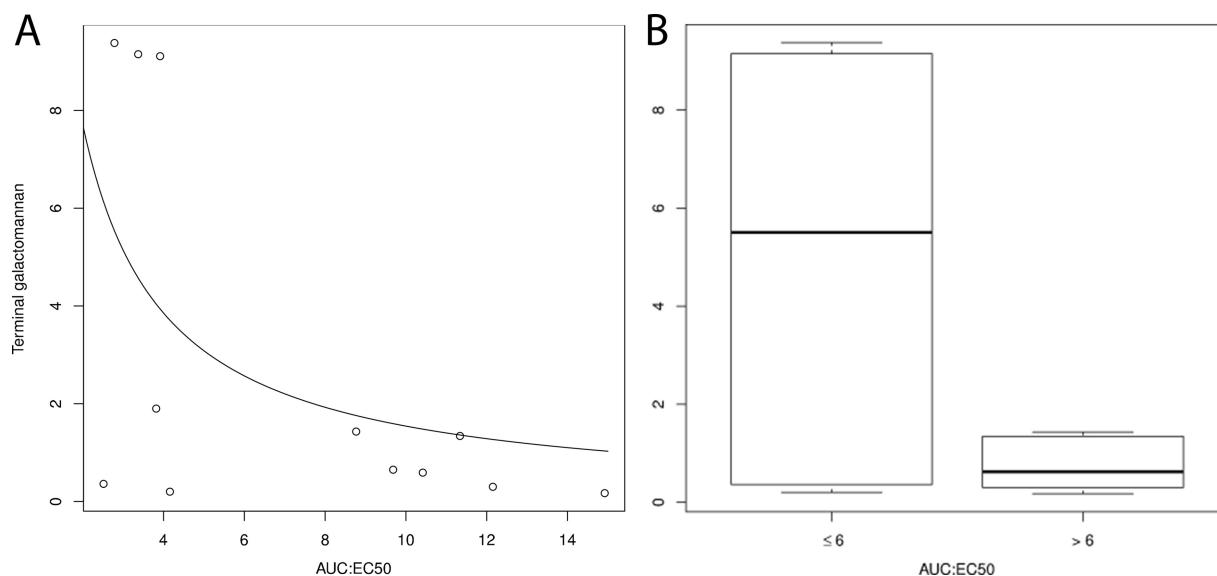


FIG 4 Relationship of voriconazole average daily AUC/EC_{50} ratio and final (i.e., last measured) galactomannan level in the 12 children. (A) Terminal galactomannan = $(AUC/EC_{50})/15.4$ ($P = 0.003$). (B) An AUC/EC_{50} ratio of >6 suggested a more consistently lower terminal galactomannan level ($P = 0.07$).

EC_{50} requires at least one measured voriconazole level and galactomannan level in a patient and requires some PK-PD modeling expertise, it captures and quantifies much of the pharmacodynamic variability that is evident in this study. Thus, the AUC/EC_{50} ratio provides an understanding of the therapeutic response in terms of drug exposure (AUC) as well as the pharmacodynamics. A high estimate for EC_{50} may be caused by factors such as a high fungal burden, the presence of antifungal resistance mechanisms, a delay in the initiation of antifungal therapy, infection within sanctuary sites, and profound immunosuppression. In this way, we view it as potentially superior to the *in vitro* MIC, which does not account for the clinical therapeutic environment within a patient. The AUC/EC_{50} ratio is a fully individualized *in vivo* estimate of drug potency, and it significantly predicted terminal galactomannan levels, even in this small study. It did not predict survival, but the majority of this cohort died from a range of causes, including the underlying disease. Furthermore, terminal galactomannan is likely a more objective reflection of *in vivo* voriconazole efficacy than survival, which is multifactorial, especially in these kinds of patients with complex underlying medical problems.

The next steps are clear. Larger, richer data sets that contain optimally informative sampling for both voriconazole and galactomannan will enable the construction of more robust pharmacokinetic-pharmacodynamic mathematical models. These models will form the basis of dual-output stochastic controllers where a clinician has the option to individualize dosing to control the serum drug concentrations, the circulating biomarker, or both. Such an advance represents a further critical step toward the provision of true individualized therapy, which is surely the ultimate goal of all clinicians treating any patient with a life-threatening invasive fungal infection. Such an approach is one key advance for better care of immunocompromised patients who usually have multiple comorbidities. Moreover, as circulating biomarkers are developed for other diseases, this approach can be applied to a wider range of infections.

ACKNOWLEDGMENTS

William Hope is supported by a Clinician Scientist Fellowship from the National Institute of Health Research (NIHR). Michael Neely is supported by NICHD grant R01 HD070886.

Jan-Willem C. Alffenaar received funding from Pfizer, MSD, and Astellas for investigator-initiated studies. William Hope has received research funding from Pfizer, Gilead, Astellas, and F2G and has acted as a consultant and/or given talks for Pfizer, Basilea, F2G, Nordic Pharma, Mayne Pharma, and Pulmocide.

FUNDING INFORMATION

National Institute of Health Research provided funding to William W. Hope under grant number CS_08_08. HHS | National Institutes of Health (NIH) provided funding to Michael Neely under grant number NICHD R01 HD070886.

REFERENCES

- Denning DW, Hope WW. 2010. Therapy for fungal diseases: opportunities and priorities. *Trends Microbiol* 18:195–204. <http://dx.doi.org/10.1016/j.tim.2010.02.004>.
- Groll AH, Castagnola E, Cesaro S, Dalle J-H, Engelhard D, Hope W, Roilides E, Styczynski J, Warris A, Lehrnbecher T. 2014. Fourth European Conference on Infections in Leukaemia (ECIL-4): guidelines for diagnosis, prevention, and treatment of invasive fungal diseases in paediatric patients with cancer or allogeneic haemopoietic stem-cell transplantation. *Lancet Oncol* 15:e327–e340. [http://dx.doi.org/10.1016/S1470-2045\(14\)70017-8](http://dx.doi.org/10.1016/S1470-2045(14)70017-8).
- Walsh TJ, Anaissie EJ, Denning DW, Herbrecht R, Kontoyiannis DP, Marr KA, Morrison VA, Segal BH, Steinbach WJ, Stevens DA, van Burik JA, Wingard JR, Patterson TF. 2008. Treatment of aspergillosis: clinical practice guidelines of the Infectious Diseases Society of America. *Clin Infect Dis* 46:327–360. <http://dx.doi.org/10.1086/525258>.
- Wang G, Lei HP, Li Z, Tan ZR, Guo D, Fan L, Chen Y, Hu DL, Wang D, Zhou HH. 2009. The CYP2C19 ultra-rapid metabolizer genotype influences the pharmacokinetics of voriconazole in healthy male volunteers. *Eur J Clin Pharmacol* 65:281–285. <http://dx.doi.org/10.1007/s00228-008-0574-7>.
- Jeu L, Piacenti FJ, Lyakhovetskiy AG, Fung HB. 2003. Voriconazole. *Clin Ther* 25:1321–1381. [http://dx.doi.org/10.1016/S0149-2918\(03\)80126-1](http://dx.doi.org/10.1016/S0149-2918(03)80126-1).
- Neely M, Rushing T, Kovacs A, Jelliffe R, Hoffman J. 2010. Voriconazole

- zole pharmacokinetics and pharmacodynamics in children. *Clin Infect Dis* 50:27–36. <http://dx.doi.org/10.1086/648679>.
7. van Wanrooy MJP, Span LFR, Rodgers MGG, van den Heuvel ER, Uges DR a, van der Werf TS, Kosterink JGW, Alfenaar J-WC. 2014. Inflammation is associated with voriconazole trough concentrations. *Antimicrob Agents Chemother* 58:7098–7101. <http://dx.doi.org/10.1128/AAC.03820-14>.
 8. Brüggemann RJM, Alfenaar JC, Blijlevens NMA, Billaud EM, Kosterink JGW, Verweij PE, Burger DM. 2009. Clinical relevance of the pharmacokinetic interactions of azole antifungal drugs with other coadministered agents. *Clin Infect Dis* 48:1441–1458. <http://dx.doi.org/10.1086/598327>.
 9. Ashbee HR, Barnes RA, Johnson EM, Richardson MD, Gorton R, Hope WW. 2014. Therapeutic drug monitoring (TDM) of antifungal agents: guidelines from the British Society for Medical Mycology. *J Antimicrob Chemother* 69:1162–1176. <http://dx.doi.org/10.1093/jac/dkt508>.
 10. Hope WW, Vanguilder M, Donnelly JP, Blijlevens NM a, Brüggemann RJM, Jelliffe RW, Neely MN, Brüggemann RJ. 2013. Software for dosage individualization of voriconazole for immunocompromised patients. *Antimicrob Agents Chemother* 57:1888–1894. <http://dx.doi.org/10.1128/AAC.02025-12>.
 11. Neely M, Margol A, Fu X, van Guilder M, Bayard D, Schumitzky A, Orbach R, Liu S, Louie S, Hope W. 2015. Achieving target voriconazole concentrations more accurately in children and adolescents. *Antimicrob Agents Chemother* 59:3090–3097. <http://dx.doi.org/10.1128/AAC.00032-15>.
 12. De Pauw B, Walsh TJ, Donnelly JP, Stevens DA, Edwards JE, Calandra T, Pappas PG, Maertens J, Lortholary O, Kauffman CA, Denning DW, Patterson TF, Maschmeyer G, Bille J, Dismukes WE, Herbrecht R, Hope WW, Kibbler CC, Kullberg BJ, Marr KA, Muñoz P, Odds FC, Perfect JR, Restrepo A, Ruhnke M, Segal BH, Sobel JD, Sorrell TC, Viscoli C, Wingard JR, Zaoutis T, Bennett JE, Pauw De B, Mun P. 2008. Revised definitions of invasive fungal disease from the European Organization for Research and Treatment of Cancer/Invasive Fungal Infections Cooperative Group and the National Institute of Allergy and Infectious Diseases Mycoses Study Group (EORTC/MSG). *Clin Infect Dis* 46:1813–1821. <http://dx.doi.org/10.1086/588660>.
 13. Hope WW, Kruhlak MJ, Lyman CA, Petraitiene R, Petraitis V, Francesconi A, Kasai M, Mickiene D, Sein T, Peter J, Kelaher AM, Hughes JE, Cotton MP, Cotten CJ, Bacher J, Tripathi S, Bermudez L, Maugel TK, Zerfas PM, Wingard JR, Drusano GL, Walsh TJ. 2007. Pathogenesis of *Aspergillus fumigatus* and the kinetics of galactomannan in an in vitro model of early invasive pulmonary aspergillosis: implications for antifungal therapy. *J Infect Dis* 195:455–466. <http://dx.doi.org/10.1086/510535>.
 14. Maertens J, Theunissen K, Verhoef G, Verschakelen J, Lagrou K, Verbeken E, Wilmer A, Verhaegen J, Boogaerts M, Eldere Van J. 2005. Galactomannan and computed tomography-based preemptive antifungal therapy in neutropenic patients at high risk for invasive fungal infection: a prospective feasibility study *Clin Infect Dis* 41:1242–1250.
 15. Nouér SA, Nucci M, Kumar NS, Graziutti M, Barlogie B, Anaissie E. 2011. Earlier response assessment in invasive aspergillosis based on the kinetics of serum aspergillus galactomannan: proposal for a new definition. *Clin Infect Dis* 53:671–676. <http://dx.doi.org/10.1093/cid/cir441>.
 16. Alfenaar JW, Wessels AM, van Hateren K, Greijdanus B, Kosterink JG, Uges DR. 2010. Method for therapeutic drug monitoring of azole antifungal drugs in human serum using LC/MS/MS. *J Chromatogr B Analyt Technol Biomed Life Sci* 878:39–44. <http://dx.doi.org/10.1016/j.jchromb.2009.11.017>.
 17. Neely MN, van Guilder MG, Yamada WM, Schumitzky A, Jelliffe RW. 2012. Accurate detection of outliers and subpopulations with Pmetrics, a nonparametric and parametric pharmacometric modeling and simulation package for R. *Ther Drug Monit* 34:467–476. <http://dx.doi.org/10.1097/FTD.0b013e31825c4ba6>.
 18. Pascual A, Calandra T, Bolay S, Buclin T, Bille J, Marchetti O. 2008. Voriconazole therapeutic drug monitoring in patients with invasive mycoses improves efficacy and safety outcomes. *Clin Infect Dis* 46:201–211. <http://dx.doi.org/10.1086/524669>.
 19. Tan K, Brayshaw N, Tomaszewski K, Troke P, Wood N. 2006. Investigation of the potential relationships between plasma voriconazole concentrations and visual adverse events or liver function test abnormalities. *J Clin Pharmacol* 46:235–243. <http://dx.doi.org/10.1177/0091270005283837>.
 20. Denning DW, Ribaud P, Milpied N, Caillot D, Herbrecht R, Thiel E, Haas A, Ruhnke M, Lode H. 2002. Efficacy and safety of voriconazole in the treatment of acute invasive aspergillosis. *Clin Infect Dis* 34:563–571. <http://dx.doi.org/10.1086/324620>.
 21. Herbrecht R, Denning DW, Patterson TF, Bennett JE, Greene RE, Oestmann JW, Kern WV, Marr KA, Ribaud P, Lortholary O, Sylvester R, Rubin RH, Wingard JR, Stark P, Durand C, Caillot D, Thiel E, Chandrasekar PH, Hodges MR, Schlamm HT, Troke PF, de Pauw B. 2002. Voriconazole versus amphotericin B for primary therapy of invasive aspergillosis. *N Engl J Med* 347:408–415. <http://dx.doi.org/10.1056/NEJMoa020191>.